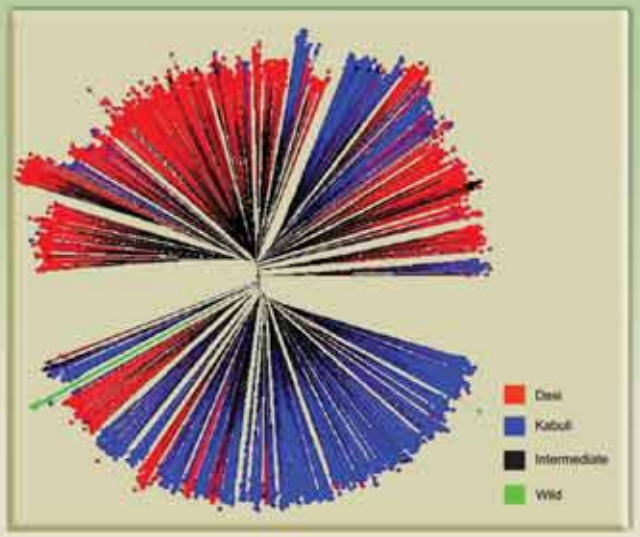
International Crops Research Institute for the Semi-Arid Tropics

Global Themes: Biotechnology and Crop Improvement 2005-2006

Archival report from genes to germplasm



Global Themes: Biotechnology and Crop Improvement 2005-2006

Archival report from genes to germplasm

ICRISAT
International Crops Research Institute for the Semi-Arid Tropics
Patancheru 502 324, Andhra Pradesh, India
icrisat@cgiar.org

ICRISAT Archival Report 2005

Projects 2, 3, 4, 5 and 6 from: Global Themes Biotechnology and Crop Improvement

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from

Global Themes Biotechnology and Crop Improvement

Project 2
Sustaining Biodiversity of Sorghum, Pearl Millet, Small Millets, Groundnut, Pigeonpea and Chickpea for Current and Future Generations

Output 1.1: Germplasm conserved, evaluated, documented and exchanged

Summary

Genetic resources are important and essential components of crop improvement. The benefits of these resources largely depend on the diversity of the collection, their adequate characterization and documentation, and user access to the collection. Some of the significant genebank activities to accomplish these objectives are as follows.

During 2005, we assembled 2633 new accessions (chickpea: 2014 and sorghum: 619) in the genebank. With these additions, the total accessions increased to 117,503 from 130 countries. A set of 483 sorghum germplasm samples, collected by a group of researchers from INRAN, UMA, ICRISAT and CIRAD in Niger in 2003, was received at Patancheru after obtaining the Germplasm Acquisition Agreement (GAA) from the Government of Niger. Efforts are underway for obtaining 424 pearl millet samples collected from this mission. A set of 622 groundnut accessions were acquired from National Agrobiological Sciences (NIAS), Ibaraki, Japan that were unique in comparison with those in the ICRISAT Genebank. Similarly, 840 chickpea accessions were acquired from USDA, Pullman,USA. Both the sets were planted for quarantine observations for possible release during 2006.

A total of 2984 accessions were processed for medium-term storage (MTS) and 8486 accessions for long-term storage (LTS). We also prepared 8225 seed samples of different crops for off-site duplication as safety back-up. Safety back-up collection was established at Niamey regional genebank for 11,791 accessions. Seed viability of 1741 accessions from MTS (three to eight years old) and 3238 accessions from LTS for (over 10 years old) was monitored. This resulted in identifying 179 accessions in MTS (<75% viability) and 69 accessions in LTS (<85% viability) for regeneration during 2006. Regenerated 3037 accessions with critical seed stock and viability for MTS; 5723 accessions for LTS and 4417 accessions for safety back-up. Additionally, 303 critical accessions were regenerated under controlled conditions of greenhouse facility. The seed health of 7582 accessions identified for LTS was tested using standard protocols and the results were documented.

We characterized 3916 accessions during 2004 rainy and 3413 during 2004–05 postrainy seasons (full characterization or collecting data on missing traits). Additionally, 3904 accessions of different crops were evaluated for special traits useful in crop improvement. Germplasm composite sets of 1000 accessions each of pigeonpea and finger millet and 850 accessions of groundnut were grown for evaluation. To enhance utilization of germplasm, mini-core sets of chickpea and groundnut were evaluated at several locations in six countries. Core set of finger millet was evaluated at three locations in India and one in Kenya and mini-core set of finger millet at one location in India. Pigeonpea core set (1290 accessions) was evaluated at ICRISAT-Patancheru.

The Genebank Information Management System was updated to meet the users' requirement. Germplasm databases of chickpea at ICARDA (Syria) and groundnut at NIAS (Japan) were compared for identifying unique accessions for the ICRISAT Genebank. Characterization and evaluation data on large number of cultivated and wild species accessions were computerized.

During the year, 30,985 germplasm samples were supplied to users on request. This included 4325 samples to scientists outside ICRISAT (22 countries and 87 requests) and 26,660 samples for use by ICRISAT scientists.

Activity 1.1.1: Assemble, conserve, and regenerate the global germplasm collection for safe storage and supply

Milestone: New germplasm collected from priority areas, and assembled germplasm safely conserved as active and base collection for utilization (2007)

During 2005, a total of 2633 new accessions were assembled in the genebank. This included 2014 accessions of chickpea (68 wild and 682 cultivated from ICARDA and 21 wild and 1243 cultivated from USDA-Pullman, USA) and 619 sorghum accessions. The sorghum accessions were those that were missing from the earlier Rockefeller Foundation Collections. These were subsequently acquired from National Seed Storage Laboratory, Fort Collins, USA. With these additions, the total number of accessions in the genebank increased to 117,503

from 130 countries. A set of 483 sorghum germplasm samples collected in Niger in 2003 by a group of researchers from INRAN, UMA, ICRISAT and CIRAD was received at Patancheru after obtaining the Germplasm Acquisition Agreement (GAA) from the Government of Niger. Efforts are underway for obtaining 424 pearl millet samples collected during this mission. A total of 622 groundnut accessions from National Agrobiological Sciences (NIAS), Tsukuba, Japan and 840 chickpea collections from USDA, Pullman, USA were identified as unique for assembly at ICRISAT Genebank. We are in the process of securing 231 pigeonpea samples that were collected in Mozambique (79), Tanzania (123) and Uganda (29) from ICRISAT Regional Program, Nairobi, Kenya.

We processed germplasm seed samples of 2984 accessions (sorghum: 279, pearl millet: 76, chickpea: 271, pigeonpea: 39, groundnut: 2036 and small millets: 283) for MTS and 8486 accessions (sorghum: 850, pearl millet: 1250, chickpea: 575, pigeonpea: 1281 and groundnut: 4530) for LTS. These were the harvests of 2004 rainy and 2004–05 postrainy season grow outs. With these additions, the total collection in LTS increased to 92,968 accessions that represent 79.1% of the entire collection.

During this year, the seed viability of 11,691 accessions for MTS and LTS was tested. This included: 1220 (sorghum), 1811 (pearl millet), 2649 (chickpea), 1281 (pigeonpea) and 4730 (groundnut) accessions. The seed viability of 1741 accessions (sorghum – 370, pearl millet – 561, pigeonpea – 570 and groundnut – 240) in MTS for 3 to 10 years; and 3238 accessions (pearl millet – 1836 and groundnut – 1402) in LTS for over 10 years was monitored. This led to identification of 179 accessions in MTS (<75% viability) and 69 accessions in LTS (<85% viability) for regeneration during 2006. A total of 7582 germplasm accessions (sorghum – 1070, chickpea – 569, pigeonpea – 1281, groundnut – 3998, and pearl millet – 664) regenerated from medium-term storage of ICRISAT Genebank were evaluated for seed health using the standard blotter method and the results were documented.

To achieve the germplasm safety back-up at other locations, we prepared seed samples of 8225 accessions (sorghum – 1932, chickpea – 2763, pigeonpea – 1208 and groundnut – 2322) from 2004 rainy season and postrainy season plantings. At regional genebank – Niamey, Niger, safety back-up collection was established for 11,791 accessions (pearl millet – 5205, groundnut – 2006 and small millets – 4580). A set of 1877 accessions of chickpea is awaiting back-up storage at ICARDA, Aleppo, Syria.

HD Upadhyaya and CLL Gowda

Milestone: Germplasm accessions with limited seed stock/viability regenerated for medium- and long-term conservation, and tested for seed health (2007)

We regenerated 3037 accessions (sorghum – 25, pearl millet – 771, chickpea – 220, pigeonpea – 1080, groundnut – 537, finger millet – 238, foxtail millet – 148, barnyard millet – 3, little millet – 3, proso millet – 2, and kodo millet – 10) for MTS and 5723 accessions representing sorghum (2000), pearl millet (769), chickpea (452), pigeonpea (427) and groundnut (2075) for LTS during the 2005. Special regenerations were carried for 4417 accessions (chickpea: 2667 and groundnut: 1750) for safety back-up. In the special facility for wild *Arachis*, 182 accessions representing 28 species were grown for seed increase. Additionally, 121 critical accessions of different crops (chickpea – 108, pigeonpea – 10, sorghum – 1 and finger millet – 2) were regenerated in the glasshouse facilities.

HD Upadhyaya and CLL Gowda

Seed health testing of germplasm accessions for medium- and long-term storage in the genebank: A total of 7582 germplasm accessions (sorghum – 1070, chickpea – 569, pigeonpea – 1281, groundnut – 3998, and pearl millet – 664) regenerated from medium-term storage of ICRISAT Genebank were evaluated for seed health using the standard blotter method. Only 233 of 7582 accessions were free from any seed-borne pathogens (sorghum – 26, pearl millet – 29, chickpea – 49, pigeonpea – 35, and groundnut – 94). Forty seed-associated fungal species in sorghum, 22 in pearl millet, 24 in chickpea, and 32 each in groundnut and pigeonpea were detected. The major fungi genera associated with seed of all five crops were species of *Cladosporium*, *Alternaria*, *Fusarium*, *Phoma*, *Curvularia*, *Bipolaris, Aspergillus, Penicillium*, *Rhizoctonia bataticola* and *Periconia*. Some of these fungi, such as species of *Alternaria, Fusarium, Curvularia, Phoma* and *Rhizoctonia* were seed-borne and thus affected seed germination up to 4.5% in sorghum, 2.7% in pearl millet, 4.2% in chickpea, 4.8% in pigeonpea and 0.75% in groundnut. Bacterial growth, in traces, was detected in all crops without any significant effects on seed viability.

RP Thakur and *HD Upadhyaya*

Milestone: Requested germplasm distributed to bona fide users for utilization (2007)

A total of 30,985 samples of in-trust germplasm accessions were distributed to scientists globally for utilization. This includes 4325 samples (329 sorghum; 369 pearl millet, 1511 chickpea; 98 pigeonpea; 339 groundnut; and 1679 small millets) to 22 countries (Table 1.1) under 87 consignments; and 26,660 samples to users within ICRISAT. The total included: sorghum – 8140, pearl millet – 2714, chickpea – 3627, pigeonpea – 3042, groundnut – 7573 and small millets – 1564. Additionally, leaf samples of germplasm composite sets (sorghum – 2300, chickpea – 2774, and pigeonpea – 1000) were provided for DNA extraction within the institute.

HD Upadhyaya, CLL Gowda and RP Thakur

Table 1.1. Germplasm samples distributed from ICRISAT-Patancheru Genebank to scientists globally during the year 2005

Country	Sorghum	Pearl millet	Chickpea	Pigeonpea	Groundnut	Small millets	Total
Bangladesh					1		1
Canada			213				213
China					188		188
Denmark	7						7
Fiji	10						10
France		49					49
Germany	37						37
Haiti	11				11		22
India	149	88	1065	46	119	1156	2623
Iran	58	7		19			84
Japan	1	8	208	3			220
Jordan		20					20
Kenya						506	506
Mexico	14						14
Niger		120					120
Pakistan		22					22
Papua New Guinea			24	16	19		59
Somalia	42						42
Spain			1				1
Thailand					1		1
USA		55		14			69
Uruguay						17	17
Total	**329**	**369**	**1511**	**98**	**339**	**1679**	**4325**

Activity 1.1.2: Undertake characterization, evaluation and documentation of germplasm to enhance its utility in crop improvement

Milestone: New germplasm characterized for morpho-agronomic characters and evaluated for special traits (2006)

During the 2005 rainy season, a total of 3916 accessions of pearl millet (512), pigeonpea (900) and groundnut (2504), and in the postrainy season, 3413 accessions of sorghum (156), pearl millet (512), chickpea (1745) and

groundnut (1000) were characterized for morpho-agronomic traits (full characterization or collecting missing data).

In addition, 3904 accessions of different crops (sorghum – 129, pearl millet – 504, finger millet – 1020, foxtail millet – 175 chickpea – 202, pigeonpea – 1000 and groundnut – 874) were grown for recording agronomic and special traits useful in crop improvement. The traits/sets in different crops include: 32 zerazera type, 62 yellow endosperm and composite collection of 35 accessions in sorghum; a core set of 504 accessions of pearl millet and composite set of 1000 accessions; 20 elite accessions in finger millet; core set of 155 and 20 elite accessions in foxtail millet; 34 large-seeded *kabuli*, 16 early-maturing, 54 salinity tolerant, 58 extra-early *kabuli*, 20 each of drought tolerant lines (large root length density and deep roots) in chickpea; composite set of 1000 accessions of pigeonpea and composite set of 850 and 24 extra-early maturing accessions in groundnut.

The finger millet core collection was evaluated at three locations in India (ICRISAT-Patancheru, Bangalore and Vizianagaram), and at one location in Kenya. Pigeonpea core set (1290 accessions) was evaluated at ICRISAT-Patancheru location.

Finger millet composite collection of 1000 accessions (ICRISAT finger millet core collection – 622, agronomically elite – 222, core collection of Indian NARS – 50, resistance to stresses – 85, grain nutrition traits – 12, and genetic diversity – 9) was characterized for important morpho-agronomic characters. A pigeonpea composite collection of 1000 accessions was grown under pollination control cages for characterization and regeneration.

Several promising sources of germplasm were identified from evaluations during 2004-05. From chickpea germplasm composite set (3000 accessions) - ICCs 8318, 17256, 8324, and 12197 (*desi*), IG 70779 (*kabuli*) and ICC 812 (pea-shaped seed) for yield (2.74–3.35 t ha^{-1}), ICCs 12034, 13821, 16641, and ICCV 96329 (*kabuli*) and ICCs 17258, 5810, and ICCV 96030 (*desi*) for early flowering and yield (33–36 days to 50% flowering and 1.18–2.02 t ha^{-1}); ICCs 12034, 7346, and 14205 for large seed size and yield (45.0–45.7 g 100^{-1} seed weight and 1.18–2.02 t ha^{-1} grain yield) among *kabuli* types and ICCs 14648, 4871, and 7672 (29.2–35.4 g 100^{-1} seed weight and 1.25–2.26 t ha^{-1}) among the *desi* types.

From new chickpea introductions (USA), lines EC no's 543533, 543598, 543584, 543593, and 543599 (all *kabuli* type) were identified for large seed size and yield (45.5–54.9 g 100^{-1} seed weight; 1.70–1.91 t ha^{-1} seed yield).

Based on the evaluation of 16 large-seeded *kabuli* chickpea accessions at six environments, lines ICCs 17109, 16670, 7344, 14194, and 8155 (35–37 days to 50% flowering; 50–57 g 100^{-1} seed weight) were identified as good sources of earliness combined with large seed size in comparison to control ICCV 2 (40 days and 27 g 100^{-1} seed). Similarly, based on the evaluation data of 28 early-maturing chickpea accessions from six environments - ICCs 16641 and 16644 (31–32 days to 50% flowering) were identified as additional sources for extra-earliness. ICCs 2859, 10232, 10629, 11160, 11180, and 14648 (1.36–1.66 t ha^{-1}) produced significantly greater seed yield than the early-maturing controls Harigantars (1.10 t ha^{-1}) and ICCV 2 (1.27 t ha^{-1}) in the pooled analyses.

From amongst 58 extra-early *kabuli* elite accessions tested, 12 accessions were early flowering (23–26 days to 50% flowering), large-seeded (27.4–35.5 g 100^{-1} seed weight) and high-yielding (2.38–2.82 t ha^{-1}) in comparison with ICCV 2 (27 days, 26.1 g, and 2.36 t ha^{-1}).

Based on eight seasons data of 21 early-maturing groundnut accessions at ICRISAT-Patancheru, the test entries produced an average pod yield of 1.08 t ha^{-1}, 8.2% more than the average of all the controls at 1240 ^{o}Cd [equivalent to 75 days after sowing (DAS) in rainy season]; and 1.46 t ha^{-1}, 12.6% more than the average of controls at 1470 ^{o}Cd (equivalent to 90 DAS) in the postrainy season. Two new sources earliness (ICG 3540 and ICG 14855) produced 22.6% and 16.8% higher pod yield at 1240 ^{o}Cd and 10.6% and 23.7% higher at 1470 ^{o}Cd than the earliest-maturing control Chico. These accessions also produced 27.9% and 21.9% higher pod yield at 1240 ^{o}Cd and 4.0% and 16.2% higher at 1470 ^{o}Cd than the control JL 24, respectively.

HD Upadhyaya and CLL Gowda

Milestone: New germplasm sources identified for water use efficiency traits in groundnut, and drought avoidance and salinity tolerance in chickpea (2005)

Groundnut composite collection consisting of 850 accessions was evaluated in an augmented design with four repeated control cultivars for pod yield potential and drought related traits. SPAD Chlorophyll Meter Reading (SCMR) and Specific Leaf Area (SLA) were recorded on these accessions of groundnut composite collection at 60 and 80 days after sowing (DAS). ICGs 2741, 5725, 5728, 6323, and 7878 were identified with high SCMR (53.9–61.0) (known to represent high water use efficiency).

Based on eight seasons data of 18 cultivated groundnut accessions for drought-related traits, ICGs 5745, 6766, 7243, 14523, and ICG 14475 (129–150 and 132–153 SLA at 60 and 80 DAS and 42–44 SCMR at both sampling dates) were identified as additional sources for drought tolerance traits in comparison to control CMSG 84-1 (144 and 150 SLA and 43 and 42 SCMR at 60 and 80 DAS, respectively).

Chickpea mini-core was evaluated for root length and root depth (which are responsible for drought tolerance). ICCs 8261, 10885, 16796, 13816, 13599, 1915, 15264, 6306, and 5337 were identified with large root length and density; and ICCs 3512, 15697, 13523, 1356, 4872, 7272, 8261, 95, 440, and 1431 for deep root system.

ICCs 15510, 4953, 7255, 14199, and 12908 (2.52–3.36 t ha^{-1}) were the best five accessions in comparison to control Jumbo 2 (1.10 t ha^{-1}) among the 54 salinity-tolerant chickpea accessions evaluated for morpho-agronomic traits during 2004-05 postrainy season. Groundnut mini-core was evaluated for salinity tolerance. ICGs 4890 and 4911 were found tolerant to salinity.

HD Upadhyaya, J Kashiwagi, Vincent Vadez, PM Gaur and CLL Gowda

Milestone: Mini-core collections of chickpea and groundnut evaluated for agronomic traits at different locations in Asia, and Southern Africa (2007)

During this year, chickpea mini-core set was evaluated at three locations in India (Patancheru, Kanpur and Ludhiana) and one location each in Canada and Japan. At the Indian Institute of Pulses Research (IIPR), Kanpur, India, 13 accessions of chickpea were selected for large seed size (>45 g 100^{-1} seed weight) for utilization in breeding programs. These are – ICCs 7344, 12033, 12034, 14194, 14195, 14196, 14197, 14199, 14204, 14203, 14204, 14205 and EC 381882.

From the groundnut mini-core set evaluated at Shandong Peanut Research Institute (SDPRI) China, several accessions were identified promising. These included – ICGs 5662, 6057, 6766, 11219 and 11855 for large seed size (78.4–105.2 g 100^{-1} seed weight); ICGs 36, 76, 118, 397, 434, 1415, 1448, 1455, 1668, 5745, 6057, 6201, 7633 and 14710 for bacterial wilt resistance (score 'zero'); ICGs, 118, 1101, 6022, 8567, 10890, 12672 and 14482 for high oil content (54.0–55.4%); ICGs 3053, 5745, 8285 and 8490 for high oleic acid (60.9–64.7%) and low linoleic acid (18.0–21.2%); and ICGs 3053, 5745 and 8490 for high oleic acid/linoleic acid ratio (3.0–3.6%). These accessions merit further tests for confirming the results.

Groundnut mini-core evaluation by Field Crops Research Center, Khon Kaen, Thailand resulted in identifying ICGs 297, 1668, 1973, 3027 and 13099 for high pod yield (3583–4168 kg ha^{-1}); ICGs 2106, 3240, 7969, 12879 and 12988 for higher shelling percentage (79.8–82.1%); and ICGs 4538, 5662, 6993, 8760 and 9777 for large seed size (77.5–100.5 g 100^{-1} seed weight). Groundnut mini-core collection was also evaluated for morpho-agronomic traits at two locations in Vietnam.

HD Upadhyaya, CLL Gowda, Vincent Vadez and Scientists from NARS

Milestone: Passport, characterization and evaluation data of germplasm documented (2006)

The existing Genebank Information Management System (GIMS) has been working well for safe and efficient handling and reporting of the data. The germplasm characterization databases of different crops updated for 16,224 accessions and documented data on different traits involving germplasm sets. These characterization data include – sorghum (931 accessions for 2004 rainy season and 417 accessions for 2004–05 postrainy season); (foxtail millet (769), barnyard millet (384), proso millet (118); chickpea (978 accessions for two traits); pigeonpea (38 traits on 6668 accessions); and groundnut (41 traits on 5959 accessions). Core collection of 622 finger millet germplasm was characterized and data was tabulated.

HD Upadhyaya

Milestone: Mini-core collection of chickpea germplasm characterized for resistance to *Ascochyta* blight (AB), *Botrytis* gray mold (BGM), wilt, dry root rot and collar rot (2006)

From the chickpea mini-core evaluations at ICRISAT-Patancheru, several promising sources of multiple resistance to biotic and abiotic stresses were identified. These include - ICC 1915 [drought + salinity + *Ascochyta* blight (AB)]; ICC 6306 (drought + AB); ICC 13816 [drought + wilt + *Botrytis* gray mold (BGM)]; ICC 15264 (drought + BGM); ICC 3512 (drought + wilt); ICC 2277 [dry root rot (DRR) + salinity)]; ICC 12328 (DRR + salinity + drought); ICCs 7272, 8261 and 13523 (drought + salinity); ICCs 8261, 10885 and 15697 (drought + salinity + BGM) and ICCs 2969, 6874, 14402 and 15567 (wilt and *Helicoverpa* pod borer).

S Pande, HD Upadhyaya and PM Gaur

Evaluation of chickpea mini-core entries for resistance to fungal diseases: Based on epidemiological principles, we have re-standardized the resistance screening methods for AB, BGM, FW and DRR. These refined techniques were used for resistance evaluation of chickpea mini-core. A total of 211 accessions of chickpea mini-core were evaluated in controlled environment for resistance to *Ascochyta* blight (AB), *Botrytis* gray mold (BGM), *Fusarium* wilt (FW) and dry root rot (DRR) diseases. Three accessions (ICC 1915, ICC 6306 and ICC 11284) were found moderately resistant (disease score 3.1 to 5 on a 1–9 rating scale, where 1 = no symptom; 9 = >75% kill) to AB under controlled environment. Fifty-five accessions were identified as moderately resistant (3.1 to 5.0 rating on 1–9 rating scale) to BGM. High levels of resistance to FW were found in several mini-core accessions under field screening. Twenty-one accessions were asymptomatic and 25 accessions had <10% mortality. Of the 211 mini-core accessions, 6 were moderately resistant (3.1 to 5 rating on a 1–9 rating scale) to dry root rot infection in blotter paper technique under laboratory conditions. Eleven accessions (ICCs 2990, 4533, 6279, 7554, 7819, 9848, 12028, 12155, 13219, 13559 and13816) were resistant to BGM and wilt; and ICC 13441 was resistant to DRR and wilt; ICC 11284 was resistant to AB and BGM; and ICC 11764 and 12328 were resistant to BGM and DRR.

S Pande, GK Kishore, HD Upadhyaya and PM Gaur

Activity 1.1.3: Assure risk-free export and import of germplasm and breeding materials

Milestone: Requested Germplasm exported for utilization and new Germplasm imported for conservation after seed health evaluation and clearance through PQ/NBPGR (Annual)

Seed health testing of breeding material and germplasm accessions for export/import: A total of 4871 seed samples of ICRISAT mandate crops comprising of breeding lines and germplasm accessions were exported to 45 countries (109 consignments). Of these, 153 samples (3%) were rejected mainly due to poor germination and association of seed-borne fungi of quarantine significance. A bulk consignment of 300 kg groundnut (cv. Asha - ICGV 86564) was exported to the Philippines. This consignment was cleared by the National Plant Protection and Training Institute, Hyderabad. A number of seed samples, including 30 of finger millet for export to Kenya that were found infected by species of Bipolaris, Curvularia and Phoma, were salvaged by treatments with Benomyl, thiram, or their combinations.

A total of 2937 seed samples (sorghum – 500, pearl millet – 450, chickpea – 1253 and groundnut – 732) were imported from 8 countries (Australia, Brazil, Israel, Japan, Malawi, Niger, USA and Vietnam). Also a special permission was obtained from Directorate of Plant Protection, Quarantine and Storage, Faridabad, India to import 2648 dried and grounded samples of rice straw (1500 samples) and pigeonpea (1148) from Philippines, Nicaragua and China for the Patancheru-based ILRI program. The National Bureau of Plant Genetic Resources (NBPGR), Hyderabad released 328 germplasm samples of sorghum (210), chickpea (103), and groundnut (15). Of the above, nine accessions of chickpea that were infected with bacteria were grown in the greenhouse. The remaining 94 accessions of chickpea were released to the consignee. Ten of the 15 accessions of groundnut did not germinate hence only five were grown in the greenhouse.

Grow-out test for imports: Chickpea germplasm (129 accessions) from Australia that were found moderately resistant to *Ascochyta* blight and *Botrytis* grey mold under growth chamber conditions in 2004, were grown during the rainy season 2005 for multiplication in the post-entry quarantine isolation area (PEQIA). Some of these accessions showed symptoms of *Fusarium* wilt, *cucumber mosaic virus* and *alfalfa mosaic virus*. These infected samples were destroyed by incineration. Only 121 disease-free accessions were harvested and released to the scientist concerned. Twenty groundnut accessions (14 cultivated and 6 wild *Arachis*) were grown in the greenhouse till harvest to observe bacterial wilt infection (*Ralstonia solanacearum*). No apparent symptoms were observed on the plants. The harvested seed was further tested for the presence of the bacterium by plating

the seed on tetrazolium chloride agar medium. All samples were found free from the bacterium and thus the samples were released to the concerned scientist.

Seventy-five chickpea accessions (24 from Syria and 51 from USA) were grown in the greenhouse for observation of bacterial infection (of unknown etiology). Seeds harvested from these accessions were tested for bacterial infection in nutrient agar medium. There were no bacterial symptoms observed either in the greenhouse or in the plated seed, and thus the samples were released.

RP Thakur

Better understanding and use of agro-biodiversity through the application of genomics and bioinformatics: Plant breeding efforts aimed towards increasing food security require genetic resources as a critical component - both for short-term gains and long-term increase in productivity. The International Agricultural Research Centers have responded to the threat of genetic erosion in economically useful plant species by developing a global network of genebanks for *ex situ* conservation of genetic diversity. Chapter 14G of Agenda 21 of the United Nations Conference on Environment and Development (UNCED) and the Global Plan of Action for the Conservation and Sustainable Utilization of Plant Genetic Resources for Food and Agriculture (GPA) endorsed by the Conference of the Parties to the Convention on Biological Diversity (CBD) underscore the importance of and the responsibilities on the large *ex situ* collections held by the CGIAR Centers, including ICRISAT.

ICRISAT assembled a large collection of germplasm of its mandate crops from threatened areas of biodiversity. The assembled germplasm needs to be maintained using appropriate procedures and through establishment of safe, efficient and cost-effective management systems. While ICRISAT collections represent 70-80% of available diversity in these crops, there is continuing need to rescue germplasm, especially wild and weedy relatives from endangered areas. NARS in developing countries also require assistance in collecting and conserving biodiversity. However, conservation is not the end by itself, but a means of making available diversity for use by the present and future generations. This requires systematic characterization and evaluation of germplasm for special traits. Traditionally, most characterization and evaluation is based on morph agronomic characters that could be easily detected and measured. However, there are many characters that are difficult to identify and are controlled by a number of genes interacting in complex ways. These include yield, time to flowering, resistance to important insect pests, diseases and abiotic stresses and quality parameters. The availability of modern biotechnological tools, particularly molecular markers, provides an opportunity to characterize germplasm for such complex traits.

Large collections of germplasm, which are difficult to handle, can be made more accessible through development of core collections, which are only 10% of entire collection but represent species diversity. ICRISAT scientists have developed core collections of all five mandate crops and finger millet. In the crops where the number of accessions is very large and core subset is also large, ICRISAT scientists have developed a strategy to select a mini-core collection (Upadhyaya and Ortiz 2001). The mini-core collection is 10% of the core collection (i.e., 1% of the entire collection) and contains almost full diversity of the entire collection. We have developed mini-core collections of chickpea (211 accessions, Upadhyaya and Ortiz 2001), groundnut (184 accessions, Upadhyaya et al. 2002) and pigeonpea (Upadhyaya et al 2006). The challenge is now to evaluate the core and mini core collections for useful traits and identify germplasm accessions as diverse parents for utilization in the breeding programs.

Wild and weedy relatives play an important role in sustaining agricultural productivity. In spite of their potential in crop breeding, very few attempts were made to utilize them for improvement of ICRISAT mandate crops, except groundnut. However, developments in biotechnology should provide new opportunities to make greater use of wild species in genetic enhancement.

This project has a global responsibility for effective protection as well as utilization of biodiversity of ICRISAT mandate crops. These primary goals will be achieved with efficient management, conservation, and the enhanced utilization of genetic resources.

Highlights of 2005

Chickpea composite collection phenotyped and genotyped

- Under the Generation Challenge Program (GCP), the chickpea composite collection (3000 accessions) was phenotyped for grain yield and related traits in an augmented design with five control cultivars during 2004-05 postrainy season. ICCs 8318, 17256, 8324, 12197, 812, and IG 70779 were the best high yielding accessions (2.74-3.35 t ha^{-1}). ICCs 12034, 13821, 16641, 17258, 5810, and ICCVs 96329 and 96030 were the earliest flowering accessions (33-36 days). ICCs 12034, 7346, and 14205 among kabuli types and ICCs 14648, 4871, and 7672 among the desi types were the largest-seeded accessions.

- The composite collection was genotyped using 35 SSRs at ICRISAT and 15 SSRs at ICARDA. Data from ICARDA is pending. The 35 SSR loci produced 1160 alleles, ranging between 15 and 58 alleles with an average of 34 alleles per SSR locus. The polymorphic information content (PIC) for the SSR loci varied from 0.47 to 0.96, with a mean of 0.84. Analyses are in progress to determine the population structure and identify the most diverse accessions for developing reference collection of 300 accessions (10% of the composite collection) for association mapping, functional genomics, gene tagging and genetic enhancement in chickpea.

Composite collections of groundnut, pigeonpea and finger millet developed

- Under the GCP, the composite collections of groundnut (850 accessions from ICRISAT and 150 from EMBRAPA), pigeonpea and finger millet (1000 accessions each) were constituted. The groundnut collection included accessions resistant to *Aspergillus flavus* seed colonization (16), various diseases (108), various insects (23) and accessions of - mini core collection (184), mini core comparators (184), mini core for Asia region (50), Asia core (60), elite/released cultivars (36), and drought tolerant (18), fresh seed dormancy (6), high and low biological nitrogen fixation (9) high shelling percentage (10), high nutritional traits (10) interspecific derivates (5), with desired agronomic traits (60) accessions genotyped earlier (18), and wilds (52) from ICRISAT, and wilds (62) and cultivated (88) from EMBRAPA.

- The pigeonpea composite collection included the mini-core collection (146), mini-core comparator (146), from core collection (236), superior morpho-agronomic traits (301), resistant to biotic stresses (74), resistant to abiotic stresses (14), elite/released cultivars (20), and 63 accessions of 7 wild species.

- The finger millet composite collection included core collection (622), plant growth aspect (114), core collection of Indian Coordinated Small Millet Improvement Project (50), desired agronomic traits (117), grain nutritional traits (12) and resistant to diseases (85). During 2006 this composite collection will be genotyped using 20 SSR markers, and the information will be used to determine population structure and identifying a reference collection of 300 accessions.

Composite collections of groundnut, pigeonpea and finger millet phenotyped

- A set of 850 groundnut accessions from ICRISAT (part of groundnut composite collection) was phenotyped for pod yield and related traits in an augmented design with four control cultivars, during 2005 rainy season. ICGs 6201, 6407, 6703, 10566 and 15042 were early flowering (19 days) in comparison to control cultivar ICGS 44 (22 days). ICGs 3027, 8352, 8285, 13920, and 13916 produced high pod yield (4.40 – 4.82 t ha^{-1}) in comparison to control cultivar ICGS 76 (4.38 t ha^{-1}) and ICGs 44 (3.88 t ha^{-1}). ICGs 5016, 4304, 8305, 8352, and 12059 were large-seeded (87 – 89 g 100 seed weight) accessions

- The pigeonpea composite collection was phenotyped in an augmented design with four control cultivars, for yield potential and related traits during 2005 rainy season. Data recording is in progress. Similarly, the finger millet composite collection was phenotyped in an augmented design with four repeated control cultivars for traits related to grain yield during 2005 rainy season. The preliminary investigation revealed that 65 finger millet accessions were promising for seed yield than the control cultivars. The early flowering accessions were IEs 4442, 4702, 6013, 588 and 4759 (42-51 days).

Composite collections of groundnut and pigeonpea genotyped

- The 850 accessions of the groundnut composite collection (ICRISAT contribution) were planted and leaf material was used to extract DNA for genotyping. Twenty SSR primer pairs were selected at ICRISAT for pre-screening groundnut mini-core accessions to identify 10 polymorphic primers. At EMBRAPA, ten other polymorphic primers have been identified and these selected 20 SSR primers are being used to fingerprint the entire composite collection using the ABI 3700. This dataset will be used for further analysis to assess

genetic diversity and then a reference collection of most diverse lines will be established that can be used for further research.

- The composite collection of pigeonpea (1000 accessions) was planted in the field and twelve plants per accession were selected for DNA extraction. Leaf samples for DNA extraction have been collected. DNA from these selected 12 plants per accession are pooled together mainly to capture within accession variation. Thirty SSR primer pairs were initially selected to pre-screen sixteen most diverse mini-core accessions to identify 20 polymorphic primer pairs, which will then be used to fingerprint the entire composite collection.

Core collections of pearl millet, pigeonpea, finger millet and foxtail millet phenotyped

- The core collections (10% of entire collection) of pearl millet (504 accessions), pigeonpea (1290), finger millet (622) and foxtail millet (155) were phenotyped during 2005 rainy season in augmented design with repeated control cultivars. The pearl millet results revealed that IP 9496 flowered in less than 45 days and IPs 10423 11937, 11947 and 17435 flowered very late (>125 days). IP 15220 and IP 10401 grew to a height of less than 40 cm. IP 15304 and IP 15257 produced 6 and 8 productive tillers. IPs 5416, 12310 and 5447 produced panicles longer than 65 cm and more than 25 mm thickness. Data processing is in progress. In pigeonpea, we selected 19 accessions in extra-early maturity group (68 days to 50% flowering, harvesting index 19.0%, shelling turnover 58%, and mean seed yield 580 kg $^{ha-1}$) in comparison to control ICPL 87 (DF 73, HI 17.1%, shelling 51.7%, and seed yield 219 kg $^{ha-1}$). Similarly, 5 early maturing, 6 medium maturing, and 29 late maturing high yielding accessions combined with other traits of economic importance were identified.

- In finger millet, IEs 2288, 3280, 3952, 5066 and 5179 (2.04-2.15 t $^{ha-1}$) were identified as high yielding accessions and IEs 501, 2322, 2957, 4759, and 6013 as early flowering accessions (49-52 days to 50% flowering). Similarly, in foxtail millet core collection, ISe 1254, 1227, 1234, 1286 and 1161 were early flowering (25-33 days) accessions. The preliminary investigation revealed that 16 germplasm accessions were promising for seed yield.

Evaluation of pigeonpea mini-core for pigeonpea sterility mosaic virus resistance

- Pigeonpea mini-core collection (146 accessions) was evaluated against two isolates of pigeonpea sterility mosaic virus (PPSMV), the B-isolate at Bangalore and P-isolate at Patancheru, India. Plants were inoculated with respective PPSMV isolates at 2-leaf stage, monitored for symptom type and percent incidence at 2 weekly intervals scoring was based on visual symptoms and plants were also tested for virus by ELISA using PPSMV-P polyclonal antibodies. ICP 8863 was used as susceptible control.

- On screening against P-isolate, 3 accessions (ICPs 7869, 14120 and 14155) showed no infection; 8 accessions showed 1-10% infection; and 11 accessions showed 11-30% infection. The rest of the accessions showed >30% infection. Five accessions (ICPs 12123, 10654, 11015, 11059 and 3046) although showed >80% infection, the genotypes expressed chlorotic ring spot symptoms, and no sterility observed in these genotypes and these can be regarded as tolerant to SMD. On screening against B-isolate, four accessions (ICPs 6123, 15185, 14569 and 14976) had <30% infection. Symptoms appeared very late. It appears that resistant sources to B-type of isolates are scarce.

Trait-specific mining of novel alleles and genes in pearl millet

- During 2005, in collaboration with researchers at the International Centre for Genetic Engineering and Biotechnology (ICGEB) and the Central University of Hyderabad, an attempt was made to generate gene-based markers associated with a pearl millet drought tolerance QTL, mapping in linkage group 2, which was mapped from donor parent, PRLT 2/89-33. This was done by isolating RNA samples from flag leaves of pairs of hybrids, near-isogenic for the drought tolerance QTL allele(s), i.e., 843A × H 77/833-2 and 843A × ICMR 01029 that had received either a fully-irrigated control treatment or a drought stress treatment initiated at panicle emergence in a greenhouse dry-down experiment. Using RNA samples collected from stressed plants of the two hybrids, a subtractive cDNA library was developed. Recombinant clones were collected and EST sequences corresponding to approximately 300 unigenes obtained. Initial annotation of the EST sequences suggested some chloroplast genome contamination of the cDNA library. So far, none of the EST sequences obtained appear to contain SSRs that might exhibit polymorphism between the near-isogenic pollinator of the two hybrids. However, such polymorphic EST-SSRs (and other polymorphic EST-based markers) could be used for exploratory allele mining in the vicinity of the pearl millet drought tolerance QTL located on linkage group 2.

Assessment of genotypic diversity in a set of agronomically superior insect resistant sorghum germplasm

- During 2005, genetic diversity was re-assessed for the set of 91 elite sorghum germplasm accessions using SSR markers. The set included 12 shoot fly and 15 stem borer resistant accessions, 9 accessions resistant to both shoot fly and stem borer, 17 midge resistant accessions, and 38 agronomically elite recurrent parents for which ICRISAT previously initiated a large-scale marker-assisted backcross program for the stay-green components of terminal drought tolerance. Twenty SSR markers were used to separate the PCR products using capillary electrophoresis that generated a total of 118 alleles with an average of 5.1 alleles per SSR locus. A high level of polymorphism was detected by 13 out of 20 (65%) SSR markers. An UPGMA (un-weighted paired group method with arithmetic averages) dendrogram was then constructed using Jaccard's similarity coefficient between each pair of accessions (which ranged from 0.28 to 1.00). The dendogram showed clustering of accessions in respect to geographical origin, race and specific traits such as resistance to specific insect pests. Based on clustering pattern, it was clear that the accessions under study are not only diverse for midge, shoot fly and stem borer resistance but also for agronomically superior recurrent parents that grouped in different clusters. However, some of the accessions with resistance to midge, shoot fly and stem borer clustered separately, suggesting that these lines might contain unexploited genetic variation for insect resistance that could be further exploited in breeding programs. This information will be useful for identifying elite recurrent parents for marker-assisted backcrossing programs to introgress insect resistance QTLs from the currently available mapping populations. Further, newly identified pairs of agronomically elite and genetically diverse insect resistant breeding lines could be used for developing new mapping populations to detect additional insect resistance QTLs.

Phenotypic and molecular characterization of sorghum germplasm in East and Central Africa

- Molecular markers provide information on the global genetic structure of the species – typically by using 20-50 SSR markers – and they provide the basis for association studies and determine entry points into vast germplasm collections. Once validated, information on marker-trait associations can be used for marker-assisted breeding. Through this project, standardized documentation, phenotypic and molecular characterization of sorghum accessions held as breeding material, international nurseries or as conserved germplasm in national gene banks of 8 ECA NARS is being analyzed. A regional composite set of 200 accessions per country is being analyzed for 25 markers and the additional diversity will be readily placed in relation to the components of the global sorghum germplasm structure. Capacity in high-throughput capillary based genotyping and low-cost PAGE systems is being built.

Phenotypic and genotypic diversity assessment of nutritional quality in sorghum and pearl millet germplasm

- The phenotypic diversity for grain micronutrient density in sorghum was assessed. The genetic variability for grain densities of Fe, Zn and phytates and high broad-sense heritabilities was observed. However, there is limited variability for beta-carotene density in sorghum, and all materials with detectable levels of this micronutrient in their grain have yellow endosperm. Grain densities of Fe and Zn were positively correlated, though each one of them had negative correlation with yield.

- Similarly, phenotypic diversity assessment for grain micronutrient density in pearl millet was assessed. Although there are significant genotype × environment interaction for pearl millet grain densities of Fe and Zn, there were good correlations between two seasons [Fe (r=0.66**) & Zn (r=0.69**)] and genotype rankings for Fe and Zn density were fairly consistent across environments and laboratories. There was wide variability for grain Fe and Zn density and like sorghum, in pearl millet too grain densities of Fe and Zn were positively correlated. A rapid, low-cost, staining protocol of Fe grain density has been optimized and can now be used for inexpensive high-throughput screening of core collections of sorghum and pearl millet.

Information management systems

- The workflow management system (LIMS/Laboratory Information Management System) came into operation in 2005 to help with the capture of genotyping data from the Applied Genomics Laboratory (AGL). This three-tier application (data layer, middleware and presentation layer) is modular in its construction. This allows further enhancements to be made if required through the use, re-use or replacement of existing modules. LIMS also incorporates automated allele binning through a Java executable of the algorithm of Cordon and Idury (1997). Testing for user acceptability of this application is currently in progress. Genotyping information from the LIMS system flows into a larger database ICRIS (Integrated ICRISAT Crop Resources Information System). The ICRIS is envisaged to be an integrated database and currently consists of three databases for genetic resource data, genotyping data and phenotypic/trait information. The database can be accessed through the user interface, tabular and graphical reports can be generated from existing data.

Genetic Diversity, Genomic Resources and Bioinformatics

Output 1.3.2: Molecular characterization and validation of mini-core germplasm collections
Milestone: Phenotypic and genotypic information on chickpea and groundnut composite collection

***Chickpea Composite Collection Genotyped*:** A composite collection of 3000 accessions representing wide spectrum of chickpea genetic diversity (biologically - 80% landraces, 11% advanced lines/cultivars, and 1% wild species and geographically – 30% from South and South-East Asia, 25% from West Asia, 22% from Mediterranean, and 5% each from Africa and America's) was developed (Global Theme – Biotechnology Archival Report 2004). The composite collection has been genotyped using high throughput assay (ABI3700) and 35 ploymorphic SSRs at ICRISAT and 15 ploymorphic SSRs at ICARDA. Data from ICARDA is awaited. The genotypic data will be used to develop a reference collection consisting of 300 accessions (10% of the composite collection) for association mapping, functional genomics, gene tagging and genetic enhancement in chickpea.

A C (language) program was developed for allele binning based on the Idury and Cardon (1977) algorithm to provide a statistical measure of the fit of the determined "raw" alleles to an expected size based on SSR repeat unit. Using this test, we found that marker TAA58, TA21, and TR28 deviated significantly (as determined by marker quality index and allelic drift) to the expected fit, and will be dropped from the analysis. The entire dataset was formatted into a binary matrix (structure format) as well, a format necessary to operate in DARwin 5 program.

The 35 SSR loci's produced 1182 alleles, ranging between 15 and 58 alleles with an average of 33.77 alleles per SSR locus (see figure 3.1 for allelic distribution of accessions over 35 SSR loci). The polymorphic information content (PIC) for the SSR loci varied from 0.474 to 0.956, with a mean of 0.839. From the allelic distribution among accessions, several SSR loci produced unique alleles in a number of accessions. For example, SSR marker NCPGR19 in ICC 9330 (456bp) and ICC 9676 (464bp); NCPGR6 in ICC 4849 (361bp); TA200 in ICC 13912 (355bp), IG 73087 (358bp), and ICC 4853 (367bp); TA206 in ICC16915 (425bp), ICC1124 (431bp), and ICC7554 (437bp); TA21 in ICC 9402 (419bp); TA28 in ICC 12283 (438bp); TA71 in ICC 95 (139bp); TaaSH in ICC 4853 (496bp); and TR43 in ICC 10466 (417bp), ICC 4075 (432bp), ICC 228 (468bp), and ICC 4853 (474bp) produced alleles that were present only on these but absent in the remaining accessions. Further analysis is in progress to determine the population structure and identify the most diverse accessions for developing the reference collection of 300 accessions.

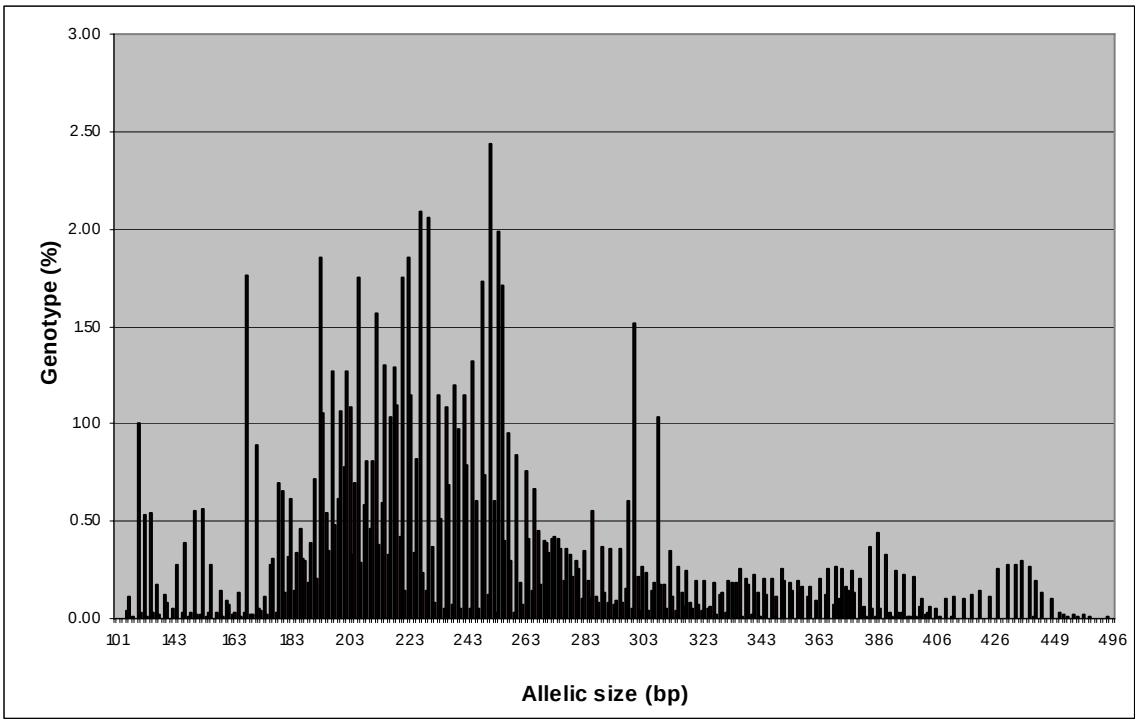

Figure 3.1. Histogram of allele frequencies for the 1182 alleles detected using 35 SSR loci in 3013 accessions

Chickpea Composite Collection Phenotyped: Chickpea composite collection consisting 3000 accessions including core/mini core collection, was phenotyped for grain yield and related traits in an augmented design with five control cultivars during 2004-05 postrainy season. ICCs 8318, 17256, 8324, 12197, 812, and IG 70779 (2.74-3.35 t ha^{-1}) were the top high yielding accessions. ICCs 12034, 13821, 16641, 17258, 5810, and ICCVs 96329 and 96030 (33-36 days to 50% flowering, 1.18-2.02 t ha^{-1}) were the earliest flowering accessions. ICCs 12034, 7346, and 14205 (45.0-45.7 g 100^{-1}seed weight and 1.18-2.02 t ha^{-1} grain yield) among kabuli types and 14648, 4871, and 7672 (29.2-35.4 g 100^{-1}seed weight and 1.25-2.26 t ha^{-1} grain yield) among the desi types were identified as the large seeded accessions.

HD Upadhyaya SL Dwivedi, PM Gaur,D Hoisington, RK Varshney, S Chandra and CLL Gowda

Groundnut Composite Collection Developed: Groundnut composite collection consisting of 850 accessions from ICRISAT and 150 from EMBRAPA representing entire collection ecologically, taxonomically, and phenotypically was developed. The composite collections includes accessions resistance to - *A. flavus* seed colonization (16), bacterial wilt (5), bud necrosis (7), peanut mottle virus (2), rosette (12), early leaf spot (7), late leaf spot (14), rust (15), stem and pod rot (9), multiple resistant (37), aphid (1), jassid (8), leaf miner (5), thrips (4), termite (5), and accessions of - mini core collection (184), mini core comparators (184), mini core for Asia region (50), Asia core (60), elite/released cultivars (36), and drought tolerant (18), fresh seed dormancy (6), high and low biological nitrogen fixation (9) high shelling percentage (10), high oil content (5), high protein content (5), interspecific derivates (5), large seeded (10), morphological variants (26), early maturity (25), accessions genotyped earlier (18), and 52 accessions of 14 wild *Arachis* species from ICRISAT, and 62 wild accessions and 88 cultivated from Empresa Brasileira de Pesquisa Agropecuária (Brazilian Agricultural Research Corporation EMBRAPA).

Groundnut Composite Collection Genotyped: 850 accessions of the composite collection were planted and leaf material was used to extract DNA for further genotyping. Twenty SSR primer pairs were pre-screened on mini-core accessions to identify 10 polymorphic primers. At EMBRAPA, ten other polymorphic primers have been identified and these selected 20 SSR primers are being used to fingerprint the entire composite collection through capillary electrophoresis using ABI 3700. GeneScan and Genotyper softwares are used to analyze the electrophoresis data to get information on alleles. This dataset will be used for further analysis to assess genetic diversity and then a reference collection (300 accessions) of most diverse lines will be established that can be used for further genotyping and phenotying studies and in the improvement programs.

Groundnut Composite Collection Phenotyped: Groundnut composite collection consisting of 850 accessions from ICRISAT was phenotyped for pod yield and related traits in an augmented design with four control cultivars, during 2005 rainy season. ICGs 6201, 6407, 6703, 10566, and 15042 were early flowering (19 days) in comparison to control cultivar ICGS 44 (22 days). ICGs 3027, 8352, 8285, 13920, and 13916 produced high pod yield (4.40 – 4.82 t ha^{-1}) in comparison to control cultivar ICGS 76 (4.38 t ha^{-1}) and ICGS 44 (3.88 t ha^{-1}). ICGs 5195 and 11427 were identified with high shelling turn over (82-83%) in comparison to ICGS 44 and ICGS 76 with 80%. ICGs 5016, 4304, 8305, 8352, and 12059 were large-seeded (87 – 89 g 100 seed weight) accessions

HD Upadhyaya, R Bhattacharjee, D Hoisington and RK Varshney

Activity 1.3.2.2: Phenotypic and genotypic diversity assessment of the sorghum, pearl millet, and finger millet core collections

Team: HD Upadhyaya, S Senthilvel, S Chandra, CT Hash, CLL Gowda, D Hoisington RK Varshney, B Jayashree and SMH Rizvi

Milestone: Phenotypic and genotypic information on sorghum and finger millet composite collection

***Sorghum Composite Collection Genotyped*:** Leaf samples of 2300 sorghum accessions were collected for DNA extraction. Fingerprinting with 50 SSR markers is in progress.

Finger Millet Composite Collection Developed: Finger millet composite collection consisting of 1000 accessions representing entire collection ecologically, taxonomically, and phenotypically was developed with the support of Generation Challenge Program. The composite collection includes accessions – core collection (622), plant compactness and aspect (114), core collection of AICSMIP (50), grain size (5), disease resistant and good plant aspect (76), morphological variance (9), early flowering (9), nutritional quality (12), grain yield (19), grain yield and size (9), grain size (13), fodder yield (8), finger blast (2), neck blast (3), neck and finger blast (4), finger length (17), finger number 9), harvest index (5), number of productive tillers (7), and short plant

height (7). During 2006 this composite collection will be genotyped at 20 SSR loci and information will be used to determine population structure and identify a reference collection of 300 accessions.

***Finger Millet Composite Collection Phenotyped*:** Composite collection of finger millet consisting 1000 accessions including core collection was phenotyped in an augmented design with four repeated control cultivars for, traits related to grain yield during 2005 rainy season. Data processing is in progress. The preliminary investigation reveals that 65 germplasm accessions were promising for seed yield than the control cultivars. The early flowering accessions were IEs 4442, 4702, 6013, 588, and 4759 (42-51 days).

HD Upadhyaya, S Senthilvel, S Chandra , CT Hash, CLL Gowda , D Hoisington and RK Varshney

Milestone: Phenotypic information on pearl millet, finger millet, and foxtail millet core/mini core collection

Pearl Millet Core Collection Phenotyped: Pearl millet core collection consisting of 504 accessions was phenotyped for grain yield and related 15 characters during 2005 rainy season. The preliminary results reveals that IP 9496 flowered in less than 45 days and IPs 10423 11937, 11947 and 17435 flowered very late (>125 days). IP 15220 and IP 10401 grew to a height of less than 40 cm. IP 15304 and IP 15257 produced 6 and 8 productive tillers. IP 5416, IP 12310 and IP 5447 produced panicles more than 65 cm long and more than 25 mm thickness. Four accessions, IP 6510, IP 6530, IP 6554 and IP 11947 scored 8 for green fodder yield potential. Data processing is in progress. Same set was planted during 2005-06 postrainy season to validate the observations.

Finger Millet Core Collection Phenotyped: Finger millet core collection consisting of 622 accessions was phenotyped in an augumented design experiment with four control cultivars, for grain yield potential and quality characters during 2004 rainy season. IEs 2288, 3280, 3952, 5066, and 5179 (2.04-2.15 t ha^{-1}) were the high yielding accessions and IEs 501, 2322, 2957, 4759, and 6013 (49-52 days flowering and 1.29-1.51 t ha^{-1} seed yield) were early flowering accessions.

Foxtail Millet Core Collection Developed and Phenotyped: Foxtail millet core collection consisting of 155 accessions was developed and phenotyped in a replicated Alpha designed trial, for grain yield and related traits during 2005 rainy season. Data processing is in progress. ISe 1254, 1227, 1234, 1286, and 1161 were early flowering (25-33 days) accessions. The preliminary investigation reveals that 16 germplasm accessions were promising for seed yield.

HD Upadhyaya ,CLL Gowda, CT Hash and D Hoisington

Milestone: Technology for trait-specific mining of novel alleles and genes in sorghum, pearl millet, chickpea and groundnut germplasm collections developed and utilized

During 2005, in collaboration with researchers at the International Centre for Genetic Engineering and Biotechnology (ICGEB) and the Central University of Hyderabad, an attempt was made to generate gene-based markers associated with a pearl millet drought tolerance QTL, mapping in linkage group 2, and mapped from donor PRLT 2/89-33. This was done by isolating RNA samples from flag leaves of pairs of hybrids near-isogenic for the drought tolerance QTL allele(s) (i.e., 843A × H 77/833-2 and 843A ×ICMR 01029), which had received either a fully-irrigated control treatment or a drought stress treatment initiated at panicle emergence in a greenhouse dry-down experiment. Using RNA samples collected from stressed plants of the two hybrids, a subtractive cDNA library was developed. Recombinant clones were collected and EST sequences corresponding to approximately 300 uni-genes obtained. Initial annotation of the EST sequences suggests some chloroplast genome contamination of the cDNA library. So far, none of the EST sequences obtained appear to contain SSRs that might exhibit polymorphism between the near-isogenic pollinator of the two hybrids. Such polymorphic EST-SSRs (and other polymorphic EST-based markers) could be used for exploratory allele mining in the vicinity of the pearl millet drought tolerance QTL on pearl millet linkage group 2.

CT Hash, ,SMH Rizvi,S Senthilvel and B Jayashree

Activity 1.3.2.3: Phenotypic and genotypic diversity assessment of the pigeonpea core collection
Team: HD Upadhyaya HD, R Bhattacharjee, D Hoisington , RK Varshney, KB Saxena and S Chandra

Milestone: Phenotypic and genotypic information on pigeonpea composite and core collections

Pigeonpea Core Collection Phenotyped: Pigeonpea core collection was phenotyped for grain yield and related traits in an alpha designed experiment during 2004 rainy season. Selected 19 accessions in extra-early maturity group (mean days to 50% flowering, 68 days, harvesting index 19.0%, shelling turnover 58%, and mean seed yield 580 kg ha^{-1}) in comparison to control ICPL 87 (DF 73, HI 17.1%, shelling 51.7%, and seed yield 219 kg ha^{-1}). Similarly, 5 early maturing, 6 medium maturing, and 29 late maturing high yielding accessions combined with other traits of economic importance were identified.

Pigeonpea Composite Collection Developed: Pigeonpea composite collection consisting of 1000 accessions representing entire collection ecologically, taxonomically, and phenotypically was developed. The composite collection includes accessions – minicore collection (146), minicore comparator (146), from core collection (236), superior morpho-agronomic traits (301), resistant to biotic stresses (74), resistant to abiotic stresses (14), elite/released cultivars (20), and 63 accessions of 7 wild species.

Pigeonpea Composite Collection Phenotyped: Pigeonpea composite collection consisting of 1000 accessions including mini core collection was phenotyped in an augmented design with four control cultivars, for yield potential and related traits during 2005 rainy season. Data recording is in progress.

Pigeonpea Composite Collection Genotyped: The composite collection was planted in the field and twelve plants per accession were selected for DNA extraction. DNA from these selected 12 plants per accession were pooled together mainly to capture within accession variation. Thirty SSR primer pairs were initially selected to pre-screen sixteen most diverse mini-core accessions to identify 20 polymorphic primer pairs, which will then be used to fingerprint the entire composite collection. The genotyping will be carried out through capillary electrophoresis using ABI3700. Further data analysis will be done to determine the genetic diversity and also to develop a reference collection of 300 accessions.

HD Upadhyaya, R Bhattacharjee , D Hoisington , RK Varshney, KB Saxena and S Chandra

Output 1.3.3 Molecular characterization of trait-based germplasm

Activity 1.3.3.2: Phenotypic and genotypic diversity assessment of stem borer, shoot fly and *Striga* resistant sorghum germplasm
Team: HC Sharma, CT Hash, RT Folkertsma , S Chandra, HD Upadhyaya , D Kiambi , D Hoisington and Sante de Villiers

Milestone: Phenotypic and genotypic information on stem borer and shoot fly resistant sorghum germplasm

During 2005, the SSR-based genetic diversity analysis of a set of 91 elite sorghum germplasm accessions was revised during drafting of the PhD thesis of Mr. S.P. Mehtre from Marathwada Agricultural University. The set of lines included 12 shoot fly resistant and 15 stem borer resistant accessions, 9 accessions resistant to both shoot fly and stem borer, 17 midge resistant accessions, and 38 agronomically elite recurrent parents for which ICRISAT previously initiated a large-scale marker-assisted backcross program for the stay-green components of terminal drought tolerance. Based upon capillary electrophoresis separation of PCR products from 20 sorghum SSR primer pairs, a total of 118 alleles were detected in this set of sorghum lines, with an average of 5.1 alleles per SSR locus, and 13 out of the 20 (65%) SSR primer pairs were able to detect a high level of polymorphism. Jaccard's similarity coefficient (which ranged from 0.28 to 1.00) between each pair of accessions was used to construct a dendrogram to determine the relationships among accessions using the un-weighted paired group method with arithmetic averages (UPGMA). The dendogram showed clustering of the accessions by geographical origin, race and specific traits such as resistance to specific insect pests. This study confirmed that the accessions studied are genetically quite diverged with sorghum lines showing midge, shoot fly and stem borer resistance clustering in different groups. In addition, a cluster of agronomically superior recurrent parents was identified that is genetically quite divergent from each of these insect resistant clusters. However, some of the accessions with resistance to midge, shoot fly and stem borer clustered separately, suggesting that these lines might contain unexploited genetic variation for insect resistance that could be exploited in breeding program. This information will be useful for identifying elite recurrent parents for marker-assisted backcrossing programs

to introgress insect resistance QTLs from the currently available mapping populations. Further, newly identified pairs of agronomically elite and genetically diverse insect resistant breeding lines could be used for developing new mapping populations to detect additional insect resistance QTLs.

HC Sharma , CT Hash, RT Folkertsma ,S Chandra and HD Upadhyaya

Milestone: Phenotypic and molecular characterization of sorghum germplasm held by ECA NARS completed (2008)

Phenotypic and molecular characterization of Sorghum germplasm in East and Central Africa:
Phenotypic information of germplasm provides the entry point for plant breeding efforts, providing information on responses to biotic and abiotic stresses and farmer- and market-preferred characteristics. Molecular markers provide information on the global genetic structure of the species – typically by using twenty to fifty neutral markers – and they provide the basis for association studies and determine entry points into vast germplasm collections. Once validated, information on marker-trait associations can be used for marker-assisted breeding. Through this project, standardized documentation, phenotypic and molecular characterization of sorghum accessions held as breeding material, international nurseries or as conserved germplasm in national gene banks of 8 ECA NARS is being analyzed using 20-50 SSR markers. These are part of the GCP set of high quality microsatellite markers that is being used for the survey of a global composite set of sorghum germplasm. A regional composite set of 200 accessions per country is being analyzed with 25 markers and the additional diversity will be readily placed in relation to the components of the global sorghum germplasm structure. As part of the project, capacity in high-throughput capillary based genotyping and low-cost PAGE systems is being built in collaborating NARS through a PhD studentship and hands-on-training of 8 Visiting Scientists.

A project planning meeting has been held and consensus built on the database structure, standardized methodologies for experimental design, data collection and documentation. Training on data entry and retrieval using Microsoft Access has also been conducted. A computerized inventory of sorghum germplasm held in Uganda and Sudan has been initiated and the selection of 200 and 400 sorghum accessions made respectively. A PhD student from Sudan has been recruited and is currently being trained in basic molecular techniques while awaiting registration at the University of Free State, South Africa. Planning is underway for a second project meeting to review status and conduct a Sorghum phenotyping workshop, including hands-on-training on data entry into the previously developed Microsoft Access Database.

D Kiambi, D Hoisington and Sante de Villiers

Activity 1.3.3.3: Phenotypic and genotypic diversity assessment of sorghum germplasm varying in flowering time and stay-green/senescence at maturity

Milestone : Allele-mining to develop allele-specific markers for all major flowering genes in sorghum completed (2008)

Genetic materials appropriate for this activity have been included in the composite germplasm set of sorghum that is being genotyped with approximately 50 well-distributed SSR loci in the Generation Challenge Program. However, to achieve this milestone substantial special-project funding is required**.**

CT Hash, RT Folkertsma, V Vadez , FR Bidinger,D Hoisington, HFW Rattunde, F Sagnard , B Clerget, S Chandra and Upadhyaya HD

Activity 1.3.3.4: Phenotypic and genotypic diversity assessment of nutritional quality in sorghum and pearl millet germplasm
Team: CT Hash , D Hoisington , BVS Reddy, S Ramesh , KN Rai, VN Kulkarni , EW Rattunde, V Vadez and Upadhyaya HD

Findings from the first two years of phenotypic diversity assessment for grain micronutrient density in sorghum in HarvestPlus, the Biofortification Challenge Program, can be summarized as follows:

- Genetic variability exists for grain densities of Fe, Zn and phytates.
- Broad-sense heritabilities are high for sorghum grain densities of Fe, Zn and phytates.
- There is limited variability for beta-carotene grain density in sorghum, and all materials with detectable levels of this micronutrient in their grain have yellow endosperm. Because of this, we recommend halting conventional phenotypic diversity assessment and marker-based genotypic diversity assessment for this trait in sorghum, allowing the limited available resources to be focused on improvement of sorghum grain densities of Fe and Zn.

- The correlation of sorghum grain densities of Fe and Zn is significant and positive, which will facilitate simultaneous improvement of the grain densities of these two micronutrients.
- There are significant negative correlations of sorghum grain densities of Fe and Zn with grain yield, probably as a result of negative correlation of sorghum grain densities of these micronutrients with grain size.
- No significant interactions of genotype × managed soil fertility (NPK) level was observed for Fe, Zn and phytate grain densities in sorghum

Similarly, findings from the first two years of phenotypic diversity assessment for grain micronutrient density in pearl millet in HarvestPlus, the Biofortification Challenge Program, can be summarized as follows**:**

- Although there are significant genotype × environment interaction for pearl millet grain densities of Fe and Zn, there were good correlations between two seasons [Fe (r=0.66**) & Zn (r=0.69**)] and genotype rankings for Fe and Zn density were fairly consistent across environments and laboratories.
- There is wide variability for grain Fe and Zn density in pearl millet, and high grain Fe and Zn density is available in elite backgrounds. Almost all micronutrient-dense genotypes identified are derived from '*Iniari*' germplasm.
- As in sorghum, pearl millet grain densities of Fe and Zn are positively correlated, so simultaneous improvement of the grain densities of these two micronutrients should be possible.
- A rapid, low-cost, staining protocol of Fe grain density has been optimized and can now be used for inexpensive high-throughput screening of core collections of sorghum and pearl millet for additional Fe-dense accessions. In pearl millet the aim of such screening would be to identify 'non-*Iniari*' germplasm having high grain density of Fe.
- Thus, there are good prospects to increase grain density levels of both Fe and Zn in pearl millet.

Based on the results of the first two years of phenotypic assessment of grain micronutrient densities in these two crops, it appears there are opportunities to:

- use high-throughput method to screen core collection for additional Fe-dense accessions of sorghum and pearl millet, which are then reasonably likely to also have high grain densities of Zn;
- assess effectiveness of enhancing Fe and Zn density in released OPVs by recurrent selection;
- initiate inheritance studies of grain Fe and Zn density, which can include mapping population development in pearl millet (initially using bulk segregant analysis for grain Fe density)
- and initiate studies of nutritional availability of the Fe and Zn in grains of sorghum and pearl millet genotypes that have high grain densities of these micronutrients.

CT Hash, D Hoisington , BVS Reddy, S Ramesh, KN Rai,VN Kulkarn, EW Rattunde , V Vadez and HD Upadhyaya

Activity 1.3.3.8: Phenotypic diversity assessment of sterility mosaic disease resistance in wild and cultivated pigeonpea germplasm
Team: Lava Kumar, HD Upadhyaya and F Waliyar

Milestone: Genetically diverse sources of resistance to SMD in pigeonpea identified

Evaluation of pigeonpea mini-core for Pigeonpea sterility mosaic virus resistance: Pigeonpea mini-core collection, comprising 146 accessions was evaluated against two isolates of pigeonpea sterility mosaic virus (PPSMV), for B-isolate at Bangalore and P-isolate at Patancheru, India. Thirty seed of each accession was sown in plastic pots in three replications and maintained in greenhouse at Patancheru. In Bangalore sowings were done in the experimental station. Plants were inoculated with respective PPSMV isolates at 2-leaf stage, and they were monitored for symptom type and percent incidence at 2 weekly intervals and scoring was based on visual symptoms and plants were also tested for virus by ELISA using PPSMV-P polyclonal antibodies. ICP 8863 was used as susceptible control.

Of 146 accessions evaluated against P-isolate, 3 accessions (ICPs 7869, 14120 and 14155) showed no infection; 8 accessions (ICP 14368, 11910, 14229, 14569, 14147, 14545, 11833 and 14471) showed 1-10% infection; and 11 accessions (ICP 14701, 14722, 15049, 14444, 14801, 14638, 14976, 13304, 14294, 11015 and 4317) showed infection between 11-30%. Rest of the accessions showed >30% infection. Five accessions (ICPs 12123, 10654, 11015, 11059 and 3046) although showed >80% infection, the genotypes expressed chlorotic ringspot symptoms, and no sterility observed in these genotypes and they can be regarded as tolerant to SMD. This experiment is still on going and final data with complete agronomic features will be presented at later date.

Against B-isolate 143 accessions were tested (no germination in case of ICPs 12105, 10559 and 12596). Only 4 of 143 accessions had <30% infection against PPSMV-B isolate (ICPs 6123, 15185, 14569 and 14976) (Table 3.1). Symptoms appeared very late in all the four genotypes.

Evaluation of pigeonpea mini-core indicates narrow base of resistance to SMD in the pigeonpea germplasm. Resistant sources to B-type of isolates are much scarce. The resistant accessions provide entry point for further evaluation of genotypes for SMD resistance. All the promising lines identified in this activity will be further validated.

Lava Kumar, HD Upadhyaya and F Waliya

Table 3.1: Pigeonpea mini-core accessions resistant to PPSMV against P and/or B isolates

% SMD infection	PPSMV-P Isolate	PPSMV-B Isolate
0	7869, 14120, 14155	None
1-10	14368, 11910, 14229, **14569,** 14147, 14545, 11833, 14471	None
11-20	14444, 14801, 14638	6123, 15185
21-30	14701, 14722, 15049, **14976**, 13304, 14294, 11015, 4317	**14569, 14976**

Remaining accessions of pigeonpea mini-core showed >30% infection
Common sources for both isolates underlined and in bold

Output 1.3.4: Molecular characterization of gene flow

Activity 1.3.4.2: Sorghum and pearl millet and their wild relatives in Asia and Africa
Team: F Sagnard, HFW Rattunde, EW Rattunde, RT Folkertsma, CT Hash, BVS Reddy, D Hoisington D and HD Upadhyaya

Milestone: Sorghum crop-to-wild gene flow in Mali and Kenya

ICRISAT leads a project on the "Environmental Risk Assessment of Genetically Engineered Sorghums in Mali and Kenya". It aims at measuring the realized amount of crop-to-wild gene flow and analyzing the farmers' practices that may limit or favour the in situ genetic introgression between cultivars and wild Sorghum populations. This is a multidisciplinary project involving population geneticists, GIS/remote sensing specialists, molecular biologists, Sorghum breeders, and social scientists from ICRISAT in Mali and Kenya, from IER and the University of Bamako in Mali, from KARI in Kenya that started in march 2005.

We found numerous morphological evidence of crop/wild introgression occurring in Mali and Kenya. The crosses made on-station between several wild and cultivated Sorghums were all successful and produced normally fertile hybrids. However, it seems that the occurrence of wild Sorghums and the presence of weedy types in farmers' field fifer between regions. In the Soudano-guinean zone of South and South-West Mali, wild and weedy types are widespread in and around farmers' fields whereas weedy sorghum are rare in the Central and Northern part of Mali where they are not mentioned as an important threat for agriculture. The amount of genetic introgression may be variable due to the phenological overlap of wild and cultivated Sorghum flowering period. Our partial results show that wild Sorghum in North Mali are more genetically differentiated from cultivated Sorghum than those from the Mandé region. They also flower earlier than the landraces cultivated in the same villages

F Sagnard, HFW Rattunde, EW Rattunde, RT Folkertsma, CT Hash, BVS Reddy, D Hoisington and HD Upadhyaya

Output 1.3.5: Assessment of phenotypic and genotypic diversity in sorghum and pearl millet in Western and Central, and Southern and Eastern Africa

Activity 1.3.5.1: Temporal evolution of genetic diversity of Sorghum and Pearl Millet in Niger between 1976 and 2003.
Team: F Sagnard, HFW Rattunde, EW Rattunde, CT Hash, D Hoisington and HD Upadhyaya

Milestone: Agromorphological and genetic marker analyses of sorghum and pearl millet collection in 79 villages across Niger

ICRISAT participated in a collaborative project on Sorghum and Pearl Millet agrobiodiversity along with French Institute for Biodiversity (IFB) including IRD, CIRAD and INRA in 2004 and 2005 to compare the genetic diversity of Sorghum and Pearl Millet collections conducted at a 27 year interval, using both molecular (32 SSR) markers and agromorphological descriptors (Fig. 3.2) The results were expected to provide new insights on the impact of climatic and anthropogenic changes on the evolution of crop diversity in the Semi-Arid regions.

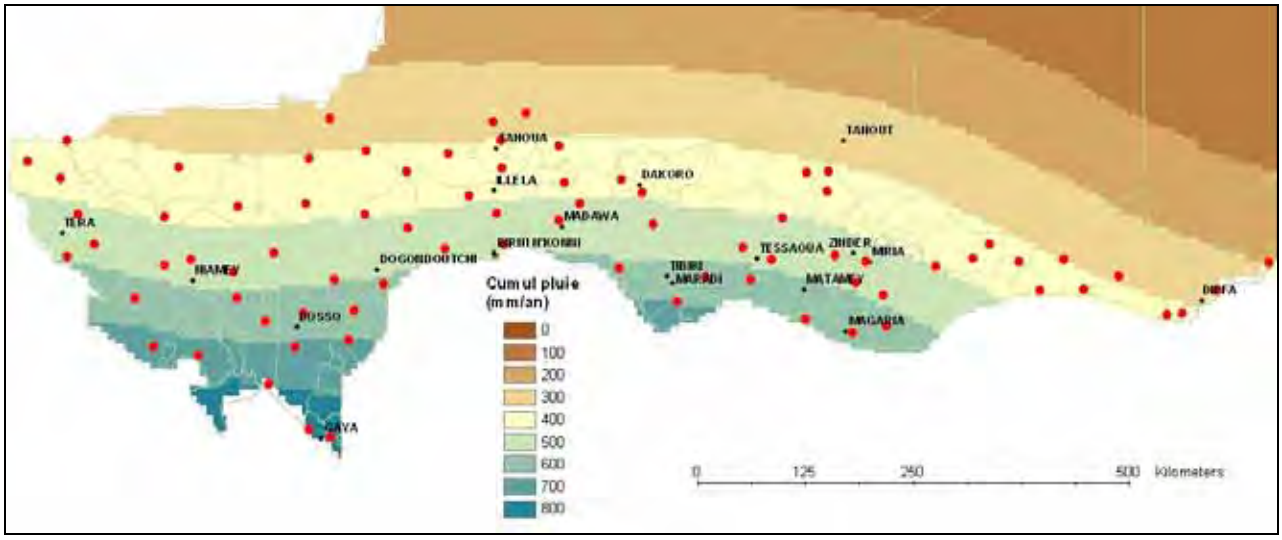

Figure 3.2 Locations of the 79 villages where Sorghum and Pearl Millet collections were undertaken in 1976 and 2003 (Bruno Gérard, ICRISAT Niamey).

In Niger, substantial diversity for agromorphological traits and SSR was observed in Sorghum. The genetic diversity is highly structured among races and among regions with the durra race predominant in Eastern Niger and the guinea race mainly located in the region where the annual rainfall exceeds 600 mm per year. We did not find any evidence of genetic erosion between 1976 and 2003 at the country level despite the occurrence of several drought events and the doubling of the population in Niger during the period of study. The low genetic differentiation (Fst = 0.003) of the two sorghum collections could indicate the strong resilience of agrobiodiversity in harsh environments and support the idea that *in situ* conservation by farmers is an efficient and complementary way to conserve the diversity of crop genetic resources. The data on genetic diversity observed at a more local scale will be analyzed in 2006.

F Sagnard, HFW Rattunde, EW Rattunde, CT Hash, D Hoisington and HD Upadhyaya

Activity 1.3.5.2: Structure of Sorghum diversity in Mali in relation to the environmental gradient and the farmers' practices.

Milestone: Assessment of inter- and intra-varietal structures of sorghum landrace diversity using agromorphological and microsatellite markers.

ICRISAT team in Mali participates in the project "Sorghum agrobiodiversity in Mali and Burkina Faso" funded by the French Fund for Global Environment (FFEM). This project promotes plant participatory and decentralized breeding using a large amount of local germplasm as a method to conciliate crop improvement and genetic resource conservation of Sorghum in its centre of diversity.

The Sorghum genetic diversity is not randomly distributed in Mali along a North-South gradient. Landraces are highly structured according to botanical races and phenology. Traits such as sensitivity to photoperiod and the tillering ability are widespread in the local Sorghums. These characters should be linked to adaptation to harsh environments of the Semi-Arid tropics. The lines produced by the IER breeding program conserve an important part of the local genetic diversity. The within seed lot genetic diversity of 10 Sorghum varieties (6 guinea

gambicum landraces, 3 guinea margaritiferum landraces and 1 commercial guinea gambicum variety) is more important in the guinea gambicum landraces than in the guinea margaritiferum landraces and the commercial variety (Table 3.2). An average of 1.25 to 3.36 alleles per locus were found. The heterozygote deficit is lower than values that have been previously published for the durra race sorghum except for the guinea margaritiferum landraces, which indicate a higher outcrossing rate for the guinea gambicum sorghums than generally admitted (up to 53 %).

All landraces are highly genetically differentiated (0,19 < Fst < 0,65), even among 2 landraces cultivated in the same field by the same farmer. This is much likely the consequence of repeated bottlenecks caused by sowing seeds collected on a limited number of individuals each year.

Table 3.2: Within seed lot genetic diversity of 10 Sorghum varieties from Mali assessed at 11 microsatellite markers

Variety ID	KAG-L	DIO-A	DOUA-B	KAG-B	POM-E	SIR-B	SOUG-A	DOUA-F	DOUA-D	SIR-E
Name	CSM63E	Kalo Sabani	Nio bléni	Doubirou	Hémé piri	Sambou	Magno gnoulê nyê	Kendé blé	Kendé diéma	Kendé
Type	Guinea gambicum Commercial	Guinea gambicum Landrace	Guinea gambicum Landrace	Guinea gambicum Landrace	Guinea gambicum Landrace	Guinea gambicum Landrace	Guinea gambicum Landrace	G. marga Landrace	G. marga Landrace	G. marga Landrace
He	0,078	0,359	0,292	0,314	0,302	0,324	0,324	0,325	0,025	0,078
A	1,636	3,455	3,273	2,727	2,909	2,545	2,364	2,636	1,273	1,909
A'	1,628	3,364	3,156	2,671	2,856	2,507	2,342	2,612	1,255	1,894
Hobs	0,051	0,248	0,082	0,136	0,069	0,076	0,129	0,038	0,004	0,008
Fis	0,346	0,309	0,720	0,567	0,798	0,766	0,602	0,894	0,852	0,898
S(eq.)	0,514	0,472	0,837	0,724	0,888	0,867	0,752	0,944	0,920	0,946

He: gene diversity, A: allelic richness, A': allelic richness corrected for the sampling size, Hobs: observed heterozygosity, Fis: Wright's fixation index, S (eq.): selfing rate at the equilibrium state under a mixed mating model assumption)

F Sagnard , HFW Rattunde, EW Rattunde, CT Hash, D Hoisington and HD Upadhyaya

Output 1.3.6: Information management and analysis

Activity 1.3.6.1: Integrated marker assisted selection system (iMAS)

Milestone: Testing of standalone application integrating different software: Integrated marker assisted selection system (iMAS)

The goal of this project is to develop an integrated decision support system, called iMAS, to seamlessly facilitate marker-assisted plant breeding by integrating freely available quality software involved in the journey from phenotyping -and genotyping of genetic entities to the identification and application of trait-linked markers, and providing simple-to-understand and use online decision guidelines to use and interpret their outputs. To achieve this goal, the project has been structured into nine activities: Analyze potentially useful free software, Select software for inclusion in iMAS, Develop iMAS system, Develop and incorporate online decision guidelines, Test iMAS system, Refine iMAS system, Develop iMAS user manual/tutorial, Release of and training in iMAS, and Consultation and support. The following 8 software were incorporated into iMAS, IRRISTAT, GMendel (and possibly MapDisto), PlabQTL and Win QTL-Cartographer, Tassel, PopMin, and GGT. The functionality of IRRISTAT, GMendel, PlabQTL, and Win QTL-Cartographer has been tested, problems identified, and corrective actions taken. The functionality of others is being tested. The draft text of online decision guidelines is under preparation.

S Chandra, B Jayashree and D Hoisington

Activity 1.3.6.2 Information management systems

Milestone: The laboratory information management system comes into operation (2005**)**

Genotyping data quality algorithm developed: The workflow management system (LIMS/Laboratory Information Management System) came into operation in 2005 to help with the capture of genotyping data from the Applied Genomics Laboratory (AGL) (can be accessed at http://www.icrisat-intranet.org/lims/user.htm).

Through the course of the year, the MS-Windows based LIMS was recoded using open source software (Java) following the interests under the Challenge Program to build platform independent applications. The database server currently runs on the Windows OS and uses the MS-SQL database for data storage, but will soon be ported to the open source PostgreSQL DB. The apache Tomcat server has been used to connect the GUI (graphic user interface) to the database. The GUI has been custom built as JSP (Java server pages) pages within the struts framework. This three-tier application is modular in its construction; incorporating four major modules: start up, sample tracking, report generation, protocols and markers. Laboratory management and data management are the two kinds of functions achieved. Being modular, the application also allows for further enhancements to be made if required, through the use, re-use or replacement of existing modules. Since the primary users of the LIMS application have been laboratory technicians and students, the goal is that the system must be easy to learn and use. The LIMS is expected to encourage best laboratory practices, through some of the data quality measures built into the application. This LIMS also incorporates automated allele binning through a Java executable of the algorithm of Idury and Cordon (1997). Testing for user acceptability of this application is currently in progress.

Genotyping information from the LIMS system flows into a larger database ICRIS (Integrated ICRISAT Crop Resources Information System). The ICRIS is envisaged to be an integrated database and currently consists of three databases for genetic resource data, genotyping data and phenotypic/trait information. The database can be accessed through the user interface and simple reports generated from the genotyping data. Graphical views of the data in the form of histograms can also be generated. The ICRIS database incorporates the platform architecture evolving from the Challenge Program activity on generic platform development for GCP databases. It is a three-tier application and uses Hibernate for ORM (object relational mapping), to link the relational database at the back end to domain models in the middle layer. The relationships within the domain model will help the user build effective queries to retrieve information from the database through the user interface.

B Jayashree and S Chandra

Activity 1.3.6.3 High Performance Computing tool box

Milestone: Pipeline of parallelized tools for in-silico marker discovery (2005)

The Paracel High Performance Computer through 2005: A comparative genomics and population genetics tool box consisting of parallelized versions of popular software programs is being put in place on the Paracel HPC at ICRISAT for all interested users. A number of software was configured to work on all four nodes (eight processors) of the HPC. All the tools are open-source, they include the sequence clustering tool 'MegaBlast', sequence assembly tool 'PCAP' with improvements over the original program, 'Polybayes' for SNP marker detection, 'SNP2CAPS' a tool to convert SNPmarkers to CAPS markers, and the software used in population genetics 'Structure'. Parallelization of these tools allows for analysis of larger datasets with considerable reduction in time than would be possible with single processors. Web pages to the HPC and applications installed in it have also been written using PHP (language used for server side html embedded scripting) and PBSWEb (a web based interface to the portable batch system used for job scheduling, which uses PHP) accessible at http://hpc.icrisat.cgiar.org/ and http://hpc.icrisat.cgiar.org/PBSWeb/ .

B Jayashree and S Chandra

Project 3
Producing more and better food of the staple cereals and legumes of the west and central African (WCA) SAT (sorghum, pearl millet and groundnut) through genetic improvement

The ICRISAT team in WCA achieved a very welcome diversification during 2005. A new pearl millet breeder has strengthened activities on a wide scale, and increased our presence and ability to work with farmers, especially in the Sahelian zone of WCA substantially. Similarly a Striga ecologist joined the team in Samanko, Mali, to focus on research efforts on evaluating options for integrating Striga management measures in collaboration with farmers, in both sorghum- and millet-based systems.

Highlights of the 2005 season were the high levels of heterosis found in a wide range of Guinea race sorghum hybrids, tested for the first on a wide scale. For the first time, we succeeded in producing Guinea race hybrid seed in isolation plots, under natural pollen shedding conditions.

Groundnut seed of a wide range of new varieties continues to be in great demand, from research partners, from farmers, and for the first time, from an emerging private seed sector. Thus the first signs of changes in the supply of improved varieties through targeted commercial activities are encouraging. Efforts to develop national Foundation Seed Units are underway that can support these developments in a sustainable way.

Pearl millet improvement research started off with a large-scale germplasm evaluation and multiplication. The germplasm originates from the West-African region, and represents a wide range of agro-ecological zones. New populations are in preparation, and plans underway to classify this material into heterotic groups.

Research on micro-nutrient nutrition of sorghum and pearl millet was established more broadly with support of the Harvest Plus Challenge Program. Nutritionist input is strongly required to achieve appropriate targeting of the work.

Output 3.1: Regionally adapted diverse breeding materials, varieties and hybrids developed

New source materials provide the basis upon which new varieties and hybrids can be developed. Progress is being made on developing new and improved source materials for sorghum in several respects. The original Guinea-race sorghum population has been and continues to be diversified, targeting specific climatic zones and their adaptation requirements, including for the longer-season Guinea zone, in collaboration with Institute of Agricultural Research (IAR), Nigeria. This intensified collaboration with Nigeria was the outcome of intense interactions between Nigerian officials and the ICRISAT team in WCA. Plans for intensifying this collaboration further are underway, along with plans for new projects.

Participatory variety testing, especially for new groundnut and sorghum varieties, is becoming institutionalized among NARS partners. Methodologies are being refined for use in Mali, Niger, Burkina Faso, Nigeria, and Senegal, and for different systems, as well as crop uses. The team has been able to put together major publications on this issue, and more is in the pipeline.

Results of multi-location sorghum trials indicate that progress in increasing grain yields over local varieties, while retaining adaptive characteristics, is actually possible. Most promising are novel shorter height guinea race sorghum varieties, and guinea race sorghum hybrids.

Enhanced Nutrient Uptake Ability

Marker-assisted breeding for phosphorus acquisition ability in pearl millet

Milestone: African pearl millet germplasm identified for P acquisition from poorly soluble P sources and development of mapping populations for QTL of P acquisition ability initiated (2006)

Mechanisms of low P acquisition: We have now set up a protocol in the lab to measure root acidification of pearl millet seedling. This protocol combines pH measurement and image analysis and could potentially be used for large screening. It basically consists of using the colorimetric relation with pH of a colour indicator. Plantlets are inserted in a 3 mm layer of 0.9% agar + bromocresol blue. Image of the plate are scanned at regular interval

during 24 hours. Agar is then melted and changes in color measured spectrophotometrically at 540 nm. Alternatively, and in a more precise way, the scanned images are converted in layers of different color, *i.e.*, different pH values. The respective surfaces are measured and H^+ extrusion calculated. This allows mapping of the H^+ extrusion. With this technique, we can potentially screen the parents of existing mapping populations of pearl millet. The current orientation of this work is to modify this method to make measurement of acidification in older plants and correlate results with measurements at the seedling stage.

Phenotyping of low P millet: The phenotyping of the testcross hybrids obtained from the combination of four different male-sterile lines and the mapping progenies of the crosses between 81B-P6 and ICMP 451-P8, and between LGD 1-B-10 and ICMP 85410-P7 was undertaken in February 2005. This was the first time that scaling up low P screening was undertaken for pearl millet at ICRISAT-Patancheru. The experimental design used was an alpha lattice with 5 replications, 1 pot per entry per replication, and 1 plant per pot. As usual, 8" pots were used and filled with a sand-low P soil mixture and 200 mg of rock phosphate per kg of mixture. Plants receiving the low P treatment appeared to be very stunted compared to those receiving the control treatment. Data analysis revealed large plant-to-plant variation within entries, little consistency across replications, and problems of plant establishment.

From there on, we decided to look back at the different trials that had been conducted over the past two years to establish this protocol. Careful examination of the previous experiments (2003-2004) conducted with the mapping population parents revealed several important factors: (i) the mean biomass response of low P plants varied a lot across experiments, reaching from as low as 5-10% of control to up to 40-50%, indicating probably that large variations occurred in the P levels of the different soil lots used in the experiments; (ii) earlier experiments carried out with urea tended to have larger biomass yield means than later experiments carried out with ammonium nitrate, showing that N source may have played an important interaction with the observed low P treatment responses (for understandable reasons); (iii) yet, there was still a good correlation between the biomass yield values obtained from the set of pearl millet mapping population parents throughout time.

After this exercise, we have then addressed four issues: (i) the differences in Olsen P in the different batches of soil, (ii) the control of plant to plant variation; (iii) the source of nitrogen; (iv) the possibility to favor initial plant establishment with early application of tiny doses of soluble P.

The issue raised in (i) was confirmed coincidentally when we decided to bypass the sand acid-washing step from our protocol to save time, resources, and avoid contamination. We ran an experiment to compare the biomass response of a set of 4 pearl millet genotypes grown in low P soil mixed (1:1 w/w) either with acid-washed sand or non acid washed sand. To our surprise, pearl millet genotypes grew about twice as well in the mixture using the acid washed sand as in the one with non-acid washed sand. Soil samples from the mixture had been kept and revealed that Olsen P was 0.9 and 1.5 ppm in the mixtures using non-acid washed and acid-washed sand, respectively. This experiment indicated that: (i) very slight differences in Olsen P triggered large differences in the plant response; (ii) plant to plant variation seen in other experiments might be due to minute differences in P availability, leading to differences in plant establishment (iii) in this experiment, plant establishment was good and subsequent plant to plant variations remained minimum.

Regarding issue (ii), we now measure soil and sand Olsen P each time a new batch is used, and we ensure that Olsen P of the soil mixture is above 1.5 ppm (lower values lead to severe problems of early plant establishment). Mixing is thoroughly done using a concrete mixer to ensure homogeneity of the soil mixture. In the low P field where soil is collected, we have also applied fertilizer P in one strip, which we can use as "inoculum" in case the Olsen P value of a batch of soil turns out too low. We also grow 3 plants per pot. Preliminary indications show that the problem of plant-to-plant variation is now under control.

Regarding issue (iii), we have carried out two separate experiments. Using a set of 4 hybrids, we have found that, indeed, urea-grown plants under low P grow significantly better than under ammonium nitrate. However, there is also a significant N × genotype interaction in a sense that one genotype grows as well under low P conditions regardless of whether N is provided from NH_4NO_3 or urea. In a second experiment, we grew the same set of genotypes in 4 different soils. Data are currently being analyzed. The visual differences were very obvious regarding the overall differences between NH_4NO_3 and urea. This means that the phenotypic screening of pearl millet mapping population progenies for differences in phosphorus acquisition ability, which is foreseen, will need to be done with both N sources.

Finally, regarding issue (iv), the possibility to add tiny amounts of P to the young seedlings was initially thought to decrease variation from plant to plant. In a preliminary experiment, we had the same 4 genotypes as above,

grown in low P soil, and we treated plants with a) nothing, b) soaking one night in a 100mM KH_2PO_4 concentration, and c) applying 10 times the amount of P contained in an average pearl millet seed in a soluble form at 4 days after sowing. For plants receiving the +P treatments, we found that shoot biomass at 40 DAS was more than double of those receiving no P treatment, and this response varied across genotypes. A repeat of this experiment has been done using three soils, and three "priming" application of soluble P: 5, 10, and 20 times the amount of P in one seed. There again, we have found an important response to early application, with maximum effect being 10 times the amount in one seed. These results are very important and exciting, as they show that a very minor early application of P gives over a 40-fold return in P acquisition by the plants. Combined with issue (iii), urea plus initial "priming" give the base for a very good and applicable package.

V Vadez

Activity 3.1.1: Develop and improve trait specific populations and breeding lines of sorghum, pearl millet and groundnut

Sorghum stay-green introgression lines derived by marker-assisted backcrossing provided to African national programs in locally adapted, farmer-preferred genetic backgrounds (2008)

Due to lack of adequate SSR marker polymorphism between stay-green drought tolerance donor parent E 36-1 and the two recurrent parents (S35 and IRAT 204), and reduction in the amount of funding provided by granting agencies for activities related to this milestone, we have focused the marker-assisted backcrossing program for the sorghum stay-green trait on donor B35 only.

During 2004 marker-assisted backcrossing of genomic regions associated with the stay-green component of terminal drought tolerance of donor parent B35 was advanced two generations in the genetic background of elite recurrent parent Kapaala = ICSV 111. By the end of 2004, BC_4F_2 seed had been produced for families expected to segregate for four of the six target stay-green QTL in this genetic background (for which suitable BC_3F_1 families were available in 2003) and for the remaining two target QTL, seed had been produced for BC_3F_2 families.

During the first half of 2005, BC_3F_2 and BC_4F_2 plants homozygous for donor parent alleles at SSR marker loci flanking each of six individual stay-green loci (*i.e.*, single-QTL introgression lines for stay-green QTLs *stg*A, *stg*B, *stg*1, *stg*2, *stg*3 and *stg*4) from donor parent B35 in the genetic background of ICSV 111 (released in Ghana as 'Kapaala') and its sub-selection S 35, were identified and their selfed seed harvested. These included ten BC_4F_2 plants expected to be homozygous for *stg*A, three BC_4F_2 plants expected to be homozygous for *stg*B, six $BC3F_2$ plants expected to be homozygous for *stg*1, ten BC_3F_2 plants expected to be homozygous for *stg*2, two BC_3F_2 and seven BC_4F_2 plants expected to be homozygous for *stg*3, and four BC_4F_2 plants expected to be homozygous for *stg*4. The corresponding BC_4F_3 and BC_3F_3 families, along with seed samples of their recurrent and donor parents, were sent to the national sorghum breeding program in Ghana (SARI) in June 2005 in time for initial rainy season seed increase for agronomic and farmer-participatory evaluation under the Water for Food Challenge Program. Further, these progenies were advanced a further generation by selfing at ICRISAT-Patancheru during the 2005-06 post-rainy season to produce seed required for future assessment of their drought tolerance, agronomic performance and grain quality. Marker data generation in 2005 required to identify the putative QTL introgression homozygotes was performed as part of the M.Sc. thesis research program of Mr. Sripathi Venkateswararao. In late 2005 he submitted a thesis to Acharya N.G. Ranga Agricultural University (ANGRAU) India, and this was defended successfully in early 2006.

Milestone: Later maturing guinea-race random-mating population of sorghum developed for Northern Guinea zone

A random-mating population was created by crossing a Nigerian Guinea-race sorghum accession IS 7978 with the Guinea Population containing the male-sterile gene ms3. IS 7978 is late maturing (flowering October 18 at ICRISAT-Mali) with large grain size (3.5 g/100 seeds). This new population was included in a five-variety on-farm trial in Kaduna state, Nigeria in 2005. Results obtained from a total of 8 villages indicated that although the grain was often appreciated, the flowering was often too early (dates between 30 September and 18 October frequently recorded). Farmers indicated this population to be of interest in for the more northerly Sudanian zone. Original F_2 seed of this population will be used in 2006 to cross to Kaura and Fara Fara varieties from the Northern Guinea zone to create a later-maturing population.

HFW Rattunde and E Weltzien

Milestone: Guinea sorghum hybrid parents tolerant/resistant to midge identified

Several Guinea-race germplasm accessions from Cameroon have shown consistent high levels of resistance to midge, both *per se* and in the hybrids they produce. Results from the 2005 Hybrid trial indicated no loss due to midge (0 to 1%) for accessions IS 15302, IS 15629 and IS 30804, all from Cameroon, whereas other accessions of similar maturity had near total loss (IS 26320 (Togo) and IS 7978 (Nigeria) had 84 and 75% losses, respectively). Likewise the hybrids produced with the Cameroon accessions as male parents also showed very high levels of resistance (0 to 2% loss), despite the female parent (IS 3534A) showing 24% loss.

HFW Rattunde and E Weltzien

Milestone: **Diversified agronomically superior dual-purpose groundnut breeding lines with improved resistance to foliar diseases and rosette**

Groundnut is an important crop for resource-poor farmers in Semi-arid West Africa, who rely on it for their economic prosperity and nutritional welfare. Nutritionally groundnuts are an excellent source of dietary protein, oil/fat, and vitamins such as thiamine, riboflavin and niacin.

Foliar diseases such as rust, early and late leaf spots and groundnut rosette disease cause significant yield losses in groundnut in WCA. In order to stabilize yields and enhance productivity development of high yielding resistant varieties is the most economic approach.

We evaluated 36 advanced groundnut-breeding lines with multiple attributes (dual purpose, tolerant to Aflatoxin contamination and resistant groundnut rosette disease) for resistance early leaf spot. Among 14 early maturing rosette resistant lines, one was highly tolerant to early leaf spot (score of 5 on 1-9 scale) at Samanko in Mali. For the dual-purpose lines, 10 had a score of 4-5 compared to the susceptible check with a score of 8. The six aflatoxin resistant lines were also tolerant to early leaf spot.

We also advanced 71 rosette resistant F_2 populations to F_3 and 47 to F_4. Bulk progenies were selected for further selection and generation advance. These populations were also scored for early leaf spots. Among the F_2 populations, 7 had a score of 5 while in the F_3 populations, 12 had a score of <5. Varieties with high pod and fodder yield with resistance to foliar diseases are expected from these improved breeding lines and populations.

BR Ntare and AT Diallo

Milestone: Multiplication and characterization of pearl millet genebank accessions of diverse geographic origin

One of the objectives of WCA pearl millet improvement program at ICRISAT-Niamey is to better exploit and to enhance access of NARS breeders and farmers to the genetic diversity of pearl millet [*Pennisetum glaucum* (L.) R. Br.] in its center of origin in West Africa. Therefore, 281 pearl millet accessions from nine countries of West and Central Africa, assembled during joint IRD/ICRISAT pearl millet collections in 1976 and 2003 and geographically covering longitudes from 8.44E to 17.28W and latitudes from 6.49N to 20.26N, were grown for seed multiplication and initial characterization in the rainy season 2005 at ICRISAT, Sadoré (Niger).

The multiplication of materials was performed by controlled pollination ("sibbing") within each accession, aiming at a minimum effective population size of 60 plants contributing to the next generation. Pollen donors were simultaneously selfed to obtain S_1 seed as an initial step of further inbred line development out of the most promising accessions. Characterization data include days to 50% flowering, plant height, panicle length and diameter, exsertion, form and compactness of the panicle, numbers of selfed and sibbed panicles, and grain weights of selfed and sibbed panicles. The determination of 100-seed weight and grain color is still underway.

Raw data indicate that the germplasm reveals significant variation for all morphological and phenological traits studied: e.g., observations ranged from 40 to 155 days to 50% flowering, 153 to 405 cm for plant height, 18 to 115 cm for panicle length, 1 to 6 cm for panicle diameter, and from minus 23 to plus 12 cm for panicle exsertion. These accessions are therefore a gold mine of variability for future breeding.

Seed multiplication through sibbing was achieved for 248 out of the 281 accessions, with the amount of seed produced ranging from 22 to 2474 g. The effective population size of ≥60 was achieved in 117 accessions (minimum of 30 sibbed panicles). Twenty five accessions were multiplied with an effective population size of less than 20 (<10 sibbed panicles), due to poor germination or lack of adaptation of the accessions. Selfed seed was produced from 232 accessions, with the seed quantities obtained ranging from 14 to 2901 g.

Small amounts of the regenerated seed will be returned to the IRD-Montpellier and ICRISAT-Niamey genebanks for long-term conservation. The complete characterization data shall be linked to the gene banks information systems. The data will mainly be used for further diversity analysis and studies of heterotic grouping of West African pearl millet genetic resources with the final aim of significantly improving pearl millet yield performance and stability in West Africa. The cooperation from IRD-Montpellier and IRD-Niamey (Drs Y Vigouroux, G Bezançon) in providing the original seed of the accessions and extra-large pollination bags is highly appreciated.

BIG Haussmann, SS Boureima and A Boubacar

Activity 3.1.2: Develop hybrid parents adapted to specific zones of cereals cultivation in WCA

Milestone: Superior Guinea-race sorghum R-lines identified

A set of 29 restorer lines of diverse geographic origin was established for creating experimental hybrids. These R-lines were identified based on prior testcross results and chosen to represent geographic and morphological diversity. Twenty restorer lines were identified from accessions in the Guinea-race Core Collection, and nine landraces or bred varieties from Mali. The restorer lines were used to produce hybrids on newly developed A-lines FambeA, IPS001A, and IS3534A. Hybrid trials were conducted in 2005 at ICRISAT-Mali (two dates of sowing), IER-Sotuba, Mali and Bengou, Niger. Later maturing restorer lines of humid West Africa (Nigeria, Cameroon, Togo) origin produced hybrids with the highest grain yields (Table 3.1). Restorer lines of intermediate maturity, although not producing the highest yielding hybrids, still provided significantly significant yield superiority over highly adapted check varieties.

Table 3.1. Mean grain yields of hybrids over four locations in West Africa, 2005, with diverse restorer lines crossed onto a common female parent FambeA

Hybrids	Origin of male	Yield (t ha^{-1})
FambeA*Fara Fara-17	Nigeria	3.0
FambeA*IS 26320tg	Togo	2.9
FambeA*IS 23206za	Zambia	2.8
FambeA*IS 7978na	Nigeria	2.8
FambeA*IS 27113zi	Zimbabwe	2.7
FambeA*GPN01 267-9-1	Mali	2.6
FambeA*Seguetana CZ	Mali	2.3
Check Varieties		
CSM335	Mali	1.8
Seguetana CZ 24/25	Mali	1.6
CSM388	Mali	1.5
SE ±		0.19

A restorer line database with agronomic characterization information and combining ability results of both Guinea-race and inter-racial restorer lines developed by IER, is being compiled jointly with IER. Origin, days to heading and plant height of identified restorer lines used for experimental hybrid production are being documented.

Activity 3.1.3: Participatory testing and release of improved sorghum, pearl millet and groundnut varieties

Milestone: Advanced yield testing of new breeding material evaluated jointly with Malian partners, and with farmer participation

Organized variety testing with farmer participation started in 2003 in 11-12 villages and on 2-3 research stations. The same 32 varieties were grown at each location.

Eight (in 2003) and seven (in 2004, 2005) of the villages were situated in Dioila district, an area of more intensive agricultural production. Many farmers are literate and well organized at a local level. Five sites were managed by the farmer organizations which form part of the Union of Cereal Producers in Dioila district (ULPC). Three were managed by village organizations which were initially formed for the management of cotton production in the respective villages, with whom the researchers had developed good working relationships through a close collaboration with the extension service of the cotton parastatal (CMDT= Compagnie Malienne du Developpement des Textiles). In the Mandé area, villages were suggested by the extension partners, who had longstanding relationships with many villages.

The participating farmers were primarily chosen by the farmers' organizations (Dioila) and the extension services (Mandé). The farmers were responsible for choosing the field for the trial and two local control varieties: one common for the whole village and one of specific interest to the farmer who provided the field. Four farmer participants together with the person providing technical assistance chose the village level check variety, usually one of the dominant varieties in the village used by many farmers. The farmers were involved in the choice of the test entries in the following manner:

1. Some varieties were retained from a precursor trial. They were retained based on farmers' choice and the yielding ability in the trials.
2. Farmers involved in the trials came to visit the ICRISAT research station during the pre-harvest period. They were shown the S2- progeny trials from the diversified Guinea-race populations, from which entries for the farmers' trials were to be selected. They scored each plot, using a score from 1-3 with color-coded paper pieces. The preference of farmers was one main criterion to choose varieties for the trials.
3. Farmers did not, however, visit the IER breeding stations, and thus experimental varieties from IER were chosen by researchers primarily. However, IER also conducts some of its selection program in close collaboration with farmers, and thus some materials have been selected by farmers in other areas of sorghum cultivation. These entries were fixed lines, which had previously been tested in multi-location station trials.

Farmers were responsible for managing the trial field and for visual evaluation for a range of traits identified previously together with them. Farmers received a basal dose of N and P fertilizer, which they applied at sowing time. The seed was treated (if the chemical was available in the local market). Each farmer grew one replication of the 32-entry trial. The 6 row plots of 5 m length were arranged in 4 ranges of eight plots each, and randomized as alpha-lattice designs with 4 plots per block. Students and local extension officers supported the farmers, particularly with sowing, plot identification, recording of observations, decision-making about management, and at harvest. Weighing the yield of each individual plot was a key responsibility of the technical support staff. They further organized the visits of other farmers to the field trials and their evaluation of the test varieties. The researchers contributed to the organization of these visits and organized the testing for processing and culinary qualities

In the other region (Mandé), where agriculture is more extensive and less cotton is grown, the same trials with 32 entries were grown in four villages. Extension agents of a local NGO and the government extension service supported the farmers. The responsibilities were shared in the same manner as described above.

Farmers' visits to the trials were organized at one research station and in at least 10 villages each year. All visiting farmers scored all varieties for their overall performance and acceptability, using a 1-3 scale and paper slips with different colors to signify each score (Christinck et al., 2005, p.96). After harvest, a two-day workshop was organized for each pair of neighbouring trial sites in order to discuss the yield results and evaluate grain and culinary qualities. On the first day the results of the yield evaluations, the farmers' selection, and other key observations were presented to the farmers who had participated in the trials as well other interested farmers from the villages concerned. The results were discussed and four varieties were chosen for the culinary trials the following day. The key activity on the second day was the evaluation of processing qualities and the culinary quality of the four best varieties in each village. All participants could also evaluate the grains of each variety visually, using the same 1-3 scoring system using different colored paper slips.

The four entries selected by the workshop participants for the culinary testing were considered to enter the second stage of testing, but only if their processing and culinary characteristics were found to be acceptable.

After harvest, and after completing the tests for culinary quality, a workshop with all farmers who had participated in the trials and other project activities was organized for each project zone (Dioila and Mandé).

Yield data were discussed and varieties finally selected for further testing. Furthermore, changes with regard to trial management, monitoring and responsibilities for the diffusion of results were discussed and decided jointly.

Results of yield trials

The results of the yield trials were very encouraging in all years (2003, 2004, 2005), in the sense that all trials could be harvested and evaluated. Only individual replications of trials had to be abandoned in a few cases.

The seasons were markedly different; in 2003, the rainy season was very good, started early and continued until mid-October in all the project areas. This even led to some difficulties caused by excessive rainfall, such as water logging. In 2004, however, the rainy season started late and ended earlier than expected, as a consequence terminal drought stress occurred, particularly in fields with lower water holding capacities. In 2005, the season started some what earlier than in 2004 and ended earlier than normal, but later than in 2004.

In 2003, varieties could be identified in each village which were more preferred by the farmers than the local check entries. The yield superiority, however, was fairly low, between 10-20% on a variety mean basis for individual villages. A number of new dwarf lines performed relatively well in these trials, and as these are not yet finished varieties, the remaining variability could be exploited in order to further improve the grain yields.

In 2004, some of the new improved varieties showed clearly superior grain yields over the farmers' check entries in both project areas, and also reached high values in the farmers' preference scoring. This was partly due to earlier maturity, an advantage under the end-of-season drought conditions encountered this year. Mean grain yield varied widely between locations, and there was also considerable variability between individual replications within the same village. This made the data evaluation more difficult. The flowering dates were only recorded at the research stations. Table 3.2 gives the results of one of the villages in the Doila area.

Table 3.2. Yield (t ha^{-1}) and preference of the best performing varieties and the controls in Wacoro village, 2004 rainy season. (The names of the best varieties are given in brackets)

Variety	Rep 1 Nonkon Dembele	Rep 2 M'Pie Dembele	Rep 3 Moussa Bengaly	Rep 4 Tiecoura Traore	Overall Wacoro	
					Yield	Preference
Mean	1.4	1.1	1.2	1.0	1.17	48%
Village Check	1.5	1.1	1.2	0.9	1.19	68%
Farmer check	1.2	1.3	1.2	1.1	1.21	85%
Best variety	**2.3** (Bolibana)	**1.7** (Lafia)	**2.2** (Kalaban)	**1.6** (Coni)	1.50 (Kalaban)	41%
2nd variety	**2.3** (Coni)	**1.6** (Quinzen)	**1.9** (Sebekoro)	**1.6** (Kalaban)	1.50 (Lafia)	51%
3rd variety	**2.0** (Magnan)	**1.5** (Koura)	**1.7** (Grinka)	**1.5** (Weli)	1.48 (Coni)	48%

There is now increasing interest in the shorter sorghum varieties, as they exhibit better stover quality than the tall varieties that are highly lignified. Farmers are also experiencing that they are easier to harvest, and tend to give higher grain yields.

E Weltzien, HFW Rattunde, I Sissoko and A Christinck

Milestone: Synthesis report on participatory variety selection in groundnut available

Investments by ICRISAT and partners have resulted in the development of a broad range of groundnut varieties. However, farmers have limited access to these varieties. The key is to make available a range of modern varieties and train farmers to efficiently produce seed of selected varieties, using appropriate technologies leading to increase rural incomes.

Over 200 participatory varietal selection (PVS) trials were conducted in 45 locations across four countries: Mali, Niger, Nigeria and Senegal. In each country farmers have selected at least one or two new groundnut varieties. Seed production schemes were initiated in each country to ensure availability of seed of these varieties. The synthesis report will document the PVS process, pathways to adoption of improved varieties, lessons learned and perspectives.

BR Ntare, J Ndjeunga, AT Diallo, HY Bissala and F Waliyar

Milestone: A synthesis report on groundnut seed systems in WCA

The availability and uptake of seed of high quality by farmers is fundamental to the transformation of predominant traditional agricultural production practices to achieve increased stability and sustainable food production in West Africa. New seeds with higher yield potential or ability to relieve constraints faced by farmers in using traditional varieties form part of the improved inputs required to increase crop production.

This technical paper summarizes information on the structure, conduct and performance of formal and informal groundnut seed supply systems in 4 countries in West Africa namely Mali, Niger, Nigeria and Senegal. It highlights a range of technical, socio-economic, institutional and policy constraints facing the groundnut seed industry in West Africa. Low and inconsistent supply of breeder seed, poor seed demand estimation, lack of or non-functional national variety release committees, inappropriate institutional arrangements and the biological features of groundnut have limited private sector entry and the performance of the groundnut seed industry. Options likely to be sustainable should focus on local village seed schemes whereas small-scale private seed entrepreneurs or community based seed systems should be encouraged to become seed entrepreneurs or engaged in the seed industry. There is evidence of vertical integration between inputs and product markets. Appropriate linkages between seed and grain producers, and grain producers and processors are necessary to drive the private sector entry in the seed industry.

J Ndjeunga, BR Ntare, F Waliyar and M Ramouch

Milestone: **Technical paper on market prospects for groundnut in WCA published**

This is a result of a study commissioned by ICRISAT, with financial support from the Common Fund for Commodities. The paper documents the principal groundnut producing countries in the international markets, global market status trends and quality requirements, recent trends in production and consumption, and strategies for increasing groundnut production in West Africa.

BR Ntare, F Waliyar, M Ramouch, E Masters and J Ndjeunga

Output 3.2: Methodologies for enhancing productivity, adaptation of sorghum pearl millet and groundnut cultivars developed

Basic research into adaptive mechanisms of the local guinea races sorghums is showing more explicitly that not only phonological changes are affected by sensitivity to the photoperiod, but also growth rate changes. However these changes do not affect the whole plant, but rather the above ground dry-matter. It seems that root growth continues unchanged during the various phenological changes of the sorghum plant, in contrast to maize. Research was started to understand better the differential responses of pearl millet varieties to different flowering dates, but also its pattern of root growth.

During 2005 a handbook on Priority setting with farmers for breeding programs and seed system activities was finalized. The book provides basic insights and concepts, but also practical tools for the field, described in sufficient detail, that they can be used directly.

Activity 3.2.1: Design sorghum and pearl millet ideotypes for a regional selection strategy for increasing productivity under production system-specific conditions

Milestone: Testing and grouping Guinea race sorghum and pearl millet, for photoperiod-sensitivity under the newly shown categories

A regional sorghum core-collection of 214 accessions was sown on the 26 June 2004 and characterized for their phyllochron during the life cycle. The goal of the study was to estimate the variability of the rate of development in sorghum, since this character and its evolution during the life cycle of the plant could give insights into how to increase the yield potential of photoperiod-sensitive varieties. The sorghum core-collection managed and studied by CIRAD has been used in this study. This study has largely demonstrated the generality

of bilinearity of the leaf appearance rate for long-duration varieties producing more than 22-23 leaves, while the leaf appearance rate remained always constant for short-duration varieties. It showed also that, for sowing dates at the end of June, varieties of the guinea race (including *margaritiferum*, *gambicum* and *conspicuum*) acquired a shorter phyllochron at emergence, than varieties from the *caudatum* race which were intermediate and varieties from the *durra* and *kafir* races which had the longest phyllochron. This suggests that varieties from the guinea race expand their first leaves faster than varieties from the other races, when sown late.

A new series of monthly sowings of 12 pearl millet varieties with a large range of photoperiod-sensitivity was initiated in June 2004. The 18th sowings has been done in December 2005. Current results show patterns similar to sorghum: (i) the duration of the vegetative phase of 9 varieties varies slightly with the sowing date, corresponding with a quantitative photoperiod-sensitivity, while for the 3 other varieties this vegetative phase has been very long for sowings done during the beginning of the year, corresponding with a qualitative or absolute photoperiod-sensitivity; (ii) when more than 25 leaves have been produced by the apex, the rates of development and growth of the plants slowed down considerably and simultaneously; (iii) the rates of development acquired at emergence doubled from sowings done on the summer solstice to those done on the winter solstice. On the other hand, the vegetative phase of the 12 pearl millet varieties tested in Bamako has been minimal for sowings done at the end of June-beginning of July, while this minimum generally occurs for sowings done in October with sorghum. This last observation is contradictory to the current theory, established under artificial lightings, which predicts that pearl millet, a short-day species, must flower later when days are the longest.

B Clerget, HFW Rattunde and B Siaka

Milestone: Enhance knowledge on dwarfing and panicle size x photoperiod-sensitivity interactions

Panicle size x photoperiod-sensitivity interactions in experimental guinea hybrids

Ten experimental guinea sorghum hybrids and their parents were sown on the beginning of June and July to assess the contribution of the yield components responsible for the heterosis observed. The hypothesis is that the heterosis is due to a larger panicles size. Harvest data showed significant heterosis for grain yield of hybrids based on the parent Fambé A. These hybrids had more grains per panicle than their male parents. For July sowings, this larger number of grains clearly results in a better harvest index, thus a larger number of grains per unit of biomass. For the June sowing, on the contrary, the harvest index of the hybrids was generally not better than the parents and the larger grain number appears to be only related with an increase of the total biomass produced by the hybrids. In June sowings, the grain size (100-grain weight) of the hybrids was also slightly larger than that of the parents. On the other hand, no difference was observed between hybrids and parents in the rates of development of the panicle nodes. They remained always equal to the rate of leaf initiation at the time of the panicle initiation. These results show that heterosis depends on the sowing date, as the expression of the yield improvement of photoperiod-sensitive varieties and hybrids varies with the sowing date. It is necessary to understand these variations in order to find better combinations.

B Clerget and S Dagnoko

Comparing the rates of aerial and root development of sorghum and maize

Tropical maize has a much better yield potential than photoperiod-sensitive guinea race sorghum. To compare their developmental strategies, a parallel study of the developments of the aerial plant and of the horizontal and vertical root fronts were carried out with locally cultivated varieties. The rates of development acquired at emergence were similar for both species. Maize expanded 22 leaves at a constant rate of leaf appearance, against 28 leaves for sorghum with a bilinear kinetic. Maize has produced 15.4 t ha^{-1} of total biomass and 7 t ha^{-1} grain within 90 days against 18 t ha^{-1} of biomass, 3.5 t ha^{-1} grain in 130 days for sorghum. The horizontal rates of root growth were 1.2 and 1.8 cm day^{-1} for maize and sorghum, respectively. The maximal vertical rates of root growth have been similar and equal to 2.8 cm day^{-1} for both species. But the strategy to conquer the vertical dimension of soil has been very different between the species: the root tip of sorghum descended quickly at a constant rate from emergence to the 62nd day, time of panicle initiation. It continued its descent at a reduced constant rate (1.2 cm day^{-1}) until grain maturity. Consequently there was no link between the rate of descent of the roots and the rate of leaf appearance, which broke on the 41st day. In maize, the vertical progress of the root tip stopped one week after germination at a depth of 20 cm. It started again only on the 21st day, at panicle initiation time, and definitively stopped on the 56th day, when internodes had finished elongating. It thus appears that (i) the rate of descent of the root tip would be related to the phenology of the aerial plant, but differs with species and (ii) the roots of sorghum continue to go down quickly even after the rate of development of the aerial plant has decreased and the stem has started to elongate. This pattern of root growth may explain the

better adaptation of late maturing guinea race sorghums to low fertility soils: during its stem elongation phase, the biomass is synthesized at a lower rate using minerals and water extracted from a larger volume of soil in comparison with maize.

B Clerget

Activity 3.2.2: Refine and adapt methodologies for farmer participation in specific stages of sorghum and pearl millet breeding

Milestone: Handbook on farmer participation in priority setting published

The handbook was published, and has been used as a basis for some training courses. The book is in high demand, and the first edition is nearly out of print.

A Christinck, E Weltzien and V Hoffmann

Output 3.3: Impact-oriented eco-friendly IPM technologies developed

During 2005 research on IPM technologies focused on two of the most complex issues faced by farmers: Aflatoxin contamination of groundnuts, and *Striga* management in dry-land cereals. For *Striga* control research on integrating control measures in a farmer participatory, joint learning was designed, and discussed with partners. We are looking forward to its implementation in 2006. For aflatoxin contamination results from widespread on-farm testing and evaluation is now available, and is leading to intense discussion on consequences, among farmers themselves, farmers and processors, as well as consumers.

Activity 1.2.1.4: Mapping *Striga* resistance in sorghum

Milestone: EST-derived markers closely linked to *Striga* resistance in sorghum identified (2006)

Sequencing of gene-rich regions of the sorghum genome has moved ahead rapidly, as evidenced by the November 2004 release by Orion Genomics to the public domain of over 500K methyl-filtered sequence reads, predicted to include at least a portion of the sequence of 95% of all sorghum genes (more than 784K sorghum DNA sequences are now available in GenBank). Interestingly, some 25K of the new methyl-filtered sequence reads contain SSR repeat motifs. Thus, the currently available sorghum DNA sequence information would allow substantial expansion of sequence-tagged microsatellite (STMS) marker resources for sorghum, and could provide polymorphic co-dominant PCR-compatible markers in large numbers across the entire sorghum genome. Even more interesting is that by applying simple, inexpensive bioinformatics protocols, we can identify with reasonable certainty where many of these new STMS markers have counterparts in the rice genome sequence, and hence can predict where they will map on the sorghum genome.

Using this approach, (as part of the PhD thesis research program of Mr. P. Ramu), sorghum EST sequences from The Institute for Genome Research (TIGR) database were searched for the presence of SSRs in their sequences. The non-redundant sorghum EST sequences with SSR motifs were searched against the rice genome sequence in the Gramene database (using BLAST). Among the hits identified, those having the highest score were checked for its location on the rice genome. Nearly 2,000 non-redundant sorghum EST sequences containing SSRs were searched against the rice genome. For each rice linkage group, 50 hits with map positions distributed across the full length of the linkage group were selected. This revealed rice chromosomal locations of similar sequences, allowing selection of a subset of 600 sorghum EST-SSR loci distributed across regions of the sorghum genome that are syntenic with each of the 12 rice chromosomes (50 each). These sorghum EST-SSR loci, if polymorphic in sorghum, are expected to provide coverage across the entire nuclear genome of sorghum. Primer pairs flanking the repeat sequences in each of these 600 sorghum EST sequences were then designed (after masking repeat regions) using the Primer3 program (www.genome.wi.mit.edu). PCR optimization and polymorphism assessment of primer pairs for these 600 candidate sorghum EST-SSR markers were taken up simultaneously using as template DNA samples from the parental lines of several ICRISAT sorghum RIL populations, including those previously used for mapping *Striga* resistance.

During 2005, a subset of the E36-1 × N 13-based RIL population has been used (by PhD student, Kassahun Bantte) to start mapping some of the polymorphic EST-SSR markers. Genotyping has been completed and mapping initiated for about 50 EST-SSR loci expected to map to four of the ten sorghum linkage groups (SBI-01, SBI-02, SBI-03, and SBI-05). Based on the marker data generated, 44 of these EST-SSR markers were mapped across all 10 sorghum linkage groups (not just those actually targeted). Among these 44 newly mapped markers, four were closely linked to the *stg*B QTL on LG B = SBI-02 (which was a primary target of this

exploratory mapping exercise). These findings will facilitate future development of additional sorghum EST-SSR markers specifically targeting QTLs for any mapped trait, including *Striga* resistance, for use in marker-assisted selection. With completion of this activity, if we need more SSR markers in a particular region of the sorghum genome, we hope to be in a position to quickly and inexpensively develop them, using the vast amount of sorghum sequence information that is rapidly becoming available.

RT Folkertsma, CT Hash, B Jayashree and BIG Haussmann

Striga resistance in sorghum transferred to elite African cultivars using marker-assisted selection. Marker-assisted backcrossing of *Striga* resistance QTLs from donor parent N 13 into the genetic background of farmer-preferred sorghum varieties from Mali, Sudan, Kenya, and Eritrea were advanced more slowly than planned during 2005. Logistical problems in moving DNA samples, and/or appropriate tissue samples for DNA isolation between national sorghum breeding program sites (where the crossing and backcrossing activities are undertaken) and the BecA facility in Nairobi, Kenya (where the SSR marker genotyping activities are undertaken), are hampering progress towards this milestone.

FR Folkertsma, S deVilliers, D Hoisington , D Kiambi and CT Hash

Arresting the scourge of Striga in sorghum in Africa by combining the strength of marker-assisted backcrossing with farmer-participatory selection: Through this project, NARS in the Kenya, Mali, Eritrea and Sudan are being assisted to strengthen *Striga* resistance of farmer-preferred sorghum varieties (FPSVs) through a combination of marker-assisted backcrossing (MAB) and farmer-participatory selection. The stability of inheritance of the transferred *Striga* resistance alleles in the FPSVs, the actual out-crossing rates in selected FPSVs, and the pollen flow of these FPSVs is being analyzed in order to develop recommendations for variety maintenance and on-farm seed production. To complement the molecular work, a socio-economic and population genetics study of the sorghum seed supply systems in the four target countries is being undertaken concurrently to guide the design of effective seed interventions by partner institutions so that improved materials efficiently reach farmers.

The first generation of backcrosses between F_1 and the farmer-preferred varieties (Hugurtay, Hiryray, Ochuti, Tabat, Wad Ahamat, Tiemarifign, and CSM 335) has been performed and BC_1F_1 generated in Kenya, Mali, Sudan, and Eritrea. In Kenya, 210 BC_1F_1 individuals were genotyped using 3 SSR markers: 63 plants were found to be heterozygous for markers in the QTL target regions – 3 plants contained 3 *Striga* QTLs, 13 had 2 *Striga* QTLs, and 47 had 1 *Striga* QTL. Confirmed hybrids were backcrossed to the FPSV recurrent parent Ochuti to produce the BC_1F_1 generation, which is currently being genotyped. In Sudan, 144 Tabat BC_1F_1 samples were genotyped and 28 plants were found to be heterozygous for markers in the QTL target regions – 5 plants contained 4 *Striga* QTLs, 4 contained alleles of 3 QTLs, 5 had 2 QTLs, and 12 had 1 QTL. Capacity for DNA isolation, quantification, PCR optimization, and agarose/PAGE with silver staining has been provided to the NARS in Mali (UB and IER), and Kenya (KARI-Katumani) through training, provision of equipment, and technical backstopping.

A gene flow experiment was set up to determine the distance of pollen flow using male-sterile lines as receptors, thus eliminating the need of large scale PCR genotyping. The flowering dates of the materials from Sudan, Kenya, and the male-sterile lines were determined and the first pollen dispersal experiment carried out using Ochuti and N 13 pollen donor and recipient, respectively. The experiment is currently being repeated at another site. A field study has been conducted on sorghum indigenous knowledge, farming system, stakeholders' perceptions and seed supply systems in Eritrea, Kenya, and Sudan. A report containing the findings of status of the seed sub-sector in Eritrea, Kenya, and Sudan has been compiled.

D Kiambi

Similarly, marker-assisted backcross introgression of stay-green QTLs from donor parent B35 into the genetic background of recurrent parent IRAT 204 advanced two generations during 2005, with the marker data generation required being performed as part of the M.Sc. thesis research program of Mr. Sripathi Venkateswararao.

Activity 3.3.1: Develop and evaluate on-farm integrated Striga control strategies in sorghum and pearl millet (in collaboration with GT Agro-ecology)

Milestone: Integrated options for Striga control in pearl millet tested and refined in two target production zones

Potential for sesame to contribute to integrated control of *Striga hermonthica* of pearl millet in the West African Sahel

Striga hermonthica is an important constraint to the production of pearl millet, a staple cereal in many parts of sub-Saharan Africa. Sesame is an important oilseed crop well adapted to the sandy soils of the West African Sahel. Intercropping of sesame and pearl millet has been reported to reduce emerged *Striga* numbers, but formal research into the potential of sesame to contribute to control the parasite is lacking. Field trials were undertaken to evaluate the potential of sesame grown in rotation with pearl millet to reduce *Striga* infestation. Emerged *Striga* numbers and *Striga* fruiting were strongly reduced on pearl millet following sesame compared to sole millet. To maximize cereal yield, soil fertility enhancement and water conservation are indispensable elements of integrated *Striga* control. The results can guide future research at a time where sesame is being promoted to diversify agricultural production in the Sahel.

DE Hess, H Dodo and E Weltzien

Integrated *Striga* management in farmers pearl millet and sorghum fields

A 6-year on-farm trial with Integrated *Striga* management strategies in Mali and Niger led to very large reductions in the number of emerged *Striga* plants as well as seed bank densities, when compared to the normal farmers' practice. Although not quantified, farmers are known to adopt parts, if not all of the measures in other fields infected with *Striga*. More detailed analyses are underway.

DE Hess, R Tabo and E Weltzien

Milestone: Biology of resistance to Striga in sorghum understood

The role of sorghum genotype in the interaction with the parasitic weed *Striga hermonthica*: The main objective of this study was to find suitable measures for the selection of breeding material (crop genotypes) with superior levels of resistance or tolerance to *Striga*. The relation between *Striga* infestation, infection and yield loss and the effect of host genotype on *Striga* parasitism and reproduction were studied for 4-10 genotypes in agar-gel, pot and field tests. *Striga* parasitism and reproduction, and the detrimental effect of *Striga* on crop yield can be significantly reduced through crop genotype choice. Maximum aboveground *Striga* number is a reliable selection measure for resistance. *Striga* flower stalk dry weight can be used to identify genotypes that reduce *Striga* reproduction. The maximum relative yield loss is a suitable selection measure for tolerance in susceptible genotypes, while for genotypes that are more resistant the relative yield loss per *Striga* infection seems more appropriate. For these tolerance measures, yield assessment of nearby uninfected controls is indispensable. Chlorophyll fluorescence, more precisely photochemical quenching and electron transport rate, may enable screening for tolerance without this requirement.

J Rodenburg, E Weltzien and D Hess

Combining the strengths of marker-assisted backcrossing and farmer-participatory selection to improve *Striga* resistance in sorghum: *Striga*-resistant sorghums would be an important component of integrated *Striga* management if resistance was available in locally adapted farmer varieties. The application of marker-assisted selection in *Striga* resistance breeding would greatly accelerate progress since field screening is difficult, complex, and often unreliable; *Striga* seed is quarantined thus confining tests to areas where *Striga* is endemic; and because some *Striga* resistance genes are recessive, increasing the time required for conventional backcrossing. QTL mapping for resistance of sorghum to S. hermonthica was performed using a population of F3:5 lines developed from the cross N13 x E36-1, where the resistant sorghum line N13 is characterized by "mechanical" resistance (Haussmann et al., 2004). Composite interval mapping detected five QTL common across five environments over two years of *Striga* resistance evaluation, with the resistance alleles deriving from N13. Since their effects were validated across environments, years and independent genotype samples, these robust QTL are excellent candidates for marker-assisted selection. In a three-year project, launched in April 2004, *Striga* resistance of farmer-preferred sorghum varieties in Eritrea, Kenya, Mali and Sudan will be enhanced through a combination of marker-assisted backcrossing and farmer-participatory selection. The impact of gene flow on the stability of the achieved *Striga* resistance will be investigated in a complementary study.

Simultaneously, a socio-economic study of the sorghum seed supply systems in these countries will be undertaken to guide the design of effective seed interventions by partner institutions so that improved materials efficiently reach farmers. Linkage with technology exchange will boost promotion of the improved varieties as component of integrated *Striga* control.

RT Folkertsma, BIG Haussmann, HK. Parzies, D Kiambi, V Hoffmann and H H Geiger

Milestone: Quantification of the *Striga hermonthica* life cycle as a tool for seed bank management

Studies on weed management in cropping systems have shown there are economical advantages in managing long-term weed population dynamics in addition to short-term management to control weeds and prevent crop yield loss. An important component of integrated weed management is monitoring and attempting to predict how cropping systems and control strategies affect long-term population dynamics of weeds. In order to be able to model long term *Striga* seed bank dynamics, steps in the life cycle such as seed bank replenishment (seed production) and seed bank depletion (seed mortality in the soil), were quantified.

In six field experiments, we tried to: (i) develop a reliable, standardized method for monitoring seed production; and (ii) determine the effect of rainy season length, seed density, host cycle length and several control strategies on aboveground demography leading to seed production. Seed bank germination and depletion of *Striga* was also measured at a site in Mali and a site in Niger during one rainy season under different crop and fallow systems.

Seed production was affected by rainy season and host cycle length, as well as by different control strategies. A five-fold increase in initial seed density did not affect seed production and data indicated possible density dependence in underground stages, although with a very high variability. There were striking differences in above-ground *Striga* appearance between years and sites considering small differences in infestation or inoculation levels of (germinable) seeds. Finally, a relation was found between allometric seed production estimates and soil seed content to a depth of 3 cm. Seed production and seed bank dynamics of *Striga* are affected by season length, host characteristics and should therefore be incorporated into population modeling to support choices of integrated control methods.

Until now it is not clear what influences emergence levels considering certain densities, although rainfall distribution in the first weeks after sowing are suggested to play an important role.

Seed bank depletion was determined using two seed burial and retrieval methods, namely (i) mesh seed bags filled with sand and *Striga* seeds and (ii) soil inoculation and sampling after which seeds were extracted by means of wet sieving and flotation. Fate of exhumed seeds was assessed by a seed press test in which empty seeds were considered to have germinated.

Seed germination contributed most to seed bank depletion under a variety of vegetative cover types including host crops, non-host trap crops, intercrops of hosts and trap crops and weedy fallow. The soil sampling method and the seed bag burial method yielded similar percentages of seed bank depletion and treatment effects showed similar trends. Combining data from previous studies on seed production with these data on seed losses indicated that seed bank reduction by suicidal germination would only be achievable if seed production and seed bank replenishment are completely prevented. The results raise questions on the specificity of trap crops and whether differences reported previously in seed bank depletion between trap and host crops are simply caused by the prevention of seed production, rather than increased (suicidal) seed germination in the soil.

With the information obtained in this study and from literature, a population model was constructed and parameterized to explore the long-term effects of management strategies on the *Striga* seed bank through scenario study. The results from this study will aid in assessing and developing promising management strategies for *Striga*.

TA van Mourik, E Weltzien and R Tabo

Activity 3.3.3: Test options to reduce Aflatoxin contamination in groundnut

Milestone: Integrated technologies to minimize aflatoxin contamination in groundnut out scaling trials established in four countries

Aflatoxin is a toxic substance produced by mold fungi (*Aspergillus flavus and A. parasiticus*) that can grow on poorly managed agricultural crops, particularly groundnuts. If eaten in sufficient quantities aflatoxin can cause

serious sicknesses that can lead to liver and several other cancers. Groundnuts for sale and export should be free from aflatoxin. Therefore appropriate crop management is essential at pre-and post harvest times.

ICRISAT and its partners have developed several technologies that can contribute to reducing risks to aflatoxin contamination. These include genetic resistance and integrated crop management practices, agronomic practices, biological control, and biotechnological interventions. A number of these technologies have been tested on-farm with farmers in Mali. ICRISAT has also developed inexpensive quantitative methods for the detection of aflatoxin in groundnut-based products and feed. The ELISA based diagnostic test is reliable, cost effective and easy to carry out. This can help NARS, NGOs, traders and exporters to undertake large scale testing of groundnut-based-foods and feed for aflatoxin.

Resistant/tolerant varieties: Past research has identified and developed groundnut varieties that are tolerant to *Aspergillus flavus* invasion and subsequent Aflatoxin contamination. The first task was to expose these varieties to groundnut farmers through participatory on farm trials/demonstrations. In such trials in the district of Kolokani and Kayes, the main groundnut procuring regions in Mali, low levels of aflatoxin contamination were recorded (Table 3.3). Similar trials/demonstrations have been extended to Niger, Nigeria and Senegal.

Table 3.3. Ranges and means of Aflatoxin content in the kernels (ppb) in tolerant varieties evaluated by 10 farmers in Kolokani during the 2004/2005 cropping season

Variety	Range (ppb)	Mean (ppb)	Pod yield (t ha^{-1})	Haulm yield (t ha^{-1})
ICG 6101	0.22-1.46	0.86	0.82	1.15
ICG 7	0.02-0.96	0.36	0.92	0.89
ICG 6222	0.51-4.27	1.86	0.82	1.13
ICGV 88274	1.64-11.29	5.87	0.72	1.07
ICGV 92093	2.17-12.45	6.71	0.86	1.07
Res check: 55-437	0.06-2.45	1.02	0.93	1.07
Susc Check Fleur 11	70.89-118.18	92.49	0.94	0.93
Local (47-10)	7.96-25.19	16.95	0.87	0.93
SE ±		1.920	0.064	0.066
CV (%)		39	23	20

Integrated management practices: **I**nfection of groundnut pod/kernel by the mold fungi occurs both in pre-and post-harvest conditions. In the pre-harvest conditions, end-of season drought is a major predisposing factor. Technologies to mitigate the effect of drought have been developed. These have been tested in two major groundnut regions of Mali (Kolokani and Kayes). The technologies are: application of lime, crop residues and farmyard manure and their combination. These treatments were applied to a resistant (55-437) and a susceptible (JL 24) variety with farmer participation. Results are presented in Tables 3.4 and 3.5. All treatments, especially, application of lime and farmyard manure significantly reduced aflatoxin contamination, especially in the susceptible variety. On average the application of lime reduced aflatoxin contamination by 84%.

Table 3.4. Aflatoxin content (ppb) in the kernels under various agronomic practices, averaged over 5 farmers in Kolokani, Mali, 2004/2005

Treatment	Variety		Pod yield (t ha^{-1})	
	55-437	JL24	55-437	JL24
Lime 50DAP	1.90	52.34	1.16	1.06
2.5 t ha^{-1} FYM	2.07	64.07	1.27	1.09
2.5 t ha^{-1} Residue	3.28	126.59	1.14	1.03
Lime + Residue	2.76	79.53	1.24	0.96
FYM + Residue	4.20	90.64	1.39	1.18
No treatment	6.21	190.84	1.00	1.07
SE ±	1.22		0.087	

Table 3.5. Aflatoxin content in the grain (ppb) under various agronomic practices, averaged over 5 farmers in Kayes 2004/2005

Treatment	Aflatoxin content (ppb)		Pod yield (t ha^{-1})	
	55-437	JL 24	55-437	JL 24
Lime 50DAP	0.12	4.20	2.208	2.204
2.5 t ha^{-1} FYM	0.26	6.76	2.460	2.468
2.5 t ha^{-1} Residue	0.79	36.71	1.952	2.080
Lime +Residue	0.36	7.36	2.004	2.081
FYM + Residue	0.94	12.10	2.576	2.460
No treatment	2.83	82.32	2.83	82.32
SE ±	1.564		0.082	

Best-bet harvesting and drying technique: Groundnuts need to be harvested at the correct time. Delays in harvesting result in over maturity leading to fungal infections and subsequent aflatoxin contamination.

Poorly dried groundnuts enhance fungal growth and aflatoxin contamination. Good storage with kernel moisture <10% does not permit fungal growth and aflatoxin contamination. Poor curing can induce fungal growth (aflatoxin contamination) and reduce seed quality for consumption, marketing and germination.

Groundnuts that are allowed to dry well immediately after harvesting tend to develop negligible levels of contamination, where as groundnuts left out but covered with haulms and leaves tend to develop alarming levels of aflatoxin contamination (Table 3.6 and 3.7). The most effective control was achieved through immediate removal of pods from the harvested plants, but this has labor constraints at the time when other farm activities are at their peak. There is a need to explore cheap dryers that can be used by farmers during the harvest period.

Table 3.6. Effect of method of drying on aflatoxin contamination (susceptible variety 47-10) averaged over 10 farmers in Kolokani, Mali, 2004/2005

	Drying method		
Name of the farmer	Traditional	Improved	% Reduction
Bagui	17.94	2.22	88
Mory	13.73	1.78	87
Seba	15.93	4.97	69
Demba	14.61	3.89	74
SE ±	1.373		
CV (%)	29%		

Table 3.7. Effect of method of harvesting and drying on aflatoxin contamination (susceptible variety, 47-10) in Kayes, Mali, 2004/2005

	Harvest/drying method		
Name of the farmer	Traditional	Improved	% Reduction
Madou	71.31	20.02	72
Savadogo	60.08	18.01	70
Yaya	58.01	21.53	63
Mamadou	79.52	28.31	64
Coumba	59.62	15.73	74
Djenaba	74.48	27.01	64
Kande	44.86	14.28	68
Seydou	12.32	1.96	84
SE ±	2.999		
CV (%)	43%		

Results indicated that proper handling of groundnut during and after harvest will reduce fungal growth and aflatoxin contamination thereby increasing the marketability of groundnuts and increasing sales and income by local groundnut farmers. There is a need for increased awareness for aflatoxin contamination and health hazards.

BR Ntare, AT Diallo, F Waliyar and O Kodio

Project 4
Producing more and better food from staple cereals (sorghum and millets) and legumes (groundnuts, chickpea and pigeonpea) at lower cost in the eastern and southern African (ESA) SAT through genetic improvement

Output 2.1: Genetically diverse and regionally adapted germplasm and breeding populations *[Increased availability of diverse germplasm sources and breeding materials]*

A large number of varieties have been released (some varieties in many countries) indicating that requests for pure seed will be high. Many varieties released by national authorities have never been multiplied and accessed by farmers. Availability of all classes of good quality seed to stakeholder groups in the seed industry is key to enhancing the impact from crop breeding and also in enhancing agricultural productivity.

In 2005 a total of 344 sorghum and 79 pearl millet lines were planted for purity assessment. Various quantities of seed were also multiplied for different classes of seed for sorghum, pearl millet and finger millet. Indexing of all groundnut varieties and germplasm at ICRISAT Lilongwe was completed. The groundnut breeder seed produced at Malawi is sufficient to produce 169 hectares of Foundation seed.

Nucleus seed of all improved cultivars of pigeonpea and chickpea was produced. Breeder seed of five long-duration and one medium-duration pigeonpea was developed at Kampi ya Mawe Research Station. Similarly, pure seed of three chickpea kabuli genotypes and one desi type was produced at Kabete Research Station. The seed is maintained under short-term storage conditions at ICRISAT-Nairobi.

Advanced lines of short-duration pigeonpea attaining 50% flowering within 65 days were observed at Kiboko Research Station (KRS) in eastern Kenya. In spite of a severe drought, the cultivars flowered and matured within 3 months at Kampi ya Mawe Research Station (KMRS) in eastern Kenya. These early maturity and ratoonability traits should enhance adoption of the new short-duration cultivars by farmers. Medium-duration cultivars that are insensitive to photoperiod were evaluated at Chitedze Research Station [CRS ($13^{o}59'$ S and $33^{o}44'$ E] in Malawi and yielded about 2 t ha^{-1} indicating success in the development of the medium-duration pigeonpea types insensitive to photoperiod and warm temperatures. In Tanzania, ICEAP 00068 having large, cream seed and ratoonability was released (as cultivar 'Tumia'). Two long-duration pigeonpea cultivars (ICEAP 00040 and ICEAP 00020) showed superior (10%) dhal recovery. ICEAP 00020 is still at the pre-release stage but ICEAP 00040 was released previously for commercial production in Malawi (as cultivar 'Kachangu').

The groundnut program centred on development and evaluation of groundnut breeding populations and breeding lines with resistance to foliar diseases. The program maintains breeding populations for groundnut rosette and ELS disease, and gene pyramiding for multiple resistance.

Preliminary sorghum yield evaluation was done on materials that were already in the F_6 and F_7 generations and were planted at Alupe, Kenya. Ten lines were selected for inclusion in regional evaluation trials based on the yield data and other agronomic characteristics such as flowering, agronomic score and midge reaction. Eleven restorer lines selected from crosses between KARI Mtama 1X SC691-14-NIG-FET were identified and advanced based on their short stature and large seed. A total of 15 experimental hybrids were developed and tested at Alupe. On-farm demonstrations were carried out with a number of improved sorghum and finger millet varieties suitable for the drought prone areas of eastern and north eastern Kenya as well as those suitable to the humid Lake Victoria zone. The best performing sorghum lines were IESV92036 and IS8193. Overall, the highest yielding and blast resistant finger millet lines were KNE688, ACC32 and KNE814.

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Team: R Folkertsma , CT Hash and EW Rattunde

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RT Folkertsma, CT Hash, B Jayashree and BIG Haussmann

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The first generation of backcrosses between F_1 and the farmer-preferred varieties (Hugurtay, Hiryray, Ochuti, Tabat, Wad Ahamat, Tiemarifign, and CSM 335) has been performed and BC_1F_1 generated in Kenya, Mali, Sudan, and Eritrea. In Kenya, 210 BC_1F_1 individuals were genotyped using 3 SSR markers: 63 plants were found to be heterozygous for markers in the QTL target regions – 3 plants contained 3 *Striga* QTLs, 13 had 2 *Striga* QTLs, and 47 had 1 *Striga* QTL. Confirmed hybrids were backcrossed to the FPSV recurrent parent Ochuti to produce the BC_1F_1 generation, which is currently being genotyped. In Sudan, 144 Tabat BC_1F_1

samples were genotyped and 28 plants were found to be heterozygous for markers in the QTL target regions – 5 plants contained 4 *Striga* QTLs, 4 contained alleles of 3 QTLs, 5 had 2 QTLs, and 12 had 1 QTL. Capacity for DNA isolation, quantification, PCR optimization, and agarose/PAGE with silver staining has been provided to the NARS in Mali (UB and IER), and Kenya (KARI-Katumani) through training, provision of equipment, and technical backstopping.

A gene flow experiment was set up to determine the distance of pollen flow using male-sterile lines as receptors, thus eliminating the need of large scale PCR genotyping. The flowering dates of the materials from Sudan, Kenya, and the male-sterile lines were determined and the first pollen dispersal experiment carried out using Ochuti and N 13 pollen donor and recipient, respectively. The experiment is currently being repeated at another site. A field study has been conducted on sorghum indigenous knowledge, farming system, stakeholders' perceptions and seed supply systems in Eritrea, Kenya, and Sudan. A report containing the findings of status of the seed sub-sector in Eritrea, Kenya, and Sudan has been compiled.

D Kiambi

Sorghum stay-green introgression lines derived by marker-assisted backcrossing provided to African national programs in locally adapted, farmer-preferred genetic backgrounds (2008).

Due to lack of adequate SSR marker polymorphism between stay-green drought tolerance donor parent E 36-1 and the two recurrent parents (S35 and IRAT 204), and reduction in the amount of funding provided by granting agencies for activities related to this milestone, we have focused the marker-assisted backcrossing program for the sorghum stay-green trait on donor B35 only.

During 2004 marker-assisted backcrossing of genomic regions associated with the stay-green component of terminal drought tolerance of donor parent B35 was advanced two generations in the genetic background of elite recurrent parent Kapaala = ICSV 111. By the end of 2004, BC_4F_2 seed had been produced for families expected to segregate for four of the six target stay-green QTL in this genetic background (for which suitable BC_3F_1 families were available in 2003) and for the remaining two target QTL, seed had been produced for BC_3F_2 families.

During the first half of 2005, BC_3F_2 and BC_4F_2 plants homozygous for donor parent alleles at SSR marker loci flanking each of six individual stay-green loci (*i.e.*, single-QTL introgression lines for stay-green QTLs *stg*A, *stg*B, *stg*1, *stg*2, *stg*3 and *stg*4) from donor parent B35 in the genetic background of ICSV 111 (released in Ghana as 'Kapaala') and its sub-selection S 35, were identified and their selfed seed harvested. These included ten BC_4F_2 plants expected to be homozygous for *stg*A, three BC_4F_2 plants expected to be homozygous for *stg*B, six $BC3F_2$ plants expected to be homozygous for *stg*1, ten BC_3F_2 plants expected to be homozygous for *stg*2, two BC_3F_2 and seven BC_4F_2 plants expected to be homozygous for *stg*3, and four BC_4F_2 plants expected to be homozygous for *stg*4. The corresponding BC_4F_3 and BC_3F_3 families, along with seed samples of their recurrent and donor parents, were sent to the national sorghum breeding program in Ghana (SARI) in June 2005 in time for initial rainy season seed increase for agronomic and farmer-participatory evaluation under the Water for Food Challenge Program. Further, these progenies were advanced a further generation by selfing at ICRISAT-Patancheru during the 2005-06 post-rainy season to produce seed required for future assessment of their drought tolerance, agronomic performance and grain quality. Marker data generation in 2005 required to identify the putative QTL introgression homozygotes was performed as part of the M.Sc. thesis research program of Mr. Sripathi Venkateswararao. In late 2005 he submitted a thesis to Acharya N.G. Ranga Agricultural University (ANGRAU) India, and this was defended successfully in early 2006.

Similarly, marker-assisted backcross introgression of stay-green QTLs from donor parent B35 into the genetic background of recurrent parent IRAT 204 advanced two generations during 2005, with the marker data generation required being performed as part of the M.Sc. thesis research program of Mr. Sripathi Venkateswararao.

We hosted a Visiting Scientist from the Indian national program (Dr Madhasudhana Rao from the National Research Center for Sorghum) for 3 months to develop marker genotype data for assessing opportunities of MABC to improve stay-green drought tolerance of elite and landrace materials of interest to Indian sorghum breeders, and another from the Universidad Autonoma de Neuvo Leon (Dr F Zavala G) for 1 month to learn how to conduct SSR-MAS for the stay-green trait.

CT Hash

Activity 2.1.1: Maintenance breeding: verify and maintain purity of released varieties, elite lines and hybrid seed parents

Milestone: At least 5 sorghum and 3 pearl millet popular varieties purified by 2007

A large number of varieties have been released and some have multiple country releases indicating that requests for pure clean stocks of seed will be large. Many varieties released by national authorities have not been multiplied and accessed by farmers. Availability of all classes of quality seed to stakeholder groups in the seed industry is key to enhancing the impact from crop breeding and also in enhancing agricultural productivity. As a CGIAR institute we have a role in ensuring that the initial seed materials that flow from ICRISAT breeding programs is pure and of the best quality as this will be used by other stakeholders in the seed chain. In 2005 all breeding materials were catalogued and computerized in the Bulawayo genebank and various sources for each variety were identified. These sources were sampled and planted out–a variety in four rows and one row for hybrid parental lines (Table 2.1). Observations were made for each source to assess uniformity, trueness to type and other distinctive traits. The best three sources for each were identified and would be used for further multiplication of the variety/parental line. The highly contaminated sources were discarded from the store.

Sorghum

- Most varieties had a uniformity level of 80%
- 18% of varieties had 100% uniformity in all sources
- Seed of Macia that some seed companies and NGOs had was absolutely not true to type
- Chokwe (ICSV112) will need to be reintroduced
- Sorghum A lines shedding pollen were insignificant
- Off types in sorghum A lines were also very small
- Each of the sorghum released varieties had a source that was 95-100% pure

Table 2.1. Maintenance breeding for sorghum and millet in ESA

Crop	Description	No of lines planted
Sorghum	Released varieties	30
Sorghum	Regional trial entries	78
Sorghum	A & B lines	163
Sorghum	Restorer lines	73
Pearl millet	Released varieties	20
Pearl millet	A & B lines	59

Pearl Millet A &B lines and varieties

- A number of pearl millet A lines were shedding pollen
- 18 pearl millet lines need to be reintroduced
- Eventually all inbred lines for pearl millet need to be reintroduced
- The released pearl millet varieties are reasonably uniform

Conclusion:

- The activity helped identify true sources of varieties and parental that were uniform and true to type materials
- Schedule for seed production need to ensure that we always have a reliable source of clean quality seed
- The clean stocks will be planted to provide nucleus seed for each variety

MA Mgonja and S Kudita

Milestone: At least 5 kg each nuclear seed of elite groundnut varieties of different maturity groups produced each year; at least 200kg of the most promising varieties under on-farm produced for large scale promotional testing with collaborators annually and at least one t of each variety under commercial production in ESA produced each year in support of commercial production

The groundnut improvement program based at Lilongwe, Malawi has developed a number of varieties that are released by NARS for farmer use in their respective countries. We also have the mandate to maintain the

breeder seed of released varieties. We completed indexing of all varieties and germplasm at ICRISAT Lilongwe. Two elite varieties ICGS 31 (released in Botswana) and ICGV-SM 99537 were completely missing. We requested and secured fresh seed of ICGS 31 from NARS Botswana and shared the same with the breeder from Patancheru for safe keeping at the ICRISAT genebank. Similarly we requested and obtained fresh seed of ICGV-SM 99537 from Zimbabwe. This seed is now being multiplied at ICRISAT-Bulawayo. A sample will be made available for ICRISAT-Malawi and the Genebank in India after harvest.

ES Monyo

Milestone: Nucleus seed of all improved cultivars of sorghum, pearl millet, groundnuts, chickpea and pigeonpea produced and made available to NARS and other partners on request

Nucleus and breeder seed production is an essential component of the breeding program. Seed production at the regional centers facilitates the conduct of regional collaborative trials as well as on-farm adaptive trials with NARS and NGOs. The breeder seed production activity is also an important activity necessary to get adequate quantities of seed for foundation and certified seed production by NGOs and the private sector. This targets needs of collaborating partners for implementation of promotion and scaling out of improved varieties

Sorghum: Nucleus seed of 27 cultivars were multiplied and 135 kg of seed obtained. Breeder seed of 8 varieties was also multiplied and about 200 kg of seed is available. Foundation seed quantities available are: Macia (625 kg); 5DX160 (150 kg); Gadam El Hamam (200 kg); Kari Mtama 1 (400 kg); and ZSV3 (120 kg).

Pearl millet: Nucleus seed of 12 varieties (107 kg) and breeder seed for 6 varieties (160 kg) was produced. Foundation seed for three varieties, namely Okashana 1 (1550 kg); PMV 3 (437 kg); ICMV221 (120 kg) are available.

Finger millet: A total of 60 kg of nucleus seed of 8 finger millet varieties and breeder/ foundation seed for U15 (60 kg); P224 (50 kg) and ACC32 (49 kg) is available.

Fodder finger millet: A total of 40 kg of finger millet seed suitable for use as fodder was multiplied and availed to collaborators in University of Warwick in UK.

MA Mgonja, S Kudita and E Muange

Groundnuts: The following breeder seed of popular varieties was produced to sustain commercial production to meet market demand; ICGV-SM 90704 (6.2 t), ICG 12991 (5.1 t), JL 24 (1.6 t), CG 7 (0.68 t), plus various quantities of nucleus seed ranging from 1–39 kg for 45 varieties in Advanced, Elite and released status.

ES Monyo

Pigeonpeas and chickpeas: Limited quantities of pigeonpea breeder seed of 6 varieties (ICEAP 00040, 00053, 00850, 00576-1, 00911 and 00557) were multiplied at Kampi ya Mawe research station. The plants were raised in an environment isolated from insect pollinators in order to maintain genetic purity.

Using single plant selections, nuclear seed of 13 (eight *desi* + five *kabuli*) chickpea genotypes was constituted at Kabete field station. These genotypes included ICCV 95423, ICCV 95423, ICCV 96329, ICCV 96329, ICCV 92318, ICCV 97105 and ICCV 97105. Further multiplication of the seed will be conducted at ICRISAT-Nairobi. At least three of these genotypes are earmarked for release by the national programs in ESA.

SN Silim and E Gwata

Activity 2.1.2: Develop and evaluate trait specific populations and breeding lines for adaptation to specific environments, pest and diseases and for product market/food safety requirements

Milestone: At least 1 short-duration, 2 medium-duration and 2 long-duration pigeonpea varieties with end user quality traits identified or released

Short-duration: Advanced lines of short-duration pigeonpea attaining 50% flowering within 65 days were observed at Kiboko research station (KRS) in eastern Kenya. The genotype ICEAP 00994 obtained 1.4 t ha^{-1} while the commercial cultivar ICPL 87091 obtained 40% less grain yield. In spite of the severe drought during the season, the cultivars flowered and matured within 3 months at Kampi yaMawe research station (KMRS) in eastern Kenya. However, the yield was relatively low, but farmers in the area usually harvest a second (ratoon) crop. The yield from the ratoon crop can be as high as 80% of the first crop. These qualities (early maturity, ratoonability) should enhance adoption of the new short-duration cultivars by farmers.

Medium-duration cultivars: Medium-duration cultivars that are insensitive to photoperiod were evaluated at Chitedze Research Station [(CRS) 13°59' S and 33°44' E] in Malawi. The highest grain yield (1.95 t ha^{-1}) was obtained for the experimental cultivar ICEAP 01160/15 compared to 0.36 t ha^{-1} of the commercial cultivar ICEAP 00068. However, the grain size (11.55 g 100 $seed^{-1}$) of this genotype was relatively small. Large grains (16.43 g) were observed for the experimental cultivar ICEAP 01172/6 which also obtained three fold higher grain yield than ICEAP 00068. In spite of the severe terminal drought experience at CRS, these improved genotypes flowered and matured before the end of May. The results obtained from this field evaluation indicated that medium-duration pigeonpea types insensitive to photoperiod and warm temperatures were developed successfully. The experimental cultivars are likely to attain higher grain yield under optimum moisture conditions.

Long-duration cultivars for *Dhal* processing: The end-user qualities for pigeonpea that are important in our region include large, cream seed and high *dhal* recovery. Compared with the industry average, two cultivars (ICEAP 00040 and ICEAP 00020) showed superior (10%) *dhal* recovery. ICEP 00020 is still at the pre-release stage but ICEAP 00040 was released previously for commercial production in Malawi (as cultivar 'Kachangu'). This germplasm will be useful in the region in pigeonpea breeding programs aimed at improving both traits.

SN Silim and E Gwata

Activity 2.1.3: Develop new improved varieties, hybrids, seed parents with end-user preferred plant and grain traits for food security and markets

Milestone: Breeding populations with resistance to ELS, Rosette, LLS and Rust developed and stability of resistance assessed

Development and evaluation of breeding populations, with resistance to foliar diseases: Activities centred around development and evaluation of groundnut breeding populations and breeding lines with resistance to foliar diseases. The program maintains populations for rosette disease and the aphid vector, ELS disease and gene pyramiding for multiple resistances.

Breeding populations with resistance to the Groundnut Rosette Virus (GRV): Breeding material and populations developed for the purpose of studying the inheritance of resistance were screened for superior progeny rows using the infector row technique.

From the 65 progeny rows, 79 single plants were selected from 42 progenies based on field observations but only 26 combined GRV resistance with yield potential. From the inheritance study, all ten progenies in the study were field selected, but upon adding the yield criteria, only one progeny will be advanced.

Breeding populations with resistance to the Groundnut Rosette Disease (GRD): A nursery consisting of 17 GRD resistant F_6 progenies was evaluated using the infector row technique. A two tier strategy was used for selection from the segregating populations. First selection was done in the field where observed superior plants were tagged and harvested. The final selection was done in the laboratory using yield as a second criterion. After combining field disease resistance with yield performance, only 6 plants from 4 progenies were finally selected for further advance.

Breeding populations with resistance to the Groundnut Aphid Vector: This study involved two nurseries in F_5 generation each with 46 progeny populations. Out of 111 plants carrying vector resistance, only 29 progeny populations combined yield potential with high levels of vector resistance. The fact that a higher proportion of resistant high yielding progenies were identified from this nursery speaks of the importance of incorporating your resistance source in an already known high yielding adaptable genotype.

Breeding populations with resistance to early leaf spot (ELS) disease: Breeding populations for ELS disease resistance ranged from F_2, F_3, and F_4 populations. The greatest problem we have with ELS is that resistance in early maturity background is very rare. Secondly, resistance is associated with poor quality including poor kernel reticulation and loss of palatability.

The F_2 nursery consisted of 443 segregating lines from germplasm crosses. Of these, 131 were selected with good levels of field resistance, but when this was complimented with yield potential only 23 were retained. The second population consisted of 101 F_3 progenies, of lines combining ELS resistance with confectionary traits – particularly seed size. Only 21 progenies combining ELS resistance and confectionary traits were retained. The

third nursery consisted of 650 F_4 progeny rows, of which 343 were identified for field resistance but only 61 retained at the final selection. The fourth nursery included 42 F_4 progeny rows of interspecific hybrids obtained through embryo rescue at ICRISAT-Patancheru. We could not find any evidence of superior performance, partly because of the very limited amount of seed per row (5-8). All 42 progenies were advanced to give opportunity for observing more plants for their reaction to ELS.

ES Monyo

Breeding populations for multiple resistance - gene pyramiding: These populations were developed for multiple stress resistance to improve yield stability of varieties. Deployment of multiple resistances is important to guard against genetic vulnerability and possible breakdown of resistances.

Key findings from the nurseries above include:

- Excellent lines combining resistance to aphid and ELS (179), aphid and rust (27) in F_7. These were promoted to progeny trials.
- Excellent breeding lines (Spanish) in with resistance to the aphid vector and GRV (49), ELS and Rosette (51), GRV and dormancy (45), and rosette and dormancy (25) identified in F_6 generation.
- Excellent Spanish single plant selections (8) resistant to aphid identified in the F_5 generation.
- Populations with a combination Aphid and ELS, Aphid and Rosette, and Aphid and GRV currently in F_3, F_4 and F_5 advanced to further generations.

High levels of ELS resistance from the populations above trace their origin to gene blocks from wild *Arachis spp.* most often associated with low yield potential. Continuous backcrossing and recovery of resistance in high yielding genetic background is therefore essential.

ES Monyo

Milestone: Wide range of varieties, hybrids and seed parents with market traits evaluated

Sorghum:

- Preliminary sorghum yield trial with 64 entries: These materials (F_6 and F_7 generations) were planted in Alupe, Kenya. Most of these materials flowered in about 68 days and had a mean yield of 3.471 t ha^{-1}. Ten lines were selected for inclusion in regional evaluation trials based on the yield data and other agronomic characteristics like flowering, agronomic score and midge reaction.

- Eleven restorer lines selected from crosses between KARI Mtama 1X SC691-14-NIG-FET selected for their short stature and large seed were identified and can be used to make high-yielding short-statured hybrids.

- A total of 15 experimental hybrids were developed at Alupe during the 2005 long rains season for evaluation along with other hybrids developed earlier.

- On-farm demonstrations: A number of improved varieties suitable for the drought prone areas of eastern and north eastern Kenya as well as those suitable to the humid Lake Victoria zone have been identified. Some of these varieties are still in the pre- release status; partly because there has been limited effort in the promotion of these improved elite sorghum varieties. In some cases there is need to submit on-farm performance data which is also a pre-requisite for their release. On-farm demonstrations were therefore established in five districts in Kenya in collaboration with partners from Kenya Agricultural Research Institute (KARI). The mother and baby trial design was used. The mother trial had 8 entries (6 improved, 1 commercial and a local variety) replicated four times. Fifteen baby trials were planted in the surrounding environments. The baby trials were not replicated. Each farmer with a baby trial had 2 improved varieties, 1 commercial and a local check. Field days were also conducted and attended by more than 500 farmers and policy makers. *Striga* was also recognized as a very serious biotic stress for sorghum that needed judicious and collaborative efforts through integrating genetic (conventional and molecular techniques) with crop and soil fertility management technologies.

Sorghum:

Data analysis for the mother trials has been completed and the best performers are Seredo, IESV92036 and IS8193 across the four sites. At least 50% of the data from the baby trials has been received and analysis has

been completed. The mean yield for the Alupe sites was 4.76 t ha^{-1}; Siaya 3.94 t ha^{-1} and Migori 0.914 t ha^{-1}. Data from Siaya are given in Table 2.2.

Table 2.2. Sorghum on-farm evaluation trials across 22 farmers in Siaya district of Kenya, 2005

Ranked from highest to lowest yielder	Name	Grain yield (t ha^{-1})	Shoot fly damage 1. None 2. Low 3. Average 4. High	Overall disease score 1. None 2. Low 3. Average 4. High	Days to 75% maturity 1. Early 2. Medium 3. Late
1	Seredo	4.540	1.6	1.9	1.0
2	Local check	4.400	1.9	2.0	2.5
3	IESV 92036	3.980	2.0	1.8	1.3
4	IS 8193	3.840	1.8	1.9	1.8
5	IESV 92022/1-SH	3.840	1.9	2.1	2.5
6	Wagita	3.800	2.0	1.9	2.5
7	IES 93042-SH	3.750	2.0	1.9	2.8
8	IESV 92055/S-SH	3.370	1.9	2.6	1.8
	Grand mean	3.940	1.9	2.0	2.0
	SE	0.989	0.20	0.46	0.52

Finger millet: Promotion of blast resistant finger millet in Western and Nyanza provinces of Kenya: With the exception of Sudan, finger millet is the most important millet in the ESA region. Finger millet is increasing in importance due to the unique nutritional components and also its marketing potential. Finger millet blast is an important disease that can cause high yield losses. Previous work characterized blast pathogen populations and also identified some finger millet resistant varieties. Promotion and demonstration of potential of improved and blast resistant varieties was carried out in Kenya and Uganda using four farmers' varieties (Acc14, 29, 32 and 44) and ICRISAT germplasm lines KNE620, 629, 688, 814 and 1149. Yields across the mother and baby trials in the two countries ranged from 1.06 t ha^{-1} to 1.85 t ha^{-1}. Overall, the highest yielding materials were KNE688; ACC32, KNE814. These were also the most blast resistant varieties.

MA Mgonja, P Kaloki and J Kibuka

Output 2.2: Regionally adapted parental lines, varieties and hybrids with market traits developed for SAT regions

ICRISAT and NARS scientists in ESA recognize the advantage in pursuing regionalized crop improvement strategies and using the **lead NARS** approach to improve efficiency in crop improvement. A number of sorghum and pearl millet Lead NARS project have been developed and are under implementation. The groundnut programs have defined priority research issues for non-confectionary groundnut for the short-duration production domain (which is by far the largest) and confectionary groundnuts for the medium- to long- duration domains.

A regional sorghum hybrid evaluation indicated the highest yields to be from SDSH93025, SDSH98008, DC75; ZWSH 1 and SDSH90003 with yields from 2.14 to 2.69 t ha-1. Sorghum cultivar evaluation including some high-yielding, large grain sorghum varieties and restorers acquired from ICRISAT India and tested in Zimbabwe for three years were evaluated in Nampula province of Mozambique to identify new materials. The line SDSL98018 the highest yielding, followed by some of the new high-yielding large grain variety SP993527. In Alupe, Kenya, the IESV92036-SH had the highest yield (5.15 t ha^{-1}), followed by SDSH93025 (4.643) and Wagita 3.789 t ha^{-1}. There are some varieties which are very competitive and can yield as well as the hybrids. The NARS breeders in Kenya have been encouraged to release the most promising varieties for farmers' access.

The ICRISAT ESA program has a range of groundnut lines of varying maturity range and grading qualities. In the short duration category the focus is on incorporation of dormancy, large seeded, yield, quality and adaptation, and selecting for drought resistance with a focus on the confectionary market. At Chitedze Research Station, several short-duration high-yielding varieties–some with yields ≥ 200% over the standard check JL 24–were identified. The best five varieties were ICGV 94536, ICGV-SM 99598, ICGV-SM 98543, ICGV-SM 00528 and ICGV-SM 98544 with kernel yields ranging from 2.5 to 2.7 t ha^{-1} compared to the control JL 24 (1.6

t ha^{-1}). In the elite short-duration drought tolerance and dormancy groundnut variety trial, the best five entries in terms of yield performance to be ICGV-SM 95598, ICGV-SM 98519, ICGV-SM 86021, ICGV-SM 95599, and ICGV 94139 with yields ranging from 2.6 to 3.1 t ha^{-1} compared to 1.6 t ha^{-1} to 2.1 t ha^{-1} for the controls. Among the best five lines with resistance to Early Leaf Spots are ICGV-SM 93541, ICGV-SM 96678, ICGV-SM 95714, ICGV-SM 95695, ICGV-SM 95740. The top five rosette resistant entries were ICGV-SM 99543, ICGV-SM 99566, ICGV-SM 01513, ICGV-SM 01514 and ICGV-SM 01506.

The Malawi national program has released a rosette resistant variety (ICGV-SM 99568) in August 2005.

There is a growing demand for chickpeas particularly kabuli types on the international markets. In ESA, both desi and kabuli types are popular with smallholder farmers because of their ability to utilize residual soil moisture. A range of chickpea genotypes fitting various agro-ecological and farming systems were evaluated. The results indicated very good grain yield potential for chickpeas in ESA. However, on-farm yields are generally low due to the broadcasting habit instead of row planting and also due to lack of adequate inputs such as pesticides.

Activity 2.2.1: Delineate agro-ecological zones; identify themes and recommendation domains for regionalized breeding approaches

Milestones: At least two lead NARS/ projects led by NARS on behalf of the region

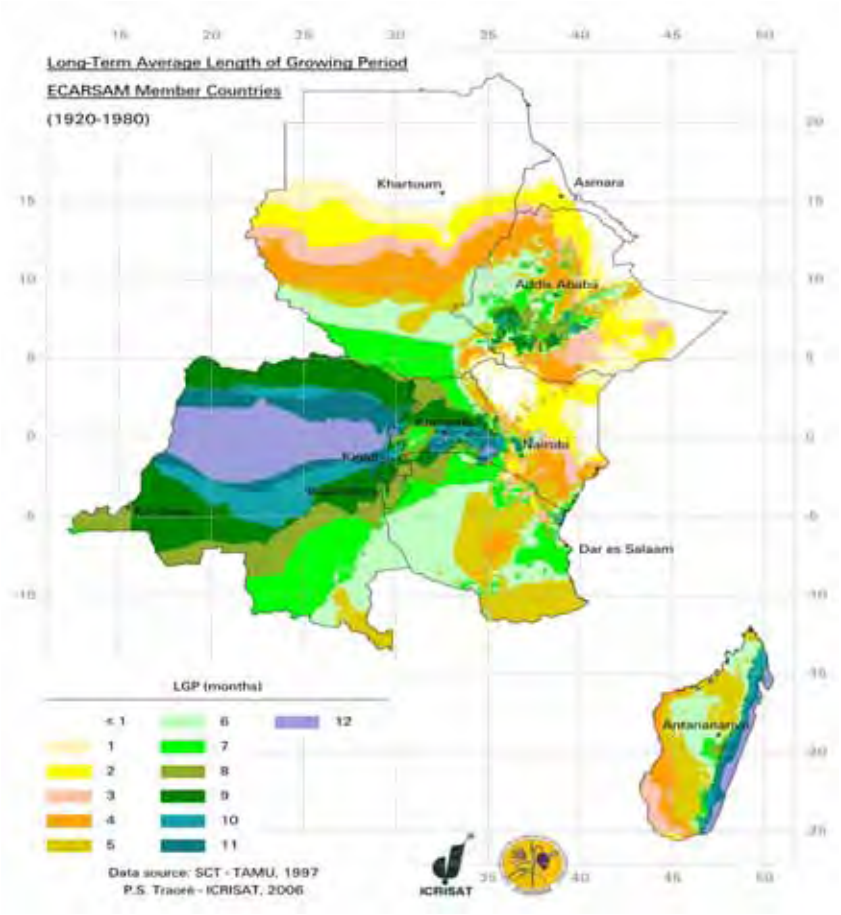

Figure 2.1. Long-term average length of growing period in Eastern and Central Africa

NARS breeders in ECA have provided climatic information for sorghum, pearl millet and finger millet growing conditions to allow AEZ and GIS mapping to determine production and recommendation domains for these crops in the region (Fig. 2.1). NARS have also provided similar information for the test sites to facilitate mapping of sites that fall within the same AEZ for initial determination of test sites for regional testing of germplasm. The ESA region in collaboration with NARS scientists in the respective Sub Regional Organization

recognize the added advantage in pursuing regionalized crop improvement strategies and using the **lead NARS** approach to improve efficiency in crop improvement. Through the ECASARM network/ASARECA and in collaboration with ICRISAT, a number of Lead NARS projects have been developed and are under implementation.

These include:

- Fighting *Striga*: Resistant genes deployed to boost sorghum productivity: Lead NARS-Eritrea
- Increasing sorghum utilization and marketability through variety identification and food products diversification: Lead NARS-Tanzania
- Integrated *Striga* management for improved sorghum production in ECA: Lead NARS- Tanzania
- Photoperiod sensitive sorghum improvement: Lead NARS-Zambia

MA Mgonja, S Traore and B Mitaru

Activity 2.2.2: Set priorities for a regionalized breeding approach for ICRISAT mandate crops

Milestone: Recommendation domains identified for at least two crops by 2007; Strategic plan and priorities set

Research priorities for legumes and oilseed crops for ASARECA region: The Association for Strengthening Agricultural Research in Eastern and Central Africa (ASARECA) has identified legumes and oilseed crops as high priority crops with potential for moving a large number of farmers out of poverty if improved research technologies were easily made available to farmers. ASARECA thus requested ICRISAT to assist in the development of a regional strategy for legumes and oilseed crops. The major oilseed crop in the region is groundnuts. As a member of the team for the development of the regional strategy, we identified four priority areas for groundnuts; yield and adaptation, foliar disease resistance, aflatoxin management and drought. Based on these, we defined priority research issues for non-confectionary groundnuts for the short-duration recommendation domain (which is by far the largest), and confectionary groundnuts for the medium- to long-duration domains. Since the non-confectionary groundnuts the major use is oil extraction, recommendation was therefore for research focus on oil content and quality bearing the following:

- Target development of short-duration cultivars which must incorporate genetic resistance to fresh seed dormancy for short-duration environments.
- For the medium- to long-duration environments need to target yield improvement, quality and confectionary types.
- For all environments improvement of groundnuts for foliar disease (Rosette, ELS, LLS and RUST) resistances and genetic diversity.

The fastest expanding intake of groundnuts is in the confectionary markets – mainly governed by consumers' preferences for taste, seed color, size, shelf life of marketed products and industrial specifications for particular size and shape. For this reason, research priorities should consider the following:

- Low oil content to avoid product rancidity.
- Low Oleic/Linoleic acid (O/L) ratio which favor long shelf life and taste.
- Consumers prefer large seeded nuts hence the challenge to develop large seeded nuts for short duration environments.
- Research efforts are needed to reduce aflatoxin.

For all types, there is need to improve genetic diversity through interspecific/wide hybridization since cultivated groundnuts lies on a narrow genetic base.

ES Monyo

Activity 2.2.3: Assist NARS to pursue regionalized sorghum and pearl millet improvement strategies

Milestone: A range of maturity groups, growth habits and quality characteristics for sorghum, pearl millet, groundnut, chickpea and pigeonpea identified to fit into various agro ecologies and farming systems (2004-2007)

The national programs in the Eastern and Southern Africa are increasingly facing declining human and financial resources for research and development work. In view of this they have had limited capacity to contribute

sorghum and pearl millet lines to the regional evaluation programs. It has therefore been necessary to complement the NARS by providing semi finished materials for preliminary evaluation to identify materials adapted to individual countries before recommending them into regional trials.

Improvement of the long season photoperiod sensitive sorghum: The long season photoperiod sensitive sorghums have over the years contributed significantly to the food security of the long season areas of central and northern Mozambique. These areas can be considered as relatively high potential because of the amount of rainfall that is received. From preliminary observations in Mozambique, the genetic diversity of the long season sorghums has declined. The activity therefore looked at the possibility of identifying long season sorghums with higher yield potentials across the targeted areas especially from Tanzania for re-introduction. Further analyses are to provide information on the use of these for developing populations and restorer lines that can be used in hybrid program. A total of 33 long season photoperiod sensitive sorghums from Tanzania, Mozambique and Zambia and two improved long season varieties (Pato and Sima) were evaluated in Nampula (Mozambique) and Naliendele (Tanzania). The number of days to 50% flowering ranged from 136 to 160 days. The improved varieties flowered in 102-110 days. The same materials were also characterized at the University of Zambia by a student form Tanzania. The materials have been assigned the Bulawayo genebank numbers and will be conserved for future crop improvement work including developing experimental hybrids.

MA Mgonja and S Kudita

Regional sorghum hybrid evaluation: A regional hybrid trial was conducted in Kenya, Uganda and Zimbabwe. The hybrids originated from the ICRISAT Zimbabwe and Kenya as well as from Pioneer Seed Company. In Kenya these were tested in Alupe and Kiboko and at ICRISAT Matopos in Zimbabwe. Yields in Alupe were higher (mean of 2.78 t ha^{-1}) than in Kiboko (trial mean of 0.81 t ha^{-1}). The yields in Alupe ranged from 1.31 t ha^{-1} to 5.15 t ha^{-1} with IESH22008 yielding the highest. In Kiboko the highest yields of 2.22 t ha^{-1} was obtained from ZWSH1. The combined analysis indicated the highest yields to be from SDSH93025; SDSH98008; DC75; ZWSH 1 and SDSH90003 Table 2.3). Hybrids from Pioneer were extra early and yields were low.

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Table 2.3. Combined analysis – sorghum hybrid evaluation in Kiboko and Alupe, Kenya, 2005

Ranked according to yield	Variety	Grain yield t ha^{-1}	Days to 50% heading	Days to 50% flowering	Plant height (cm)	Head length (cm)	Exsertion
1	SDSH 93025	2.69	55	60	157.4	34.0	9.0
2	SDSH 98008	2.53	52	57	163.1	34.9	9.2
3	DC 75	2.38	53	58	136.4	31.2	8.5
4	ZWSH 1	2.38	61	63	163.5	28.2	9.6
5	SDSH 93021	2.13	54	59	149.3	34.2	4.1
6	SDSH 98012	1.81	63	67	142.6	30.1	7.6
7	SDSH 90003	1.76	56	60	135.5	30.0	6.1
8	SDSH 94001	1.73	56	61	147.1	28.5	9.3
9	SDSH 98022	1.64	57	61	142.9	30.7	5.1
10	SDSH 94003	1.64	54	59	144.2	31.8	6.5
11	GV 3017	1.61	50	55	124.4	22.4	4.1
12	SDSH 94011	1.53	56	61	138.2	27.9	11.2
13	SDSH 48	1.51	59	64	152.5	29.3	9.2
14	SDSH 98006	1.19	63	68	152.6	32.1	8.2
15	NS 5511	1.18	57	61	105.5	25.2	4.6
16	BSH 1	1.09	59	63	138.3	27.6	7.7
17	SDSH 98001	1.00	66	72	149.3	28.9	9.9

Ranked according to yield	Variety	Grain yield t ha^{-1}	Days to 50% heading	Days to 50% flowering	Plant height (cm)	Head length (cm)	Exsertion
18	SDSH 409	0.89	55	59	120.4	29.1	8.5
19	SDSH 93024	0.35	55	60	143.0	31.4	7.5
20	S4-8601	0.25	54	60	138.0	32.0	7.6
Grand mean		1.63	56.7	61.5	142.4	29.9	7.7
SE		0.54	2.31	2.32	10.38	2.38	2.80
CV (%)		33.1	4.1	3.8	7.3	8.0	36.5

Sorghum cultivar evaluation in Mozambique and Kenya: Twenty five cultivars including some high yielding, large grain sorghum varieties and restorers acquired from ICRISAT India (and tested in Zimbabwe for three years) were evaluated in Nampula province of Mozambique to identify new materials for in-country evaluation. The line SDSL98018 was the highest yielding, followed by some of the new high yielding bold grain variety SP993527 (Table 2.4). Macia and Sima were not among the top ten varieties. Sima was the lowest performer and it was also the latest to flower, an indication that it did not fit the length of growing period for the site.

Table 2.4. Sorghum cultivar evaluation in Nampula Mozambique, 2005

Ranked from highest to lowest yielder	Name	Grain mass (t ha^{-1})	Initial plant stand	Days to 50% flowering	Plant height (cm)
1	SDSL98018	2.01	27.35	65.66	122.2
2	SP993527	1.57	21	69.46	111.5
3	ICSV89094	1.47	27.82	69.79	145
4	SDSL98021	1.47	19.76	69.61	216.6
5	SP993529	1.40	22.37	67.25	126.1
6	SP993442-1	1.37	23.43	67.38	114.1
7	ICSV89106	1.35	19.26	70.6	129.4
8	ICSV89117	1.13	23.86	69.66	142
9	ICSV93041	1.13	18.61	67.06	146.8
10	ICSR161	1.11	16.33	67.99	116.5
11	SP993371-3	1.04	25.93	73.17	125.7
12	SP993532	0.93	28.83	65.29	126.6
13	Macia	0.91	21.33	66.33	122.2
14	SP993522-1	0.86	16.8	72.33	113.5
15	SP995214	0.67	14.39	65.73	96
16	ICSV500	0.65	22.75	68.59	146.2
17	SP993515	0.62	9.21	72.33	97.3
18	SP993314	0.61	20.12	67.68	145.7
19	ICSV547	0.56	13.01	68.93	148.9
20	SP993520-1	0.54	6.93	71.74	111.6
21	ICSV91010	0.48	14.75	68.33	98.8
22	ICSV492	0.42	22.45	67.75	127.9

Ranked from highest to lowest yielder	Name	Grain mass (t ha^{-1})	Initial plant stand	Days to 50% flowering	Plant height (cm)
23	SP993531	0.40	3.37	68.1	93
24	ICSV382	0.32	8.77	69.21	147.6
25	Sima	0.12	9.24	74.68	165.4
Grand mean		0.926	18.31	68.99	129.5
SE		0.5061	8.393	4.22	18.27
CV (%)		54.7	45.8	6.1	14.1

In Alupe, Kenya, 25 cultivars were evaluated for performance to identify varieties and hybrids that could be incorporated into the regional trials. The IESV92036-SH had the highest yield 5.15 t ha^{-1} followed by SDSH93025 (4.64 t ha^{-1}) and Wagita 3.79 t ha^{-1} (Table 2.5). The hybrids SDSH93021, SDSH 98008, and varieties Seredo and IS8193 had yields above 3.2 t ha^{-1}. There are some varieties which are very competitive and can yield as well as the hybrids. There is need to identify hybrids that are much more superior to the hybrids if hybrids are to be taken up by the farmers and the industry.

Table 2.5. Sorghum cultivar evaluation in Alupe, Kenya, 2005

Ranked from highest to lowest yielder	Name	Grain yield (t ha^{-1})	Establishment count/plot	Days to 50% flowering	Plant height (cm)
1	IESV 92036-SH	5.15	50	66	224.2
2	SDSH 93025	4.64	47	59	205.2
3	WAGITA	3.79	46	67	196.1
4	ASINGE	3.60	49	71	250.5
5	SDSH 98008	3.58	48	58	199.8
6	SDSH 93021	3.54	30	59	194.0
7	IS 8193	3.36	47	66	175.0
8	SEREDO	3.26	46	61	163.6
9	SDSH 98012	3.10	45	65	179.7
10	DC 75	3.03	40	56	170.8
11	ZWSH 1	2.92	36	59	206.7
12	IESV 93042-SH	2.80	44	69	197.1
13	SDSH 94011	2.74	29	60	168.9
14	SDSH 94003	2.73	31	58	169.7
15	SDSH 98022	2.59	39	60	195.5
16	SDSH 90003	2.54	22	57	159.9
17	SDSH 94001	2.44	29	58	183.2
18	GV 3017	2.08	43	54	144.5
19	NS 5511	2.06	47	58	139.0
20	SDSH 98006	1.98	22	67	192.2
21	SDSH 48	1.81	31	59	183.0
22	BSH 1	1.55	47	59	183.2
23	SDSH 93024	1.48	23	63	170.6
24	SDSH 409	1.43	7	57	144.9
25	SDSH 98001	1.31	37	69	204.9
Grand mean		2.781	37.4	60.4	184.1
SE		0.673	5.20	1.70	12.11
CV (%)		24.20	13.90	2.80	6.60

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Milestone: 5–10 high-yielding short-duration groundnut varieties with confectionary market traits identified for promotion to on-farm testing in ESA region

Short-duration groundnut varieties with improved grain and grading qualities for the domestic and export markets: The ICRISAT ESA program maintains groundnut lines of varying maturity range and seed qualities. In the short-duration category our focus is on incorporation of dormancy, large seeded, high yield and adaptation, and selecting for drought resistance with a focus on the confectionary market. Rainfall at Chitedze Research Station ended pre-maturely (673.3 mm). The crop was therefore subjected to severe end of season drought. At Ngabu Research Station the main drought screening center, effective rain was less than 400 mm, also ending first week of February just when the nurseries were beginning to flower. The nurseries were thus severely stressed. No variety survived the heat (temperatures easily reaches 40 deg.) and long extended drought at Ngabu Research Station in Southern Malawi. This location provides for an excellent natural environment for drought screening. Zero yields were recorded for most entries. However, entries that at least survived death were identified for further observation. Ngabu Research Station provides for an excellent heat and drought screening natural environment for ESA region.

At Chitedze Research Station, several short-duration high-yielding varieties – some with yields ≥ 200% over the standard check JL 24 were identified.

Advanced adaptability and quality groundnut variety trial (Spanish): Some lines outperformed those of the control varieties (JL 24, Nyanda and Sellie). The top five varieties (out of 33 test lines) were ICGV-SM 03552, ICGV-SM 03573, ICGV-SM 03564, ICGV-SM 03559, ICGV-SM 03560 (yields 2794–3043 kg ha^{-1} vs 1299 for JL 24) under low disease pressure. All entries showed susceptibility to ELS with scores ranging from 6–9. Sellie had the highest ELS score (9). Though susceptible to ELS, yields were not affected much because of short-duration.

Elite aflatoxin resistance groundnut variety trial (Spanish): Thirteen short-duration genotypes were evaluated and compared to three controls, J11, JL 24 and Nyanda. ICGV 95456 performed exceptionally well in pod yield (3222 kg ha^{-1}), haulms weight (1819 kg ha^{-1}), kernel yield (2028 kg ha^{-1}), 100 seed mass (43.05 g), compared to J11 the resistant check (pod yield 1611 kg ha^{-1}, haulms yield 832 kg ha^{-1}, and kernel yield 1192 kg ha^{-1}). Results from germination tests conducted prior to harvesting showed 0% germination indicating fresh seed dormancy which is not normally the case with most Spanish varieties. ELS incidence was severe and none of the materials tested showed resistance to the disease. Nyanda was the worst hit with an ELS score of 9.0.

Elite very short-duration groundnut variety trial (Spanish): Fourteen short-duration test lines and two controls which included JL 24 and ICG 12991 were evaluated. With the exception of two lines, all the varieties evaluated performed better that the controls. The best five varieties were ICGV 94536, ICGV-SM 99598, ICGV-SM 98543, ICGV-SM 00528 and ICGV-SM 98544 with kernel yields ranging from 2464–2681 kg ha^{-1} compared to the control JL 24 (1616 kg ha^{-1}). In all the entries, ELS incidence was severe.

Elite short-duration drought tolerance and dormancy groundnut variety trial: Twenty breeding lines were evaluated and compared to 4 controls (ICG 12991, Malimba, Nyanda and JL 24). The best five entries in terms of yield performance were ICGV-SM 95598, ICGV-SM 98519, ICGV-SM 86021, ICGV-SM 95599, and ICGV 94139 with kernel yields ranging from 2633-3080 kg ha^{-1} (compared to 1662-2101 kg ha-1 for the controls). ICGV-SM 95599 and ICGV-SM 98519 also showed good levels of resistance to ELS (score 5.0).

ES Monyo

Milestone: At least 2 groundnut varieties with resistance to rosette, ELS, LLS and rust incorporating market desired traits identified for release in ESA

Evaluation and promotion of varieties for food security and market traits: Various breeding lines and varieties were evaluated at Chitedze Research Station, Malawi for rosette and ELS under high and low disease pressure. High rosette and ELS disease pressure was induced using the infector row technique.

Elite rosette resistance groundnut variety trial (Spanish): Twenty three elite breeding lines and 2 controls were evaluated under high rosette disease pressure. The top five entries were ICGV-SM 99543, ICGV-SM 99566, ICGV-SM 01513, ICGV-SM 01514 and ICGV-SM 01506 with kernel yields ranging from 929–1183 kg ha^{-1}. The susceptible control JL 24 produced just 202 kg ha^{-1}, while the resistant control ICG 12991 managed 942 kg ha^{-1}. Rosette disease incidences for the resistant varieties ranged from 0–2.2% while incidence on JL 24

was 81%. The resistant control ICG 12991 had 1.5% incidence. Unfortunately all materials were susceptible to ELS.

The Malawi national program released a new rosette resistant line ICGV-SM 99568 in August 2005. As can be seen from the performance results above, better higher yielding lines are in the pipeline ready for release in the next 2-3 years.

Elite early leaf spots resistance groundnut variety trial: Fourteen elite breeding lines were evaluated under high ELS disease pressure. Among the best five lines were ICGV-SM 93541, ICGV-SM 96678, ICGV-SM 95714, ICGV-SM 95695, ICGV-SM 95740 with kernel yields ranging from 705–947 kg ha^{-1}, compared to the best resistant control Valencia R2 (600 kg ha^{-1}) and the susceptible check JL24 (428 kg ha^{-1}). We therefore have elite germplasm in the pipeline that are ≥25% superior to the current best.

ES Monyo

Milestone: Evaluate a range of chickpea genotypes fitting various agro-ecological and farming systems

Adaptation of chickpeas: There is a growing demand for chickpeas particularly *kabuli* types on the international markets. In ESA, both *desi* and *kabuli* types are popular with smallholder farmers partly because of their ability to utilize residual moisture. Two field trials of chickpeas were conducted in Kenya and Mozambique. In Kenya both *desi* and *kabuli* types flowered with 60 days. The *desi* type cultivar ICCV 00108 attained the highest grain yield (3.5 t ha^{-1}) while ICCV 00302 achieved the highest grain yield among the *kabuli* types.

Similarly in Mozambique, ICCV 97128 (*desi* type) attained the highest grain yield (4.3 t ha^{-1}) which was 25% higher than the mean grain yield (3.5 t ha^{-1}) of the trial. The highest grain yield among the *kabuli* types was 3.6 t ha^{-1}. The *kabuli* cultivar ICCV 92318 attained the largest grain size (100 grain weight = 42.3 g). The results of the two trials indicated very good grain yield potential for chickpeas in ESA as represented by the two locations. ICRISAT-Nairobi with partners in the Ethiopia national program conducted on-farm technology demonstration [under the using five pilot learning sites (Ude, Hidi, Qurqura, Dire and Godino)] located in Ada district (Fig. 2.6). In addition, smallholder chickpea farmers in the ESA region generally lack adequate inputs such as pesticides.

SN Silim and E Gwata

Table 2.6. Agronomic performance of chickpea experimental cultivars during the 2005 cropping season at Kabete Research Station, Kenya

Cultivar	Kabuli/Desi	Days to 50% flower	Seed weight (g 100 $seed^{-1}$)	Grain yield (t ha^{-1})
ICCV 00108	D	63	26.9	3.5
ICCV 97125	D	65	23.8	3.1
ICCV 97107	D	64	22.3	2.8
ICCV 97201	D	62	25.5	2.7
ICCV 97110	D	62	23.3	2.7
Ngara local	D	63	19.9	2.5
ICCV 97031	D	64	24.0	2.4
ICCV 97114	D	69	27.4	2.4
ICCV 97126	D	65	25.7	2.4
ICCV 97128	D	67	24.1	2.4
ICCV 00302	K	59	29.3	2.3
ICCV 92311	K	59	31.4	2.2
ICCV 97306	K	57	35.6	2.0
ICCV 97206	D	70	25.8	2.0
ICCV 97406	K	67	23.9	1.9
ICCV 97115	D	69	21.4	1.9
ICCV 92944	D	61	26.2	1.9

Cultivar	Days to Kabuli/Desi	Seed weight 50% flower	Grain yield (g 100 seed^{-1})	(t ha^{-1})
ICCV 00402	K	59	27.7	1.9
ICCV 95311	K	54	36.5	1.9
ICCV 97033	D	64	23.8	1.9
ICCV 00104	D	61	30.4	1.9
ICCV 97105	D	63	24.6	1.8
ICCV 96329	K	54	33.7	1.8
ICCV 00305	K	58	26.9	1.6
ICCV 95423	K	63	35.3	1.4
ICCV 92318	K	54	33.3	1.3
Mean	-	62.2±2.5	27.3±2.1	2.2±0.8
CV (%)	-	4.0	7.6	37.0

Output 2.3: Participatory methods and technologies for crop improvement and IPM developed and tested

ICRISAT, in collaboration with the Tanzania NARS, provided seed of improved long-duration pigeonpea cultivars for on-farm trials in the northern region particularly in Babati district, Tanzania. On-farm variety evaluation and farmer participation resulted in wide adoption of ICEAP 00040 and ICEAP 00053 in the district.

Activity 2.3.1: Evaluate and promote through participatory methods, new improved pigeonpea, sorghum and pearl millet varieties, hybrids and seed parents, and groundnut varieties with resistance to major pests and diseases

Milestone: IPM options in field, and plant products efficacy in storage pests of pigeonpea, sorghum and millets evaluated (2007)

Farmer participation and technology dissemination: ICRISAT in collaboration with the Selian Agricultural Research Institute (SARI) and Ilonga Agricultural Research Institute (Tanzania) distributed seed of improved long-duration pigeonpea cultivars for on-farm trials in the northern region, particularly in Babati district. The varieties were evaluated on-farm through farmer-participatory methods. On-farm meetings between ICRISAT scientists and pigeonpea farmers were conducted in the Babati district. ICEAP 00040 and ICEAP 00053 have been adopted widely in the district partly because of their acceptable agronomic, market qualities and resistance to *fusarium* wilt disease.

SN Silim and E Gwata

Output 2.4: Technical backstopping provided to regional networks and projects

The information on the regionally released sorghum cultivars in the SADC region has been published, providing description of the varieties and breeding history and characteristics. More than 200 copies were distributed to NARS partners in ESA. We contributed in the SADC-SSSN meetings to finalize the protocol for regional variety testing and registration. Adequate seed quantities have been multiplied for sorghum varieties Sima, Macia and ICSV112 as well as for two pearl millet varieties Okashana1 and PMV3 to meet the requirements on seed quantities prior to regional variety registration.

A template for a web-based seed catalog to support regional variety registration in the Southern Africa Development Community (SADC), the East African Community (EAC), and in West Africa (a joint initiative involving the West African Economic and Monetary Union (WAEMU), the Economic Community of West African States (ECOWAS), and the Institut du Sahel (INSAH)/Comité Inter-Etat de la Lutte Contre la Secheresse au Sahel (CILSS) was designed. This catalog will contain information generated from Distinctness, Uniformity, Stability (DUS) tests and results obtained from Value for Commercial and Use (VCU) tests needed for regional registration.

A Foundation Seed Unit in Mozambique – Unidade de Sementes Basica (USEBA) –addresses the lack of availability of publicly developed varieties. In 2005 a detailed business plan was drawn up for USEBA to become self-sustaining. Based on estimated demand for foundation seed it was showed that full-cost recovery was possible after three years. The Mozambique and Malawi examples are being used to learn about how the seed supply constraint limiting adoption of public-sector developed varieties can be overcome, and to apply

these lessons in the development of more sustainable seed supply systems that are urgently required in many countries of sub-Saharan Africa and indeed elsewhere. The harmonization of seed policies is being pursued independently in three sub-regions of sub-Saharan Africa (SSA) under different auspices, and with funding support from different donors.

Technical backstopping and implementation strategies, capacity building, and information were provided to regional networks: ECARSAM, SMINET SADC –FANR and SSSN.

Activity 2.4.1: Provide technical backstopping and implementation strategies, capacity building, and information to regional networks: *ECARSAM, SADC–FANR, SSSN*

Milestone: By 2007, at least 3 sorghum (Macia, ICSV112 and Sima) and 2 pearl millet (Okashana1 and PMV3) varieties with regional adaptation and multiple release status published in the Regional Variety catalogue

The information on the regionally adapted sorghum and pearl millet in the Southern Africa Development Community (SADC) region has been published, providing description of the varieties and breeding history and characteristics. More than 200 copies were distributed to NARS partners in ESA. We participated in the SADC Seed Security Network (SSSN) meeting deliberating on finalizing the protocol for regional variety testing and registration. The protocol will be submitted to the policy makers in the SADC region for ratification. Adequate seed quantities have been multiplied for sorghum varieties Sima, Macia and ICSV112 as well as for two pearl millet varieties Okashana1 and PMV3 to meet the requirements on seed quantities prior to regional variety registration.

MA Mgonja and S Kudita

Seed catalog: A template for a web-based seed catalog to support regional variety registration in the Southern Africa Development Community (SADC), the East African Community (EAC), and in West Africa (a joint initiative involving the West African Economic and Monetary Union (WAEMU), the Economic Community of West African States (ECOWAS), and the Institut du Sahel (INSAH)/Comité Inter-Etat de la Lutte Contre la Secheresse au Sahel (CILSS) was designed. This catalog will contain information generated from Distinctness, Uniformity, Stability (DUS) tests and results obtained from Value for Commercial Use (VCU) tests needed for regional registration. It is also planned to include maps indicating zones of adaptation of specific varieties based on a common set of mega environments that is sufficiently flexible to cater for the broad range of crops that are being considered for regional registration in each of the three regions.

Catalog management will be the responsibility of the designated authority in each region, and will be done through a web-based management content system only accessible by the designated authority with administration privileges. The catalog will list both commercial and public-sector developed varieties; and information on availability of commercial seed and of basic and foundation seed of public-sector developed varieties produced by Foundation Seed Enterprises (FSEs) will be allowed for a fee to offset the costs of maintaining the seed catalogs.

RB Jones

Foundation Seed Enterprises: **I**n 2003 ICRISAT was asked to establish a Foundation Seed Unit in Mozambique – Unidade de Sementes Basica (USEBA) – to address the lack of availability of public-sector developed varieties. This unit was established as a project under ICRISAT management drawing upon lessons from the seed revolving fund previously established in Malawi for the dissemination of improved groundnut and pigeonpea varieties developed by ICRISAT's regional groundnut and pigeonpea breeding programs, which has achieved considerable success in raising adoption levels of improved varieties. In 2005 a detailed business plan was drawn up for USEBA to determine the likelihood of this unit becoming self-sustaining and based on estimated demand for foundation seed showed that after three years full-cost recovery was possible. Detailed costings were developed for procurement of seed processing equipment and for its operation and management that were then presented to the Mozambique Government and accepted.

The Mozambique and Malawi examples are being used to learn about how the seed supply constraint limiting adoption of public-sector developed varieties can be overcome, and to apply these lessons in the development of more sustainable seed supply systems that are urgently required in many countries of sub-Saharan Africa and indeed elsewhere.

RB Jones

Harmonization of seed policies*:* The harmonization of seed policies is being pursued independently in three sub-regions of sub-Saharan Africa (SSA) under different auspices, and with funding support from different donors. The regions, countries, regional economic communities (RECs), and institutions involved in these initiatives are listed in Table 2.7.

A four-day workshop was jointly organized by the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) and Iowa State University (ISU) with support from the "Agricultural Policy Harmonization Project (APHP)" and the program for the Sustainable Commercialization of Seed Systems in sub-Saharan Africa (SCOSA) to review the status of seed harmonization efforts in the three sub-regions of SSA, and then to develop detailed work plans.

The synthesis of the seed policy situation is that all three regions are working on the development of a regional variety testing and release system and common seed certification standards (both field and laboratory) of selected crops. The SADC and EAC regions are in addition pursuing quarantine pest lists based on available scientific knowledge and WAEMU would like to initiate this effort. With regard to Plant Variety Protection (PVP), the EAC region is working toward the development of PVP laws for Uganda and Tanzania, while SADC is interested in developing draft PVP laws for their member countries. The states of West Africa have already drafted a model PVP law intended for regional use and this is expected to be adopted by member states in the near future. With respect to accreditation, the EAC has initiated discussions on this topic. SADC also appears very interested, while WAEMU needs to have additional consultations, given the limited level of seed industry development in their region.

Table 2.7. Regions, countries, regional economic communities (RECs), and supporting organizations involved in harmonization of seed policies

Region	Southern Africa	Eastern and Central Africa	West Africa
Regional Economic Community	Southern Africa Development Community (SADC)	East African Community (EAC) and the Common Market for Eastern and Southern Africa (COMESA)	West Africa Economic and Monetary Union (WAEMU)
Countries	13 countries: Angola, Botswana, Democratic Republic of Congo, Lesotho, Malawi, Mauritius, Mozambique, Namibia, South Africa, Swaziland, Tanzania, Zambia, and Zimbabwe	3 countries under phase one (1999): Kenya, Tanzania and Uganda (EAC) 7 countries under phase two (2001): Burundi, Eritrea, Ethiopia, Rwanda and Sudan (2003) the Democratic Republic of Congo and Madagascar	8 countries: Benin, Burkina Faso, Cote d'Ivoire, Guinea Bissau, Mali, Niger, Senegal, and Togo
Supporting organization	SADC Seed Security Network (SSSN)	Eastern and Central Africa Program for Agricultural Policy Analysis (ECAPAPA)	The International Fertilizer Development Center (IFDC)

RB Jones

Milestone: Agreement on initiation of regional testing for cereals (sorghum and pearl millet) and legumes (groundnut, pigeonpea and chickpea) (2006)

Sorghum hybrid development has not been an integral part of the sorghum and pearl millet research work in ICRISAT-Nairobi. In southern Africa, however, there has been substantial work of developing and testing of hybrids. The release of hybrids by NARS breeders in SADC has been relatively minimal compared to release of OPVs. Private seed companies have developed and released comparatively more hybrids. Recently, there has been an increasing interest from the NARS and private company breeders to conduct hybrid evaluation. NARS collaborators in Uganda, Zimbabwe and Kenya were provided with a set each of sorghum regional hybrid trials. The model currently in operation in India will be adopted for the ESA region to enhance crop improvement efficiency and also to exploit genetic gains from hybrids. The last regional cultivars evaluations were done during the time of the former network EARSAM in the 1990s. After that there has not been any regular formalized germplasm sharing and comparison of breeding materials developed by different breeding institutions. Based on experiences from the 15 years regional cultivars evaluation in southern Africa, a format has been designed for NARS breeders to provide geographical, climatic and biophysical characteristics of their

test sites for sorghum and pearl millet. The information has been provided to the GIS expert to help map target areas for breeding and to stratify test sites for use variety evaluation in Multi Environment Trials (MET).

MA Mgonja and B Mitaru

Activity 2.4.2: Strengthen linkages and partnerships between projects, activities and networks operational within and beyond the ESA region

Milestone: ECARSAM priorities set

Sorghum and millets are the third most important crops in the region, cultivated over 13 million hectares of land. Productivity of these crops is, however, low because of constraints affecting the production to consumption continuum, and limited resources of the national agricultural research systems (NARS) in the region. The East and Central Africa Regional Sorghum and Millet Network (ECARSAM) was initiated in 2003 as one of the networks of the Association for Strengthening Agricultural Research in East and Central Africa (ASARECA), building on the achievements of the former network, the Eastern Africa Regional Sorghum and Millet Network (EARSAM), which ran from 1982 to 1993. The principal goal of the network is to achieve increased economic growth and improved livelihoods in the Eastern and Central Africa (ECA) while enhancing the quality of the environment.

The national systems in ECA cannot simultaneously address all the research needs hence the need to prioritize the constraints and address them accordingly through networking to achieve efficient utilization of resources. The priority setting document is thus an outcome of several consultations involving participation of stakeholders at different levels. The major constraints were identified and these formed the ECARSAM research themes.

1. low productivity;
2. high post-harvest handling losses;
3. limited processing and utilization;
4. limited market for sorghum and millets;
5. unfavorable terms of credit and policy framework;
6. limited capacity-building and institutional development; and
7. limited knowledge and information exchange.

Researchers in ECARSAM are implementing research and development activities in the seven themes funded through ASARECA competitive grants.

B Mitaru

Milestone: Contribution to the annual ECARSAM steering committee meetings and activities and information sharing 2005

ECARSAM is one of the networks/programs and projects (NPP) in the ASARECA region specifically responsible for sorghum and millet research and development for Eastern and Central Africa covering 10 countries. It was revived in September 2003. The ICRISAT–ESA cereal improvement program network work very closely with ECARSAM stakeholders and the ICRISAT cereal breeder provides technical backstopping to the ECARSAM network. Collaboration has also continued with the SADC–FANR. Some of the backstopping activities for 2005 included:

ICRISAT scientists have provided technical backstopping support to ECARSAM and SADC–FANR as follows:

- Participation in stakeholder and Steering Committee meetings for ECARSAM
- Backstopping support to develop competitive proposals for the ASARECA Competitive Grant System in June 2005
- Reviewing of Concept Notes and Proposals that were to be funded under CGS stream B (ECARSAM) in November 2005
- Participated in a proposal development on diversified uses of sorghum targeting funding by the Volcani Institute in Israel
- Supported in the design and implementation of on-farm research demonstration for promoting improved sorghum and blast resistant finger millet varieties in Kenya and Uganda

- Organized an International Finger Millet Workshop in September 2005 to share results on the promotion of blast resistant finger millet varieties and articulate future finger millet research and funding
- Participation at the ECARSAM stakeholders workshop 8-12 November where the ECARSAM priorities and strategies were finalized and documented
- Assisted the ECARSAM network in developing the Finger Millet proposal on "*Facilitating the promotion of improved and blast resistant finger millet varieties to enhance production*" targeting Kenya and Uganda that was submitted and approved for funding by DFID as a collaborative activity between NRI, ICRISAT and the Kenya and Uganda NARS partners
- Assisted and provided technical backstopping to SADC-FANR in organizing a Task Force that deliberated on identification of Sub Regional Organization - Pilot Learning Sites for the Sub Sahara Africa/FARA Challenge Program
- Germplasm for sorghum and millets has been provided to Sudan, Republic of South Africa, Mozambique, Botswana, Madagascar, Ethiopia, NGOs in Zimbabwe, Tanzania, Kenya and Uganda
- Information on the regionally adapted sorghum and pearl millet in the SADC region has been published, providing description of the varieties and breeding history and characteristics. Adequate seed quantities have been multiplied to meet the requirements on seed quantities prior to regional variety registration
- Attended SADC Seed Security Network (SSSN) meeting deliberating and finalizing the protocol for regional variety registration for submission to the policy makers in the SADC region.

MA Mgonja and B Mitaru

Milestone: Capacity and training workshops

- In collaboration and with support from ECARSAM, 26 NARS scientists from 6 countries of the ECA region participated in a training workshop on "Developing winning proposals and technical writing".
- A Tanzanian student graduated at the University of Zambia (MSc Plant Breeding) having worked on the characterization of the long season sorghum varieties using both morphological and molecular markers.
- Sorghum and Pearl millet Seed Production raining was provided to 12 farmers is Eastern Kenya.
- A training workshop for 35 extension staff, farmers and NGO collaborators was conducted at Matopos, Zimbabwe in November 2005 to enable them in implementation of the on-farm and on-station trials in Zimbabwe. This project takes the Integrated Genetic and Natural Resource Management (IGNRM) approach where improved drought tolerant crop varieties for sorghum, maize and pearl millet are integrated with soil fertility and rainwater management technologies for increased productivity.

MA Mgonja and B Mitaru

Milestone: Establish a management framework for the Challenge Program on Water for Food – Project No.1 (CPWFPN1) on crop varieties, soil fertility and water management

ICRISAT is leading and managing Challenge Program Project no.1 (CPWFPN1) for the Limpopo Basin. The Project takes the approach on "Integrated Genetic and Natural Resource Management (IGNRM)". A number of tasks were accomplished in 2005:

- Completed negotiations between ICRISAT and CPWF and finalized MOUs between ICRISAT and the partners institutions
- Organized the inception workshop for the CPWFPN1 in January 2005 at Polokwane, South Africa
- Established project management team and held planning meetings in March and October 2005
- Facilitated development of workplans and strategies for implementation
- Drafted proceedings for the inception workshop
- Milestones for year 1 achieved and report submitted in December 2005
- Assisted in planning and setting up on farm demonstration that integrate crop varieties, soil fertility and water management techniques in Zimbabwe and Mozambique
- Completed the agro-ecological analysis and stratification of sites and an abstract of the paper is given

Agro-ecological Analysis and Stratification of Research Sites of the Limpopo Catchment for Verification of Crops, Soil Fertility and Rainwater Management Technology Options

ICRISAT is collaborating with CIAT, CIMMYT, IWMI and NARES partners of Zimbabwe, South Africa and Mozambique in the implementation of the CPWFPN1. The project is building on past investments and achievements for developed technologies in the areas of crops, soil fertility and water management. It is on the premise that diversified cereal and legume crops, soil fertility and rainwater management options can be combined to reduce risks and improve productivity. Public and private institutional arrangements that allow for output market linkages can be used to enhance profitability and sustainability of smallholder agriculture in the Limpopo basin. Options for participatory technology dissemination and model-based decision support tools can be deployed to promote the benefits more widely.

Identification of benchmark representative test sites was considered an important factor for accurate and efficient technology verification. Participants in the CPWFPN1 proposed 25 sites from which to choose representative sites. The major problem was how to cover the maximum diversity of agro-ecological environment in the Limpopo catchment. Agro- ecological analysis and stratification of research sites was conducted in order to have a minimum coverage of existing experimental sites that would be representative of farmers' fields.

The climates of 25 existing experimental sites within the Limpopo Basin (Table 2.8) were clustered using FloraMap. The extent of the adaptation range for each cluster and site was determined using Homologue. The soil characteristics, land cover, and population were determined from existing data sets. Local access to major markets was mapped using the CIAT Accessibility Wizard. Protected areas were eliminated from the analysis. The length and reliability of the growing season was mapped for the whole catchment using MarkSim. A water balance model was used to calculate potential growing season for rainfed crops over 100 simulated years and the proportion of failed season was mapped.

The 25 sites were stratified into 5 clusters (Fig. 2.2) and each site was processed to give a map of its climatic influence. An example for cluster 4 is in Figure 2.3.

Each site was carefully chosen to represent the maximum environmental range. The exercise quickly eliminated non-representative sites. A consensus was reached on the favored sites where environmental representativity and research infrastructure were maximized. These results provided an objective basis for selection of a few representative benchmark test sites for crop-soil fertility-water productivity technology and also for wide dissemination of results within and beyond the basin.

MA Mgonja

Table 2.8. Stratification of the proposed test sites for the Challenge Program Project No.1

Site	Latitude	Longitude	Agency[1]	Country[2]	Cluster
Chokwe	-24.53	32.98	CP17	MOZ	1
Mabalane	-23.80	33.60	CP1+17	MOZ	1
Macia	-25.03	33.10	*	MOZ	1
Massingir	-23.80	32.20	CP1+17	MOZ	1
Xai Xai	-25.10	33.50	CP1+17	MOZ	1
Xilembene	-24.60	33.20	CP1+17	MOZ	1
Giyani	-23.33	30.73	LDA	RSA	2
Makulele	-22.86	30.92	LDA	RSA	2
Matibi	-22.08	30.65	*	ZIM	2
Mbahela	-22.81	30.45	LDA	RSA	2
Mopane	-22.60	29.85	*	RSA	2
Mtetengwe	-22.00	30.00	CP17	ZIM	2
Musina	-22.34	30.04	LDA	RSA	2
Filabusi	-20.80	29.30	CP17	ZIM	3
Insiza	-21.42	29.42	*	ZIM	3
Mwenezi	-21.42	30.73	*	ZIM	3
Bochum	-23.30	29.12	LDA	RSA	4

Site	Latitude	Longitude	Agency[1]	Country[2]	Cluster
Burgersfort	-24.62	30.33	MDA	RSA	4
Mafefe	-24.17	30.08	CP Wet	RSA	4
Mashushu	-24.32	29.65	LDA	RSA	4
Nebo	-23.03	29.85	*	RSA	4
Sikororo	-24.20	30.42	CP17	RSA	4
Strydkraal	-24.47	29.74	LDA	RSA	4
Tzaneen	-23.77	30.16	LDA	RSA	4
Spitzkop	-23.77	29.85	LDA	RSA	5

1. The Codes with CP are Challenge Program on Water for Food Project Nos (CPWFPN) – CP1=CPWFPN1(ICRISAT), CP17=CPWFPN17(WaterNet), CP Wet=CPWFPN30(Wetland), LDA=Limpopo Department of Agriculture, MDA=Mpumalanga Department of Agriculture.
2. MOZ=Mozambique, RSA=Republic of South Africa, ZIM=Zimbabwe.

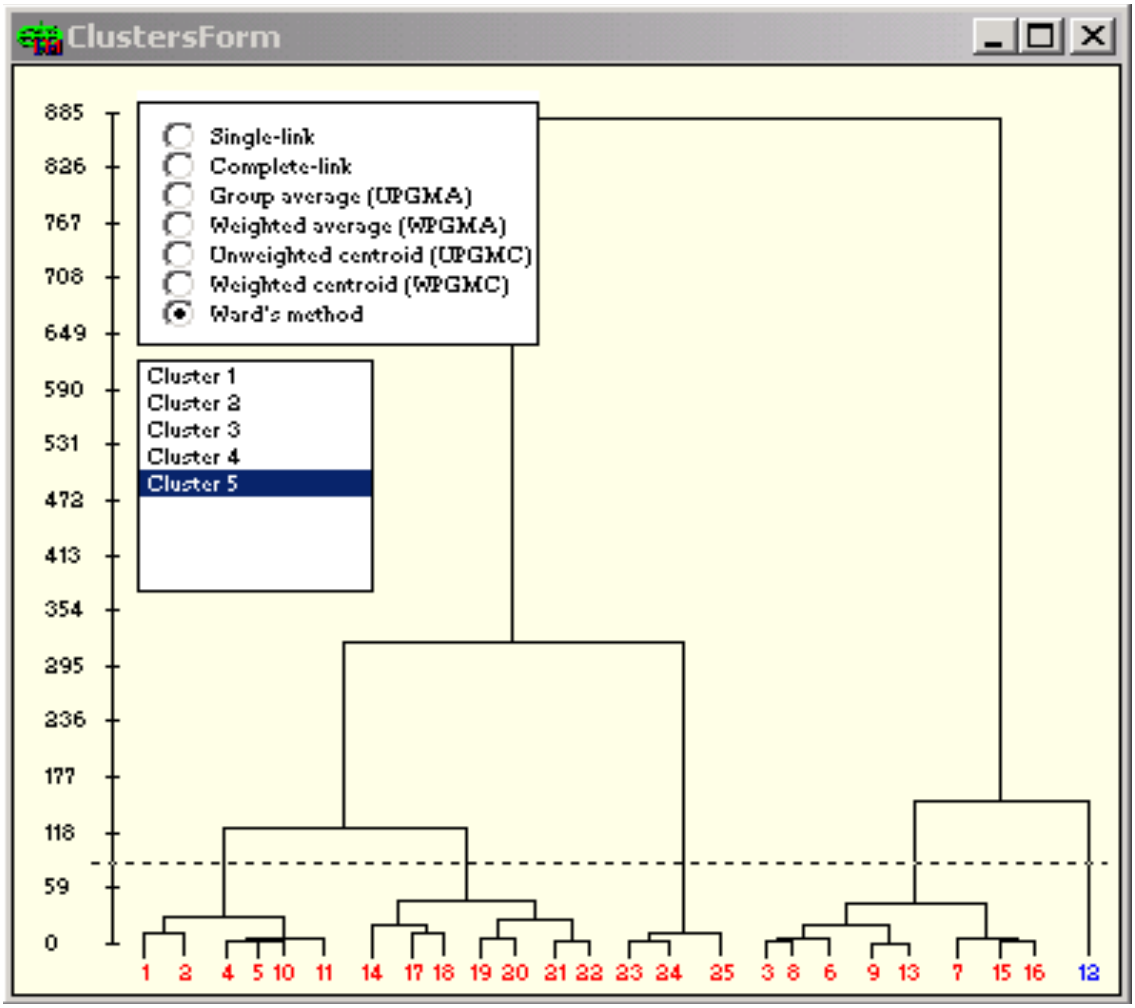


Figure 2.2. Stratification of Limpopo Basin sites into five major groups according to climatic variable

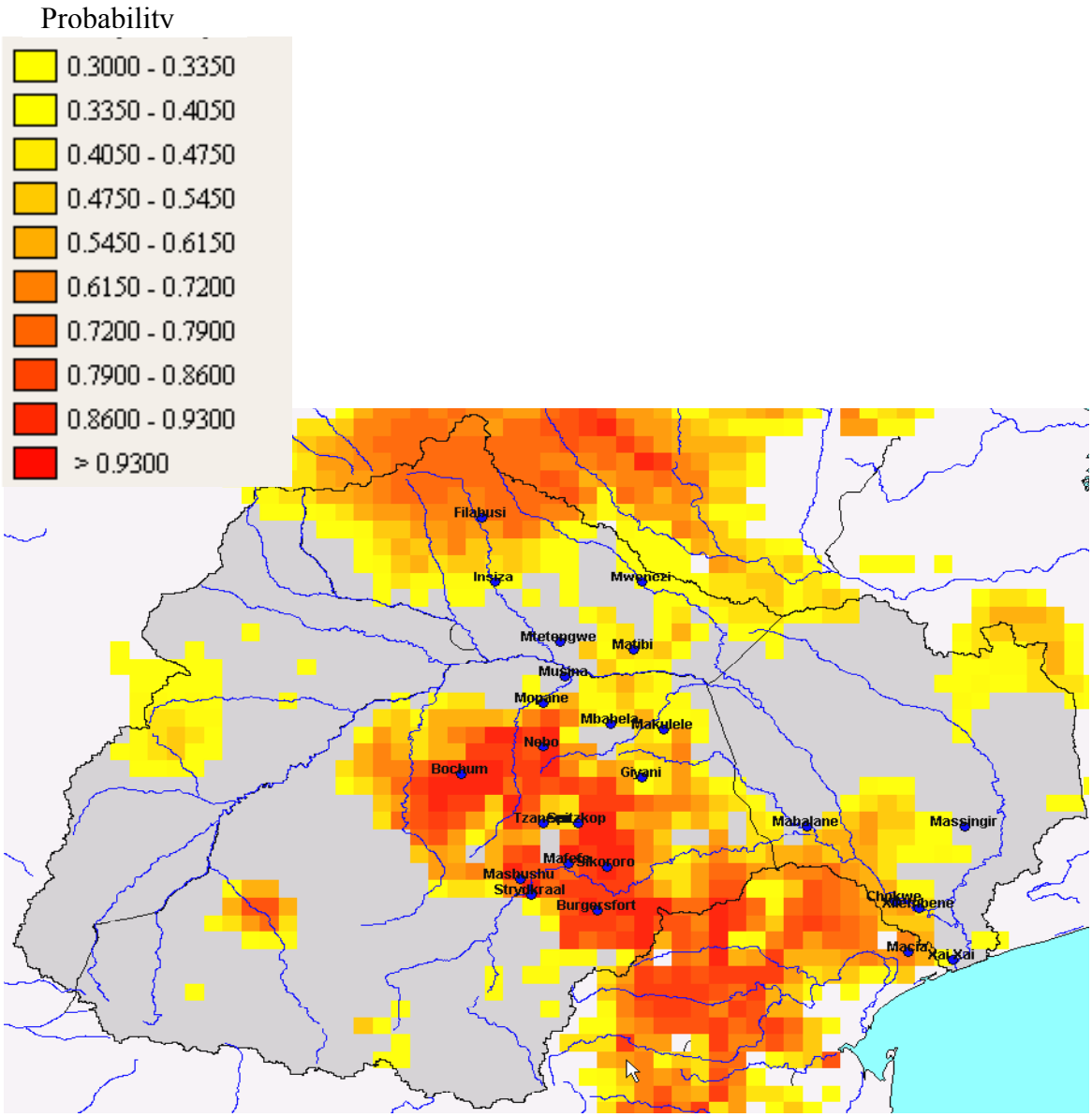


Figure 2.3. Areas of influence for cluster 4 site grouping.

Output 2.5: Regional/sub-regional genebanks maintained

ICRISAT Zimbabwe: Genebank upgrading and germplasm rejuvenation was completed in Kenya. The Tanzania sorghum landraces have been assigned Bulawayo Genebank numbers and preserved. Cluster analysis based on morphological characters revealed two major distinct groups with two subgroups in each. Molecular marker analysis using SSR revealed variations among 41 genotypes, which were grouped into 11 clusters and was able to separate all the landraces. Three sorghum controls, N13, Ochuti and Adiwo each formed independent clusters. Markers Xgap84 and Xtxp320 had high number of alleles than other markers (7 and 8 alleles, respectively). There were no genotype specific makers with the exceptions of Ochuti and N13 controls. Analyses showed significant variations among landraces, clustering was not by area of collection and molecular markers were more efficient.

ICRISAT Kenya: 106 accessions of sorghum and 529 accessions of African finger millet germplasm were characterized. Accessions for 1081 sorghum, 144 accessions of pigeonpea and 36 chickpea have been rejuvenated and seed is being processed for storage.

Pigeonpea germplasm, originally collected from Tanzania, was characterized for morphological and agronomic traits in Kenya (at Kabete and Kampi yaMawe field stations) and Tanzania (at Ilonga research station). The accessions collected from all the four regions in Tanzania generally flowered later when grown in Kenya (at

Kabete and KMRS) compared with Ilonga. The germplasm from the northern region flowered significantly late at all three locations.

Activity 2.5.1: Complete upgrading of regional/sub-regional genebanks

Milestone: Genebank upgrading and germplasm rejuvenation

- Purchased additional equipments required in the genebank and to facilitate operationalization of the installed drier in Nairobi, Kenya
- A medium term storage room for germplasm has been completed at Kiboko, Kenya
- A seed drying room and a drier installed for drying seed for medium storage
- Equipped the Nairobi (Kenya) storage facility with trays for holding storage bottles for ease of tracking the accessions

MA Mgonja and E Muange

Activity 2.5.3: Rejuvenate, characterize, and share information with partners in the region to enhance utilization of the germplasm and usefulness of the genebanks

Milestone: Rejuvenation, collection in identified gaps, and characterization of sorghum, pearl millet and pigeonpea accessions

Bulawayo:

- The Tanzania sorghum landraces have been assigned Bulawayo Genebank numbers and preserved
- 702 sorghum and millet accessions were moved from Bulawayo to Nairobi for rejuvenation in 2006
- Monitored viability of 800 sorghum germplasm accessions. Variability was >85%.

Germplasm sharing: 36 sorghum lines to Mozambique; 86 sorghum and pearl millet entries to Kenya; 32 sorghum breeding lines to Zimbabwe; 66 pearl millet landraces to Zimbabwe.

MA Mgonja and S Kudita

Nairobi:

- 106 accessions of sorghum were characterized and seed was processed for storage
- 529 accessions of African finger millet germplasm were characterized and materials with some good performance will be organized for preliminary yield evaluation
- Accessions for 1081 sorghum, 144 accessions of pigeonpea and 36 chickpea have been rejuvenated and seed is being processed for storage
- 575 accessions of sorghum were rejuvenated and 83% produced seed. Seed multiplication will have to be repeated to get adequate quantities for storage
- Number of accessions preserved: Finger millet (379); chickpea (90); groundnut (19); pearl millet (33); sorghum (1243); pigeonpea (203)
- Number of accessions requiring further rejuvenation due to limited seed amounts has been determined for each crop species

MA Mgonja and E Muange

Milestone: Rejuvenate pigeonpea germplasm for medium-term storage

Rejuvenation of pigeonpea germplasm: A total of 227 pigeonpea genotypes in short-, medium- and long-duration maturity groups were rejuvenated at Kiboko (Kenya). For each genotype, approximately 50 g of seed were harvested and processed for medium-term storage at Kiboko.

Characterization of introduced accessions: Pigeonpea germplasm originally collected from Tanzania was characterized for morphological and agronomic traits in Kenya (at Kabete and Kampi yaMawe Research Stations) and in Tanzania (at Ilonga Research Station). At Kampi yaMawe, the accessions were raised in insect pollinator-free conditions in order to avoid cross-pollination associated with pigeonpea. The germplasm was evaluated in the open field at Kabete and Ilonga.

The accessions collected from all the four regions in Tanzania generally flowered later when grown in Kenya at Kabete and Kampi yaMawe compared with Ilonga (Table 2.9). The germplasm from the northern region flowered significantly late at all three locations. This information is useful, particularly for synchronizing prospective parental lines for hybridization in pigeonpea breeding. The grain yield of the accessions did not

exceed 0.5 t ha^{-1} at both Kiboko and KM. However, at Ilonga, the mean grain yield ranged between 0.5- 2.1 t ha^{-1} (Table 2.9). This could be attributed to the combined effect of genetic and environmental factors. Similarly, the grain size as measured by 100-seed weight, was higher at Kabete than at both KMRS and Ilonga (Table 2.9). Grain color among these accessions was largely white but speckled grains were also observed. The number of branches was highest (11) at Ilonga and lowest (5) at Kampi yaMawe. Similarly, a reduction in the number of pods was also observed at Kampi yaMawe. Likely, this was due to the moisture stress conditions prevailing at the location.

Table 2.9. Duration to 50% flowering (days), seed weight (g 100 seed^{-1}) and grain yield (t ha^{-1}) of pigeonpea germplasm originating from Tanzania evaluated during 2005

	Duration to 50% flower (days)			Seed weight (g 100 seed^{-1})			Grain yield (t ha^{-1})		
Origin	Kabete	KYM[1]	Ilonga	Kabete	KYM	Ilonga	Kabete	KYM	Ilonga
Coastal belt	97-125	100-149	92-105	14.5	13.2	11.8	1.7	0.3	1.7
Eastern region	99-150	104-223	95-118	15.8	14.7	13.4	2.2	0.3	2.1
Southern region	95-139	103-195	90-123	16.2	14.9	14.8	1.6	0.3	1.7
Northern highlands	118-176	127-234	212-160	16.2	15.2	10.0	2.9	0.2	0.5
Medium duration (Check)	106	92	96	16.5	15.4	14.2	1.9	0.3	1.5
Long duration (Check)	91	202	122	15.5	18.7	14.7	1.9	0.8	1.2
Mean±S.E.	103±5	133±16	108±5	15.7±1.1	14.8±2.1	13.0±1.6	2.1±0.6	0.3±0.1	1.5±0.5

1. KYM=Kampi yaMawe.

SN Silim and E Gwata

Project 5/6
Producing more and better food at lower cost of staple hybrid and open-pollinated cereals and legumes in the Asian SAT (sorghum, pearl millet, pigeonpea, chickpea, and groundnut) through genetic improvement

Enhanced resistance to insect pests and diseases via the application of genomics, genetic engineering, wide-hybridization and diagnostics

Insect pests, diseases, and the parasitic weed, *Striga* are serious constraints to increase production, productivity, and utilization of sorghum, pearl millet, chickpea, pigeonpea, and groundnut in the SAT. Crop losses due to these pests have been estimated at over US$ 7.4 billion annually. While *Helicoverpa* control is heavily based on insecticides, chemical control of shoot and panicle feeding insects on cereals is beyond the reach of resource poor farmers in the SAT regions in Asia, Africa, and Latin America. For many diseases and *Striga,* cost effective technologies are yet to be worked out. Current sensitivities about environmental pollution, human health and pest resurgence are a consequence of improper use of synthetic pesticides. Host-plant resistance, natural plant products, bio-pesticides, natural enemies and agronomic practices offer a potentially viable option for integrated pest management (IPM). They are relatively safe for the non-target organisms and human beings. Use of modern biotechnological tools such as marker assisted selection, genetic transformation and wide hybridization for developing crop cultivars with resistance to insect pests and diseases will have a great bearing on future pest management programs. Insect and disease modeling, decision support systems, and remote sensing would contribute to scaling up and dissemination of the IPM technologies. Current research projects in biotechnology, crop improvement and natural resource management focus on the major pests such as pod borers (*Helicoverpa, Maruca*, and *Melanagromyza*), Fusarium wilt and sterility mosaic in pigeonpea; *Helicoverpa*, Wilt and Botrytis gray mold in chickpea; rossette virus, foliar diseases, aflatoxins and leaf miner in groundnut; *Striga*, grain molds, shoot fly, stem borers, midge and head bugs in sorghum; and downy mildew, stem borer and head miner in pearl millet. IPM promotion and capacity building are significant components of research at ICRISAT. The outputs from this research will lead to sustainable management of insect pests and diseases of cereals and legumes based cropping systems, thereby improving the livelihoods of poor people in SAT.

Highlights for 2005

The mapping stem borer/midge resistance in sorghum

- Mapping population based on 296B × IS 18551 (270 lines) was evaluated for resistance to spotted stem borer, *Chilo partellus*, under artificial infestation. Leaf damage rating (DR) varied from 4.0 – 8.0 (1 = <10% leaf area damaged, and 9 = >80% leaf area damaged) and deadheart formation ranged from 5.6 to 90.3% in the mapping population.

- The mapping population, ICSV 745 × PB 15881-3, was evaluated for resistance to sorghum midge, *Stenodiplosis sorghicola*. Midge damage in the mapping population ranged from 2.0 to 9.0 (on a 1 to 9 rating scale, where 1= <10% spikelets with midge damage and 9 = >80% spikelets with midge damage) as compared to 1.0 in the resistance check, ICSV 197, and 7.7 in the susceptible check. Putative QTLs associated with resistance to these insects have been identified.

Mapping shoot-fly resistance in sorghum

- A linkage map based on 296B × IS 18551 has been developed that covers 2165 cM. Composite interval mapping revealed the presence of putative QTLs for all important shoot fly resistance traits, accounting for 6 to 36% of observed phenotypic variances for these traits.

- Three of the four target QTLs from the donor parent IS 18551 were introgressed to the BC_4F_2 generation in the genetic background of shoot fly susceptible elite sorghum hybrid seed parental line, 296B; and all four QTLs were advanced to the BC_4F_2 in the genetic background of shoot fly susceptible elite sorghum hybrid seed parental linem BTx623.

Mapping *Striga* resistance in sorghum

- The non-redundant sorghum EST sequences with SSR motifs were searched against the rice genome sequence in the *Gramene* database (using BLAST). A subset of 600 sorghum EST-SSR loci distributed across regions of the sorghum genome that are syntenic with each of the 12 rice chromosomes (50 each)

was selected. A subset of the E36-1 × N 13-based RIL population was used to start mapping some of the polymorphic EST-SSR markers.

- NARS in the Kenya, Mali, Eritrea and Sudan are being assisted to strengthen *Striga* resistance of farmer-preferred sorghum varieties (FPSVs) through a combination of marker-assisted backcrossing (MAB) and farmer-participatory selection. The stability of inheritance of the transferred *Striga* resistance alleles in the FPSVs, the actual out-crossing rates in selected FPSVs, and pollen flow of these FPSVs is being analyzed in order to develop recommendations for variety maintenance and on-farm seed production.

Mapping downy mildew resistance in pearl millet

- The year 2005 saw the first official release for seed multiplication and cultivation of a product of marker-assisted selection, "HHB 67 Improved" for downy mildew resistance in pearl millet. This pearl millet hybrid was identified for release Haryana, India and was later identified (in July) at the national level for cultivation in the arid zone of northwestern India, where extra-early maturing pearl millet hybrid "HHB 67" is been cultivated on nearly 500,000 ha annually.

- QTL analysis of the mapping population progeny derived from cross 841B-P3 × 863B-P2 detected a major QTL on linkage group 4 accounting for nearly 60% of the observed variation in downy mildew incidence. Two resistance QTLs of small effect were also detected (one each on LG 1 and LG 7)

Mapping *Helicoverpa* resistance in chickpea

- Two RIL mapping populations from the interspecific crosses of *C. arietinum* × *C. reticulatum* (ICC 3137 × IG 72953 and ICC3137 × IG 72953) for mapping of QTLs for resistance to the pod borer, *Helicoverpa armigera* were advanced to F_4. Crosses were also made between ICCC 37 and ICC 506EB for initiating development of an intraspecific mapping population for mapping resistance to *H. armigera.*

Mapping *Ascochyta* blight and *Fusarium* wilt resistance in chickpea

- A linkage map of chickpea with 84 markers (82 SSRs and 2 ESTs) was constructed using one F_2 population of an intraspecific cross of ICCV 04516 (AB resistant) and Pb 7 (AB susceptible). The map length was 724 cM with an average density of 8.62 cM. Three AB resistance QTLs were mapped, 1 on LG 3, and 2 and 3 on LG 4. The QTLs identified on LG 4 were validated in another population.

- A set of 84 RILs derived from the cross C 104 ×WR 315 was genotyped using 73 SSR markers and screened for resistance to Fusarium wilt (FW) race-1. Forty-five markers were assigned to nine linkage groups. The gene for FW race 1 was mapped on linkage group 2. Three SSR markers, TA37, TA 200, and TR2 were closely linked to the resistance gene. The former was located at a distance of 0.4 cM, while the latter two markers were located at a distance of 3.5 cM.

Mechanisms, diversity, stability and inheritance of resistance to *Helicoverpa* in wild relatives of chickpea and pigeonpea

- In pigeonpea, Larval Weights of *Helicoverpa armigera* larvae were <20 mg when reared on diets having pod powder of *C. acutifolius*, *C. sericeus* (ICPW 160*), C. scarabaeoides* (except ICPW 137, ICPW 141, and ICPW 152, *P. scariosa, C. platycarpus*, and *R. aurea)* compared to 53.3 mg on ICPL 87 and 44.0 mg on ICPL 332. Larvae took >25 days to complete the development when reared on the artificial diet impregnated with lyophilized pod powder of *C. acutifolius* (ICPW 2), *C. lineatus* (ICPW 41), *C. sericeus*, *C. scarabaeoides* (except those reared on ICPW 125), *P. scariosa* (ICPW 207), *R. aurea* (ICPW 210), *D. ferruginea*, and *C. platycarpus* (ICPW 68) as compared to 15.7 days on ICPL 87, 23.3 days on ICPL 332, and 12.7 days on artificial control diet.

Introgression of genes conferring resistance to *Helicoverpa* from wild relatives of pigeonpea and chickpea

- Advanced generation diploid (F_1BC_4) and tetraploid (F_4-F_5) derivatives from the cross, *C. platycarpus* × *C. Cajan,* were screened for resistance to *H. armigera* under unprotected field conditions. Damage due to *H. armigera* in the wild parent, *C. platycarpus*, was <1%. Damage in cultivated parent, ICPL 87 was 80%. Damage in diploid derivatives was nearly 20% and in tetraploid derivatives 8%. Nearly 25% pod damage was recorded in the second and third flush in the diploid hybrids.

- In chickpea six lines from the cross ICCV 92318 × IG 72934 suffered nearly 20% pod damage, while one line showed <10% pod damage.

Wide hybridization for resistance to Botrytis gray mold and Ascochyta blight in chickpea

- Over 100 wild *Cicer* accessions were evaluated for résistance to BGM. Three accessions of C. bijugum, 21 accessions of *C. judaicum* and one accession of *C. reticulatum* were found to be resistant to BGM with a mean disease score of < 2.5 (on a 1 to 9 rating scale) as compared to 9.0 in the susceptible check, JG 62

- Wild *Cicer* accessions IG 17159 and IG 73074, which have moderate level of resistance to *Botrytis* gray mold were used in the crossing program. Screening data suggested that the BGM in wild *Cicer* accessions, IG 17159 and IG 73074, was monogenic and recessive.

Wide hybridization for resistance to early/late leaf spots and Aflatoxins in groundnut

- A total of 105 lines of advanced generation interspecific derivatives involving *A. cardenasii*, *A. stenosperma*, *A. kempf-mercadoi* and *A. diogoi* were screened against late leaf spot (LLS) under field conditions. Nineteen lines from *A. hypogaea* × *A. cardenasii*, and one line from *A. hypogaea* × *A. dura* × *A. hypogaea* showed high levels of resistance to LLS (score of 2 (a 1 to 9 scale).

- To generate interspecific amphidiploids, wild *Arachis* from different sections were crossed with *A. hypogaea*. F_1 hybrids were obtained between *A. hypogaea* and *A. diogoi* (section Arachis), BC_1 and BC_2 hybrids were obtained between *A. hypogaea* × *A. chiquitana* (section *Procumbentes*), and two BC_2 hybrid were obtained between A. *hypogaea* × *A. kretschmeri* (section *Procumbentes*).

Transgenic resistance to *Helicoverpa* in pigeonpea and chickpea

- Transgenic pigeonpea plants carrying the *cry1Ac* gene were evaluated during 2004/05 cropping seasons under field conditions. The leaves of three transgenic events reduced the larval weight by 40% after 4 days of infestation as compared to that on the control plants. Pod bioassay of two transgenic events with third-instar *H. armigera* indicated weight gain of 156 to 263% as compared to 357 to 461% in control plants.

- Over 20 transgenic events of chickpea with *cry1Ab* and *cry1A*c genes were bio-assayed for resistance to *H. armigera.* There was approximately 35 to 40% reduction in *H. armigera* larval weight on transgenic chickpea plants as compared to that control plants.

Transgenic groundnut resistance to fungal pathogens and viruses

- For the Indian peanut clump virus (IPCV), contained field trial of the selected 5 events of groundnut transgenic plants having coat protein or replicase genes was carried out at ICRISAT, Patancheru. Two transgenic plants showed initial infection (tested virus positive in ELISA), but in subsequent assays were virus negative.

- For the peanut bud necrosis virus (PBNV), the transgenic groundnut (cv. JL 24) plants of 48 independent events were produced with two vector constructs encoding nucleocapsid protein (NP) gene of PBNV through two different transformation systems. Thirty-five independent events were evaluated for resistance to PBNV by using 100-times dilution of the virus inoculum. Of these, 24 lines showed less incidence (30%) compared to untransformed controls (100%).

- Transgenic groundnut containing rice chitinase gene was evaluated for *A. flavus* resistance by *in vitro* seed inoculation. Seed infection rate in these varied from 0 to 100%. Twenty-two plants that had 0 to ≤10% seed colonization were selected and advanced to T_4 generation, which are under testing.

Diagnostic tools for mycotoxins and viruses

- Exposure of humans and animals to Aflatoxin (AFB1) results from the consumption of contaminated food. AFB1 covalently binds to lysine moiety of serum albumin (AFB1-lys adduct). AFB1-lys adduct has been identified as a useful biomarker to determine the human exposure to aflatoxins, and to assess the exposure risk among various sections of population. An indirect ELISA has been developed for quantitative estimation of AFB1-lys adducts in human serum albumin. ELISA test was validated using reference antibodies by analyzing Hepatitis B virus positive samples.

Characterization of pigeonpea sterility mosaic virus (PPSMV) isolates and development of diagnostic tools

- PPSMV isolates can broadly be grouped as B and P types. The PPSMV isolates within each group have distinct physico-chemical characteristics. The B type isolates can overcome host-plant resistance selected

against P types. The B types occur in northern and southern regions, and P types occur in the central regions of India.

Non-target effects of transgenic crops on beneficial natural enemies of crop pests

- There were no adverse effects of Bt toxins on the fecundity of the *H. armigera* larval parasitoid Campoletis *chloride*. The adverse effects of Bt toxins on *C. chloride* were through early mortality of *H. armigera* larvae, and not through direct effects of the Bt protein no traces of Bt toxin protein were found in the *C. chloride* cocoons and adults with the ELISA test.

Output 1.2.1: Marker-assisted selection for enhanced resistance to insect pests and diseases

Activity 1.2.1.1: Mapping stem borer/midge resistance in sorghum
Team: HC Sharma HC, CT Hash and BVS Reddy

Milestone: Mapping population evaluated for resistance to stem borer

***Reciprocal crosses of RILs for stem borer resistance [ICSV 745 × PB 15520-1 and ICSV 745 × PB 15881-3] advanced*:** During the 2004 *rabi* season, a total of 935 $F_{3:9}$ progenies of stem borer RILs and their bulks were advanced to $F_{3:10}$: 272 $F_{3:9}$ of ICSV 745 (stem borer-susceptible, but midge-resistant and agronomically elite line) × PB 15520 (stem borer-resistant, but midge-susceptible and agronomically elite line), 300 $F_{3:9}$ of PB 15520 × ICSV 745, and 363 $F_{3:9}$ of ICSV 745 × PB 15881-3 (stem borer-resistant, but midge-susceptible and agronomically elite line).

BVS Reddy

***Evaluation of mapping population for resistance to spotted stem borer,* Chilo partellus:** Mapping population based on 296B ×IS 18551 (270 lines) was evaluated for resistance to spotted stem borer, *Chilo partellus,* under artificial infestation in a balanced design with three replications. Data were recorded on leaf feeding, deadheart formation, leaf glossiness, days to panicle initiation, recovery resistance, and agronomic score. Leaf damage rating (DR) varied from 4.0 – 8.0 in the mapping population (1 = <10% leaf area damaged, and 9 = >80% leaf area damaged) as compared to 5.5 in IS 2205 – the resistant check, and 8.5 in ICSV 1 - the susceptible check. IS 18551 suffered a DR of 6.5, while 296B suffered a DR of 8.0. Deadheart formation ranged from 5.6 to 90.3% in the mapping population, 24.1% in the resistant check - IS 2205, and 63.0% in the susceptible check, ICSV 1. The susceptible parent 296B showed 71.3% deadheart formation compared to 25.5% in the resistant parent – IS 18551. Leaf glossiness score varied from 1.0 – 5.0 in the mapping population (1 = highly glossy, and 5 = non-glossy) as compared to 1.0 in IS 2205 – the resistant check, and 5.0 in the susceptible check - ICSV 1. Leaf glossiness score was 5.0 in 296B and 1.0 in IS 18551.

The mapping population, ICSV 745 × PB 15881-3, was evaluated for resistance to sorghum midge, *Stenodiplosis sorghicola* during the 2004/05 post-rainy season. There were three replications in a balanced randomized complete block design (RCBD). Data were recorded on midge damage on a 1 to 9 rating scale (1= <10% spikelets with midge damage, and 9 = >80% spikelets with midge damage). Midge damage in the mapping population ranged from 2.0 to 9.0 compared to 1.0 in the resistance check, ICSV 197, and 7.7 in the susceptible check, Swarna. The resistant (ICSV 745) and the susceptible parents (PB 15881-3) suffered a midge DR of 2.0 and 6.3, respectively.

HC Sharma, BVS Reddy and CT Hash

Activity 1.2.1.2: Mapping shoot-fly resistance in sorghum
Team: CT Hash, BVS Reddy, HC Sharma and S Chandra

Milestone: RILs and backcross populations advanced to facilitate marker-assisted selection for shoot fly resistance (2008)

***Marker-assisted selection for resistance to shoot fly, Atherigona soccata in sorghum*.** Sorghum RILs [252 RILs from the cross 296B (susceptible) × IS 18551 (resistant)] were phenotyped and genotyped to understand the genetics of shoot fly resistance and locate chromosomal regions harboring putative quantitative trait loci (QTLs) for shoot fly resistance and related component traits [PhD thesis, "QTL analysis for shoot fly resistance in sorghum [*Sorghum bicolor* (L.) Moench", by SP Deshpande, submitted in 2005 to Marathwada Agricultural University (MAU)-Parbhani, Maharashtra, India]. After genotyping the RIL population with SSR markers, phenotypic observations of off-type progenies were confirmed. As a result, the effective size of the mapping

population was reduced to 213 RILs, for which phenotypic screening data (from two field screens for shoot fly resistance conducted at MAU-Parbhani) could be combined with the marker data set (111 SSR loci) for linkage analysis and QTL mapping.

The linkage map for this RIL population covers 2165.8 cM (Haldane units), providing at least partial coverage of all ten sorghum linkage groups, but regions with poor marker coverage remain. Composite interval mapping revealed the presence of putative QTLs for all important shoot fly resistance traits, accounting for 6 to 36% of observed phenotypic variances for these traits. One major QTL for glossiness was detected on LG J, accounting for 33% of observed phenotypic variation. Minor QTLs for seedling vigor, deadhearts , and seedling height were also detected. Co-localization of a QTL for trichome density on the upper leaf surface (explaining 20% of observed phenotypic variation) with a QTL for trichome density on the lower leaf surface (explaining 25% of observed phenotypic variation) indicated similarities in genetic control of trichome densities on either side of sorghum leaf blades. The results obtained largely confirmed the previous information based on the BTx623 × IS 18551 mapping population. It would be desirable to fill some of the remaining gaps in the linkage map, if appropriate markers can be identified, and repeat the QTL analyses with all available shoot fly resistance screening data sets for the 213-entry subset of the (296B × IS 18551)-based sorghum RIL population.

QTL mapping of shoot fly resistance, based on two phenotypic screens conducted at ICRISAT-Patancheru of the sorghum RIL mapping population based on the cross 296B × IS 18551, which had been genotyped previously, was also undertaken [PhD thesis, "Genetic diversity analysis, QTL mapping and marker-assisted selection for shoot fly resistance in sorghum [*Sorghum bicolor* (L.) Moench" by SP Mehtre, submitted in 2006 to MAU, Maharashtra, Parbhani, India] . Analysis of the phenotypic data was performed using the residual maximum likelihood algorithum (ReML), which provides the best linear unbiased predictors (BLUPs) of performance of the genotypes. The BLUPs of 213 uniform RILs, along with their genotypic data from 111 marker loci, were used for QTL analysis. Parental and RIL BLUPs revealed wide variation in phenotypic values for shoot fly resistance and its component traits in each of the screening environments. Wide variation was observed in the RIL population for shoot fly resistance component traits such as leaf glossiness, trichome density (upper and lower surfaces of seedling leaf blades), seedling vigor, oviposition preference, and deadhearts. These traits can be used as simple criteria to select for resistance to shoot fly in sorghum. The genotypic variances for shoot fly resistance traits were significant in each of the screening environments as well as across-environments. Glossiness intensity, trichome density (both upper and lower surfaces of seedling leaf blades), oviposition preference, deadhearts, and seedling vigor showed consistent heritability (broad-sense) estimates in individual screening environments, but low to moderate heritability estimates across environments,indicating that these traits are under genetic control, but there is a substantial role of genotype (G) × environment (E) interaction in expression of these traits.

QTL analysis was performed using the composite interval mapping (CIM) method implemented in PLABQTL version 1.1, which uses a regression approach. This revealed presence of putative QTLs for shoot fly resistance and its component traits including leaf glossiness, oviposition preference, deadhearts, and trichome density. The portion of observed phenotypic variance explained by different putative QTLs varied from 6 to 34%. Glossiness intensity was largely controlled by a major QTL on LG J (= SBI-05), accounting for 34% of observed phenotypic variation, and one minor QTL on LG G (= SBI-10), accounting for 8% of observed phenotypic variation across environments. After adjusting for QTL × environmental interaction, these two QTLs explained 31% of genetic variation in glossiness intensity in this population. Resistant parent IS 18551 contributed to additive genetic effects for increased glossiness at both of these QTLs. For oviposition preference and deadhearts, two common QTLs (one on LG F = SBI-09 and one on LG G) were also mapped in across-environments analysis. Together, these QTLs explained 17% phenotypic variation in oviposition preference and 19% for deadhearts in across-environments analysis. Significant QTL × environmental interactions were observed for these QTLs for oviposition preference and deadhearts. The QTL mapped on LG G for deadhearts and oviposition preference co-localized with a major QTL for trichome density (upper and lower surfaces of seedling leaf blades) and a minor QTL for glossiness intensity. The QTL mapped on LG F for deadhearts and oviposition preference co-localized with a minor QTL for trichome density on the lower leaf surface. For trichome density on the upper leaf surface, one QTL was detected on LG G accounting for 30% of observed phenotypic variance in across-environments analysis. This QTL co-localized with a QTL for trichome density on the lower leaf surface, and explained nearly 27% of observed phenotypic variance across two screening environments. The major QTL for glossiness intensity and a minor QTL for oviposition preference (LG J = SBI-05) and a major QTL for trichome density and minor QTLs for glossiness, deadhearts, and oviposition (LG G = SBI-10) detected in this study have previously been mapped at the same location in another sorghum RIL population derived from the cross BTx623 × IS 18551. This confirmed that these chromosomal regions might be harboring candidate genes contributing to shoot fly resistance in IS 18551.

During 2005, SSR-marker-assisted backcrossing of putative shoot fly resistance QTLs from donor parent IS 18551 into the genetic backgrounds of three hybrid parental lines (20B, 28B, and KR 192) of interest to the sorghum breeding program at the Marathwada Agricultural University-Parbhani, Maharashtra, India, were advanced to the BC_3F_1 seed generation. For this purpose, around 224 BC_2F_1 plants from five populations were genotyped at the seedling stage with 10 SSR marker loci linked to four targeted QTLs (one each on LG A = SBI-01, LG E = SBI-07, LG G = SBI-10, and LG J = SBI-05) associated with shoot fly resistance traits. Nearly 100 heterozygous plants, which had one, two, or more QTL introgression(s), were selected and crossed as a female parent with the selfed progeny of their respective recurrent parents to generate BC_3F_1 progenies.

SSR-marker-assisted backcrossing of putative shoot fly resistance QTLs into the genetic background of the susceptible parents of the two ICRISAT sorghum shoot fly mapping populations, i.e., BTx623 and 296B, were advanced during 2005. By the end of 2005, three of the four target QTLs from donor parent IS 18551 were advanced to the BC_4F_2 seed generation in the genetic background of shoot fly susceptible elite sorghum hybrid seed parental line 296B, and all four QTLs were advanced to the BC_4F_2 seed generation in the genetic background of shoot fly susceptible elite sorghum hybrid seed parental line BTx623. It is expected that the first BC_4F_2 shoot fly resistance QTL introgression homozygotes will be identified by mid-2006.

RT Folkertsma, BVS Reddy, HC Sharma , CT Hash, S Chandra and S Senthilvel

Activity 1.2.1.3 : Mapping head bug/grain mold resistance in sorghum
Team: RP Thakur, CT Hash, HC Sharma, BVS Reddy and EW Rattunde

RILs developed for grain mold resistance: Two grain mold RIL populations; 352 $F_{3:4}$ progenies of IS 23599 (grain mold resistant line) × AKMS 14B (grain mold susceptible line), and 348 $F_{3:4}$ progenies of IS 25017 (grain mold resistant line) × KR 188 (grain mold susceptible line), were advanced to $F_{3:5}$ progenies in the 2004/05 *rabi* season. Limited information available so far indicates that SSR marker polymorphism between parental line pairs of these populations may not be adequate to permit QTL mapping with sorghum SSR markers currently available in the public domain. Assessment of a sample of the RILs for variation in plant height and flowering time, as well as panicle compactness, in an appropriate grain mold screening environment is recommended as previously published attempts in the USA to map QTLs for the related grain weathering complex were largely ineffective as all "grain mold resistance" QTLs identified were either associated with alleles for tall plant height at dwarfing gene loci (that contributed to drier microenvironments for panicles borne by taller progenies in the RIL population) or with grain hardness QTLs.

BVS Reddy, S Ramesh, RT Folkertsma, CT Hash and RP Thakur

Activity 1.2.1.5: Mapping downy mildew resistance in pearl millet
Team: CT Hash and RP Thakur

Milestone: Cultivars derived through MAS for downy mildew resistance released for cultivation to the farmers (2008)

First product of pearl millet marker-assisted selection, "HHB 67 Improved", eleased for cultivation in India.
The year 2005 saw the first official release for seed multiplication and cultivation of a product of marker-assisted selection "HHB 67 Improved" for downy mildew resistance in pearl millet. This pearl millet hybrid was identified for state release in the Indian state of Haryana in January 2005, and was later identified (in July) at the national level for cultivation in the arid zone of northwestern India, where extra-early maturing pearl millet hybrid "HHB 67" has been cultivated on nearly 500,000 ha annually.

"HHB 67 Improved" is the first product of marker-assisted selection, other than transgenic Bt cotton hybrids, to be approved for release in India. The new hybrid was bred by backcrossing additional downy mildew resistance into the seed parents (using conventional greenhouse seedling screening procedures and resistance donor ICML 22) and the pollinator parent (using RFLP-based marker-assisted selection and resistance donor ICMP 451-P6) of "HHB 67", identifying BC_4F_3 families homozygous for resistance, and then performing line × tester experiments to identify agronomically superior hybrid combinations similar to "HHB 67", but having improved downy mildew resistance. The superior combinations were then tested multilocationally in on-station collaborative, state and national trials (2001-2004) with a smaller number being tested extensively in on-farm trials in Haryana (2003-2004), to identify the hybrid combination that has ultimately been released for cultivation as a high-yielding and disease-resistant replacement for "HHB 67".

Large quantities of breeder seed of the parental lines of "HHB 67 Improved" were distributed in 2005 to public and private seed agencies by ICRISAT and Haryana Agricultural University following approval of the hybrid's release by the central government in India. Due to regulatory delays in the release process, much of the breeder seed is being used for certified seed production of the new hybrid, rather than multiplication of foundation seed of the parental lines, that is normally used for producing certified seed. However, this should ensure that certified seed is available to sow >50,000 ha with "HHB 67 Improved" during the 2006 rainy season.

CT Hash and RP Thakur

Greenhouse screening for resistance to downy mildew: A total of 1443 lines, including inbreds, hybrids and self bulks of different populations were evaluated against three pathotypes (Sg 139 from Jodhpur, Sg 409 from Patancheru, and Sg 298 from New Delhi) of downy mildew pathogen, *Sclerospora graminicola* in 12 experiments. All screenings were carriedout in a completely randomized design having 3 replications, with 1 to 2 pots per replication, and 35 to 40 seedlings per pot. In each set, appropriate resistant and susceptible controls were included. In most of the sets, the common susceptible check 7042S recorded 95 to 100% disease incidence, indicating high effectiveness of the screening technique. The results of screening with frequencies of lines falling into different downy mildew incidence classes are presented in Table 2.1**,** and brief descriptions of material screened are given below. Lines showing ≤10% incidence were considered resistant.

Hybrids: A total of 53 hybrids made on 841B-like drought tolerance QTL introgression lines based on donor 863B-P2 were evaluated against three pathotypes (Sg 139 from Jodhpur, Sg 409 from Patancheru, and Sg 298 from New Delhi). Of these, 17 hybrids were resistant to Jodhpur pathotype, 5 to Patancheru pathotype, and 32 to New Delhi pathotype (Table 2.1). As it was considered useful to evaluate the performance of the resistant hybrids in their respective adaptation zones for further selection, the entire set of hybrids was also evaluated for agronomic performance in field trials at Central Arid Zone Research Institute (CAZRI), Jodhpur, Rajasthan, India, and Rajasthan Agricultural University, Regional Research Station (RAU-RRS), Nagaur, Rajasthan, India, during the rainy seasons of 2004 and 2005.

841B-like inbred lines: Fifteen 841B-like drought tolerance QTL introgression lines based on donor 863B-P2 were evaluated against three pathotypes (Sg 139 from Jodhpur, Sg 409 from Patancheru, and Sg 298 from New Delhi). Only one line each was resistant to the Jodhpur and Patancheru pathotypes, while 2 were resistant to the New Delhi pathotype. 863B remained highly resistant to all the three pathotypes while the remaining lines were susceptible. This indicated that 841B is now susceptible to this set of downy mildew pathotypes, which are broadly representative of the most virulent field isolates in India, and that the major downy mildew resistance QTL(s) from 863B are still effective against these pathotypes, and are not linked to the putative drought tolerance QTL on linkage group 2 of 863B. Pyramiding of downy mildew resistance and drought tolerance QTLs in the background of 841B will be necessary before an applied product can come out of the drought tolerance backcrossing program.

843B-like inbred lines: A total of 44 lines in the common background of 843B, but derived from backcrossing with downy mildew resistance donors ICML 22, P 7-3, and ICMP 85410 were evaluated against Patancheru (Sg 409), Jodhpur (Sg 139), and New Delhi (Sg 298) pathotypes. Only 4, 14, and 10 lines were resistant to pathotypes Sg 409, Sg 139, and Sg 298, respectively.

H 77/833-2-like inbred lines: Of 30 lines with a background of H 77/833-2 derived from downy mildew resistance donor ICMP 451-P6 and drought tolerance donor PRLT 2/89-33 evaluated against Patancheru (Sg 409), Jodhpur (Sg 139) and New Delhi (Sg 292) pathotypes, none showed resistance to Jodhpur (Sg 139) pathotype, while only 1 and 4 lines were resistant to Patancheru and New Delhi pathotypes, respectively. This indicated that H 77/833-2, ICMP 451-P6, and PRLT 2/89-33 are susceptible to these pathotypes, which are broadly representative of the most virulent field isolates in India; and pyramiding of downy mildew resistance and drought tolerance QTLs in the background of H 77/833-2 will be necessary before an applied product can come out of the drought tolerance backcrossing program. Crosses pyramiding the available drought tolerance QTL with downy mildew resistance QTLs in the background of H 77/833-2 were advanced by one generation.

Improvement of stover quality: A total of 61 breeding lines being used for improving the stover quality of elite pearl millet seed parent maintainer lines were evaluated against Jamnagar (Sg 200), Jodhpur (Sg 139), Durgapura (Sg 212), and Patancheru (Sg 409) pathotypes. Only 2 lines (863B-P2 and ICMB 99022) were disease free, 18 were resistant to all three pathotypes, and 9 were resistant to 2 pathotypes. The results indicated that the elite seed parent maintainer lines selected for stover quality improvement also need to be improved for downy mildew resistance. As 863B-P2 is being used as one of the stover quality donor parents, and it is also highly resistant to the full range of pathotypes screened, it should be possible to select more downy mildew

resistant derivatives from the segregating progenies using either conventional selection with seedling greenhouse screens of potted seedlings or RFLP/SSR/morphological marker-assisted selection.

Segregating breeding populations: *BC_2F_1s (81B × IP 18293) × 81B*: Of 40 $B_{C1}F_2/BC_2F_1$ progenies from [(81B × IP 18293) × 81B] evaluated against New Delhi pathotype (Sg 298), only 3 progenies were resistant (≤10% incidence) compared to 45 and 89% incidence on IP 18293 and 81B, respectively. It seems that the resistance donor IP 18293 is now susceptible to the New Delhi pathotype, and needs to be confirmed. Disease incidence in the segregating progenies ranged from 6 to 97%, with an operational heritability >0.90. Segregation patterns permitted identification of BC_2F_1 progenies apparently carrying one or more partially effective resistance genes from donor IP 18293. Disease-free seedlings from eight BC_2F_1 progenies in four families were transplanted to the field and advanced a generation by selfing and backcrossing to produce families of BC_2F_2/BC_3F_1 progeny pairs, each expected to segregate for one or more partially effective resistance genes from IP 19283 in the background of 81B. This map-directed conventional backcrossing program is expected to produce near-isogenic lines carrying single additional downy mildew resistance genes from IP 18293 in the genetic background of 81B.

***BC_2F_1s (843B × IP 18293) × 843B*:** Of 52 BC_1F_2/BC_2F_1 progenies from [(843B × IP 18293) × 843B] × 843B evaluated against New Delhi pathotype (Sg 298), none of the progenies were resistant (and none was expected), while IP 18293 recorded 45 and 843B 98% disease incidence, respectively. Disease incidence in the segregating progenies ranged from 10 to 99%, with an operational heritability of >0.90. Segregation patterns permitted identification of BC_2F_1 progenies apparently carrying one or more partially effective resistance genes from donor IP 18293. Disease-free seedlings from six BC_2F_1 progenies in four families were transplanted in the field and advanced a generation by selfing and backcrossing to produce families of BC_2F_2/BC_3F_1 progeny pairs, each expected to segregate for one or more partially effective resistance genes from IP 19283 in the background of 843B. This map-directed conventional backcrossing program is expected to produce near-isogenic lines carrying single additional downy mildew resistance genes from IP 18293 in the genetic background of 843B.

***BC_2F_2/BC_3F_1s (PT 732B × P 1449-2) × 81B*:** Of 169 BC_2F_2/BC_3F_1 progenies from [(PT 732B × P 1449-2) × 81B] evaluated against New Delhi pathotype (Sg 298), none of the progenies was uniformly resistant (and none was expected). Downy mildew incidence levels of 29, 79, and 100% were recorded on the resistance donor P 1449-2, and elite seed parents 81B and PT 732B, respectively. Disease incidence in the segregating progenies ranged from 30 to 100%, with an operational heritability >0.90. Segregation patterns permitted identification of BC_3F_1 progenies apparently carrying one partially effective resistance gene from donor P 1449-2. A small subset of the BC_3F_1 progenies (seven progenies distributed across five of the six families screened) was selected for re-screening against Sg 298 in early 2006. Disease-free seedlings from one or more of the selected progenies in these families will then be transplanted in the field and advanced a generation by selfing and backcrossing to produce families of BC_3F_2/BC_4F_1 progeny pairs, each expected to segregate for one or more partially effective resistance genes from P 1449-2 in the background of 81B. This map-directed conventional backcrossing program is expected to produce near-isogenic lines carrying single additional downy mildew resistance genes from P 1449-2 in the genetic background of 81B.

***BC_2F2/BC_3F_1s (PT 732B × P 1449-2) × 843B*:** Of 389 BC_2F_2/BC_3F_1 progenies from (PT 732B × P 1449-2) × 843B evaluated against New Delhi pathotype (Sg 298), no individual segregating progeny was found uniformly resistant (and none was expected). Downy mildew incidence of 11, 96, and 99% was recorded on resistance donor P 1449-2, and on elite seed parents 843B and PT 732B, respectively. Disease incidence in the segregating progenies ranged from 10 to 100%, with an operational heritability >0.90. Segregation patterns permitted identification of BC_3F_1 progenies apparently carrying one or two partially effective resistance genes from donor P 1449-2. A small subset of the BC_3F_1 progenies (18 progenies distributed across 8 of the 16 families screened) was selected for re-screening against Sg 298 in early 2006. Disease-free seedlings from one or more of the progenies in each of these selected families will then be transplanted in the field and advanced a generation by selfing and backcrossing to produce families of BC_3F_2/BC_4F_1 progeny pairs, each expected to segregate for one or more partially effective resistance genes from P 1449-2 in the background of 843B. This map-directed conventional backcrossing program is expected to produce near-isogenic lines carrying single additional downy mildew resistance genes from P 1449-2 in the genetic background of 843B.

BC_3F_2/BC_4F_1s (PT 732B × P 1449-2) × PT 732B: Of 144 BC_3F_2/BC_4F_1 progenies from [(PT 732B × P 1449-2) × PT 732B] evaluated against New Delhi pathotype, 28 progenies (19%) were resistant, while 42 and 100% downy mildew incidence was recorded on the resistance donor P 1449-2 and the recurrent parent PT 732B, respectively. Disease incidence in the segregating progenies ranged from 0 to 100%, with an operational heritability >0.90. Segregation patterns permitted identification of BC_4F_1 progenies apparently carrying one,

two, or three partially effective resistance genes from the donor P 1449-2. A small subset of the BC_4F_1 progenies (23 progenies distributed across all four families screened) was selected for re-screening against Sg 298 in 2006. Disease-free seedlings from one or more of the progenies in each of these selected families will then be transplanted in the field and advanced a generation by selfing and backcrossing to produce families of BC_4F_2/BC_5F_1 progeny pairs, each expected to segregate for one or more partially effective resistance genes from P 1449-2 in the background of PT 732B. This map-directed conventional backcrossing program is expected to produce near-isogenic lines carrying one or more additional downy mildew resistance genes from P 1449-2 in the genetic background of PT 732B.

Mapping population progeny F_4 self bulks (PT 732B-P2 × P 1449-2-P1): Of 131 F_4 self bulk mapping population progenies derived from cross [PT 732B-P2 × P 1449-2-P1], which were evaluated against the more virulent new Patancheru pathotype (Sg 409), none were resistant, while 80 and 100% disease incidence was recorded on parental lines P 1449-2-P1 and PT 732B-P2, respectively. The limited variation detected was less than expected based upon prior screens of the parental lines of this pearl millet mapping population. It was expected that this screen would permit QTL mapping of resistance from donor parent P 1449-2, which could be incorporated into the genetic background of four elite seed parent maintainer lines (at least two of which are now highly susceptible to this pathotype) into which stover quality QTLs from P 1449-2 are also being backcrossed. However, as no resistance genes are segregating in this mapping population with effects on Sg 409 that are large enough to warrant backcrossing, QTL mapping using this phenotyping data set has not yet been attempted.

Mapping population progeny F_4 self bulks (841B-P3 × 863B-P2): Of 160 F_4 self bulk mapping population progenies derived from cross 841B-P3 × 863B-P2, which were evaluated against the more virulent new Patancheru pathotype (Sg 409), only 16 progenies (10%) were found resistant. Disease incidence on the parental lines was 94% on 841B-P3 and 11% on 863B-P2, while 100% disease incidence was observed on the susceptible control, 7042S.

QTL analysis of the mapping population progeny means by composite interval mapping procedures detected a major QTL on linkage group 4 (near RFLP marker locus *Xpsm*265 in the vicinity of the d_2 dwarfing gene locus) accounting for nearly 60% of the observed variation in downy mildew incidence among the mapping population progenies in this screen, with resistant parent 863B-P2 providing the favorable allele (having an additive effect that reduces downy mildew incidence by 32%). Two resistance QTLs of small effect, with favorable alleles from 841B-P3, were also detected (one each on linkage group 1 and linkage group 7).

While it may prove difficult in the short term to recombine resistance at this QTL with the semi-dwarf plant height that is preferred for hybrid seed parent lines, due to its linkage with a tall allele at the d_2 dwarfing gene locus, it will be relatively simple to backcross this gene into the genetic backgrounds of elite d_2 dwarf seed parent maintainer lines (as has already been initiated for recurrent parent ICMB 89111-P2). This can be done by making backcrosses from tall (D_2/d_2) BC_nF_1 plants (as pollen parent) onto the d_2-dwarf recurrent parent (as stigma parent), passing the resulting $BC_{n+1}F_1$ progenies through a severe greenhouse seedling screen against downy mildew isolate Sg409, and transplanting the disease-free segregates in the field (along with non-inoculated recurrent parent seedlings) for generation advance. At the end of this combined conventional greenhouse screening and morphological marker-assisted backcrossing program, it will be necessary to use a large $BC_{n+m}F_2$ population to find the expected rare individuals that have both d_2 dwarf plant height and are heterozygous for the resistance allele at the linked downy mildew resistance gene. A further generation of head-to-row advance, with conventional screening, will then allow identification of $BC_{n+m}F_3$ progenies that are homozygous for both d_2 dwarf plant height and the downy mildew resistance gene. When such plants are used as resistance donors, the downy mildew resistance allele will be tightly linked with the recessive dwarf plant height gene, and molecular marker-assisted selection combined with conventional screening will prove to be best alternative for introgressing this 863B resistance allele into other d_2 dwarf genetic backgrounds that are susceptible to Sg 409.

Mapping population progeny F_4 self bulks (ICMB 89111B-P6 × ICMB 90111B-P6): Of 206 F_4 self bulk mapping population progenies derived from cross ICMB 89111B-P6 × ICMB 90111B-P6, which were evaluated against the virulent Patancheru pathotype (Sg 409), only 17 progenies (8%) were found resistant, while 100% disease incidence was recorded on susceptible parent ICMB 89111B-P6 and 11% on resistant parent ICMB 90111B-P6. QTL analysis of this data set is in progress.

Screening for resistance to downy mildew under field conditions: Screening downy mildew resistance of all breeding materials was conducted in a completely randomized block design with 3 replications, and plots of 2

rows × 4 m, using 7042S as the standard susceptible control. All screening trials were quite successful with 7042S recording 95 to 99% disease incidence. Lines scoring ≤10% incidence were considered resistant. A total of 99 inbred lines, hybrids, and/or open-pollinated varieties from 4 trials were screened, of which 34 were found resistant (Table 2.1). The details of each trial are provided below, comparing results from greenhouse screening of the same materials against the more virulent new Patancheru pathotype (Sg 409).

841B-like inbreds: Thirteen 841B-like drought tolerance QTL introgression lines based on donor 863B-P2, and their two parental lines (841B-P3 and 863B-P2) were evaluated. Of the 15 trial entries, 14 were resistant in this field screen. In comparison, greenhouse screen of the same lines against Patancheru pathotype Sg 409, only resistant parent 863B-P2 was considered highly resistant, while 841B-P3 and its thirteen near-isogenic derivatives were all considered susceptible. The observed differences between field and greenhouse screening are somewhat troubling, but indicative of the reasons that the greenhouse screen is preferred for selection purposes while the field screen is now restricted to a confirmatory role. The combined results from field and greenhouse screening of this set of inbreds indicated that pyramiding of downy mildew resistance and drought tolerance QTLs in the background of 841B (based on molecular marker-assisted selection for the linkage group 2 drought tolerance QTL from 863B and molecular marker-assisted selection or greenhouse seedling screening for the linkage group 4 downy mildew resistance QTL from 863B) will be necessary before an applied product can come out of the drought tolerance backcrossing program in this genetic background.

843B-like inbreds: Of 44 inbred lines screened, including backcross derivatives of recurrent parent 843B with resistance donors ICML 22, P 7-3, and ICMP 85410, along with their parents, only 12 were resistant in the downy mildew nursery. In comparison, greenhouse screen of this same set of materials against Patancheru pathotype Sg 409 identified only 4 inbreds as resistant. In general, most of the near-isogenic single-QTL introgression lines in this set of materials did not have sufficiently improved downy mildew resistance to warrant widespread use in applied hybrid breeding programs. Resistance gene pyramiding (based upon marker-assisted selection or greenhouse screening of potted seedlings against one or more highly virulent pathogen isolates) and/or other resistance deployment strategies are required to generate materials for applied use. However, several of the materials in this set of inbreds could be used as near-isogenic host differential lines for characterizing pathogen virulence variation.

H 77/833-2-like inbreds: Of 30 entries screened in the downy mildew nursery in the H 77/833-2-like inbreds trial, only 2 were considered resistant. Results of this field screen were similar to the greenhouse screen of these same materials against Patancheru pathotype Sg 409, in which only 1 resistant line was identified.

ACIAR pearl millet stover quality project trial: This trial contained a pair of near-isogenic hybrids differing for a genomic block controlling disease reaction to downy mildew and rust, two pairs of composite populations near-isogenic for normal and mutant alleles at a brown midrib gene, a pair of composite populations near-isogenic for a portion of pearl millet linkage group 7 associated with a putative stover quality QTL, and two released dual-purpose open-pollinated varieties. Of the 10 trial entries screened in the downy mildew nursery, 6 were resistant. No clear associations were observed between disease reaction and expected differences in ruminant livestock nutritional quality of the pairs of entries in this trial.

CT Hash,T Nepolean ,S Senthilvel and RP Thakur

Table 2.1: Summary of 2005 phenotyping of pearl millet mapping population progenies and products of conventional and marker-assisted backcrossing for resistance to different pathotypes (*Pat = Patancheru, Jdp = Jodhpur, Ndl = New Delhi) of pearl millet downy mildew (*Sclerospora graminicola*) (ICRISAT, Patancheru, India)

Material	Pathotype*	No of lines	Number of lines in each DM incidence class					
			0 (%)	1-5 (%)	6-10 (%)	11-20 (%)	21-30 (%)	>30 (%)
Greenhouse screening								
Hybrids of 841B-like drought tolerance QTL introgression lines based on donor 863B-P2	Jdp (Sg 139)	53	6	5	6	16	13	7
	Pat (Sg 409)	53	4	0	1	10	9	29
	Ndl (Sg 298)	53	6	17	9	15	5	1
B-lines from 841B-like drought tolerance QTL introgression backcrossing based on donor 863B-P2	Jdp (Sg 139)	15	1	0	0	0	0	14
	Pat (Sg 409)	15	1	0	0	0	0	14
	Ndl (Sg 298)	15	1	0	1	0	4	9

843B-like inbred lines	Pat (Sg 409)	44	0	3	1	2	2	36
	Jdp (Sg 139)	44	5	7	2	0	0	30
	Ndl (Sg 298)	44	0	2	8	1	1	32
H 77/833-2-like inbred lines	Pat (Sg 409)	30	0	0	1	1	2	26
	Jdp (Sg 139)	30	0	0	0	0	0	30
	Ndl (Sg 298)	30	1	0	3	2	1	23
ACIAR pearl millet stover quality project trial	Pat (Sg 409)	10	0	0	1	3	2	4
	Jdp (Sg 139)	10	0	1	2	3	0	4
	Ndl (Sg 298)	10	0	2	2	1	2	3
BC_1F_2/BC_2F_1 progenies from (81B × IP 18293) × 81B	Ndl (Sg 298)	40	0	0	3	3	10	24
BC_1F_2/BC_2F_1 progenies from (843B × IP 18293) × 843B	Ndl (Sg 298)	52	0	0	0	4	7	41
BC_2F_2/BC_3F_1 progenies from (PT 732B × IP 1449-2) × 81B	Ndl (Sg 298)	169	0	0	0	0	4	165
BC_2F_2/BC_3F_1 progenies from (PT 732B × IP 1449-2) × 843B	Ndl (Sg 298)	389	0	0	0	5	16	368
BC_3F_2/BC_4F_1 progenies from (PT 732B × IP 1449-2) × PT 732B	Ndl (Sg 298)	144	8	5	15	21	16	79
F_4 self bulks (PT 732B-P2 × IP 1449-2-P1)	Pat (Sg 409)	131	0	0	0	0	0	131
F_4 self bulks (841B-P3 × 863B-P2)	Pat (Sg 409)	160	0	2	14	17	14	113
F_4 self bulks (89111B-P6 × 90111B-P6)	Pat (Sg 409)	206	0	10	7	17	15	157
Field Screening								
841B-like inbred lines		15	1	11	2	0	0	1
843B-like inbreds lines		44	0	6	6	7	8	17
H 77/833-2-like inbred lines		30	0	1	1	7	6	15
ACIAR pearl millet stover quality project trial		10	0	4	2	2	2	0

Activity 1.2.1.6: Mapping *Helicoverpa* resistance in chickpea
Team: HC Sharma, PM Gaur and D Hoisington

Milestone: Mapping of pod borer resistance in chickpea (2007)

Development of interspecific mapping populations: Two RIL mapping populations are being developed from the interspecific crosses of *C. arietinum* × *C. reticulatum* (ICC 3137 × IG 72953 and ICC 3137 × IG 72953) for mapping of QTLs for resistance to the pod borer, *Helicoverpa armigera*. The crosses were advanced by one generation (F_1) in the field in the crop season, and by two generations (F_2 and F_3) in the greenhouse during the off-season. There are 210 RILs in the cross ICC 3137 × IG 72953 and 260 RILs in the cross ICC 3137 × IG 729353. Crosses were also made between ICCC 37 and ICC 506EB for initiating development of an intraspecific mapping population for mapping resistance to *H. armigera*.

PM Gaur, HC Sharma and TJ Ridsdill-Smith

Evaluation of mapping population (ICC 506 × Vijay) for resistance to Helicoverpa armigera: The mapping population ICC 506 × Vijay (200 lines) was evaluated for resistance to *H. armigera* under natural infestation in the field. There were three replications in a randomized complete block design. Observations were recorded on leaf damage, numbers of eggs laid, larval density, number of pods, pods damaged, and grain yield. The leaf damage rating ranged from 1.0 to 8.0 in the mapping population, 1.7 in Vijay, 1.0 in ICC 506, and 3.2 in ICCC 37 (1 = <10% leaf area damaged, and 9 = >80% leaf area damaged). The overall resistance scores were 2.7 and

2.0 in Vijay and ICC 506, respectively, 5.2 in ICCC 37, and 2.0 to 6.7 in the mapping population. Percentage pod damage ranged from 6.9 to 34.8% in the mapping population, 22.2% in Vijay, 5.8% in ICC 506, and 25.6% in ICCC 37. This population is being evaluated for one more season to use this data for identifying molecular markers associated with resistance to *H. armigera.*

Evaluation of Cicer reticulatum × C. arietinum interspecific mapping populations for resistance to Helicoverpa armigera: The F_2s of the interspecific populations involving *Cicer reticulatum* × *C. arietinum* (IG 72933 × ICC 3137, and IG 72953 × ICC 3137) were planted in the field, and evaluated for resistance to *H. armigera* using the detached leaf assay. There were five replications for each plant in a randomized complete block design. Data were recorded on leaf feeding (1 = <10% leaf area damaged, and 9 = >80% leaf area damaged), larval survival, and larval weight. In the mapping population based on IG 72933 × ICC 3137 (298 progenies + 3 checks), the leaf damage rating ranged from 1.7 to 8.0 in the mapping population, 3.5 in IG 72933 and 3.3 in ICC 3137, and 2.6 in ICC 506. Larval survival ranged from 24 to 99% in the mapping population, 62% in ICC 3137, 69% in IG 72933, and 65% in ICC 506. Larval weights ranged from 1.63 to 12.93 mg in the mapping population, 7.88 mg in ICC 3137, 5.23 mg in IG 72933, and 5.477 mg in ICC 506. In the mapping population based on IG 72953 × ICC 3137 (110 lines + 3 checks), the leaf damage rating was 1.7 to 6.9 in the mapping population, 3.9 in IG 72953 and 3.4 in ICC 3137, and 3.3 in ICC 506. Larval survival ranged from 49 to 92% in the mapping population, 61% in ICC 3137, 58% in IG 72953, and 56% in ICC 506. Larval weights ranged from 4.592 to 15. 318 mg in the mapping population, 9.647 mg in ICC 3137, 4.691 mg in IG 72953, and 7.552 mg in ICC 506. DNA extracted from this population has been sent to CAMBIA for genotyping.

HC Sharma, PM Gaur and TJ Ridsdill-Smith

Activity 1.2.1.7: Mapping *Ascochyta* blight and *Fusarium* wilt resistance in chickpea
Team: S Pande, PM Gaur and D Hoisington

Milestone: Markers identified for *Botrytis* gray mold and *Fusarium* wilt resistance and validated (2005)

Phenotyping of recombinant inbred and germplasm lines for BGM resistance: Recombinant inbred lines (RILs) of chickpea developed from a cross ICCV 2/ICCV 10 × ICC 1496 were evaluated for BGM resistance under controlled environment conditions. Phenotyping of 222 F_3 RILs and 250 F_7 RILs of ICCV 2 × ICC 1496, and 254 F_3 and 250 F_7 RILs of ICCV 10 × ICC 1496 was conducted using conditions optimum for disease development. Ten-day-old seedlings of the test material along with JG 62 as a susceptible check were inoculated by uniform foliar spray of inoculum (3×10^5 conidia ml^{-1}). Inoculated seedlings were incubated in a growth room at 15°C and 100% RH. At 20 days after inoculation (DAI), disease severity was measured on a 1 to 9 rating scale (1 = >10% healthy plants, and 9 = >75% plants killed). RIL ICCX-000049-F_2 (10P)-847 derived from ICCV 10 × ICC 1496 was found to be resistant (<3 rating) to BGM. Distribution of moderately resistant (3.1 to) lines in each set was as follows: RILs of F_3 ICCV 10 × ICC 1496 - 179 lines; F_7 ICCV 2 × ICC 1496 - 78 lines; F_7 ICCV 10 × ICC 1496 - 17 lines.

A set of 59 chickpea lines that had distinct genotypic profiles were tested for BGM resistance under controlled environment conditions as mentioned above. Among these, ICC 1496, ICCV 93513, ICCV 96852, and ICCV 98503 were moderately resistant to BGM with a mean disease score of 3.1 to 5.

S Pande, PM Gaur and GK Kishore

Identification of molecular markers for resistance to Aschochyta blight: A linkage map of chickpea with 84 markers (82 SSRs and 2 ESTs) was constructed using F_2 population of an intraspecific cross of ICCV 04516 (AB resistant) and Pb 7 (AB susceptible). The map length was 724.4 cM with an average density of 8.62 cM. Three AB resistance QTLs were mapped, QTL1 on LG 3, and QTL 2 and QTL 3 on LG 4. The QTLs identified on LG 4 were validated in another population. A set of 84 RILs derived from the cross C 104 × WR 315 was genotyped using 73 SSR markers and screened for resistance to *Fusarium* wilt (FW) race-1. Forty-five markers were assigned to nine linkage groups. The gene for FW race 1 was mapped on linkage group 2. Three SSR markers, TA37, TA 200, and TR2 were closely linked to the resistance gene. The former was located at a distance of 0.4 cM, while the latter two markers were located at a distance of 3.5 cM. Efforts were made to expand the genome map of chickpea and mapping of *Fusarium* resistance genes using ICCV 2 × JG 62 RILs. The 126 RILs of ICCV 2 × JG 62 were screened for 12 additional polymorphic markers. Now the data is available for 206 markers on these RILs.

PM Gaur and S Pande

Activity 1.2.1.8: Mapping *Fusarium* wilt resistance in pigeonpea
Team: D Hoisington , S Pande and KB Saxena

Milestone: Markers for *Fusarium* wilt (*Fusarium udum*) resistance in pigeonpea, and fodder and feed quality in groundnut and pigeonpea mapped and verified in unrelated populations

Standardization of phenotyping for Fusarium wilt resistance in pigeonpea: A reliable and reproducible technique for wilt resistance screening was standardized using three pigeonpea cultivars: ICP 2376 susceptible, and ICP 8863 and ICPL 87119 – resistant to wilt. Using root dip technique, the effect of inoculum concentration of *F. udum* on wilt development in these cultivars was studied. Eight-day-old seedlings raised in sterile sand were uprooted and the roots dipped in a conidial suspension of *F. udum* (Patancheru isolate) from eight-day-old shake culture. Conidial concentrations used for this study were: 1×10^4, 2.5×10^4, 5×10^4, 1×10^5, 2×10^5, and 3×10^5 conidia ml^{-1}. Root-inoculated seedlings were transplanted in pre-irrigated 15 cm diameter plastic pots filled with sterilized sand and black soil (4: 1). Minimum and maximum temperatures in the greenhouse were 18 to 20 and 26 to 29°C, respectively. Incidence of wilt in each treatment was recorded periodically up to 30 days after inoculation. Conidial concentration had a direct effect on the incidence of wilt. Incidence of wilt was highest at the conidial concentration of 1×10^5 and above. Cultivar ICP 2376 had 100% wilt, while ICP 8863 had 0% wilt, and ICPL 87119 had 20% wilt at 1×10^5 conidia ml^{-1} 15 days after inoculation (DAI). By 30 DAI, the mortality in ICP 8863 and ICPL 87119 increased to 20% and 40%, respectively. With a decrease in inoculum concentration, percentage mortality decreased and incubation period increased in all the three cultivars. Using the standardized root dip inoculation method and optimum inoculum concentration, phenotyping of recombinant inbred lines for wilt resistance from the cross ICP 2376 × ICPL 87119 and parents was carried out to map *Fusarium* wilt resistance.

S Pande and KB Saxena

Output 1.2.2 Exploitation of wild relatives of crops for increasing the levels and diversifying the basis of resistance to insect pests and diseases

Activity 1.2.2.1: Mechanisms, diversity, stability, and inheritance of resistance to *Helicoverpa* in wild relatives of chickpea and pigeonpea
Team: HC Sharma and SL Clements

Milestone: Mechanisms or resistance to *Helicoverpa armigera* in wild relatives of chickpea and pigeonpea studied.

Mechanisms of resistance to Helicoverpa armigera in wild relatives of chickpea: In the absence of high levels of resistance to *H. armigera* in the cultivated germplasm of chickpea, we evaluated 25 *Cicer reticulatum* accessions for resistance to this pest. There were two replications in a randomized complete block design under field conditions. Under multi-choice conditions in the field, ten accessions showed lower leaf damage and lower numbers of eggs and/or larvae of *H. armigera*. Of these, IG 69960, IG 72934 and IG 72936 showed significantly lower leaf feeding than the cultivated genotypes and/or other accessions at the vegetative and reproductive stages. Larval weight was lower or comparable to that on *C. bijugum* (IG 70019) and *C. judaicum* (IG 70032) in *C. reticulatum* accessions IG 72933, IG 72934, IG 72936, and IG 72953 at the seedling stage, and on IG 69960 and IG 72934 at the flowering stage. Less than 7 larvae survived (out of 15) on IG 70020, IG 72940, IG 72948, IG 72949, and IG 72964 compared to 12 on ICC 506. Developmental period was prolonged by 3 to 8 days on *C. reticulatum* accessions compared to that on ICCC 37. Less than five larvae pupated on the *C. reticulatum* accessions (except IG 72958 and ICC 17163) compared to 11 in ICCC 37. Accessions showing lower leaf feeding and adverse effects on the survival and development can be used in increasing the levels and diversifying the basis of resistance to *H. armigera* in chickpea.

HC Sharma, SL Clemens and TJ Ridsdill-Smith

Mechanisms of resistance to Helicoverpa armigera in wild relatives of pigeonpea: There was considerable variation in oviposition preference of the female moths on different accessions of the same species. *Cajanus albicans* (ICPW 13) and *C. scarabaeoides* (ICPW 90, ICPW 94, ICPW 116, and ICPW 137) were non-preferred for oviposition (<100 eggs per female) compared to the cultivated pigeonpea, ICPL 87 (334 eggs), whereas some of the accessions belonging to *C. acutifolius* (ICPW 2), *C. cajanifolius* (ICPW 28 and ICPW 29), *C. sericeus* (ICPW 160), *C. lineatus* (ICPW 40), *C. scarabaeoides* (ICPW 281), *Dunbaria ferruginea* (ICPW 178), and *Flemingia bracteata* (ICPW 192) were highly preferred as a substrate for oviposition (236 to 425 eggs).

Larval weights were <20 mg when reared on diets having pod powder of *C. acutifolius*, *C. sericeus* (ICPW 160), *C. scarabaeoides* (except ICPW 137, ICPW 141, and ICPW 152), *P. scariosa*, *C. platycarpus*, and *R. aurea* compared to 53.3 mg on ICPL 87 and 44.0 mg on ICPL 332. Larvae took >25 days to complete the development when reared on the artificial diet impregnated with lyophilized pod powder of *C. acutifolius* (ICPW 2), *C. lineatus* (ICPW 41), *C. sericeus*, *C. scarabaeoides* (except those reared on ICPW 125), *P. scariosa* (ICPW 207), *R. aurea* (ICPW 210), *D. ferruginea*, and *C. platycarpus* (ICPW 68) as compared to 15.7 days on ICPL 87, 23.3 days on ICPL 332, and 12.7 days on artificial control diet.

HC Sharma

Activity 1.2.2.2: Introgression of genes conferring resistance to *Helicoverpa* from wild relatives of pigeonpea and chickpea
Team: N Mallikarjuna, PM Gaur, HC Sharma and HD Upadhyaya

Wide crosses for pod borer resistance in pigeonpea: To transfer pod borer resistance genes from *C. scarabaeoides* to cultigens, crosses between resistant wild *Cajanus* and cultigens were attempted during the 2004 rainy season. The F_{1s} of the crosses, ICPW 94-P_1 ×ICP 28 P_1, ICPW 125-P_1 × ICP 26-P_1, ICPW 130-P_1 × ICP 26-P_2, and ICP 26-P_3 × ICP 14770-P_1 along with their parents were evaluated during the 2005 rainy season. Samples for DNA extraction have been collected. Thirty-eight resistant F_5 progenies were selected during 2004 rainy season. One single plant from each progeny has been selected for bi-parental mating to improve on deficient traits for resistance and productivity.

HD Upadhyaya

Evaluation of progenies for resistance to pod borer from the secondary and tertiary gene pool of Cicer: Nine lines were selected from the cross ICCV 92318 × IG 72933 with less than 25% *Helicoverpa armigera* damage from 2004 screening experiment and screened in 2005 for *H. armigera* under unprotected field conditions. One line showed pod damage of 10%, and one line with 25%. In the rest of the lines, the damage was between 25-75%. Six lines were selected from the cross ICCV 92318 × IG 72934, from 2004 screening experiment which had *H. armigera* damage of 20%, one line showed a damage rating of less 10%. In the rest of the lines the damage rating was between 25-50%. Twenty one lines from the cross ICCV 92318 × IG 72937 were selected from 2004 *H. armigera* screening experiment. Two lines did not show any pod damage (0%), 2 lines had <10% damage, and 3 lines had 20 to 25% damage.

Cicer pinnatifidum and *C. bijugum* were crossed with chickpea cultivars KAK-2, ICCV 2, ICCV 10, ICCV 92318, and JGK 1. Embryo rescue techniques were used to save aborting embryos from all cross combinations. Crosses with *C. pinnatifidum* did not produce normal green hybrids with any of the cultivars. *C. bijugum*, which has multiple disease resistance, produced green hybrid plants with KAK-2 and ICCV 2. This is a breakthrough in chickpea wide crosses, as hybrids have not been reported earlier with *C. bijugum*. Study of trichomes was undertaken on *C. bijugum* (IG 70006), *C. judaicum* (IG 70038) and *C. pinnatifidum* (ICCW 37), the wild *Cicer* used in the crossing program. Two types of trichomes were observed. Type A was non-glandular, and present on *C. bijugum* (IG 70006), *C. judaicum* (IG 70038), *C. pinnatifidum* (IG 17152) and *C. arietinum*. Type B trichome had varied number of neck cells and was present on *C. bijugum* (ILWC 69), *C. judaicum* (IG 17150), *C. pinnatifidum* (ICCW 37) and *C. arietinum*. Hence according to the present study there is no major difference between the wild species and the cultivated chickpea used in the study based on the presence of type A and B trichomes.

Wide hybridization in pigeonpea involving compatible Cajanus gene pool: Over 6550 plants were obtained as progeny from 287 selections made in 2004-2005 (Figure 2.1). In 31 selections, pod damage was < 20%, and many of the plants had a damage rating of <12% (Figure 2.2). Some of the selections had tan colored pods. From the total of 6550 plants 41 plants were found to be high yielders with the number of pods ranging from 400 to 900, and the number of seeds ranging from 1040 to 2300 plant^{-1}.

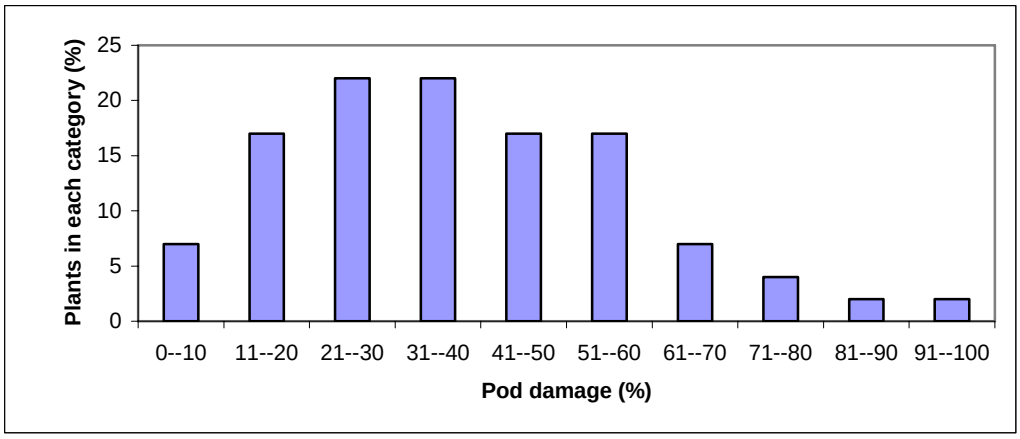


Figure 2.1. Frequencies of pigeonpea plants with different levels of Helicoverpa armigera damage in progenies derived from compatible gene pool.

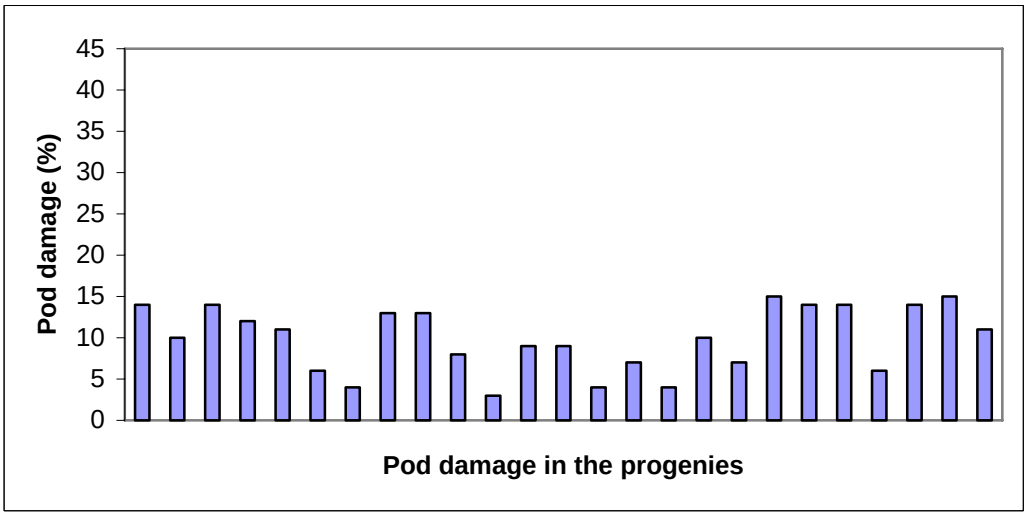


Figure 2. 2. Helicoverpa damage in plants of the progeny 8329 (Cajanus cajan cv, ICPL 2 × C. acutifolius)

N Mallikarjuna and HC Sharma

Wide hybridization in pigeonpea involving incompatible gene pool: Advanced generation diploid (F_1BC_4) and tetraploid (F_4-F_5) derivatives from the cross *C. platycarpus* × *C. cajan* were screened for *H. armigera* under unprotected field conditions. The hybrids flowered between 70-73 days compared to 80 in the cultivated parent ICPL 85010, and 55 days in *C. platycarpus*. Pod set was earlier in the progenies when compared to cultivated parent. Damage due to *H. armigera* in the first flush, in the wild parent *C. platycarpus*, was <1%. Damage in cultivated parent ICPL 87 was 80 %. Damage in diploid derivatives was 20% and damage in tetraploid derivatives was 8%. Nearly 25% pod damage was recorded in the second and third flush in the diploid hybrids.

N Mallikarjuna and HC Sharma

Interspecific crosses and advanced generation lines evaluated for resistance to Helicoverpa armigera: Selections from the interspecific crosses (derived from *Cajanus scarabaeoides, C. sericeus* and *C. acutifolius*) were evaluated for resistance to *H. armigera:* Forty selections derived from the crosses involving *C. scarabaeoides, C. lanceolatus, C. albicans, C. trinervius, C. sericeus* and *C. acutifolius*, along with resistant (ICPL 332, ICP 7035, and ICPL 88039) and susceptible (ICPL 87 and ICPL 87119) checks were evaluated for resistance to *H. armigera* under field conditions. There were three replications in a randomized complete block design. Data were recorded on pod damage rating, percentage pod damage, and healthy pods. Pod damage rating ranged from 2.0 to 9.0 in the test material compared to 9.0 in ICPL 87, 4.5 in ICPL 87119, and 3.2 in ICPL 332. Selection numbers 1, 6, 7, 17, 18, 26, 33, 34, 35, and 38 had lower pod borer damage (DR <5.0, and pod damage

<30%), and more number of healthy pods plant^{-1} (>50 per plant) compared to a DR of 9.0, 73% pod damage, and <15 pods plant^{-1} in ICPL 87. The ICPL 87119 had a pod damage rating of 4.5, and 44% pod damage.

HC Sharma and HD Upadhayaya

Activity 1.2.2.3: Wide hybridization for resistance to *Botrytis* gray mold and *Ascochyta* blight in chickpea.
Team: N Mallikarjuna, S Pande and PM Gaur

Milestone: Resistance to *Ascochyta* blight and *Botrytis* gray mold introgressed in to chickpea cultivars through interspecific hybridization.

Identification of BGM resistance in wild Cicer accessions: *Botrytis* gray mold (BGM) caused by *Botrytis cinerea* is a serious biotic constraint of chickpea in eastern Indo-Gangetic plains of India, Nepal, and Bangladesh. Extensive screening of germplasm and breeding material failed to detect high levels of resistance to BGM. By following standardized screening procedures, 118 wild *Cicer* accessions were evaluated for résistance to BGM. Three accessions of *C. bijugum* (IG 69981, 70022, and 70023), 21 accessions of *C. judaicum* (ICCs 17148, 17149, 17151, 17193, 17194, 17204, 17205, IGs 69959, 69969, 69977, 69986, 69987, 70000, 70030, 70032, 70033, 70034, 70037, 70038, 72931, and 72932), and one accession of *C. reticulatum* (IG 72959) were found to be resistant to BGM with a mean disease score of < 2.5 on a 1 to 9 rating scale as compared to 9.0 in the susceptible check, JG 62.

Intensive screening was carried out on 155 accessions of wild *Cicer* belonging to different species in 2004. From these, BGM resistant wild *Cicer* accessions IG 73074 and IG 17159 *(C. echinospermum*) were crossed with cultivated chickpea. F_1 and F_2 plants were screened for resistance to BGM under simulated conditions. Amongst 74 hybrids derived from the cross chickpea × IG 17159, 25 had a score of 3, and 2 hybrids had a score of 2 on a scale of 1 to 9. In the cross chickpea × IG 73074, 20 hybrids were screened for BGM and 50% of the hybrids had a score of 3. These will be advanced in 2006 and screened for resistance to BGM.

S Pande, N Mallikarjuna and GK Kishore

Activity 1.2.2.4 : Wide hybridization for resistance to early/late leaf spots and Aflatoxins in groundnut
Team: N Mallikarjuna and F Waliyar

A total of 105 lines of advanced generation interspepific derivatives involving *A. cardenasii*, *A. stenosperma*, *A. kempf-mercadoi* and *A. diogoi* were screened for disease reaction against late leaf spot (LLS) under field conditions during 2005 rainy season. Field trials were laid out in a broad-bed-and-furrow (BBF) system, size of each plot was 1.5 × 4 m, with inter-row spacing of 30 cm. Plant spacing of 10 cm was maintained for each line, and the experiment was un-replicated. TMV 2, a highly susceptible cultivar to LLS was used as an infector row after every five-test rows. Chemical sprays were used to control insect pests. At 50 days after sowing, plots were inoculated by spraying conidial suspension of *Phaeoisariopsis personata* urediniospores. After inoculation, perfo-irrigation was provided daily for 15 min in the evening for 30 days to create high humidity required for disease development. LLS incidence and severity was scored on a 1 to 9 rating scale at intervals of 15 days from 75 to 105 days after sowing.. Out of 113 interspecific derivatives, 19 lines from *A. hypogaea* × *A. cardenasii*, and one line from *A. hypogaea* × *A. dura* × *A. hypogaea* showed high levels of resistance to LLS with a score of 2 (on 1 to –9) scale. From 4 wide crosses, 58 lines had a score of 3, 21 had a score of 4, Promising lines with a disease rating of 3 and less will be advanced for further testing.

In the experiment to generate interspecific amphidiploids, wild Arachis from different sections were crossed with A. hypogaea. F_1 hybrids were obtained between A. hypogaea and A. diogoi (section Arachis), BC_1 and BC_2 hybrids were obtained between A. hypogaea × A. chiquitana (section Procumbentes), and two BC_2 hybrid was obtained between A. hypogaea × A. kretschmeri (section Procumbentes).

F Waliyar and N Mallikarjuna

Output 1.2.3: . Transgenic resistance to insect pests and diseases

Activity 1.2.3.1: Transgenic resistance to *Helicoverpa* in pigeonpea and chickpea
Team: KK Sharma and HC Sharma

Milestone: Transgenic plants resistant to *Helicoverpa armigera* evaluated

Putative transgenic plants carrying cry1Ac evaluated for resistance to Helicoverpa armigera under greenhouse and field conditions: Transgenic pigeonpea plants were developed by introducing the synthetic *cry1Ac* gene through *Agrobacterium tumefaciens*-mediated genetic transformation. Transgenic pigeonpea plants carrying the *cry1Ac* gene were evaluated during 2004/05 cropping seasons under contained conditions using a completely randomized block design, and there were three replications. The selected transgenic plants were bio-assayed with neonate and third-instar *H. armigera* larvae under laboratory conditions. The leaves of transgenic pigeonpea events ICPL 88039-10-3-5, ICPL 88039-15-4-2, ICPL 88039-13-1-1 (6.17 to 6.78 mg per larva) reduced the larval weight (by 40%) after 4 days of infestation as compared to that on the control plants (10.56 mg per larva). In flower bioassay, three transgenic events showed 40 to 49.2% mortality as compared to 32% on control plants. Pod bioassay of ICPL 88039-13-1-1 and ICPL 88039-15-4-2 transgenic events with third-instar *H. armigera* indicated weight gain of 156.4 to 263.2% as compared to 357.4 to 461.4% in control plants.

Evaluation of transgenic chickpea plants plants for resistance to Helicoverpa armigera: Over 20 transgenic events of chickpea with *cry1Ab* and *cry1Ac* genes were bio-assayed for resistance to *H. armigera.* Selected events and their progenies were also tested extensively under field and contained greenhouse conditions. There was approximately 35 to 40% reduction in *H. armigera* larval weight on transgenic chickpea plants as compared to control plants. In transgenic plants, larval weight ranged from 4.14 to 5.13 mg per larva as compared to 8.96 to 9.13 mg per larva on control plants. The selected plants have also been re-evaluated using detached leaf bioassay and no-choice cage technique under greenhouse conditions. The plants showing consistent biological activity will be planted under contained field conditions.

HC Sharma and KK Sharma

Activity 1.2.3.3: Transgenic groundnut resistance to fungal pathogens and viruses
Team: KK Sharma, F Waliyar and P Lava Kumar

Evaluation of transgenic groundnut for Indian peanut clump virus (IPCV) resistance: For the Indian peanut clump virus (IPCV), permission to carry out contained field trial of the selected 5 events of groundnut transgenic plants having coat protein or replicase genes was obtained from DBT. Contained field evaluation against IPCV was initiated at ICRISAT during the monsoon season of 2005. Five IPCV-H transgenics events IPCV cp1, IPCV cp12, IPCV cp46, IPCV rep6, and IPCV, rep21 were evaluated in a IPCV sick plot (Field# RCW17A) during 2005 rainy season at ICRISAT. Test plants along with control (JL 24) were sown in a split-plot design. Every test plant was assayed by DAS-ELISA at two weeks intervals, and plants were observed for symptoms. IPCV infection was very low in transgenic plants and also in controls (about 5%). Maximum number of infected plants were in IPCV cp46. However, two transgenic plants, IPCV cp12, and IPCV cp46, showed initial infection (tested virus positive in ELISA), but in subsequent assays were virus negative. It is likely that these two plants have potential resistance to IPCV. Due to delay in the rainy season and erratic and heavy rainfall during the plant growth season, a very low level (~10%) disease incidence was observed in the test plots, which did not allow conclusive validation of the results.

Standardization of mechanical inoculation procedure for IPCV: Evaluation of transgenic plants for IPCV resistance under field conditions was found to be difficult due to skewed inoculum distribution in the soil and erratic infection rate. Therefore, a reliable procedure for mechanical inoculation of IPCV was developed using innoculum prepared from IPCV-infected French bean sap extract at 1: 10 (w/v) dilution. This resulted in up to 90% infection and plants showed typical symptoms in 18 to 23 days after inoculation (Table 2.2). This procedure has proved to be very effective in evaluating IPCV resistance in transgenic plants, and is also applicable for testing resistance groundnut genotypes for resistance to this virus.

Evaluation for resistance to IPCV under greenhouse conditions: Using mechanical sap inoculation procedure, promising transgenic events IPCV cp12 (402 R1), IPCV cp1 (101-1 R2), IPCV rep6 (109-1 R3), IPCV rep21 (411-R2), and IPCV cp46 (304 R3) were evaluated for resistance (fifteen seeds per event, 3 seeds per pot). At the three-leaf stage, 12-plants were mechanically inoculated with 1: 10 (w/v) extract prepared from IPCV-infected French bean. Non-transgenic JL 24 plants were used as controls. Plants were monitored for virus infection by testing newly emerging leaves at weekly intervals by ELISA. At 3 weeks post inoculation (wpi), 8 to 50% of the plants tested positive, and infection peaked at 6 wpi (75 to 92%) (Figure 2.3). At 8 wpi, virus was not detected in emerging leaves of some plants leading to reduction in percent infection Nine transgenic plants tested negative to virus. Plants, rep21 p#3 and 9; cp12 p#16; cp1 p#38, during 5 and 6 wpi tested positive to virus, but in subsequent assays, were found to be negative (Table 2.3). It is likely that these 9 transgenic plants may have good resistance to IPCV.

Lava Kumar, KK Sharma, Farid Waliyar and SN Nigam

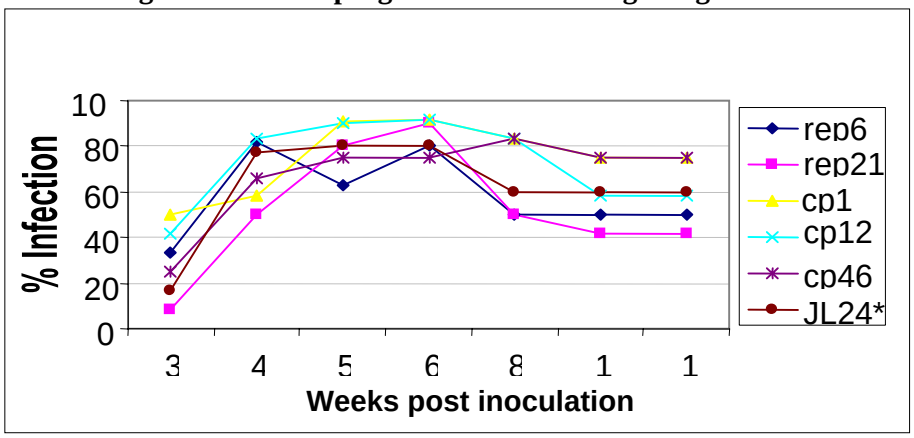
Table 2.2 Percent transmission of IPCV (Hyderabad isolate) by mechanical sap inoculation to groundnut (JL 24)			
Incubation period[1]	Infection[2] (%)	Temperature[3]	
		Max	Min
18	6/7 (85)	30.0	21.0
19	13/20 (65)	31.0	21.0
21	9/14 (64)	31.0	21.0
20	16/17 (94)	30.5	20.0
18	12/20 (60)	30.0	18.0
18	14/20 (70)	29.5	18.4
21	6/11 (55)	29.3	18.0
20	14/17 (82)	29.6	17.9
21	14/25 (56)	30.0	16.1
23	0/25 (0)	29.6	11.3
21	9/24 (37)	29.6	12.4
23	2/6 (33)	29.7	13.6
23	14/24 (58)	29.8	14.0
21	13/26 (50)	30.0	15.5
20	14/26 (54)	29.3	16.8
22	21/28 (75)	30.3	16.5
20.5	177/286 (62)	29.9	16.9

[1]Maximum number of days at which all the virus infected plants showed symptoms; [2]Infection confirmed by DAS-ELISA; [3]Mean temperature recorded during days to infection.

Table 2.3 Promising transgenic events for resistance to IPCV								
Event	Weeks after inoculation						At harvest	Symptoms
	3	4	5	6	8	10		
IPCV rep6 (P# 71)	-	-	-	-	-	-	-	S
IPCV rep21 (P# 3)	-	-	+	+	-	-	-	NS
IPCV rep21 (P# 9)	-	-	+	-	-	-	-	SS
IPCV rep21 (P# 11)	-	-	-	-	-	-	-	SS
IPCV rep21 (P# 12)	-	-	-	-	-	-	-	NS
IPCV cp1 (P# 38)	-	-	+	+	-	-	-	MM
IPCV cp1 (P# 40)	-	-	-	-	-	-	-	S
IPCV cp12 (P# 16)	-	-	+	+	-	-	-	SS
IPCV cp46 (P# 46)	-	-	-	-	-	-	-	S

S = Stunting. NS = No symptoms. SS = Severe stunting (clump) mm = Mild mosaic no stunting. All test plants were evaluated for virus by ELISA using IPCV antibodies.

Figure 2. 3 IPCV progress curve in transgenic groundnut



*All infected plants showed mosaic symptoms and stunting. IPCV incidence increased and reached peak during 6th week after inoculation, and from 8th week, virus in emerging leaves of some plants were not detected thus contributing to reduction in incidence. *JL24 is control*

Contained greenhouse and on-station contained field evaluation of peanut bud necrosis virus (PBNV) resistant transgenic plants: For the peanut bud necrosis virus (PBNV), the transgenic groundnut (cv. JL 24) plants of 48 independent events were produced with two vector constructs encoding nucleocapsid protein (NP) gene of PBNV through two different transformation systems. Integration of the transgene and stable genetic transformants in the T_0 and T_1 generations were assessed by PCR, RT-PCR, and Southern blot for coding regions of PBNV (NP) and *hpt*. Thirty-five independent events of T_1 generation were evaluated for resistance to PBNV under P_2 greenhouse conditions by using 100-times dilution of the virus inoculum. Of these, 24 lines showed less incidence (30%) compared to untransformed controls (100%). Transgenic plants were also evaluated in on-station contained field testing during the 2005 rainy season. Twenty-four transgenic events and 4 controls, were planted in RCBD design. Plants were sown in the month of August to take advantage of high thrip activity, and planting was done sparsely to create bare-earth effect to attract viruliferous thrips (30 cm gap between plants; 50 cm gap between rows). Field trial was monitored at fortnightly intervals for symptoms and virus infection. Controls recorded 80 to 100% infection. In transgenic plants, percent infection ranged between 37.5 to 100%. Event# PBNV (B)-1 had the lowest percent infection (37.5%) at the time of harvest. (Table 2.4).

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Table 2.4. On-station contained field evaluation of transgenic plants for resistance to PBNV during 2005 rainy season

	Plants infected / tested	**Infection (%)**
PBNV (B)-10	19/23	82.6
PBNV (B)-7	17/24	70.8
PBNV (B)-6	16/24	66.7
PBNV (B)-4	16/24	66.7
PBNV (B)-3	17/23	73.9
PBNV (B)-1	9/24	37.5
Control JL 24	17/24	70.8
PBNV (B)-11	14/24	58.3
PBNV (B)-12	20/23	86.9
PBNV (B)-14	19/24	79.1
PBNV (B)-20	19/24	79.1
Control JL 24	18/24	70.8
PBNV (B)-21	20/24	83.3
PBNV (A)-A	18/24	75.0
PBNV (A)-G	21/24	87.5
PBNV (A)-F	20/24	83.3
PBNV (A)-E	19/24	79.1
PBNV (A)-D	23/23	100
Control JL 24	20/24	83.3
PBNV (A)-C	16/23	69.6
PBNV (A)-B	22/23	95.6
PBNV (A)-H	22/22	100
PBNV (A)-I	18/24	75.0
PBNV (A)-J	15/24	62.5
PBNV (A)-K	18/24	75.0
PBNV (A)-L	22/24	91.6
PBNV (A)-N	16/22	72.7
Control JL 24	19/24	79.1
SE±		2.85

Development of transgenic plants for resistance to tobacco streak virus (TSV): The binary vector containing the coat protein gene of TSV (pCAMBIA 2300 TSVcp) was obtained from the Donald Danforth Plant Science Center (DDPSC) under a collaborative project supported by ABSPII. By using the *Agrobacterium*-mediated genetic transformation of cotyledonary explants, 50 putative transgenic events were produced and transferred to the containment greenhouse. The putative transgenics were confirmed through PCR and RTPCR and Western blots, and 15 events were advanced to T_1 generation in the containment greenhouse. The recovery frequency of transgenic events was 80%.

Sai Vishnu Priya, S Arockiasam and KK Sharma

Evaluation of transgenic groundnut containing rice chitinase gene for resistance to Aspergillus flavus by in vitro seed inoculation assay: Molecular characterization based on PCR of over 30 putative transgenic plants of groundnut transformed with rice chitinase gene in T_3 generation was carriedout. Preliminary bioassays with *Aspergillus flavus* (by inoculating the seeds of the selected 10 events) indictaed that 2 events had 0 to 10% incidence in terms of seed infection.

Transgenic groundnut containing rice chitinase gene was evaluated for *A. flavus* resistance by *in vitro* seed inoculation method. Seeds were inoculated with the *A. flavus* spores and incubated at 28 ºC for five days under high humid conditions. Promising T_1 events were advanced to T_2 generation. Seed from 170 plants were evaluated for *A. flavus* seed colonization. Seeds from 22 transgenic plants (event# 12, 18, 23, 24, 27, 29, 30, 31, 36, 44) that had seed infection of 0 to 10% were regenerated and advanced to T_3 (Table 2.5). Seed from 315 T_3 plants were tested for *A. flavus* seed colonization (Table 2.6). Seed infection rate in these varied from 0 to 100% (Figure 2.4). Twenty-two plants (event# 23, 27, 29, 31, 36, and 44) that had 0 to ≤10% seed colonization were selected and advanced to T_4 generation, which are under testing.

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Table 2.5. Transgenic groundnut selected for advancing from T_2 to T_3 generation

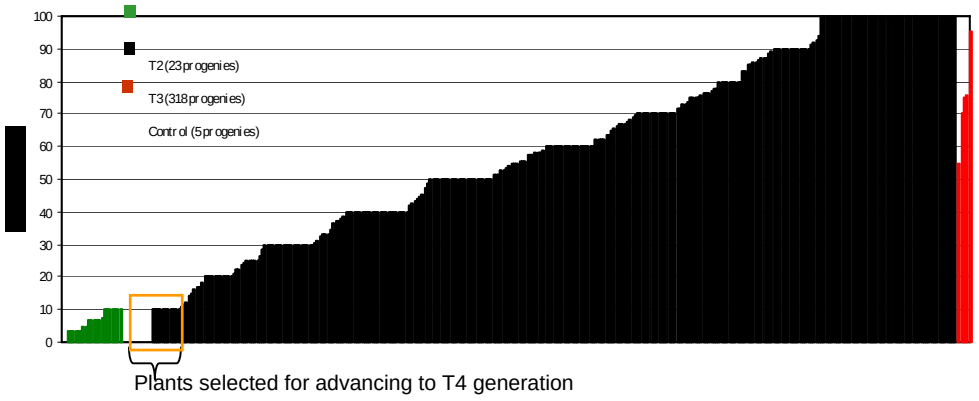
Event	***A. flavus* infection (%)**
12_12	0.00
18-6	3.33
29-12	3.33
36-3	3.33
44-2	3.33
44-11	3.70
27-13	4.76
23-11	5.00
23-10	6.67
29-4	6.67
29-6	6.67
30-7	6.67
44-7	6.67
29-1	7.41
29-3	8.89
23-12	10.00
24-1	10.00
27-12	10.00
30-1	10.00
30-4	10.00
31-3	10.00
36-2	10.00
Control	74.00
SE ±	0.65

Table 2.6 Transgenic groundnut selected for resistance to Aflatoxin advancing from T_3 to T_4 generation

Event identity	***A. flavus* colonization (%)**
23-11-1	0.00
29-3-4	0.00
29-3-5	0.00
29-6-2	0.00
29-12-1	0.00
31-3-15	0.00
31-3-17	0.00
36-2-4	0.00
36-2-14	0.00
44-7-20	0.00

Event identity	*A. flavus* colonization (%)
44-11-2	0.00
23-12-8	10.00
27-12-4	10.00
29-6-11	10.00
31-3-16	10.00
36-2-1	10.00
36-2-2	10.00
36-2-9	10.00
36-2-15	10.00
36-2-21	10.00
44-7-14	10.00
44-7-21	10.00
Control	95.00
SE±	1.14

Figure 2.4. Aspergillus flavus colonization (%) in transgenic groundnut (T2 and T3)



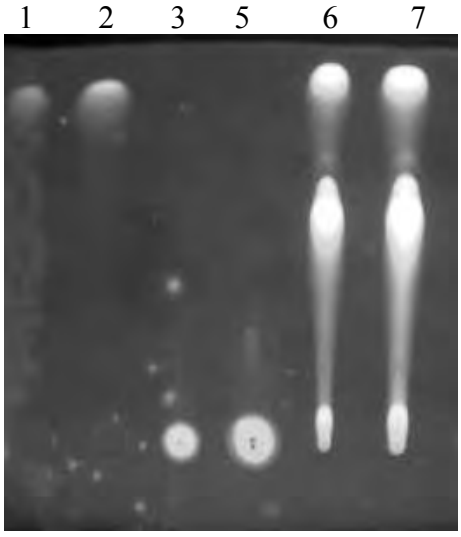


Figure 2.5. Evaluation of AFB1-ovalbumin standards by thin layer chromatography.

AFB1-ovalbumin adduct has no relative mobility, and thus fluorescence appears at the spot (lanes 3 and 4); and multiple spots in organic phase (lanes 6 and 7) suggests partial and unreacted AFB1

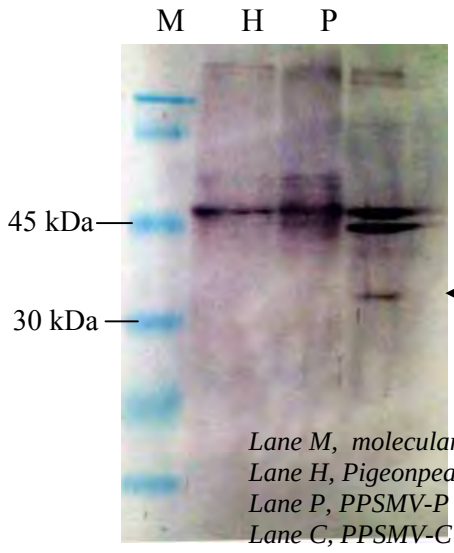


Figure 2.6. Western immuno blot assay using PPSMV-C polyclonal antibodies

The antibodies reacted with the PPSMV-C, but not with PPSMV-P. Note that the 50 kDa protein common in all preparations is large subunit of Rubesco, which co-precipitates with the virus protein.

Output 1.2.4: Diagnostic tools and bio-safety assessment of the products of biotechnology

Activity 1.2.4.1: Diagnostic tools for mycotoxins and viruses

ELISA for the detection of aflatoxins in human serum: Exposure of humans and animals to Aflatoxin (AFB1) results from the consumption of contaminated food. AFB1 covalently binds to lysine moiety of serum albumin (AFB1-lys adduct). AFB1-lys adduct has been identified as a useful biomarker to determine the human exposure to aflatoxins, and to assess the exposure risk among various sections of population. For this purpose, an indirect competitive ELISA was developed for quantitative estimation of AFB1-lys adducts in human serum albumin. Protocols for preparation of standards, AFB1-lys, and AFB1-ovalbumin adducts were developed using AFB1-8,9 epoxide (Figure 2.5). These were used at various concentrations to standardize ELISA procedure using rabbit polyclonal antibodies produced at ICRISAT, and also with the antibodies obtained from the University of Columbia, USA (which was used as reference for comparative assay). A simple method for extraction of albumin from serum fraction was standardized and used for estimating AFB1-lysine concentration by ELISA. The ELISA can detect up to 1.9 pg AFB1-lys per 75 µl serum sample using reference antibody. The assay has 97.4% recovery of AFB1-lysine adduct in artificially spiked samples. However, antibodies produced at ICRISAT resulted in weak reaction and were not useful for detection of AFB1-lys adduct. ELISA test was

Table 2.7 Concentration of AFB1-lysine pg mg^{-1} albumin (two replications)

HBV positive sample	AFB1-lys (pg mg^{-1} albumin)
1.116	0
1.165	0
1.2	0
1.20	0
0.976	30.18
1.089	0
1.198	0
1.205	0
1.198	0
1.11	0
1.042	8.5
1.162	0
1.181	0
0.98	19.42
1.024	5.14
1.121	0
1.134	0
1.121	0
1.131	0
1.081	0
Healthy control	0

validated using reference antibodies by analyzing Hepatitis B virus positive samples (Table 2.7). This revealed AFB1-lys levels in 4 samples (Table 2.7). This test will be validated by large scale testing of samples and new antibodies for AFB1-lys detection will be produced.

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Polyclonal antibodies to PPSMV-C isolate: Polyclonal antibodies were produced against the C isolate. Antibody titer in DAS-ELISA is 1: 2000 for detecting the virus in leaf extracts (1: 10 w/v). These antibodies strongly reacted with a 35 kDa protein of C isolate (Figure 2.6), but not with the P isolate. Using these antibodies, it is now possible to distinguish the C and P isolates. The antibodies will be validated by testing a large number of samples.

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Activity 1.2.4.3: Characterization of pigeonpea sterility mosaic virus (PPSMV) isolates and development of diagnostic tools

Using differential pigeonpea genotypes, PPSMV isolates from southern [Bangalore (B), Coimbatore (C)], central [Dharwad (D), Gulbarga (G), and Patancheru (P)] and northern [Varanasi (V)] India were studied. The genotypes inoculated with isolates P, D, and G showed similar phenotypic reaction, and were regarded as 'P' types (Table 2.8). The genotypes inoculated with B, C, and V isolates showed similar phenotypic reaction, but distinct from the P-types, and were regarded as 'B' type. The phenotypic reaction of C isolate was similar to that of B, but they differed in size of nucleoprotein (35 and 32 kDa, respectively). The B and P isolates have similar sized nucleoprotein, but have different phenotypic reaction on differential hosts. The study indicated occurrence of several PPSMV isolates with varying degrees of properties. Based on severity, various PPSMV isolates occurring in the Indian subcontinent can broadly be grouped as B and P types. The PPSMV isolates within each group have distinct physico-chemical characteristics. The B type isolates can overcome host-plant resistance selected against P types. The B types occur in northern and southern regions, and P types occur in the central regions of India.

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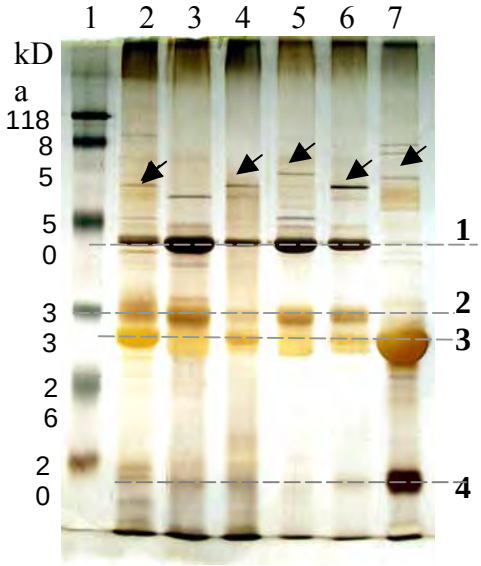
Studies on Helicoverpa armigera Nuclearpolyhedro virus (HaNPV) isolates in India: Six isolates of HaNPV obtained from different geographic locations from India were characterized [GAU-HaNPV, UASD-HaNPV, AK-HaNPV, TNAU-HaNPV, PAU-HaNPV,and ICRISAT-HaNPV]. These preparations were analyzed in 12% SDS-PAGE. Three major proteins of c 50, 32, and 30 kDa were detected in the page gels (Figure 2.7). The 50 and 32 kDa proteins were not detected in the GAU-HaNPV (Figure 2.7). The motilities of the 32 and 30 kDa protein slightly differed among various isolates. This could be due to variation in amino acid sequence or due to glycosilation. Viral DNA isolated from partially purified preparations was unsuitable for RFLP analysis.



Lane 1, Molecular weight marker
Lane 2, ICRISAT
Lane 3, UASD
Lane 4, TN
Lane 5, AK
Lane 6, PAU
Lane 7, GAU

Figure 2.7. Protein profiles of different isolates of HaNPV
Note that three major proteins (indicated as 1, 2, 3 and 4) are present in all but GAU isolate. The molecular weights of the major proteins are nearly similar, but not identical (also see Table 25). GAU sample (lane 7) is unique in that it lacks the c 32 and c 42 kDa protein. Several minor proteins are also seen in the gel (indicated with arrows), but there presence was not consistent in purified preparations.

kDa protein slightly differed among various isolates. This could be due to variation in amino acid sequence or due to glycosilation. Viral DNA isolated from partially purified preparations was unsuitable for RFLP analysis. Oligonucleotide primers will be designed to amplify, Bro-B gene and Hr-5 repeat region, which are known to be variable among various isolates, to study the diversity at the genome level.

Table 2.8. Response of pigeonpea genotypes to infection with P and Type B isolates of pigeonpea sterility mosaic virus occurring in India

Genotype	Type P isolates			Type B isolates		
	Patancheru (P)	Gulbarga (G)	Dharwad (D)	Bangalore (B)	Coimbatore (C)	Varanasi (V)
ICP 2376	RS	RS	RS	**SM**	**SM**	**SM**
ICP 7035	NS	NS	NS	NS	NS	NS
ICP 8862	NS	NS	NS	**MM**	**MM**	**MM**
ICP 8863	SM	SM	SM	SM	SM	SM

SM = Severe mosaic. MM = Mild mosaic. RS = Chlorotic ring spots.

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Activity 1.2.4.4.: Non-target effects of transgenic crops on beneficial natural enemies of crop pests
Team: HC Sharma and KK Sharma

Effect of Bt toxins and transgenic plants on the survival and development of the parasitoid, Campoletis chlorideae, and the coccinellid predator, Cheilomenes sexmaculatus, and other non-target insects. Four *Bt*-transgenic and the non-transgenic cotton genotypes were planted in a replicated trial under field conditions to have a better understanding of risk assessment approach for non-target arthropods. There was <10% parasitization of *H. armigera* eggs by *Trichogramma* spp. in both transgenic and nontransgenic cotton genotypes. The larval and pupal periods were longer, and larval weight and adult emergence of *C. chloridae* was lower on transgenic cotton genotypes (RCH 2 and Mech 12) as compared to their nontransgenic counterparts. A total of 40 insect specimens (25 from transgenic and 15 from non-transgenic cotton) were colleted, and were tested for the presence of *Bt*-toxin using qualitative ELISA. Among the 25 insect species colleted from transgenic plots, 7 showed high levels of *Bt*-toxin, 9 showed low levels, and the remaining 9 had no *Bt*-toxin. The transgenic cotton genotypes under sprayed and unsprayed conditions had more number of maturing bolls, good opened bolls, and lower damage in the green fruiting bodies by *H. armigera* than their nontransgenic counterparts at maturity.

Cry1Ac (LC_{50}) intoxicated *H. armigera* larvae increased the larval period of *C. chlorideae* by two days, and reduced the cocoon formation, adult emergence, and adult weight. There was no effect of *Bt* toxins on the fecundity of *C. chlorideae*. The adverse effects of *Bt* toxins on *C. chlorideae* were through early mortality of *H. armigera* larvae, and not through direct effects of the *Bt* toxins, since no traces of the toxins were found in the *C. chlorideae* cocoons and adults with the ELISA test. Studies on the host preference of *H. armigera* and it's larval parasitoid, *C. chlorideae* among six host crops of *H. armigera* (cotton, groundnut, chickpea, pigeonpea, sorghum, and pearl millet) indicated greater recovery of *H. armigera* larvae on pigeonpea and chickpea. But, chickpea was the least preferred host of *C. chlorideae* for the parasitization of *H. armigera* larvae. There was significant influence of host insects (*H. armigera*, *H. assulta*, *Spodoptera litura*, *S. exigua*, *Achoea janata*, and *Mythimna separata*) on the developmental biology of *C. chlorideae*. Two other insect species tested as xxxx (*Corcyra cephalonica* and *Sesamia inferens*) were not parasitized by the parasitoid, *C. chlorideae*. The larval period was significantly longer on *A. janata*, *S. exigua*, and *M. separata* (10.5 to 11.7 days) as compared to those reared on *H. armigera*, *H. assulta*, and *S. litura* (8.1 to 8.5 days). *Campoletis chlorideae* had longer pupal period on *A. janata* than that on *H. armigera*. The cocoon formation and adult emergence of the parasitoid were also highest on *H. armigera* (81.6 and 70.1%) than that on other hosts. The culture of the coccinellid predator, *Cheilomenes sexmaculatus*, and its aphid host, *Aphis craccivora*, has been established in the greenhouse to initiate studies on non-target effects of *Bt* toxins on generalist predators in the eco-system. An experiment was also conducted on the mating behavior of *C. chlorideae*. There were no significant differences in parasitism efficiency of the mated or unmated females. However, the unmated females produced only male offspring.

HC Sharma and MK Dhillon

Compatibility of Bt transgenic plants with the entomopathogenic fungi: In an attempt to study genetic diversity and identify virulent strains of entomopathogenic fungi for pest management, we evaluated several fungal isolates for *H. armigera* management in collaboration with Andhra University, Vishakhapatnam, Andhra Pradesh, India. The BB 2 isolate of *Beauveria bassiana* (1×10^7 conidia per ml) in combination with neem oil (0.3%) resulted in 87.7% mortality of *H. armigera* neonate larvae (74.4% larvae with mycosis) compared to

42.2% mortality with with neem oil alone, and 70% with *B. bassiana* alone. In another experiment, BB 2 and ITCC 4688 strains of *B. bassiana* and PADP 11 and PADP strains of *Nomuraea rileyi* were evaluated for their bio-efficacy in combination with an emulsifier, rapeseed oil, linseed oil, trehalose, skim milk powder, and carboxy methyl cellulose. The *B. bassiana* strain ITCC 4688 in combination with emulsified rapeseed oil resulted in 93.66% mortality of second-instar larvae of *H. armigera* compared to 85.33% mortality with the fungus conidia alone. The *N. rileyi* strain PADP 11 in combination with emulsifiable linseed oils resulted in 89.9% mortality of second-instar larvae of *H. armigera* compared to 86.66% mortality with the fungus alone. However, maximum mycosis (95.45%) was observed with fungus conidia in carboxy methyl cellulose. In case of tobacco caterpillar, *S. litura*, maximum mortality (91.5%) of the second-instar larvae was observed with *B. bassiana* PADP 11 conidia with emulsified linseed oil compared to 81.5% mortality with the fungal conidia alone, while maximum mycosis (90.17%) was observed with fungal conidia in carboxy methyl cellulose and emulsifier.

Uma Devi and HC Sharma

Improved abiotic stress tolerance, agronomic and quality traits via the application of genomics, genetic engineering and wide-hybridization

Drought is globally the most important constraint to crop productivity and with predictions of greater water scarcity in the future, drought is likely to remain the number one constraint. As options for irrigation are often not available in the semi-arid tropics (SAT), it is critical that genetic enhancement strategies focus on maximizing extraction of available soil moisture and improving the efficiency of water use in crop establishment, growth, biomass production and seed yield. Genetic improvement for drought tolerance has always been a challenge to conventional breeding approaches that rely on selection for yield in drought-stressed environments. The large genotype × environment interaction for yield in natural stress environments often makes direct selection for yield ineffective. Biotechnological tools provide targeted approaches for improving component traits of drought tolerance, which should be more effective than the conventional breeding methods in developing drought tolerant germplasm.

This global project focuses on identifying molecular markers for the quantitative trait loci (QTLs) controlling traits contributing to drought tolerance/avoidance in pearl millet, sorghum, chickpea and groundnut, and on the marker-assisted introgression of these QTLs into adapted cultivars/farmer parental varieties, elite breeding lines. Based on available information on their relative importance in conferring drought tolerance/avoidance, we have selected traits for improvement in each crop, for example, stay green in sorghum, deep and vigorous roots in chickpea, and water use efficiency in groundnut. In pearl millet, we have pursued QTLs associated with maintenance of grain yield under terminal drought situations.

The global project is also evaluating transgenic technology for developing drought tolerant plants in chickpea and groundnut. The genes presently being tested include *DREB1A* and *P5CSF129A*. The transcription factor, *DREB1A* driven by a drought-responsive *rd29A* promoter is expected to enhance tolerance to several abiotic stresses, such as drought, chilling temperature and salinity. The gene *P5CSF129A* increases proline accumulation and improves tolerance to osmotic stress.

Another important abiotic stress being targeted is soil salinity, which is commonly found in arid regions that rely on irrigation for agriculture. We are pursuing efforts to identify sources of salinity tolerance in all five ICRISAT mandate crops. Existing mapping populations have been developed in pearl millet that will be used for identification of QTLs for salinity tolerance. In sorghum, new mapping populations are being developed for this purpose. Similar work will be initiated in ICRISAT mandate legume crops when suitable parents are identified for development of mapping populations.

We are also seeking to improve the ability to obtain and utilize key soil nutrients in our mandate crops. For example, we have found significant variation in phosphorous (P) acquisition ability from low P soils in pearl millet. We are investigating the underlying mechanisms in P acquisition ability of plants and making efforts to identify markers for this important trait.

In the past, the breeding efforts in crop improvement have largely focused on genetic enhancement of yield potential and resistance to biotic and abiotic stresses. The emphasis on quality traits is increasing in recent years and is expected to increase in the coming years. We are presently targeting the improvement of the pro-vitamin A (β-carotene) content in pearl millet, groundnut and pigeonpea; methionine content in pigeonpea; stover ruminant quality in pearl millet; and feed and fodder quality in groundnut and pigeonpea. Marker-assisted

breeding will be used for improvement of all these traits, except for β-carotene content in groundnut and pigeonpea, and methionine content in pigeonpea, for which transgenic technology is being used.

The concerted and focused efforts underway will lead to development of diversified breeding populations/lines of ICRISAT mandate crops with improved tolerance to abiotic stresses, improved nutrient uptake and utilization, improved nutritional quality of grain, and improved quality of feed, fodder and stover. We will make these available to all public and private sector plant breeders and seed producers globally, who will, in turn, use these to produce locally adapted varieties with improved tolerances and quality traits for SAT farmers who depend upon our crops for their livelihoods.

Highlights for 2005

Marker-assisted introgression of drought tolerance QTLs in pearl millet

- Experiments were conducted to test whether roots have any role to play in the QTLs identified for terminal drought tolerance in pearl millet. The roots of tolerant genotypes reached a length of up to 240 cm in those plants exposed to water stress, whereas no sensitive pearl millet genotype had roots longer than 210 cm under water stress. The pearl millet genotypes having tolerance to terminal drought also had larger "investments" in deeper rooting than the drought sensitive pearl millet genotypes. Expressed as a percentage of roots in the 0-30 cm layer (to compare to shallow rooting), tolerant pearl millet genotypes had 15-20% of the roots present in soil layers deeper than 150 cm, whereas sensitive pearl millet had less than 5% of roots deeper than 150 cm.

- Products of marker assisted backcrossing (MABC) were advanced by one generation for development of drought tolerance QTL-NILs in the background of elite maintainer line 841B having introgressed alleles from LG 1 and LG 6 of donor 863B. Products of MABC for drought tolerance QTL-NILs with LG 2 substitutions from donor 863B in the background of elite maintainer line 841B were tested in second year. Conducted fourth year of testing of products of MABC for drought tolerance QTL-NILs with LG 2 substitutions from donor PRLT 2/89-33 in the background of elite restorer line H 77/833-2. Products of MABC for drought tolerance QTL from LG 2 of donor PRLT 2/89-33 in background of elite pollinator H 77/833-2 were retested.

Marker-assisted breeding for stay-green QTLs in sorghum

- Experiments were conducted to test whether roots have any role to play in the QTLs earlier identified for terminal drought tolerance in sorghum (QTLs that were detected for the stay-green component of terminal drought tolerance). Roots of stay-green trait donors and stay-green QTL introgressed materials reached a length of at least 270 cm under water stress, whereas senescent genotypes ISIAP Dorado and R 16 had a maximum root length of 210 cm. Again, expressed as percentages of roots in the 0-30 cm layer stay-green sorghums had 15-35% of the roots present in soil layers deeper than 150 cm, whereas senescent sorghums had only 5-10% of roots deeper than 150 cm.

- Progenies from MABC introgression of several putative stay-green QTL into released cultivars ISIAP Dorado were evaluated. Genetic differences in stay green expression were clear in the case of the highly senescent R 16, but less so in the case of the moderately senescent ISIAP Dorado. None of the backcross progenies expressed the stay-green trait to the same degree as the donor parent B35, but most have only one or two stay-green QTL, in contrast to B35, which has a larger number (six or more).

- BC_3F_2 and BC_4F_2 plants homozygous for donor parent alleles at SSR marker loci flanking each of six individual stay-green loci (i.e., single-QTL introgression lines for stay-green QTLs stgA, stgB, stg1, stg2, stg3 and stg4) from donor parent B35 in the genetic background of ICSV 111 (released in Ghana as 'Kapaala') and its sub-selection S 35, were identified and their selfed seed harvested. These included ten BC_4F_2 plants expected to be homozygous for stgA, three BC_4F_2 plants expected to be homozygous for stgB, six BC_3F_2 plants expected to be homozygous for stg1, ten BC_3F_2 plants expected to be homozygous for stg2, two BC_3F_2 and seven BC_4F_2 plants expected to be homozygous for stg3, and four BC_4F_2 plants expected to be homozygous for stg4. The corresponding BC_4F_3 and BC_3F_3 families, along with seed samples of their recurrent and donor parents, were sent to the national sorghum breeding program in Ghana (SARI) for agronomic and farmer-participatory evaluation.

Marker-assisted breeding for drought-avoidance root traits in chickpea

- Two experiments were conducted to assess how much and at what stage deep rooting actually contribute to water extraction, in particular for conferring tolerance to terminal drought. Three genotypes with deep and

profuse rooting (ICC4958, Annigeri and ICC8261), and two genotypes with shallow rooting (ICC1882 and ICC283) were used. Preliminary results show that deep rooted genotypes extract more water from the 60-90 cm layer at 25 days after sowing and the 90-120 cm layer at 34 DAS, than shallow rooted genotypes.

- The genetic components of root characteristics were investigated through generation mean analysis, using six basic populations (P1, P2, F_1, F_2, BC_1, and BC_2) of two crosses (ICC283 × ICC8261 and ICC4958 × ICC1882). The additive gene effect played an important role in controlling root length density (RLD) in both the crosses.

- A RIL mapping population developed from the cross ICC4958 (large roots) × ICC1882 (small roots) was evaluated for root traits. The plants were grown in cylinder culture systems and the root traits were studied at 35 DAS. The root system of each entry was divided into 4 pieces of different soil depths (0-30 cm, 30-60 cm, 60-90 cm; and 90-120 cm) and scanned to obtain digital images. Later, the dry weight was recorded. All digital images were analyzed using the image analysis software WinRhizo.

Marker assisted breeding for water use efficiency in groundnut

- The two parents TAG24 (low TE) and ICGV86031 (high TE) were tested along with 318 RIL F8 progenies from their cross. Preliminary analysis indicated a good correlation with data gathered in 2004. This was so despite the fact that the range of TE values obtained in 2005 (1.9 – 2.4) was less than the range obtained in 2004 (2.5 – 3.7). In 2005, we also received the delta $\Delta^{13}C$ data from the 2004 phenotyping experiment and found the well-reported negative correlation between TE and $\Delta^{13}C$.

- Parents (TAG24 and ICGV86031) of the RIL population phenotyped for WUE were screened with 463 SSR markers. Thirty-five markers were found to be polymorphic and were screened on 318 RILs.

- Eighteen groundnut accessions with 7 control cultivars were evaluated for pod yield potential and other traits related to drought. ICGs 5745, 6766, 7243, and 14523, ICG 14475 had 129-150 and 132-153 Specific Leaf Area (SLA) at 60 and 80 days after sowing (DAS) and 42-44 SPAD Chlorophyll Meter Reading (SCMR) at both DAS. ICG 2773 and ICG 5827 produced greater pod yield than the high yielding control cultivars ICG 44 and ICG 76. SCMR and SLA data were recorded groundnut for composite collection consisting of 850 accessions at 60 and 80 DAS. ICGs 2741, 5725, 5728, 6323, and 7878 were identified for high SCMR (53.9 – 61.0).

Transgenic drought tolerance in chickpea and groundnut

- Seven events of the C235 chickpea genotype transformed with *P5CSF* gene were tested in our common dry-down protocol. Higher cumulative transpiration values were recorded in all the transgenics than in C235 wild type under water stress. The total transpirable soil water (TTSW), i.e., the amount of water that plants were able to extract from the soil, was higher in all transgenics than in the wild type under water deficit. Although we found significant differences in TE, we found no advantage of the transgene for TE compare to the wild type in any of the 7 events tested.

- Five transgenic groundnut lines from parent JL24, containing a *DREB1A* gene, plus JL24 were tested for the response of transpiration to progressive soil drying. Results are very promising for several reasons: (i) the transgenic lines RD2 shows a significantly lower FTSW threshold where transpiration declines than JL24; (ii) The transpiration efficiency of this line is circa 50% higher than JL24; (iii) The TE of most transgenic lines is higher under well-watered conditions and it seems to correlate with different stomatal behavior, i.e. stomatal conductance (Gs) was lower in transgenics, in full agreement with the theory explaining differences in TE; (iv) There was a close correlation (R^2=0.87) between the FTSW threshold where transpiration begins to decline and TE, i.e. genotypes with more conservative behavior and early stomatal closure, like JL24, had lower TE than lines having late stomatal closure, like transgenic line 2; and (v) There was no correlation between TE and delta $\Delta^{13}C$.

Mapping salinity tolerance in pearl millet and sorghum

- The 24 parental lines of 12 existing ICRISAT pearl millet mapping populations were further assessed under salinity and three contrasting pairs of parents were identified. These mapping populations are being used to identify QTLs associated with salinity tolerance. Forty TRAP markers based on gene sequences for three enzymes expected to be involved in salinity stress tolerance were developed and mapped.

- SSR-marker-based genetic diversity assessment was initiated for of a set of 30 sorghum genotypes identified as either sensitive or tolerant to salinity stress. Initial results, based on information generated

using 76 SSR marker primer pairs, indicate that there is substantial genetic diversity within the range of productive and apparently salinity tolerant breeding lines and elite varieties tested, and that sufficient marker polymorphism is available to allow genotyping of mapping populations that could be produced by crossing selected pairs of salinity sensitive and tolerant genotypes.

Mapping salinity tolerance in chickpea, groundnut and pigeonpea

- A large screening of chickpea genotypes was screened for salinity tolerance. A 6-fold range of variation in the seed yield per pot was observed. Some genotypes had a 20% higher seed yield under salinity than the best released variety for salinity tolerance, CSG8962. The parents of ICCV 2 × JG 62 mapping population showed high contrast for seed yield under salinity. Thus, this mapping population will be used to map QTLs for salinity tolerance.

- A total of 288 groundnut genotypes were tested for salinity tolerance. A 5-fold variation in pod numbers across genotypes was found. Breeding line ICCV92206 was among the best. A poor relation between pod number and seed yield was observed.

- In pigeonpea, a set of genotypes, including newly set up mini-core collection, wild relatives (*acutifolius*, *platycarpus*, *sericeus*, *albicans*), wild derivative from the breeding program, and few hybrids were screened for salinity tolerance. Wide variation for biomass production was found at vegetative stage (biomass at 50 DAS), with a large contrast between accessions *C. scaraboides* and of *C. sericeus*. This opens the possibility to develop mapping populations from interspecific crosses.

Marker-assisted breeding for phosphorus acquisition ability in pearl millet

- A protocol was developed to measure root acidification of pearl millet seedlings in the lab. This protocol combines pH measurement and image analysis and could potentially be used for large screening. The current orientation of this work is to modify this method to make measurement of acidification in older plants and correlate results with measurements at the seedling stage.

- The phenotyping of the testcross hybrids obtained from the combination of four different male-sterile lines and the mapping progenies of the crosses between 81B-P6 and ICMP 451-P8, and between LGD 1-B-10 and ICMP 85410-P7 was undertaken. Plants receiving the low P treatment appeared to be very stunted compared to those receiving the control treatment.

- A series of experiments were conducted to address four issues (i) the differences in Olsen P in the different batches of soil, (ii) the control of plant to plant variation; (iii) the source of nitrogen; and(iv) the possibility to favor initial plant establishment with early application of tiny doses of soluble P.

Genetic engineering of groundnut for enhancement of β-carotene and of pigeonpea for enhancement of methionine and β-carotene

- *Agrobacterium*-mediated genetic transformation of the selected genotypes of groundnut was carried out by using newly constructed binary vectors containing ß-carotene genes (*psy1* and *crtB*). The putative transgenic groundnut shoots obtained under antibiotic selection pressure were elongated for rooting followed by their transfer to the containment greenhouse.

- The primary putative transformed pigeonpea shoots obtained under antibiotic selection pressure are being analyzed by PCR analysis for the presence of *SSA* and *psy1* genes. The expression of the *SSA* gene in transgenic tobacco (used as a modal system) was verified by RT-PCR analysis of the T1 seeds.

Improved pearl millet stover quality

- QTL analysis was performed by composite interval mapping for stover quality traits of individual stover fractions (stem, sheath and leaf blade) as well as whole stover samples. For stem fraction digestibility and metabolizable energy content, a major QTL was identified on LG 2, at a position similar to that for drought tolerance alleles from the 863B parent (suggesting it should be simple to combine improved stem digestibility with greater drought tolerance in the genetic background of elite seed parent maintainer line ICMB 841). Another major stover quality QTL was mapped to LG 5, which controlled significant proportions of phenotypic variation for *in vitro* true digestibility and for metabolizable energy content.

- For the putative LG 7 stover quality QTL, RFLP and SSR marker data were used to identify introgression homozygotes for the associated region from donor parent 863B in the genetic background of ICMB 841. Several such plants were identified and their selfed seed harvested for future testcross hybrid production.

- We completed the second and final cycle of FS progeny selection for improved stover quality in the released pearl millet variety ICMV 221. As in the first cycle, there were highly significant differences for all stover quality traits and agronomic variables measured, among the 280 FS progeny evaluated. Selected progenies will be recombined to form both grain and dual-purpose versions of the original variety with improved stover nutritional quality

Output 1.1.1: Enhanced Drought Tolerance in Pearl Millet and Sorghum

Activity 1.1.1.1: Marker-assisted introgression of drought tolerance QTLs in pearl millet
Team: CT Hash, V Vadez, F Bidinger, SMH Rizvi and S Chandra

Milestone: Root traits in LG 2 drought tolerance QTL-NIL of pearl millet characterized (2006)

The main purpose of experiments conducted in 2005 was to test whether roots have any role to play in the QTLs identified for terminal drought tolerance in pearl millet (QTLs that were detected for differences in panicle harvest index, PNHI, under conditions of terminal drought stress). We used four contrasting parents (drought sensitive H 77/833-2 and 841B, and drought tolerant PRLT 2/89-33 and 863B), plus two QTL-NILs in which a major drought tolerance QTL from LG 2 has been introgressed from donor parent PRLT 2/89-33 into the genetic background of H77-833/2 via several cycles of marker-assisted backcrossing (ICMR 01029 and ICMR 01031).

In a first experiment, we grew plants in 18 cm diameter 120 cm deep PVC cylinders, during September-December 2004. Twenty replicated plants were grown for each genotype, under well-watered conditions for the first 2 weeks after sowing. Thereafter, half of the plants were exposed to water stress by withholding irrigation while the other half was kept under well-watered conditions (control). One set of plants (5 replicated plants per genotype in stress and control) was harvested at 35 DAS (days after sowing) and the second set was harvested at 67 DAS. The second experiment was planted during March-April 2005. This experiment was basically a repeat from the first experiment, except that cylinders used were 200 cm long (same diameter), the second harvest was done at 63 DAS, and the genotypes evaluated included inbred lines 841B and 863B, the parental lines of a second pearl millet terminal drought tolerance mapping population. At harvest, the entire root system was gently pulled out after washing away the soil, its entire length measured and thereafter sliced in 30 cm portions to measure dry weight and root length density of the different portions.

Data of the first and second experiments were very consistent with each other, despite the differences in cylinder length, and therefore results of the second experiment are discussed. In pearl millet, roots of tolerant genotypes reached a length of up to 240 cm in the second harvest in those plants exposed to water stress, whereas no sensitive pearl millet genotype had roots longer than 210 cm under water stress. We also looked at the root dry weight in each of the 30 cm layers. We found that all 4 pearl millet genotypes having tolerance to terminal drought had also larger "investments" in deeper rooting than the drought sensitive pearl millet genotypes. Expressed as a percentage of roots in the 0-30 cm layer (to compare to shallow rooting), tolerant pearl millet genotypes had 15-20% of the roots present in soil layers deeper than 150 cm, whereas sensitive pearl millet had less than 5% of roots deeper than 150 cm.

These results are very encouraging as they show a significant and consistent involvement of roots in the terminal drought tolerance QTLs of pearl millet. The protocol used to pinpoint these differences can be further refined.

***V** Vadez*

Milestone: The leaf gas exchange response (photosynthesis, Gs) to drought and TE assessed in LG 2 drought tolerance QTL NIL of pearl millet (2006)

One experiment was carried out to compare the response to soil drying and transpiration efficiency of various parental lines of pearl millet genotypes contrasting for terminal drought tolerance. Pearl millet genotypes used were ICMR 1029, ICMR 1031, H 77/833-2, and PRLT 2/89-33. In this experiment, carried out during October-November 2004, we found that pearl millet genotypes differed very little for the threshold of soil moisture

where transpiration starts to decline. We also measured transpiration efficiency in that experiment and found that all the genotypes had very similar TE.

V Vadez

Milestone: Products of markers-assisted backcrossing (MABC) for drought avoidance QTLs tested in pearl millet (2006)

Products of MABC were advanced by one generation for development of drought tolerance QTL-NILs in the background of elite maintainer line 841B having introgressed alleles from LG 1 and LG 6 of donor 863B. Products of MABC for drought tolerance QTL-NILs with LG 2 substitutions from donor 863B in the background of elite maintainer line 841B were tested in second year. We conducted the fourth year of testing of products of MABC for drought tolerance QTL-NILs with LG 2 substitutions from donor PRLT 2/89-33 in the background of elite restorer line H 77/833-2. Products of MABC for drought tolerance QTL from LG 2 of donor PRLT 2/89-33 in background of elite pollinator H77/833-2 were retested. Obtained special project funding extension of 10 months to continue research in this activity area (supported by the DFID PSP pearl millet marker-assisted breeding project). The project activities are being conducted with national program partners. Six new CAZRI drought tolerance populations were advanced to F_2/BC_1F_1 population pairs and sent to CAZRI and the AICPMIP.

CT Hash, S Senthivel , P Satish Kumar and SMH Rizvi

Activity 1.1.1.2: Marker-assisted breeding for stay-green QTLs in sorghum
Team: CT Hash, V Vadez, F Bidinger, SMH Rizvi, R Folkertsma, M Mgonja, F Rattunde and S Chandra

Milestone: Techniques standardized for large-scale screening of root traits in sorghum.

The main purpose of experiments conducted was to test whether roots have any role to play in the QTLs thus far identified for terminal drought tolerance in sorghum (QTLs that were detected for the stay-green component of terminal drought tolerance). We used two elite drought-sensitive parents [ISIAP Dorado (partially senescent) and R 16 (highly senescent)], two stay-green donor parents (B35 and E 36-1), and two derivatives of R 16 thought to carry 2 or 3 major stay-green QTLs (RSG 03123 with drought tolerance from donor B35 and RSG 04012 with drought tolerance from donor E 36-1).

In the first experiment, we grew plants in 18 cm diameter and 120 cm deep PVC cylinders, during September-Dec 2004. Twenty plants were grown for each genotype, under well-watered conditions for the first 2 weeks after sowing. Thereafter, half of the plants were exposed to water stress by withholding irrigation while the other half was kept under well-watered conditions (control). One set of plants (5 replicated plants per genotype in stress and control) was harvested at 35 DAS and the second set was harvested at 67 DAS. The second experiment was conducted during March-April 2005. This experiment was basically a repeat from the first experiment, except that cylinders used were 200 cm long (same diameter), and the second harvest was done at 63 DAS. Genotypes ISIAP Dorado, E 36-1, and RSG 04012 were included in the second experiment. At harvest, the entire root system was gently pulled out of the cylinder after washing away the soil, its entire length measured and thereafter sliced in 30 cm portions to measure dry weight and root length density in these different portions.

Data of the first and second experiments were very consistent with each other, despite the difference in cylinder length, and therefore results of the second experiment are discussed. We found that roots of stay-green trait donors and stay-green QTL introgressed materials reached all a length of at least 270 cm in the second harvest under water stress, whereas senescent genotypes ISIAP Dorado and R 16 had a maximum root length of 210 cm. Again, expressed as percentages of roots in the 0-30 cm layer (to compare to shallow rooting), stay-green sorghums had 15-35% of the roots present in soil layers deeper than 150 cm, whereas senescent sorghums had only 5-10% of roots deeper than 150 cm. Interestingly, stay-green and senescent materials did not vary for root length under well-watered conditions.

These results are very encouraging as they show a significant and consistent involvement of roots in the stay-green QTLs associated with terminal drought tolerance in sorghum. The protocol used to pinpoint these differences can be further refined. As an attempt to do so, a third experiment was started with sorghum, where stress was imposed at 21, 35, 49, and 69 (flowering) days after sowing. At each date of stress imposition, one set of plants (5 replicated cylinders per genotype) was pre-harvested and two additional sets were submitted to drought and well-watered conditions. Harvest of the two sets undertaking treatment was done once wilting symptoms were visible on all plants, usually 3-4 weeks after treatment imposition.

V Vadez

Milestone: First products of marker-assisted backcrossing of stay-green QTL from donor E 36-1 into the backgrounds of S35 and IRAT 204 field-tested to assess terminal drought tolerance (2008)

We evaluated sets of progenies from MABC introgression of several putative stay-green QTL into released cultivars ISIAP Dorado (BC_3F_4 lines, with donor parent B35) and R 16 (BC_1F_3 lines, with donor parents B35 and E 36-1) in both supplementally irrigated and dryland environments in the postrainy season of 2004-2005. The objectives were to assess the expression of stay-green in the backcross progenies under postrainy season conditions, to compare the agronomic expression of the recurrent parents and their backcross derivatives, and to evaluate the effects of stay-green expression on grain filling and grain yield under the characteristic post-flowering drought stress of the postrainy season. Environmental (moisture) differences in stay-green expression were strong in both sets of materials; genetic differences in stay green expression were clear in the case of the highly senescent R 16, but less so in the case of the moderately senescent ISIAP Dorado. For example, the percent green leaf area 2/3 of the way through grain filling in the R 16 derivatives ranged from 7% to 29% in the dryland environment, compared to 34% to 57% in the ISIAP Dorado trial (in which stress was interrupted by a rain shower in mid grain filling). None of the backcross progenies expressed the stay-green trait to the same degree as the donor parent B35, but most have only one or two stay-green QTL, in contrast to B35, which has a larger number (six or more).

Many of the R 16 derivatives differed agronomically from the recurrent parent in height and flowering time, and some in grain size and stover yield. The majority were similar in grain yield to R 16 in both environments. However, most of the differences reflect the differences in the donor parents and R 16, and should disappear with further backcrosses. The derivatives of ISIAP Dorado were generally more similar to the recurrent parent (as would be expected from BC_3 materials), except for later flowering, and some tendency for the derivatives to have a greater stover yield and a smaller grain size. Stay-green expression during the second half of the grain filling period was related to relative (dryland environment/supplementally irrigated environment) grain size *(i.e.*, to completeness of grain filling) in both sets of materials: $r^2 = 0.61$ ($P < 0.01$) for R 16 (Figure 1.1) and $r^2 = 0.59$ ($P<0.01$) for ISIAP Dorado. The effect of differences in stay-green expression on grain size carried over into a significant effect on grain yield in the case of R16 ($r^2 = 0.48$) but not in the case of the ISIAP Dorado derivatives ($r^2 = 0.10$). We hypothesize that the difference in stay green effect in the two trials was due to both genetic (R16 is normally considerably more senescent than is ISIAP Dorado) and environmental (the rain shower during mid grain filling in the ISIAP Dorado trial) reasons. Selected R 16 evaluation will be repeated in 2005-2006, and

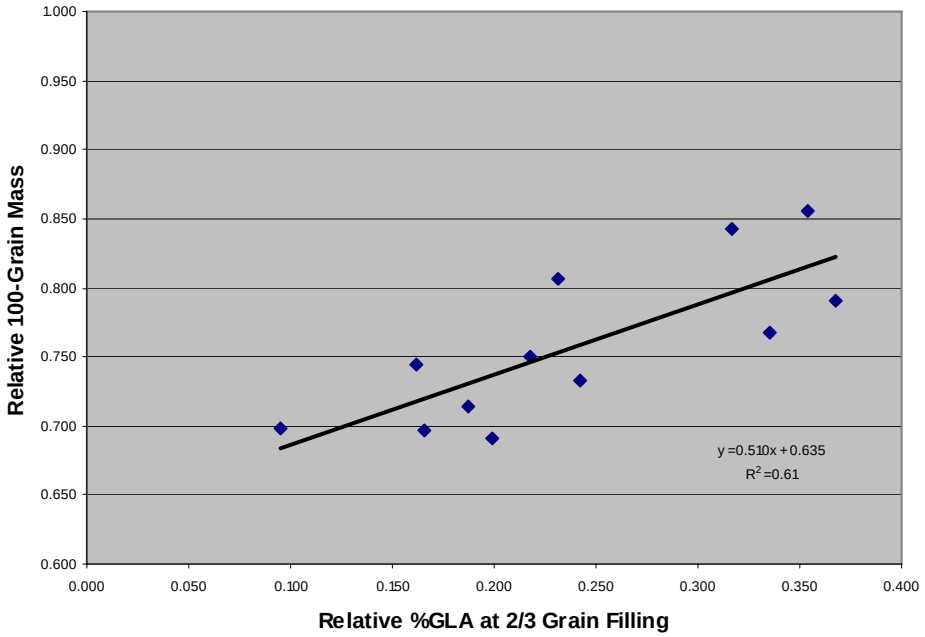

Figure 1.1. Relative (dryland/supplementally irrigated environments) 100-grain mass as a function of relative (dryland/supplementally irrigated environments) percent green leaf area (%GLA) at two-thirds of the way through grain filling for R 16 and its derivatives

better R 16 derivatives from 2004-2005 trial were backcrossed to R16 twice during 2005 prior to possible re-evaluation during 2006-2007.

FR Bidinger and CT Hash

Milestone: Sorghum stay-green introgression lines derived by marker-assisted backcrossing provided to African national programs in locally adapted, farmer-preferred genetic backgrounds (2008)

Due to lack of adequate SSR marker polymorphism between stay-green drought tolerance donor parent E 36-1 and the two recurrent parents (S35 and IRAT 204), and reduction in the amount of funding provided by granting agencies for activities related to this milestone, we have focused the marker-assisted backcrossing program for the sorghum stay-green trait on donor B35 only.

During 2004 marker-assisted backcrossing of genomic regions associated with the stay-green component of terminal drought tolerance of donor parent B35 was advanced two generations in the genetic background of elite recurrent parent Kapaala = ICSV 111. By the end of 2004, BC_4F_2 seed had been produced for families expected to segregate for four of the six target stay-green QTL in this genetic background (for which suitable BC_3F_1 families were available in 2003) and for the remaining two target QTL, seed had been produced for BC_3F_2 families.

During the first half of 2005, BC_3F_2 and BC_4F_2 plants homozygous for donor parent alleles at SSR marker loci flanking each of six individual stay-green loci (*i.e.*, single-QTL introgression lines for stay-green QTLs *stg*A, *stg*B, *stg*1, *stg*2, *stg*3 and *stg*4) from donor parent B35 in the genetic background of ICSV 111 (released in Ghana as 'Kapaala') and its sub-selection S 35, were identified and their selfed seed harvested. These included ten BC_4F_2 plants expected to be homozygous for *stg*A, three BC_4F_2 plants expected to be homozygous for *stg*B, six $BC3F_2$ plants expected to be homozygous for *stg*1, ten BC_3F_2 plants expected to be homozygous for *stg*2, two BC_3F_2 and seven BC_4F_2 plants expected to be homozygous for *stg*3, and four BC_4F_2 plants expected to be homozygous for *stg*4. The corresponding BC_4F_3 and BC_3F_3 families, along with seed samples of their recurrent and donor parents, were sent to the national sorghum breeding program in Ghana (SARI) in June 2005 in time for initial rainy season seed increase for agronomic and farmer-participatory evaluation under the Water for Food Challenge Program. Further, these progenies were advanced a further generation by selfing at ICRISAT-Patancheru during the 2005-06 post-rainy season to produce seed required for future assessment of their drought tolerance, agronomic performance and grain quality. Marker data generation in 2005 required to identify the putative QTL introgression homozygotes was performed as part of the M.Sc. thesis research program of Mr. Sripathi Venkateswararao. In late 2005 he submitted a thesis to Acharya N.G. Ranga Agricultural University (ANGRAU) India, and this was defended successfully in early 2006.

Similarly, marker-assisted backcross introgression of stay-green QTLs from donor parent B35 into the genetic background of recurrent parent IRAT 204 advanced two generations during 2005, with the marker data generation required being performed as part of the M.Sc. thesis research program of Mr. Sripathi Venkateswararao.

We hosted a Visiting Scientist from the Indian national program (Dr Madhasudhana Rao from the National Research Center for Sorghum) for 3 months to develop marker genotype data for assessing opportunities of MABC to improve stay-green drought tolerance of elite and landrace materials of interest to Indian sorghum breeders, and another from the Universidad Autonoma de Neuvo Leon (Dr F Zavala G) for 1 month to learn how to conduct SSR-MAS for the stay-green trait.

CT Hash

Output 1.1.2 : Enhanced Drought Tolerance in Chickpea and Groundnut

Activity 1.1.2.1: Marker-assisted breeding for drought-avoidance root traits in chickpea
Team: PM Gaur, V Vadez , J Kashiwagi, RK Varshney, D Hoisington and S Chandra

Milestone : Markers for drought-avoidance root traits in chickpea identified and verified in unrelated populations (2006*).*

Drought avoidance root traits in chickpea further dissected: Lot of knowledge on root traits in chickpea has been gathered in the past two decades. However, we have little understanding of how roots work and how much and at what stage deep rooting actually contribute to water extraction, in particular for conferring tolerance to terminal drought. Two experiments were designed to start addressing this issue, using three genotypes with deep

and profuse rooting (ICC4958, Annigeri and ICC8261), and two genotypes with shallow rooting (ICC1882 and ICC283). Plants were grown in cylinders made of assembled 30 cm portions of 10 cm diameter PVC tubes. Plants were grown under well-watered conditions for the first two weeks after sowing, after that irrigation was withdrawn in half of the tubes. Harvests were performed at different times after stress imposition and each time, tubes were split into 30 cm portions (0-30, 30-60, 60-90, and 90-120). Roots in each portion were separated. After separation, soil was homogenized quickly and a soil aliquot was taken, weighted, dried at 80ºC for two days and weighted again to determine water content. The main purpose of the experiment was to relate root length density in the different cylinder portions to water content remaining. Preliminary interpretation in the first experiment shows that deep rooted genotypes extract more water from the 60-90 cm layer at 25 DAS, and, the rooting profile moving further down, thereafter extract more water from the 90-120 cm layer at 34 DAS, than shallow rooted genotypes. These results need to be confirmed and refined but they have the potential to greatly simplify the screening of root traits. One particular interest of this technique would be to study how water uptake by deep rooting plants contributes to providing water at that time of seed filling.

V Vadez

Estimation of gene effects for drought avoidance root traits: The genetic components of root characteristics were investigated through generation mean analysis, using the means of six basic populations (P_1, P_2, F_1, F_2, BC_1, and BC_2) of two crosses (ICC283 × ICC8261 and ICC4958 × ICC1882). The additive gene effect played important role in controlling root length density (RLD) in both the crosses. Although the magnitudes of additive gene effects and additive × additive gene interactions of total RLD were different between two crosses, the direction of the gene effect was consistent towards increasing RLD in both the crosses, except 30-60 cm and 60-90 cm depth in ICC4958 × ICC1882. Although the magnitude of additive gene effect for RLD was relatively small, the cross ICC283 × ICC8261 was better than ICC4958 × ICC1882 to use in selection for larger total RLD, especially RLD in 30-60 cm depth, which is important for the seed yield under the terminal drought. The cross ICC283 × ICC8261 also showed substantial additive and additive × additive epistasis for the RLD in deeper soil section, which have greater impacts on yield under severe terminal drought. Recurrent selection in later generations would be effective for developing chickpea genotypes with higher root length density from the cross ICC283 × ICC8261 because of the relative amounts of dominance and epistasis effects.

J Kashiwagi, PM Gaur and S Chandra

Phenotyping of new mapping population for drought-avoidance root traits: A RIL mapping population developed from the cross ICC4958 (large roots) × ICC1882 (small roots) was evaluated for root traits. The plants were grown in cylinder culture systems and the root traits were studied at 35 days after sowing. The root system of each entry was divided into 4 pieces in different soil depth (0-30 cm, 30-60 cm, 60-90 cm, and 90-120 cm) and scanned to obtain the digital images. Later the dry weight was recorded. All digital images were analyzed using the image analysis software WinRhizo. The parental lines are being screened for identification of polymorphic markers.

J Kashiwagi, PM Gaur and RK Varshney

Activity 1.1.2.2: Marker assisted breeding for water use efficiency in groundnut
Team: SN Nigam, HD Upadhyaya, V Vadez, D Hoisington, RK Varshney and S Chandra

Milestone: QTLs conferring water use efficiency identified in groundnut (2007)

Phenotyping of mapping population for TE: A repeat of the phenotyping experiment carried out in 2004 was performed during the January-March period. The two parents TAG24 (low TE) and ICGV86031 (high TE) were tested along with 318 RIL F8 progenies from their cross. Experimental design used was a 8 × 40 lattice design with 5 replications. Data have also been sent to the biometric unit for spatial analysis and control whether unknown patterns of variation are likely to occur in the experiment. Another similar set of plants was cultivated to evaluate the pre-treatment biomass. Preliminary analysis has been done and shows a good correlation with data gathered in 2004. This was so despite the range of TE values obtained in 2005 (1.9 – 2.4) were far less than the range of values obtained in 2004 (2.5 – 3.7). This was probably related to the fact that a late rain (45 mm) came and refilled the field capacity a few days only before planned harvest. Plants recovered from drought and this may have contributed to those being the most susceptible to intermittent drought and having low TE. In 2005 we also received the $\Delta^{13}C$ data from the 2004 phenotyping experiment and found the well-reported negative correlation between TE and $\Delta^{13}C$.

V Vadez

Genotyping of mapping population with SSR markers: Parents (TAG24 and ICGV86031) of the RIL population phenotyped for WUE in groundnut were screened with 463 SSR markers. Thirty-five markers were

found to be polymorphic and were screened on 318 RILs. Another 450 SSR are available and will be screened in 2006.

RK Varshney

New sources of variability for drought tolerance traits: Evaluated 18 groundnut accessions with 7 control cultivars in a lattice design during 2004-05 post-rainy season, for pod yield potential and other traits related to drought. ICGs 5745, 6766, 7243, and 14523, ICG 14475 (129-150 and 132-153 Specific Leaf Area (SLA) at 60 and 80 days after sowing (DAS) and 42-44 SPAD Chlorophyll Meter Reading (SCMR) at both DAS) were identified as additional sources related to drought tolerant traits in comparison to control CMSG 84-1 (144 and 150 SLA and 43 and 42 SCMR at 60 and 80 DAS, respectively). ICGs 2773 and 5827 (3.39-3.43 t ha^{-1}) produced greater pod yield than the high yielding control cultivars ICGS 44 (2.87 t ha^{-1}) and ICGS 76 (3.21 t ha^{-1}). Leaf samples from these 18 accessions and control cultivars have been collected for DNA extraction. Finger printing using 20 SSR markers is in progress.

Groundnut composite collection consisting of 850 accessions was evaluated in an augmented design with four repeated control cultivars for pod yield potential and drought related traits. SCMR and SLA data was recorded on these accessions of groundnut composite collection at 60 and 80 DAS. ICGs 2741, 5725, 5728, 6323, and 7878 were identified for high SCMR (53.9 – 61.0). Leaf samples have been collected for DNA extraction. Finger printing using 20 SSR markers is in progress. Data for other agronomic traits is in process.

HD Upadhyaya

Activity 1.1.2.3: Transgenic drought tolerance in chickpea and groundnut
Team: KK Sharma, V Vadez, PM Gaur and SN Nigam

Milestone: Selected transgenic lines of chickpea and groundnut characterized for drought tolerance traits (2005*)*

Transgenic chickpea (P5CSF) characterized under drought: Seven events of C235 chickpea genotypes transformed with P5CSF gene were tested in our common dry-down protocol, in which we measured transpiration efficiency (g biomass per kg of water transpired, TE), and the soil moisture level (measured with the fraction of transpirable soil water, FTSW) where transpiration declines. Growth was slightly but significantly retarded in all the transgenic events at the time of pre-treatment harvest (about 30 DAS), whereby shoot dry weight of transgenic was in the range of 1.40-1.80 g $plant^{-1}$, compared to 2.45 g $plant^{-1}$ in C235. This trend was maintained at the end of the experiment under well-watered conditions where all transgenics kept a lower biomass (4.2 – 5.9g) than wild type C235 (6.6g). Under water stress, the final biomass was also somewhat higher in C235 (3.85g) than in all trangenics except one (3.20-3.55), although the transgenics had performed relatively better under drought than under well-watered conditions. This was consistent with the higher cumulative transpiration values in all the transgenics than in C235 wild type under water stress. This result fully agrees with the osmotic adjustment hypothesis, which predicts that increased turgor would help stomata to remain open to maintain transpiration rate high. It also agrees with the finding that the total transpirable soil water (TTSW), i.e. the amount of water that plants were able to extract from the soil, was higher in all the P5CSF transgenics than in the wild type under water deficit.

Although we found significant differences in TE, we found no advantage of P5CSF for TE compare to the wild type in any of the 7 events of P5CSF that were tested. All the TE values were in a narrow range of 2.10 – 2.80 g biomass kg^{-1} water transpired in well-watered plants and C235 had the highest values (2.80 g kg^{-1}). In agreement with this, we found large differences in the stomatal conductance (Gs) under well-watered conditions, with P5CSF transgenics having 3-4-fold higher Gs than C235. This correlated well with leave temperature, which was lower in transgenics than in C235, and with TE ($R^2 = 0.75$), whereby lower Gs correlated with higher TE, in agreement with the theory. Under water stress, we found that all the transgenics had a later initiation of stomatal closure compare to C235. TE values were in the range of 2.70 – 3.70 g kg^{-1} and C235 was in the middle of that range.

P5CSF is supposed to enhance proline production and confer drought protection of major physiological processes. We measured proline in drought stress plants but found very little differences between C235 and the transgenics. Confirmation measurements are currently being re-assessed as there was a doubt about the quality of the standards being used. We also hypothesized that the transgene contributing to osmoticum production may help keeping root growth active by maintaining turgor in root tips. Results were contrary to our expectation since C235 had larger roots than all transgenics both under well watered and water stress. However, root mass was relatively less reduced under drought stress compare to C235, indicating that root growth was somewhat improved in transgenic P5CSF under water stress.

Although there was no advantage of P5CSF for TE, the differences found in TE and in a number of aspects such as Gs, TTSW, etc. make the P5CSF transgene very interesting material to investigate certain important physiological mechanisms such as the process of water extraction by roots, or TE.

V Vadez

Transgenic groundnut (DREB1A) characterized for leaf gas exchange and TE: Two experiments using 5 transgenic groundnut lines from parent JL24, inserted with the DREB1A gene, plus JL24 have been tested for the response of transpiration to progressive soil drying. These 5 events have been chosen from a preliminary screening of 14 events + wild type JL24, in which a dendrogram based on the number of days to reach FTSW=0 and the FTSW threshold where transpiration declines. From this dendrogram, we have selected one event out of the 5 main clusters, therefore choosing events ranging from very similar to very different from the wild type. This is a major contrast with earlier transgenic studies where preference is given for transgenics having an extremely contrasting phenotype compare to the wild type.

Results are very promising for several reasons: (i) The transgenic lines RD2 shows a significantly lower FTSW threshold where transpiration declines than JL24; (ii) The transpiration efficiency of this line is circa 50% higher than JL24; (iii) The TE of most transgenic lines is higher under well-watered conditions and it seems to correlate with different stomatal behavior, i.e. stomatal conductance (Gs) was lower in transgenics, in full agreement with the theory explaining differences in TE; (iv) There was a close correlation (R^2=0.87) between the FTSW threshold where transpiration begins to decline and TE, i.e. genotypes with more conservative behavior and early stomatal closure, like JL24, had lower TE than lines having late stomatal closure, like transgenic line 2; (v) There was no correlation between TE and $\Delta^{13}C$. We do not have a clear explanation for this, even after consulting the experts in that domain. However, we interpret that the lack of relation could be explained by differences in the midday stomatal opening, whereby transgenics might close up stomata during the period of maximum VPD (usually 12noon-3pm) more so than JL24, thereby saving water. This behavior might be sufficient to have better water use, while not long enough to materialize in measurable difference in the $\Delta^{13}C$ discrimination ratio.

With the range of variation in TE obtained with transgenic lines, these data give new ground for exploring the physiological basis for high TE. We have gathered such preliminary data. We are now planning new experiments to: (i) document the root behavior of transgenic plant under stress; (ii) compare TE in the most promising transgenics to that of the best germplasm accessions available so far; (iii) study the stomatal behavior of transgenics under drought; (iv) study the recovery from stress between the transgenics and the wild type; (v) prepare a first evaluation of these transgenics for yield under drought in order to relate how their performance in TE, one of the component of the yield architecture, actually convert into yield.

V Vadez

Output 1.1.3: Enhanced Salinity Tolerance

Activity 1.1.3.1: Mapping salinity tolerance in pearl millet and sorghum
Team: CT Hash, V Vadez, KN Rai, BVS Reddy and S Chandra

Milestones: Mechanisms of tolerance to salinity in sorghum and pearl millet identified (2005)
Contrast for salinity tolerance between parents of pearl millet mapping populations confirmed (2005).

We confirmed previously observed contrasts for salinity tolerance using a set of sensitive and tolerant pearl millet and sorghum genotypes. Results agreed well with previous trials. In this trial, carried out in the greenhouse during a time of the year where vapor pressure deficit (VPD) is low, we found that the ratio of biomass achieved under salinity to that of control was about 40-50%, in contrast to about 10% in a previous trial conducted in outdoor conditions with these same genotypes, in a season with much higher evaporative demand (April-June). These results show the necessity to take careful consideration of the VPD while doing salinity response experiments.

A set of pearl millet breeding lines was tested again to confirm the salinity tolerance in some of them. In this trial, we also wanted to compare the evaluation for maximum forage production at booting stage, and the grain production at maturity, as it seems from earlier dataset that there is a poor correlation between booting stage evaluation and grain yield. We found a fairly good correlation between the biomass production at booting stage and the grain and fodder yield production at harvest, indicating that harvest at booting would suffice for screening pearl millet germplasm.

The 24 parental lines of 12 existing ICRISAT pearl millet mapping populations were further assessed under salinity and we have identified three contrasting pairs of parents, which will be used to identify QTL associated with salinity tolerance in pearl millet. Our activities will now focus on identifying a suitable pollinator.

A set of sorghum breeding material is also under evaluation to confirm the salinity tolerance of some material. As for pearl millet, we also would like to compare the evaluation at booting stage and that at maturity. This experiment is currently underway.

V Vadez

Mapping QTLs for salinity tolerance in pearl millet: The PhD thesis research program of Ms Rupasree M advanced, assessing opportunities to exploit existing pearl millet mapping populations for mapping components of salinity tolerance in pearl millet. She was able to develop and map over 40 TRAP markers based on gene sequences for three enzymes expected to be involved in salinity stress tolerance, and to phenotype the mapping population in which these were mapped for salinity tolerance during germination and early seedling growth, and map a few QTLs for these under control and salinity stress (150 mM NaCl) conditions. The data analysis is on-going.

CT Hash

Assessment of molecular diversity in a set of sorghum germplasm with variable tolerance to salinity: SSR-marker-based genetic diversity assessment was initiated for of a set of 30 sorghum genotypes identified as either sensitive to salinity stress under pot and field conditions (5 entries) or as tolerant to salinity stress under pot conditions and productive under saline field conditions (25 entries). Initial results, based on information generated using 76 SSR marker primer pairs, indicate 1) that there is substantial genetic diversity within the range of productive and apparently salinity tolerant breeding lines and elite varieties tested, 2) that sufficient marker polymorphism is available to allow genotyping of mapping population progeny sets that could be produced by crossing selected pairs of salinity sensitive and apparently salinity tolerant genotypes, and 3) that there is surprising similarity the marker genotype level (apparently identical PCR products for 63 of 75 tested SSR primer pairs) between elite varieties S 35 and NTJ 2, both of which were selected as productive under saline field conditions, and which apparently share common '*zera-zera*' landrace germplasm in their ancestry. Based on these results, crossing necessary to initiate formation of new sorghum mapping populations specifically targeting salinity tolerance can begin.

CT Hash

Activity 1.1.3.2: Mapping salinity tolerance in chickpea, groundnut and pigeonpea
Team: V Vadez, PM Gaur, SN Nigam, KB Saxena, HD Upadhyaya, N Mallikarjuna, D Hoisington and S Chandra

Milestones:Salinity tolerant lines identified in chickpea, groundnut and pigeonpea (2005)
Mechanisms of tolerance to salinity identified in chickpea groundnut and pigeonpea (2006).

Chickpea: A large collection of chickpea genotypes was screened for salinity tolerance. Four sets of plants were used. Two sets were grown in 10.5" pots filled up with 10 kg black soil, treated with either 0 or 80 mM NaCl solution at sowing (sufficient to saturate the field capacity), thereafter watered with soft water up to maturity. Two other sets were planted in 6.5" pots containing 3 kg of soil, treated with either 0 or 100 mM NaCla solution at sowing, thereafter watered with soft water up to harvest at 50 DAS. The set of 272 genotypes included the entire mini-core collection of ICRISAT, breeding lines, wild derivatives of chickpea, and all chickpea genotypes reported in the literature as being tolerant to salinity or sodicity.

There was a good correlation between the biomass data obtained at 50 DAS under salinity and those obtained with 252 common genotypes in 2004, indicating that our screening facility provides good and reliable data.

Although it has been reported several times in the past that there was little genotypic contrast in chickpea for salinity tolerance, we found over a 6-fold range of variation in the seed yield per pot. In particular, we found that 6-8 genotypes had a 20% higher seed yield under salinity than the best released variety for salinity tolerance, CSG8962. We also tested the parents of existing mapping populations. We found that ICCV2 and JG 62 were among the most contrasting genotypes for seed yield under salinity, with ICCV2 being the most sensitive whereas JG62 was among the most tolerant.

Investigation of possible tolerance mechanisms was investigated. We found a good correlation between seed yield under salinity and seed yield under control, showing that yield potential was partially determining the yield

under saline conditions. Sodium was analyzed in shoot tissue and showed that there was a negative correlation between biomass and Na accumulation. However, there was no significant correlation between shoot Na accumulation and seed yield. We found that desi type were usually more tolerant than Kabuli types to salinity. We found that seed yield per pot was well correlated to the number of pods per plants under salinity but poorly correlated to the seed size, showing that salinity effect played a role probably more during the pod setting and/or grain formation that during seed development and filling. We also found that maturity was significantly related to salinity tolerance, in a form of an inverse parabola. Extra early genotypes had fairly low seed yield under salinity. There was an extremum flowering time for salinity tolerance of about 54 DAS, after which there was a sharp decrease in seed yield of later genotypes. Last but not least, we did not find any correlation between the biomass at 50 DAS and the seed yield at maturity, meaning that subsequent screening for salinity tolerance in chickpea need to be done up to yield.

Groundnut: A repeat experiment using 6 genotypes was carried out to finalize the standardization of the screening protocol. It showed that an application of a 100-125 mM NaCl solution to saturate the field capacity of Alfisol was a suitable treatment to find good phenotypic contrast in groundnut. In particular, we found that tolerant groundnut keep the ability to expand leaves. By contrast, there seemed to be no relation between the biomass produced under salinity and the accumulation of Na. As in sorghum, it seems that groundnuts are able to accumulate large amounts of Na in the stem. Whether this could be used as a screen for salinity tolerance has not been tested.

Based on this protocol, a large screening was performed in the salinity screening facility in outdoors conditions. A total of 288 genotypes were tested, including the entire mini-core, breeding lines, as well as genotypes that we selected based on their origin (passport data) from the Chaco area from North Argentina, South-East Bolivia and West Paraguay, an area known to be affected by soil salinity. The screening was carried out during the April-June period. Plants were grown in 10.5" pots filled up with 10 kg black soil, treated with either 0 or 100 mM NaCl solution at sowing (sufficient to saturate the field capacity, but applied in three split doses during the initial 12 days after sowing). Because other experiments were planned at the same time in the screening facility, we could only evaluate the yield by the pod number at harvest (60 DAS). Nevertheless, we found here also a 5-6-fold variation in pod numbers across genotypes. Breeding line ICCV92206 was among the best. In peanut also we found a poor relation between pod number and seed yield. A repeat of this experiment is foreseen for early 2006, in which we expect to measure yield.

Pigeonpea: Same as in groundnut, a repeat experiment was done to finalize the standardization of the screening protocol, whereby 75 mM NaCl turned out to be the most suitable treatment to reveal genotypic contrast in pigeonpea. Interestingly, we found that SCMR reading could be a good screen for salinity tolerance in pigeonpea.

Large screening was carried out for salinity tolerance in pigeonpea. Pigeonpea is among the legumes that are the most sensitive to salinity stress. In the 1990's several papers have come up showing the relative tolerance of wild relatives of pigeonpea such as *Cajanus platycarpus*, and *C. sericeus*. The purpose of this initial large screening was to probe the range of variation in the response to salt stress using a large range of material from the germplasm bank. This range included the newly set up mini-core collection of pigeonpea, wild relatives *acutifolius*, *platycarpus*, *sericeus, albicans,* etc., wild derivatives from the breeding program, and a few hybrids. In this we also made an effort to select pigeonpea having differences in crop duration (from short to long duration). We found large contrast for biomass production at vegetative stage (biomass at 50 DAS), with large contrast between accessions of *C. scaraboides* and of *C. sericeus*. This opens the possibility to develop mapping populations of wild relatives.

V Vadez

Output 1.1.4: Enhanced Nutrient Uptake Ability
Activity 1.1.4.1: Marker-assisted breeding for phosphorus acquisition ability in pearl millet
Team: CT Hash, V Vadez and S Chandra

Milestone: African pearl millet germplasm identified for P acquisition from poorly soluble P sources and development of mapping populations for QTL of P acquisition ability initiated (2006)

Mechanisms of low P acquisition: We have now set up a protocol in the lab to measure root acidification of pearl millet seedling. This protocol combines pH measurement and image analysis and could potentially be used for large screening. It basically consists of using the colorimetric relation with pH of a colour indicator. Plantlets are inserted in a 3 mm layer of 0.9% agar + bromocresol blue. Image of the plate are scanned at regular interval

during 24 hours. Agar is then melted and changes in color measured spectrophotometrically at 540 nm. Alternatively, and in a more precise way, the scanned images are converted in layers of different color, *i.e.*, different pH values. The respective surfaces are measured and H^+ extrusion calculated. This allows mapping of the H^+ extrusion. With this technique, we can potentially screen the parents of existing mapping populations of pearl millet. The current orientation of this work is to modify this method to make measurement of acidification in older plants and correlate results with measurements at the seedling stage.

Phenotyping of low P millet: The phenotyping of the testcross hybrids obtained from the combination of four different male-sterile lines and the mapping progenies of the crosses between 81B-P6 and ICMP 451-P8, and between LGD 1-B-10 and ICMP 85410-P7 was undertaken in February 2005. This was the first time that scaling up low P screening was undertaken for pearl millet at ICRISAT-Patancheru. The experimental design used was an alpha lattice with 5 replications, 1 pot per entry per replication, and 1 plant per pot. As usual, 8" pots were used and filled with a sand-low P soil mixture and 200 mg of rock phosphate per kg of mixture. Plants receiving the low P treatment appeared to be very stunted compared to those receiving the control treatment. Data analysis revealed large plant-to-plant variation within entries, little consistency across replications, and problems of plant establishment.

From there on, we decided to look back at the different trials that had been conducted over the past two years to establish this protocol. Careful examination of the previous experiments (2003-2004) conducted with the mapping population parents revealed several important factors: (i) the mean biomass response of low P plants varied a lot across experiments, reaching from as low as 5-10% of control to up to 40-50%, indicating probably that large variations occurred in the P levels of the different soil lots used in the experiments; (ii) earlier experiments carried out with urea tended to have larger biomass yield means than later experiments carried out with ammonium nitrate, showing that N source may have played an important interaction with the observed low P treatment responses (for understandable reasons); (iii) yet, there was still a good correlation between the biomass yield values obtained from the set of pearl millet mapping population parents throughout time.

After this exercise, we have then addressed four issues: (i) the differences in Olsen P in the different batches of soil, (ii) the control of plant to plant variation; (iii) the source of nitrogen; (iv) the possibility to favor initial plant establishment with early application of tiny doses of soluble P.

The issue raised in (i) was confirmed coincidentally when we decided to bypass the sand acid-washing step from our protocol to save time, resources, and avoid contamination. We ran an experiment to compare the biomass response of a set of 4 pearl millet genotypes grown in low P soil mixed (1:1 w/w) either with acid-washed sand or non acid washed sand. To our surprise, pearl millet genotypes grew about twice as well in the mixture using the acid washed sand as in the one with non-acid washed sand. Soil samples from the mixture had been kept and revealed that Olsen P was 0.9 and 1.5 ppm in the mixtures using non-acid washed and acid-washed sand, respectively. This experiment indicated that: (i) very slight differences in Olsen P triggered large differences in the plant response; (ii) plant to plant variation seen in other experiments might be due to minute differences in P availability, leading to differences in plant establishment (iii) in this experiment, plant establishment was good and subsequent plant to plant variations remained minimum.

Regarding issue (ii), we now measure soil and sand Olsen P each time a new batch is used, and we ensure that Olsen P of the soil mixture is above 1.5 ppm (lower values lead to severe problems of early plant establishment). Mixing is thoroughly done using a concrete mixer to ensure homogeneity of the soil mixture. In the low P field where soil is collected, we have also applied fertilizer P in one strip, which we can use as "inoculum" in case the Olsen P value of a batch of soil turns out too low. We also grow 3 plants per pot. Preliminary indications show that the problem of plant-to-plant variation is now under control.

Regarding issue (iii), we have carried out two separate experiments. Using a set of 4 hybrids, we have found that, indeed, urea-grown plants under low P grow significantly better than under ammonium nitrate. However, there is also a significant N × genotype interaction in a sense that one genotype grows as well under low P conditions regardless of whether N is provided from NH_4NO_3 or urea. In a second experiment, we grew the same set of genotypes in 4 different soils. Data are currently being analyzed. The visual differences were very obvious regarding the overall differences between NH_4NO_3 and urea. This means that the phenotypic screening of pearl millet mapping population progenies for differences in phosphorus acquisition ability, which is foreseen, will need to be done with both N sources.

Finally, regarding issue (iv), the possibility to add tiny amounts of P to the young seedlings was initially thought to decrease variation from plant to plant. In a preliminary experiment, we had the same 4 genotypes as above,

grown in low P soil, and we treated plants with a) nothing, b) soaking one night in a 100mM KH_2PO_4 concentration, and c) applying 10 times the amount of P contained in an average pearl millet seed in a soluble form at 4 days after sowing. For plants receiving the +P treatments, we found that shoot biomass at 40 DAS was more than double of those receiving no P treatment, and this response varied across genotypes. A repeat of this experiment has been done using three soils, and three "priming" application of soluble P: 5, 10, and 20 times the amount of P in one seed. There again, we have found an important response to early application, with maximum effect being 10 times the amount in one seed. These results are very important and exciting, as they show that a very minor early application of P gives over a 40-fold return in P acquisition by the plants. Combined with issue (iii), urea plus initial "priming" give the base for a very good and applicable package.

V Vadez

Output 1.1.5: Improved Nutritional Quality

Activity 1.1.5.1: Marker-assisted genetic improvement of grain carotenoid content in pearl millet
Team: CT Hash, KN Rai and S Chandra

Milestones: Markers for grain carotenoid content mapped in pearl millet (2008)
MAS for enhancement of grain carotenoid content in pearl millet initiated (2008)

Attempts to complete NIRS calibration for pearl millet grain carotenoid content following HPLC repairs were unsuccessful as correlation between two HPLC runs was not significant for the panel of 90 S1 and inbred progenies. No further work was undertaken in 2005 as required special project funding was not available, and no future work is projected unless special project funding is forth-coming.

CT Hash

Activity 1.1.5.2: Genetic engineering of groundnut for enhancement of β-carotene and of pigeonpea for enhancement of methionine and β-carotene
Team: A Vanamala, KK Sharma, SN Nigam and KB Saxena

Milestones: Transgenic groundnut lines with over-production of β-carotene evaluated under controlled field conditions (2007).
Transgenic pigeonpea lines with over production of methionine and β-carotene evaluated under controlled field conditions (2007).

Gene constructions of ß-carotene into plant expression vectors: The in-house cloned genes encoding for phytoene synthases, *psy*1 from maize and *crt*B from *Erwinia herbicola,* involved in ß-carotene biosynthesis were sequenced and the gene sequences were confirmed by alignment and blast analysis. The coding sequences of these genes were initially fused to constitutive expression promoter (CaMV 35S), sub-cloned into T-DNA region of the binary vector pCAMBIA 2300 and mobilized into *Agrobacterium tumefaciens* strain C 58 for genetic transformation studies. The expressions of the in house cloned genes were verified initially by transforming tobacco as a model system. Subsequently, the coding sequences of *psy1* or *crt*B genes were fused with the oleosin promoter to drive gene expression in the oil bodies of matured seeds. These gene constructs were further sub-cloned into plant expression vector pCAMBIA 2300 and mobilized into *Agrobacterium* C 58 strain for genetic transformation of groundnut and pigeonpea. To generate marker-free transgenic plants, these gene constructs were also sub-cloned into binary vectors of pCAMBIA 2300 (minus *npt*II) and 2 T-DNA.

Construction of plant expression vectors containing sunflower seed albumin gene (SSA): The methionine-rich sunflower seed albumin gene (*SSA*) driven by the vicillin promoter (for seed specific expression), a 3.2 kb *Eco*RI fragment of pLT4 plasmid, was sub-cloned into binary plasmids pHS723, pCAMBIA 1301 and pCAMBIA 1302, and mobilized into *Agrobacterium* strain C 58 for genetic transformation of pigeonpea. Similarly, the same gene construct is being sub-cloned into pCAMBIA 2300:φ*npt*II binary vector to produce marker-free transgenic plants.

Development and analysis of putative groundnut transgenic events carrying β-carotene genes: *Agrobacterium*-mediated genetic transformation of the selected genotypes of groundnut was carried out by using with newly constructed binary vectors containing ß-carotene genes (*psy*1 and *crt*B) for obtaining high frequency of transformants. The putative transgenic groundnut shoots obtained under antibiotic selection pressure were elongated for rooting followed by their transfer to containment greenhouse.

The putative transgenic T0 groundnut plants growing in the containment greenhouse were analyzed molecularly for the integration and presence of the transgenes by using PCR with gene specific primers and Southern hybridization for the copy number. Initial molecular characterization from few putative transgenic groundnut plants showed the integration of *psy*1 gene. The transgene expression and the presence of m-RNA transcripts (*psy*1) were observed in few putative groundnut transgenic events by RT- PCR analysis and these events are being advanced for further generations.

Development of pigeonpea transgenic events for enhanced level methionine: The binary vector gene constructs containing *SSA* gene along with vicillin promoter (pHS723:SSA) in *Agrobacterium tumefaciens* strain C 58 is being regularly used for genetic transformation of pigeonpea for obtaining enhanced level of seed methionine content. In separate studies, *Agrobacterium*-mediated genetic transformation for selected genotypes of pigeonpea is being carried out regularly using with newly constructed binary vectors containing maize *psy*1 gene for generating transgenic events with enhanced level of ß-carotene. The putative transgenic pigeonpea shoots obtained under antibiotic selection pressure are being elongated for rooting and transfer to the containment greenhouse for further analysis.

The primers specific to the coding sequence of *SSA* and *psy*1 genes were designed and conditions for PCR amplification optimized. The primary putative transformed pigeonpea shoots obtained under antibiotic selection pressure are being molecularly analyzed by PCR analysis for the presence of *SSA* and *psy*1 genes. The expression of *SSA* gene in transgenic tobacco was verified (used as a modal system) by RT-PCR analysis in the T1 seeds of tobacco.

A Vanamala, KK Sharma, , M Rai, SN Nigam and KB Saxena

Output 1.1.6 : Improved feed and fodder quality

Activity 1.1.6.1: Improved pearl millet stover quality
Team: CT Hash, B Blummel, F Bidinger, R Folkertsma, M Mgonja, F Rattunde and S Chandra

Milestone: Genotype and genotype × environment effects on stover quality documented, heritability of different stover quality components assessed, and sampling methods for efficient assessment of stover quality established in pearl millet (2006)

Management and genotype effects on pearl millet stover quantity and quality: Farmers have two basic options for improving both the quantity of stover produced and its nutritional quality – intensifying crop management to increase production and/or feed value of stover, and choice of cultivar type and/or specific cultivar to exploit genetic differences in quantity and/or quality of stover produced. We conducted a preliminary evaluation of both options for pearl millet, as baseline information for a funded project on the genetic improvement of millet stover quality.

The most significant management option available to farmers for increasing pearl millet stover digestible dry matter (DDM) yield and stover metabolizable energy (ME) yield is adequate fertilization. Although increased fertilizer application had small, but significant, negative effects on stover digestibility, sugar concentration and ME, these were more than offset by large increases in total stover dry matter production. Higher fertility also had a major positive effect on stover nitrogen concentration, which should have a significant effect on animal weight gain (especially where stover is fed without supplementation with either a concentrate or a higher N legume straw). Increasing plant population, in contrast, had little effect on either stover quality or yield. The low population treatment still produced nearly the same biomass and stover yields as the high population treatment, despite a two-fold difference in plant numbers. Although stover from the lower plant population treatment did have slightly numerically higher values for almost all stover quality traits, but differences were only significant for N % and digestibility, and these had little effect on stover DDM or ME productivity.

Choice of cultivar type (landrace, open-pollinated dual-purpose cultivar, or F_1 hybrid) did have a significant effect on both stover productivity and quality. The dual-purpose cultivars (and the landraces) had higher stover productivity and significantly higher digestibility, sugar concentration and ME than the hybrids, but this was at the cost of a significantly lower grain yield. The most important finding was that there were no strong negative relationships between the most important stover quality traits and either grain or stover yield, however, so there is not bar to combining high grain yields with high stover yields with at least average stover quality. Several dual-purpose hybrids in the trial (notably HHB 60 and the old ICRISAT release ICMH 451), capitalized on this, producing DDM and ME yields on par with the best open-pollinated dual-purpose cultivars and landraces, with a significantly higher grain yield. The results suggest that there is no reason why targeted breeding of dual-

purpose hybrids (stover and grain yield) with improved stover quality should not be successful. Such hybrids should also maximize returns to investment in increased fertilization, though increased grain and quality stover yields.

FR Bidinger and M Blümmel

Milestone: Additional markers for key components of pearl millet stover quality (digestibility, N content, intake, etc.) identified (2006)

Linkage map construction and QTL mapping: New SSR, EST (EST-SSR and SSCP-SNP) and TRAP markers (developed by the University of Nebraska, USA; JIC, Norwich, UK; and ICRISAT-Patancheru, India) were added into the existing linkage map of the pearl millet mapping population based on cross ICMB 841 × 863B, which was comprised of the previously available polymorphic RFLP and SSR markers. Markers were assigned to different linkage groups based on marker recombination and ordered within linkage groups to minimize the frequency of candidate errors. After removing colinear markers and those exhibiting distorted segregation patterns, seven linkage groups were constructed using 76 polymorphic RFLP, SSR and EST markers.

QTL analysis was performed by composite interval mapping for stover quality traits of individual stover fractions (stem, sheath, and leaf blade) as well as whole stover samples. Stems are a major and important constituent among the different stover fractions because they contribute more mass to the livestock feed. For stem fraction digestibility and metabolizable energy content, a major QTL was identified on LG 2, at a position similar to that for drought tolerance alleles from the 863B parent (suggesting it should be simple to combine improved stem digestibility with greater drought tolerance in the genetic background of elite seed parent maintainer line ICMB 841). Another major stover quality QTL was mapped to LG 5, which controlled significant proportions of phenotypic variation for *in vitro* true digestibility and for metabolizable energy content. In contrast, the ICMB 841 alleles for QTLs on LG 6 were favorable for gas volume, true organic matter degradability, nitrogen content and metabolizable energy content of stover stem and sheath fractions.

T Nepolean, ,S Senthilvel, CT Hash and M Blümmel

Milestone: Elite hybrid parental lines of pearl millet with improved stover quality through marker-assisted backcrossing available for evaluation (2007*)*

***Advancing QTL introgression lines*:** During the 2004/05 post-rainy season, 16 F_1 hybrids of selected hybrid seed parent maintainer lines (ICMB 95111, ICMB 95222, HMS 7B and ICMB 93333) and parents of two stover quality mapping populations (ICMB 841, 863B, PT 732B and P1449-2) were grown in the field. Each hybrid combination was backcrossed with the recurrent parent and resulting in a total of 16 F_1 parent × recurrent parent BC_1F_1 combinations. Two plant × plant crosses from each combination were selected to advance the BC_1F_1 generation. Two crosses from each of the BC_1F_1 hybrid combinations were sown and raised during the rainy season of 2005. BC_1F_1 hybrids were backcrossed, plant × plant, with their respective recurrent parents to produce BC_2F_1 seeds. The BC_1F_1 generations of 16 crosses were screened using polymorphic SSR and TRAP markers, to assess their segregation patterns and select the heterozygous QTL introgression genotypes from various crosses for generation advance.

For the putative LG 7 stover quality QTL, RFLP and SSR marker data were used to identify introgression homozygotes for the associated region from donor parent 863B in the genetic background of ICMB 841. Several such plants were identified and their selfed seed harvested for future testcross hybrid production.

The revised QTL analysis reported above indicated that at least some fo the introgression lines for the LG 2 drought tolerance QTL of 863B in the genetic background of ICMB 841 are likely to carry a QTL for improved stover quality.

T Nepolean and CT Hash

Milestone: Two cycles of recurrent selection for improved stover quality (using NIRS analysis) completed in two arid zone landrace-based populations in pearl millet (2007*)*

Evaluation of ICMV 221 cycle 2 FS (full-sib) progenies: We completed the second and final cycle of FS progeny selection for improved stover quality in the released pearl millet variety ICMV 221. The objective of the exercise is to measure the genetic progress that can be made in improving the ruminant nutritional quality of pearl millet stover using simple recurrent selection methodology and rapid near-infrared reflectance spectroscopy (NIRS) assessment of key stover quality parameters. As in the first cycle, there were highly

significant differences for all stover quality traits and agronomic variables measured, among the 280 FS progeny evaluated. For example stover *in vitro* organic matter digestibility ranged from 37 to 46%, stover metabolizable energy from 5.2 to 6.7 mega joule kg^{-1}, stover nitrogen from 0.67 to 1.25 %, and stover yield from 123 to 432 g m^{-2}. Selected progenies will be recombined to form both grain and dual-purpose versions of the original variety with improved stover nutritional quality, to compare to the original ICMV 221. Individual progenies will also be used to develop high stover quality trait lines for various research purposes, including evaluating the effect of improving parental stover quality on hybrid stover quality.

FR Bidinger and M Blümmel

Milestones: Stover quality of experimental pearl millet hybrids of BC_5 products evaluated in multilocational field trials (2008)
Stover quality of experimental varieties from selected arid zone populations in multilocational field trials evaluated in pearl millet (2008)

Response to a single cycle of selection for stover yield and quality: *We* random-mated selected progenies from the first cycle of improvement of the stover quality of the pearl millet variety ICMV 221 (see above) to produce stover quality trait-based experimental varieties to assess the potential for reselecting an existing variety to improve its stover quality. Selection was done jointly for a high or low value of the quality trait, plus grain and stover yield within one SED for mean of the whole set of progenies. In addition we made specific grain or dual purpose versions of the original variety with as good a stover quality as was possible within a high grain or grain + stover yield objective. All six experimental varieties and the C0 and C1 versions of the original ICMV 221 were tested in replicated small plots for productivity and *in vitro* quality analysis by NIRS, and a subset was grown in replicate large plots for *in vivo* quality analysis in a replicated sheep feeding trial.

The reselected grain type experimental variety did not differ significantly from the parent variety, either in agronomic performance (apart from some offsetting changes in yield components) or in stover quality in either the *in vitro* or *in vivo* measurements. The dual propose experimental variety, in contrast, exceeded the parent variety in stover yield by 12% ($P < 0.05$) and in grain yield by 5% (NS). More interestingly, although it did not differ from the parent variety in the *in vitro* quality evaluations, it was significantly better than the parent for organic matter digestibility (56.6% vs. 54.4%) in the *in vivo* evaluation.

Selection for high and low stover digestibility and for high stover nitrogen generally resulted in small and non-significant reductions in both grain and stover yield. The exception was the experimental variety made on the basis of low stover nitrogen, which resulted in a 6% ($P<0.05$) increase in biomass and a 10% ($P<0.05$) increase in grain yield. Selection for and against stover digestibility and stover N% resulted in small, but non-significant, changes in these parameters, in the direction of selection in the *in vitro* stover quality assessment, but few other changes in quality. In the *in vivo* assessment however, the high digestibility selection had a significantly higher digestibility (57.5%) than the parent variety (54.5%).

FR Bidinger and M Blümmel

Output 1.2: Diverse range of populations, breeding lines and potential hybrid parents developed, evaluated and disseminated

Summary

The research carried out under this generic output includes a wide range of activities in the two cereals and three legumes, which constitute a major part of research in this regional project. Significant progress was made in both strategic and applied research areas dealing with focused germplasm evaluation and utilization, development of genetically diverse and productive breeding lines and populations with resistance to biotic stress factors and tolerance to drought, and cytoplasmic-genic male sterility (CMS) systems with both medium- and long-term perspectives to enhance sustained productivity of these crops.

In sorghum, a landrace (IS 23526) was identified that had Brix reading similar to the sweet-sorghum check variety SSV 84, but it had 65% more sugar yield and flowered a week earlier. Several other landraces with sugar yield comparable to SSV 84 were also identified. Germplasm lines identified for high levels of shoot fly and grain mold resistance were crossed with elite breeding lines possessing moderate resistance to these traits to further build the resistance levels in the breeding materials. Germplasm and breeding lines with high levels of both iron (Fe) and zinc (Zn) and salinity tolerance were identified, and it was shown that though nitrogen application had a significant effect on increasing the Fe levels, the genotype × nitrogen interaction was not significant. Several advanced B-lines and R-lines with grain yield and grain size higher than or comparable to

the widely used commercial checks (B-line 296B and R-line RS 29) were developed. Grain mold research confirmed the earlier results that the probability of producing mold-resistant hybrids is higher from resistant × susceptible crosses, followed by resistant × resistant crosses, and that mold-resistant hybrids or parental lines had higher levels of flavan-4-ols and lower levels of ergosterol than the susceptible ones. A comparison of hybrids based on the A_1 and A_2 cytoplasm showed that there was no significant difference between these two cytoplasms for resistance to either grain mold or shoot fly, or for grain yield. Also, A-lines with these two cytoplasms had similar grain yield in a diverse range of genetic backgrounds, while those with other CMS systems [A_3, A_1 (M), A_4 (VZM) and A_4 (G)] had lower grain yield.

In pearl millet, a topcross hybrid based on a germplasm accession as a pollen parent, that had earlier been found outyielding a commercial sorghum-sudan grass hybrid, again showed its yield superiority with 36% higher dry forage yield. Interestingly, open-pollinated varieties (OPVs) developed from germplasm-derived progenies were identified that were comparable to or had higher dry forage yield than this high-yielding pearl millet hybrid. Utilization of large-seeded and long panicle germplasm led to the development of early generation progenies that had >15 g of 1000-seed mass and 60–80 cm of panicle length. Two high-yielding (both grain and dry fodder) germplasm accessions with high levels of salinity tolerance were also identified. Improved populations, largely based on the iniari germplasm, with high levels of both Fe and Zn were identified, with some of these populations showing more than 2-fold within-population variability. A simple staining method using Perls Prussian Blue was standardized for rapid and cost-effective preliminary selection of germplasm with high Fe content. Early-generation progenies derived from composites targeted for arid Rajasthan were developed that were of early to mid-early maturity and had high levels of downy mildew (DM) resistance to the most virulent Jodhpur pathotype of the DM pathogen population. Trait-based breeding of potential seed parents led to the identification of a large number of advanced generation progenies with high yield potential and DM resistance to 1–2 DM pathotypes. High-yielding and DM resistant advanced generation progenies, mostly of medium maturity group, from improved populations (composites and OPVs) were also developed. Efforts were increased to develop seed parents with A_4 and A_5 cytoplasm and their respective restorers. Nine male-sterile lines (3 A_1 cytoplasm and 6 A_4 cytoplasm) of diverse morphological characteristics and with DM resistance to at least two diverse pathotypes of DM were designated and disseminated. Fourteen A_1-system elite restorers were converted into their A_4 restorer versions, and 39 A_1-system elite restorers were converted into their A_5-restorer versions. Based on the mean performance of isonuclear hybrids evaluated across two locations for two years, it was shown that there was no difference between the A_1 and A_5 cytoplasm for grain yield and other agronomic traits. A comprehensive genetical study of five diverse CMS systems involving 45 F_2 populations and their corresponding backcrosses was completed. The results showed generally trigenic control of male sterility in each CMS system with varying interactions.

In pigeonpea, sources of high levels of salinity tolerance were identified in improved seed parents, cultivated germplasm of pigeonpea and a wild species, C. scarabaeoides*. Several advanced breeding lines with resistance to both* Fusarium *wilt and sterility mosaic were identified. Testcross evaluation of pigeonpea hybrids showed high frequency of restorers (96%), which enhances hybrid breeding efficiency with this CMS system. Pigeonpea hybrids of three maturity groups (extra-short, short and medium duration) evaluated at Patancheru showed hybrid seed yield advantage ranging from 50 to 200% over the check varieties of comparable maturity. Hybrid yield advantages of similar order in these three maturity groups were found in multilocation trials conducted at 4–6 locations.*

In chickpea, new germplasm sources of resistance to Fusarium *wilt (FW),* Botrytis *gray mold (BGM),* Ascochyta *blight (AB), dry root rot, and collar root rot were identified. Also, breeding lines with higher seed yield and larger seed size than controls and combining high levels of resistance to* Fusarium *wilt were identified both in* desi *and* kabuli *groups. Some of the advanced breeding lines with high levels of resistance to either* Ascochyta *blight or* Botrytis *gray mold were identified in* desi *chickpea. A genetical research found two loci governing the number of flowers per axis. Seed-borne nature of the fungus causing* Fusarium *wilt was confirmed. A detached leaf method developed for rapid laboratory screening for* Helicoverpa *resistance was found effective in case of chickpea but not in case of pigeonpea. HPLC profile of leaf exudates showed significant negative correlation between malic acid and pod damage, oxalic acid and leaf damage, acetic acid and larval weight as well as leaf and pod damage, and citric acid and pod damage.*

In groundnut, seven germplasm accessions of two wild species (5 of A. duranensis *and 2 of* A. stenosperma*) were identified that had systemic infection to Tobacco streak virus, and two of these were also highly resistant to both late leaf spot and rust. The foliar disease resistance breeding produced several advanced breeding lines with high pod yield combined with moderate levels of resistance to late leaf spot, and high levels of resistance to rust in both Spanish Bunch and Virginia Bunch groups. Several advanced breeding lines with high pod yield*

and high levels of resistance to aflatoxin were developed. Some of the resistant lines had as low as 2.9–4.2 µg kg^{-1} of aflatoxin content. Drought-tolerance breeding program led to the development of several advanced breeding lines with high pod yield under rainfed condition, some of which outyielded the control even under irrigated condition. High-yielding advanced breeding lines with high oil content were also identified.

Activity 1.2.1: Evaluate and introgress new germplasm sources of variability for yield components, resistance to biotic and abiotic stresses and quality traits

Milestone: Germplasm lines of sorghum and pearl millet with large seed, and high fodder yield and quality traits identified and introgressed (2005)

Race- and trait-based B-lines: In order to diversify the hybrid parental lines for grain yield and other traits, high-yielding B-lines were crossed with germplasm lines belonging to different races and having specific traits. The resulting crosses were advanced with selection for different race-specific traits while maintaining desired maturity and grain yield. The promising F_4 progenies with maintainer reaction were utilized for conversion into A-lines with A_1 and A_2 cytoplasmic-nuclear male sterility (CMS) systems. These are in the various stages of conversion.

Diversification of sweet sorghum hybrid parents: Considering that hybrids have high biomass yielding ability, sweet-stalk hybrid parents' research is being given strategic importance at ICRISAT-Patancheru. New germplasm lines were evaluated in replicated trials for sweet-stalk biomass yield, to identify promising sweet sorghum germplasm lines for use in introgression into the available grain sorghum hybrid parents in order to diversify them for sweet-stalk traits.

A total of 98 landraces and varieties, along with the controls SSV 74 and SSV 84, were evaluated in the 2004–05 postrainy season for stalk sugar content and biomass yield. Based on the performance for these traits, 45 lines were selected and evaluated along with controls NSSH 104, SSV 74 and SSV 84 during the 2005 rainy season. Brix reading was taken 18 days after 50% flowering. The sugar yield, based on Brix reading and juice yield, was estimated. One of the landraces, IS 23526 (5.8 t ha^{-1}) significantly out-performed the control SSV 84 for sugar yield (3.5 t ha^{-1}). This line, besides being early by 7 days, had high Brix reading (19.5%), which was comparable to the control SSV 84 (19.4%). The sugar yield potential of several other test lines, RSSV 106 (4.8 t ha^{-1}), NSS 254 (4.3 t ha^{-1}), IS 18521 (4.1 t ha^{-1}) and IS 4617 (3.9 t ha^{-1}), was comparable to that of SSV 84 (3.5 t ha^{-1}). The promising landraces will be introgressed into available hybrid parents.

BVS Reddy and S Ramesh

Shoot fly resistance breeding: Shoot fly is one of the major biotic constraints in both rainy and postrainy seasons. Considering that the available seed parents bred for shoot fly resistance (SFR) possess moderate resistance levels and grain yield potential, efforts are being made to diversify seed parents for SFR.

In order to diversify hybrid parents for SFR, 13 germplasm lines (IS 18551, IS 923, IS 1057, IS 1071, IS 1082, IS 1096, IS 2394, IS 4663, IS 5072, IS 4664, IS 5470, IS 5636 and IS 18369) with high levels of SFR were crossed onto shoot fly resistant breeding lines (8), advanced backcross progenies in conversion program (8) and new B-lines (3) (on A_1 cytoplasm) and the resulting F_1s (29, 26 and 10, respectively) were evaluated during the 2005 rainy season. From these, 25, 13 and 7 F_2s, respectively were selected.

From 14 F_1s generated from the crosses between shoot fly resistant B-lines and postrainy varieties, 4 F_2s were selected. Further, 31 F_2s derived from the crosses between shoot fly resistant breeding lines and elite B-lines, and 11 F_2s derived from the crosses between shoot fly resistant breeding lines and high-yielding varieties were evaluated during the 2005 rainy season and produced 17 F_3 and 9 F_3 progenies, respectively. Four F_4s were produced from 24 F_3 progenies of shoot fly resistant B-lines × shoot fly resistant B-lines crosses during the 2005 rainy season. All these progenies are being advanced with selection during the 2005–06 postrainy season.

BVS Reddy, S Ramesh and HC Sharma

Grain mold resistance breeding: Grain mold is one of the major biotic constraints in rainy season in India. Efforts are being made to diversify the hybrid seed parents for grain mold resistance (GMR), as the available hybrid seed parents possess moderate resistance levels and grain yield potential.

Several crosses were made involving grain mold-resistant B-lines, varieties and landraces and their selections. A total of 94 F_3s derived from these crosses were evaluated during the 2005 rainy season for grain yield and grain size, plant height and maturity, and 27 F_4s were produced. These 27 F_4s will be evaluated for grain mold

resistance (GMR) during 2006 rainy season and those found resistant will be testcrossed onto A_1 and A_2 CMS systems for conversion into A-lines.

BVS Reddy, S Ramesh and RP Thakur

Germplasm evaluation and introgression in pearl millet:

High biomass yield: Increased attention to develop hybrid parents or varieties for forage purposes has prompted genetic diversification of forage breeding material by exploiting germplasm accessions of diverse origin for high biomass yield. About 77 progenies of a wide flowering range (52–85 days) were used to constitute six OPVs (ICMV 05111-ICMV 05666). Each OPV was developed from intercrossing of 4–26 progenies selected for forage purpose during the previous year. Important traits considered while making these varietal groups were plant height, stem thickness, tillering and leafiness. Another variety (ICMV 05777) was developed from a germplasm accession (IP 6073) from the Central African Republic. Random mating of 38 S_3 progenies of an OPV (CO 8) resulted in ICMV 05888. Similarly, ICMV 05999 was developed from random mating of 91 S_1 progenies of another OPV (RMFB). All these nine OPVs were evaluated along with seven controls [comprising 2 forage hybrids, 3 dual-purpose hybrids, 1 open-pollinated variety (WC-C 75) and a sorghum-sudan grass hybrid (GK 908)]. High dry forage yielding ability of an earlier identified promising hybrid, ICMA 00999 × IP 17315 was again confirmed as it produced 36% more dry forage (on oven dried basis) than the sorghum-sudan grass hybrid GK 908 (11.7 t ha^{-1}) at 80-day harvest. Of the nine varieties, five had 12.8–17.1 t ha^{-1} dry forage yield (15.8 t ha^{-1} for ICMA 00999 × IP 17315) at 80-day harvest. Four of the five high-yielding varieties flowered in 61–73 days (GK 908 flowering in 72 days and ICMA 00999 × IP 17315 in 69 days). These OPVs, if found promising in multilocational trials, have potential of being directly released as open-pollinated forage varieties, and also as promising germplasm for use as pollinators of topcross hybrids. In addition to the varietal trial, a set of 21 new germplasm accessions was visually evaluated for forage yield, and 10 were selected for further evaluation and utilization. To introgress earliness in high biomass yield backgrounds, six germplasm-derived photosensitive improved populations were crossed with an extra-early-maturing composite (EEBC) and an early-maturing B-line (834B) that has high early seedling vigor. These 12 hybrids were evaluated and four were selected based on the visual assessment of earliness and high biomass yield for further evaluation and utilization.

Large seed size: Although seed parents having up to 14.5 g 1000^{-1} seed mass have already been developed, the quest for developing hybrid parents with still larger seed size (16–20 g 1000^{-1} seed mass) in diverse genetic backgrounds is in progress. About 370 F_4 progenies derived from the crosses involving lines derived from three large-seeded germplasm accessions in their parentage were evaluated and 174 progenies were selected based on the visual assessment of grain size and agronomic potential to generate 430 F_5 progenies. Of the selected 174 progenies, 16% flowered in 51–60 days (ICMB 96555 flowering in 52 days), of which 12 had >15 g 1000^{-1} seed mass. Additionally, 90 F_3s were selected (mostly d_2 dwarf) out of 229 planted based on visual assessment of large seed size and agronomic potential and 40% of the selected progenies flowered in 51–60 days (ICMB 96555 flowering in 52 days). Of these, 17 progenies had >15 g 1000^{-1} seed mass. About 40% of the large-seeded F_5 progenies and 47% three-way F_3 progenies with >15 g 1000^{-1} seed mass (4 had even up to 20 g 1000^{-1} seed mass) were of mid-late to late maturing type, indicating the necessity of crossing elite early and mid-early lines to mobilize the large seeded trait in the commercially exploitable background.

Panicle length: Panicle length is a highly heritable and important grain yield component. Cultivated pearl millet hybrids possess panicle length not exceeding 30 cm. Great potential exists in developing long panicle hybrid parents by using germplasm accessions with >100 cm panicle length. Our attempts till now reveal that although such accessions were very good source of panicle length, the genetic drag in terms of poor exertion, tall plant height, poor tillering, obvious late maturity, and more importantly, poor spikelet density was very high. Amongst the total 700 progenies (F_4–F_6 and beyond) produced during the 2005 summer season, based on the panicle length and other desirable features, only 166 long-panicled progenies having high agronomic scores were evaluated, of which 78 progenies were selected based on the visual assessment to generate 195 progenies for further evaluation. Progenies with panicle length up to a maximum of 82 cm, flowering in 56–70 days, were monoculm (not tillering), and had poor to medium spikelet density. Interestingly, some degree of earliness was evident in the progenies, ie, 39% of 78 progenies flowered in 46–55 days (NCd_2 flowered in 48 days), which had panicle length up to 60 cm. About 350 out of 700 advanced generation progenies were also screened against Durgapura pathotype under high disease pressure in the greenhouse condition (>95% incidence in susceptible controls ICMP 451 and 843B) and 41% were highly resistant (0–10% DM incidence). In order to improve spikelet density and tillering, 124 hybrids produced from crossing a set of 4 compact panicle lines, 3 good exsertion lines and 6 high-tillering lines with 9 long panicle progenies were evaluated, and 73 were selected and classified into five groups based on the panicle and plant traits, such as thick and long panicle (31

F_1s), medium-long to thin panicles (18 F_1s), dwarf-tillering (13 F_1s), short-height tillering (14 F_1s) and compact panicles (8 F_1s). Some of them were included in more than one group. In addition, we evaluated 116 germplasm accessions (compactness score 7–9 on a 1-9 scale, with 1 = loose and 9 = very compact, head length >25 cm and panicle girth >25 mm as per the genetic resources characterization data) as new sources of long and compact panicle traits and 18 were selected producing 25 S_1 progenies for further evaluation and utilization. The selected progenies flowered in 53–67 days (NCd_2 flowering in 51 days).

White seed color: White grains are expected to diversify uses of pearl millet in food industry. However, the main bottleneck is the non-availability of germplasm with white grain color in photoperiod-insensitive genetic background. About 75 white-grain F_3 progenies derived from 15 F_2s developed through crosses involving four germplasm-derived lines from three photosensitive germplasm accessions were evaluated and 38 were selected based on the visual assessment for white grain color and agronomic potential to generate 95 F_4 progenies. About 28% of the selected F_3 progenies flowered in 56–65 days (54 days for ICMB 94222). All the F_4 progenies will be further evaluated to produce early to mid-late maturing progenies with white grain so as to transfer the same into agronomically superior genetic backgrounds.

KN Rai, VN Kulkarni, HD Upadhyaya and M Blümmel

Milestone: Germplasm lines of sorghum and pearl millet with higher levels of Fe, Zn and ß-carotene contents; and salinity tolerance identified and introgressed (2006)

Micronutrient density: Hybrid parents of high-yielding commercial hybrids (10 B-lines and 14 R-lines) and 6 high-yielding released varieties were crossed with germplasm lines having high levels of β-carotene (3 germplasm lines), Zinc (2 germplasm lines) and Iron (1 germplasm line and 5 breeding lines) that were identified based on the evaluation of 86 diverse hybrid parents, varieties and germplasm lines. A total of 176 F_1s were obtained: 32 F_1s from the crosses between high-yielding breeding lines and the germplasm lines rich in β-carotene, 27 F_1s between high-yielding breeding lines and the germplasm lines rich in Zn and 117 F_1s between high-yielding breeding lines and the germplasm lines rich in Fe contents. These are being advanced during the 2005–06 postrainy season.

BVS Reddy and S Ramesh

Grain Fe and Zn density in germplasm accessions of pearl millet: The sibbed seeds of 20 diverse germplasm accessions (part of a trial of 120 entries) from both summer and rainy seasons (2004) were analyzed at the National Institute of Nutrition, Hyderabad, India, for grain Fe and Zn density. The mean Fe density ranged from 34 to 54 ppm and Zn from 35 to 49 ppm. Three accessions (IP 6764, IP 8964 and IP 12240) had the highest Fe of 50–55 ppm and 4 accessions (IP 3122, IP 3859, IP 9453 and IP 8964) had the highest Zn of 48–49 ppm across two seasons. The germplasm accession, IP 8964 had both higher Fe and Zn density. The levels of Fe and Zn in these accessions were significantly lower than those in the elite breeding lines with higher grain Fe and Zn. There was highly significant correlation between Fe and Zn density ($r = 0.79$; $P < 0.01$).

Grain and fodder yield of salinity tolerant germplasm of pearl millet in saline soils: A set of 15 germplasm accessions, identified as salinity tolerant based on three years (2002–04) evaluation at ICBA was evaluated at 10 dS m^{-1} salinity level at Gangavathi, Karnataka, India, during 2005 rainy season (second season) for grain and fodder yield. Early stages of crop growth was affected due to continuous rainfall that delayed the weeding operation resulting in 33% lower grain yield and 50% lower fodder yield compared to 2004 rainy season. Two germplasm accessions, IP 22269 and IP 6098, were identified for high grain (1210 and 1168 kg ha^{-1}) and fodder yield (3000 and 3500 kg ha^{-1}) compared to control Raj 171 (1008 kg ha^{-1} grain yield and 2583 kg ha^{-1} fodder yield); and three promising accessions, IP 3616, IP 6105 and IP 6101 (3083–3167 kg ha^{-1}) for high fodder yield compared to Raj 171 during 2005-rainy season. Based on the two seasons data the same two germplasm accessions (IP 22269 and IP 6098) were identified for grain (1411 and 1389 kg ha^{-1}) and fodder yield (5667 and 4806 kg ha^{-1}) compared to control Raj 171 and 2 of the 3 identified during 2005 (IP 3616 and IP 6105) for only fodder yield (5445 and 4196 kg ha^{-1}) as these had similar grain yield and higher fodder yield compared to control Raj 171 (1493 kg ha^{-1} grain and 3514 kg ha^{-1} fodder yield).

KN Rai, VN Kulkarni, V Vadez, HD Upadhyaya, P Pathak and TJ Rego

Milestone: New germplasm sources with different resistance mechanisms to *Helicoverpa*; resistance to *Ascochyta* blight (AB), *Botrytis* gray mold (BGM), wilt and root rot, and drought-avoidance root traits identified and introgressed in chickpea (2006)

Mechanism and inheritance of resistance to *Helicoverpa* in chickpea: Genetics of resistance to pod borer (*Helicoverpa armigera)* in chickpea was focussed on studying the nature of gene action and maternal effects,

plant resistance mechanisms and interaction of different components of resistance and grain yield. Eight *desi* [ICC 12475 (ICC 506), ICC 12476, ICC 12477, ICC 12478, ICC 12479, ICC 4918, ICC 12426 or ICCC 37 and ICC 3137] and one kabuli [ICCV 2 (ICC 12968)] parents were selected based on earlier screening trials to study the genetics of resistance, using full diallel. ICCV 2 was the earliest to flower and mature followed by ICC 4918, ICCC 37, ICC 12478 and ICC 12477, while ICC 12479, ICC 12476 and ICC 3137 were late to flower and mature. ICC 12478 suffered significantly lower damage, followed by ICC 506, ICC 12479 and ICC 12477. ICC 3137 was highly susceptible and recorded lowest seed yield. Most of the crosses with ICC 506, ICC 12478 and ICC 12479 suffered low damage, while those with ICC 3137 suffered higher damage. ICCC 37 recorded higher yield, followed by ICC 12479 and ICC 12476.

Inheritance of resistance: Gene action and maternal effects were estimated from the full diallel trial. Additive gene action was predominant for days to initial flowering, days to 50% flowering, days to maturity, pod borer damage (%), pods plant^{-1}, seeds plant^{-1}, seeds pod^{-1} and 100-seed weight. While non-additive gene action was important for yield plant^{-1}, total plot yield and yield (kg ha^{-1}). The additive: dominance (A:D) ratio was greater than unity for days to 50% flowering, days to maturity, pod borer damage (%), pods plant^{-1}, seeds plant^{-1}, seeds pod^{-1} and 100-seed weight, indicating over dominance, while for yield plant^{-1} and yield (kg ha^{-1}), the ratio was less than unity, indicating partial dominance. There was no maternal inheritance for maturity-related traits, pod borer damage, and grain yield. The hybrid, ICC 12476 × ICCC 37 showed positive and significant specific combining ability (SCA) effects for seeds pod^{-1}, but the reciprocal hybrid ICCC 37 × ICC 12476 showed negatively significant SCA effects for number of seeds pod^{-1}. So the hybrid ICCC 37 × ICC 12476 may be showing cytoplasmic effect for the number of seeds pod^{-1}.

Mechanisms of resistance: The three mechanisms of resistance *viz.,* non-preference for oviposition, antibiosis and tolerance to *H. armigera* in chickpea genotypes were studied under laboratory, greenhouse and field conditions. Oviposition studies under no-choice, dual choice and multi-choice laboratory and multi-choice field conditions revealed that the resistant control genotype, ICC 506 recorded lowest number of eggs, followed by ICC 12476, ICC 12477 and ICC 12478. The highest oviposition was observed on the susceptible genotypes, ICC 12426 and ICC 4918. The genotypes ICC 12475, ICC 12476, ICC 12477, ICC 12478 and ICC 12479 were least preferred by *H. armigera* females for oviposition compared to ICC 4918, ICC 3137 and ICCV 2. In detached leaf assay studies, the survival rate and larval weights were lowest on the resistant control, ICC 12475 (ICC 506), followed by ICC 12476, ICC 12477, ICC 12478 and ICC 12479, suggesting that water-soluble compounds in the leaf exudates (malic and oxalic acid) were primarily responsible for resistance to *H. armigera*.

Tolerance: The genotypes ICC 12476, ICC 12477, ICC 12478 and ICC 12479 were found to be resistant and their levels of resistance were comparable to the resistant control, ICC 12475 under no-choice cage conditions. Under un-infested conditions, the per plant yield was greater in ICC 12426 followed by ICC 12478 and Annigeri. The resistant cultivars ICC 12478 and ICC 12475 recorded higher yield than the rest of the cultivars. At the podding stage of the crop, when plants were infested with the third instar larvae, the recovery resistance was very poor, as most of the plants were damaged.

Larvae fed on leaf material and on artificial diet with lyophilized leaf and pod powder recorded lowest larval and pupal weights and prolonged larval and pupal periods on the resistant genotype, ICC 506. Highest growth index, adult index, oviposition index and pupal index were recorded on ICC 12426 and ICC 4918, while the lowest on the resistant control, ICC 12475.

High Performance Liquid Chromatography (HPLC) profile of leaf exudates showed that the malic acid was negatively correlated with damage rating at flowering (–0.28*), at maturity (–0.32**) and pod damage (–0.22*). Oxalic acid showed negative significant correlation with damage rating in detached leaf assay (–0.22*). Acetic acid showed a negative correlation with larval weight (-0.45*), damage rating at flowering (–0.33**) and maturity (–0.26*). Citric acid showed negative and significant correlation with damage rating at flowering (–0.23*).

The genotypes, ICC 12476, ICC 12477, ICC 12478, ICC 12479 and ICCV 2 were on par with the resistant control, ICC 12475 for pod borer damage under protected conditions. ICC 12475, ICC 12426, ICC 12478 and ICC 12479 recorded higher grain yield under unprotected conditions. The genotypes ICC 12475 (3.77%) and ICC 12478 (6.59%) recorded the lowest reduction in grain yield under unprotected conditions, indicating the presence of tolerance mechanism in chickpea to *H. armigera.* The tolerant lines can be used in further breeding programs and the mechanisms responsible for the resistance can be exploited to develop resistant varieties.

Correlation of different components of resistance with grain yield showed significant positive correlation under protected conditions between number of larvae and eggs (0.89**), leaf damage and egg number (0.82*), yield plant^{-1} and egg number (0.77*), yield plant^{-1} and larva number (0.76*) and pod damage (%) and larval number (0.91**). Significant negative correlation was recorded between yield plant^{-1} and borer damage (%) (–0.79*), under unprotected conditions. These correlations and interaction of different components of resistance and grain yield will help in gene pyramiding.

CLL Gowda and HC Sharma

Resistance to *Helicoverpa* introgressed into diverse chickpea breeding lines: One thousand five hundred and eighty six progenies (363 F_7 progenies and 721 F_8 progenies from single-crosses, and 502 F_7 progenies from four-way crosses) were sown under natural infestation of *Helicoverpa* larvae to select progenies for resistance. Selections were visually made for plants with early maturity, lesser pod damage and higher yields. We selected 1161 progenies (236 F_7 and 490 F_8 progenies from single-crosses and 435 F_7 progenies from four-way crosses) for progeny testing next year.

CLL Gowda

International Chickpea Screening Nursery – *Helicoverpa* Resistance (ICSN-HR): Using reliable field screening techniques developed at ICRISAT for screening against *Helicoverpa*, several resistant sources have been identified. The resistant (less susceptible) sources identified in field screening were used in crosses to transfer resistance in high-yielding varieties. Pedigree selection for low borer damage under pesticide-free conditions was found effective in identifying pod borer resistant lines. This trial is intended to share material showing resistance to *Helicoverpa* with the collaborating scientists of the national programs. Most lines are of short to medium duration, adapted to environments similar to southern and central India (16 to 22°N latitudes). The objective is to evaluate promising *Helicoverpa* resistant selections in varying environments and to provide an opportunity to NARS partners for selections for use as parents or as end products suitable for various conditions. The trial with 15 chickpea genotypes, including two controls, was sent to two collaborators.

CLL Gowda

Development of diverse range of breeding populations in chickpea: A total of 100 crosses were made during 2004. These included 71 crosses for AB resistance, 18 for BGM resistance, 3 each for *Helicoverpa* resistance, extra-large seeded kabuli lines and genetic studies. The resistance donor parents included ICCV 04516, ICCV 04538, ICC 3996, ICC 12004, ICC 12965, ICC 14917, PBG 5 and GL 90135 for resistance to AB; ICCV 98502 and ICCV 98503 for resistance to BGM; and IG 72933, IG 72953 and ICC 506 EB for resistance to *Helicoverpa* pod borer. Forty-one of these crosses were made in greenhouse during off-season and thus F_1s from these could be grown during the crop season 2004/05 along with 222 crosses made during the crop season 2003/04. Thus, a total of 263 F_1s were grown during crop season 2004/05. These included 227 (119 *desi* × *desi*, 103 *kabuli* × *kabuli* and 5 *desi* × *kabuli*) for improvement of yield, seed traits and resistance to *Fusarium* wilt; 32 for enhancing resistance to AB; and 4 for enhancing resistance to *Helicoverpa* pod borer. Fifty-four new crosses were made in greenhouse during the off-season. These included 18 crosses involving newly identified AB resistant line ICCV 04502, 8 crosses involving salinity-tolerant lines (ICC 2580 and L 550), 8 crosses involving lines with deeper and vigorous root systems (ICC 4958 and ICC 8261), 3 crosses involving dry root rot resistant parents (MPJG 98-9023, MPJG 98-11151), 8 crosses involving super-early line ICCV 96029, 7 crosses involving extra-large-seeded *kabuli* lines, and 2 crosses involving multipinnate line ICC 5714. Seventy crosses were advanced by one generation (60 F_1s and 10 F_2s) and 38 crosses by two generations (F_1 and F_2 from 20 crosses, and F_2 and F_3 from 10 crosses) during the off-season in greenhouse. The cross made between cultivated chickpea line ICC 506 EB and of *C. reticulatum* (IG 72953) for combining different mechanisms of resistance to *Helicoverpa* was advanced by two generations (F_2 and F_3) in the off-season.

PM Gaur

Evaluation of germplasm lines for AB resistance under controlled environment conditions: *Ascochyta* blight (AB) caused by *Ascochyta rabiei* is an important foliar disease of chickpea that can cause complete loss of grain yield. Epidemics of AB are frequently associated with prevailing cool and humid weather and incidence of this disease in chickpea growing areas is spatially separated. Few resistant sources for AB are available, but most of them are susceptible to *Fusarium* wilt. Hence, continued attempts are being made at ICRISAT-Patancheru under controlled environment conditions for identification of additional sources of resistance.

Resistance screening technique: Resistance screening in controlled environment enables a rapid identification for resistance to AB. For identification of resistance, 10-day-old seedlings raised in plastic trays (30 × 20 × 5 cm) filled with sterile sand and vermiculite (4:1) were spray-inoculated with a conidial suspension (5×10^4 conidia ml^{-1}) of *A. rabiei* multiplied on autoclaved *kabuli* chickpea seed. One line of susceptible cultivar Pb 7

was planted in each tray as control. Inoculated seedlings were incubated at 20 ± 2°C and 100% RH for 96 h. Thereafter, 100% RH was provided for 8–16 h a day. Disease severity was scored at 10 days after inoculation on 1–9 rating scale where 1 = no disease and 9 = >75% of the plants killed.

AB resistance in germplasm lines: Using the above standard procedure, we evaluated 344 germplasm accessions twice for resistance to AB. ILC 3864 and FLIP 83-23C were resistant to AB with a mean disease severity of ≤3.0 rating on 1–9 rating scale and 80 lines were moderately resistant (disease rating 3.1–5.0) in comparison to susceptible control (9.0 rating). Promising lines were included in *Ascochyta* Blight Nursery and will be tested in different locations in India.

***Ascochyta* Blight Nursery (ABN):** Thirty-six germplasm entries, identified as promising for AB resistance in the controlled environment evaluation, were included in ABN during 2004/05 season. The nursery was evaluated under field conditions at Dhaulakuan (CSKHPKV), Gurdaspur (PAU), Ludhiana (PAU), Hisar (CCSHAU) and ICRISAT-Patancheru (under controlled environment conditions) in India. Each entry was planted in two replications with one row 2–4 m long in each replication (2 m long in Gurdaspur, Ludhiana, Hisar; and 4 m long in Dhaulakuan). Artificial inoculations with conidial suspension were done at flowering and pod initiation stage of the crop in all locations. Susceptible cultivar Pb 7 was found susceptible at all the locations tested. All the entries were found highly susceptible to AB at Hisar. Fifteen were found moderately resistant (3.1 to 5.0 on 1–9 rating scale) at ICRISAT-Patancheru, Dhaulakuan, Gurdaspur and Ludhiana. Cultivars ICC 652, 15976, 15980 were found highly susceptible at Gurdaspur, while they were moderately resistant (<5 rating) at other locations.

Evaluation of germplasm lines for BGM resistance under controlled environment conditions

Botrytis gray mold (BGM) caused by *Botrytis cinerea* is another destructive foliar disease that can cause complete loss of grain yield in chickpea. Frequent epidemics of BGM are associated with cool and humid weather followed by frequent winter rains. Growing resistant sources is the most economical way to manage this disease. But adequate levels of resistance to BGM are not available in cultivated chickpea. Hence, continued attempts are being made at ICRISAT for identification of sources of resistance to BGM.

Resistance screening technique: A reliable and reproducible screening technique for BGM resistance in controlled environment has been established. Eight to ten-day-old seedlings of test lines, along with JG 62 as a susceptible control were inoculated with conidia of *B. cinerea* (3×10^5 conidia ml^{-1}) multiplied on autoclaved merigold flowers. Inoculated plants were maintained at 15 ± 2°C and 100% RH with a 12 h photoperiod. BGM severity was recorded on a 1–9 rating scale at 20 days after inoculation.

BGM resistance in germplasm lines: One hundred and sixty one promising lines identified for BGM during 1985 to 2000 were evaluated for BGM resistance in controlled environment. A set of 60 germplasm lines, selected based on the diversity in their genotyping profiles were also tested for BGM resistance. Of the 161 germplasm lines tested, 35 were found promising (<5 rating on 1–9 scale). Of the 60 diverse germplasm, 4 lines were found moderately resistant (<5 rating on 1–9 scale).

***Botrytis* Gray Mold Nursery (BGMN):** The nursery consisted of 29 BGM promising entries identified in the controlled environment evaluation at ICRISAT-Patancheru during 2004/05 season. One local susceptible cultivar was also included for comparison. The entries of this nursery were evaluated at ICRISAT-Patancheru, Pantnagar (GBPUA&T), Gurdaspur (PAU) and Ludhiana (PAU) in India; Tarahara in Nepal; and Ishrudi and Jessore in Bangladesh. The entries were evaluated under controlled environment conditions at ICRISAT-Patancheru. Each entry was planted in two replications with one row (2–4 m long) in each replication. Artificial inoculations with conidial suspension were done at flowering and pod initiation stage of the crop in all the locations except in Tarahara, Nepal. Data from all the locations were received, except from Bangladesh. Susceptible cultivar H 208 showed susceptible reaction in all the locations in India and Nepal. BGMN at Gurdaspur location was planted in AB nursery and hence the reaction of these entries was not considered. Two entries (ICC 1069 and ICCL 87322) at Pantnagar; 10 entries (ICC 8509, 12339, ICCVs 89302, 98505, ICCL 86215, ICCX 860030-BP-BP, ICCX 880030-BP-BP-6PN-BPN-BP, ICCX 860023-BP-BP-BP-3P-BH-1H-BH, ICCX 860029-BH-1PN-BPN-B and ICCX-880355-BH-BP-5H-BH) at ICRISAT-Patancheru, India and three entries (ICCs 8509, 12512 and 12952) at Tarahara, Nepal had <5 rating on 1–9 rating scale. None was resistant at Ludhiana, India.

Evaluation of germplasm lines for wilt resistance under field conditions

Wilt caused by *Fusarium oxysporum* f. sp. *ciceri* (FOC) is a very serious soil-borne disease in most of the chickpea growing areas in the world. Growing resistant lines is the best way to manage this disease. Though wilt resistant lines are available, their resistance frequently breaks down at other locations due to existence of physiological races in the pathogen. Hence we continued to identify lines with broad-based resistance.

Resistance screening for wilt: Large-scale evaluation of breeding and germplasm lines for resistance to wilt was conducted in a wilt sick plot at ICRISAT-Patancheru. Wilted chickpea plants were chopped and incorporated in the field every year to maintain threshold levels of the fungus. A multiple disease sick plot (MDSP) consisting of pathogens of wilt (*Fusarium oxysporum* f.sp. *ciceri*) followed by dry root rot (*Rhizoctonia bataticola*), collar rot (*Sclerotium rolfsii*), black root rot (*Fusarium solani*) and wet root rot (*Rhizoctonia solani*) is also available at ICRISAT-Patancheru for evaluating wilt promising material for wilt and root rots. Each test entry was planted in two rows 4 m long with two replications. Early-wilting susceptible cultivar ICC 4951 was sown after every four test rows, and late-wilting cultivar ICC 5003 and resistant cultivar ICC 11322 were sown alternately after every 12 test rows (ie, after every 15 rows), along with test material for proper comparison. Periodical observations on number of wilt and root rot infected plants were recorded. Lines showing <10% incidence were considered resistant and their resistance was confirmed in the greenhouse and laboratory using individual screening techniques.

Wilt and root rot resistance in germplasm lines: One hundred and forty-one advanced wilt and root rot promising selections were further evaluated for resistance to wilt and root rots in multiple disease sick plot (MDSP). Of the 141 lines evaluated, 70 had combined resistance to wilt, DRR and collar rot under field conditions.

Chickpea wilt observation nursery (CWON): Twenty-eight wilt promising entries identified at ICRISAT-Patancheru and two susceptible controls were included in this nursery that was evaluated at 12 locations in India: Akola, Dharwad, Gurdaspur, ICRISAT-Patancheru, Hisar, Hazribagh, Jabalpur, Junagadh, Ludhiana, Raipur, New Delhi, and Sehore during 2004/05 season. Each entry was planted in two replications in one row of 4 m in each replication. Data on wilt was recorded twice at flowering and at maturity stages of the crop. Data from 10 locations were received (Gurdaspur, ICRISAT, Hisar, Hazribagh, Jabalpur, Junagadh, Ludhiana, Raipur, New Delhi and Sehore) and compiled. Wilt sick nursery at Ludhiana consisted of mixture of three soil-borne diseases, foot rot (*Operculella padwickii*), black root rot (*F. solani*) and wilt (*F. oxysporum* f.sp. *ciceri*). Since foot rot and black root rot were dominant, most of the lines were found free from wilt. Hence, the reaction of the entries in this location was ignored. The susceptible cultivar ICC 4951 was susceptible at all the locations except Gurdaspur where its incidence was 27.6%. ICCX-950106-F4-43P-BP was resistant in seven locations; and ICCs 12467, 14409, 14433, and 11322 were resistant in six locations.

S Pande, PM Gaur, J Narayana Rao and GK Kishore

Quantification of fungal pathogens of chickpea in wilt and multiple disease sick plots: In the second year of conducting this experiment (initiated in the year 2003/04) to quantify fungal pathogens of wilt complex of chickpea in wilt (BIL 3C) and multiple disease sick plots (BIL 1) at ICRISAT-Patancheru, soil samples were collected and processed exactly like previous year before planting and after harvesting of chickpea crop in these two fields. As in the previous year, number of fungal colonies g^{-1} soil was counted at 48 to 96 h after incubation. Number of sclerotia of *S. rolfsii* was quantified by using rapid floatation technique. As in the previous year, fungal colonies of the wilt and root rot were high in the samples collected after harvest of the crop in both fields. Number of *Fusarium oxysporum* f.sp. *ciceri* (FOC) colonies was around 1300 g^{-1} soil before planting and increased enormously during the crop growth and reached to around 3000 g^{-1} soil after harvest of the crop in both fields. Colonies of FOC were recovered up to 75 cm depth before planting and up to 100 cm depth immediately after harvest of the crop in both fields. In BIL 1, colonies of *Fusarium solani* and *Rhizoctonia bataticola* were, respectively, around 310 and 190 g^{-1} of surface soil, collected before planting. These two pathogens too multiplied during the cropping period and doubled after harvest of the crop. Both these pathogens were recovered up to 50 cm depth before planting and up to 65 cm after the harvest of the crop in BIL 1. About three sclerotia of *S. rolfsii* 10 g^{-1} soil were recovered from the surface soil collected before planting while its number increased to 6.5 at the end of the crop season in this field. As in the previous year, negligible number of these root rot pathogens was observed in wilt sick plot. Number of colonies of wilt and root rot fungi decreased as the depth increased. It was observed that the number of colonies of wilt complex fungi increased from two to three folds after harvest of the crop during February than October (before planting). The wilt and multiple disease sick plots are kept fallow every year from February to October. It was evident that during this period, the number of colonies of these fungi is reduced drastically and the reduction may be due to the absence

of the chickpea crop. However, the reduction is not below threshold levels of the pathogens as the susceptible control planted during October, killed with in the stipulated time (<30 days after sowing).

Succession of fungal pathogens of chickpea in wilt and multiple disease sick plots: This is the second year of conducting this experiment (initiated in the year 2003/04) to find out the sequence of occurrence of wilt complex diseases in chickpea in wilt and multiple disease sick plots. Methodology, including cultivars (JG 62, L 550 and WR 315), sampling and isolations on nutrient-rich (potato dextrose agar) and semi-synthetic media (czapek dox agar) were similar as in the previous year in both fields (Archival Report 2004). As in the previous year, isolations were made from root tip, root hair, epidermis and cortex, vascular bundles, and collar region in each cultivar at 10-day interval from both fields.

Multiple-disease sick plot: Wilt fungus was found dominant from seedling to harvesting stage of the crop. Isolation on both the media indicated that FOC was recorded from all the root parts from 20 days after sowing in highly susceptible (early wilter) and moderately susceptible (late wilter) cultivars and continued to be present till the death of the plants. All the plants of the cultivar L 550 (late wilter) wilted at 90 days after sowing (DAS). Highly susceptible cultivar JG 62 completely wilted in 30 days after sowing in this field. FOC was found in root tip and root hairs in resistant cultivar WR 315 at 50 days after sowing and continued till maturity. This late infection and restriction of the fungus at root tip in this cultivar may be due to its resistance to the wilt pathogen. Moreover, the plants of this cultivar remained healthy till maturity in this field indicating its resistance to this pathogen. Black root rot fungus (*Fusarium solaris*), attacks the crop between 20 and 30 days after sowing (ie, in seedling stage) when the soil moisture is high. Soil moisture stress and warm temperatures encourage the dry root rot (*Rhizoctonia bataticola*) fungus causing rotting of the roots. During this season, dry root rot appeared from 40 DAS till maturity in both late wilting L 550 and resistant WR 315.

Wilt sick plot: Similar to multiple disease sick plot, FOC was recorded from 20 days after sowing in all the root parts in early (JG 62) and late wilting (L550) cultivars. Susceptible cultivar JG 62 died completely within 30 days after sowing. FOC was observed from all the root parts up to 90 days in L 550 and later all the plants wilted. Resistant cultivar WR 315 yielded FOC only from root tip and root hair from 40 days after sowing as in multiple disease sick plot. Though basically it is a wilt sick plot, low intensities of black root rot fungus was recorded up to 30 days after sowing (ie, in seedling stage) was recorded. Similarly, dry root rot fungus was observed from 50 days after sowing till maturity in both late wilting L 550 and resistant WR 315.

S Pande and J Naranayana Rao

Detection of seed-borne nature of *Fusarium oxysporum* f.sp. *ciceri*: It is reported that the wilt fungus is transmitted through seed. To confirm the seed-borne nature of the wilt fungus, an experiment was conducted using chickpea seeds collected from different plants that wilted prior to maturity (late wilting) from wilt sick plot and from healthy plants of wilt susceptible cultivar JG 62 from wilt-free field during 2004 crop season. These seeds were air dried at room temperature, bulked separately and stored at 5°C in the refrigerator in the laboratory. An experiment to detect the seed born nature of FOC was conducted at ICRISAT- Patancheru. Four hundred seeds (100 seeds in each replication) were taken from wilted and healthy lots, surface sterilized with 2.5% clorox for 5 min and plated onto the modified czepek dox agar medium in 10 cm glass petri plates @ 10 seeds per plate. All the plates were incubated at 25°C with 12 h light and 12 h dark periods for seven days. About 22% of the seeds collected from wilted plants yielded FOC and no fungus was observed from seeds collected from healthy plants. This indicated that FOC was seed-borne and present in wilted seeds collected from late wilting plants.

S Pande

Seed treatment with fungicides to control seed-borne inoculum of *Fusarium oxysporum* f. sp. *Ciceri*: Chickpea seeds collected from wilted plants were treated with six fungicides and their combinations (1:1 commercial formulations) @ 2.5 μg kg^{-1} seed to control seed-borne inoculum of *Fusarium oxysporum* f. sp. *ciceri* (FOC). The treatments (fungicides and their combinations) included in this study were Bavistin, Benlate, Captan, Indofil M 45, Kavach, Thiram, Benlate + Thiram, Bavistin + Thiram, Bavistin + Indofil M 45, Captan + Thiram, Indofil M 45 + Thiram and Kavach + Thiram. Three hundred fungicide-treated seeds (3 replications with 100 seeds in one replication) from each treatment were plated on czepek dox agar medium @ 10 seeds per plate. Equal number of untreated seeds was also plated for proper control. All the plates were incubated in Percival incubator at 25°C with 12 h light and 12 h dark for seven days. FOC was present in 26.5% of untreated seeds (control treatment). FOC colonies were low in the seeds treated with fungicide combinations than individual fungicides and control. Lowest number of FOC colonies were recorded in the seeds treated with Benlate + Thiram (3.3%) followed by Bavistin + Thiram (5.3%), and Bavistin + Indofil M 45 (5.7%). Since

thiram is unavailable to most of the farmers in several villages, it is more convenient to the farmers to use Bavistin + Indofil M 45.

S Pande

Establishment of *Fusarium oxysporum* f. sp. *ciceri* in the soil through seed transmission

It was evident that the wilt fungus is transmitted through seed harvested from wilt-affected plants (mostly late wilting). Wilt incidence has been increasing considerably in farmers' fields over the years. To find out the establishment of *Fusarium oxysporum* f. sp. *ciceri* in soil through infested seeds, we conducted an experiment consisting of three serial sowings representing three crop seasons in the same pot (undisturbed soil) under greenhouse conditions.

First sowing representing first crop season: One hundred seeds collected from wilted plants during 2004 crop season were sown in small (5 cm dia) ice-cream cups (pot) filled with sterilized soil + sand mixture (1:1). One seed was sown in each pot to avoid contamination. Seeds collected from healthy plants of the cultivar JG 62 were used as control for proper comparison. Seedlings were observed for 45 days for wilt symptoms. Data on number of plants wilted and days to wilt were recorded. Isolations were made from all the dead plants on *fusarium* selective medium (Czepak dox agar) for confirmation of wilt. At the end of the experiment, small quantity of soil from each pot was taken, dried and assessed for *fusarium* propagules on selective medium. Of the 100 plants, 19 plants exhibited wilt symptoms in 35 to 45 days after sowing. FOC was recovered from all the wilted plants on selective medium. About 100 to 250 propagules of FOC per gram of soil were recorded from the pots where wilt was observed.

Second sowing representing second crop season: Above-ground parts of all the plants from all the pots were removed by cutting at collar region so that the root system of each plant remained in soil. Soil in the pots was stirred with sterilized iron rod and pots were undisturbed for a week. Seeds collected from healthy plants of the cultivar JG 62 were planted @ one seed in one pot and observed for 45 days for wilt symptoms. Data on all the parameters were exactly recorded as in the first sowing. Plants in all the 19 pots, where wilt was observed in the first sowing plus five more new pots (total 24 pots), had wilt symptoms at the end of the experiment. It took about 25–30 days for all the 19 pots (old) and around 35 days for the five new pots to exhibit wilt symptoms in the second sowing. Isolation of wilted plants on selective medium yielded FOC. Number of propagules of FOC multiplied very fast in the pots during the crop season and reached to 900 to 1600 propagules g^{-1} of soil.

Third sowing representing third crop season: Roots of each plant were chopped and incorporated in soil in the pot and stirred with sterilized iron rod. Seven days later, chickpea seeds collected from healthy plants of the cultivar JG 62 were sown @ one seed in one pot and observed for 45 days for wilt symptoms. Data on all the parameters was recorded as in the first sowing. Wilt symptoms appeared in all the 24 pots which had wilt in the second sowing. Wilt was observed between 14 and 18 days after sowing, which was much earlier, compared to first and second sowings. All the wilted plants yielded FOC on selective medium. Soil collected from the pots from where wilt was observed, had 1900 to 3150 FOC propagules g^{-1} of soil and no propagules of FOC were observed in other pots.

S Pande

Milestone: New germplasm sources of resistance to biotic and abiotic stresses, and confectionery traits identified and introgressed in groundnut (2004)

Screening groundnut breeding lines for foliar disease resistance [late leaf spot (LLS) (*Phaeoisariopsis personata*) and rust (*Puccinia arachidis*)]

Resistance of advanced breeding lines to LLS and rust: Six replicated yield trials (Elite foliar diseases resistant groundnut variety erect bunch (Spanish (var. vulgaris) and Valencia (var. fastigiata)) (EFDRGVT-SB), Elite foliar diseases resistant groundnut variety spreading bunch (Virginia bunch (var. hypogaea)) (EFDRGVT-VB), Advanced foliar diseases resistant groundnut variety Spanish bunch (AFDRGVT-SB), Advanced foliar diseases resistant groundnut variety Virginia bunch (AFDRGVT-VB), Preliminary foliar diseases resistant groundnut variety Spanish bunch (PFDRGVT-SB) and Preliminary foliar diseases resistant groundnut variety Virginia bunch (PFDRGVT-VB), consisting of 109 advanced breeding lines were screened for resistance to rust and LLS in an artificial sick plot containing trial inoculum and infector-rows. Experiment was laid out in a broad-bed-and-furrow (BBF) system with two replications. Chemical sprays were used to control insect pests. At 50 days after sowing, plots were inoculated by spraying the infected and test rows with 20 L of mixed conidial suspension of *P. personata* and *P. arachis dis* urediniospores. After inoculation, perfo-irrigation was

provided daily for 15 min in the evening hours for 30 days to increase humidity required for disease development. Diseases (LLS and rust) were scored based on a 1–9 rating scale at 75, 90 and 105 days after sowing (1 = no disease, 9 = 81-100% foliage damage).

Development of LLS and rust was uniform throughout the infector rows and 100% infection was observed in the susceptible controls. Of the 109 breeding lines, ICGV 05122 showed good resistance to LLS with an overall disease severity score of 4.0. Another two lines (ICGV 05089 and ICGV 05096) had a score of 4.5; 25 lines scored 5.0; and 7 lines scored 5.5. Remaining lines were found to be susceptible (disease score 6–8) at 103 days after sowing. Seventy-three lines showed good resistance to rust (disease score of 1 to 2); 26 lines scored 2.5 to 3; and the remaining advanced breeding lines scored >4 (on a 1–9 scale) at 103 days after sowing.

Resistance of F_2–F_6 population to LLS and rust: During the 2005 rainy season, 239 lines from 5 breeding populations (F_2–F_6) were evaluated for resistance to LLS and rust under field conditions. Test plants were evaluated based on their disease severities at the time of harvest measured on 1–9 disease rating scale. A few single plants from F_6 population showed high levels of resistance to LLS (disease score 1 to 3). Remaining lines scored >4 on 1–9 rating scale.

Inheritance of late leaf spot resistance: Progenies of three crosses (ICGV 11337 × JL 24, ICGV 13919 × JL 24 and ICGV 11337 × ICGV 13919) planted in replicated trial with their reciprocals along with 10 generations in 60 cm inter-row space and 10 cm space between plants with two germplasm parents and one susceptible cultivar (total 2440 single plants) were screened for resistance to LLS under field and controlled environment conditions during 2005 season.

Plants in field trials were spray-inoculated with conidial suspension of *Phaeoisariopsis personat* at 50 days after sowing. After inoculation, perfo-irrigation was provided daily for 15 min in the evening hours for 30 days. Subsequently, percent leaf area damage and percentage defoliation was measured in each plant. Plant reaction to LLS was recorded based on a 1–9 rating scale at 75, 90 and 105 days after sowing (DAS). In all three crosses, 89 progenies showed high levels of resistance to LLS with a score of 2.0; 225 had a score of 3.0; 205 observed score of 4.0; and 140 recorded score of 5.0 at 105 days after sowing. Remaining lines scored >6.0 score. Mean disease severity of all the plants in each generation showed that generation 2 (ICGV 11337 in Cross 1 and ICGV 13919 in Cross 2 and 3) was most resistant to LLS. Also, cross 3 (ICGV 11337 × ICGV 13919) recorded lower disease score in all the generations than other two crosses.

The fully expanded quadrifoliate leaves of 45-day-old plants (third or fourth from top) of each line were excised and planted in sand cultures (roughly 1.5 cm thick) prepared in plastic trays (39.5 cm × 29 cm × 7 cm) for evaluation under controlled environmental conditions. In each tray, 20 leaflets were planted and trays were covered with plastic bags and incubated in the growth chamber at 24°C temperature and 85% relative humidity. LLS inoculum (30,000 spores/ml) was sprayed on both the surfaces of each leaflet inserted in the sand plates. Leaves were sprayed with water daily up to 5 days after inoculation. Observations were taken as shown below:

- Incubation period: Every alternative day from 5 days after inoculation
- Latent period: Every alternative day from 5–37 days after inoculation
- Lesion number: Every alternative day from 5–21 and 30 days after inoculation
- Percent leaf area damage: Every alternative day from 5–21 and 30 days after inoculation
- Lesion diameter: 25 days after inoculation

Some plants showed latent period (LP) of 30–36 days after inoculation and 2–5 lesions with minimum lesion diameter (<0.5 mm). The screening results for LLS indicated that the resistance levels were higher in cross ICGV 11337 × ICGV 13919 then in cross ICGV 11337 × JL 24 and ICGV 13919 × JL 24. These lines during field evaluation scored 2–4 (on 1–9 rating scale), indicating that they contain good resistance to LLS.

Farid Waliyar and SN Nigam

Screening of wild *Arachis* species for resistance to *Tobacco streak virus* (TSV) in the glasshouse: Twenty-three accessions of wild *Arachis* species were screened twice in the glasshouse. Promising accessions were further evaluated. Seven accessions, viz., ICG 8139, 8200, 11550, 8195, 8203 and 8205 belonging to *A. duranensis* and ICG 13210 belonging to *A. stenosperma* showed consistent resistance to TSV. These accessions did not show any systemic infection. Two accessions (ICG 8139 and 11550) also possessed high levels of

resistance to rust and late leaf spot and thus can be used in interspecific crosses to develop multiple disease-resistant groundnut varieties.

Farid Waliyar, P Lava Kumar and SN Nigam

Effect of virus titer and date of inoculation on infectivity of *Peanut bud necrosis virus* (PBNV) *Tobacco streak virus* (TSV) and *Indian peanut clump virus* (IPCV): Experiments were conducted in the greenhouse with groundnut variety JL 24 to determine the effect of plant growth stage on infection with PBNV, TSV, and IPCV. Plants were inoculated at 10, 20 and 30 days after germination with three dilutions 1:10, 1:50 and 1:100 (leaf wt./buffer) of inoculum. Results indicated that IPCV, PBNV and TSV infection in groundnut can occur at 10 and 20 days after germination and result 100% infection. In case of plants inoculated at 30 days after germination, infection reduced by about 50% and symptoms development was delayed for all the three viruses. However, 90% of the test plants inoculated with IPCV were infected and showed mild symptoms. These results have implications on field protection of susceptible host plants and also in assessing host resistance against these viruses.

P Lava Kumar and Farid Waliyar

Survey for seed-borne groundnut virus diseases in genetic resources seed multiplication fields: In 2004 rainy season, germplasm lines were scored for seed-borne virus diseases in RP7 field at ICRISAT-Patancheru. Nineteen suspected plants were tagged and tested by ELISA. None contained any infected plants. All plants were uprooted and burned. During the post-rainy 2004/2005 survey, 20 suspected plants were tagged and tested by ELISA for PMV and PStV. Two plants (one in ICG 8323 and another one in ICG 10171) were positive to PMV. All plants were uprooted and burned.

In 2004 rainy season, one more field (RP5) was inspected and 14 suspected plants were found. They were tagged and tested by ELISA for the presence of PMV and PStV but none contained any virus. All the suspected plants were destroyed. In postrainy 2004/2005, 47 suspected plants were tagged and tested by ELISA for PMV, PStV but none contained virus. These suspected plants were destroyed.

P Lava Kumar and Farid Waliyar

Activity 1.2.2: Develop diverse range of populations and breeding lines with improved yield potential, resistance to biotic and abiotic stresses and better quality

Milestone: High-yielding sorghum lines with large grains and resistance to grain mold and shoot fly developed (2006)

Breeding for high grain yield and large grain size: A total of 74 F_2s derived from high-yielding B-lines (HYB) × HYB, Lustrous B-lines × HYB, new B-lines × advanced progenies and sweet-stalk B-lines × sweet-stalk B-lines crosses and 924 F_3s derived from HYB × HYB crosses were evaluated during the 2005 rainy season. Based on grain size, plant height and overall agronomic visual score, 78 F_3s and 173 F_4s, respectively were produced, which are being evaluated during the 2005–06 postrainy season.

Several early-maturing advanced progenies derived from the crosses between postrainy season adapted varieties, high-yielding B-lines and landraces were evaluated during the 2004–05 postrainy season. A total of 44 F_6 progenies from 50 F_5 progenies, 530 F_5 progenies from 651 F_4 progenies and 140 F_4 progenies from 956 F_3 progenies, 63 F_7 progenies from 9 selections from participatory plant breeding program were selected and are being advanced through selection. These will be testcrossed on A_1 system A-lines to assess their male-sterility maintainer or fertility restorer reaction during the 2005–06 postrainy season.

BVS Reddy and S Ramesh

Grain mold-resistance (GMR) breeding

Testcrossing of agronomically superior grain mold-resistant advanced breeding lines: A total of 100 breeding lines previously screened for GMR that included B-lines, R-lines and varieties were evaluated during 2005 rainy season and 62 lines were selected based on the plant agronomic appearance. The varieties are being testcrossed onto A_1 and A_2-based A-lines during the 2005–06 postrainy season to assess their male-sterility maintainer or male-fertility restoration reaction.

A set of 72 F_5 progenies (derived from 102 F_4s of the crosses between grain mold resistant landraces, high-yielding B-lines (HYB) lines and grain mold resistant breeding lines) and another set of 17 F_5 progenies derived

from crosses between GMR B-lines and HYB-lines were selected for GMR during the 2005 rainy season. Among the selected progenies, days to 50% flowering ranged from 61 to 75 days and plant height from 1.3 to 2.3 m. These will be further selected and testcrossed on A_1 and A_2 CMS systems to assess their male-sterility maintainer or male-fertility restoration reaction.

BVS Reddy, S Ramesh and RP Thakur

Evaluation of advanced breeding lines for grain mold resistance: Enhancing the level of grain mold resistance in breeding lines continues to be a priority. Ninety-six F_5 lines, selected from crosses involving 9 elite B-lines and 8 mold tolerant lines were evaluated for grain mold resistance in the field nursery. Each entry was grown unreplicated in 2 row-plots of 4 m long and overhead sprinklers were provided on rain-free days for grain mold infection and development from flowering to physiological maturity. In each entry 10 uniformly flowering plants were tagged. Panicle grain mold rating (PGMR) was recorded at physiological maturity on threshed grain mold rating (TGMR) on bulk threshed grain of 10 tagged panicle per plot using a 1–9 scale (1 = no mold, 2 = 1–5%, 3 = 6–10%, 4 = 11–20%, 5 = 21–30%, 6 = 31–40%, 7 = 41–50%, 8 = 51–75% and 9 = >75% grains molded). Considering the mean grain mold severity rating of 1–4 (up to 20% infection) as resistant and 4–9 as susceptible classes, 9 lines were found resistant and the remaining were susceptible, while the susceptible controls Bulk Y and 296B had scores of 8.4 and 9.0, respectively.

RP Thakur and BVS Reddy

Evaluation of single plant selections for grain mold resistance: One hundred single plant selections from the 2004 rainy season were evaluated in a RCB design with two replications, one row per replication in the grain mold nursery as described above. Both PGMR and TGMR scores revealed that 56 lines were resistant (≤4.0 score) compared to ≤1.4 in resistant control, IS 8545 and 9.0 in susceptible control, SPV 104. This shows a significant improvement in grain mold resistance level within one generation of pedigree selection. We have selected 124 single plants for further evaluation and selection.

RP Thakur

Evaluation of hybrids and their parents for grain mold resistance stability: Under the Indian Council of Agriculture Research (ICAR) (All India Coordinated Sorghum Improvement Project)-ICRISAT partnership project, we evaluated the resistance stability of 29 hybrids and their 18 parental lines through a collaborative Sorghum Grain Mold Resistance Stability Nursery (SGMRSN). In addition to the 47 test entries, the nursery consisted of two resistant (IS 25017 and IS 14384) and one susceptible (CSH 16) controls. The 50-entry nursery was conducted at five Indian locations (Coimbatore, Dharwad, Parbhani, Palem and Patancheru) in a RCB design with two replications. Each entry was planted in 2-row plots of 4 m. Sprinkler irrigation was provided from flowering to physiological maturity of the crop for infection and mold development. Five plants with uniform flowering were tagged in each row (10 plants plot^{-1}), and the panicle grain mold rating (PGMR) was recorded at physiological maturity and threshed grain mold rating (TGMR) on bulked threshed grain of 10 tagged panicles per plot with the help of a magnifying glass using a 1–5 scale (1 = no mold, 2 = 1–10%, 3 = 11–25%, 4 = 26–50% and 5 = >50% grains molded) on the tagged panicles. Data on agronomic traits, such as plant height, days to 50% flowering, glumes cover, and grain hardness were also recorded. The data from Coimbatore were not considered since insects damaged the nursery at the time of grain development.

The analysis of variance indicated highly significant ($P < 0.001$) effects of location, genotype, and their interaction on mold severity scores. The mean PGMR of the 47 test entries across four locations varied from 2.0 to 3.7 compared to 1.4 to 2.2 in resistant controls and 3.4 in the susceptible control. The mean PGMR score across entries was highest at Patancheru (3.6) followed by Palem (3.0), Dharwad (2.9) and the lowest was at Parbhani (1.6). The mean TGMR of test entries across the four locations varied from 2.4 to 4.5 compared to 1.7 to 2.8 in the resistant controls and 4.2 in the susceptible control. The mean TGMR score across entries was highest again at Patancheru (3.9) and lowest at Parbhani (2.9). Considering the mean grain mold severity rating of 1–2.5 (up to 20% infection) as resistant and 2.5–5 as susceptible classes (on the 1–5 scale) both for PGMR and TGMR, 9 of the 28 hybrids and 4 of the 19 parental lines were resistant across locations and thus these seemed to have resistance stability against grain mold pathogens. Further evaluation of these hybrids and parental lines would be required to confirm the results.

RP Thakur and BVS Reddy

Shoot fly resistance (SFR) breeding: A total of 81 advanced (F_6) progenies derived from the crosses between HYB and shoot fly resistant breeding lines were screened for SFR using interlard fish meal screening technique during the 2005 rainy season. Eight lines which supported lower deadhearts (DH) at 17 days after emergence (DAE) (31% DH compared to 57% DH in the susceptible control, 296B) were selected. These will be testcrossed onto A_1 and A_2 cytoplasms during 2005-06 postrainy season.

From the advanced progenies evaluated during the 2004-05 postrainy season, 80 F_4s, 174 F_5s, and 17 F_6s were selected and are being advanced with selection. These will be testcrossed onto A_1 and A_2 CMS systems during the 2005–06 postrainy season. The 15 stabilized lines earlier confirmed as maintainers of A_1 and with SFR (39–57% DH compared to 42% DH in 296B at 21 DAE) for postrainy season adaptation were advanced for conversion into male-sterile lines during the 2005-06 postrainy season.

BVS Reddy, HC Sharma and S Ramesh

Milestone: Sorghum and pearl millet breeding lines (from existing collections) having high levels of Fe, Zn and β-carotene content; and salinity tolerance identified, and characterized for their yield potential and agronomic traits (2006)

Micronutrient density: A total of 40 lines with high and low differentials were selected and evaluated for their stability for agronomic traits and grain Fe, Zn and β-carotene contents under three applied nitrogen levels (120 kg, 80 kg and 40 kg N ha^{-1}) during the 2004–05 postrainy season. The results indicated significant genetic variability for grain Fe and Zn contents. The genotype × N interaction was not significant, indicating the selections made at one fertility level perform similarly at other fertility levels. The mean grain Fe content of the lines appeared to be highest when grown under recommended dose of nitrogen fertilization, while it was significantly lower when the lines were grown under low levels of N fertilization. The grain Zn content did not vary with the levels of N fertilization. These results indicate that the lines bred for high grain Fe and Zn contents need to be grown in recommended dose of managed fertility level to derive maximum benefit.

Based on the mean grain Fe, Zn and phytates contents of the entries across two locations [ICRISAT-Patancheru and National Research Centre for Sorghum (NRCS), Hyderabad], 12 entries (which include the lines with high and low Fe and Zn contents) have been selected to assess their stability across different soil Fe and Zn fertilization and soil types (alfisols and vertisols) at ICRISAT-Patancheru during 2005–06 postrainy season.

BVS Reddy and S Ramesh

Salinity tolerance: Promising hybrid parents and varieties were selected for soil salinity tolerance based on the pot culture experiments under induced salinity at ICRISAT-Patancheru. These selected lines were evaluated in two trials: (i) 21 varieties + 1 B-line + 2 R-lines + 1 hybrid + 5 susceptible controls and (ii) 33 hybrids + 22 parents (5 B-lines + 13 varieties + 4 restorers) were sown at Agriculture Research Station (ARS), Gangavathi, Karnataka, India under soil salinity – stress condition (10 dS m^{-1}) and at ICRISAT-Patancheru in pot culture. The trials at Gangavathi were vitiated due to poor plant stand resulting from heavy rains and shoot fly infestation. However, both trials were replanted. The results from both ARS, Gangavathi (field) and ICRISAT (pot culture) experiments are awaited. Twelve hybrid parents/varieties (ICSV112, GD65008, S35, JJ1041, ICSV 93046, ICSV 93034, CSV 15, NTJ 2, ICSR 170, ICSV 745, SPV 1022 and ICSB 406) were sent for testing in saline coastal areas by Central Rice Research Institute (CRRI), Cuttack, Orissa, India. These lines were found tolerant to salinity in pot-screening carried out at ICRISAT-Patancheru.

Seed of 15 salinity-tolerant entries (ICSV 93046, ICSV 745, SP 47513, SP 39262, SP 47529, S 35, ICSV 93048, ICSV 112, SP 47519, SP 39105, ICSR 93034, SP 39053, SP 40567, SP 47503 and SP 39007) was supplied to International Center for Biosaline Agriculture (ICBA), Dubai, for further screening under salinity stress in field conditions.

BVS Reddy, S Ramesh and V Vadez

Grain and fodder yield of salinity-tolerant improved populations of pearl millet in saline soils: Fourteen improved populations selected as salinity tolerant by ICBA were re-evaluated at 10 dS m^{-1} salinity level at Gangavathi, Karnataka, India, during 2005-rainy season (second season) for grain and dry fodder yield along with two OPVs (ICTP 8203 and Raj 171) and a pollinator (ICMP 451) as controls. HHVBC-tall had 1922 kg ha^{-1} of grain yield, which was significantly higher than two popular controls (90% more than Raj 171 and 122% more than ICTP 8203) with almost similar maturity (56 days to flower) compared to controls (54 days to flower in ICTP 8203 and 59 in Raj 171) during 2005. It also produced 48% more dry fodder yield than the high fodder yielding control Raj 171 (2583 kg ha^{-1}). Two more populations (Sudan Population III and Dauro genepool) were identified for higher grain yield (1508 and 1347 kg ha^{-1}) and fodder yield (2833 and 4000 kg ha^{-1}). Leonis genepool produced significantly higher fodder yield (132% more) than Raj 171. Based on the two seasons data, the promising dual-purpose populations adapted to saline conditions were HHVBC-tall, Sudan Population III, Dauro genepool, and the control Raj 171 (with 1452 to 1996 kg ha^{-1} of grain yield and 4009 to 4941 kg ha^{-1} of fodder yield). Another population, Leonis genepool (6117 kg ha^{-1}) was identified only for fodder purpose.

Evaluation of pearl millet hybrid parents for grain and fodder yield in saline soils: Nine salinity-tolerant and seven sensitive hybrid parents selected based on ICRISAT-Patancheru results and first year evaluation at Gangavathi were again evaluated at 10 dS m^{-1} salinity level at Gangavathi, Karnataka, India for grain and fodder yield during 2005 rainy season. Among the seed parents, the tolerant parent ICMB 95222 produced 130% more grain and 286% more fodder yield than the sensitive control ICMB 94111 (536 kg ha^{-1} grain and 2750 kg ha^{-1} of fodder yield) during 2005 rainy season. Other promising seed parents for grain yield were ICMB 01222 and 841B (74-79% more than ICMB 94111) and for fodder yield were 863B and ICMB 96333 (59-79% more than ICMB 94111). Among the restorer parents, the tolerant parents ICMP 451 and CZP 9621 had high grain (938 and 882 kg ha^{-1}) and fodder (6667 and 5000 kg ha^{-1}) yield compared to sensitive control H 77/833-2 (363 kg grain and 3667 kg fodder ha^{-1}). Based on the two seasons data, the seed parent ICMB 95222 was identified for high grain (1164 kg ha^{-1}) and fodder (6794 kg ha^{-1}) yield, ICMB 01222 only for high grain yield (1265 kg ha^{-1}), and ICMB 96333 and 863B only for fodder yield (about 3980 kg ha^{-1}). Among the restorer parents, ICMP 451 was identified for high grain (1311 kg ha^{-1}) and fodder (6721 kg ha^{-1}) yield and HTP 94/54 only for fodder yield (5478 kg ha^{-1}). These hybrid parents could be used in the production of high-yielding hybrids tolerant to salinity.

Inheritance pattern of salinity tolerance in pearl millet: Four hybrids produced from two salt-tolerant lines (ICMB 95333 and ICMP 451) and two sensitive lines (81B and ICMB 94111) along with the four parents and another high yielding hybrid identified previous years field trials were evaluated at ICRISAT-Patancheru in pot culture under saline and control conditions to determine the pattern of inheritance of salinity tolerance and to know whether salinity tolerance of one parent is enough to produce tolerant hybrids. Under saline conditions, flowering was delayed by 10 days compared to non-saline control condition (50 day to flower). Hybrids had significantly higher panicle weight than parents in both saline and control conditions and higher biomass weight only in saline condition. The two hybrids involving both tolerant parents had 10–27% higher panicle weight (26.9 and 30.9 g $plant^{-1}$), and the two hybrids with one tolerant parent had 7–9% higher panicle weight (25.9 and 26.4 g $plant^{-1}$) compared to the hybrid with two sensitive parents (24.3 g $plant^{-1}$) under saline conditions. The same conclusion did not hold good with respect to biomass yield. But for the parents 81B (sensitive parent) and ICMP 452 (tolerant parent), which were affected by downy mildew, the salinity susceptible index (calculated on the basis of panicle weight) was generally lower in the hybrids with at least one tolerant parent (0.91–0.98), and in tolerant parent (0.99) compared to the hybrid with two sensitive parents (1.01) and sensitive parent (1.21). This limited set of data indicated the dominance of salinity tolerance over sensitivity and the need of at least one tolerant parent for production of salinity-tolerant high-yielding hybrid.

Comparison of HHVBC sub-selections of pearl millet for salinity tolerance: Salinity tolerance of HHVBC-tall has been confirmed at both ICBA-Dubai and ICRISAT-Patancheru. Based on one-year field trial, it has also been found very productive in saline field conditions at Gangavathi, Karnataka, India. HHVDBC is d2 dwarf version of HHVBC-tall. Four HHVDBC-derived sub-populations earlier selected for different heights and panicle thickness (dwarf, medium height, tall and thick panicle) along with C_0 cycle bulk of HHVBC-tall were evaluated in pot culture at ICRISAT and in field condition at Gangavathi to assess the differences, if any, among these sub-populations for salinity tolerance, measured in terms of both grain and fodder yield. Significant differences were observed among HHVDBC-derived sub-populations for grain and fodder yield at Gangavathi, and for biomass yield and panicle weight in saline and control conditions of pot experiments at ICRISAT. HHVBC-tall C_1 produced 33% higher grain (2308 kg ha^{-1}) and 114% higher dry fodder yield (6250 kg ha^{-1}) than the HHVBC-tall C_0 (1734 kg ha^{-1} grain and 2917 kg ha^{-1} fodder). Other promising sub-population identified for both grain and dry fodder yield was HHVDBC-thick (31% more grain and 85% more fodder than HHVBC-tall C_0), and for only fodder yield was HHVDBC-medium height (6250 kg ha^{-1}). In pot culture experiments under saline conditions, all the sub-populations produced significantly higher panicle weight compared to sensitive control (13.1g $plant^{-1}$). Amongst the sub-populations, HHVBC-tall C_0 and HHVBC-tall C_1 produced higher biomass both in saline (37.2 and 32.5 g $plant^{-1}$) and control (73.9 and 76.5 g $plant^{-1}$) conditions, and higher panicle weight under saline conditions (28.4 and 26.7 g $plant^{-1}$) compared to other populations, which had 51.0–67.9 and 25.9–31.3 g $plant^{-1}$ of biomass in control and saline conditions respectively and 21.4–23.8 g $plant^{-1}$ panicle weight in saline conditions. Based on these results, HHVBC-tall C_1 was identified for higher grain and fodder yield under saline conditions.

Grain Fe and Zn density in improved populations of pearl millet: The trial consisting of 69 improved populations from diverse origin [ICRISAT-Asia (33), ICRISAT-WCA (32) and ICRISAT-ESA (4)] conducted during 2004 rainy season was repeated during 2005 summer season. As indicated by the trial means, the grain Fe was 12% higher during the rainy season (58 ppm). On the contrary, grain Zn was 11% lower during summer season (40 ppm). Significant genetic variability with approximate two-fold variation for both grain Fe (42–80 ppm) and Zn (27–50 ppm) was observed. Significant season × genotype interaction was noticed. The correlation between Fe and Zn was highly significant ($r = 0.76$; $P < 0.01$). Based on the mean data of two seasons, 7

populations having high Fe (>70 ppm Fe) and 8 populations having high Zn (>45 ppm Zn) were identified, and 6 of these common populations ICTP 8203, GGP Bulk (C_0), CGP, IAC-ISC-TCP-4, PVGGT-5 and Ugandi were identified for both high grain Fe and Zn density.

Intra-population variability for grain Fe and Zn density in pearl millet: One released variety each from Africa (GB 8735) and India (AIMP 92901) were identified for high grain Fe and Zn based on the two seasons evaluation of 30 populations (in the 120 entries trial) during 2004, and 4 populations (CGP, GGP bulk, ICTP 8203 and PVGGP 6) were identified based on evaluation of 69 populations during 2004 rainy season, for studying intra-population variability and deriving progenies with still higher Fe and Zn than the parental populations. Significant variability for both Fe and Zn was evident among the 64 S_3 progenies of AIMP 92901 and 68 S_2 progenies of GB 8735 (previously derived). The range in Fe and Zn density in AIMP 92901 and GB 8735 progenies showed approximately two and half fold variation for Fe (35–104 ppm in AIMP 92901 and 40–105 ppm in GB 8735) and two-fold variation for Zn (29–68 ppm in AIMP 92901 and 29-60 ppm in GB 8735) (Figs. 1.1 and 1.2). Significant positive correlations ($P < 0.01$) between Fe and Zn were observed in AIMP 92901 ($r = 0.68$) and in GB 8735 ($r = 0.76$). Based on the data, top nine progenies of both AIMP 92901 and GB 8735 were selected separately for Fe and Zn for random mating during 2006 summer to initiate recurrent selection for high Fe and high Zn. The top three progenies for both micronutrients from both the populations will be crossed to four B-lines (counterpart of designated A-lines) with high Fe and Zn to produce hybrids with still higher Fe and Zn. Field testing of 50 S_1 progenies derived from the remaining four populations is undergoing to study the intra-population variability for grain Fe and Zn.

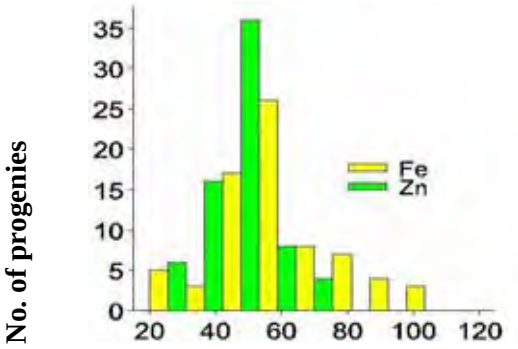

Figure 1.1. Frequency distribution of AIMP 92901 (S_3) progenies of pearl millet for grain Fe and Zn density, rainy season 2005.

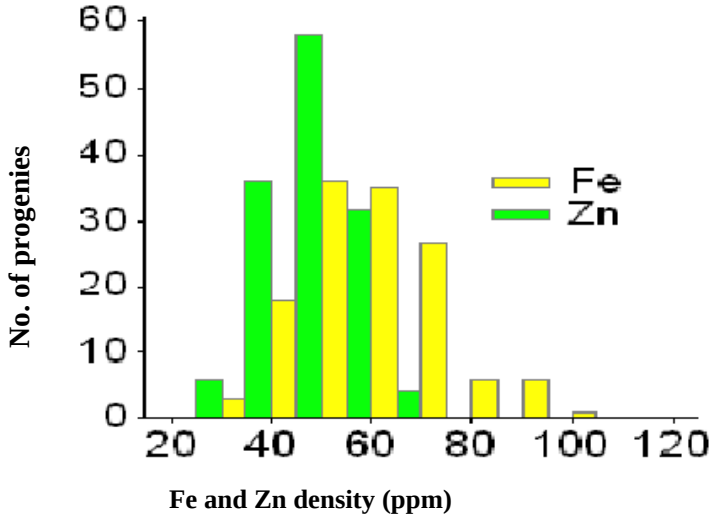

Figure 1.2. Frequency distribution of GB 8735 (S_2) progenies of pearl millet for grain Fe and Zn density, rainy season 2005

Assessment of stability of Fe and Zn density in selected genotypes of pearl millet: Thirty lines that included 14 high (51–76 ppm Fe and 48–65 ppm Zn), 8 medium (41–45 ppm Fe and 41–46 ppm Zn), 6 low lines (30–38 ppm Fe and 24–34 ppm Zn) and 2 controls selected from the two seasons screening trial during 2004 were evaluated during 2005 summer and the same trial was repeated in rainy season 2005 for assessment of stability. The analysis of sibbed seeds of these 30 entries from summer season at National Institute of Nutrition, Hyderabad, indicated stability of the genotypes for grain Fe and Zn. The genotypes with high Fe and Zn remained high in the summer trials and majority of low remained low.

Inheritance of grain Fe and Zn density in pearl millet: From the two seasons screening of 120 entries during 2004, four lines each with high (Fe 60–75 ppm and Zn 56–65 ppm), medium (Fe 42–45 ppm and Zn 41–47 ppm) and low (Fe 30–36 ppm and Zn 24–34 ppm) density were identified and 12 × 12 full diallel crosses (including reciprocals) were generated during 2005 summer. First season evaluation of the diallel set of crosses was completed during 2005 rainy season. The sibbed seeds of three replicates of 132 F_1s and 12 parents were sent to NIN for laboratory analysis.

Compare the cost effectiveness of alternative Fe and Zn assessment protocols in pearl millet:
A three-pronged approach has been followed to reduce the cost of grain Fe and Zn analysis; (i) the reduction of the cost involved in the grain sample production, (ii) the use of staining technique for quicker and cheaper analysis and (iii) the management of analytical cost of Fe and Zn through estimation of inter-laboratory correlations.

Comparison of selfed, sibbed and open-pollinated seed sources for Fe and Zn density: Since production of selfed seed for micronutrient analysis is much more cost-effective than producing sibbed seed, Fe and Zn density in these two seed sources were estimated to examine if there is any correlation between the two seed sources, and hence a possible guide for more cost-effective seed production in the future. Open- pollinated (OP), sibbed and selfed grain samples of 30 entries (15 inbred lines and 15 populations with a wide range of Fe and Zn levels) produced from the trial consisting of 120 entries, conducted during the 2004 summer season were analyzed for Fe and Zn density. Results showed highly significant correlation ($P < 0.01$) between selfed and sibbed seed both for Fe ($r = 0.68$) and Zn ($r = 0.78$). The correlation of OP seed with sibbed and selfed seed were 0.43 and 0.44 ($P < 0.05$) for Fe and 0.50 and 0.53 for Zn ($P < 0.01$). The initial results indicated that the grain samples obtained through selfing could be used in the grain Fe and Zn analysis. The three types of grain samples of the same 30 entries produced during the 2004 rainy season will be analyzed to confirm these results. Also, selfed, sibbed and open-pollinated grain samples of 30 populations produced during 2004 rainy season and 2005-summer season (from the trial consisting of 69 populations) will be analyzed to determine the relationship between three types of grain samples for Fe and Zn.

A rapid staining method of screening for grain Fe density: A simple, rapid and economic method of identification of high grain Fe genotypes was developed using Perls' Prussian stain. This protocol has great utility in screening large number of germplasm accessions or improved breeding lines within shortest possible time and with minimum cost (the chemical cost of analysis of each sample is $0.01), and helps in discarding the low-density lines.

Inter-laboratory correlations of grain Fe and Zn density: A randomly selected sample of 12 genotypes with a wide range of Fe and Zn levels as revealed from the lab analysis at NIN, Hyderabad were also analyzed at ICRISAT-Patancheru, India and at the Waite Analytical laboratory, Adelaide University, Australia. The correlations for Fe and Zn density among the laboratories were highly significant ($r = 0.77$ to 0.98; $P < 0.01$), indicating dependability of the results from any of these three laboratories. Since, ICRISAT-Patancheru laboratory charges for the analysis of Fe and Zn are very low (US$1.4) compared to NIN (US$20) and Waite (US$7), whenever there is large number of samples, initial screening will be done at ICRISAT-Patancheru and those with high Fe and Zn density will be analyzed in other laboratories for confirmation of the ICRISAT-laboratory results.

KN Rai, VN Kulkarni, V Vadez, P Pathak and TJ Rego

Milestone: High-yielding and DM resistant trait-specific (diverse maturity, large grains, large panicles, high tillering) advanced breeding lines of pearl millet developed (2007)

Development of hybrid parents adapted to arid Rajasthan, India: Development of high-yielding hybrids adapted to arid Rajasthan is a great challenge that could be achieved through the development of hybrid parents adapted to this region. Breeding early-maturing seed parents with resistance to downy mildew (DM) against the most virulent Jodhpur pathotype is a strategy that is expected to produce desired results. Two approaches have

been followed to develop potential seed parents; (i) exploitation of ICRISAT-CAZRI B-composite (ICCZBC) and (ii) pedigree breeding in promising B× B crosses. Potential restorer progenies development is based on (i) the exploitation of Mandor Restorer Composite (MRC) and (ii) improved populations adapted to arid Rajasthan region.

Development of seed parent progenies adapted to western Rajasthan, India: Development of seed parent progenies was set in motion two years ago through the constitution of ICCZBC based on 91 crosses selected from the diallel set of 20 parents previously selected for Rajasthan adaptation. About 1100 S_1-S_2 progenies derived from ICCZBC were evaluated against Jodhpur pathotype under high disease pressure in greenhouse condition (>90% DM incidence in susceptible controls 843B and ICMP 451), of which 45% were highly resistant (0-10% DM incidence). Out of 200 DM resistant S_2 progenies (derived from previous evaluation at Patancheru) evaluated in the rainy season, 77 were selected, and 50 of these flowered in 41-50 days (843B flowering in 41 days and 81B in 55 days). Of the 174 progenies evaluated in the summer drought nursery, 78 were selected based the agronomic potential, of which 50 flowered in 41-50 days (843B flowered in 43 days and 842B in 50 days).

Seed parent progeny development targeted to arid Rajasthan (India) also got an early start through pedigree breeding in 18 promising B x B crosses (mostly from the diallel set used to constitute ICCZBC) and now we are in the process of identifying advanced generation DM resistant progenies. In this context, 1316 F_5 progenies derived from B × B crosses were evaluated for DM resistance against Jodhpur pathotype under high disease pressure in greenhouse condition (>90% DM incidence in susceptible controls 843A and ICMP 451), of which 85% were found to be resistant (0–20% DM incidence). Based on DM resistance and visually assessed agronomic potential, 708 progenies were selected and evaluated during the rainy season, of which 263 were selected. Of these, about 220 flowered in 41–50 days (843B flowered in 39 days, 842B in 45 days and 81B in 51 days). Additionally, 300 F_5 progenies selected at CAZRI, Jodhpur (India) were also screened against Jodhpur pathotype, of which 82% had 0–20% DM incidence. Of the 60 S_2 progenies evaluated in the summer drought nursery, 30 were selected based on the visually assessed agronomic potential, of which 17 flowered in 41-50 days (843B flowered in 43 days, ICMB 95111 in 49 days and 842B in 50 days).

Early-maturing potential B-line trial: Breeding early-maturing hybrids has been largely dependent on male-sterile line 843A, which is the earliest-maturing, and it has good-tillering, large grain size and good combining ability, but is susceptible to DM. In breeding early-maturing seed parents, 843B has been used extensively. Genetic diversification for earliness is underway through exploitation of Extra-Early-Dwarf B-Composite (EEDBC). Out of the 22 EEDBC-derived early-flowering B-lines (40-48 days) evaluated, 14 progenies flowered in 41–46 days (ICMB 95444 flowered in 44 days and 843B in 42 days). Five of them were selected based on visual assessment of agronomic potential, of which 3 were resistant to both Jalna and Durgapura pathotypes with less than 15% DM incidence under high disease pressure in the greenhouse condition (80–91% DM incidence in susceptible controls).

Development of restorers adapted to western Rajasthan, India: Deriving progenies from the populations adapted to western Rajasthan has been the strategy followed to develop restorers for this region. In this respect, high-yielding Rajasthan Composite Bajra-2 (RCB 2), early-maturing Mandor Restorer Composite (MRC) and ICMS 7704 have been exploited. More than 100 advanced generation progenies (S_6–S_8) from MRC/RCB 2/ICMS 7704, and an additional 28 progenies from MRC selected from the summer drought nursery were evaluated during rainy season, of which 60 were selected. MRC-derived progenies were affected by water-logging in early stages of crop growth, resulting in delayed flowering as indicated by the late flowering of the controls. Of the 53 progenies evaluated 20 were selected, of which 16 flowered in 46–55 days. Twenty-five early-maturing MRC progenies with maturity similar to that of H 77/833-2 (45 days to 50% flower) were selected to further evaluate under post-flowering drought condition. Of the 14 selected progenies from RCB 2/ICMS 7704, 8 flowered in 40–50 days (ICMR 356 flowered in 46 days and ICMP 451 in 49 days).

MRC progeny trial: Mandor Restorer Composite (MRC) has been exploited to derive restorers suitable for producing early-maturing and high-tillering hybrids similar to HHB 67. Twenty-eight advanced generation (S_7–S_8) progenies derived from early-maturing MRC were evaluated at Bawal, Jamnagar and Patancheru, India in replicated trials. At Bawal, 9 progenies were selected based on the visual assessment of the agronomic potential and 5 of these were also selected at Patancheru. Additionally, 7 more progenies were selected at Patancheru. Also, 5 progenies with superior agronomic potential than the controls were selected at Jamnagar. Eight of the selected progenies at Bawal flowered in 44–51 days (H 77/833-2 flowering in 45 days and RIB 3135-18 in 50 days) and 9 of them selected at Patancheru flowered in 44–49 days (45 days for both the controls). All of the selected progenies at Jamnagar flowered late (2–13 days late) compared to the control H 77/833-2 (47 days).

Development of trait-specific breeding lines: Grouping of advanced generation progenies based on visually distinguishable desirable phenotypic characters (target traits) [such as high-tillering, long panicle, thick panicle, large seed size, compact panicle, etc.] enhances the effectiveness of visual selection by making the within-group comparison of lines easier for specific traits. During such trait-specific grouping, variability for non-target trait is also maintained to develop lines with multiple desirable traits without compromising on DM resistance. In addition to the trait-specific grouping, advanced generation progenies are also grouped into those resembling specific plant types of commonly used A-/R-lines such as 81A type, ICMA 89111 type, 843A type, ICMR 356 type, etc., which are easily recognized by breeders and the seed producers.

Trait-specific seed parent progenies: About 2200 advanced generation (F_5/S_5 and beyond) seed parent progenies belonging to 9 different trait-specific groups [ranging from 27 (forage type progenies) to 792 (early-generation long panicle progenies)] were evaluated. About 960 progenies (ranging from 25 to 244 in various groups) were selected based on the visual assessment of agronomic potential and target trait of respective groups for further testing. Early-flowering type was the newly constituted group with 65 diverse progenies evaluated, and 43 selected based on the visual assessment of agronomic potential. Of these, 72% flowered in 45 days and earlier (843B flowering in 39 days and 841B in 46 days). A large proportion of the selected progenies (65–82%) from large seed, high-tillering, very dwarf, compact panicle, erect-type and forage-type groups flowered in 46–55 days (ICMB 89111 flowering in 46 days and 81B in 51–53 days). Proportions of such progenies were less in long panicle and thick-panicle groups, indicating the need for introducing earliness in these progenies. The selected long-panicle progenies had panicle length ranging from 30 to 60 cm. Similarly the selected thick-panicle progenies had panicle diameter of 40–50 mm.

About 290 progenies typical to plant type of some of the most widely used and diverse hybrid parents such as 81A, 843A, ICMA 88004, ICMA 89111 and ICMR 356 were evaluated, and 254 were selected based on the visual assessment of agronomic potential and plant type. Of these, 81% belonged to early to mid-late group, flowering in 41–50 days (81B flowering in 53 days, ICMB 89111 in 45 days, 843B in 39 days). In addition to these, nearly 1700 preliminary and elite potential progenies were evaluated to select 667 progenies of various maturity classes based on the visual assessment of grain yield potential. Of these, 4% were in early group, flowering in 45 days; 35% were in mid-early group, flowering in 46-50 days; and 42% were in mid-late group, flowering in 51–55 days (81B flowering in 51 days, ICMB 89111 in 45 days and 843B in 40 days).

Screening for DM resistance in greenhouse condition under high disease pressure (83–100% disease incidence in susceptible controls) against Durgapura pathotype showed 20–35% progenies having no DM incidence in very dwarf, compact panicle and erect groups, and 1–10% DM incidence in 10–20% of the other progenies. About 40% of the progenies averaged over 8 specific plant type groups were free from DM and about 20% progenies had less (1–10%) incidence, indicating large proportion of progenies being DM resistant. Amongst the early-generation long panicle progenies and advanced generation B × B progenies, which are yet to be classified into different trait-specific/plant type groups, about 40% were DM resistant (0–10% incidence).

Thick panicle B-line trial: High Head Volume Dwarf B-Composite (HHVDBC) is a good source for deriving lines with thick panicles, which is an important yield component. Twenty-three advanced generation (S_5) progenies derived from HHVDBC were evaluated and nine thick panicle progenies having more than 40 mm panicle diameter with higher grain yield potential (based on the agronomic score) compared to the control ICMB 01222 (32 mm diameter) were selected. Of these, three progenies flowered in 51–53 days (ICMB 01222 flowered in 50 days). Four out of nine selected progenies had at least 30 cm panicle length.

Trait-specific restorer parent progenies: Similar to the trait-specific grouping of seed parent progenies, the grouping of restorer progenies based on different target traits and specific plant type is underway. About 690 advanced generation progenies (S_8 and beyond) from 9 different trait-specific groups (ranging from 16 in the large seeded group to 141 in the early-maturing group) were evaluated. Based on the visual assessment of yield potential and agronomic traits, 162 progenies from only 6 trait-specific groups were selected as the remaining groups were affected by water-logging during the early stage of the crop growth, making their evaluation ineffective. Flowering in these groups was delayed by 7–10 days as indicated by ICMR 356 flowering in 53 days (flowers in 45–46 days under normal conditions). About 60% of the progenies (46–55 days to flower) in these trait-specific groups were of similar maturity as ICMR 356. In the process of updating the trait-specific groups, about 90 progenies were added in dual-purpose group and 40 in the stay-green group. An extra-early group with 36 progenies was constituted. Additionally, 49 early-maturing and genetically diverse lines flowering in 38–53 days were selected to initiate intercrossing among them to constitute an Extra-early restorer composite.

IPC 804 is a good restorer line of short plant height, sturdy and erect plant type, and long panicles. Thus we constituted IPC 804 plant type group with 189 progenies. Additionally, we also constituted IPC 804–medium height group with 115 progenies. These two groups along with 42 ICMR 356 type progenies were evaluated and 96 were selected based on the visual assessment of agronomic potential and plant type. About 62% of the selected progenies flowered in 46–60 days (ICMR 356 flowering in 53 days and IPC 804 in 55 days). In the process of updating specific plant type groups, 29 progenies were added in ICMR 356 type. In addition to these, about 154 progenies with very high agronomic performance from diverse groups of material were selected for further evaluation.

Genetic diversification of restorer lines

Exploitation of OPVs for restorer line development: High-yielding improved populations adapted to different agro-ecological conditions serve as primary sources for deriving restorer progenies. These diverse populations recognized for different traits help in selection of progenies of various morpho-physiological characters such as earliness, height, tillering, panicle and seed traits and dual-purpose traits. We evaluated early and advanced generation progenies obtained from different populations for their agronomic potential.

About 800 population progenies (S_1–S_4) derived from 12 diverse populations were evaluated and 286 were selected based on the visually-assessed agronomic potential. Of these, 12% progenies from JBV 2 and JBV 3 flowered in 40–50 days (ICMR 451 flowering in 48 days and HTP 94/54 in 54 days) and 13% of the selected from other populations flowered in 46–60 days.

Restorer progenies derived from population intercrosses: A total of 216 (F_3–F_5) progenies with high agronomic score (selected out from 600 progenies selected during 2005-summer) derived from MRC × long panicle, Jakharana × ESRC crosses and from population intercrosses were evaluated and 93 were selected to generate 247 progenies. Forty-six of the selected progenies flowered in 46–50 days (ICMR 356 flowering in 47 days and ICMP 451 in 48 days). Jakharana × ESRC progenies (154) were evaluated against Durgapura pathotype under high disease pressure in greenhouse condition (90% in ICMP451 and 100% in H 77/833-2), of which 53% of the progenies had 0–10% DM incidence**.**

Pedigree breeding of restorers of A_5 CMS system: Restorer development for A_5 CMS system is unique as A_5 restorers are rare in the germplasm. Therefore, a different approach involving incorporation of A_5 restorer genes(s) in diverse genetic backgrounds was followed to breed restorers of this CMS system. During 2004 rainy season, we evaluated 190 hybrids derived from crossing 8 elite male-fertile inbreds in A_5 cytoplasmic background as female parents with 31 high-yielding population progenies as male parents. Based on grain yield potential, agronomic traits and fertility restoration, 14 hybrids (involving 7 female parents and 10 male parents) were selected and their F_2 populations were produced to initiate pedigree selection for A_5 restorer line during the previous year. From these 14 F_2 populations evaluated during the 2005 summer season, 8 populations were selected based on the visual assessment of the promising segregants with respect to yield potential, agronomic traits and male-fertility restoration. We selected 117 plants from these 8 populations, which were advanced to F_3 generation. Also, 20 progenies of diverse pedigree from restorer breeding program were crossed with the individual plants of all 14 F_2s to diversify the genetic base of A_5 restorers program. More than 90 crosses made on the selected 8 F_2 populations were harvested for further evaluation/selection.

Potential restorer progenies: This group consisted of 395 progenies (S_4-S_6 and beyond) that were selected during summer and rainy season 2004 from different groups of breeding materials. These were evaluated during 2005 rainy season, of which 103 were selected based on the visually assessed grain yield potential and other agronomic traits to generate about 170 progenies. Of the selected progenies, 38 flowered in 46–55 days (ICMR 356 flowering in 55 days and ICMP 451 in 54 days). All 395 progenies were evaluated against Durgapura and Jalna pathotypes under high disease pressure in greenhouse condition (>95% in incidence in controls). Of these, 24% progenies were resistant (0–10% DM incidence) to Durgapura pathotype and 33% to Jalna pathotype.

Elite restorer progenies: About 470 progenies that included 288 diverse progenies (S_4–S_6 beyond) with high agronomic score selected from different groups of materials during 2005 summer season and the remnant seed source of 170 progenies were further evaluated during the rainy season. Since a major part of this block was affected by water logging, only 56 progenies were selected, and 18 of these flowered in 46-55 days (ICMR 356 flowering in 56 days and ICMP 451 in 54 days). The 288 progenies selected in 2005 summer were evaluated against Durgapura and Jalna pathotypes under high disease pressure in greenhouse condition (90–100% in

susceptible controls). Of these, 32% of the progenies were resistant (0–10% DM incidence) to Durgapura pathotype and 37% to Jalna pathoype.

RCB2/ICMS 7704 Progenies trial: Exploitation of RCB 2 and ICMS 7704 is expected to produce restorer lines of dual-purpose hybrids. Nine progenies derived each from RCB 2 and ICMS 7704 were evaluated at Jamnagar and Patancheru in replicated trials. At Jamnagar, all the 5 progenies identified for good agronomic potential based on the visual assessment were from RCB 2, of which 4 flowered in 44–50 days (ICMP 451 flowering in 50 days and ICMR 356 in 44 days). At Patancheru, amongst 7 progenies (3 derived from RCB 2 and 4 from ICMS 7704) selected based on the agronomic score, 4 flowered in 46–52 days (ICMP 451 flowering in 49 days and ICMR 356 in 46 days). A progeny with the highest agronomic score (ICMS 7704 S1-103-2-3-2-2-2-4-B) derived from ICMS 7704 that flowered in 56 days having good panicle length (29 cm) was the best dual-purpose progeny. It was also identified for conversion into A-line of forage hybrids.

Dual-purpose restorer progenies trial: To expand the genetic base of dual-purpose restorers exploitation of a wide range of populations through inbreeding and selection has been one of the strategies in restorer line development. Twenty-eight previously selected advanced generation progenies derived from 13 different populations were evaluated at Patancheru, Hisar and Jamnagar. At Hisar, 6 progenies (with 4 score, where 1 = poor; 5 = best) were selected based on their visually assessed grain yield potential and other agronomic traits compared to controls ICMP 451 and HTP 94/54 (both with 3 score). Of these, 4 were also selected at Jamnagar. Additionally, 12 progenies (with 3–5 score) having superior scores than controls (both 2) were also selected at Jamnagar. At Patancheru, 13 progenies that had agronomic scores similar to or better than the controls (both 4) were selected. Of these, 3 were best with 5 score. Flowering behavior of the progenies at Patancheru and Jamnagar were similar compared to Hisar (11–14 days later). Selected progenies at Hisar flowered in 57–71 days (63 days for ICMP 451 and 71 for HTP 94/54) and at Jamnagar these flowered in 43–57 days (49 days for ICMP 451 and 56 for HTP 94/54). Amongst the 3 best progenies identified at Patancheru, 2 flowered in 52 days (49 days for ICMP 451 and 52 for HTP 94/54) and third was late by a week. Of the remaining 10 progenies, eight flowered in 46–53 days. Almost all the selected progenies in all the locations had ≥150 cm plant height.

KN Rai, VN Kulkarni and RP Thakur

Downy mildew resistance in advanced generation progenies from selected B × B crosses targeted for western Rajasthan, India

Maintaining high levels of downy mildew resistance in breeding lines is the key to the development of stable A-, B- and R-lines and their hybrids. Keeping this in view we screened large number of advanced generation breeding lines targeted for Rajasthan, India against two major pathotypes, Durgapura (Sg 212) and Jodhpur (Sg 139) in the greenhouse.

- A total of 1228 advanced breeding lines including 131 $F_{5,}$ 375 F_6 progenies from B × B crosses, and 722 trait-specific progenies were screened against Durgapura pathotype in an unreplicated trial (one pot with 35–40 seedlings per line). Of these, 18% lines were disease free and 19% showed high levels of resistance (≤10% incidence).
- Similarly, a total of 2734 advanced breeding lines including 1616 F_5 progenies from B × B diallel crosses, and 440 S_1 and 678 S_2 progenies from ICCZ BC C were screened against Jodhpur pathotype. Of these, 37% were disease free, and 23% showed high levels of resistance (≤10% incidence).
- A total of 352 progenies from high head volume dwarf composite (HHVBC) were screened against Jalna (Sg 150) and 359 against Durgapura (Sg 212) pathotypes. Of these, 11% were disease free and 6% showed high levels of resistance (≤10% incidence) to Jalna pathotype; 15% lines were disease free and 8% were resistant to Durgapura pathotype.

RP Thakur and KN Rai

Hybrid parents adapted to the arid zone

Combining ability of Mandor Restorer Composite (MRC) progenies under arid zone conditions: Forty-one lines from MRC (selected based on DM resistance and agronomic value at ICRISAT-Patancheru) were testcrossed to five seed parents and the resulting hybrids were evaluated for general combining ability (GCA) at the Rajasthan Agricultural University station at Nagaur, Rajasthan, India. The mean grain yield of the trial, under severe drought stress, was only 341 kg ha^{-1}, but the differences among MRC line for the mean hybrid yields was highly significant (range of 201 to 522 kg ha^{-1}). The best of the control restorers (arid zone landrace-derived topcross pollinators) had average hybrid yields of 401 and 427 kg ha^{-1}). MRC lines with a positive and significant ($P < 0.05$) GCA were detected for all traits measured: percent productive panicles (2), biomass

(5), harvest index (11), grain yield (7), stover yield (5) and panicle harvest index (8). GCA for biomass was associated with GCA for both grain yield ($r = 0.64$, $P < 0.001$) and stover yield ($r = 0.93$, $P < 0.0001$), and GCAs for grain and stover yields were positively correlated ($r = 0.35$, $P < 0.10$). This suggests that we could identify lines from the MRC with positive GCA for both grain and stover yields, but that both are dependant upon a positive GCA for total biomass. The most useful finding is that despite the selection of the MRC lines under favorable conditions at Patancheru (rather than in the arid zone), it was possible to identify superior lines in a very drought stressed environment. This is likely due to the selection of restorers with adaptation to the arid zone as the parents of the original MRC. Combining ability of landrace-based restorer populations in arid zone conditions: We continued the evaluation of the GCA of the landrace-based restorer populations that we bred with the objective of making a sample of the best of arid zone landrace germplasm more readily available for the breeding of restorers for the arid zone. The eleven populations plus four controls were crossed to four arid zone adapted seed parents, and the resulting hybrids (plus four single-cross hybrid controls) were evaluated at the Rajasthan Agricultural University station at Nagaur, India. Trial mean grain yield, under severe drought stress, was 338 kg ha^{-1}; with a range in individual entry grain yields of 182 to 528 kg ha^{-1}. Two of the restorer populations tested had a significant ($P < 0.05$) positive GCA for grain yield (the Jakharana restorer population and the control inbred restorer RIB 3135-18) and three populations had a significant positive GCA for stover yield (Barmer, Jakharana, Jakharana × ESRC restorer populations). These same three populations, plus RIB 3135 had a significant GCA for total biomass productivity, which we consider to be the best measure of adaptation to severe stress conditions, and hence to the arid zone. The single-cross hybrid controls as a group had grain yields well above the trial average (405 vs. 338 kg ha^{-1}), but stover yields well below the trial average (1190 vs. 1410 kg ha^{-1}), indicating that the current conventional hybrids, selected mainly for grain yield, do not fully meet the needs of arid zone farmers for both grain and stover. This trial will be continued in order to sample a wider range of environments before recommending individual restorer populations for actual use in breeding programs.

GCA for biomass of B × B crosses in arid zone conditions: Earlier research suggested that a positive GCA for biomass productivity under arid zone environments is the most desirable characteristic in new seed parents for this zone, as only such seed parents can produce hybrids with simultaneous improvement in both grain and stover yield. We made a complete diallel among twelve B-lines thought to be adapted to the arid zone to identify maintainer germplasm with this ability. We selected 23 B × B F_1s from the diallel and testcrossed these (and two inbred A-line controls) to four arid zone restorers, to evaluate the general combining ability of the selected F_1s for biomass productivity. The resulting 100 testcrosses were evaluated at the Rajasthan Agricultural University station at Nagaur, India. Yields were relatively low because of the severe stress (trial mean grain and stover yields were 473 and 1910 kg ha^{-1}), but there were significant differences among B × B F_1s for most traits. Only one of the crosses had a significant ($P < 0.05$) positive GCA for biomass under these conditions. However, this did result in significant positive GCAs for both grain and stover yields. Interestingly, this was a cross between two B-lines which themselves had a positive GCA for biomass – ICMB 91444 and ICMB 93333, both of whom contain West African parentage. We will continue with the B × B cross evaluations to sample a wider range of environments, but will also start inbreeding the ICMB 91444 × ICMB 93333 cross to produce progenies for selection for GCA for biomass in the arid zone.

Selection of restorers from the Early Rajasthan population: The Early Rajasthan Population (ERP) was bred from 30 selected S_1 progenies from four early-maturing landraces from western Rajasthan. The base population has performed very well under stress and is the source of a released variety CZP 9802, as well as an early maturing restorer population for the arid zone. Because of the obvious value of the ERP for the arid zone, we initiated the process of deriving inbred lines from the restorer version of the population, as potential R-lines. In order to assure that the most valuable characteristic of the ERP – its adaptation to the arid zone – was retained, we conducted a replicated yield test of 192 ERP S_1 progenies at the Rajasthan Agricultural University station at Nagaur, India to select desirable progenies. Due to the severe stress, the mean grain yield of the trial was very low (246 kg ha^{-1}), but differences among progenies in grain yield (15 to 700 kg ha^{-1}) were highly significant. Importantly, there were also highly significant differences among progenies for key indicators of adaptation to severe stress – percent panicles with seed (28 to 100%) and panicle harvest index (29 to 78%) – as well as in time to flowering (41 to 62 days). Selection of progenies was based mainly on drought escape/tolerance traits rather than grain yield: percentage panicles with seed ≥90%, panicle harvest index ≥55% and flowering ≤52 days.

FR Bidinger

Stover productivity and quality of arid zone adapted seed parents: Because of the importance of animal products to the income of arid zone farmers, new cultivars of pearl millet for the arid zone should not only produce grain and stover yields superior to those of the landraces now grown, but the ruminant nutritional

quality of their stover should be at least equal to that of the current landraces. As not much is known about nutritional quality of either the traditional landraces or of potential alternative hybrid cultivars, we conducted an experiment to assess stover quality in a set of six typical landraces and their eighteen topcross hybrids made on three arid zone adapted seed parents. Over four environments in 2003 and 2004, the yields of the landrace-based topcross hybrids exceeded that of the landrace pollinators by an average of 17% in the case of grain yield and 7% in the case of stover yield. There was no difference in nitrogen content of the landrace and topcross hybrid stover, and small (1–2%) but significant differences in the *in vitro* organic matter digestibility and metabolizable energy, in favor of the topcross hybrids. Combining the differences in stover productivity and quality between the landraces and the topcross hybrids resulted in a 13–14% increase in stover digestible dry matter, metabolizable energy and nitrogen per ha. Under the typical dry season, restricted intake feeding system of the arid zone (restricted due to limited stover supplies) these differences should translate into economic increases in animal productivity. Therefore, there is no *a priori* reason to be concerned about negative effects of arid zone hybrids, made with adapted parental lines, on either stover productivity or quality.

FR Bidinger and M Blümmel

Milestone: Pigeonpea breeding lines with resistance to *Helicoverpa*, wilt and sterility mosaic developed (2006)

Screening for wilt and sterility mosaic (SM) resistance: A total of 46 advanced breeding lines, 391 F_1 hybrids, and 62 backcross populations were screened in the wilt and sterility mosaic sick plots. The disease incidence was very high. Among the inbred lines, 45 were found resistant to both wilt and sterility mosaic diseases. In each line, five plants were selfed to produce pure seed. In the backcross populations, hand pollinations were done on the resistant segregants to advance the generation. Among the hybrids, 105 were found resistant to both wilt and sterility mosaic diseases, while 47 were resistant to wilt and 95 were resistant to sterility mosaic disease.

KB Saxena

Screening for salinity tolerance: Besides various management options, the development of salinity-tolerant varieties is the best option for saline areas. A protocol has been standardized at ICRISAT-Patancheru to screen pigeonpea genotypes for salinity tolerance, and a concentration of 75 mM was found to be critical for screening. SCMR (SPAD chlorophyll meter reading) was positively associated with higher biomass under salinity and this trait could be used as an early indicator for salinity tolerance. The shoot biomass data were analyzed using the residual maximum likelihood method. Among the wild species two accessions (ICPW 87 and ICPW 94) from *C. scarabaeoides* were most tolerant. Of the cultivated pigeonpea (*C. cajan*) accessions, ICP 13991, 14974, 13997, and 11412 exhibited high level of tolerance and ICP 13625, 13996, 14175, 11414, and 11420 showed high susceptibility. Among the maintainers, ICPB 2051, 2030 and 2039 were found tolerant while ICPB 2032 was highly susceptible.

N Srivastava and Vincent Vadez

Screening pigeonpea accessions and breeding lines for broad-based resistance against sterility mosaic isolates: Thirty-eight pigeonpea breeding lines developed at ICRISAT using broad-based sterility mosaic disease (SMD) resistant variety, ICP 7035, as one of the parents, were evaluated on-station for resistance against SMD during 2003–05. All these lines are of short to medium duration maturity (100–160 days to mature) types. From the 38 breeding lines, 12 promising lines (ICP 14404, 16166, 11719, 16169, 14478, 16165, 14834, 11632, 95029, 14399, 16294 and 16293) were evaluated during 2005-06 in on-station trials at Bangalore, Bidar and Gulbarga, Karnataka, India, against SMV-P and SMV-B isolates, respectively, to assess the maturity and resistance to SMD. Test plants were grown in SMD sick plots to allow infection to take place at young stage (12–15 days old plants). Local cultivars ICP 8863 and TTB 7 were used as susceptible controls, and these showed >80% infection.

At Bidar and Gulbarga, all the 12 genotypes attained 80% maturity in 140 days and showed good resistance to PPSMV-P isolate endemic in this region. Seven of 12 accessions showed no infection. In the remaining genotypes, SMD incidence ranged between 7.5 and 33%. All the 12 genotypes were selected by the local scientists for further validation. The seven genotypes (ICP 16166, 11719, 16169, 16293, 14834, 11632 and 95029) that showed no infection will be evaluated in the coordinated trials and on-farm. Evaluation of these genotypes against PPSMV-B isolate at Bangalore revealed that all the 12 accessions were infected (30–90% incidence). Although all the genotypes were infected, most of the genotypes produced flowers. In particular, ICP 14834, 16165, 11719, 14478, 11632 and 14399 (incidence 30–40%) showed apparently normal flowering. Five accessions, ICP 11719, 14834, 11632, 14399, 14478 and 16165, performed well against both PPSMV-P and PPSMV-B isolates.

On-farm trials to promote SMD resistant varieties: About 300 on-farm trials were organized with three medium-duration pigeonpea varieties, ICP 7035 (broad-based SMD and wilt resistant), ICP 87051 and ICPL 96058 (resistant to PPSMV-P isolate and wilt for central India). ICP 7035 was released for farmer cultivation by ICRISAT and University of Agriculture Sciences, Bangalore, India during May 2005.

Ten on-farm trials were conducted in three talukas (Chincholi, Aland and Gulbarga) in Gulbarga district of Karnataka (1 acre for test genotype and 0.5 acre for local control). Test varieties were ICP 7035 (3 farmers) and ICPL 96058 (7 farmers) with local controls, ICP 8863 and Asha. Sowings were done in second week of July. Genotypes were found performing well. All these trials were conducted with inputs, for commercial scale production.

Two hundred on-farm trials of ICPL 96058 were conducted in 15 villages of Mahaboobnagar district in Andhra Pradesh, India. Sowings were done in third week of July. No local controls were included in these trials. These trials were managed as per the traditional farmers practice. All the farmers in these trials are smallholders. Seeds were treated to prevent wilt infection. IPM methods were followed for pest control.

Twenty-five on-farm trials of ICPL 96058, ICP 7035 and ICP 87051 were organized in eight districts (Medak, Ranga Reddy, Kammam, Adilabad, Nizamabad, Nellore, Nalgonda and Warangal of Andhra Pradesh, India). Three varieties were supplied to farmers. ICP 8863 and other local varieties were included as controls.

Thirty-two frontline demonstrations of ICP 7035 were organized in four districts of southern Karnataka. These demonstrations were conducted in association with the State Extension Education Unit for the promotion of variety in diverse regions. Forty-eight on-farm trials of ICP 7035 were conducted in the SMD epidemic areas in three districts of Karnataka.

All the farmer-managed trials were conducted as per the native farmer methods. Seed was provided to the farmers. During the course of the trials, visits were made to monitor the farms, for the evaluation of performance, to obtain farmers opinion and to monitor SMD incidence in the region. All trial locations received good rainfall and varieties performed well. No SMD incidence was noticed on disease resistant varieties.

Survey for SMD incidence: In August 2005, a roving survey for SMD incidence was conducted to monitor 4–6 weeks old pigeonpea crop (most susceptible stage) in Gulbarga and Bidar districts (SMD hotspots) of Karnataka, India. Twenty-nine villages in four talukas (Aland, Gulbarga, Jewargi, Chittapur) of Gulbarga district; and 30 villages in five talukas (Basavakalyan, Bhalki, Bidar, Aurad, Humnabad) of Bidar district were surveyed. Most fields were free from SMD incidence (based on visual symptoms) although farmer-grown cultivars in these regions are highly susceptible to SMD. In few fields, especially in Bidar, around 1% SMD incidence was observed. Mite population was non-existent on the infected plants. Therefore, those infected plants may not act as source for secondary spread. Farmers were asked to remove the affected plants. Generally, frequent rains suppress multiplication of eriophyid mite vector and this restricts disease spread.

Training to farmers and field days: Training courses were conducted for farmers on IPM to manage SMD, wilt and pod borer. These included preparation of plant-based extracts for pest control and training in good agriculture practices and means to enhance pigeonpea productivity. Emphasis on management of SMD and wilt was through cultivation of resistant genotypes; and for pod borer management, a combination of traditional methods and judicious application of chemical sprays. Training courses were organized as one-day events, at Agriculture Research Stations, NGO training centers, and on-farm during field visits. In these events, farmers were given lectures in local language. We also demonstrated the affects of SMD on pigeonpea and performance of resistant varieties. Farmers selected for seed village program were given training in pigeonpea crop management to ensure quality seed production. Farmer field days were organized to demonstrate on-farm performance of pigeonpea varieties to the farmers. All the farmers training programs and field days were organized in collaboration with NGOs and State Extension Education Units. Information bulletins were prepared in local language and supplied to the farmers in such events.

Seed village system: Seed villages were established at 12 locations for the multiplication of ICPL 96058 and ICP 7035. These were established in association with local NGOs and Extension Education Units in Andhra Pradesh and Karnataka states in India. In this, a network of designated farmers multiplies the pigeonpea genotypes for seed purpose. Standard crop management practices were followed, external inputs such as fertilizer, one time irrigation was given at flowering, and isolation distance was maintained. The seed produced was collected and sold on par with the market price to the local farmers. The money generated will be used (as revolving fund) to continue the seed production next year. This system ensures timely availability of quality

pigeonpea seed to the farmers at right time. At present, each seed village has the capacity to produce 100 to 200 kg seed for each season. This target will be enhanced in due course. Assured supply of quality seed at right time is encouraging farmers to participate in this program.

P Lava Kumar, Farid Waliyar and KB Saxena

Sterility mosaic (SM) caused by pigeonpea sterility mosaic virus (PPSMV) and *fusarium* wilt (FW) caused by *Fusarium udum* are destructive diseases of pigeonpea with an estimated annual yield losses of >US $400 million. Deployment of host-plant resistance is the best way for economical management of these diseases. **Combined resistance screening for sterility mosaic and *fusarium* wilt:** Wilt sick plot for evaluation of breeding and germplasm lines for resistance to wilt was developed at ICRISAT-Patancheru. Chopped wilted pigeonpea plants are incorporated in the field every year to maintain threshold level of *F. udum*. Test material planted in wilt sick plot are inoculated using leaf staple technique at two-leaf stage with SM-infested leaves for SM infection. These techniques provide a simultaneous identification of wilt and SM resistant lines. Susceptible cultivars ICP 2376 for wilt (resistant to SM) and ICP 8863 for SM (resistant to wilt) were planted as indicator rows after every 10 test rows. Incidence of wilt and SM were recorded thrice at seedling, flowering and pod filling, and at maturity stages of the crop. Pigeonpea lines with <10% incidence to individual disease were considered as resistant.

Wilt and SM resistance in germplasm lines: Seventeen lines for wilt and SM resistance, six advanced lines for wilt and 26 advanced lines for SM were evaluated for combined resistance to FW and SM at ICRISAT-Patancheru. Good sources of combined resistance against wilt and SM diseases were observed. Nine lines (BDN 2010, BSMR 736, BSMR 846, KPL 43, KPL 44, MAL 3, ICP 9174, ICP 11438 and ICP 12290) were found resistant to both FW and SM diseases. Of the six advanced wilt promising lines, ICH 732 and ICP 6997 had resistant reaction (<10%) to both the diseases. Among the 26 SM promising lines, six lines (PT 1037, 2032, 2033, 2035, ICP 8090, ICP 8103) had combined resistance to both FW and SM. Additionally, seven lines were found asymptomatic and 10 were resistant (<10%) to SM.

Wilt and SM resistance in advanced breeding and inbred lines: Thirty advanced breeding and inbred lines were evaluated for combined resistance to FW and SM following standard evaluation techniques. Among the test lines, three lines ICPL 20097, ICPL 20098, ICPL 20099 were asymptomatic to both FW and SM. Twenty-three lines were found resistant to both diseases (<10% SM and wilt diseases).

Wilt and SM resistance in pigeonpea international nursery: Ten entries included in pigeonpea international nursery (PIN) were screened for combined resistance to both FW and SM following standard screening techniques. Six lines (ICPLs 87119, 96053, 96058, 96061, 99044 and 99050) were resistant to both FW and SM diseases

S Pande and KB Saxena

Wilt and SM resistance in *Helicoverpa* tolerant lines: Seventeen pigeonpea lines that were tolerant to pod borer, *Helicoverpa armigera*, were tested for their resistance to FW and SM diseases. ICP 7035 was asymptomatic to SM and ICPL 20042 was resistant to FW (<10% disease incidence). However, none of the *Helicoverpa* tolerant lines were resistant to both FW and SM diseases.

S Pande and HC Sharma

Disease Screening for Collaborators in pigeonpea

ICAR-ICRISAT collaborative research on pigeonpea wilt: Under ICAR-ICRISAT collaboration, 49 entries [32 entries in Advanced Variety Trial (AVT) and 17 entries in Initial Varietal Trial (IVT)] from Indian Institute of Pulses Research (IIPR), Kanpur, were received for evaluating for SM and wilt resistance under standard field evaluation techniques. Of the 17 IVT promising selections from IIPR Kanpur, two lines (MAL 23 and Bahar) were found asymptomatic, and MAL 13 was found resistant to both the diseases. Additionally four lines [ICP 7119 (HY 3C), BRG 3, MAL 18 and MAL 20] were resistant to SM alone. Among 32 AVT entries, only KPL 96053 was found resistant (<10%) to both FW and SM.

Acharya NG Ranga Agricultural University (ANGRAU)-ICRISAT collaboration for Wilt and SM resistance in advanced breeding lines: One hundred and four advanced breeding populations and progenies received from ANGRAU-Warangal, India were evaluated for SM and wilt resistance following standard field evaluation techniques. Of the 104 breeding lines, five lines WB 119, F29/7, F 5-98-1-21-2-3-1, F5-98-8-4-1-1-2

and F-98-2-6-2 were asymptomatic and 19 lines were resistant to SM. One line, F5-98-2-7-1-2-1 had combined resistance (<10%) to both SM and wilt.

S Pande

Confirmation of wilt resistance of advanced wilt promising germplasm lines using root dip and pot screening techniques under greenhouse environment

Root dip technique: Twenty-three pigeonpea lines, which were found resistant under field environment for three consecutive years, were tested for confirmation of their resistance to wilt under controlled environment (greenhouse). A susceptible control ICP 2376 was included for proper comparison. Seedlings of each entry were raised separately in polythene bags (8 × 20 cm) filled with sterilized sand in the greenhouse. Eight-day-old shake culture of the fungus, *Fusarium udum* (ICRISAT isolate) and eight-day-old seedlings were used for inoculation. Conidial concentration used for this study was 2×10^5 ml^{-1}. Roots of five seedlings of each entry were trimmed, dipped in the conidial inoculum for 30 seconds and transplanted in pre-irrigated 15 cm (diameter) plastic pots filled with sterilized sand and black soil mixture (4:1). Each pot represented one replication and three pots were kept for each entry. Uninoculated controls for each entry were maintained. The experiment was kept for 30 days for wilt symptoms. Wilt-susceptible cultivar ICP 2376 had 100% wilt within 12 days after inoculation (DAI). Of the 23 cultivars tested, five cultivars, ICP 12749, ICPL 94062, IPA 40, V 71A and V 102 were free from wilt.

Sick-pot technique: The above experiment was repeated using sick-pot technique in the greenhouse to correlate the efficiency of the technique for wilt screening under controlled environment. Pure culture of *Fusarium udum* (ICRISAT isolate) was multiplied on sand-pigeonpea meal medium (90 g pure sand, 10 g pigeonpea granules and about 20 ml distilled water in 250 ml conical flasks and autoclaved at 20 lb pressure for one hour) and incubated at 25°C for 15 days. Whole contents of the flask were taken out after 15 days of incubation and uniformly powdered. About 200 g of the inoculum was mixed in two kg soil (sterilized black soil + sand mixture in 1:1) filled in 15 cm plastic pots. All the pots were kept moist with light irrigation for two days to allow the fungus to settle. Thus, the soil in the pots was made wilt sick for evaluation of pigeonpea lines for wilt resistance. Seedlings were raised in polythene bags as in the root dip technique. Eight-day-old seedlings grown on ploythene bags were taken out and transplanted in the *F. udum* sick pots @ five seedlings per pot. These pots were kept for each entry and each pot represented one replication. Pots were watered as and when required and observed for 30 days for wilt symptoms.

Results were similar to root dip technique, confirming the correlation between the techniques. Since both techniques gave similar results, root dip technique, which is easy to handle, can be used for confirmation of wilt resistance in greenhouse.

S Pande

Resistance to *Helicoverpa* introgressed into diverse chickpea breeding lines (2006)

Breeding lines for resistance to *Helicoverpa:* One thousand five hundred and eighty six progenies (363 F_7 progenies and 721 F_8 progenies from single-crosses, and 502 F_7 progenies from four-way crosses) were sown under natural infestation of *Helicoverpa* larvae to select progenies for resistance. Selections were visually made for plants with early maturity, lesser pod damage and higher yields. We selected 1161 progenies (236 F_7 and 490 F_8 progenies from single-crosses and 435 F_7 progenies from four-way crosses) for progeny testing next year.

CLL Gowda

International Chickpea Screening Nursery – *Helicoverpa* Resistance (ICSN-HR): Using reliable field screening techniques developed at ICRISAT for screening against *Helicoverpa*, several resistant sources have been identified. The resistant (less susceptible) sources identified in field screening were used in crosses to transfer resistance in high-yielding varieties. Pedigree selection for low borer damage under pesticide-free conditions was found effective in identifying pod borer resistant lines. This trial is intended to share material showing resistance to *Helicoverpa* with the collaborating scientists of the national programs. Most lines are of short to medium duration, adapted to environments similar to southern and central India (16 to 22°N latitudes). The objective is to evaluate promising *Helicoverpa* resistant selections in varying environments and to provide an opportunity to NARS partners for selections for use as parents or as end products suitable for various conditions. The trial with 15 chickpea genotypes, including two controls, was sent to two collaborators.

CLL Gowda

Milestone: Diversified breeding lines with resistance to *Ascochyta* blight (AB), *Botrytis* gray mold (BGM), wilt, root rot, and drought avoidance root traits developed (2006)

Development of high-yielding *Fusarium* wilt resistant breeding lines in chickpea: A total of 71 advanced breeding lines (F_6 onwards) of *desi* chickpea were evaluated in one preliminary yield trial (PYT) and two advanced yield trials (AYTs) along with two controls, JG 11 and ICCC 37. Three lines (ICCX-970047-BP-BP-P49-BP-BP, ICCX-970047-BP-BP-P64-BP-BP and ICCX-970047-BP-BP-P68-BP-BP) in PYT and one line (ICCX-970047-BP-BP-P46-BP-BP) in AYT gave 6 to 15% higher yield than the best control JG 11 and had high resistance to FW (0 to 10% mortality). These lines were as early as JG 11 in maturity (94 to 96 days) and had larger seed (24 to 25 g 100^{-1} seed as compared to 20 g 100^{-1} seed of JG 11).

In *kabuli* chickpea, 42 advanced breeding lines were evaluated in one PYT and one AYT along with the controls KAK 2 and Vihar. Though several breeding lines gave higher yield (up to 25%) than the best control KAK 2, only one line (ICCX970075-BP-BP-P27-BP-BP) had the best combination of maturity duration (94 days), seed size (32 g 100^{-1} seed), yield (1560 kg ha^{-1}) and resistance to FW (3.9% mortality) and was superior to KAK 2 (97 days to maturity, 32 g 100^{-1} seed weight, 1400 kg ha^{-1} yield, and 20% mortality from FW).

PM Gaur and S Pande

Development of high-yielding *Ascochyta* blight (AB) and *Botrytis* gray mold (BGM) resistant lines in chickpea: Fifty AB/BGM resistant advanced breeding lines of *desi* chickpea were evaluated along with 10 controls (8 promising cultivars/breeding lines from Western Australia - Sona, Moti, Sonali, Rupali, WACPE 2078, WACPE 2098, WACPE 2099, ICCV 96836; and 2 cultivars from India - JG 11, ICCV 10), in a RBD with 3 replications under a special project funded by the Council of Grain Grower Organization Ltd (COGGO), Western Australia. The entries were screened against *Fusarium* wilt (FW) in a wilt-sick nursery and for AB and BGM under controlled environment conditions. There was no line that showed excellence in all attributes. Eleven lines showed high resistance to AB (score between 2.0 to 3.0 on 1–9 scale, where 1 = highly resistant and 9 = highly susceptible) and 12 lines showed high resistance (score between 3.7 and 4.0) to BGM. One line (ICCV 04526) had high resistance to both the diseases (2.3 score for AB and 4.0 score for BGM). Nine lines gave 10 to 30% higher yield over the best control (WACPE 2099) from Western Australia, which yielded 2.0 t ha^{-1}. However, no line gave higher yield than JG 11, the higher-yielding control from India.

PM Gaur, S Pande and CLL Gowda

Multiplication of BGM and AB promising chickpea lines: One hundred and seventeen BGM and AB promising lines identified in the earlier controlled environment evaluation were multiplied in a vertisol field at ICRISAT-Patancheru. These lines, originally received from Australia (NSW Agriculture), differed in number of days for flowering and pod maturity. In each line, 20–200 g of seed was harvested and tested to reconfirm BGM and AB resistance in controlled environment.

Reconfirmation of BGM resistance: Twelve-day-old seedlings raised in sand-vermiculite mixture (4:1) were used for artificial inoculation. A susceptible control JG 62 was used as indicator in each tray along with nine test entries. The experiment was conducted with 24 plants in three replications, ie, eight plants per replication. Seedlings were transferred to the controlled environment at 15°C and 100% RH and allowed to acclimatize for 24 h before inoculation. Seedlings were then inoculated with conidial suspension of *B. cinerea* @ 3×10^5 conidia ml^{-1} using the standard weather conditions (15°C and 100% RH with 12 h light and 12 h dark). Disease severity was quantified on a 1–9 rating scale at 20 days after inoculation. Based on the mean disease score, individual chickpea lines was categorized as immune (disease score 1.0), resistant (disease score 1.1–3.0), moderately resistant (disease score 3.1–5.0), susceptible (disease score 5.1–7.0) and highly susceptible (disease score 7.1–9.0). Of the 117 lines evaluated, 108 were moderately resistant, and 36 of these had a minimum disease severity 4.0 on 1–9 rating scale.

Reconfirmation of AB resistance: The lines mentioned above were also screened for AB to confirm their resistance to AB under controlled environment at ICRISAT-Patancheru. Seedlings were raised in sand-vermiculate mixture (4:1) for 12 days in greenhouse and transferred to controlled environment at 20°C and 100% RH. Seedlings were allowed to acclimatize for 24 h and then inoculated with *A. rabiei* conidial suspension of 5×10^4 conidia ml^{-1} and incubated at 20°C and 100% RH. Disease severity was measured on a 1–9 rating scale at regular intervals up to 14 DAI. The experiment was conducted in three replications with eight plants in each replication. Among 117 lines evaluated for AB resistance, high levels of disease resistance were observed in several lines. Seventy-three lines were resistant (disease score of 1 to 3 on a 1–9 rating scale) and

38 lines were moderately resistant (disease score 3.1 to 5.0) to AB infection. Of the 73 resistant lines, 19 lines had a score of ≤2.0 on a 1–9 rating scale.

Suresh Pande and G Krishna Kishore

Australia-ICRISAT collaborative research on chickpea

***Botrytis* gray mold:** In collaboration with Department of Agriculture, Western Australia (DAWA), Center for Legumes in Mediterranean Agriculture (CLIMA) and Council of Grain Growers Organization (COGGO), Australia, 60 chickpea improved breeding lines were evaluated for *Botrytis* gray mold resistance to BGM in controlled environment at ICRISAT-Patancheru. Ten-day-old seedlings of test lines, along with JG 62 as a susceptible control were inoculated with conidial suspension of *B. cinerea* (3×10^5 conidia ml^{-1}) multiplied on autoclaved merigold flowers. Inoculated plants were maintained at 15 ± 2°C and 100% RH with a 12 h photoperiod. BGM severity was recorded on a 1–9 rating scale at 20 days after inoculation. Based on the repeated resistance evaluations, 12 lines had score 4.2–5.0 and were identified as moderately resistant to BGM disease.

***Ascochyta* blight:** The same 60 lines tested for BGM were also screened for AB resistance under controlled environment. Seedlings were raised in sand-vermiculate mixture (4:1) for 12 days in greenhouse and transferred to controlled environment at 20°C and 100% RH. Seedlings were allowed to acclimatize for 24 h and then inoculated with *A. rabiei* conidial suspension of 5×10^4 conidia ml^{-1}. Disease severity was measured on a 1–9 rating scale at regular intervals up to 14 DAI. The experiment was conducted in three replications with eight plants in each replication. Among 60 lines evaluated for AB resistance, 13 had score of <3.0 rating and were identified as resistant, while 19 liens were identified as moderately resistant (3.1–5.0 rating).

Wilt: The above 60 breeding lines were also evaluated in wilt sick plot for identification of wilt resistance. Each line was planted in one-row of 4 m with two replications. Susceptible cultivars ICC 4951 (early wilter) and ICC 5003 (late wilter) had 100% wilt in both the replications.

Of the 60 entries, four entries ICCVs 93505, 93705, 95702 and 96853 had <10% wilt and were identified as resistant cultivars. The remaining 56 entries were susceptible with wilt incidence ranging from 11 to 100 %.

Suresh Pande and PM Gaur

Evaluation of diverse breeding populations to *fusarium* wilt: In collaboration with breeders at ICRISAT-Patancheru, four F_2 populations, 13 F_3 populations, 112 trial entries and 114 crossing-block entries were evaluated for wilt resistance in wilt-sick plot. Test entries also included lines from preliminary yield trial (PYT)-*desi* (21 entries), PYT-*kabuli* (24 entries), advanced yield trial (AYT)-*desi* (49 entries) and AYT-*kabuli* (18 entries). Three released *desi* cultivars ICC 37, JG 1 and Annegiri were used as controls in PYT and AYT *desi* trials; and two *kabuli* cultivars Vihar and KAK 2 as controls in PYT and AYT *kabuli* trials. Wilt-susceptible cultivar ICC 4951 after every three test rows and wilt resistant cultivar ICC 11322 after every 12 rows (ie, after every 9 test rows) were planted along with test material for proper comparison. FOC propagules were monitored every year before and after planting of the crop, and threshold levels of the pathogen was maintained by incorporating chopped wilted plants every year. Data on number of wilted plants were recorded at seedling, pod filling and maturity stages of the crop. Lines showing <10% wilt were considered as resistant and advanced for further evaluation. Wilt incidence in susceptible control, ICC 4951 was 100% within 30 days after sowing while it was <5% in resistant ICC 11322 at harvest across the field. Individual healthy plants were selected from F_2 and F_3 populations to advance the single plant progenies. Two entries (ICCX 970047-BP-BP-P49-BP-BP and ICCX 970047-BP-BP-P68-BP-BP) in PYT-*desi* trial; and four entries (ICCX 980061- F4- P22-BP, ICCX 980068-F4-P10-BP, ICCX 980068-F4-P13-BP and ICCX 980074-F4-P23-BP) in PYT-*kabuli*, had <10% wilt incidence. AYT-*desi*, ICCX-970047-BP-BP-P46-BP-BP was asymptomatic to wilt, while ICCX 970047-BP-BP-P72-BP-BP had 10% wilt incidence. One entry in AYT-*kabuli*, ICCX-970075-BP-BP-P16-BP-BP was asymptomatic to wilt, while ICCX-970075-BP-BP-P27-BP-BP had <10% wilt incidence.

S Pande and PM Gaur

Screening of advanced wilt promising lines for wilt and root rots: Advanced wilt promising selections were further evaluated for resistance to wilt and root rots in multiple disease sick plot (MDSP). MDSP at ICRISAT-Patancheru consisted of wilt (*Fusarium oxysporum* f.sp. *ciceri*) (dominant) followed by dry root rot (*Rhizoctonia bataticola*), collar rot (*Sclerotium rolfsii*), black root rot (*Fusarium solani*) and wet root rot (*Rhizoctonia solani*) pathogens. During this current season, 23 promising germplasm and breeding lines for wilt, four lines wilt plus dry root rot (DRR), 61 advanced promising breeding selections for wilt, 24 *desi* and 11 *kabuli* advanced promising selections from Kanpur for wilt and 18 wilt resistant germplasm lines were evaluated

for resistance to wilt, DRR and collar rot in BIL 1. Wilt-susceptible early wilter JG 62 after every 4 test rows, wilt susceptible late wilter K 850 and wilt resistant WR 315 alternately were planted after every 12 test rows along with test material for proper comparison and also inoculum buildup/multiplication in this field. Inoculum levels of these pathogens were monitored before and after the chickpea crop every year. Observations on number of plants killed were recorded at seedling, flowering-pod filling and near maturity stages of the crop. Of the 23 advanced wilt promising germplasm and released lines, 11 lines (ICCs 338, 11223, 12243, 14344, 14391, 14434, 15949, 16124, IPC 98-51, KAK-2 and ICCX-850621-BH-BH-1H-BH-BH were found resistant to wilt, DRR and collar rot under field conditions. All the four wilt plus DRR promising lines tested, ICCs 14432, 14449, ICCX 850636-BH-26H-BH and ICCX 830235-BH-BH-5H were found resistant to wilt, DRR and collar rot diseases. Of the 61 advanced wilt promising breeding lines, 29 had combined resistance to wilt, DRR and collar rot under field conditions. Of the 24 *desi* and 11 *kabuli* selections from IIPR, Kanpur, 15 lines from *desi* and one *kabuli* line MPJGK 99-115 showed resistance to all the three diseases under field conditions. Of the 18 wilt resistant lines, 10 lines had resistance to wilt.

S Pande

ICAR-ICRISAT collaborative research on chickpea wilt and *Ascochyta* blight: Under ICRISAT-IIPR collaboration, lines from 12 trials, IVT 1 (LS) with 8 entries, AVT 1 (LS) 8 entries, AVT 2 (LS) 8 entries, IVT (RF) 11 entries, AVT 1 (RF) 10 entries, IVT 1 (bold) 11 entries, high input trial 9 entries, AVT 2 (*kabuli*) 6 entries, IVT (*kabuli*) 11 entries, AVT 1 (*desi*) 3 entries, IVT 1 (*desi*) 12 entries, and National Nursery (43 entries) were evaluated against wilt, following standard field evaluation techniques in wilt sick plot. NNAB with 49 entries was screened for *Ascochyta* blight resistance in controlled environment. Cultivar IDG 11 from IVT *desi* was found asymptomatic to wilt. Among the other lines, ALG 7 from AVT 1 (LS), A2LG 7 from AVT 2(LS), IRFG 10 from IVT (RF), ARFG 5, ARFG 8 from AVT 1 (RF), IBG 10 from IVT 1 (bold) were found resistant (<10%). In the National Nursery, resistant entries (<10% incidence) lines were, NNW 4, NNW 5, NNW 6, NNW 7, NNW 8, NNW 9, NNW 10, NNW 11, NNW 12, NNW 16, NNW 19, NNW 29 and NNW 33.

S Pande

Milestone: Diversified oil-type groundnut breeding lines of different duration with improved yield potential and resistance to biotic and abiotic stresses developed (2006)

Foliar diseases resistant breeding lines and varieties: Late leaf spot (LLS) and rust are the two most important foliar fungal diseases of groundnut in Asia. Together they cause up to 70% loss in pod yield besides adversely affecting the seed and fodder quality. Sixty-seven crosses were made (49 in the 2004/05 postrainy season and 18 in the 2005 rainy season) to generate populations for foliar disease resistance and high pod yield with desirable agronomic traits. The advanced breeding lines and different populations (from earlier crosses) were screened in the foliar disease screening nursery. The populations/progenies were scored for rust and LLS incidence on a scale of 1–9 (where 1 = no disease and 9 = >80% damaged foliage) at the time of harvest. In 418 F_2–F_7 foliar disease resistant breeding population grown in the 2004/05 postrainy season in a foliar disease nursery, 10 single plant and 394 bulk selections were made based on foliar disease resistance, pod yield and other desirable agronomic traits. The promising selections came from ICGV 92069 × ICGV 93184 and ICGV 96177 × ICGV 94088 crosses. Among 239 progeny bulks grown in an infector-row foliar disease screening nursery in the 2005 rainy season, 75 single plants and 266 bulk selections were made based on disease reaction, pod yield and other desirable agronomic traits. The promising selections, among others, came from ICGV 92069 × ICGV 93184 and ICGV 96246 × 92R/75 crosses.

In the 2004/05 postrainy season, 84 foliar disease resistant advanced breeding lines were evaluated in 4 replicated yield trials for foliar diseases and for desirable pod and seed characters in a disease-screening nursery. The severity of foliar diseases in the screening nursery was low. In the Elite (Spanish Bunch, SB) trial, 3 lines were significantly superior to the control ICGV 86590 (3.6 t ha^{-1} ± 0.22 t ha^{-1}). ICGV 02410 (4.7 t ha^{-1}) produced the highest yield in the trial. In the Elite (Virginia Bunch, VB) trial, ICGV 02446 produced significantly higher yield (4.9 t ha^{-1}) than the best control ICGS 76 (3.8 ± 0.21 t ha^{-1}). In the Preliminary (SB) trial, 14 lines produced significantly higher pod yields (3.8–4.6 t ha^{-1}) than the best control cultivar ICGS 44 (2.8 ± 0.34 t ha^{-1}). In the Preliminary (VB) trial, three lines (ICGV # 04093, 04087 and 04094) produced significantly higher pod yield (4.4, 4.2 and 4.1 t ha^{-1}, respectively) than the highest-yielding control ICGS 76 (3.5 ± 0.17 t ha^{-1}). In 2005 rainy season, 133 advanced breeding lines (in 6 different replicated yield trials) and 108 advanced breeding lines (in 2 augmented trials) were evaluated. The elite and advanced yield trials were evaluated under both irrigated and rainfed conditions. The preliminary trials were evaluated only under irrigated conditions. The infector-row system (for disease screening) was adopted only under irrigated conditions. The irrigated trials were artificially inoculated twice to create uniform disease (LLS and rust) pressure and were

scored for disease reaction on a 1–9 scale on 72, 88 and 103 DAS. The rainfed trials had only natural disease infection and were not scored for disease reaction.

Under irrigated conditions, in Elite Trial (SB), three lines significantly outyielded (3.4–3.1 ± 0.26 t ha^{-1}) the highest-yielding control ICGS 44 (2.3 t ha^{-1}). In this trial, ICGV 02410 gave the highest pod yield of 3.4 t ha^{-1} with a disease score of 6.0 for LLS and 2 for rust compared with an 8.0 score for LLS and 7.5 score for rust in ICGS 44 at 103 DAS. Under rainfed conditions, four lines significantly outyielded (3.7–3.3 ± 0.14 t ha^{-1}) the highest-yielding control ICGV 86590 (2.8 t pods ha^{-1}). Of these, ICGV # 02410 and 02415 significantly outyielded the controls in both irrigated and rainfed trials. In Elite Trial (VB), evaluated under irrigated conditions, ICGV 02446 (3.6 ± 0.10 t ha^{-1}) gave significantly higher pod yield than the highest-yielding control ICGV 86699 (3.0 t ha^{-1}). ICGV 02446 scored 7 for LLS and 2 for rust compared with a 5.5 for LLS and 1.5 for rust of ICGV 86699 at 103 DAS. Six lines significantly outyielded (3.6–3.1 ± 0.15 t ha^{-1}) the highest-yielding control ICGV 86699 (2.4 t pods ha^{-1}) when the trial was grown under rainfed conditions. ICGV 02446 gave the highest pod yield under both rainfed and irrigated conditions. In Advanced Trial (SB), 14 lines produced significantly higher pod yield (3.7–2.9 ± 0.21 t ha^{-1}) than the highest-yielding control ICGV 86590 (2.0 t ha^{-1}). In this trial, the best yielding line ICGV 04093 had a score of 6.5 for LLS and 2 for rust compared with score 8.0 for LLS and 2 for rust in ICGV 86590. In the same trial under rainfed conditions, 14 lines significantly outyielded (4.0–3.4 ± 0.10 t ha^{-1}) the highest-yielding control ICGV 86590 (3.0 t pods ha^{-1}). In this trial, ICGV 04060 gave the highest pod yield (4.0 t ha^{-1}). Ten lines performed well under both irrigated and rainfed conditions.

In Preliminary Trial (SB), six lines significantly outyielded (3.3–2.8 ± 0.22 t ha^{-1}) the highest-yielding control ICGV 86590 (2.0 t ha^{-1}). The highest yielding test line ICGV 05097 scored a 5 for LLS and 2 for rust compared with 7.5 for LLS and 1.5 for rust in ICGV 86590.

In Augmented Trial-1, 4 lines out of the 72 (adjusted pod yield = 3.52–3.25 t ha^{-1}, rust score = 2.0, LLS score = 8.0–5.0) outyielded the highest-yielding control JL 24 (2.66 t ha^{-1}, 8.0, 8.5; LSI = 0.51) and the resistant control ICGV 86699 (1.80 t ha^{-1}, 2.0, 5.0; LSI = 0.51). The best entry in this trial was ICGX 000134 (3.5 t ha^{-1}; 2.0, 5.0). In Augmented Trial-2, no entry among the 28 test entries evaluated could outyield the best and the resistant controls. Nine foliar disease resistant lines were selected in the 2004/05 postrainy seasons for inclusion in international trials. The seed of these lines was multiplied in 2005 rainy season.

Two late leaf spot-resistant germplasm lines (ICG 11337 and ICG 13919) and a susceptible variety JL 24 were used as parents for studying the inheritance of components of late leaf spot resistance. Parents, F_1, F_2 and backcross generations were screened in the field and growth chambers for different components of resistance. Detailed observations, on incubation period, latent period, lesion number, % leaf area damage and lesion diameter were recorded by the pathology group on the detached leaves in the growth chambers. The field observations included % defoliation at 75, 90 and 105 days after planting and associated disease scores on a 1–9 scale. Data compilation is in progress.

***A. flavus*/aflatoxin resistant breeding lines and varieties:** Aflatoxin contamination of groundnut seeds caused by *Aspergillus flavus* poses serious health hazards to humans and livestock, and hampers international trade. Nine new crosses were made in the 2005 rainy season to develop aflatoxin-tolerant breeding lines. In the 2005/2006 postrainy season, 393 F_3–F_8 bulk and 196 single plant selections were planted for further selection.

Thirty–five advanced breeding lines were evaluated in 3 trials during the 2004/05 postrainy season in *A. flavus* sick plot to screen for tolerance to aflatoxin production. ICGV 02226 and ICGV 02234 showed low aflatoxin content (2.89 and 4.21 µg kg^{-1}, respectively) compared to the zonal control Somnath (445.42 µg kg^{-1}). In the 2005 rainy season, 106 advanced breeding lines were evaluated in 5 replicated trials and 380 advanced breeding lines in 3 augmented trials. The elite and advanced trials were grown under both irrigated and rainfed conditions. The preliminary and augmented trials were conducted only under irrigation. All the replicated trials were also grown in an *A. flavus* sick plot to record observations on pre-harvest seed infection and aflatoxin production. The results on pre-harvest seed infection and aflatoxin contamination are awaited.

In Elite Trial, four lines (ICGV # 01099, 01073, 02194 and 01060) outyielded (3.3–2.8 ± 0.17 t ha^{-1}) the highest yielding control ICGS 11 (2.2 t ha^{-1}) and the resistant control J 11 (1.5 t ha^{-1}) under irrigated conditions. Under rainfed conditions, 9 lines gave significantly higher pod yield (2.4–1.7 ± 0.01 t ha^{-1}) than the highest-yielding control ICGS 11 (1.4 t ha^{-1}) and the resistant control J 11 (1.0 t ha^{-1}). ICGV 01060 performed the best under rainfed conditions. All the four lines performing well under irrigation also did well under rainfed conditions. In Advanced Trial (SB) under irrigated conditions, ICGV # 03311 and 03315 (3.0 ± 0.11 t ha^{-1}) outyielded the

highest-yielding control ICGS 11 (2.2 t ha^{-1}) and resistant control J 11 (1.4 t ha^{-1}). The same two lines also outyielded (2.3 ± 0.06 t ha^{-1}) the highest-yielding control ICGS 11 (1.6 t ha^{-1}) and resistant control J 11 (1.1 t ha^{-1}) under rainfed conditions. In Advanced Trial (Dark Green Leaves) evaluated under irrigated conditions, ICGV 03398 c(2.9 ± 0.23 t ha^{-1}) outyielded the highest-yielding control ICGS 11 (1.8 t ha^{-1}) and the resistant control J 11 (1.2 t ha^{-1}). The same line also outyielded (2.0 ± 0.12 t ha^{-1}) the controls ICGS 11 (1.4 t ha^{-1}) and J 11 (1.0 t ha^{-1}) when evaluated under rainfed conditions. In Preliminary Trial-1, seven lines produced significantly higher pod yield (3.7–2.2 ± 0.13 t ha^{-1}) than the highest-yielding control ICGS 11 (1.8 t ha^{-1}) and the resistant control J 11 (1.5 t ha^{-1}) under irrigated conditions.

In Augmented Trial-1, 14 lines (adjusted pod yield = 3.96–2.83 t ha^{-1}) out of 65 test lines outyielded the highest-yielding control ICGS 76 (2.13 t ha^{-1}; LSI = 0.50). ICGX 000109 - treatment 60 (3.9 t ha^{-1}) was the best entry in the trial. In Augmented Trial-2, out of 120 test entries, 19 lines (adjusted pod yield = 2.63–1.76 t ha^{-1}) produced significantly higher yield than the highest-yielding control ICGS 76 (1.30 t ha^{-1}; LSI = 0.44). Three top entries in this trial were ICGX 000021 (2.63 t ha^{-1}), ICGX 000103 (2.5 t ha^{-1}) and ICGX 000021 (2.3 t ha^{-1}). In Augmented Trial-3, only ICGX 000109 (3.30 t ha^{-1}) outyielded the best control ICGS 76 (1.80 t ha^{-1}; LSI = 0.89).

SN Nigam, R Aruna and Farid Waliyar

Drought resistant breeding lines and varieties: Drought reduces not only the pod and haulm yields but also affects the quality of the produce. It predisposes the crop to aflatoxin contamination by *A. flavus* infection. It can occur at any stage of the crop growth. Sixty crosses were made (49 in the 2004/05 postrainy season and 11 in the 2005 rainy season) between the breeding lines with high water use efficiency traits and new germplasm sources (ICG # 1834, 1891, 5100, 5341, 5465, 8230) to generate populations for selection under moisture stress.

In 710 F_2–F_7 drought tolerant breeding populations grown under imposed mid-season moisture stress (65–100 DAE), 1 single plant and 607 bulks were selected based on pod yield and other desirable agronomic traits. The promising selections came from (ICGV 96294 × ICGV 92004), (ICGV 87846 × ICGV 99240) and (ICGV 87846 × (ICGV 87290 × ICGV 87846) crosses. In 2005 rainy season, 223 single plants and 327 bulks based on pod yield and other desirable agronomic traits were selected from 467 F_2–F_7 drought-tolerant breeding populations. Crosses (ICGV 92069 × ICGV 93184) × (ICGS 44 × ICGV 76) and (ICGV 92069 × ICGV 93184) × ICGV 98300, among others, gave the most promising selections.

Among the 71 drought-tolerant advanced breeding lines evaluated in 5 replicated yield trials in the 2004/05 postrainy season under imposed mid-season moisture stress conditions and under full irrigated conditions, only one line ICGV 03115 in the advanced drought trial (VB) with pod yield of 2.9 t ha^{-1}, 76% shelling outturn and 39 g 100-seed weight outyielded ICGS 76 (2.2 ± 0.16 t ha^{-1}; 57 %; 40 g) under imposed stress. ICGV # 99029, 92267 and 99054 (3.1–3.3 t ha^{-1}) outperformed JL 24 (2.5 ± 0.27 t ha^{-1}) under fully irrigated conditions in the Elite on-farm trial.

In 2005 rainy season, 53-advanced breeding lines along with controls were evaluated in 4 replicated trials. The elite and advanced trials were grown under both irrigated and rainfed conditions whereas preliminary trials were grown only under rainfed conditions. Another 52 advanced breeding lines were evaluated in an augmented design under irrigated conditions only. In Elite Trial (SB) under irrigated conditions, eight lines produced significantly higher pod yield (5.1–4.2 ± 0.18 t ha^{-1}) than the highest-yielding control ICGV 00350 (3.2 t ha^{-1}). In this trial, the highest-yielding line ICGV 03063 gave 70% shelling outturn with 44 g of 100-seed weight compared with 68% shelling outturn and 35 g 100-seed weight of ICGV 00350. The same lines significantly outyielded (4.9–4.0 ± 0.10 t ha^{-1}) the highest yielding control ICGV 00350 (2.7 t ha^{-1}) when evaluated under rainfed conditions. In this trial, the highest-yielding line ICGV 03064 gave 60% shelling outturn with 38 g of 100-seed weight compared with 67% shelling outturn and 34 g 100-seed weight of ICGV 00350. ICGV # 03064, 03061, 03063 and 03056 were among the top five under both the growing conditions. In Preliminary Trial (SB), 10 lines significantly outyielded (5.1–4.2 ± 0.25 t ha^{-1}) the control ICGV 00350 (3.4 t ha^{-1}). In this trial, the highest yielding line ICGV 05153 gave 64% shelling outturn with 37 g 100-seed weight compared with 65% shelling outturn and 36 g 100-seed weight of ICGV 00350. In Augmented Trial-1, conducted with 48 test entries under irrigated conditions, 37 lines (adjusted pod yield = 5.83–3.63 t ha^{-1}) outyielded the control ICGV 00350 (3.40 t ha^{-1}; LSI = 0.13). The best entry in this trial was ICGX 000052 (5.8 t ha^{-1}). Twelve drought-tolerant lines were selected in the 2004/05 postrainy seasons for inclusion in international trials.

Three hundred twenty F_9 RILs and their parents (ICGV 86031 and TAG 24) were phenotyped twice in the pot culture for water-use-efficiency traits. Parental polymorphism is also being assessed with SSR markers in collaboration with GT-Biotechnology scientists. A diverse set of genotypes (189) has been selected to initiate

association mapping. The set would be genotyped with SSR markers and phenotyped for different morphological traits of interest.

SN Nigam, R Aruna and Vincent Vadez

High oil content breeding lines and varieties: Varieties adapted to local agroecological conditions are required for enhancing productivity. Lines with high oil content are required in countries where groundnut is mostly crushed for edible oil. In Elite Trial (High Oil Content), 12 lines gave significantly higher pod yield (4.9-4.1 ± 0.16 t ha^{-1}) than the highest-yielding control ICGV 86590 (3.7 t ha^{-1}) when evaluated under irrigated conditions. In this trial, ICGV 99033 gave the highest pod yield (4.9 t ha^{-1}) with 66% shelling outturn and 50 g of 100-seed weight compared with 69% shelling outturn and 40 g of 100-seed weight of control ICGV 86590. The oil content in this trial ranged from 54.7 to 44.7% and the protein content from 26.9 to 18.4% in the 2004/2005 postrainy season. Eighteen lines had oil content ≥50%. The oil content in ICGV 86590 was 49.1%. Although ICGV 00017 had the highest oil content in the 2004/2005 postrainy season (54.7%), its yield was low in the 2005 rainy season (2.4 t ha^{-1}). Seven of the 12 significantly higher yielding lines had oil content between 52.5% and 50.1%. In evaluation under rainfed conditions, only 7 lines significantly outyielded (pod yield:-4.2–3.8 ± 0.15 t ha^{-1}) but 33 entries (49.3–55.5%) had higher oil content than the highest-yielding control ICGV 86590 (3.3 t ha^{-1}; 46.8%). ICGV 00434 gave the highest pod yield (4.2 t ha^{-1}) and ICGV 00351 had the maximum oil content (55.5%) in this trial. ICGV # 99017, 01273, and 01270 were among the top five under both growing conditions.

Medium-duration breeding lines and varieties: Varieties which take 110–120 days to mature at ICRISAT-Patancheru, fall in the medium-duration group. Seventeen crosses were made in the 2005 rainy season to generate new medium-duration populations for selection for high yield and desirable agronomical characteristics. In the 574 F_2–F_7 medium-duration breeding populations grown under high input conditions, 78 single plants and 642 bulks were selected based on high yield and other pod and seed characters. Among others, the most promising selections came from (ICGV 93023 × ICGV 92088) and (ICGV 93023 × ICGV 99160) crosses. These selections (572) were sown in the 2005 rainy season for further selection. Among these, 463 bulks and 293 single plants were selected based on high yield and other desirable agronomic traits**.** The most promising selections, among others, came from (TAG 24 × ICGV 98300), (JL 24 × ICGV 98300), (TAG 24 × ICGV 99032) and (TMV 2 × ICGV 98300) crosses. These selections (756) were sown in the 2005/2006 postrainy season for further evaluation and selection.

In the 2004/05 postrainy season, 128 medium-duration advanced breeding lines were evaluated in 5 replicated yield trials. In the Elite (SB) trial, three lines ICGV # 02323, 02322 and 03015 outyielded (5.6, 5.4 and 5.0 t ha^{-1}, respectively) the best control ICGV 86590 (3.8 ± 0.31 t ha^{-1}). In the Advanced (VB) trial, four lines ICGV # 03043, 03042, 03015 and 03037 produced significantly higher pod yield (5.5, 5.4, 5.2 and 5.1 t ha^{-1}, respectively) compared with ICGS 76 (3.7 ± 0.28 t ha^{-1}). In the Preliminary (SB) trial, two lines ICGV # 04112 and 04115 produced significantly higher pod yield (5.8 and 5.7 ± 0.42 t ha^{-1}) than the highest-yielding control ICGV 86590 (4.26 t ha^{-1}). In the Preliminary (VB) trial, seven lines produced significantly higher pod yield (3.9–4.9 ± 0.26 t ha^{-1}) than the highest-yielding control ICGV 86325 (3.1 t ha^{-1}). In the 2005 rainy season, 193 advanced breeding lines in 6 replicated trials and 76 lines in 2 augmented trials were evaluated for pod yield and other agronomic traits. Elite and advanced trials were grown under both irrigated and rainfed conditions, while all other trials were grown under irrigated conditions only.

In Elite Trial (SB), all the 16 lines produced significantly higher pod yield (4.8–3.2 ± 0.20 t ha^{-1}) than the highest-yielding control ICGS 44 (2.4 t ha^{-1}) under irrigated conditions. In this trial, ICGV 03043 gave the highest pod yield (4.8 t ha^{-1}) with 65% shelling outturn and 36 g of 100-seed weight compared with 69% shelling outturn and 36 g of 100- seed weight of control ICGS 44. In the same trial conducted under rainfed conditions, 15 lines significantly outyielded (3.8–2.5 ± 0.18 t ha^{-1}) the highest-yielding control ICGS 44 (1.9 t ha^{-1}). In this trial, ICGV 03016 produced the highest pod yield (3.8 t ha^{-1}). ICGV # 03043, 03014, 03042 and 03016 were common in the top five under both the growing conditions. In Advanced Trial (SB), evaluated under irrigated conditions, 14 lines significantly outyielded (5.2–3.4 ± 0.18 t ha^{-1}) the highest-yielding control ICGS 44 (2.6 t ha^{-1}). In this trial, ICGV 04122 gave the highest pod yield (5.2 t ha^{-1}) with 64% shelling outturn and 47 g of 100-seed weight compared with 69% shelling outturn and 34 g of 100-seed weight of ICGS 44. In the same trial under rainfed conditions, 15 lines significantly outyielded (4.5–2.2 ± 0.18 t ha^{-1}) the highest-yielding control ICGS 11 (1.6 t pods ha^{-1}). ICGV 04122 gave the highest pod yield (4.5 t ha^{-1}) in rainfed trial also. ICGV # 04122, 04148, 04112 and 04126 were common in the top five under both the growing conditions.

In Advanced Trial (VB), evaluated under irrigated conditions, 12 lines produced significantly higher pod yield (4.9–3.6 ± 0.19 t ha^{-1}) than the highest-yielding control ICGV 86325 (2.6 t ha^{-1}). In this trial ICGV 04149 gave

the highest pod yield (4.9 t ha^{-1}) with 75% shelling outturn and 49 g of 100-seed weight compared with 75% shelling outturn and 45 g of 100-seed weight of ICGV 86325. When this trial was evaluated under rainfed conditions, the same 12 lines significantly outyielded (4.6–3.1 ± 0.13 t ha^{-1}) the highest-yielding control ICGV 86325 (2.5 t ha^{-1}). ICGV 04141 gave the highest pod yield (4.6 t ha^{-1}) in rainfed trial. ICGV # 04141, 04149, 04142 and 03035 were common in the top five under both the growing conditions.

In Preliminary Trial (SB), evaluated under irrigated conditions, 32 lines produced significantly higher pod yield (4.9–2.9 ± 0.20 t ha^{-1}) than the highest yielding control ICGV 95070 (2.8 t ha^{-1}). In this trial, ICGV 05034 gave the highest pod yield (4.9 t ha^{-1}) with 66% shelling outturn and 44 g of 100-seed weight compared with 67% shelling outturn and 37 g of 100-seed weight of ICGV 95070. In Preliminary Trial (VB), 11 lines significantly outyielded (4.7–3.3 ± 0.19 t ha^{-1}) the best control ICGS 76 (2.8 t ha^{-1}). In this trial, ICGV 05063 gave the highest pod yield (4.8 t ha^{-1}) with 72% shelling outturn and 64 g of 100-seed weight compared with 68% shelling outturn and 35 g of 100-seed weight of ICGS 76.

In Augmented Trial-1, 15 of the 44 lines significantly outyielded (adjusted pod yield = 5.05–3.58 t ha^{-1}) the best control ICGS 44 (2.94 t ha^{-1}; LSI = 0.59). In this trial, ICGX 000127 (5.1 t ha^{-1}) and ICGX 010029 (4.8 t ha^{-1}) were the best performers. In Augmented Trial-2, among the 24 test entries, 7 lines significantly outyielded (adjusted pod yield = 3.55–2.25 t ha^{-1}) the best control ICGV 86325 (1.76 t ha^{-1}; LSI = 0.43). Among the test lines, ICGX 000032 (3.6 t ha^{-1}) and ICGX 000124 (2.9 t ha^{-1}) were top ranking two lines. Eight medium duration lines were selected in the 2004/05 postrainy seasons for inclusion in international trials. The seed of these lines has been multiplied in the 2005 rainy season.

Confectionery type breeding lines and varieties: The food use of groundnut has been increasing over the years. Seed size, shape, uniformity and color assume importance when groundnut is used for direct consumption. Fifty eight new crosses (33 in the 2004/05 postrainy season and 25 in the 2005 rainy season) were made to generate populations for selection for confectionery traits, high yield and other desirable characters. ICGV # 95179, 00451, 00440, 94215 and ICG # 6767, 6670 and 1651 were the new parents used to develop new breeding populations.

Of the 665 F_2–F_7 confectionery breeding populations grown under high input conditions, 25 single plants and 279 bulks were selected for confectionery traits and high pod yield. The promising selections came from (ICGV 98408 × ICGV 88386) and (ICGV 96236 × ICGV 88386) crosses. These 304 selections (269 segregating populations and 35 in yield trials) were sown in the 2005 rainy season under high input conditions. In the 269 F_2–F_7 confectionery breeding populations grown under high input conditions in the 2005 rainy season, we made 261 bulk selections based on superior agronomic and confectionery traits. The promising selections, among others, came from ICGV 96016 × Sunoleic and ICGV 99085 × ICGV 86564 crosses.

Thirty-five advanced breeding lines were evaluated in the 2004/05 postrainy season in 3 replicated trials. Five lines (ICGV # 02242, 02234, 02227, 02233 and 02226) in the Elite (SB) trial (3.3–3.9 ± 0.27 t ha^{-1}) and seven lines (3.0–4.7 ± 0.32 t ha^{-1}) in the preliminary yield (SB) trial significantly outyielded the control Somnath (2.2 t ha^{-1}). Fifty-one confectionery advanced breeding lines (including controls) were evaluated in three replicated yield trials under high input conditions during the 2005 rainy season. In Preliminary Trial (SB), ICGV 05174 (pod yield = 3.5 ± 0.20 t ha^{-1}; 100-seed weight = 58 g) gave significantly higher pod yield than the highest-yielding control ICGV 97045 (2.8 t ha^{-1}; 57 g). In Preliminary Trial (VB), ICGV 05200 (3.8 ± 0.15 t ha^{-1}; 69 g) produced significantly higher pod yield than the highest-yielding control ICGV 98432 (3.2 t ha^{-1}; 67 g). Fourteen confectionery elite breeding lines were selected in the 2004/05 postrainy season for inclusion in international trials. Their seed has been multiplied in the 2005 rainy season.

Short-duration breeding lines and varieties: Short-duration (<100 days) varieties are needed in areas where the growing season is short and the crop suffers from end-of-season drought. Such varieties are also suitable in multiple cropping systems, and rice-fallow conditions to utilize residual moisture. Five new crosses were made in the 2005 rainy season to generate breeding populations for selection for short-duration, high yield and other desirable agronomic characteristics. Selections (648 bulks and 520 single plants) made during the 2004/2005 postrainy season were sown during the 2005/2006 postrainy season for further selection.

We evaluated 114 lines (including controls) in 5 replicated trials and 360 lines in 3 augmented trials for yield and other agronomic traits under irrigated conditions in the 2005 rainy season. The elite and advanced yield trials were also evaluated under rainfed conditions. All the trials conducted under both irrigated and rainfed conditions were harvested when the crop accumulated 1470 °Cd equivalent to 90 days after sowing (DAS) at Patancheru. Under irrigation, in Elite Trial (SB), 12 lines significantly outyielded (pod yield = 2.3–1.8 ± 0.12 t

ha^{-1}) the control JL 24 (pod yield = 1.4 t ha^{-1}; shelling outturn = 62%; 100-seed weight = 29 g). ICGV 02099 (2.3 t ha^{-1}; 66%; 37g) was the best entry among the 12 lines. In the same trial under rainfed conditions, 10 lines outyielded (1.8–1.4 ± 0.09 t ha^{-1}) the highest yielding control Chico (1.1 t ha^{-1}). The best entry in the trial was ICGV 02099 (1.8 t ha^{-1}). ICGV # 02099, 02022, 02144 and 02126 were among the top five under both the growing conditions. In irrigated Elite Trial (VB), 3 lines (ICGV # 98294, 98292 and 98293) gave significantly higher pod yield (2.9-2.6 ± 0.10 t ha^{-1}) than the best control ICGS 76 (2.1 t ha^{-1}; 66%; 42 g). When the same entries were evaluated under rainfed conditions, ICGV # 98292 (2.7 ± 0.09 t ha^{-1}), 98293 (2.5 ± 0.09 t ha^{-1}) and 98287 (1.9 ± 0.09 t ha^{-1}), outyielded the control ICGS 76 (1.5 t ha^{-1}). ICGV # 98292 and 98293 were common in the top three under both the growing conditions. In Elite Trial (large seeds) evaluated under irrigated conditions, none of the test entries significantly outyielded the best control ICGS 44 (2.0 t ha^{-1}; 64%; 40 g) for pod yield. However, ICGV # 01232 (45 g), 02131 (43 g), 01234 (43 g) and 99258 (42 g) recorded a higher 100-seed weight than ICGS 44. In the same trial under rainfed conditions, five lines outyielded (2.2–1.9 ± 0.16 t ha^{-1}) the highest-yielding control Somnath (1.4 t ha^{-1}). Only ICGV 01234 performed well under both the growing conditions.

In Advanced Trial (SB), 10 lines produced significantly higher pod yield (2.5–1.9 ± 0.09 t ha^{-1}) than the highest yielding control JL 24 (1.6 t ha^{-1}; 63% shelling outturn; 33 g 100 seed weight) and Chico (1.5 t ha^{-1}; 58 %; 31.0 g). The best entry was ICGV 03206 (2.5 t ha^{-1}; 71%; 37 g). When the entries were evaluated under rainfed conditions, 12 lines outyielded (1.9–1.5 ± 0.09 t ha^{-1}) the control Chico (1.2 t ha^{-1}). The best test line was ICGV 03207 (1.9 t ha^{-1}). ICGV # 03208 and 03210 were common in the top five under both the growing conditions. In Preliminary Trial, 10 lines significantly outyielded (2.0–1.6 ± 0.10 t ha^{-1}) the highest-yielding control Chico (1.3 t ha^{-1}; 69%; 23g) under irrigated conditions. The best entry in the trial was ICGV 04022 (2.0 t ha^{-1}; 64%; 36 g).

A special trial was formulated to compare the performance of top-yielding short-duration varieties developed over the years at ICRISAT Center. The trial consisted of 44 test entries and was grown under irrigated conditions only. It was harvested 90 DAS. Thirty-four entries gave significantly higher pod yield than the Indian national control JL 24 (0.86 ± 0.15 t ha^{-1}). Among the test lines only ICGV 99195 (2.4 t ha^{-1}; 64%; 34 g) gave significantly higher yield than ICGV 91114 (1.8 t ha^{-1}; 67%; 33 g), which is becoming popular among the farmers in India.

In Augmented Trial-1, 83 test lines (adjusted pod yield = 2.88–1.60 t ha^{-1}) out of 110 significantly outyielded the control Robut 33-1 (1.26 t ha^{-1}, LSI = 0.33). The top three entries in this trial were ICGX 000096 (2.9 t ha^{-1}), ICGX 000096 (2.8 t ha^{-1}) and ICGX 000014 (2.6 t ha^{-1}). In Augmented Trial-2, 59 out of 95 test lines (adjusted pod yield = 2.70–1.43 t ha^{-1}) significantly outyielded the control Robut 33-1 (1.02 t ha^{-1}, LSI= 0.39). The best entry in this trial was ICGX 000101 (2.7 t ha^{-1}). In Augmented Trial-3, we evaluated 140 test lines. Of these, 18 produced significantly higher yield (adjusted pod yield = 2.79–2.09 t ha^{-1}) than the highest-yielding control Robut 33-1 (1.57 t ha^{-1}, LSI = 0.51). ICGX 000003 (2.8 t ha^{-1}) and ICGX 000014 (2.7 t ha^{-1}) were the top two entries in this trial.

Adaptation to Anantapur (India) conditions: Breeding lines, specifically bred for Anantapur (Andhra Pradesh, India) conditions were evaluated in two different trials. In Trial-1, 43 test entries selected from previous trials and 6 controls were evaluated in a 7 × 7 lattice design, both under irrigated and rainfed conditions at ICRISAT-Patancheru. In this trial, six entries (3.1–2.9 ± 0.0.16 t ha^{-1}) outyielded the best control TAG 24 (2.4 t ha^{-1}) under irrigated conditions. Under rainfed conditions also, six lines produced significantly higher pod yield (2.6–2.3 ± 0.13 t ha^{-1}) than the highest-yielding control TAG 24 (2.0 t ha^{-1}). Three lines, ICGX 020006-treatment 4, ICGX 020006 and ICGX 020021, were common in the top five under both the growing conditions. In Trial-2, 36 test lines along with 9 controls were evaluated in an augmented design under irrigated conditions. In this trial, nine lines (adjusted pod yield = 4.19–2.89 t ha^{-1}) outyielded the best control TAG 24 (2.36 t ha^{-1}, LSI= 0.41). The top entry in this trial was ICGV 87846 followed by ICGV 99029 (4.06 t ha^{-1}).

SN Nigam and R Aruna

Aflatoxin resistance in germplasm and drought-tolerant breeding lines: Aflatoxins are toxic, immuno-suppressive, mutagenic carcinogens and can cause various health effects including liver and other cancers in humans and animals. This problem can be resolved by using cultivars that resist the toxin-producing fungus (*Aspergillus flavus*) infection and aflatoxin production.

One hundred-thirteen advanced breeding lines were evaluated during 2004–05 postrainy season for *A. flavus* seed infection and aflatoxin contamination. These lines comprised of six trials based on category of the materials and tested in three replications. The materials were screened in the sick plot. *A. flavus* inoculum was

applied four times during the crop growth period, and end-of-season drought was imposed to facilitate the seed infection. At harvest, seed samples from each plot were collected separately; and analyzed for *A. flavus* seed infection using blotter plate method, and aflatoxin contamination by indirect competitive ELISA method. *A. flavus* infection ranged from 2.7% to 69.3% and aflatoxin contamination in these lines was 1.6 to 4849 µg kg^{-1}. Six of the 24 resistant lines (ICGV 01002, 01094, 01096, 01156, 02191 and 02194) showed <5 µg kg^{-1} aflatoxin. All the eight advanced dark green leaf lines were susceptible (>100 µg kg^{-1}) to aflatoxin contamination. Among 21 advanced Spanish bunch varieties, only two lines (ICGV 03319 and 03341), and one (ICGV 03389) out of 10 advanced Virginia bunch varieties showed <5 µg kg^{-1} aflatoxin.

One hundred-thirty advanced breeding lines (in six trials with four replications) were screened in the sick plot for resistance to *Aspergillus flavus* infection and aflatoxin contamination under artificially inoculated conditions during the 2005 rainy season. After harvest, the produce was dried under natural sunlight with the pods stripped manually. The samples are being processed for *A. flavus* infection and aflatoxin contamination.

Farid Waliyar and SN Nigam

Milestone: Diversified groundnut breeding lines with improved confectionery traits developed

Advanced confectionery groundnut lines assessed for mycotoxins: Aflatoxin resistance in confectionery groundnut is essential because the groundnuts are consumed directly as food. Also, aflatoxin resistance in confectionery groundnut is crucial from international trade point of view.

Twenty-six confectionery groundnut lines (Spanish and Virginia type) comprising two trials in three replications were screened during 2004–05 postrainy season in sick plot under artificially inoculated conditions. The inoculum multiplied on sorghum/maize/pearl millet seed was applied four times during the crop growth period. The predisposing end-of-season (30 days before harvest) drought was imposed to facilitate the fungal infection. Harvesting was done by lifting the plants and the produce was dried in sunlight before the kernels were shelled. *A. flavus* infection was determined using blotter plate method, and aflatoxin content was estimated by ELISA method. In Spanish bunch confectionery types, *A. flavus* infection ranged from 4 to 69% and aflatoxin content ranged from 2–1172 µg kg^{-1}. Three (ICGV 02226, 02229 and 02234) of the 13 elite Spanish bunch confectionery groundnut lines were resistant (<5 µg kg^{-1}). In Virginia bunch type, *A. flavus* infection and aflatoxin contamination ranged from 11 to 37% and 88 to 2103 µg kg^{-1}, respectively.

Farid Waliyar and SN Nigam

Activity 1.2.3: Develop regionally adapted parental lines of potential hybrids in sorghum, pearl millet and pigeonpea

Milestone: High-yielding male-sterile lines of sorghum with temperature insensitivity and resistance to grain mold and shoot fly developed (2007)

Race-specific and trait-based B-lines: In a program to develop high-yielding race-specific and grain mold and shoot fly resistant A/B-lines, several F_4 progenies with maintainer reaction were used for conversion into A-lines. These are in various stages of conversion.

New B–lines trial: The lines in advanced stages of conversion on A_1 and A_2 CMS systems (25 B on A_1 CMS + 2 controls; 22 B on A_2 CMS + 2 controls) were evaluated in a preliminary B-lines trial during the 2005 rainy season. The results revealed superiority of some of the A_1-based B-lines such as SP 2315 (3.9 t ha^{-1}) SP 2317 (3.8 t ha^{-1}) SP 2313 (3.7 t ha^{-1}) SP 2359 (3.7 t ha^{-1}) and SP 2305 (3.6 t ha^{-1}) over the control ICSB 52 (2.4 t ha^{-1}) with comparable maturity. These were numerically superior to another control 296B (2.8 t ha^{-1}) for grain yield. Similarly, several A_2-based B-lines such as SP 2785 (3.6 t ha^{-1}) SP 2895 (3.5 t ha^{-1}) SP 2859 (3.5 t ha^{-1}) SP 2853 (3.5 t ha^{-1}) SP 2779 (3.4t ha^{-1}) SP 2873 (3.3 t ha^{-1}) and SP 2783 (3.2 t ha^{-1}) were superior over the control ICSB 52 (2.0 tha^{-1}) with comparable maturity. These were also numerically superior to another control 296B (3.0 t ha^{-1}) for grain yield. Though none of the A_1-based B-lines were superior to the control ICSB 52 for grain size (3.1 g 100^{-1} grains), 15 B-lines with a grain size ranging from 2.4 to 2.7 g 100^{-1} grains were significantly superior to 296B (2.0 g 100^{-1} grains). Amongst the A_2-based B-lines, SP 2779 (3.0 g 100^{-1} grains) was significantly superior to the controls, ICSB 52 (2.7 g 100^{-1} grains) and 296B (2.1 g 100^{-1} grains) for grain size. Five B-lines (2.4 to 2.9 g 100^{-1} grains) were significantly superior to 296 B for grain size.

Elite A_1-system B-lines Trial (EBT): An EBT consisting of 14 high-yielding B-lines selected from the results of Advanced B-line Trial (conducted during the 2004 rainy season) was conducted during the 2005 rainy season. One of the test B-lines, ICSB 25005 (5.4 t ha^{-1}) was found to be exceptionally superior to the control 296B (2.1 t

ha^{-1}) for grain yield with comparable maturity. ICSB 25003 (3.4 t ha^{-1}) and ICSB 25002 (3.1 t ha^{-1}) were other B-lines, which outyielded the control 296B (2.1 t ha^{-1}). Four B-lines ICSB 25001 (2.8 g 100^{-1} grains), ICSB 25005 (2.5 g 100^{-1} grains), ICSB 25002 (2.4 g 100^{-1} grains) and ICSB 25003 (2.3 g 100^{-1} grains) were significantly superior to 296B (2.0 g 100^{-1} grains) for grain size.

Stability of male-sterile lines: Hybrid seed production with CMS lines is usually undertaken in August-November sowings. As a result, the flowering in the seed production plots coincides with temperatures as low as 10^{o}C during December–January and maximum temperatures as high as 45^{o}C during March–April. It is known that female sterility (due to stigma non-receptivity or inadequate pollen tube germination and its growth) leads to poor seed set at low temperatures. On the other hand, male sterility of seed parents often breaks down at high temperatures. Under such circumstances, the quality of hybrid seed deteriorates with large number of selfed seeds leading to lower commercial yields of hybrids. Therefore, information on the stability of male-sterility at high temperatures in the male-sterile lines is useful.

All the designated A-, B- and R-lines, developed at ICRISAT-Patancheru, are being characterized as per Distinctiveness, Uniformity and Stability (DUS) test guidelines in phases. A total of 277 A-lines that were characterized in the first phase were evaluated for stability of male sterility during 2005 summer season along with the controls 296A and 27A. The maximum temperature during the flowering period ranged from 40 to 43^{o}C. The stability of male-sterility was assessed using the criteria of seed set % under bagging. The results indicated that while most of the A-lines (204) showed no seed set, three A-lines (ICSB 40, ICSB 641 and ICSB 91001) showed 2 to 5% seed set and the remaining 70 lines showed above 5% seed set under bagging. The results indicate that male-sterility is a threshold trait requiring specific environment (ie, particular temperature regime) for complete expression. Also, expression of male-sterility depends on nuclear genetic background of A-lines. The lines which are sensitive to high temperature for the expression of male-sterility should be avoided for seed production during summer season in locations with high air temperature. All the designated A-lines that are being characterized in the second phase will also be evaluated for stability of male-sterility under high temperature during 2006 summer season.

BVS Reddy and S Ramesh

Milestone: High-yielding and DM resistant male-sterile lines of pearl millet developed and characterized (2007)

Seed parents development: Fully converted 5–9 A-lines in different cytoplasmic backgrounds and their counterpart B-lines selected for high yield potential, agronomic eliteness and high levels of DM resistance are designated every year for dissemination. Such a flow of A-/B-lines has been possible due to continuous addition of B-lines in the conversion program.

The 2005-series seed parents: Nine 2005-series A-lines having diverse nuclear and two cytoplasmic backgrounds (3 A_1 cytoplasms and 6 A_4 cytoplasms) were designated for distribution worldwide. All the lines were d_2 dwarf with a wide range of maturity (39–51 days to 50% flowering), panicle length (11–35 cm), panicle thickness (20–32 mm diameter), tillering ability (1–4 tiller plant^{-1}) and seed size (5.5–12 g 1000^{-1} seed mass). Important among these were ICMA 05555, a large-seeded (12 g 1000^{-1} seed mass compared to 10.9 g of 843B) A_1 CMS system male-sterile line, having thick-panicles (32 mm diameter compared to 23 mm of 843B), with maturity duration similar to that of 81B (50 days to 50% flower), and producing highest grain yield, 77% more than the control 81B (1355 kg ha^{-1}); ICMA 05111 and ICMA 05999 (both A_4 CMS) were high-yielding (16–51% more grain yield than 81B), high-tillering (about 4 tillers plant^{-1}, similar to 843B); ICMA 05888 (A_4) with long panicles (35 cm compared to 22 cm of 81B) producing 34% more yield than 81B; ICMA 05777 was early-maturing with 39 days to 50% flower (similar to 843B); ICMA 05222 and ICMA 05333 (both A_4) were high-yielding (about 25% more gain yield than 81B) and had thick panicles (30 mm diameter).

Eight of these male-sterile were highly resistant to DM with 0–10% incidence to at least three of the five diverse pathotypes (Jodhpur, Jalna, Jamnagar, Durgapura and Patancheru) under high disease pressure in the greenhouse condition (92–100% DM incidence in susceptible controls). Of these, ICMB 05333 was resistant to all five pathotypes, two to four pathotypes, five to three pathotypes, and one to two pathotypes.

A-/B-lines under backcrossing: Backcross conversion of 87 B-lines into 98 A-lines with different cytoplasmic backgrounds (29 A_1, 67 A_4 and 2 A_5) reached the advanced stage (BC_5 and beyond) with 32 candidate A/B pairs identified for 2006-series A-lines. We also evaluated 228 B-lines in early generation backcrossing and selected 168 (88 A_1, 106 A_4 and 61 A_5) for advancing to BC_2–BC_3. First backcross was made with 127 B-lines, of which 61 B-lines and their backcross progenies (34 A_1, 26 A_4 and 36 A_5) were selected for further backcrossing.

Conversion of 34 elite maintainers of A_1 CMS system into A_4-system A-lines was completed and third backcross of these maintainers was completed to convert them into A_5-system A-lines. Above-mentioned crosses clearly indicate gradual shift from A_1 system A-lines to A_4 and A_5 system A-lines.

Marker-assisted backcross breeding to pyramid DM resistance genes in ICMA 89111: Attempts to transfer DM resistant QTL from 863B into ICMB 89111, a high-tillering male-sterile line with moderate DM resistance have been underway to further improve its DM resistance. In a continuing effort to pyramid DM resistance genes in ICMB 89111, 257 BC_5F_3 progenies (mostly d_2 dwarf) were evaluated, of which 97 progenies were selected based on visual assessment for phenotypic resemblance to ICMB 89111. About 22% of the 97 selected progenies flowered in 46–50 days, while 73% flowered in 51–55 days (ICMB 89111-P_2 flowered in 52 days and ICMB 89111 flowered in 45 days).

Screening seed parents for multiple disease resistance: Six A/B pairs, earlier bred (between 1988 and 1993) for both DM and smut resistance, were field evaluated for smut incidence under high disease pressure in the artificially inoculated smut nursery at Patancheru (90–100% smut in controls 81A/B and 841A/B). Three of these were highly resistant to smut (<5% severity) and one of these (ICMA 92777) was also highly resistant to DM with <5% incidence against Durgapura and Patancheru pathotypes under high disease pressure in the greenhouse condition (63–96% DM incidence in susceptible controls). Others had smut incidence in the range of 27–40%. It is interesting that a selection within ICMA 92777 made by a private seed company is the seed parent of one of the most popular hybrids in India.

Characterization of hybrid parents for performance *per se*: The evaluation of grain yield and yield contributing traits of counterpart B-lines of most of the designated A-lines developed up to 2001 had been completed. Recently designated B-lines, especially those developed during 2002–04 and other lines missing in the earlier evaluations, were grouped into d_2 dwarf and medium height groups and evaluation was done in replicated trials during the rainy season 2004 and summer season 2005.

D_2 dwarf B-lines: Average grain yield of the trial having 26 d_2 dwarf B-lines (counterparts of the designated A-lines) over two seasons ranged from 1441 to 2787 kg ha^{-1} with the yield level during summer being 63% more than the rainy season (1649 kg ha^{-1}). The correlation of grain yield between two seasons was positive and significant (r = 0.60**), indicating that rankings of the B-line followed broadly similar pattern in both the seasons. Three B-lines (43 days to 50% flower) were early-maturing, similar to that of 843B (41 days to 50% flower) and produced 18–47% of higher grain yield than 843B (1441 kg ha^{-1}). All the 18 B-lines yielding 15–58% more grain than the control 81B across two seasons were also significantly superior either in one or both the seasons, and 15 of them flowered earlier than 81B (56 days 50% flower). Of these high-yielding 18 B-lines, 13 had larger grain size >10.0 g 1000^{-1} seed mass compared to 81B (7.0 g 1000^{-1} seed mass) and 3 had even larger grain size (12.3–14.0 g 1000^{-1} seed mass) than 843B (11.7 g 1000^{-1} seed mass).

Medium height B-lines: Of the 18 medium-height B-lines (counterparts of the designated A-lines) evaluated for two seasons for field performance, 8 lines had 2721–3163 kg ha^{-1} of mean grain yield (2636 kg ha^{-1} for ICMB 88004 used as control) and flowered in 47–56 days (45 days for ICMB 88004). From amongst 8 high-yielding lines, 6 had larger grain size (11.6–14.3 g 1000^{-1} seed mass) than ICMB 88004 (11.4 g 1000^{-1} seed mass).

Characterization of hybrid parents for DUS traits: In order to document the designated hybrid parents to prevent IPR infringement, designated A-lines and some of the important and widely distributed R-lines were characterized for DUS traits. Characterization of 99 A/B pairs (designated until 2004 series) for 26 DUS traits (that included 17 essential traits) was completed for two seasons. Additional 9 A/B pairs of 2005-series were also characterized during the 2005 rainy season. Characterization of 43 selected entries from ICRISAT pollinator collection (IPC lines) for the same number of traits was completed for two seasons. Additional, 41 IPC lines were also characterized during the 2005 rainy season. Characterization data, along with details will be published in the International Sorghum and Millet Newsletter, and also placed on the ICRISAT webpage.

Agronomic performance of F_1 seed parents: An earlier study had shown that a male-sterile F_1 (ICMA 95111 × ICMB 97444) derived from two morphologically similar but genetically diverse seed parents (DM resistant versions of 843B) had 22% higher grain yield than its high-yielding parental line. To assess the combining ability of male-sterile F_1 in comparison with the parental male sterile lines, 4 three-way hybrids made on a male-sterile F_1 with 4 restorers, their 8 single-cross hybrids made on the two parental male-sterile lines, and 4 single-cross hybrids made on 843A (control) were evaluated. Three-way hybrids either had practically similar or slightly lower grain yield levels than the respective single-cross hybrids. Phenotypically, three-way hybrids were

as good as single-crosses as indicated by the similar standard deviation values for days to flowering, plant height and panicle parameters. This is due to the morphologically similar parents of male-sterile F_1 and to some extent, masking effect of male parents. Thus, the male-sterile F_1s besides their higher seed yield in the hybrid seed production plots, provide a good avenue in the production of three-way hybrids that may have grain yield comparable to single-cross hybrids, and a mechanism for more effective resistance gene deployment.

KN Rai, VN Kulkarni and RP Thakur

Downy mildew resistance in B-lines: Twenty-four 2005 series B-lines were evaluated against five pathotypes – Jodhpur (Sg 139), Jalna (Sg 150), Jamnagar (Sg 200), Durgapura (Sg 212) and Patancheru (Sg 409) in a completely randomized design with two replications in greenhouse. Of the 24 lines, 16, 10, 14, 15 and 4 were resistant (≤10% incidence) to Jodhpur, Jalna, Jamnagar, Durgapura and Patancheru pathotypes, respectively relative to 96–100% incidence in the susceptible control 7042S. Four B-lines were resistant (≤10% incidence) to all five pathotypes, 2 lines to four pathotypes, 7 lines to three pathotypes and 3 lines to two pathotypes. The remaining 6 lines had differential disease incidence to all five pathotypes. The 4 B-lines that were resistant to all five pathotypes could be used in resistance breeding program as seed parents.

RP Thakur and KN Rai

2006-series restorers: Restorer line development envisages designating 5 to 9 restorers every year similar to the approach followed for the A-/B-lines. Sixty-three A_4 restorer progenies and 64 A_1 restorer progenies were identified that are being further evaluated for fertility restoration, agronomic eliteness and DM resistance to designate them during 2006.

Backcross breeding to develop restorers of A_4 and A_5 CMS systems: Several A_4-system male-sterile lines have been developed and disseminated, and many more can be rapidly developed. The development of A-lines with A_5 cytoplasm can be even faster. The utility of these A-lines in hybrid development, however, is considerably constrained by the lack of their restorers. We have been converting 49 elite inbreds (32 from ICRISAT, 15 from six national program institutions, and one each from two private seed companies) into A_4 and A_5 restorer versions. Fourteen A_1-system R-lines have now been converted into A_4-system R-lines, and 39 A_1-system R-lines into A_5-system R-lines.

Population breeding of restorers of A_4 CMS systems: Identification of A_4 restorer progenies from those derived from the populations possessing moderate levels of A_4 restorers has been pursued as one of the objectives, leading to identification of 68 progenies during previous year, based on visually assessed grain yield potential and agronomic traits. These potential A_4 restorers were testcrossed on different A_4-system A-lines (3–9) to test their fertility restoration in different genetic backgrounds. Testcross data on fertility/sterility reaction confirmed 19 of these being A_4 restorers (flowering in 51–61 days compared to 51 days for ICMP 451). More than 60 progenies were derived from these 19 selected progenies, which were crossed to 6 A_4 system A-lines to reconfirm their fertility reaction before designating them. These progenies were also evaluated for DM resistance against Durgapura and Jalna pathotypes under high disease pressure in greenhouse condition (90–100% in susceptible controls). Of these, 34% were resistant (0–10% DM incidence) to Durgapura pathotype and 16% were resistant to Jalna pathotype.

Hybrid performance of seed parents and restorer parents: Although ICRISAT focuses its majority of efforts on developing hybrid parents, some of the designated parental lines are also evaluated for their hybrid yield potential. This activity is expected to speed up the hybrid breeding process by zeroing down to few parents or to the progenies with the parents of successful hybrids in the pedigree. While these hybrid evaluation trials aid in replacing existing hybrid parents, these also serve as channel for cost-effective breeding of high-yielding hybrids.

Combining ability of seed parents and potential restorers: A set of 11 A-lines (6 A_1, 4 A_4 and 1 A_5) that included 4 established A-lines (81A, 843A, 863A and ICMA 88004) were crossed with 10 potential restorer progenies previously selected for their grain yield potential and agronomic traits in a line × tester design for estimation of combining ability for grain yield and yield-related traits. The crosses (110) and parents were planted in separate but adjoining blocks, replicated two times in 1-row plots of 5 m length. From amongst the A-lines, 6 had positive (though not significant) general combining ability (gca) effects for grain yield. Of these, four were new lines and two of these long panicle A-lines (ICMA 04111 and ICMA 04777) had significant positive gca effects for panicle length and days to 50% flowering. These lines have been developed as improvement over 81A. The other two high-tillering lines had significant positive gca effect for tillering significant negative gca effect for days to 50% flower. Two potential restorers had positive significant gca effect for grain yield, along with another four having positive gca effect (though not significant). Among these 6 A-

lines, 2 had negative significant gca effects for days to flower and 2 had positive significant gca for 1000-seed mass.

Among the 110 hybrids, 14 hybrids had at least 5% more grain yield than the highest-yielding control 7688 (4980 kg ha^{-1}). Four hybrids involved potential R-line ICMS 77004-S1-52-3-1-2-1-2-1 and 4 hybrids involved 81A as female parent. Of these 14 hybrids, 12 flowered in 44-47 days (46 days for 7688 and 45 days for PB 106). All the high-yielding hybrids possessed large grain size (10.6-13.5 g 1000^{-1} seed mass) than the control 7688 (10.4 g 1000^{-1} seed mass). As the results are based on un-replicated one-row plots, these hybrids should be evaluated in different locations to confirm their grain yield potential.

Hybrids observation nursery: About 308 hybrids involving 23 A-lines of different plant types (81A, 863A, ICMA 88004 and ICMA 89111 types) and 20 potential restorers were planted in an observation nursery in un-replicated one-row plots. Hybrids were visually assessed for grain yield potential and other agronomic traits (scores 1 = poor and 5 = best). Both A-lines and potential restorers that were involved in production of more than one high-yielding hybrids were identified. Two high-tillering A-lines were involved in producing 4 or more high-yielding hybrids (ICMA 02111 involved in 5 hybrids and ICMA 04999 involved in 4 hybrids). Three 863A type male-sterile lines along with 863A itself, and ICMA 94111 (ICMA 88004 type) and ICMA 04777 (81A type) were involved in producing 2 high-yielding hybrids each. From amongst 7 potential restorers involved in production of at least 2 high-yielding hybrids, a thick and long panicle potential restorer produced 10 high-yielding hybrids, followed by a MRC-derived progeny and ICMS 7704-derived progeny, each involved in 4 hybrids each. These hybrid parents were identified on the basis of limited testing of hybrids that needs to be evaluated systematically in larger plot size in multilocation trials.

Performance of hybrids with probable adaptation to arid Rajasthan, India: Early maturity and high-tillering are the two traits considered important in breeding hybrids adapted to arid Rajasthan. Hence, breeding hybrid parents mostly revolves around basic phenotypes of the commercial hybrids grown in the region. Two early-maturing male-sterile lines with high-tillering (similar to 843A) and 843A itself were crossed to twenty-two H 77/833-2 type MRC-derived advanced generation progenies and these hybrids were evaluated at Patancheru in an observation nursery with HHB 67, HHB 67-2 and ICMR 356 as controls. Grain yield was recorded in un-replicated one-row plots. About 25 high-yielding hybrids were identified. Of these, five hybrid which had maturity similar to HHB 67 and HHB 67-2 (38–39 days to 50% flower), outyielded HHB 67 by 33–41% and HHB 67–2 by 24–32%. Additional 12 hybrids (41–43 days to 50% flower) that had maturity similar to that of ICMH 356 (41 day to 50% flower) outyielded the latter by 7–13%. Most of these hybrids were either uniformly fertile or segregated for fertile (F) and sterile (S) plants. Twelve hybrids were based on ICMA 96111 and 10 hybrids on ICMA 03666. Similarly, 4 hybrids each were based on the MRC progenies. These hybrids need to be evaluated in arid Rajasthan for confirmation of their yield potential and adaptation.

KN Rai, VN Kulkarni and RP Thakur

Milestone: High-yielding and disease resistant dual-restorers of sorghum and pearl millet developed (Annual)

Restorer progenies adapted to rainy season: A total of 457 F_2s derived from 1560 crosses included in several elite R-line × R-line half-diallel crosses were evaluated during the 2005 rainy season and 605 F_3s were selected based on grain yield potential and grain size on visual observation basis. Besides these, a total of 900 F_5 progenies derived from various crosses and their testcrosses were evaluated during the 2005 rainy season. Several F_6 selections with male-fertility restoration reaction were made for the following traits on A_1: brown midrib - 74, sweet-stalk - 11, high-yielding - 5, pop sorghum - 2, lustrous grain - 12, waxy leaf - 1; on A2: brown midrib - 48, sweet-stalk - 23, high grain yield - 2 and high forage yield - 18.

Restorer progenies adapted to postrainy season: From the 306 F_3s derived from R- × R-crosses that were evaluated during the 2004–05 postrainy season, 140 F_4s were selected. These will be advanced through selection and testcrossed on A_1 and A_2 during the 2005–06 postrainy season.

Restorer lines (R-lines): The advanced breeding lines with male-fertility restoration reaction on A_1 CMS system were evaluated in replicated yield trials. Initially, the lines are evaluated in Preliminary R-lines Trial (PRT) and those found promising are evaluated in Advanced R-lines Trial (ART). The superior lines from ART are evaluated in Elite R-line Trial (ERT). The promising lines with good plant type (tall stature with tan plant color and longer panicles with white bold grains) derived from various programs are tested in Preliminary Varietal Trial (PVT) and Advanced Varietal Trial (AVT). The results of these trials are summarized below.

PRT: None of the 22 test R-lines in PRT was significantly superior to the controls RS 29 and ICSR 89058 for grain yield. However, some of the test R-lines such as SP 3821 (3.7 t ha^{-1}), SP 3750 (3.5 t ha^{-1}), SP 3822 (3.4t ha^{-1}), SP 3646 (3.4 t ha^{-1}) and SP 3647 (3.3 t ha^{-1}) were comparable to the controls, RS 29 (3.3 t ha^{-1}) and ICSR 89058 (3.1 t ha^{-1}) for grain yield. These R-lines (2.0 to 2.2 g 100^{-1} grains) were comparable to the controls RS 29 (2.1 g 100^{-1} grains) and ICSR 89058 (2.0 g 100^{-1} grains) for grain size. These will enhance in diversifying the R-line gene pool as these are derived from diverse parentage.

ART: The promising R-lines (70) derived from various trait-specific groups were evaluated in ART. Seventeen R-lines significantly out performed (grain yield ranging from 4.0 t ha^{-1} to 3.4 t ha^{-1}) the control RS 29 (2.3 t ha^{-1}) for grain yield with comparable maturity. Two of the test R-lines, SP 6637 (4.0 t ha^{-1}) and SP 6617 (3.9 t ha^{-1}) were significantly superior to the control ICSR 89058 (2.7 t ha^{-1}) and several others (grain yield ranging from 3.6 t ha^{-1} to 2.6 t ha^{-1}) were on par with control ICSR 89058 (2.7 t ha^{-1}). One test R-line SP 6660 (2.4 g 100^{-1} grains) had significantly larger grains than the controls RS 29 (2.1 g 100^{-1} grains) and ICSR 89058 (2.1 g 100^{-1} grains). These 17 lines were designated in 2005.

ERT: None of the test R-lines were significantly superior to the controls RS 29 (2.8 t ha^{-1}) and ICSR 89058 (3.7 t ha^{-1}) for grain yield. Nevertheless, three of the seven test R-lines, ICSR 24002 (3.2 t ha^{-1}), ICSR 24005 (3.2 t ha^{-1}) and ICSR 24009 (3.1 t ha^{-1}) were on par with the control RS 29 (2.8 t ha^{-1}) and were of comparable maturity. ICSR 24007 (2.9 g 100^{-1} grains) had significantly larger grains than the controls RS 29 (2.2 g 100^{-1} grains) and ICSR 89058 (2.3 g 100^{-1} grains). The grain size of some of the R-lines, ICSR 24002 (2.4 g 100^{-1} grains), ICSR 24005 (2.3 g 100^{-1} grains), ICSR 24009 (2.2 g 100^{-1} grains) and ICSR 24008 (2.5g 100^{-1} grains) were comparable to those of controls, RS 29 (2.2 g 100^{-1} grains) and ICSR 89058 (2.3 g 100^{-1} grains). These were designated in 2004 as they are expected to contribute to diversity.

PVT: None of the test varieties significantly outyielded the control varieties. Nevertheless, one test variety SP 5116 (3.1 t ha^{-1}) was comparable with the controls JJ 1041 (3.2 t ha^{-1}) and CSV 15 (3.5 t ha^{-1}) with similar maturity period. The varieties such as SP 5116 (2.4 g 100^{-1} grains), SP 5110 (2.9 g 100^{-1} grains) SP 5106 (2.7 g 100^{-1} grains) SP 5109 (3.0 g 100^{-1} grains) and SP 5127 (3.1 g 100^{-1} grains) had significantly larger grains than those of the controls JJ 1041 (2.1 g 100^{-1} grains) and CSV 15 (2.1 g 100^{-1} grains).

AVT: Though none of the test varieties out performed the control varieties JJ 1041 and CSV 15, some of the varieties such as ICSV 24022 (3.7 t ha^{-1}), ICSV 24001(3.6 t ha^{-1}), ICSV 24010 (3.5 t ha^{-1}), ICSV 24023 (3.5 t ha^{-1}) and ICSV 24012 (3.4 t ha^{-1}) were numerically superior to the control JJ 1041 (3.3 t ha^{-1}). All these, except ICSV 24001 had significantly larger grains (2.9 to 2.9 g 100^{-1} grains) than those of the controls JJ 1041 (2.1 g 100^{-1} grains) and CSV 15 (2.1 g 100^{-1} grains). These were designated in 2004. The selected lines will be evaluated for foliar disease resistance.

BVS Reddy and S Ramesh

Evaluation of new and diverse 2005-series pearl millet R-lines against multiple pathotypes of downy mildew: Downy mildew resistance in R-lines (as in A- and B-lines) is important to breed resistant hybrids. A total of 734 restorer progenies including 416 potential R-lines and 318 elite R-lines were screened in greenhouse against Durgapura (Sg 212) and Jalna (Sg 150) pathotypes in an unreplicated single pot with 35–40 seedlings/pot/line. Of these, 12% lines were disease free and 14% lines had 1–10% incidence to Durgapura pathotype; 17% lines were disease free and 16% lines had 1–10% incidence) to Jalna pathotype; and 7% to both Durgapura and Jalna pathotypes.

Of the 713 advanced progenies including 363 long panicle progenies (F_{5-8}), 162 S_{3-4} Jhakrana × ESRC, and 188 RCB2 (S_{2-8}) screened against Durgapura pathotype in the same manner as mentioned above, 19% progenies were disease free and 21% had 1–10% disease incidence. Similarly, 134 progenies from the GB 8735 screened against Jalna pathotype, 7% were disease free and 25% had 1–10% incidence.

A total of 374 R-lines, including 298 elite restorers and 76 A_4 restorers were screened in the downy mildew disease nursery in a single replication (1 row of 4 m long). Of these, 32% lines were disease free and 22% had 1–10% incidence at the soft dough stage.

RP Thakur and KN Rai

Milestone: Stable and diversified male-sterile lines and their restorers with resistance to wilt and sterility mosaic diseases developed in pigeonpea (2006)

Search for new fertility restorers and male-sterility maintainers: In any dynamic hybrid program, significant genetic diversity among R- and A-lines is essential and to achieve this, we evaluated 282 new hybrid combinations with A_4 cytoplasm. Among these, 271 hybrids were found to restore pollen fertility and only 11 maintained male-sterility. This confirms the previous year's observation of high frequency (96%) of fertility restoration in A_4 cytoplasm. All the male-sterile F_1s combinations were backcrossed to their recurrent patents for generating BC_1F_1 progenies.

New hybrid combinations: To identify high-yielding hybrids and new male-sterility maintainers, an attempt was made to develop new hybrid combinations. With the experience gained in the past 2–3 years, we decided to give emphasis to only A_4 hybrids. A total of 397 F_1 hybrids were made with hand pollination. This included 232 short-duration, 164 medium-duration and one long-duration hybrids. Only 42 new combinations were tried with the A-lines of A_2 cytoplasm. A_1 cytoplasm was used only in 20 short-duration combinations.

KB Saxena

Evaluation of CMS lines and their restorers for SM and wilt resistance: By following standard field screening methods, 122 pigeonpea cytoplasmic male-sterile lines and their restorers were evaluated for SM and wilt resistance in wilt and SM nursery. One line CMS 99044 was asymptomatic (0%) and 9 lines had <10% incidence for both wilt and SM diseases. Additional two lines ICPA 2014 and ICPA 2037 were asymptomatic for wilt and 18 lines were asymptomatic for SM.
S Pande and KB Saxena

Milestone: Promising pigeonpea hybrids in different maturity groups with resistance to wilt and sterility mosaic identified and hybrid seed production technology developed (2006)

Evaluation of F_1 hybrids: During 2005 rainy season, a total of 287 experimental hybrids were evaluated in 30 trials. Of these, 282 hybrids were developed on A-lines with A_4 cytoplasm. Of the 31 short-duration hybrids evaluated, 11 were found promising with yield advantage over the best control UPAS 120 ranging between 31 to 207%. ICPH 3310 recorded the highest yield of 4580 kg ha^{-1}. Among the Maruti-maturity group (160 days) hybrids, ICPH 2733 (71% superiority), ICPH 3366 (62% superiority), and ICPH 2671 (59% superiority) were outstanding. In the Asha maturity group (180–200 days) the highest yield of 3364 kg ha^{-1} (102% superiority) was recorded by ICPH 2741. The other promising hybrids in this group were ICPH 3489, ICPH 3479, ICPH 3401, ICPH 2786, and ICPH 3464. Twenty-four hybrids were also resistant to both wilt and sterility mosaic. These hybrids will be re-evaluated for their yield performance and disease resistance.

KB Saxena

Evaluation of hybrids and their parents for resistance to wilt and SM: Four hundred and eleven hybrids and their parents were evaluated for SM and wilt resistance following standard field screening technique. Of these, 33 hybrids and advanced breeding lines (ICPH 2319, ICPH 2897, ICPH 2899, ICPH 2900, ICPH 2326, ICPH 2327, ICPH 2336, ICPH 2897, ICPH 2898, ICPH 2899, ICPH 2900, ICPH 2352, ICPH 2911, ICPH 2913, ICPH 2914, ICPH 2915, ICPH 2916, ICPH 2903, ICEA P00020, GUPH 1126-4, IPH 487-2, ICP 11174, ICP 11376, ICPL 99050, ICPL 96053, ICPL 96058, ICPL 96052, ICPL 87119, ICPL 20125, ICPL 20094, ICPL 20096, ICPL 20098 and ICPL 20099) were symptomatic to both SM and wilt, while 28 were resistant to both diseases (<10% SM and wilt diseases). Additionally, seven lines (ICP 15045, ICP 11376-5, ICP 8863, ICPL 87119, ICPL 20107, ICPL 99048 and ICPL 20128) were asymptomatic, while 13 were resistant (<10% to wilt). Similarly, 69 entries were asymptomatic to SM, while 30 had <10% SM incidence.

S Pande and KB Saxena

Activity 1.2.4: Conduct strategic research to improve the efficiency of genetic enhancement

Milestone: Breeding methods of producing sorghum hybrids with sweet stalk and resistance to grain mold/shoot fly developed (2007)

Evaluation of sorghum hybrids for grain mold resistance: To develop grain mold resistant hybrids and to study the association of various agronomic and morphological traits (days to 50% flowering, plant height, panicle shape, glumes cover, glume color and grain color) to grain mold resistance, 168 F_1s were developed by crossing 8 A-lines (ICSA 369, -370, -371, -400, -384, -382, -52, -101) and 21 testers (IS 41720, -41397, -41675, -18758C-618-2, -18758C-618-3, -30469C-140-2, -30469-1508-2, -84, ICSV 96105, -96094, SPV 462, ICSR

89013, -91011, -89018, -89058, -92001, -91019, -91029, PVK 801, GD 65055, -65028) in the 2004-2005 postrainy season. These 168 hybrids along with their parents and controls (Bulk Y, IS 25017, IS 20, IS 14384, PVK 801, CSH 16) were screened for grain mold resistance in 2 replications (2 row-plots of 4 m long/ replication) in a RCB design at ICRISAT-Patancheru in the 2005 rainy season. Grain mold screening was done under field conditions using overhead sprinkler irrigation from flowering to physiological maturity. Grain mold severity (PGMR) was recorded on 10 uniformly flowered-tagged plants in each plot using a 1–9 scale at physiological maturity. Mold scoring was also done on threshed bulk grain (TGMR) from the tagged panicles using the same 1–9 scale.

Based on grain mold severity rating of 1–4 as resistant and 4.1 to 9 as susceptible the entries were classified into these two groups. Both PGMR and TGMR scores on susceptible controls (SPV 104, CSH 16, Bulk Y) were >7.0, while resistant controls (IS 25017, IS 20 and IS 14384) showed ratings of 1.0. The hybrids were classified into four categories according to resistance (R) and susceptibility (S) of parental lines as R × R, R × S, S × R and S × S. All the 24 hybrids of R × R cross were resistant (≤4.0 score) both for PGMR and TGMR; of 60 hybrids of R × S cross 42 were resistant; of 24 hybrids of S ×R cross 16 were resistant and of 60 hybrids of S × S cross 13 were resistant. These results confirm our earlier findings that there is high probability of producing grain mold resistant hybrids when both parents are resistant, and low probability when both parents are susceptible. Plant height, grain and glumes color do contribute to resistance due to barrier to infection by mold fungi.

Biochemical analyses for ergosterol and flavan-4-ols were carried out for 25 hybrids (15 resistant and 10 susceptible) and their parental lines. Susceptible hybrids showed higher level of ergosterol (20.6 µg g^{-1}) than the resistant hybrids (8.7 µg g^{-1}) both in white- and red-grain backgrounds. Similarly, susceptible parental lines showed higher amounts of ergosterol than the resistant lines. In contrast to ergosterol, higher levels of flavan-4-ols were found in resistant hybrids ($2.04A_{550}$ g^{-1}) than in susceptible hybrids ($1.31A_{550}$ g^{-1}), and red-grain hybrids had more flavan-4-ols than the white-grain. Similarly, resistant parents showed higher level of flavan-4-ols ($5.27A_{550}$ g^{-1}) than the susceptible ones ($0.66A_{550}$ g^{-1}).

RP Thakur and BVS Reddy

Milestone: Character association and breeding efficiency of alternate CMS systems in sorghum and pearl millet quantified (2006)

The need for cytoplasmic diversification of A-lines (and hybrids) to mitigate the potential risk of unforeseen disease and insect pest outbreaks associated with cytoplasmic uniformity of cytoplasmic-nuclear male sterility (CMS)-based hybrids is a common knowledge. Cytoplasmic diversification also enhances the opportunities for diversifying the nuclear genetic base of A-lines as some of the outstanding restorers of one cytoplasm are found to be maintainers of other cytoplasms. However, in pursuit of diversifying the CMS base of hybrid seed parents, and hence the hybrids, the performance of hybrid seed parents and the hybrids based on alternative CMS systems for grain yield and other agronomic traits of importance cannot be compromised. Therefore, a series of studies were made to assess the efficiency of A_2 CMS system in comparison to the widely used A_1 CMS system in terms of mean performance, combining ability and heterosis for grain yield and other agronomic traits and responses to shoot fly infestation and grain mold infection at ICRISAT-Patancheru.

Grain yield and other agronomic traits: The studies on the evaluation of A_1 and A_2-based two sets of 18 isonuclear hybrids each during 2001 and 2002 rainy seasons and 2002–03 and 2003–04 postrainy seasons at ICRISAT-Patancheru revealed that A_2 CMS system is as efficient as A_1 (with a slight edge of A_2 over A_1) for commercial exploitation in terms of the development of heterotic hybrids for both rainy and postrainy season adaptation.

BVS Reddy and S Ramesh

Responses to shoot fly infestation: Two sets each of six isonuclear, alloplasmic A-lines with A_1 and A_2 cytoplasms in six different nuclear genetic backgrounds (Set 1 consisted of lines ICSB17, ICSB 37, ICSB 38, ICSB 42, ICSB 88001 and ICSB 88005 and Set 2 consisted of lines ICSB 11, ICSB 26, ICSB 88004, ICSB 18757, PM 17467B and PM 7061B) were crossed with three dual-restorers (ICSR 93001, ICSR 92003 and ICSR 93031) to produce two sets of 36 hybrids each. The two sets of hybrids along with the parents were screened (for the second time) for their responses to shoot fly infestation using infestor-row technique during 2005 rainy season at ICRISAT- Patancheru in a split-split plot design in three replications. The percentage of hybrids and the parents showing deadheart (DH) symptoms at 21 days after sowing (DAS) as a response to shoot fly infestation was estimated. As mean squares due to interaction of A-lines and R-lines with year and cytoplasm were non-significant, the *gca* effects of A-lines and mean performance and *sca* effects of hybrids in

A_1 and A_2 cytoplasm backgrounds for mean DH% were estimated based on the combined data over 2004 and 2005 rainy seasons and the results are discussed below.

CMS effects on *gca* effects of A-lines: Significant CMS effects on *gca* of A-lines for shoot fly deadheart % were observed in only one nuclear genetic background (ICSA 88005) in set I and in two nuclear genetic backgrounds (ICSA 26 and ICSA 18757) in set II, and there were no definite trends favoring any particular CMS system. While the A-line ICSA 88005 in A_1 CMS system was a better general combiner compared to that in A_2 CMS system; the A-lines, ICSA 26 and ICSA 18757 in A_2 CMS system were better general combiners compared to those in A_1 CMS system.

CMS effects on *per se* performance and *sca* effects of crosses: As is true for *gca* effects, significant CMS effects on mean deadheart % of crosses were observed only in two nuclear genetic backgrounds both in set I (ICSA 17 × ICSR 93001 and ICSA 88005 × ICSR 93001) and Set II (ICSA 26 × ICSR 93001 and ICSA 18757 × ICSR 93031) with no definite trend favoring any particular CMS system. As far as *sca* effects were concerned, significant CMS effects were noticed only in one nuclear genetic background (ICSA 88005 × ICSR 93001) in set I. The *sca* effects of A_1 and A_2 CMS-based crosses were comparable in all nuclear genetic backgrounds in set II. The results clearly indicated that, by and large, A_1 and A_2 CMS-based A-lines as well as crosses were comparable for responses to shoot fly deadhearts %. Where significant CMS effects on *gca* and *sca* effects for shoot fly deadhearts % were detected, the magnitude varied with the nuclear genetic background with no definite trend favoring any particular CMS system.

Thus, considering that A_1 and A_2-based hybrids are comparable for grain yield potential, grain traits, maturity, and for responses to shoot fly, A_2 offers immediate option for the required diversification of CMS base of hybrid parents and hence hybrids to prevent eventual risk associated with the use of single cytoplasm (A_1-based hybrids) to stabilize the yield potential of hybrids.

BVS Reddy, HC Sharma and S Ramesh

Responses to grain mold infection: The two sets of isonuclear, alloplasmic hybrids (which were tested for responses to shoot fly infestation) along with the parents were evaluated (for second time) for their response to grain mold infection under sprinkler irrigation during 2005 rainy season at ICRISAT-Patancheru in a split-split-plot design in three replications. The hybrids and the parents were scored for average panicle grain mold rating (PGMR) taken on 10 panicles using a 1 to 9 scale, where 1 = no mold, 2 = 1–5%, 3 = 6–10%, 4 = 11–20%, 5 = 21–30%, 6 = 31–40%, 7 = 41–50%, 8 = 51–75%, 9 = >75% panicle surface area colonized by grain mold fungi. The *gca* effects of A-lines and mean performance and *sca* effects of crosses in A_1 and A_2 cytoplasm backgrounds for mean PGMR scores were estimated based on the combined data of 2004 and 2005 rainy seasons and the results are discussed below.

CMS effects on combining ability and mean performance: Cytoplasms seldom had any influence either on *gca* effects of A-lines or *sca* effects of hybrid combinations for PGMR in any of the nuclear genetic backgrounds. However, significant differences between individual A_1 and A_2 cytoplasm-based hybrids for mean PGMR severity scores were observed in two nuclear genetic backgrounds (PM 17467A × ICSR 93001 and ICSA 11 × ICSR 92003) in set II for mean PGMR severity scores, though no definite pattern of association of PGMR severity scores with a particular cytoplasm was observed. For example, while A_1 cytoplasm-based hybrid (PM 17467A × ICSR 93001) showed significantly lower PGMR scores than those based on A_2 cytoplasm, A_2 cytoplasm-based hybrid (ICSA 11 × ICSR 92003) showed significantly lower PGMR scores than those based on A_1 cytoplasm. However, when mean PGMR scores over all the hybrids were considered, there were no differences between A_1 and A_2 cytoplasm-based hybrids.

Thus, considering that A_1 and A_2-based hybrids are comparable for grain yield potential and grain traits, and maturity, and for responses to grain mold, A_2 offers immediate option for the required diversification of CMS base of hybrid parents and hence hybrids to prevent any potential risk associated with the use of single cytoplasm (A_1)-based hybrids to stabilize the yield potential of hybrids.

BVS Reddy, RP Thakur and S Ramesh

Breeding efficiency of male-sterile cytoplasms (A_1 and A_2) *vs.* fertile (B-line) cytoplasm in hybrid combinations: Two sets each of 36 isonuclear (A × R) hybrids (36 in A_1 and 36 in A_2 CMS backgrounds) were made by crossing isonuclear, alloplasmic (A_1 and A_2) A-lines in 12 nuclear genetic backgrounds with three dual restorer (R)-lines. The male-fertile counterparts of the 12 male-sterile lines were emasculated and crossed with the same three dual R-lines and produced 36 B × R hybrids. The two sets of 36 A × R and one set of 36 B × R crosses were evaluated at ICRISAT-Patancheru during 2005 rainy season in split-split-plot design using three

replications with R-lines in main plots, A-lines as sub-plots and cytoplasms in sub-sub-plots. The 12 A-lines and their B-lines were evaluated in a separate trial using randomized complete block design with three replications. Sufficient care was taken for adequate supply of pollen grains to A-lines for meaningful comparison of yield performance of A-lines vs B-lines.

The comparison of A × R and B × R crosses (in both A_1 and A_2 backgrounds) indicated that, while there were no differences between A × R and B × R crosses for days to 50% flowering, A × R (both A_1 and A_2) crosses were significantly taller (by 0.2m in A_1 and by 0.1m in A_2 backgrounds) and manifested higher grain yield (by 0.7 t ha^{-1} in A_1 and by 0.9 t ha^{-1} in A_2 backgrounds) compared to those of B × R crosses when average performance of A_1 and A_2-based A × R and B × R hybrids as separate groups was considered. However, when grain size was considered, A × R (in A_1 background) crosses were significantly bolder (by 0.08 g 100^{-1} seed) than B × R crosses, while in A_2 background there were no differences between A × R and B × R crosses.

Significant cytoplasm effects were observed for all the traits except grain size when individual nuclear genetic background of A × R (both A_1 and A_2) and B × R crosses were examined. Where significant cytoplasm effects were detected in some of the nuclear genetic backgrounds, not only the magnitudes of differences between A × R and B × R crosses varied with nuclear genetic backgrounds, but also were small to have any practical importance for days to 50% flowering, plant height and grain size. Also, there was no definite trend favoring either A × R or B × R crosses for any trait, ie, while A × R crosses, besides being early, were taller and possessed larger grains compared to those of B × R crosses in a few nuclear genetic backgrounds, the reverse was true in few other nuclear genetic backgrounds. However, in most of the nuclear genetic backgrounds, A × R (both A_1 and A_2) crosses were significantly superior to B × R crosses for grain yield. Thus, the use of CMS-based hybrids for commercial exploitation of heterosis is not only justified by the feasibility of large and economy-scale hybrid seed production, but also for superior grain yield.

BVS Reddy and S Ramesh

Breeding efficiency of male-sterile cytoplasms [A_1, A_2, A_3, A_4 (M), A_4 (VZM) and A_4 (G)] vs fertile (B-line) cytoplasm: The isonuclear alloplasmic A-lines in eight nuclear genetic backgrounds (ICSA/B 11, ICSA/B 17, ICSA/B 26, ICSA/B 37, ICSA/B 38, ICSA/B 42, ICSA/B 88001 and ICSA/B 88004) along with their B-lines were evaluated in a randomized complete block design with three replications at ICRISAT-Patancheru during 2005 rainy season. Precautions were taken for the sufficient supply of pollen grains to A-lines for meaningful assessment of A-lines vs their B-lines for grain yield.

The analysis of variance indicated significant variability among the A-lines for days to 50% flowering, plant height and grain yield. While there were significant differences among male-sterility-inducing cytoplasms and between male-sterility inducing cytoplasms and their B-lines for all the traits, the magnitudes of differences for days to 50% flowering and plant height were small to have any practical significance. Also, there was no definite trend in association of days to 50% flowering and plant height with either sterile or fertile cytoplasm. In general, the grain yield potential of A_1- and A_2-based A-lines in most of the nuclear genetic backgrounds was comparable (which is advantageous from hybrid seed production point of view) to that of B-lines. However, grain yield potential of other cytoplasms [A_3, A_4 (M), A_4 (VZM), and A_4 (G)]-based A-lines were significantly lower than that of A_1 and A_2-based A-lines as well as B-lines. For grain size, A-lines based on all the six cytoplasms comparable to B-lines.

BVS Reddy and S Ramesh

Effect of A_1 and A_2 cytoplasm on grain mold resistance: To understand the effects of different cytoplasm on mold resistance, a set of 72 hybrids were made by crossing A_1 and A_2 CMS lines in the genetic backgrounds of 12 B-lines (ICSB 17, -37, -38, -42, -88001, -88005, ICSB-11, -26, -88004, ISB 18757, PM 17467 and PM 7061B) with three R-lines (ICSR 93001, ICSR 92003 and ICSR 93031). Two experiments, each consisting of 36 hybrids, 9 parental lines and 4 controls were conducted in a RCB design with two replications. Each entry was planted in 2-row plots of 4 m long. Overhead sprinkler irrigation was provided on rain-free days for the mold infection and development. Panicle gain mold rating (PGMR) was recorded using a 1–9 scale (1 = no mold and 9 = >75% molded grains on a panicle) on the 10-tagged panicles per plot and threshed grain mold rating (TGMR) was recorded on the bulk grain from the tagged panicles per plot. These experiments were conducted in 2004 and repeated during the 2005 rainy season for further confirmation of the earlier results.

The results showed that all the 12 B-lines were susceptible (>4.0 score) with the mean PGMR and TGMR scores ranging from 5.0 (ICSB 11) to 9.0 (ICSB 42). Of the three R-lines, only ICSR 93001 was resistant and the other two had severity scores of 6.0 to 7.5 compared to score 8.3 of the susceptible control (Bulk Y) and score 1.0 of the resistant control line (IS 14384). In experiment 1, all the 36 hybrids were susceptible with grain mold ratings

ranging from 5.0 to 9.0. In experiment 2, 29 of the 36 hybrids were resistant and the remaining 7 susceptible without indicating any pattern. Thus, in both experiments, there were no clear effects of A_1 and A_2 cytoplasm on grain mold severity ratings of their hybrids. These results are similar to those obtained in 2004.

RP Thakur and BVS Reddy

CMS effect on grain yield: Continued efforts in diversification of cytoplasmic-nuclear male-sterility in pearl millet have lead to the identification of more stable CMS systems such as A_4 (0.0–0.3% pollen shedders) and A_5 (no pollen shedders), compared to A_1 CMS systems (0.0–2.5% pollen shedder). These new CMS systems also have higher maintainer frequency in both African and ICRISAT-bred improved populations and hence, provide great opportunities for genetic diversification of A-lines Evaluation of isonuclear hybrids in six environments (2 year × 3 locations) had already shown that the hybrids with the A_4 cytoplasm produce only 5% less grain yield than A_1-hybrids. We evaluated 45 hybrids developed by crossing two isonuclear A-lines ($A_1$and A_5 cytoplasm) and their maintainers (fertile cytoplasm) in each of the three genetic backgrounds (81B, 5054B and ICMB 88004) as female parents with five dual-restorers as male parents. A replicated trial of these 45 hybrids was conducted at two locations (ICRISAT-Patancheru, Millet Research Station, Jamnagar, India) for two years. The results showed that there was no significant difference between the hybrids of A_1 and A_5 CMS systems, with the A_5-hybrids giving 97–102% mean grain yield of the A_1-system hybrids. Also, the hybrids of the two CMS systems were very similar with respect to plant height, time to flower, panicle length and tillering. This indicated that A_5 cytoplasm has no adverse effect on grain yield and other important agronomic traits.

Bi-directional recurrent selection for maintainer and restorer frequencies: A bi-directional recurrent selection for fertility and sterility reactions of the A_1 and A_4 CMS systems was conducted for five cycles in Early Smut Resistant Composite II (ESRC II) and for three cycles in the OPV Raj 171 to assess the selection response of maintainer and restorer frequencies in two populations. Male-sterile lines $81A_1$ and $81A_4$ were used as testers to evaluate fertility restoration and sterility maintenance properties of plants of these populations in their testcrosses during the selection process. Recurrent selection bulks of these populations (in the maintainer and restorer streams and with respect to A_1 as well as A_4 CMS system) along with the original C_0 bulks of both populations were crossed onto A_1- and A_4-system male-sterile lines in three genetic backgrounds ($81A_1$ and $81A_4$, $5054A_1$ and $5054A_4$, and $ICMA_1$ 88004 and $ICMA_4$ 88004). The resulting top cross hybrids were first evaluated for the frequency of male-sterile plants (a measure of maintainer frequency) and male-fertile plants (a measure of restorer frequency) during the rainy season 2004 and the same was repeated during the summer 2005.

Genetic changes in maintainer and restorer frequencies: The results of the 2004 evaluation were confirmed during the summer 2005. The results based on the two seasons evaluations revealed that two selection cycles in ESRC II were effective in rapidly increasing the mean frequency of maintainers from 29% (C_0 bulk) to 90% (C_2 bulk) with respect to the A_1 CMS system, and from 42% (C_0 bulk) to 99% (C_2 bulk) with respect to the A_4 CMS system in the topcross hybrids made on $81A_1$ and $81A_4$ CMS systems when selection was carried for improving maintainer frequency. Similarly, the mean frequency of restorers increased from 71% in C_0 bulk to 96% in the C_3 bulk with respect to the A_1 CMS system, and from 58 to 96% in the C_2 bulk with respect to the A_4 CMS system. In Raj 171, one cycle of selection increased the mean frequency of maintainers from 22% (C_0 bulk) to 98% (C_1 bulk) with respect to theA_1 CMS system, and from 49% (C_0 bulk) to 99% (C_1 bulk) with respect to the A_4 CMS system. Similarly, one cycle of selection for fertility restoration increased the mean frequency of restorers from 78% (C_0 bulk) to 98% (C_1 bulk) with respect to the A_1 CMS system, and from 46% (C_0 bulk) to 96% (C_1 bulk) with respect to the A_4 CMS system. Broadly speaking, the results showed that both CMS systems were equally effective in genetic improvement of both populations for fertility restoration as well as for sterility maintenance reaction. Results of topcross hybrids made on the A_1 and A_4 system A-lines in the genetic background of 5054B and ICMB 88004 were broadly supportive of these findings on the patterns of genetic changes for restorer and maintainer frequency.

Associated changes in grain yield and agronomic traits associated with recurrent selection: To examine the influence of bi-directional selection for male fertility restoration and sterility reaction in ESRC II and Raj 171 on grain yield and agronomic traits, different cycle bulks of both the populations were evaluated. The ESRC II trial consisted of 21 bulks (C_0 bulk, and five bulks each for the restorer and maintainer stream of each of the two CMS system). Similarly, Raj 171 trial consisted of 13 bulks. First replicated trials were conducted separately for ESRC II and Raj 171 selection bulks during the rainy season 2004 and the same was repeated during the summer season 2005 at Patancheru. The results of 2004 evaluations were confirmed during 2005. The results based on the two seasons evaluation revealed that the selection either for fertility restoration or for sterility maintenance in ESRC II had no adverse effect on the mean grain yield with respect to the A_1 CMS system or for sterility maintenance reaction with respect to the A_4 CMS system. There were indications of significant decline

in the mean grain yield in the C_4 and C_5 restorer bulks of the A_4 CMS system. Changes in other traits such as time to flower, plant height, panicle length, tillering ability and seed weight were non- significant. Results from Raj 171 trial also showed that recurrent selection for fertility/sterility traits with respect to either of the two CMS systems had no adverse effect on the mean grain yield and agronomic traits.

Genetics of panicle and seed size: Genetic manipulation of yield-related traits is a common approach followed to improve grain yield in crop plants. Inheritance of such traits plays a key role in deciding the selection strategy. Hitherto, studies on inheritance pattern of panicle and seed traits have been conducted with genotypes having panicle length not more than 30 cm long, panicle diameter not more than 30 mm and 1000-grain mass not more than 12 g. But, pearl millet improvement program at ICRISAT-Patancheru has produced breeding lines having panicle length >60 cm, panicle diameter >45 mm and 1000-grain mass >16 g, which are expected to enter hybrid programs of SAT regions in the near future. Hence, the study of genetic analysis of these traits in the changed character state by means of generation means and triple test cross analysis was planned. During the 2005-rainy season, contrasting inbred lines with almost similar maturity and diverse genetic background for panicle length (long panicle ranging from 55 cm to 82 cm and short panicle ranging from 14.5 cm to 17.9 cm), panicle diameter (thick panicle ranging 42 cm to 61 cm and thin panicle ranging 15 mm to 19 mm) and seed size (large-seed ranging from 16.4 to 19.2 g and small-seed ranging from 4.1 to 7.5 g for 1000-seed mass) were selected based on morphological data. Six contrasting parents in each group were crossed to generate 9 F_1s (3 in each group) during the 2005 post rainy season.

KN Rai, VN Kulkarni and RP Thakur

Milestone: Genetics of fertility restoration on diverse CMS systems investigated in sorghum and pearl millet (2007)

Appropriate breeding materials for genetic studies: Isonuclear alloplasmic A-lines based on the widely used A_1 (*milo*) cytoplasm and alternative cytoplasms (non-*milo*) and their common R-lines (on one nuclear genetic background) have been developed. These are appropriate genetic material for conducting strategic research in areas such as assessing the effects of non-*milo* cytoplasms on mean performance and combining ability of A-lines and hybrids for agronomic traits as well as resistance to biotic and abiotic constraints in comparison to widely exploited *milo* cytoplasm, and inheritance of male fertility restoration of *milo* and non-*milo* cytoplasms without any confounding effects of nuclear genetic background of A-lines and R-lines. Such strategic information is essential for assessing feasibility of utilization of alternative cytoplasms for diversifying cytoplasm base of sorghum seed parents and hence hybrids and for improving the efficiency of breeding R-lines for different CMS systems.

Two common R-lines (ICSR 94453 and IS 33844-5) on A_1, A_2, A_3, A_4 CMS systems were crossed with A_1, A_2, A_3, A_4 (M), A_4 (VZM), A_4 (G)-based A-lines in six nuclear genetic backgrounds (ICSA 11, ICSA 37, ICSA 38, ICSA 42, ICSA 88001 and ICSA 88004) during 2005 rainy season. As the seed could not be obtained in all possible combinations due to continuous rain and pest attack, the same set was planted during the 2005–06 postrainy season for obtaining F_1s.

BVS Reddy and S Ramesh

Genetics of CMS systems: Genetics of fertility restoration of diverse cytoplasmic-nuclear male-sterility (CMS) systems in pearl millet was studied in F_2, BC_1 and BC_2 populations of the crosses involving isonuclear A-lines of the five diverse CMS systems (A_1, A_4, A_{egp}, A_5 and A_v) in three diverse nuclear genetic backgrounds (81B, 5054B and ICMB 88004), and six pollen parents restoring the male fertility of hybrids based on any one, two or three male-sterile cytoplasm. Linkage between the fertility restorer genes of the A_1 and A_4 CMS systems, allelism among the fertility restorer genes of these CMS systems and molecular markers linked to fertility restorer genes of the A_1 and A_4 CMS systems were also studied. In a majority of crosses across the CMS systems, fertility restoration was governed by a trigenic inheritance mechanism, represented either by dominant alleles of one basic gene and two duplicate-complimentary genes (F_2 ratio 45:19 and BC_1 ratio 1:1) or dominant alleles of any two of the three duplicate-complimentary genes (F_2 ratio 54:10 and BC_1 ratio 3:1). In few other crosses, different trigenic mechanisms with F_2 ratio of 57F:7S and 63F:1S and corresponding BC_1 ratio of 3F:1S and 7F:1S, respectively, were also observed. Although monogenic and digenic (F_2 ratio 15F:1S and 9F:7S and BC_1 ratio 3F:1S and 1F:3S) ratios were also observed in a few crosses, these resulted from the segregation of one or two genes out of the three involved in the trigenic inheritance. Segregation patterns of testcrosses from individual plants of F_2 and BC_1 populations derived from two B × R crosses were broadly supportive of the trigenic inheritance mechanism. Test of allelism studied from the fertility/sterility reaction of the three-way hybrids obtained by crossing A-lines with the F_1s of inter-crosses among three restorer lines (IPC 1518, IPC 511 and IPC 804) indicated the presence of same alleles of all the fertility restorer genes for the A_1 CMS system,

whereas different alleles are indicated for the A_4 system. Joint segregation analysis revealed the presence of linkage between the fertility restorer genes of A_1 and A_4 CMS systems. A linkage map of 708.8 cM was constructed using 397 individuals and 36 molecular (SSR and RFLP) and morphological markers in the F_2 mapping population derived from the cross 81B × IPC 804. For the A_1 CMS system, two QTL (*Rf1a* and *Rf1b*) and for the A_4 system, three QTL (*Rf4a, Rf4b* and *Rf4c*) were identified with different unlinked genomic regions involved in the fertility restoration of these CMS systems. Based on the overall inheritance pattern observed, possible genotypes of the A-lines irrespective of CMS background were assigned as *rf*_a *rf*_a *rf*_b *rf*_b *Rf*_c *Rf*_c or *rf*_a *rf*_a *rf*_b *rf*_b *rf*_c *rf*_c and of the restorer lines as *Rf*_a *Rf*_a *Rf*_b *Rf*_b *rf*_c *rf*_c or *Rf*_a *Rf*_a *Rf*_b *Rf*_b *Rf*_c *Rf*_c (underscore to be replaced with the numbers '1', '4' or '5' or alphabet 'e' or 'v' denoting the CMS systems). The information emanating from the study has implications in the breeding of maintainer and restorer lines of diverse CMS systems.

KN Rai, VN Kulkarni and Dev Vart Yadav

Milestone: Changes in virulence patterns in pearl millet downy mildew pathogen populations and effectiveness of resistance genes deployment assessed (2006)

The pearl millet downy mildew pathogen, *Sclerospora graminicola*, is a highly dynamic organism, hence its virulence shift is monitored through on-farm survey and multilocation virulence nursery.

On-farm downy mildew survey in Gujarat, India: An Indian Council of Agriculture Research-ICRISAT collaborative downy mildew survey was conducted in 70 pearl millet fields in 18 talukas of 8 districts (Ahmadabad, Anand, Banaskanta, Gandhinagar, Kheda, Mehasana, Rajkot and Surendranagar) in Gujarat, India during September. About 30% of the fields had disease, with the incidence ranging from traces to 70%. However, no disease was observed in Rajkot and Surendranagar districts. Public sector hybrids, GHB 558 and –577 had mean incidence of 3% and 12%, respectively, whereas private sector hybrids (Gowri, Nandi 3, -5, PG and several unknowns) had mean incidence of 2 to 14% with a range of 0 to 71%. Some other hybrids, such as Pioneer 7688, Proagro 9330 and -4444 were disease-free.

RP Thakur

Evaluation of new isolates from Gujarat, India: Fourteen oosporic samples collected from six districts of Gujarat (Kheda – 3, Anand – 2, Jamnagar – 1, Mehsana – 1, Banaskantha – 5 and Gandhinagar – 2) from different susceptible hybrids were evaluated for their oospore content (oospores g^{-1} of leaf powder) and viability using TTC (Triphenyl tetrazolium chloride) method. Four samples did not contain oospores whereas the 10 samples had varied number of oospores from 1.2×10^6 g^{-1} leaf powder from Narsandha, Kheda district (designated Sg 433) to 68.8×10^6 g^{-1} leaf powder from Sunav, Anand district (designated Sg 435). There was significant difference in the viability of oospores, ranging from 29% in Sg 435 (Sunav, Anand) to 54% in Sg 437 (Jamnagar) and Sg 442 (Tarana, Gandhinagar). These nine isolates have been established on a susceptible genotype, 7042S for further studies on pathogenicity and virulence.

RP Thakur

Pearl Millet Downy Mildew Virulence Nursery (PMDMVN): The PMDMVN-2005 consisting of 23 test entries and one local resistant and one local susceptible controls was established at 12 locations (Durgapura, Mandor, Fatehpur Sekhawati, Hisar, Anand, Jamnagar, Aurangabad, Dhule, Gwalior, Patancheru, Mysore and Coimbatore) in India under the ICAR-ICRISAT partnership project. The nursery was conducted in a RCB design with three replications. Each entry was grown in 2-row plots of 4 m. Downy mildew incidence data were recorded at 30- and 60-days after seedling emergence. The data from Fatehpur Sekhawati and Aurangabad were not considered because of low disease pressure (<50% on the susceptible control). At 60-days, the mean disease incidence across 10 locations ranged from 77 to 99% on the susceptible control 7042S, indicating adequate disease pressure at all the 10 locations. The pathogen population at Anand was more virulent, infecting 12 of the 23 test entries by recording higher disease incidence than the trial mean, followed by Mysore, Patancheru, Jamnagar, Durgapura, Dhule and Coimbatore locations. The pathogen population at Mandor appeared to be least virulent with only one entry having more incidence than the trial mean. Only two entries (IP 18292 and IP 18293) showed high level of resistance stability with mean incidence of 4–6% across locations. Among the new B-lines, ICMB 93333 was the most resistant with a range of 0 to 7% incidence. Other four B-lines (ICMB 99013, ICMB 94555, ICMB 95444 and ICMB 97111) were also found resistant with mean incidence ≤8%.

RP Thakur

Milestone: Pathogenic nature of and variability in sorghum grain mold fungi determined, and greenhouse screening technique refined (2006)

Refine greenhouse-screening technique to identify resistance to the major individual grain mold pathogens: In order to develop an effective grain mold resistance breeding program, it is important to identify resistance to the individual pathogens. This could be done by artificially inoculating the sorghum panicle at the right stage and providing congenial condition for infection establishment and disease development. With this objective in view, a greenhouse grain mold-screening technique has been developed that involves: (i) spray inoculation of pot-gown sorghum plants at full anthesis with spore suspension (1×10^6 spores mL^{-1}) with the target pathogen; (ii) provide panicle wetness (95–100% RH) for 48 h by misting following inoculation at 25–28°C to facilitate infection establishment; (iii) again provide panicle wetness at physiological maturity (PM) for 72 h by misting to promote grain colonization by the pathogen; (iv) score for visual grain mold severity as percentage grain colonized on a panicle at physiological maturity; and (v) determine grain infection severity by plating grains using the blotter method.

Using this technique, a set of 20 sorghum lines including one resistant and one susceptible controls were screened for infection by *Fusarium verticilloides, Curvularia lunata* and *Alternaria alternata* in three different sets of experiment. Results indicated differential susceptibility levels of sorghum lines to individual fungi. Based on grain colonization scores, ICSB 370-2 was found to be resistant (<10% colonization) to *Fusarium verticilloides* and *A. alternata* whereas SGMR 40-1-2-3 was resistant to *C. lunata* and *A. alternata*.

RP Thakur

Analyze *Fusarium* isolates for their fumonisins production potential: A preliminary identification of the 17 isolates (from ICRISAT-Patancheru) based on morphology, crosses with tester isolates, and AFLP markers had revealed the presence of five *Fusarium* species in the sorghum grain mold complex (WFO Morasas, Medical Research Council, South Africa and JF Lesley, Kansas State University). These were: *F. verticillioides, F. proliferatum, F. thapsinum, F. sacchari* and *F. andiyazi*. Among these, the frequency of *F. proliferatum* was highest, followed by *F. verticillioides*, *F. thapsinum*, and *F. andiyazi*. In order to determine the frequency of these species in the grain mold complex, we attempted to evaluate large number of *Fusarium* isolates collected from different locations in India.

Of the 948 cultures of *Fusarium* spp. obtained from sorghum grain mold variability nursery conducted at five locations in India during 2002–04, a total of 682 were characterized during the past 2 years by comparing the growth patterns and pigmentation (images of abaxial and adaxial surfaces on PDA plates) with those of the reference cultures, and were grouped into six species. The across-location mean frequency of occurrence was highest for *F. proliferatum* (48%), followed by *F. thapsinum* (33%) and the lowest was for *F. sacchari* (2%). Among the locations, the frequency of occurrence of *F. proliferatum* was highest at Parbhani (66%) and lowest at Surat (28%); for *F. thapsinum* it was highest at Patancheru (50%), and lowest at Palem (23%); and for *F. verticillioides*, it was highest at Surat (16%), and lowest at Parbhani (5%). The results indicate variation in frequency distribution of *Fusarium* species in different sorghum growing areas and thus resistance identification to different *Fusarium* species should be planned accordingly.

Fumonisins estimation in *Fusarium* isolates: Fumonisins (FB_1 and FB_2) estimation of 12 isolates by HPLC at Iowa State University, USA revealed that *F. proliferatum* produced the highest levels of FB_1 (7.56 µg g^{-1} grain) and FB_2 (8.7480 µg g^{-1} grain), followed by other strains. Several other strains of each species produced FB_1 in various levels, but those of *F. sacchari*, *F. andiyazi*, and some strains of *F. proliferatum*, and *F. verticillioides* did not produce FB_2. Among the 682 *Fusarium* isolates assayed for their fumonisins production potential using competitive ELISA method at ICRISAT-Patancheru, the range of FB_1 production levels across the five locations varied from 0–811 µg kg^{-1} grain by *F. sacchari* to 0–4765 µg kg^{-1} grain by *F. proliferatum*, followed by *F. thapsinum*. Among five locations, isolates from Surat produced relatively low levels of FB_1 compared to those from other locations. Further investigations are needed to understand the role of environment on fungal growth and fumonisins production levels.

RP Thakur

Milestone: *Helicoverpa* resistance screening techniques refined, and mechanism and inheritance resistance studied in chickpea and pigeonpea (2006)

Mechanism and inheritance of resistance to *Helicoverpa* in chickpea: Genetics of resistance to pod borer (*Helicoverpa armigera)* in chickpea was focussed on studying the nature of gene action and maternal effects, plant resistance mechanisms and interaction of different components of resistance and grain yield. Eight *desi*

[ICC 12475 (ICC 506), ICC 12476, ICC 12477, ICC 12478, ICC 12479, ICC 4918, ICC 12426 or ICCC 37 and ICC 3137] and one *kabuli* [ICCV 2 (ICC 12968)] parents were selected based on earlier screening trials to study the genetics of resistance, using a full diallel. ICCV 2 was the earliest to flower and mature followed by ICC 4918, ICCC 37, ICC 12478 and ICC 12477, while ICC 12479, ICC 12476 and ICC 3137 were late to flower and mature. ICC 12478 suffered significantly lower pod borer damage, followed by ICC 506, ICC 12479 and ICC 12477. ICC 3137 was highly susceptible and recorded lowest seed yield. Most of the crosses with ICC 506, ICC 12478 and ICC 12479 suffered low damage, while those with ICC 3137 suffered higher damage. ICCC 37 recorded higher yield, followed by ICC 12479 and ICC 12476.

Inheritance of resistance: Gene action and maternal effects were estimated from the full diallel trial. Additive gene action was predominant for days to initial flowering, days to 50% flowering, days to maturity, pod borer damage (%), pods plant^{-1}, seeds plant^{-1}, seeds pod^{-1} and 100-seed weight. While non-additive gene action was important for yield plant^{-1}, total plot yield and yield (kg ha^{-1}). The additive: dominance (A:D) ratio was greater than unity for days to 50% flowering, days to maturity, pod borer damage (%), pods plant^{-1}, seeds plant^{-1}, seeds pod^{-1} and 100-seed weight, indicating over dominance, while for yield plant^{-1} and yield (kg ha^{-1}), the ratio was less than unity, indicating partial dominance. There was no maternal inheritance for maturity-related traits, pod borer damage, and grain yield. The hybrid, ICC 12476 × ICCC 37 showed positive and significant specific combining ability (SCA) effects for seeds pod^{-1}, but the reciprocal hybrid ICCC 37 × ICC 12476 showed negatively significant SCA effects for number of seeds pod^{-1}. So the hybrid ICCC 37 × ICC 12476 may be showing cytoplasmic effect for the number of seeds pod^{-1}.

Mechanisms of resistance: The three mechanisms of resistance *viz.,* non-preference for oviposition, antibiosis and tolerance to *H. armigera* in chickpea genotypes were studied under laboratory, greenhouse and field conditions. Oviposition studies under no-choice, dual choice and multi-choice laboratory and multi-choice field conditions revealed that the resistant control genotype, ICC 506 recorded lowest number of eggs, followed by ICC 12476, ICC 12477 and ICC 12478. The highest oviposition was observed on the susceptible genotypes, ICC 12426 and ICC 4918. The genotypes ICC 506, ICC 12476, ICC 12477, ICC 12478 and ICC 12479 were least preferred by *H. armigera* females for oviposition compared to ICC 4918, ICC 3137 and ICCV 2. In detached leaf assay studies, the survival rate and larval weights were lowest on the resistant control, ICC 12475 (ICC 506), followed by ICC 12476, ICC 12477, ICC 12478 and ICC 12479, suggesting that water-soluble compounds in the leaf exudates (malic and oxalic acid) were primarily responsible for resistance to *H. armigera*.

Tolerance: The genotypes ICC 12476, ICC 12477, ICC 12478 and ICC 12479 were found to be resistant and their levels of resistance were comparable to the resistant control, ICC 12475 (ICC 506) under no-choice cage conditions. Under un-infested conditions, the per plant yield was greater in ICC 12426 followed by ICC 12478 and Annigeri. The resistant cultivars ICC 12478 and ICC 12475 recorded higher yield than the rest of the cultivars. At the podding stage of the crop, when plants were infested with the third instar larvae, the recovery resistance was very poor, as most of the plants were damaged.

Larvae fed on leaf material and on artificial diet with lyophilized leaf and pod powder recorded lowest larval and pupal weights and prolonged larval and pupal periods on the resistant genotype, ICC 506. Highest growth index, adult index, oviposition index and pupal index were recorded on ICC 12426 and ICC 4918, while the lowest on the resistant control, ICC 12475 (ICC 506).

High Performance Liquid Chromatography (HPLC) profile of leaf exudates showed that the malic acid was negatively correlated with damage rating at flowering (–0.28, PL0.05), at maturity (–0.32, PL0.01) and pod damage (–0.22, PL0.05). Oxalic acid showed negative significant correlation with damage rating in detached leaf assay (–0.22, PL0.05). Acetic acid showed a negative correlation with larval weight (-0.45, PL0.05), damage rating at flowering (–0.33, PL0.01) and maturity (–0.26, PL0.05). Citric acid showed negative and significant correlation with damage rating at flowering (–0.23, PL0.05).

The genotypes, ICC 12476, ICC 12477, ICC 12478, ICC 12479 and ICCV 2 were on par with the resistant control, ICC 12475 for pod borer damage under protected conditions. ICC 12475, ICC 12426, ICC 12478 and ICC 12479 recorded higher grain yield under unprotected conditions. The genotypes ICC 12475 (3.77%) and ICC 12478 (6.59%) recorded the lowest reduction in grain yield under unprotected conditions, indicating the presence of tolerance mechanism in chickpea to *H. armigera.* The tolerant lines can be used in further breeding programs and the mechanisms responsible for the resistance can be exploited to develop resistant varieties.

Correlation of different components of resistance with grain yield showed significant positive correlation under protected conditions between number of larvae and eggs (0.89, PL0.01), leaf damage and egg number (0.82,

PL0.05), yield plant^{-1} and egg number (0.77, PL0.05), yield plant^{-1} and larva number (0.76, PL0.05) and pod damage (%) and larval number (0.91, PL0.01). Significant negative correlation was recorded between yield plant^{-1} and borer damage (%) (–0.79, PL0.05), under unprotected conditions. These correlations and interaction of different components of resistance and grain yield will help in gene pyramiding.

CLL Gowda and HC Sharma

Milestone: Allelic relationship of genes for early flowering and other agronomically important traits in chickpea established (2005)

Allelic relationships, penetrance and expressivity of genes controlling number of flowers per axis in chickpea: A double-flowered (two flowers per pedicel) line ICC 4929, a triple-flowered line IPC 99-18 and a multi-flowered line JGM 7 were intercrossed in all possible combinations and flowering behavior of parents, F_1s and F_2s was studied to establish allelic relationships, penetrance and expressivity of genes controlling number of flowers per axis in chickpea. The F_1s from the double-flowered × triple-flowered cross were double-flowered, whereas F_1s from double-flowered × multi-flowered and triple-flowered × multi-flowered crosses were single-flowered. The F_2s from double-flowered × triple-flowered cross gave a good fit to a 3:1 ratio for double-flowered and triple-flowered plants. The F_2s from double-flowered x multi-flowered cross segregated in a ratio of 9:3:3:1 for single-flowered, double-flowered, multi-flowered and double- and multi-flowered plants. The F_2s from triple-flowered x multi-flowered cross segregated in a ratio 9:3:4 for single-flowered, triple-flowered and multi-flowered plants. The results clearly established that two loci control number of flowers per axis in chickpea. The double-flowered and triple-flowered traits are controlled by a single-locus (***Sfl***) and the allele for double-flowered trait (***sfl$_d$***) is dominant over the allele for triple-flowered trait (***sfl$_t$*).** The multi-flowered trait is controlled by a different gene (***cym*)**. Single-flowered plants have dominant alleles at the both the loci (***Sfl_ Cym_***). The double-flower, the triple-flower and the multi-flower traits showed complete penetrance, but variable expressivity. The expressivity was 96.3% for double-flower and 76.4% for double-pod in ICC 4929; 81.2% for triple-flower and 0.0% for triple-pod in IPC 99-18; and 51.3% for multi-flower and 24.7% for multi-pod in JGM 7. Average number of flowers per axis and average number of pods per axis were higher in multi-flowered line JGM 7 than double-flowered line ICC 4929 and triple-flowered line IPC 99-18.

PM Gaur

Allelic relationships of genes controlling stem fasciation in chickpea: Several spontaneous mutants (ICC 2042, ICC 5645, ICC 14871) and one induced mutant (JGM 2) for stem fasciation are known in chickpea. A study was conducted to establish allelic relationships of genes controlling stem fasciation. The four mutants were crossed in all possible combinations. The F_1s from the crosses ICC 2042 × ICC 5645, ICC 2042 × ICC 14871 and ICC 5645 × ICC 14871 had fascinated stem and bred true in F_2. This indicated the presence of a common gene for stem fasciation in the three spontaneous mutants. The F_1s of the crosses of the induced mutant JGM 2 with all spontaneous mutants had normal F_1 plants and segregated in a ratio of 9 normal: 7 fasciated plants in F_2. Thus, the gene for stem fasciation in the induced mutant JGM 7 is not allelic to the common gene for stem fasciation in spontaneous mutants (ICC 2042, ICC 5645 and ICC 14871). The two genes in dominant condition produce normal phenotype. Fasciated plants showed significantly higher mean values for pods per plant and yield per plant than normal plants in F_2 of JGM-2 × ICC 2042. However, in F_2 of JGM-2 × ICC 5645 and JGM-2 × ICC 14871 normal plants had higher mean values than the fascinated plants.

PM Gaur

Milestone: Pathotypes of *Ascochyta* blight (AB), *Botrytis* gray mold (BGM) and *Fusarium* wilts in chickpea and pigeonpea characterized and host plant differentials identified (2006)

Morphological and pathogenic characterization of *A. rabiei*: *Ascochyta rabiei*-infected chickpea plants were collected from 13 locations in northwest plain zones of India during different crop seasons. Following standard mycological procedures, 16 pathogen isolates were observed. Single-spore isolates of these cultures were obtained and stored on potato dextrose agar at 4°C. Morphological and pathogenic characters of these single-spore isolates were studied. Isolates differed in their morphology, pycnidial color (brown to slate grey), formation of pycnidiospores (5.5×10^4 to 3.1×10^5 cm^{-2}) and pycnidial size (156.0 × 116.0 µm to 263.5 × 231.5 µm).

Morphological and pathogenic characterization of *B. cinerea*: To determine the genetic diversity of *B. cinerea* isolates and improve the efficiency of breeding for resistance, the fungus was collected from infected chickpea plants from different locations in Nepal, Bangladesh and India. Thirty-two *B. cinerea* isolates infecting chickpea, lentil and marigold were collected using BGM-specific medium containing tannic acid. Preliminary analysis of eight Indian isolates using 20 RAPD primers (decamers) categorized them into two

distinct groups. For further detailed molecular analysis using SSR markers, pure cultures of these isolates were sent to University of Melbourne, Australia.

S Pande and GK Kishore

Morphological and pathogenic characterization of races of *Fusarium oxysporum* f.sp. *ciceri*: Breakdown of host-plant resistance to *Fusarium* wilt is often reported due to the existence of different races of the pathogen. We initiated characterization of *F. oxysporum* f. sp. *ciceri* isolates from major chickpea growing areas of India. A total of 64 isolates of *Fusarium* (60 isolates collected from different locations from India during 1995–2004 and 4 race cultures reported during 1980) were used for characterization of the fungus. Microscopic observations of all these cultures indicated that some of them were black root rot pathogen *F. solani.* Therefore, pathogenicity test using susceptible cultivar JG 62 was conducted to segregate wilt, root rot and non-pathogenic groups. Standard root dip technique was employed for conducting pathogenicity test. Fifteen seedlings of the cultivar JG 62 were dipped in the inoculum of each isolate and transplanted in three pre-irrigated, 15 cm (diameter) plastic pots @ five plants per pot. Each pot represented one replication and all the pots in each replication were arranged in completely randomized block design. Outmost care was taken to avoid cross contamination while preparing the inoculum and during inoculation. The seedlings were observed for wilt symptoms for 30 days.

Three types of reactions were recorded from all these 64 isolates. Nineteen isolates [4 isolates from ICRISAT (AP), 4 from Hisar (Haryana), 1 from Gulbarga (Karnataka), 2 from Rahuri (Maharashtra), 1 from Ludhiana (Punjab), 2 from Pantnagar (Uttaranchal), 3 from Kanpur (Uttar Pradesh), Race 1 (Hyderabad) and Race 2 (Kanpur)] expressed initial wilt symptoms caused by *Fusarium oxysporum* f. sp. *ciceri* (FOC) between 9 and 18 days after inoculation, while 27 isolates produced black root rot symptoms caused by *Fusarium solani.* The remaining 19 isolates were found non-pathogenic to chickpea. Single-spore isolates were obtained from all these pathogenic isolates of FOC following standard mycological techniques. All the single-spore isolates of FOC were tested for their virulence on a susceptible cultivar JG 62 and aggressive isolates were stored in paraffin oil at 4°C for further studies.

Since representation of FOC cultures was not properly covered across the country, collection of isolates was continued in 2004/05 season. Accordingly, 25 isolates were collected from 8 locations in 7 states of India [Dholi (Bihar), Junagadh (Gujarat), Dhaulakuan (Himachal Pradesh), Sehore (Madhya Pradesh), Badnapur (Maharashtra), Ludhiana, Gurdaspur (Punjab), Gaziabad (Uttar Pradesh)]. Pathogenicity tests and single spore isolations are in progress.

S Pande

Morphological and pathogenic characterization of races of *Fusarium udum*: *Fusarium* wilt-infected pigeonpea plants were collected from nine locations in India during 2004 season. *F. udum* was isolated in the laboratory from each sample, using specific medium. These isolates were tested for their virulence on a common susceptible cultivar ICP 2376. All these nine strains (Patancheru, Badnapur, Bangalore, Gulbarga, Akola, Khargoan, Muradnagar, Warangal and Varanasi) were purified by single-spore cutting. In preliminary investigations, morphological differences were observed among different isolates, and pathological differences are being studied in detail.

S Pande and GK Kishore

Milestone: Physiological mechanisms and traits involved in drought avoidance and salinity tolerance characterized in chickpea (2005)

Methodology development for water use efficiency (WUE): In some C_3 crops, an association between water use efficiency (WUE) and Δ(leaf discrimination against ^{13}C) has been used as an indirect selection criterion. However, there is no report so far on the applicability of this technique to chickpea. Therefore, $\Delta^{13}C$ method has been under test on 10 diverse chickpea genotypes grown in pots in controlled glasshouse facility. In collaboration with Japan International Research Center for Agricultural Science (JIRCAS), it was shown that WUE of chickpea could be estimated by using $\Delta^{13}C$ technique. In 2005, leaf samples of chickpea from the mini-core collection were collected that will be sent to JIRCAS for the analysis.

J Kashiwagi

Milestone: Photoperiod-temperature responses of CMS lines assessed (2006)

Stability of CMS lines: For production of high yielding hybrids, it is essential to identify stable CMS lines. Three CMS lines with diverse cytoplasm were selected for this study. These include ICPA 2067 (A_1 cytoplasm),

ICPA 2052 (A_2 cytoplasm), and ICPA 2039 (A_4 cytoplasm). These lines were evaluated at Parbhani and Patancheru during 2004 and 2005 rainy season. Anthers were squashed in 2% aceto-carmine and three microscopic fields were examined for each floral bud. The number of sterile and fertile pollen grains was counted in each microscopic field at one-week interval. The observations showed that ICPA 2039 was the most stable CMS line at both locations. In case of ICPA 2052, five out of 49 plants showed fertile pollen grains (5–30%) and all other plants were sterile throughout the season at Patancheru. More or less similar results were observed at Parbhani. ICPA 2067 was found to be the most sensitive to environmental factors, as 24 out of 28 male-sterile plants, reverted to male-fertility at Patancheru. At Parbhani also, 24 out of 36 male-sterile plants reverted to male-fertility. The reasons for these changes are being examined.

Cytological study conducted at Parbhani showed that the breakdown of tapetum layer at the time of tetrad formation was the reason for male-sterility in the plants. In case of fertile genotypes the tapetum layer remained until the formation of pollen grains. In the environment-sensitive genotypes, the phenomenon of tapetum breakdown is affected by changes in the temperature and photoperiod and hence these lines converted to fertility.

VA Dalvi and KB Saxena

Output 1.3: Effective and eco-friendly IPM technologies designed and evaluated for legumes

Summary

Although the development of improved cultivars with high yield potential and resistance to major insect pests and diseases is a major part of research in this project, these efforts are backed by development and integration of other components of the integrated pest management (IPM), especially in the legumes. Earlier studies had clearly established the effectiveness of Helicoverpa *nuclear polyhedrosis virus (HNPV) for* Helicoverpa *management. A study addressing the problem of bad odour (malodor) and persistence of its virulence during storage showed that of the several preservatives, 10% acetone or 10% ethyl alcohol were most effective for more than six months of storage and substantial suppression of malodor. Also, adopting a simple, affordable farm-level technique, large-scale production of red hairy NPV (RHNPV) with 10000 larvae was successfully carried out at the farm level to demonstrate the usefulness of this technology for highly migratory red hairy caterpillar management. Botanical as well as microbial bio-pesticides (bacterial and fungal origin) have been found to prevent crop damage by insect pests. Research showed a high degree of differential compatibility between the botanicals and microorganisms, implying that identification of botanicals compatible with microorganisms is essential. Alternatively, selection for microbial strains compatible with desired botanicals would also have long-term beneficial effects for these biological options for managing insect pests. On-farm evaluation of an IDM technology that packages compost, Trichoderma and gypsum was found to be highly effective with 99% reduction in aflatoxin contamination, leading to 2 $\mu g\ kg^{-1}$ of aflatoxin in groundnut.*

Activity 1.3.1: Develop cost-effective and eco-friendly components of IPM technologies

Milestone: *Metarhizium* evaluated for the management of groundnut leafminer and *Maruca* (2005)

Evaluation of various insecticides for the management of *Maruca* through field studies conducted at ICRISAT-Patancheru during rainy season 2005 revealed maximum population reduction (82%) with Spinosad (Tracer) followed by indoxacarb (72%), monocrotophos (40%) and *metarhizium* (20%) 48 h after application. Five days after the application of treatments there was 90% population reduction with Spinosad compared to 85% with indoxacarb followed by 22% with monocrotophos. The larval population in *metarihzium* treatment was on par with control (6 larvae $plant^{-1}$).

GV Ranga Rao

The Year 4 work plan of IPM and IDM farmer participatory trials of IFAD TAG 532-ICRISAT project at various locations in four countries was successfully implemented. The IPM package consisting of botanicals, bio-agents, chemicals and pheromone traps for different legumes is now ready for up scaling in India, Nepal and Vietnam. The use of bio-agent in disease control is also gaining popularity among farmers.

SN Nigam and GV Ranga Rao

Milestone: Components of Aflatoxin (groundnut) and *Helicoverpa* (pulses) management technologies evaluated (2005)

Compatibility of entomopathogenic microorganisms and botanicals: *Bacillus subtilis* strain BCB 19 of a bacteria and a fungal entomopathogen *Metarrhizium anisopliae* strain GVR had earlier been developed for

biological control of *Helicoverpa armigera*. Botanicals have often been used experimentally to prevent crop damage by this and other insects. Therefore, there was an interest to learn if these two microorganisms would survive in the presence of four widely uses botanicals - *Azadirachta indica*, *Gliricidia sepium*, *Calotropis procera*, and *Nerium odorum* in field studies. Hot-water extract and bio-extract (wash of compost prepared from foliage of a given botanical) each of the four botanicals were included in the study. To assess compatibility, both a botanical extract and water suspension of a given microorganism were mixed at a concentration/level recommended for spray and the different mixtures were kept at 26ºC in a glass bottle. Samples from the different mixtures were drawn for determining population of a given microorganism on day 1 and day 7. The study revealed that the strain BCB 19 survived in both types of extracts of all the four botanicals (5.49 to 7.03 $\log_{10}$ mL^{-1}) up to seven days (Table 1.2). In hot-water extracts of two botanicals (*Calotropis procera*, and *Nerium odorum*), population of BCB 19 increased by about 10-times when assessed after seven days. But the fungus *Metarrhizium anisopliae* strain GVR survived well (4.48 to 5.04 $\log_{10}$ mL^{-1}) for seven days, only in bio-extract but not in the hot-water extract of the four botanicals. Thus, identification of botanicals compatible with desired microorganisms is essential. Alternatively, selection for microbial strains compatible with desired botanicals would also be a good idea to have long-term beneficial effects of these biological options of managing insect pests.

Table 1.2. Population ($\log_{10}$ mL^{-1}) of *Bacillus subtilis* (strain BCB 19) and *Metarhizium anisopliae* in selected botanicals

		BCB 19			Metarhizium	
Treatment	Name of botanical	Day 1	Day 7		Day 1	Day 7
Biowash	*Azadirachta indica*	5.70	5.62		5.00	4.48
	Gliricidia sepium	5.63	5.58		5.04	4.78
	Calotropis procera	5.53	5.49		4.78	4.90
	Nerium odorum	5.41	5.52		4.70	4.70
Hot water	*Azadirachta indica*	5.28	6.45		4.30	ND[1]
Extract	*Gliricidia sepium*	6.51	6.36		4.78	ND
	Calotropis procera	5.96	6.86		4.90	ND
	Nerium odorum	5.79	7.03		4.95	ND
1 = ND = Not detected at dilution 10^3.						

Population of plant growth promoting rhizobacteria (PGPR) in compost samples and market products: Current trends in agriculture are focused on the diminution of the use of chemical pesticides and inorganic fertilizers. This is strongly indicated by the fact that crops on about 31 million ha (76,326 ha in India) are currently grown without such input (www.orgprints.org, 23 March 2006) on farms widely known to follow organic farming (OF) practices, with third party certification by accredited agencies. Without chemicals, growing good crops on such farms were difficult to believe. High microbial activity of plant growth promoting rhizobacteria (PGPR) in the major inputs (eg, compost) used by farmers was hypothesized as one plausible reason of the high yields. Most of those OF practitioners that we visited used products involving cow dung and, therefore, was included in this study. Compost samples were obtained from seven farmers' fields following OF practices. One sample (with three replications) was from the compost prepared at ICRISAT-Patancheru using the method widely used by organic farmers. Population of four different groups of bacteria (siderophore producers, nitrogen fixers (*Azotobacter* like), phosphate (P) solubilizers, and *Pseudomonas fluorescens* (indicator of bacteria with ability to suppress disease-causing fungi) was identified in these samples. All these bacteria are generally classified as PGPR. Some PGPR are known to function inside plant tissue after entering through roots and express beneficial traits. Several PGPR bacteria are also sold as biofertilizers. We obtained market products of such bacteria from five private sector companies in India and evaluated the population of the relevant bacteria in these products. Results suggested surprisingly high population of beneficial bacteria in composts and lower than expected (10^7 per g carrier or per mL in liquid formulations, as per regulatory system in India) in market samples. Most compost samples were rich in all the three groups of agriculturally beneficial bacteria that were determined - siderophore producers (6.07–8.11 $\log_{10}$ g^{-1}), P-solubilizers (<2.00-7.85 $\log_{10}$ g^{-1}), and *P. fluroscens* (<3.0–6.89 $\log_{10}$ g^{-1}). Also the cow dung and Amrit Paani (another product used widely by organic farmers and prepared using cow dung) were rich in all these bacteria. On the contrary, most products from biofertilizer companies had lower than the expected (10^7 per g carrier-based or per mL liquid-based products) population of desired bacteria. Market products were studied only for three groups of bacteria - nitrogen fixers (3.30–7.45 $\log_{10}$ g^{-1}), P-solubilizers (4.70–6.62 $\log_{10}$ g^{-1}), and *P. fluorescens* (2.0–6.81 $\log_{10}$ g^{-1}). The poor quality of the market

products thus raises an important question on the justification for spending resources on doing research on PGPR for putting them on market place, when farmers' traditional knowledge products are already rich in such beneficial bacteria. Another important implication of these results is on the manner in which the inputs such as composts are presently used by farmers – by heaping it generally in hot summer when they have spare time before soil incorporation. What happens to the population of these beneficial bacteria will be an interesting piece of investigation.

OP Rupela

Management options for aflatoxin control: To develop low-cost options for the management of aflatoxin contamination in groundnut, a field trial was laid-out at ICRISAT center during 2005 rainy season. The trial comprised of four treatments (application of compost, gypsum and their combination and control) and planted in six replications using randomized complete block design. A susceptible cultivar (JL 24) was used in the trial. Highly toxigenic strain (AF 11–4) of *A. flavus* multiplied on maize/sorghum/pearl millet seed was broadcasted in the field before sowing, followed by row application of inoculum at fortnight intervals starting from 25 days after sowing. Terminal drought was imposed 30 days before harvest to facilitate the seed infection and aflatoxins contamination. Harvesting at 110 days after sowing was done by up-rooting the plant and the produce was dried under sunlight for 5–7 days before the pods were stripped. An increase of 2–11% in pod and haulm yields were observed in treatments over the control (with 857 and 2026 kg ha^{-1} pod and haulm yields, respectively). *A. flavus* infection (0–2%) and aflatoxin contamination (2–4 μg kg^{-1}) was very low in all the graded samples, except in damaged seed. It is very difficult to draw any conclusion of the treatment effect with this kind of data. Low level of *A. flavus* seed infection and aflatoxin contamination could be due to failure to impose the terminal moisture stress conditions in the field as there were continuous rains during the imposed drought stress period.

Farid Waliyar

The validation of chickpea pod borer forecast model revealed the relation between pheromone trap catches and oviposition and larval load in the following 1-3 week period. Pheromone trap catches during 2003–04 postrainy season were at peak (52 moths/trap/day) during first week of December which resulted in peak larval activity (4.5 larvae $plant^{-1}$) in the third week of December. This had given a clear indication of 15 days interval between moth catch and the damage. However, the adult activity during 2004–05 postrainy season was much less (<10 moths/tarp/day) and the consequent lower larval population (1.2 larvae $plant^{-1}$) during the third week of December, when crop was at reproductive phase.

GV Ranga Rao

Milestone: Technologies for mass production, storage and utilization of NPV, bacteria and fungi pathogenic to insect-pests developed (2006)

Helicoverpa nuclear polyhedrosis virus (HNPV) is one of the critical inputs of *Helicoverpa* IPM strategy. Several strains of HNPV are in use in the management of this pest. Of the HNPV strains from six locations (ICRISAT-Patancheru, Ludhiana, Junagadh, Akola, Dharwad and Coimbatore), ICRISAT-Patancheru strain was found to be more virulent based on the lowest LC50 values (0.54×10^8) on fifth day. The studies conducted on their DNA characterization indicated the presence of three to four major polypeptides viz., VP42.32 ± 0.92, VP34.74 ± 0.27, VP31.77 ± 0.44, VP30.66 ± 0.27 in all the strains except the strain from Junagadh, which had an extra polypeptide with 19 ± 1.41 kDa and several minor polypeptides. These studies provided clear-cut evidence on the degree of virulence of various strains. The virulence strains identified would be of immense value for increasing the biotic potential of HNPV under field conditions.

The production and storage of HNPV under various conditions often encountered the problems of bad smell (mal-odor) and persistence of virulence over long periods. In order to address this constraint, studies conducted in evaluating different preservatives for efficient long-term storage of HNPV indicated that virus with 10% acetone resulted in 73% mortality of larvae 10 months after storage, followed by 70%, 63%, 57%, 53%, 47% with 10% ethyl alcohol, 10% phenol, 10% dettol, 10% methanol and 10% ethyl acetate, respectively. Though most of these preservatives showed good response to HNPV storage up to six months, 10% acetone and 10% ethyl alcohol were found to be effective for more than six months while suppressing the malodor substantially. These preservatives are easily available and affordable with an extra input of Rs. 3–12 ha^{-1}. The results obtained can be effectively utilized by the industry and farming communities.

Adopting a simple, affordable farm-level technique, large-scale production of red hairy NPV (RHNPV) with 10000 larvae was successfully carried at the farm level during 2005. This technology would be an added asset in

managing the outbreaks of sporadic and highly migratory red hairy caterpillar (RHC) using RHNPV bio-pesticide.

GV Ranga Rao

Activity 1.3.2: Integrated IPM components and validate for effective pest/disease management

Milestone: Low-cost agro-practices for management of groundnut aflatoxin contamination and HPR integrated and tested in farmers fields (2005)

Several research reports indicate that cultural practices such as application of farm yard manure, gypsum, crop residues, and application of several bio-control agents such as non-toxigenic strains of *A. flavus*, *Trichoderma*, *Bacillus* and *Pseudomonas* reduce the aflatoxin contamination. Hence the components, viz., compost, gypsum and *Trichoderma viride* alone and their combination were tested in participatory on-farm trials. The trial was conducted in 10 farmers' fields at Ontillu, M.C. Palem, and Mullaguruvaripalli villages in Pileru. The following components were tested at each farmer's field by adopting plot size of 10×10 m^2. Compost was incorporated in the soil after field preparation, *Trichoderma* was applied in the soil before sowing and gypsum was applied at flowering time. The plantings were carried out during the second fortnight of July using local variety TMV 2 which is very susceptible to aflatoxin contamination.

Components:

1. Application of compost @ 5 t ha^{-1}
2. Application of Trichoderma @ 100 kg ha^{-1}
3. Application of gypsum @ 500 kg ha^{-1}
4. Compost + Trichoderma + Gypsum application
5. Farmers practice (control): At Pileru: Farmers apply neither farm yard manure nor fertilizer whereas at Anantapur farmers applied Muriate of potash, urea and single super phosphate.

In Anantapur district, only *Trichoderma viride* was tested at Rekulakunta village in 10 farmers' fields. *Trichoderma* was applied adjacent to the rows one week after germination. Harvesting was done by uprooting the plants that were field dried. Later, the pods were stripped manually and pod and haulm yields were recorded. Pod samples from each plot were drawn for toxin estimation. After drawing the bulk sample, the damaged pods were sorted out then remaining pods were shelled and sorted into large and small kernels. Results for the bulk samples are presented and other category kernel's processing is in progress.

At Pileru, no significant difference was observed among the treatments with regard to pod and haulm yields. Very low yields were obtained in all the treatments, which may be due to uneven distribution of rainfall that resulted in poor pegging and pod development and pod loss at the time of harvest. Bulk seed samples from all the plots were used for aflatoxin estimation using ELISA. Results on aflatoxin contamination levels in different treatments are very encouraging. Highest aflatoxin contamination 369 μg kg^{-1} was observed in untreated control plot. All the four treatments responded to the treatments by reducing aflatoxin contamination. Highest reduction in aflatoxin contamination (99% showing only 2 μg kg^{-1} of aflatoxin) was observed in the plots with compost when *Trichoderma* and gypsum were applied together, followed by individual application of gypsum, compost and *Trichoderma* over the control plots. Application of compost, *Trichoderma* and gypsum are known to reduce *A. flavus* seed infection and aflatoxin contamination. In the present study, all the individual treatments responded well to reduce the aflatoxin contamination and combination of treatment showed the confounding effect for the reduction of aflatoxin contamination in groundnut.

In Anantapur area, the results indicated that there was 13% increase in pod yield in *Trichoderma*-treated plots over the control plot yield (590 kg ha^{-1}) and there was not much difference in haulm yield. However, there was no aflatoxin contamination in all the 10 treated and control plots.

Farid Waliyar

Phytosanitary aspects of maize for aflatoxin contamination: To assess the aflatoxin situation in maize, surveys were undertaken in the major maize producing districts (Karimnagar, Nizamabad and Medak) of Andhra Pradesh, India. Maize (cobs and kernels) samples were collected 1–7 days after harvest, from rainy season and postrainy season crops grown in 2004–05 and also from various storages for aflatoxin analysis. Maize kernels were analyzed for the total aflatoxin content by ELISA. Of 1151 samples of rainy season crop analyzed, 6% contained >20 μg kg^{-1} aflatoxin (above permissible limit) and about 40% of the samples tested positive to aflatoxins, but the toxin content in these were within the permissible limits of 1–20 μg kg^{-1}. Of 310 maize samples analyzed from postrainy season crop, 8.6% contained >20 μg kg^{-1} of aflatoxins and 46.7%

contained aflatoxins within permissible limit, and 44.7% samples tested negative. Out of 100 maize samples analyzed from storages, 20% contained >20 µg kg^{-1} aflatoxin, 76% contained permissible levels and a 4% samples were free from aflatoxins. Sixty percent of the samples analyzed tested positive to aflatoxin, but the toxin content was higher than permissible limits in only 9% of the samples. Being predisposed to *A. flavus* infestation, aflatoxin concentration may increase in the contaminated samples during storage. This indeed was reflected in stored samples, wherein 96% of the samples tested positive to aflatoxin, with 20% having >20 µg kg^{-1} aflatoxin.

A roving survey was undertaken to monitor the status of maize cultivation and to obtain information on general sowing practices of farmers in Karimnagar and Nizamabad districts to implement 'good agriculture practices (GAPs)' for maize cultivation, and to use these data for comparative purposes. Farmers fields for experimental studies to implement GAPs were selected in Gundannapalli, Vattemla, and Shatrajpalli villages in Karimnagar. Two farmers from each of these villages were selected to implement GAPs and to study the affect on pre- and post-harvest aflatoxin contamination.

Farid Waliyar, SV Reddy and P Lava Kumar

Output 1.4: Alternative uses and food/feed quality of crop produce researched and promoted

Summary

Increasing the grain yield through genetic enhancement and IPM technologies contributes to farm income and thus to improved livelihoods. Commercialization of the crop produce through quality improvement and alternative food/feed options can further contribute to these efforts of livelihood improvement. Such commercialization requires inter-institutional alliances. Thus, a DFID-funded sorghum project on "Exploring market opportunities through research, industry and users coalition: sorghum poultry feed" was successfully completed. Based on the experience in this project, which operated on a limited scale in Andhra Pradesh (India) villages, another project, funded by CFC, was launched, which pursues this research on larger scale in more number of villages in Andhra Pradesh and Maharashtra states of India (both for sorghum and pearl millet); and also includes villages in China and Thailand (for sorghum). This project is intended to accelerate the utilization of sorghum and pearl millet grains (as supplement to maize) in poultry feed and bring better economic returns to farmers cultivating these crops. Similarly, considering the growing need for alternative fodder resources for the livestock, forage research was initiated to address this demand and bring in better economic returns to farmers. Single-cross hybrids of sorghum and topcross hybrids of pearl millet were identified that had significantly higher green forage yields than the commercial sorghum-sudan grass hybrids. Further, several three-way sorghum hybrids were identified that had significantly higher green forage yield than the commercial sorghum-sudan grass hybrids. Some of these sorghum hybrids also had higher stalk sugar content and more than four times grain yield than the sorghum-sudan grass hybrids. Considering the growing importance of sweet sorghum for ethanol production to address the energy requirements, breeding materials and hybrids of sorghum having sugar yield and millable cane yield higher than the control variety SSV 84 were identified. Groundnut variety ICGV 91114 is increasingly becoming popular with farmers in the drought-prone Anantapur district of Andhra Pradesh. An on-farm IDM research involving fungicidal spray showed that pod and haulm yields of this variety can be increased by 30–40% over the non-IDM treatment; and rust severity came down to <5 from rating 7 in the non-IDM treatment on 1–9 scale, which consequently leads to improved fodder quality. Economic analysis indicated higher grosss returns from adoption of ICGV 91114 due to higher pod and haulm yields as well as higher milk yields from animals fed with this variety.

Activity 1.4.1: Documentation and synthesis of available knowledge on utilization patterns and food and feed safety

Milestone: Current knowledge on alternate uses of sorghum and pearl millet synthesized (2005)

Project completion summary report was prepared for the project DFID/NRIL – Exploring marketing opportunities through a research industry, and users coalition: sorghum poultry feed and submitted to DFID.

CLL Gowda, BVS Reddy, P Parthasarathy Rao and Farid Waliyar

Milestone: Poultry feed efficiency of sorghum and pearl millet-based rations assessed (2007)

A strong coalition between sorghum grain producers, poultry feed manufacturers, poultry federation and research institutions, and market linkages between sorghum grain producers and poultry feed manufacturers was established for enhanced use of sorghum in poultry feed. The results were compiled and published in the form of

a poster "**Sorghum-based poultry feed rations: a potential alternative to maize**" and a research article titled "**Performance of layers on sorghum-based poultry feed rations".** Further, the interviews conducted by Crop Post-Harvest Program, UK with various stake holders (scientists, project participating farmers, poultry feed manufacturers) in DFID funded project have been published as a booklet titled "Behind the market".

BVS Reddy, P Parthasarathy Rao and RP Thakur

Milestones: Food and feed safety issues of low-aflatoxin groundnut lines addressed (2006)

On-farm participatory varietals selections have been carried out for three years in Andhra Pradesh, India including 2005 rainy season (DFID aflatoxin project). Four varieties (ICGV 91278, 91328, 94379, 94434) in Anantapur area and five varieties (ICGV 91114, 91341, 93305, 94379, 94434) in Pileru area (selected by farmers from 14 originally tested) were evaluated for their yield and aflatoxin contamination. The trials were planted in 18 farmer's fields in six villages each of the two districts. Performance of the four selected groundnut improved varieties was better in all the 18 farmer's fields in six villages in Anantapur district and produced higher pod and haulm yield than the control TMV 2. Highest pod yield (1029 kg ha^{-1}) was obtained with ICGV 94434 in Cherlopalli village. The variety ICGV 94434 produced 40–43% higher pod yield in three villages and in remaining three villages it produced 23–34% higher pod yield than the control TMV 2. ICGV 94379 produced 26% and 40% higher pod yield in two villages. Overall, the mean pod yield of ICGV 94434 was 34% higher and the remaining three varieties produced 15–17% higher pod yield than the control TMV 2, which yielded 590 kg ha^{-1}. Similarly, mean haulm yield increased by 12 to 23%. The aflatoxin contamination was almost nil in two villages (Danduvaripalli and Gummalagunta) and higher level of aflatoxin contamination was observed in West Narsapuram and Cherlopalli. At West Narsapuram post-harvest rains caused delay in drying of the produce and finally resulted in high level (13–241 μg kg^{-1}) of aflatoxin contamination. ICGV 94379 was tolerant (<4.0 μg kg^{-1}) in five of the six villages and it also produced 17% higher pod yield than the control TMV 2. Overall mean (six villages pooled) of aflatoxin contamination indicated that all the four improved varieties showed reduction in aflatoxin contamination ranging from 40 to 73% over the control. The highest reduction (73%) in toxin contamination was recorded in ICGV 94379. Considering the complex nature of the aflatoxin problem in groundnut, the overall mean of the six villages indicated that the improved varieties showed good tolerance to aflatoxin contamination. In addition, these lines produced 15–34% higher pod and haulm yields than the local control TMV 2.

In Piler area, pod and haulm yields ranged from 226 to 1255 and 816 to 2654 kg ha^{-1}, respectively across the six villages. All the improved varieties produced higher pod and haulm yields than control TMV 2 in all the villages. In M.C. Palem, four of the five improved varieties produced significantly higher pod yield. Highest mean pod yield (593 kg ha^{-1}) and haulm yield (1933 kg ha^{-1}) was obtained in ICGV 94379, followed by pod yield of 586 kg ha^{-1} and haulm yield of 1835 kg ha^{-1} in ICGV 91114. On average, 16–61% increase in mean pod yield and 30–54% increase in mean haulm yield was recorded. Aflatoxin contamination ranged from 0 to 869 μg kg^{-1} across the villages and improved varieties showed lower level (90 to 242 μg kg^{-1}) of aflatoxin contamination than the control TMV 2 (310 μg kg^{-1}). In general, the aflatoxin contamination in Piler area was higher than the normal situation because of the continuous rains during pre-harvest, harvest and post-harvest stages. The improved varieties were developed with aflatoxin resistance mainly for pre-harvest situations. Since the improved varieties were exposed to adverse post-harvest rains, delayed pod drying, these became vulnerable to aflatoxin contamination. However, even in this adverse post-harvest environmental situation, there was about 36–73% reduction in overall mean aflatoxin levels in improved varieties than the control TMV 2. The highest (73%) mean aflatoxin reduction was observed in ICGV 91341, followed by 67% in ICGV 91114. ICGV 91114 recorded 59% higher pod yield than TMV 2. Therefore, our approach to combine resistance with other management practices is of major importance. In this context, low-cost aflatoxin management technologies showed encouraging results. Using some of the management practices, aflatoxin contamination was reduced by 99%.

Collective ownership of threshers by poorer farmers in the villages was due to awareness of aflatoxins and to promote early pod separation. The involvement of local NGOs and Department of Agriculture in the process also substantially increased their awareness and interest in combating the aflatoxin problem. ICRISAT and NGOs effort in facilitating the process established method of collective use of farm machinery. This helped to empower and enhance the livelihood opportunities of the women and poor farmers by increasing their incomes, as well as helping to produce groundnuts and groundnut fodder with reduced levels of aflatoxin content. The project's objective of promoting early pod stripping will also be sustainable after the projects withdrawn.

From a policy perspective, the Aflatoxin Awareness Panel's activities were instrumental in positively motivating the Government of Andhra Pradesh, India to pay conscious attention to aflatoxin awareness and

aflatoxin detection. It has complimented ICRISAT's efforts to influence the Government to set up an aflatoxin analysis laboratory in Anantapur, which was completely funded by the State Government. Now farmers in Anantapur will be able to check their farm products to target different markets and benefit from a better price.

Many awareness programs such as newspapers, flyers, TV programs, meetings and field demonstrations helped to increase awareness in the primary project area but also in many other regions in Asia and Africa. A British Broadcasting Corporation (BBC) program on ICRISAT activities was recorded for telecast around the world. As secondary impact of DFID investment in aflatoxin research, is the establishment of ELISA detection facilities and training of appropriate staff in Malawi and Mozambique is important. The access to this technology helped farmers' associations to successfully export aflatoxin-free groundnut. This technology will be further transferred to other countries in Asia and Africa.

Farid Waliyar

Activity 1.4.2: Develop technological options and institutional alliances to enhance market demand for crop produce

Milestone: Institutional alliances to promote the use of sorghum in poultry feed developed (2006)

In March 2005, a project supported by DFID on "Exploring marketing opportunities through research, industry and users coalition: sorghum poultry feed" was successfully concluded involving selected villages of Ranga Reddy and Mahabubnagar districts of Andhra Pradesh, India. Encouraged by the success, another project to up-scale the findings by expanding the coverage to more areas -Udityal in Andhra Pradesh, Parbhani and Beed districts in Maharashtra in India; Beizhen, Heishan and Yi provinces in China; and Suphan Buri, Kanchana Buri and Nakon Sawan provinces in Thailand–was successfully negotiated with CFC/FAO and received the approval for funding. The project implementation was launched in May 2005. The work, which involves coalition building among various stakeholders, is in progress and the highlights of progress achieved during year1 are described below.

Based on work plans of the project in year-1, in Andhra Pradesh, two clusters-Udityal cluster (6 villages) and Palavai cluster (5 villages) comprising of 668 farmers were selected in Mahabubnagar district; and in Maharashtra three clusters-Koak cluster (5 villages) in Parbhani district, Anjanpur cluster (6 villages) in Ambajogai Taluk and Patoda cluster (5 villages) in Patoda Taluk in Beed district comprising of 1050 farmers were selected in India. Two clusters one each in Beizhen, Heishan and Yi provinces in China, and Suphan Buri, Kanchana Buri and Nakon Sawan provinces in Thailand were identified.

ICRISAT, the Project Executing Agency (PEA) with the support of coalition partners' facilitated the formation of farmers associations in all the clusters. The capacity building needs of the partners and farmers were identified and strengthened by conducting various training programs. Initially, farmers were made aware of improved cultivars and production technologies for sorghum and pearl millet for enhancing the grain production. Several training programs, exposure visits, and on-farm meetings were conducted on the use of improved crop production technologies. Innovative input chain and marketing linkages were developed for marketing surplus grain. Farmers' capacity on grading, bulking and marketing of surplus grain produce were built. As a result of this, farmers from one of the clusters (Anjanpur) in Maharashtra (India) could market their sorghum grain at 10% higher than the prevailing price in the market.

A Alur, Ch Ravinder Reddy, BVS Reddy, P Parthasarathy Rao and CLL Gowda

Activity 1.4.3: Improve fodder yield and quality for enhanced ruminant nutrition

Milestone: Effective IPM technologies to enhance fodder quality of dual-purpose and pest resistant sorghum and groundnut varieties validated (2005)

On-station evaluation of early and medium-maturing groundnut cultivars for higher quality and quantity of haulms and pods

Groundnut provides valuable edible oil for human and nutritious fodder for cattle in Deccan Plateau, India. Pod and fodder yields are very low in groundnut due to several diseases. Among these diseases, late leaf spot [(LLS) *Phaeoisariopsis personata*] and rust (*Puccinia arachidis*) are most destructive and often cause severe losses in quantity and quality in farmers' fields. Cultivars having moderate resistance to foliar diseases, when combined with moderate levels of management (economical use of fungicide), produce higher quantities of healthy fodder as well as pods. Healthy fodder has high digestibility and increases milk yield in dairy cattle. Therefore, to

identify high-yielding dual-purpose groundnut cultivars with economical level of foliar disease management, a replicated trial consisting of 10 early- and six medium-maturing genotypes with six disease management levels was conducted at ICRISAT-Patancheru during 2004 rainy season. Highly susceptible cultivar TMV 2 was included as one of the entries for comparison. Plot size was four rows of 9 m with 60 × 10 cm inter- and intra-row spacing. Disease management levels using fungicide chlorothalonil (Kavach @ 2 g L^{-1} water) were: (1) no spray, (2) one fungicide spray at 60 days after sowing (DAS), (3) two fungicide sprays at 60 and 75 DAS, (4) three fungicide sprays at 60, 75 and 90 DAS (for medium-maturing cultivars only), (5) weather-based advisory system using leaf wetness counter and (6) continuous fungicide spray from 30 DAS till maturity with 10-day interval. Design of the experiment was split-plot with spray schedules as main plots and genotypes as sub-plots. Severity of foliar diseases was recorded on 1–9 rating scale at regular intervals of crop growth. At harvest, dry weights of fodder and pods were recorded and yield per hectare was calculated.

One fungicide spray at 60 DAS for early maturing and two fungicide sprays at 60 and 75 DAS for medium-maturing genotypes gave higher yields and healthy fodder with low foliar disease severities. Lowest foliar disease severities (3.7 to 4.3 for LLS and 5.3 for rust) and highest healthy fodder (2.0 to 2.5 t ha^{-1}) and pod yields (1.46 to 1.6 t ha^{-1}) were recorded in ICGV 99201, ICGV 99206 among early-maturing cultivars. Among medium-maturing cultivars, lowest disease severity (up to 4.0 for both LLS and rust), highest fodder (up to 3.8 t ha^{-1}) and pod (1.59 to 1.74 t ha^{-1}) yields were obtained in ICGV 99032 and ICGV 99054.

S Pande

On-farm validation and promotion of integrated disease management (IDM) technology in groundnut

One hundred and twenty one farmers were selected from five villages, Jalalapuram (43), Lingareddypalli (46), Talupuru (22), Antaraganga (5) and Jonnalakothapalli (5) in Anantapur, India in the year 2004 rainy season to promote IDM technology that involved fungicidal spray schedule mentioned above. Nearly 80% of the trials during this season were sown with short-duration dual-purpose cultivar ICGV 91114, as this cultivar was preferred by several farmers. Other cultivars included in these trials were ICGV 89104 (early-maturing); and ICGV 92020 and ICGV 92093 (medium-maturing). All these trials were conducted in close collaboration with District Agricultural Advisory and Transfer of Technology Center (DAATTC), Acharya NG Ranga Agricultural University (ANGRAU), Anantapur and Accion Fraterna/RDT-Anantapur. Each trial was planted in 0.2–0.3 ha and was compared with local cultivar, (JL 24/TMV 2) for pod and fodder yields. In addition to these trials, farmers in several villages in the states of Andhra Pradesh, Karnataka and Tamil Nadu (India) adopted the technology during 2004 rainy season. Plantings were completed by 12 July 2004 due to arrival of timely monsoons in all the selected villages. Fortunately, unlike the previous years, there was a good distribution of rains and pod and fodder yields were high compared to previous years. Mean severity of foliar diseases across the villages in the most preferred cultivar, ICGV 91114 with IDM was <5 rating on 1–9 rating scale and fodder (haulm) and pod yields, respectively, were 2.46 and 2.08 t ha^{-1}, while severity of foliar diseases in non-IDM plots was around 7 rating and haulm and pod yields were 1.94 and 1.41 t ha^{-1}. Thus, the dual-purpose cultivar ICGV 91114 was found suitable under rainfed cultivation in Anantapur district of Deccan Plateau, Andhra Pradesh, India.

S Pande

Promotion of improved foliar disease resistant groundnut cultivars in the Deccan Plateau of India

Groundnut is grown extensively in the Deccan Plateau of India (75% of cropped area) by resource poor farmers; and groundnut haulms are the major source of home grown fodder for their animals. However, Anantapur district (Andhra Pradesh) in the heart of the Deccan Plateau faces fodder shortage of around 10 to 15% of the total requirement varying from 240,000 t in a drought year to 75,000 t in a normal year. Within the district, in about one-fourth of the 63 mandals (divisions), the fodder shortage is more acute. These mandals fall in the low to medium rainfall category and are located mainly in the western and central part of the district. The state government is trying to mitigate the fodder shortage through various programs that include community fodder cultivation on tank beds, supply of straw on subsidy, cattle camps, demonstrating the use of azolla (an alternate and less expensive livestock feed with unique combination of proteins, minerals, vitamins and essential amino acids), supply of fodder seed for cultivation on private lands, etc. However, these programs are not able to mitigate the shortage and a majority of farmers are buying crop residues, mainly paddy straw. The preference for paddy straw is twofold: as a cereal supplement to groundnut haulm, and as a cover to protect the staked groundnut haulm from rains. A majority of those purchased paddy straw were marginal and small farmers.

The main source of supply is from the command area of Tungabhadra river high-level canal irrigation (HLCI) that passes through the central and northwestern part of the district. Here paddy accounts for more than 10–15% of

cropped area. Villages within a radius of 50–60 km of these command areas buy their supplies of paddy straw immediately after harvest. During drought years, villages at a distance of 150–200 km also get their supplies from the canal irrigation areas. Additionally, during summer months and acute drought years, the state government procures paddy straw from the coastal districts that are 400–500 km away.

Since farmers were keen to augment their own fodder resources, they identified new groundnut cultivar ICGV 91114, resistant to foliar diseases to meet their multiple requirements ie, higher pod yield and haulm yield to improve food security and increased incomes from sale of pods, haulms, and dairy products. Economic analysis indicated higher gross returns from adoption of ICGV 91114 due to higher pod yields, higher haulm yields, and higher milk yield from animals fed with these haulms.

Based on reconnaissance surveys in 2005, it was found that the improved groundnut cultivar ICGV 91114 is grown in more than 120 villages covering 4 districts in Andhra Pradesh state and 3 districts in Karnataka state. Two-thirds of the villages are located in Anantapur district. The location of these villages was recorded using GPS instrument. A majority of the villages are located in the low to medium rainfall mandals indicating the importance of this cultivar for resource poor farmers in marginal environments. Tracking these villages at some future point would give insights into factors leading to faster spread in some locations as compared to others. The spread of the improved cultivar as documented is, however, an underestimate since information on adoption was not recorded from several villages located in the hinterlands, ie, far away from state or national highways.

The potential impacts on marginal and small farmers would depend on the linkages between small farmers, the public sector actors (seed sector, extension, marketing, etc.) and other stakeholders in the groundnut and the milk economy (dairy cooperatives, etc.). The linkage between adoption of the improved technology, higher incomes, asset acquisition reinvestment in agriculture and improvement in overall quality of life will have to wait until the technology is adopted on a larger scale at the household level. At present, it is observed that a majority of the farmers are risk averse and hence are adopting the new technology only on 10–20% of their cropped area.

S Pande and P Parthasarathy Rao

Milestone: Village level seed multiplication system established and effect of plant diseases on yield and nutritive value of crop residues of groundnut and sorghum assessed (2005)

To study the effect of plant diseases on yield and nutritive value of groundnut haulms in relation to animal health, 72 haulm samples from six groundnut varieties (ICGS 44, ICGS 11, DRG 12, ICGV 86325, ICGV 92020 and ICGV 92093) were collected and processed by ELISA for aflatoxin contamination. The aflatoxin contamination in these samples ranged from 0 to 33 $\mu g\ kg^{-1}$ and about 25% of the samples were found to be contaminated with the toxin. Among the six groundnut varieties, all the 15 haulm samples from ICGV 86325 were contaminated in the range of 1–233 $\mu g\ kg^{-1}$. In remaining varieties, the aflatoxin contamination level was very low.

Milk samples from the feeding trials (conducted with farmers in Anantapur district, India) were collected and analyzed by ELISA for aflatoxin M1 contamination. Aflatoxin M1 concentration in these samples ranged from 0 to 15 $\mu g\ kg^{-1}$. Of the 328 samples tested, 42% contained >0.5 $\mu g\ kg^{-1}$ (non-permissible level) of aflatoxin M1 contamination.

Farid Waliyar

Establishment of village-level seed system for groundnut in Anantapur district, India: Groundnut is grown extensively in the Deccan Plateau of India by resource-poor farmers. Seed is very important and expensive component of groundnut cultivation. There is no organized seed system for groundnut in this region. Few farmers store their rainy season produce as seed for the following season. Most of the poor farmers depend on large farmers or government agencies for the seed. Since seed to grain multiplication ratio is very low in groundnut, it is not possible for any single agency to supply the seed to all the groundnut growing farmers. Therefore, a village-level seed multiplication system was established in four villages in the district Anantapur in collaboration with District Agricultural Advisory and Transfer of Technology Center (DAATTC), ANGRAU-Anantapur, and Rural Development Trust (RDT) NGOs, during 2004 post rainy season. Six farmers in the village Jalalapuram, nine farmers in the village Lingareddypally, four farmers in the village Talupuru and three farmers in the village Vasanthapuram in the district Anantapur participated in the seed multiplication of the cultivar ICGV 91114 in 12 ha during the 2004 postrainy season. The produce was sold to many other farmers as seed in all these villages.

Establishment of village-level seed system for chickpea in Nepal: Chickpea is one of the important grain legume crops in Nepal and is being cultivated profitably in rice-fallow lands. Non-availability of seeds of improved chickpea varieties is one of the constraints for low yields in this country. Seed is an important component of Nepalese farming systems. Formal seed system is lacking in this country and farmers obtain seed for planting mostly from previous harvest and thus there is no difference between seed and grain. Resource poor farmers obtain seed from others or purchase from local market. Therefore, to ensure continuous supply of good quality seed, we initiated farmer seed system in collaboration with National Grain Legumes Research Program (NGLRP) and National Oilseeds Research Program (NORP) for producing the seed of improved variety Avarodhi during 2004/05 postrainy season. This system is run by a few farmer groups/self help groups (SHG). Nine farmers in the Rajahar village, Nawalparasi district, and six farmers in the village Bardibas, Mohattari district multiplied the cultivars Avarodhi and Tara during 2004 postrainy season. Grain yield in Avarodhi was 1620 kg ha^{-1} in the Bardibas village and 818 kg ha^{-1} in the Rajahar village, while that of Tara was 800 kg ha^{-1} in the village Rajahar and 500 kg ha^{-1} in the village Bardibas. All the seed was sold to other farmers in the village for sowing in the 2005 postrainy season. Thus, the village-level seed system was found successful in these two villages in Nepal.

On-station evaluation of the effect of plant diseases on yield and nutritive value of crop residues of dual-purpose groundnut cultivars: Late leaf spot [(LLS) *Phaeoisariopsis personata*] and rust (*Puccinia arachidis*) are destructive diseases and cause severe losses in yield and nutritive quality of crop residues. Management of these foliar diseases increases the yield as well as it enhances the quality of the crop residue. Therefore, a replicated trial consisting of 10 early- and six medium-maturing genotypes with five disease management levels to identify high-yielding dual-purpose groundnut cultivars was conducted at ICRISAT-Patancheru during 2004 rainy season. Highly susceptible cultivar TMV 2 was included as one of the entries for proper comparison. Plot size was four rows of 9 m with 60 × 10 cm inter- and intra-row spacing. Disease management levels using fungicide chlorothalonil (Kavach @ 2 g L^{-1} water) were (1) no spray, (2) one fungicide spray at 60 days after sowing (DAS), (3) two fungicide sprays at 60 and 75 DAS, (for medium-maturing cultivars only), (4) continuous fungicide spray from 30 DAS till maturity with 10-day interval. Design of the experiment was split-plot, with spray schedules as main plots and genotypes as sub-plots. Severity of foliar diseases was recorded on 1–9 rating scale at regular intervals of crop growth. The quality of crop residue, ie, haulms was found superior in fungicide-sprayed plots of early and medium maturing cultivars than unsprayed plots of TMV 2. Foliar disease severity rating was up to 5 on a 1–9 rating scale in the plots that received one fungicide spray at 60 DAS (early-maturing cultivars) and two fungicide sprays at 60 and 75 DAS (medium-maturing cultivars) compared to 9.0 rating in unsprayed plots of susceptible TMV 2. Among early-maturing cultivars with one fungicide spray at 60 DAS, haulm and pod yields were significantly higher in ICGV 99201 (2.40 and 1.65 t ha^{-1}) and ICGV 99206 (2.5 and 1.46 t ha^{-1}) than TMV 2 (1.79 and 0.81 t ha^{-1}). Among medium-maturing genotypes, with two sprays at 60 and 75 DAS, haulm and pod yields respectively were 3.8 and 1.74 t ha^{-1} in ICGV 99032 and 3.9 and 1.74 t ha^{-1} in ICGV 99054 compared to 1.79 and 0.81 t ha^{-1} in TMV 2.

Suresh Pande

Milestone: High-tillering sorghum population further improved for high biomass yield and quality traits (2006)

Population improvement with recurrent selection involving male-sterility-inducing gene (ms_3), besides offering greater opportunity for increasing the frequency of existing desirable genes/traits, would allow introgression of other genes/traits of importance. Such improved and/or introgressed trait-specific population would provide valuable base material for the development of broad-based trait-specific hybrid parents and high-yielding varieties. At ICRISAT-Patancheru, a high tillering population (ICSP-HT) is being improved for high tillering, sweet-stalk trait and resistance to foliar diseases.

The sweet-stalk varieties, SSV 74 and SSV 84 were crossed to male sterile plants of ICSP-HT population to introgress sweet-stalk trait. The ICSP-HT population (C_9) bulk and F_2 crosses bulk (derived from crossing SSV 84 and SSV 74 with male-steriles of ICSP-HT population) were evaluated in rainy season 2005. Selections were made separately for male-sterile and male-fertile plants in the population as well as in bulks. The bulk was advanced to F_3 and the population bulk was advanced to C_{10}. From population bulk, 181 male-sterile and 86 male-fertile plants were selected. From ICSP-HT population × SSV 84 cross bulk, 31 male-sterile and 35 male-fertile plants were selected; and from ICSP-HT population × SSV 74 cross bulk, 57 male-sterile and 61 male-fertile plants were selected. The C_{10} population bulk and F_3 crosses bulks was reconstituted by mixing the seed of male-sterile and male-fertile plants in 3:1 ratio (separately for each bulk).

BVS Reddy and S Ramesh

Milestone: Sorghum and pearl millet hybrid parents with high forage yield potential and forage quality developed (2006)

In recent years, there has been a growing demand for fodder resources due to spurt in dairy industry in India, especially in peri-urban areas. Because of its quick growth, ratoonability and tolerance to biotic and abiotic stresses, sorghum is an excellent candidate crop for meeting the increasing demand for both green and dry fodder (stover).

Germplasm lines and the breeding lines (290) were evaluated during the 2004-05 postrainy season for high stalk-sugar and biomass. Some of the trait-specific B-lines developed earlier in different programs were evaluated for biomass production ability and stalk-sugar content. Also, some hybrids made in the postrainy season were evaluated for the above two traits in the summer nursery. Besides these, tillering lines selected from different programs were evaluated for green fodder yield and stalk-sugar content. Based on these activities, the following trials (constituted from selected lines from the postrainy season) were conducted in the 2005 rainy season.

Sweet sorghum preliminary varieties/restorers trial (SSPVRT): The germplasm and the breeding lines (290) were evaluated in an un-replicated nursery; and 42 sweet-stalk varieties and restorers were evaluated in a replicated trial during the 2004-05 postrainy season for stalk-sugar content and biomass. Based on the performance for these traits, 99 lines were selected and evaluated in a replicated trial along with controls NSSH 104, SSV 74 and SSV 84 during 2005 rainy season. Brix reading was taken at 18 days after 50% flowering. The sugar yield was estimated based on Brix reading and juice yield. Some of the breeding progenies, SP 4481-1 (6.5 t ha^{-1}), SP 4484-2 (6.4 t ha^{-1}) SP 4504-3 (6.1 t ha^{-1}), SP 4484-3 (5.6 t ha^{-1}) and SP 4511-3 (5.4 t ha^{-1}) were significantly superior to the control SSV 84 (3.2 t ha^{-1}) for sugar yield based on Brix reading and juice yield. The millable cane yield of these progenies (64.9 to 102.8 t ha^{-1}) was also significantly superior to that of SSV 84 (49.8 t ha^{-1}). These progenies will be further advanced with selection.

Sweet sorghum advanced B-lines trial (SSABLT): A total of 67 high-yielding and trait-specific B-lines developed earlier in different programs were evaluated for biomass production ability and stalk-sugar content along with the controls SSV 74, SSV 84 and 296B in a sweet-stalk preliminary B-lines trial. Based on the Brix reading at maturity, 43 B-lines were selected and evaluated in SSABLT along with the same controls during 2005 rainy season. None of the B-lines performed better than the controls SSV 74 and SSV 84 for sugar yield based on Brix reading and juice yield. However, 13 B-lines (1.4 to 2.7 t ha^{-1}) significantly outperformed the control 296B (0.6 t ha^{-1}) for sugar yield.

Sweet sorghum preliminary hybrid (non-tillering) trial (SSPHT): A total of 158 hybrids involving 25 female parents and 14 male parents were evaluated during 2005 rainy season. Based on the sugar yield estimated at flowering stage, top performing 25% of the lines were selected and the sweet-stalk parameters were recorded at maturity. The hybrid ICSA 675 × SSV 74 (6.7 t ha^{-1}) was significantly superior to NSSH 104 (4.3 t ha^{-1}) for sugar yield, and comparable for total soluble sugars (12.1% as against 13.5% in NSSH 104) . Eleven hybrids (4.4 to 5.5 t ha^{-1}) were numerically superior to NSSH 104 for sugar yield.

Sweet sorghum advanced hybrids (non-tillering) trial (SSAHT): A total of 30 selected sorghum hybrids were evaluated along with the controls SSV 74, SSV 84 and NSSH 104 during 2005 rainy season. None of the hybrids were comparable to the control NSSH 104 (5.5 t ha^{-1}) for sugar yield. However, nine hybrids (25.2 to 35.1 t ha^{-1}) were comparable to NSSH 104 (32.7 t ha^{-1}) for juice yield, and three hybrids (15.8 to 17.2%) were comparable to NSSH 104 (18.0%) for Brix reading at maturity.

Sorghum forage and tillering lines trial (SFTT): High tillering lines (61) selected from different programs were evaluated along with the controls SSG 59-3, GK 908 and GK 911 for green fodder yield and stalk-sugar content at 50% flowering during 2005 rainy season. A total of 22 hybrids (89.2 to 160.3 t ha^{-1}) significantly outperformed the controls SSG 59-3 (51.7 t ha^{-1}), GK 908 (60.5 t ha^{-1}) and GK 911 (60.4 t ha^{-1}) for fresh fodder yield. The stalk-sugar content of 15 of these 22 lines (11.2 to 15.8 Brix) significantly outperformed the control GK 908 (Brix 8.1%). These lines are promising for further selection and multilocation testing.

Three-way cross trial: Most of the private sector seed companies develop and market three-way hybrids for forage purposes, using grain sorghum male-sterile lines as seed parents and sudan grass lines as male parents. In order to evaluate the potential of ICRISAT-developed grain sorghum male-sterile lines and forage restorer lines/varieties in hybrid combinations, 87 three-way hybrids were made by crossing forage/sweet-stalk varieties onto male-sterile single cross F_1s (obtained by crossing grain sorghum A-lines with non-isogenic B-lines). These

were evaluated in a replicated trial for forage yield potential and stalk-sugar content at grain maturity during the 2005 rainy season. Forty-seven hybrids (28.8 to 46.6 t ha^{-1}) outyielded popular control variety SSG 59-3 (21.7 t ha^{-1}) for green fodder yield. The Brix reading of these selected hybrids ranged from 5.5 to 18.4% against 12.9% in SSG 59-3. Two test hybrids [(ICSA 351 × ICSB 394) × ICSV 25263] (Brix 18.4%) and [(ICSA 73 × ICSB 369) × ICSR 93025] (Brix 17.6%) had significantly higher stalk-sugar content than SSG 59-3 (Brix 12.9%). Further, stalk-sugar content of 30 hybrids (Brix ranging from 17.1 to 12.9% was either numerically higher or on par with of SSG 59-3 (Brix 12.9%). The grain yield potential of many of these hybrids (0.8 to 2.7 t ha^{-1}) was significantly superior to that of the control SSG 59-3 (0.2 t ha^{-1}).

BVS Reddy, M Blümmel and S Ramesh

Hybrid yield potential of forage-type male-sterile lines: Among the seed parents (A-lines) developed for grain/dual-purpose hybrid production, eight A-lines had potential as seed parents of forage hybrids. Three of these male-sterile lines (ICMA 89111, ICMA 00999 and ICMA 03222) were crossed with each of the nine forage populations to produce 27 experimental hybrids, which were evaluated along with seven controls comprising 2 pearl millet forage hybrids, 3 dual-purpose hybrids, 1 OPV (WC-C 75) and 1 sorghum-sudan grass hybrid (GK 908) for forage yield at 80-day harvest. From amongst the controls, earlier identified forage pearl millet hybrid ICMA 00999 × IP 17315 (14.3 t ha^{-1}) had 13% higher dry forage yield than GK 908 (12.7 t ha^{-1}). Of the test hybrids, seven hybrids (3 on ICMA 89111 and 4 on ICMA 00999) had 14.3-16.3 t ha^{-1} dry forage yield. All these experimental hybrids flowered in 64–78 days (74 days for the control hybrid). Forage varieties such as ICMV 05222 and ICMV 05111 (both tall, thick stem type) had significant positive gca effects for dry forage yield at 80-day harvest, plant height and days to 50% flower, indicating delayed flowering and plant height were associated with increased dry forage yield. The highest yielding hybrid was again on ICMA 00999 that involved ICMV 05222 as the population pollen parent.

KN Rai, VN Kulkarni and M Blümmel

Output 1.5: New technologies evaluated, disseminated and their impact documented

Summary

Crop production and protection technologies developed at research stations must be validated on-farm for their relevance in terms of yield gains, farmers' acceptance and cost effectiveness. This should be backed by technology transfer efforts to enhance the pace and scale of their adoption. Impact assessment must constitute an integral part of technology development and dissemination.

An Integrated Crop Management (ICM) technology for chickpea that consists of a BGM-tolerant cultivar ICC 14344 (Avarodhi), fungicidal treatment, wider row spacing, application of rhizobium, DAP fertilizer, and judicious use of pesticides to control BGM and pests was adopted by 20,000 farmers in rice-fallow lands in 14 districts of Terai region of Nepal (Fig. 1.3). In addition, 250 farmers planted these ICM trials in two villages in central Nepal. Six ICRISAT-bred chickpea varieties were evaluated in on-farm participatory variety selection in Uttar Pradesh, Haryana and Uttaranchal states of India, where chickpea was intercropped with sugarcane. BGM severity was lower and grain yields were higher in all the improved cultivars as compared to the locals. The large-seeded kabuli variety KAK 2 emerged as the most preferred variety. An impact assessment report on chickpea adoption in Ehiopia was published. Results of a survey conducted with 300 households in four chickpea growing districts in central Ethiopia showed 6 to 66% variation in adoption across the districts, which resulted largely from the lack of awareness of improved varieties among the farmers. Of the five varieties that had been adopted, Mariye had the largest coverage. Important characteristics influencing adoption were high yield, early maturity and drought tolerance.

Groundnut variety ICGV 91114 has been rapidly spreading in the Anantapur district of Andhra Pradesh, grown by 450 farmers in 45 villages in 371 ha just for seed increase. Farmers' preference for this variety over the traditional TMV 2 is due to its earliness, ability to tolerate mid-season and terminal drought, tolerance to pests and diseases, more number of uniform pods, and higher pod and haulm yields. This variety has now also spread to three districts of the neighboring Karnataka state.

Results of an initial evaluation of short-duration pigeonpea in the rice-fallow areas of northern Philippines appeared promising as are the prospects of vegetable pigeonpea for local consumption, and perennial pigeonpea for rehabilitation of degraded lands.

*Two International Chickpea Screening Nurseries, (ICSN-*Desi *and ICSN-*Kabuli*), were evaluated by NARS scientists during 2004/05. Thirty sets of ICSN-*Desi *and 29 sets of ICSN-*Kabuli *were distributed to NARS*

scientists of 9 countries for evaluation during 2005/06. Forty-one sets of International Groundnut Varieties Trials (4 foliar diseases resistant, 8 drought tolerant, 7 short-duration, 7 medium-duration, 3 aflatoxin resistant and 12 confectionery) were supplied to collaborators in Afghanistan, Bangladesh, China, India, Indonesia, Philippines, South Africa and Sudan. Four sets of a new international trial, formulated with red-seeded groundnut varieties and advanced breeding lines, were dispatched to the Project Facilitation Unit (PFU), Tashkent, Uzbekistan. These sets will be evaluated in Uzbekistan, Tajikistan, Armenia and Georgia.

On specific seed requests (including selections made during the Scientists Field Days), we supplied 12,659 seed samples of sorghum (5648), pearl millet (4273), chickpea (1853), and pigeonpea (885). Also, we supplied 527 breeding lines and 237 segregating populations of groundnut. Eight varieties of chickpea (3) and groundnut (5), and one hybrid of sorghum were released by NARS during 2005.

Activity 1.5.1: Farmer participatory research and development

Milestone: Effectiveness of farmers' participatory selection of sorghum for postrainy season adaptation evaluated (2005)

In order to assess the effectiveness of farmers' participatory selection for postrainy season adaptation, a replicated trial consisting of 52 lines (26 farmer selection and 26 breeder selection) adapted to postrainy season that was selected from F_6 generations were evaluated at ICRISAT-Patancheru; Regional Agricultural Research Station, Bijapur; and Akola (India) during the 2004–05 postrainy season. Data received from Bijapur along with Patancheru were analyzed and the results are reported below. The data from Akola were not received.

Data were recorded for days to 50% flowering, plant height (m), stay-green score (taken on a scale 1 to 5 at harvest, where 1 = >75% green, 2 = up to 75%, 3 =2 6–50%, 4 = 10–25% green, and 5 = <10% green), lodging score (taken on a scale 1 to 5 where 1 = no lodging, 2 = up to 25% lodged, 3 = 26–50%, 4 = 51–75%, 5 = >75% of plants lodged), grain lustre and shape score (taken on a scale 1 to 5, where 1 = lustre and globular like M 35-1, 2 = lustre but not globular, 3 = less lustre but globular, 4 = less lustre and flat and 5 = less lustre and flat with beak), grain size (g 100^{-1}) and grain yield (t ha^{-1}). At both Bijapur and Patancheru, the mean grain yield of breeders' selections (Patancheru: 3.2 t ha^{-1}, Bijapur: 1.1 t ha^{-1}) was marginally superior to that of farmers' selections (Patancheru: 2.7 t ha^{-1}, Bijapur: 1.1 t ha^{-1}). However, grain size of farmers' selections (Patancheru: 3.2 g 100^{-1}, Bijapur: 3.3 g 100^{-1}) were marginally superior to that of breeders' selections (Patancheru: 3.1 g 100^{-1}, Bijapur: 3.0 g 100^{-1}). Farmers' selections were more tolerant to lodging and senescence and were late maturing than breeders' selections at both the locations. However, for grain lustre, contrasting results were obtained in two locations. While, farmers' selections were more lustrous at Bijapur, breeders' selections were more lustrous at ICRISAT-Patancheru. From these results, it is clear that farmers prefer bold grains and also fodder traits such as stay-green and non-lodging. Breeder selections were based on grain yield, early maturity and tan plant color. However, the differences between farmers' and breeder's selections for these traits were marginal to be of any significance. Based on the performance of these genotypes for grain yield, grain lustre and size and fodder yield, 4 lines in the farmers' selection group and 5 lines in the breeders' selection group were selected for further evaluation.

BVS Reddy and S Ramesh

Milestone: Farmer-participatory on-farm research on PVS and ICM in legumes implemented (2005)

The Year 4 work plan of farmer participatory varietal selection trials of IFAD TAG 532-ICRISAT project at various locations in four countries (China, Nepal, India and Vietnam) was successfully implemented. At ICRISAT-Patancheru, during 2004/05 postrainy season, 9.7 t of 8 varieties and during 2005 rainy season, 3.5 t breeder seed of three varieties was produced and supplied to different seed producing agencies.

The improved variety ICGV 91114 is winning over TMV 2 in Anantapur district, India. The variety is now being grown by 450 farmers in 45 villages spread over 371 ha in the district for seed increase. During the field visits in different villages and farmer-scientist interaction meetings, farmers rated this variety as superior to TMV 2 because of its earliness, ability to withstand mid- and end-of-season droughts, tolerance to diseases and pests, more numbers of uniform pods and higher pod and haulm yields. About 6.5 t seed of farmer-preferred groundnut varieties were made available to collaborators in Anantapur, Chhattisgarh, Gujarat and Orissa states of India.

A short-duration groundnut variety ICGV 92195, proposed by Maharana Pratap University of Agriculture and Technology (MPUA&T), Udaipur, Rajasthan, India was released by Central Varietal Release Committee as

'Pratap Mungphali-2' for zone II (Rajasthan and Gujarat) in India. National Agricultural Research Council released two high-yielding varieties, ICGV 86300 and ICGV 90173 as Rajarshi and Baidehi, respectively, in Nepal. ICGV 93468, a short-duration variety is proposed for release as 'Avtar' in Uttar Pradesh, India for spring season cultivation. This variety is already popular with the farmers and was cultivated on 59,000 ha during the 2005 spring season. A confectionery variety, AK 303, selected from a F_4 population [(ICGV 88384 × JL 24) × (ICGV 88438 × ICG 5240) F_1)], is proposed for release in Maharashtra. Of the material supplied to cooperators in India in the past, 47 lines were included in multi-location trials in different states. Of the 21 new varieties proposed for inclusion in All India Coordinated Varietal Trials, 10 varieties either have ICRISAT supplied germplasm or breeding lines in their parentage, or are direct introductions (ICGV 91114 by ARS, Anantapur; ICGV 98223 by MPUA&T, Udaipur; and four large-seeded varieties ICGV 96110, ICGV 97045, ICGV 98396 and ICGV 98412) by the Project Coordinator of All India Groundnut Improvement Project.

After one more year of on-farm trials, Ministry of Agriculture, Forestry and Fisheries (MAFF) intend to propose two groundnut varieties (ICGV 88438 and ICGV 95278) for release in East Timor. This year we have supplied 10 new varieties consisting of ICGV# 95070, 96172, 95069, 96165 and 97128 (high yielding) and 98379, 98381, 98378, 98375 and 99017 (foliar disease resistant) for evaluation in East-Timor.

SN Nigam and R Aruna

Training camps in IPM and HNPV production: During the 2005, in order to strengthen the NARS capacity, 240 farmers and 314 researchers were given training in IPM with special emphasis on HNPV production. Under Development Marketplace (DM) 2005 so far 104 (78 farmers and 26 NARS staff) members of the farming community were given in-depth training in HNPV production at ICRISAT Center, Patancheru.

GV Ranga Rao

Monitoring and evaluation of on-farm participatory chickpea ICM trials in Nepal: Chickpea is the most important grain legume in Nepal. Its production has drastically decreased since last two decades in this country due to wilt, root rots, botrytis gray mold (BGM), Helicoverpa pod borer, boron deficiency and poor nodulation. Technologies are available to manage these constraints. These single factor management measures were integrated as an integrated crop management (ICM) technology for the overall management of chickpea crop in Nepal. The ICM technology was evaluated and scaled up in Terai region of Nepal through participatory approaches by involving large number of farmers. In collaboration with scientists from Nepal Agricultural Research Council (NARC), 250 farmers participatory ICM trials were planted in two villages, Rajahar (50 trials) and Bardibas (200 trials) of Central Nepal during 2004/05 season. These trials were in addition to about 20,000 farmers growing chickpea in rice-fallow lands in 14 districts in Nepal (Fig. 1.3). The technology consisted of BGM-tolerant cultivar ICC 14344 (Avarodhi), fungicide seed treatment, wider row spacing, application of Rhizobium, fertilizer (DAP), and judicious use of pesticides to control BGM and pod borer. Plant stand was significantly higher in ICM plots than non-ICM plots across the locations. Severities of BGM and pod borer damage were significantly lower in ICM plots than non-ICM plots in all the trials in all locations. Mean grain yield was 1.35 t ha^{-1} (0.8 to 2.0 t ha^{-1}) in the cultivar Avarodhi and 1.47 t ha^{-1} in the cultivar Tara across the locations.

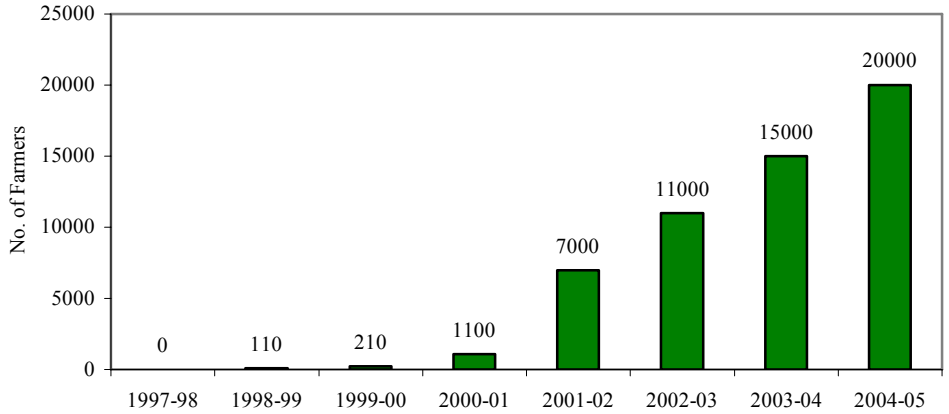

Figure 1.3. Adoption of ICM technology of chickpea in Nepal

Monitoring and evaluation of chickpea on-farm ICM and PVS trials in north western plains of India: Under crop diversification and resource conservation technology of Rice-Wheat Consortium (RWC), two chickpea trials [ICM trial and participatory varietal selection (PVS) trial] were conducted in collaboration with the Project Directorate for Cropping Systems Research (PDCSR), Modipuram, India in several farmers fields in the villages Barkhanda, Mohammadpur, Kazampur (district Ghaziabad), Ielnna (Bulendshahar district), Uttar Pradesh; Taprana (district Karnal) Dungali, Sirsali, Adhoya, (district Kurukshetra, Haryana), village Kitcha [district Udham Singh Nagar, Uttaranchal (UA)] and also at the research farm of Project Directorate for Cropping Systems Research (PDCSR) and Agricultural and Processed Food Products Exports Development Authority (APEDA), Modipuram, Uttar Pradesh, India.

ICM trial: Growing chickpea with ICM technology as a highly remunerative crop after rice for diversifying rice wheat cropping system was advocated in several villages in Indo-Gangetic Plain (IGP) of India, Nepal and Bangladesh. ICM technology consisted of improved high-yielding cultivar, fungicide seed treatment, wider row spacing, and need-based application of pesticides to manage diseases and insects. In most of the locations, chickpea was planted either on beds or by zero tillage, as intercrop with sugarcane in the furrows. BGM was observed (<4 rating on 1–9 rating scale) in IPM plots, while it had 7 rating in non-IPM plots across the locations. The low disease in IPM plots was attributed to timely application of fungicide Bavistin. Mean grain yield was almost double in IPM plots than non-IPM plots across locations.

Participatory varietal selection trial: Six ICRISAT-bred improved chickpea varieties (ICCV 2, JG 11, ICCV 10, ICCC 37, KAK 2 and Vihar) were included in a participatory varietal selection (PVS) trial that was conducted in several villages in participatory mode in farmers' fields in the states of UP, Haryana, Uttaranchal and also at research farms of PDCSR and APEDA, Modipuram. In several locations, chickpea was planted in the furrows as intercrop with sugarcane. Severity of BGM was recorded low (up to 5 rating) in all the improved cultivars compared to local cultivar (up to 8 rating). Mean grain yields were high in all the improved cultivars than local cultivars in all the trials. Of the improved cultivars, KAK 2 was most liked by many farmers.

PDCSR, Modipuram: All the six entries of the PVS trial ICCV 10, JG 11, ICCC 37, KAK 2, ICCV 2 and Vihar were planted as paired rows on beds and on flat at the research farm of PDCSR, Modipuram. Of all the cultivars, KAK 2, ICCV 2 and ICCV 10 performed well with low disease severities and high grain yield on broad beds than on flat.

APEDA, Modipuram: Of the five cultivars, HC 1, ICCV 10, Pusa 1053, Pusa 1088 (K), Pusa 1103, cultivar ICCV 10 planted under zero tillage after rice. Vegetative growth of the cultivar PUSA 1088 planted on beds after land preparation was luxurious with abundant podding. PUSA 1088 and ICCV 10 had higher grain yield than other cultivars.

Monitoring and evaluation of on-farm participatory groundnut IDM trials in Andhra Pradesh: Groundnut is the major crop in Anantapur district (India) and is grown in >700,000 ha every year. Late leaf spot and rust are destructive diseases in groundnut and cause severe losses in quality and quantity of haulms and pods. Groundnut haulms are the major source of fodder for cattle in this district. To manage these two diseases, integrated disease management (IDM) technology, developed at ICRISAT-Patancheru was evaluated, promoted and scaled up in the state of Andhra Pradesh, India, by involving large number of farmers in a participatory approach. The technology consisted of combination of improved dual-purpose groundnut cultivar with moderate levels of host plant-resistance, fungicide seed treatment and judicious use of fungicide, chlorothalonil (Kavach). One fungicide spray at 60 days after sowing (DAS) for early-maturing cultivars ICGVs 91114, 89104; and two sprays at 60 and 75 DAS for medium-maturing cultivars, ICGVs 92020, 92093 were scheduled for all IDM trials. One hundred and twenty one farmers' participatory IDM trials in five villages (46 farmers in Lingareddypalli, 43 farmers in Jalalapuram, 22 in Talupuru, 5 in Anataraganga and 5 in Jonnalakothapalli) in the Anantapur district (India) were conducted in collaboration with District Agricultural Advisory and Transfer of Technology Center (DAATTC) during 2004 rainy season. In addition to these trials, several farmers in 20 villages in the states of Andhra Pradesh, Karnataka and Tamil Nadu adopted the technology during 2004 rainy season. During this year, the weather was congenial for groundnut production in Anantapur. Severity of foliar diseases was comparatively low in IDM plots in all the cultivars than non-IDM plots of local cultivar in all the villages. Mean LLS and rust severities had up to 5 rating in IDM plots compared to around 7 rating in non-IDM plots across the villages. Haulm yields were 2. 46 t ha^{-1} in IDM and 1.94 t ha^{-1} in non-IDM plots, while pod yields were 2.08 t ha^{-1} in IDM and 1.41 kg ha^{-1} in non-IDM plots across the villages.

Farmers training program on IDM of foliar diseases in groundnut, Anantapur, Andhra Pradesh, India: A training program on the management of foliar diseases of groundnut was conducted at Anantapur (India) from

28 to 29 June 2004 at the DAATT Center. About 94 farmers (50 farmers from Lingareddypalli, 30 from Talupuru, seven each from Antaraganga and Jonnalakothapalli) from four partner villages attended this program. In the fifth village Jalalapuram, as there was good rain and all the farmers were busy in groundnut sowings, and the program was conducted in the evening in the village itself. About 96 farmers both participating and non-participating attended the program in this village. The program was conducted in local language (Telugu). Its main theme was "healthy groundnut, more pods, and more milk" through integrated disease management (IDM). The program included discussions on foliar and soil-borne diseases, and aflatoxin contamination and their management. Additionally, improved package and practices of groundnut cultivation and principles of farmers' participatory research and its impact were also discussed. IDM promotional material on groundnut diseases and aflatoxin management in Telugu was distributed to the farmers.

Farmers training program on IPM/ICM in chickpea in the villages Rajahar (Nawalparasi), Bardibas (Mohattari) and Lalbandi (Sarlahi), Nepal

Rajahar (Nawalparasi): A training course on production technologies for chickpea was conducted in the village Rajahar, district Nawalparasi on 1 October 2004 in collaboration with NGLRP and FORWARD (NGO). Twenty-seven participating farmers and several non-participating farmers attended this program. Topics covered in this program were, agronomic management, major disease and insect pests and their management, seed production and post harvest technologies for chickpea. Additionally, a field day was conducted by NGLRP in the village Rajahar, district Nawalparasi on 24 March 2005. About 26 farmers from the village Rajahar and 10 scientists from NGLRP, FORWARD and Agriculture Service Center participated the field day. Field trials on integrated pest management (IPM), participatory varietal selection (PVS), and seed multiplication were visited by the group. There was good interaction between farmers, researchers and extension personnel.

Bardibas (Mohattari) and Lalbandi (Sarlahi): Hands-on training on improved packages for chickpea production with special emphasis on IPM was conducted by NORP, Nawalpur in the villages Bardibas (Mohattari) on 18 January 2005 and Lalbandi (Sarlahi) on 19 January 2005. About 25 farmers participated in each village. In addition to the general discussion on the production practices in chickpea, farmers were specially educated about the diseases, insect pests and their management, seed production and storage. Farmers showed keen interest in learning IDM practices and adopting them. Farmers' field days were conducted by NORP in Bardibas on 19 March 2005 and in Lalbandi on 20 March 2005. About 25 farmers in the village Bardibas and 29 farmers in the village Lalbandi attended the field day. All the farmers in both the villages were impressed with the performance of IPM technology and the cultivar Avarodhi.

S Pande

Activity 1.5.2: Monitor and document the impact of improved technologies

Milestone: Utilization of ICRISAT-bred parental lines of sorghum and pearl millet by NARS and private sector and adoption of their hybrids assessed in India (2006)

Seed Producers' Sorghum Hybrid Trial (SPSHT) Report: The public and private sector scientists utilize ICRISAT-bred seed parents for developing commercial hybrids. To assess the performance of sorghum hybrids produced by different private sector organizations, "Seed producers' sorghum hybrid trials" are being coordinated in postrainy and rainy seasons. Under this activity, SPSHT 2004-05 Postrainy trial (9 entries, 4 locations) and SPSHT 2005 Rainy trial (9 entries-Tulasi 207, Kaveri 901, Kaveri 2244, MLSH 57, BSH 10, BSH 15, CSH 18, CSH 20 and local), at 5 locations (Ankur Seeds, Bagpur; Ankur Seeds, Dharwad; Basant Agrotech, Akola; Emergent Genetics, Jalna and ICRISAT- Patancheru) were conducted.

The results of postrainy season SPSHT-2004-05 showed that the hybrid performance differed with locations and no single hybrid performed better in all the locations for any of the traits such as grain yield, grain size, and days to 50% flowering. Two hybrids SR 344 and MLSH 117 at Dharwad; all the hybrids except MLSH 117 at Patancheru; and two hybrids, Kuber and SR 351 at Bijapur were significantly superior to the controls used in the respective locations. Three hybrids, EGSH 64, 746611 and SR 344 were on par with local control at Jalna. The hybrid Kuber along with superior grain yield performance also had large grains than the rest of the hybrids at three of the four locations.

The results of rainy season SPSHT-2005 showed that no single hybrid excelled in all the locations for grain yield, but they were on par with the control CSH 20 at all the locations. However, the hybrids MLSH 57, BSH 10 and BSH 15 outperformed the control CSH 18 significantly in three of the five locations. However, all the hybrids were on par with the control CSH 20 at all the locations except Kaveri 901 in two locations (Nagpur and

Akola) where it showed poor performance. MLSH 57 and BSH 15 were highest yielders at two locations each. The hybrids MLSH 57, Kaveri 901 and Kaveri 2244 had significantly larger grains than both the controls CSH 18 and CSH 20 at two or more locations. The summary reports on the results of postrainy season SPSHT-2004-05 and rainy season SPSHT-2005 were distributed to consortium partners.

BVS Reddy and S Ramesh

Impact assessment of breeding materials: A questionnaire was developed to assess the impact of ICRISAT-developed sorghum hybrid parents and breeding materials and strategic research information in a systematic manner. A Special Project Scientist was appointed at ICRISAT for the purpose.

BVS Reddy and S Ramesh

Impact assessment: In order to assess the utilization ICRISAT-bred parental lines of pearl millet by the private sector and adoption of their hybrids in India, impact assessment was initiated. Questionnaires were prepared in consultation with the Hybrid Seed Parents Research Consortium Advisory Committee and ICRISAT's socio-economists, and circulated to private sector members for their response during the consultation meeting. An Impact Assessment Economist was appointed to conduct this study.

CLL Gowda, KN Rai, BVS Reddy, KB Saxena, VN Kulkarni and S Ramesh

Milestone: Farmers' acceptance of ICM technologies including improved varieties in chickpea, pigeonpea and groundnut documented (2005)

Impact of ICM technologies including improved varieties in legumes documented: During the year, project locations in China, India, Nepal and Vietnam were visited to monitor on-farm farmer participatory trials. At each location, farmer-scientist interaction meetings were organized to assess farmers' response to improved technologies. Farmers showed tremendous response to the improved technology at all the locations visited.

SN Nigam and R Aruna

Milestone: Impact assessment of chickpea cultivars in Ethiopia completed (2005)

Adoption studies of improved chickpea varieties in Ethiopia: A report of the study conducted in Ethiopia to assess the adoption of improved chickpea varieties and the constraints to adoption of improved varieties was published in 2005. Chickpea is one of the most important pulse crops in Ethiopia, the largest chickpea growing country in Africa with a share of over 40% in the chickpea production. Bulk of the Ethiopian chickpea production is used for human consumption. Thus, chickpea is an important source of dietary protein, fiber and minerals for many Ethiopians, particularly the rural poor. Over 300 households were surveyed in four chickpea growing districts of central Ethiopia representing major chickpea production areas of the country. The adoption of improved varieties varied considerably (6 to 66%) from one district to another, mainly because of variations in the awareness of improved varieties among farmers. The variety *Mariye* had the highest adoption followed by *Shasho, Dubie*, *Arerti* and *Worku*. Important characteristics of improved varieties that were liked by farmers included drought tolerance, high yield and early maturity. The non-availability of the seed of improved varieties has also contributed to the low adoption rate of improved varieties. Extension activities have to be enhanced and made more effective to increase the farmers' awareness about the improved varieties, and an efficient seed production and distribution system needs to be established in the country. Of course, efforts are needed to develop improved varieties resistant to abiotic and biotic stresses, with high yield potential and seed traits preferred by the market.

PM Gaur, CLL Gowda, MCS Bantilan and HA Freeman

Activity 1.5.3: Exchange improved techniques and new knowledge with ARIs, NARS, NGOs, private sector, and farmer groups

ICAR-ICRISAT groundnut scientists' meet: ICAR-ICRISAT Groundnut Scientists meet was organized at ICRISAT during 2–3 October 2005. Forty-four scientists from different ICAR institutions and state agricultural universities participated in the meet. ICRISAT scientists working on different aspects of groundnut crop improvement presented the highlights in their area of work. Participants visited groundnut research fields and made selections in the breeding material for different traits of interest. The participants selected 527 advanced breeding lines and 237 segregating populations. Material has been dispatched to the respective NARS partners.

Forty-one sets of international trials (4 foliar diseases resistant, 8 drought-tolerant, 7 short-duration, 7 medium-duration, 3 aflatoxin tolerant and 12 confectionery) were supplied to cooperators in Afghanistan, Bangladesh, China, India, Indonesia, Philippines, South Africa and Sudan. We supplied 164 advanced breeding populations

to cooperators in eight countries (Afghanistan, Fiji Islands, India, Iran, Philippines, South Africa, East Timor and Uzbekistan).

SN Nigam and R Aruna

Milestone: Seeds of parental lines, breeding populations, and advanced breeding lines and varieties multiplied and distributed to researchers, collaborators and farmers on request (Annual)

Sorghum scientists' field days: Field visits were arranged for public and private sector scientists. Sorghum Scientists' field day was organized at ICRISAT-Patancheru on 10 January 2005 for public sector scientists and on 24 February 2005 for public and private sector scientists for selection of the breeding materials.

Seed supplies: A total of 5648 seed samples of hybrid parents/breeding lines were shared with scientists (public and private sector), NGOs and farmers based on specific requests. Of the 5648 seed samples, 2663 were shared with public sector scientists, 1313 with private sector scientists and 721 with NGOs and farmers in India. The rest of the seed samples (951) were shared with NARS (public and private sector scientists) outside India. The seed of selected breeding material are being supplied as per new guidelines of hybrid parents' research consortium to scientists in private sector.

BVS Reddy

Seed production and supply: We produced 325 kg breeder seed of ICTP 8203, and 158 kg supplied from the reserve seed stock. We also supplied 126 kg breeder seed of three hybrid parental lines. In addition, 70 kg breeder seed of two seed parental lines (ICMA/B 94555 and 842A/B) were multiplied. We provided 471 seed samples (432 in India and 39 overseas) of breeding lines and hybrid parents. In response to 2004 Scientists' Field Day requests, we supplied 2174 pearl millet seed samples to 14 public-sector organizations and 1628 seed samples to 22 private sector organizations. Three new seed companies joined the Hybrid Parents Research

Consortium in 2005, and we supplied 345 seed samples for lines selected during the 2005 summer season.

KN Rai and VN Kulkarni

Seed production and distribution: Breeder seed of three short-duration varieties ICPL 88039 (795 kg), ICPL 87 (25 kg) and ICPL 87091 (25 kg); and four medium-duration varieties ICPL 87119 (1055 kg), ICPL 85063 (675 kg), ICPL 8863 (460 kg), and ICP 7035 (250 kg) was multiplied in isolation. This seed will be used to meet the national and international seed requirements.

During the 2005, a total of 2870 kg seed was supplied to 40 NARS and private seed sector partners. A total of 885 pigeonpea seed samples were supplied to private seed companies and NARS in India and other countries. This includes 490 samples of hybrids, 212 CMS A/B-lines, and 183 fertility restorer lines.

Promotion of pigeonpea in the Philippines: Under a new initiative ICRISAT and the Philippines Government will collaborate in promoting pigeonpea in northern regions of the Philippines. The initial trials of short-duration lines ICPL 88039 and ICPL 88034 have shown promise in rice-fallow system. During the 2006, a number of demonstrations have been planned. Also, it is proposed to promote vegetable pigeonpea for local consumption and to try to rehabilitate degraded lands with perennial pigeonpeas. We have supplied about 100 kg seed of six varieties to the Philippines to undertake the testing program in different agro-ecological zones.

KB Saxena

Chickpea scientists' meet organized at ICRISAT-Patancheru: A one-day Chickpea Scientists' Meet was organized at ICRISAT on 6 January 2005 for the scientists of National Agricultural Research System (NARS) of India. The meeting was attended by 45 scientists that included 28 Indian NARS scientists from 12 states and 17 ICRISAT scientists. The participants visited various experiments on physiology, pathology, entomology, genetic resources, wide hybridization, genetics and breeding of chickpea and the visit facilitated interaction between ICRISAT and NARS scientists. The NARS scientists selected germplasm and breeding materials of their interests. ICRISAT supplied 1853 chickpea seed samples indented by NARS after the crop harvest.

PM Gaur and CLL Gowda

Multiplication and distribution of seeds of chickpea advanced breeding lines and cultivars: Two International Chickpea Screening Nurseries (ICSN-*Desi* and ICSN-*Kabuli*) were evaluated by NARS scientists during 2004/05. A total of 75 sets (37 of ICSN-*Desi* and 38 of ICSN-*Kabuli*) were supplied to 12 countries – Australia (2), Bangladesh (2), Canada (8), China (4), Ethiopia (6), India (41), Iran (2), Israel (1), Mexico (1), Myanmar (2), Nepal (4) and South Africa (2). The results were received from 20 locations for ICSN-*Desi* and from 19

locations for ICSN-*Kabuli*. The results received from Indian locations were compiled in a report and distributed to Indian NARS during annual group meet of All India Coordinated Research Project on Chickpea held at Kolkata during September 2005. A total of 59 sets of ICSN (30 of ICSN-*Desi* and 29 of ICSN-*Kabuli*) were distributed to NARS during 2005 for 2005/06 crop season.

About 29.0 t breeder seed of ICRISAT-related chickpea varieties ICCV 2 (7.58 t), ICCV 10 (5.76 t), ICCC 37 (18.5 t), KAK 2 (1.92 t), JG 11 (0.56 t), and JGK 1 (0.25 t) was produced. The seed was first distributed to various agencies as per the allotment from Government of India, and the remaining seed was distributed to farmers and NGOs.

PM Gaur

Milestone: ICRISAT partnerships with NARS, networks and regional fora strengthened (Annual)

Collaborative evaluation of nursery for aphid resistance: Aphid infestation was severe during the 2001 rainy season and postrainy seasons in India, especially several parts of Maharashtra. The infestation was severe in the 2001 rainy and postrainy seasons and resistant seed parents were identified by scoring the infestation damage on 1 to 5 scale, where 1 = free from aphids and 5 = >60% leaf area damaged. Based on the subsequent screenings in 2002, 2003 and 2004, an aphid nursery consisting of 62 aphid resistant lines along with two controls was constituted and was sent for evaluation in India.

Collaborative evaluation of selected lines for micronutrient density: A total of 40 sorghum lines with high and low grain Fe and Zn contents selected based on the 2004 postrainy season evaluation of 86 diverse sorghum lines were sent to National Research Centre for Sorghum (NRCS), Hyderabad, India during the 2004–05 postrainy season for yield performance and grain Fe and Zn contents at NRCS, Hyderabad. The results revealed significant genetic variability for grain Fe and Zn contents. Some of the landraces such as IS 7780 (38.5 ppm), IS 1192 (32.8 ppm), IS 24868 (32.8 ppm) and high-yielding B-line such as ICSB 561 (32.6 ppm) and ICSB 484 (29 ppm) had significantly higher Zn content compared to the trial mean value (25.4ppm). Similarly, lines such as ICSR 40 (56.7 ppm), PVK 801 (50.2 ppm), ICSB 561 (49.6 ppm), IS 152 (47.6ppm), ICSB 675 (46.2 ppm), ICSB 52 (45.6 ppm) and ICSB 38 (44.4 ppm) had significantly higher Fe content compared to the trial mean value (37.6 ppm).

BVS Reddy

Pearl millet trials and nurseries for NARS: Under the ICAR-ICRISAT partnership project, 5 trials related to hybrid parents research were sent to 10 locations and two nurseries were sent to 8 locations. Of these, 2 B-line trials and 3 R-line trials were conducted at Patancheru. Dialog was initiated with the All India Coordinated Pearl Millet Improvement Project Coordinator to strengthen the research partnership in areas such as forage hybrids, hybrids for arid Rajasthan, salinity tolerance, biofortification, seed production in Rajasthan, alternative use of pearl millet grain for alcohol production, and training and publications.

Pearl Millet Consortium Hybrid Trial: The trial consisting of 17 test hybrids from 11 private seed companies, along with three controls: ICMH 356 (ICRISAT), PB 106 (Proagro) and 7688 (Pioneer) was conducted at three diverse locations with varying productivity levels showed that overall none of the hybrids outperformed the best control hybrid 7688 either for grain yield or dry fodder yield. At Aurangabad, two hybrids had significantly higher grain yield, with BBH-111 having 20% more grain yield and Kaveri-456 having 13% more grain yield than highest-yielding control 7688 (4505 kg ha^{-1}). The same two high grain yielding hybrids also had significantly higher dry fodder yield, with BBH-111 having 32% more fodder yield and Kaveri-456 having 9% more fodder yield than 7688 (7484 kg ha^{-1})**.** Although statistically not higher than 7688 at any of the locations, the average grain yield of GSMH-55 was identical to that of the highest-yielding hybrid Kaveri-456. It is recognized that the limited data set from just three locations is inadequate to make any definitive conclusions about the performance of these hybrids. It is suggested that the Consortium Hybrid Trial in the future be conducted at 8–10 locations.

KN Rai and VN Kulkarni

Milestone: Technical information and pubic awareness literature/documents developed and disseminated (Annual)

The 2005 issue of International *Arachis* Newsletter with 20 articles from 4 countries and news and views items from different parts of the world was published on time.

Researchers from different countries [China (2), Nepal (3), Iran (1), the Philippines (3), Uzbekistan (1), Vietnam (2), India (2)] were trained in different groundnut breeding and seed production technologies. Queries related to different aspects of groundnut from farmers and students were attended to on various occasions. Three posters and two success story flyers on various aspects of groundnut were prepared for different occasions and information sharing.

Farmer friendly literature on ICM in groundnut, pigeonpea and chickpea: A 2-page handout in Telugu and English entitled 'ICGV 91114 – The Alternative to Groundnut in Anantapur is Better Groundnut' was prepared and distributed to farmers in Anantapur, Andhra Pradesh. It also contained information on low-cost production technologies.

SN Nigam and R Aruna

ICRISAT Archival Report 2006

Projects 2, 3, 4, 5 and 6

from

Global Themes Biotechnology and Crop Improvement

Project 2
Sustaining biodiversity of Sorghum, Pearl Millet, Small Millets, Groundnut, Pigeonpea and Chickpea for current and future generations

Output A: Germplasm of staple crops assembled and conserved and an additional 5% of germplasm characterized annually and documented for utilization

MTP Output Targets 2006:

- *500 new sorghum accessions from USA*
- *Unrestricted access to and movement of staple crop germplasm ensured*
- *Germplasm accessions regenerated for longterm conservation and distribution (at Patancheru genebank)*
- *Safety copy of germplasm at Niamey genebank conserved and regenerated as appropriate (groundnut 2,000 accessions and pearl millet 5,000 accessions)*

Output target A.1: New germplasm of staple crops assembled for conservation and utilization (2009)

Activity A.1.1: Identify gaps and priority areas for germplasm of staple crops

Milestone A.1.1.1: Global databases of chickpea and pigeonpea compared to identify unique germplasm (HDU, 2007)

Germplasm databases of chickpea at ICRISAT and ICARDA were compared and 500 accessions including wild relatives were identified as unique for inclusion in the genebank. Similarly, we identified from the chickpea collection maintained at WSU, Pullman, USA 839 accessions as unique for ICRISAT. The chickpea germplasm collections maintained at National Centre for Plant Genetic Resources of Ukraine (NCPGRU), Ukraine were compared with our collection and identified 313 accessions as unique for inclusion in our collection.

HD Upadhyaya

Milestone A.1.1.2: Priorities areas identified for chickpea and pigeonpea for collection/assembly in collaboration with NARS (HDU/NARS scientists, 2008)

Milestone A.1.1.3: Sorghum germplasm from USDA (500 no's), Pearl millet from Niger (400 accessions) and pigeonpea collections from Tanzania, Uganda and Mozambique (200 accessions) assembled (HDU/CLLG, 2008)

National Bureau of Plant genetic Resources (NBPGR), India has released 483 germplasm samples of sorghum received from Niger for growing in Post Entry Quarantine Isolation area (PEQIA) for observation and subsequent release. We are awaiting import permits from the Government of India for acquiring pearl millet (424) collections from Niger and pigeonpea (231) collections from Tanzania, Uganda and Mozambique presently held at Niamey and Nairobi genebanks, respectively.

HD Upadhyaya and CLL Gowda

Milestone A.1.1.4: Global databases of groundnut and sorghum compared to identify unique germplasm (HDU, 2008)

Sorghum germplasm databases of ICRISAT and the USDA maintained at NSSL, Fort Collins, USA was compared and identified 2,708 accessions of Rockefeller collection that were missing from our collection. A set of 619 accessions from the newly identified set of 2708 accessions, was received from NSSL and added to the collection. We are making efforts for obtaining the rest of the identified material for filling the existing gaps in the collection.

The germplasm database of Chinese Academy of Agricultural Sciences (CAAS), China was compared with our database and identified 250 accessions of sorghum belonging to race Caudatum and subrace Kaoliang and 250 accessions of groundnut germplasm belonging to botanical variety hirsuta as unique for ICRISAT genebank.

HD Upadhyaya

Milestone A.1.1.5: Priorities areas identified for groundnut and sorghum for collection/assembly in collaboration with NARS (HDU/NARS scientists, 2009)

Output target A.2: Assembled germplasm of staple crops characterized for utilization (2009)

Activity A.2.1: Characterize new germplasm for important morpho-agronomic traits

Milestone A.2.1.1: Sorghum germplasm from Niger (450 accessions), chickpea germplasm from ICARDA (500 accessions) and groundnut germplasm from Japan assembled and characterized for morpho-agronomic traits (HDU/CLLG, 2008)

Recording data on 483 sorghum accessions originating from Niger is in progress in PEQIA. NBPGR, India released a total of 839 chickpea germplasm samples provided by the Washington State University, Pullman, USA that were identified as unique for ICRISAT genebank. From this set, we planted 747 cultivated types during the post-rainy season of 2006 for recording morpho-agronomic traits and seed increase. Seed samples of 62 (annual types) out of 70 wild chickpea seed samples from this set are planted in a glasshouse under extended light for seed increase. We received 622-groundnut germplasm samples for further inspection and release by NBPGR. These accessions were identified as unique from the national collections of groundnut maintained at National Institute of Agricultural sciences (NIAS), Japan.

HD Upadhyaya and CLL Gowda

Milestone A.2.1.2: Sorghum germplasm from USDA, pearl millet collections from Niger (400 accessions), and pigeonpea collections from Tanzania, Uganda and Mozambique (200 no's) characterized (HDU/CLLG, 2009)

We are awaiting import permits from the Government of India for acquiring pearl millet (424) collections from Niger and pigeonpea (231) collections from Tanzania, Uganda and Mozambique presently held at Niamey and Nairobi genebanks, respectively.

HD Upadhyaya and CLL Gowda

A total of 123 pigeonpea landraces collected from farmers' fields in four pigeonpea growing regions of Tanzania were characterized and evaluated for 16 qualitative and 14 quantitative descriptors; and their response across three pigeonpea growing environments in Tanzania and Kenya determined. Polymorphism in the qualitative traits was relatively low among accessions and across collection regions. Collections from the northern highlands exhibited lower diversity in qualitative descriptors, especially physical grain characters, relative to the other three regions, an indication of farmer selection in response to market preferences. There were significant differences in agronomic traits among accessions and in G x E interaction. Grain yield had positive significant correlations with pods per plant, pod yield, racemes per plant and both primary and secondary branches per plant, traits that were also correlated with plant height. High broad-sense heritability was recorded for days to flower, days to maturity, plant height, raceme number and 100 seed mass. Principal component analysis separated variability among landraces according to days to flower, days to maturity, plant height, number of primary and secondary branches, and number of racemes per plant. Similarly, cluster analysis separated the accessions into six groups based on the same traits. There was close clustering within and between materials from the coastal zone, eastern plains and southern plains with the northern accessions distinctly separated and with wide dispersion within them. Overall, two diversity clusters were evident with coastal, eastern and southern landraces in one diversity cluster and northern highlands landraces in another cluster. This diversity grouping established potential heterotic groups in this pigeonpea germplasm, which may be used in crosses to generate new cultivars adapted to different pigeonpea growing environments with consumer acceptability. The grouping may also form a basis of forming a core collection of this germplasm representing the variability available in Tanzania.

S Silim and E Manyasa

Milestone A.2.1.3: Morphoagronomical characterization of 420 Sorghum landraces and 84 wild types from Sorghum growing areas in Mali (except Gao region) with a special focus on adaptive traits (cycle, photoperiod sensitivity, tillering, stay green) (FS, PST, NARS, 2006)

420 cultivated and 84 wild entries were planted at 3 sowing dates (22/06 and 22/07 at ICRISAT Samanko Station, 2006/07 in IER Sotuba Station) to measure the phenological characters with 8 individuals to represent one entry per replication and 2 replications per sowing date. The seedling emergence was good for cultivated sorghums and more heterogeneous for wild/weedy sorghums due to seed dormancy. Twelve thousand plants

were measured for 15 quantitative characters whereas qualitative morphological traits were measured at the entry level on one plant in each replication. The racial identification showed that guinea gambicum (53%), guinea margaritiferum (16%) and the sweet sorghums belonging to the bicolor race (12%) are the dominant sorghum groups in Mali. The highest variation was observed for cycle duration (sowing-flag leaf emergence duration varying from 48 days to 122 days). Most of landraces are partially or completely photoperiod sensitive. Stay green is mainly explained by the date of flowering rather than other environmental factors with late maturing varieties keeping green leaves longer. The information provided by SSR and DArT markers on the same material, combined with the observed quantitative trait variability within the 3 dominant sorghum groups in Mali, should allow for promising genetic association studies.

F Sagnard and PS Traoré (in collaboration with IER)

Milestone A.2.1.4: Morphoagronomical patterns of Sorghum diversity in Mali to understand large adaptive trends and identify new interesting local germplasm for breeding programs published (FS, PST + NARS, 2008)

Output target A.3: Germplasm sets of staple crops evaluated for useful traits (2009)

Activity A.3.1: Evaluate germplasm sets of staple crops for agronomic characters and special traits for utilization

Milestone: A.3.1.1: Sets of germplasm in staple crops evaluated to identify sources for yield and other quality traits (HDU/CLLG/Scientists - Crop Improvement, Annual)

Chickpea:
Drought tolerant lines: Twenty accessions selected during 2004-2005 season for root depth, a trait related to drought in chickpea were evaluated with four control cultivars (Annigeri, ICC 4958, ICCV 2, ICC 12237) for yield potential and other agronomic traits in a replicated trial. Among the deep-rooted accessions ICC 1356 (3555 kg ha^{-1}) produced 36.9 % greater seed yield than the drought tolerant control ICC 4958 (2596 kg ha^{-1}) and 27.4% greater seed yield than the highest yielding control cultivar Annigeri (2790 kg ha^{-1}). Similarly, ICC 1431 (2967 kg ha^{-1}) and ICC 95 (3124 kg ha^{-1}) produced 14.3% and 20.3% higher seed yield than ICC 4958 and 6.3% and 12.0% higher seed yield than Annigeri, respectively.

In another experiment with 20 accessions selected during 2004-2005 season for root length density, another trait related to drought in chickpea were evaluated with four control cultivars (Annigeri, ICC 4958, ICCV 2, ICC 12237) for yield potential and other agronomic traits in a replicated trial. Among the accessions with largest root length density, ICC 13816 (3120 kg ha^{-1}) produced seed yield similar to the drought tolerant control ICC 4958 (3095 kg ha^{-1}) and the highest yielding control cultivar Annigeri (3230 kg ha^{-1}).

Large-seeded Kabuli Types: Evaluated 34 large-seeded kabuli chickpea accessions originating from diverse geographical regions with four control cultivars (ICCV 2, KAK 2, JGK 1, L 550) in a replicated trial. ICC 14214 (2063 kg ha^{-1}; 53 g 100-seed weight) produced 7.1% greater seed yield and 35.1% higher seed weight than the large seeded and high yielding control cultivar KAK 2 (1927 kg ha^{-1}; 39 g 100-seed weight). Similarly, ICC 6243 (2359 kg ha^{-1}; 38 g 100-seed weight) and ICC 16803 (2072 kg ha^{-1}; 40 g 100-seed weight) produced 7.5 to 22.4% higher seed yields with similar seed weight to KAK 2. ICC 6210 (1925 kg ha^{-1}; 48 g 100-seed weight), ICC 7347 (1943 kg ha^{-1}; 51 g 100-seed weight), and ICC 14203 (1921 kg ha^{-1}; 56 g 100-seed weight) produced similar seed yield with 23.1-45.6% greater seed weight than KAK 2.

Evaluated 16 large-seeded kabuli chickpea accessions, selected from newly assembled accessions, with four control cultivars (ICCV 2, KAK 2, JGK 1, L 550), in another experiment. ICC 17457 (2468.2 kg ha^{-1}; 54.5g 100-seed weight) and ICC 17458 (2216 kg ha-1; 47g 100-seed weight) produced 28.0% and 14.9% greater seed yield with 47.3% and 28.1% greater 100-seed weight than the large seeded and high yielding control cultivar KAK 2 (1929 kg ha^{-1}; 37 g 100-seed weight).

Early Maturing Lines: Seventeen early-maturing germplasm accessions and three control cultivars (ICCV 2, Annigeri, ICCV 96029) were evaluated in a replicated yield trial for seed yield related agronomic traits. ICC 16347 (91 days; 1751 kg ha^{-1}) matured earlier and produced similar seed yield as the early-maturing control ICCV 2 (97 days; 1772 kg ha^{-1}). ICCs 5829, 11916, 13925, and 14368 (1962 – 2434 kg $ha^{-1;}$ 100 days) produced 10.7% to 37.4% greater seed yield and matured similar to ICCV 2.

Extra-Early Kabuli Types: Evaluated 58 elite extra-early maturing kabuli germplasm lines and four control cultivars (KAK 2, L 550, ICCV 2, JGK 1) under normal and late sown environments. Under normal sown

environment two entries (2130 – 2327 kg ha^{-1}) produced higher seed yield, flowered earlier (30-33 days), matured in similar days (96-97 days) with similar seed size (38–39 g) to high-yielding large-seeded control cultivar KAK 2 (2016 kg ha^{-1}; 36 days flowering; 97 days to maturity; 37 g 100-seed weight). One of these two also produced (1212 kg ha^{-1}) higher seed yield and took similar days to flowering and maturity with similar seed size to control KAK 2 (904 kg ha^{-1}) under late planted conditions.

Salinity Tolerant: Evaluated 52 salinity tolerant chickpea germplasm accessions with ICCV 2 and salinity tolerant control cultivar Jumbo 2. ICCs 4953, 5003, 10552, 10575, 12339, 13124, and 14595 and ICCV 95311 (3023–3678 kg ha^{-1}) produced significantly greater seed yield than ICCV 2 (2147 kg ha^{-1}) and Jumbo 2 (2089 kg ha^{-1}).

Evaluation of Newly Assembled Germplasm: Evaluating 747 newly assembled chickpea germplasm accessions from USA and five control cultivars in an Augmented design trial during 2006-2007. Data recording is in progress.
500 accessions from ICRISAT genebank and 418 accessions from ICARDA genebank are being evaluated with five control cultivars in an Augmented design trial during 2006-2007. Data recording is in progress.

Pigeonpea:
Evaluated 1000 accessions with four control cultivars (ICP 6971, ICP 7221, ICP 8863, ICP 11543) in an augmented design trial. The preliminary analysis indicate that ICPs 15391, 15014, 15012, 15021, and 16335 (50-54 days) were early flowering, ICPs 14459, 10915, 10904, 10906, and 10914 (41-49 cm) were short statured and ideal for mechanical harvesting, ICPs 7952, 9450, 9558, 2372, and 14225 (183–235) were with higher number of racemes, ICPs 9450, 4167, 11970, 14225, and 7952 (423–471) had higher number of pods plant per plant, ICPs 7035, 12825, 12746, 7407, and 13799 (19 – 23 g 100-seed weight) had large-sized seeds. ICPs 7952, 11737, 1, 13203, and 13483 (240–270 g yield plant^{-1}) were high yielding accessions. Harvest index was higher in ICPs 8835, 1209, 2624, 3602, and 11605 (50%- 65%).

Groundnut:
Drought Tolerant: Eighteen germplasm accessions with high SPAD and low SLA, the traits related to drought tolerance (Upadhyaya, 2005) were evaluated for pod yield and other traits related to yield in a replicated trial. Eight accessions (3.49–4.43 t ha $^{-1}$) produced higher pod yields than the control cultivar ICGS 76 (3.46 t ha $^{-1}$). ICGs 6766 and 14475 (3.49–3.83 t ha $^{-1}$) produced higher seed yield with greater seed size (71-83g) and similar shelling percentage (67-70%) to the control ICGS 76. (66g 100-seed weight; 70% shelling).

Evaluated 960 accessions in augmented design with four control cultivars (Gangapuri, M 13, ICGS 44, ICGS 76). Among the sub sp. *hypogaea* 274 entries (2.80–4.19 t ha $^{-1}$) produce higher pod yields than the control ICGS 76 (2.69 t ha $^{-1}$) and 249 entries (2.80–4.19 t ha $^{-1}$) produced higher pod yield than control cultivar M 13 (2.78 t ha $^{-1}$). ICGs 8795 (4.19 t ha $^{-1}$) and 9116 (3.84 t ha $^{-1}$) were significantly better than both the controls. Among the sub sp. *fastigiata* 32 entries (3.23–4.0 t ha $^{-1}$) produced significantly greater pod yields than the high yielding control ICGS 44 (2.19 t ha $^{-1}$). ICGs 1561, 2272, 11285 and ICGVs 04075 and 01276 (3.50-4.0 t ha $^{-1}$) were the top five entries.

Pearl Millet:
1000 accessions are being evaluated with three control cultivars, IP 3616, IP 17862, IP 22281, in augmented design trial during 2006. Data recording is in progress.

HD Upadhyaya and CLL Gowda

Characterized 360 early to medium maturity pearl millet landraces for agro-morphological traits and grain yield in rainy season 2006 at six locations in Senegal, Mali, Burkina Faso, Niger and Nigeria, and data made available to NARS partners. 64 late-maturing pearl millet landraces characterized for agro-morphological traits and grain yield in rainy season 2006 at three locations in Senegal, Mali, and Niger, and data made available to NARS partners. Preliminary characterization of 280 pearl millet landraces published in the International Sorghum and Millet Newsletter (47:110-112). The global pearl millet core collection was grown in a replicated trial at the ICRISAT Sahelian Center near Niamey, Niger, in the Rainy Season 2006. Ms Jenny Coral Padilla, MSc student from University of Hohenheim, Stuttgart, Germany, was involved in the characterization. Data analysis and MSc thesis write-up are underway.

BIG Haussmann

Milestone: A.3.1.2: New/assembled germplasm characterized/evaluated for important traits to fill gaps in characterization data (HDU, Annual)

In chickpea, we characterized 500 accessions for plant pigmentation and days to 50% flowering. In groundnut, over 2000 accessions were characterized for updating 30 traits in the databases. In sorghum, 385 accessions for which data was missing for days to 50% flowering and plant height were characterized during 2006 rainy season. We characterized during 2005-2006 post-rainy season, 235 sorghum germplasm accessions representing zerazeras and composite collections, for which data on important morpho-agronomic characters was incomplete. A total of 512 pearl millet accessions planted during 2005 post-rainy were characterized for four traits (days to 50% flowering, plant height, panicle length, panicle thickness). In pigeonpea, characterized 640 accessions during 2005 rainy season for which characterization data was incomplete and a total of 866 accessions were planted for characterization during 2006 rainy season for collecting data on missing traits.

HD Upadhyaya

Milestone: A.3.1.3: New germplasm sources identified for target insect pests and diseases in different crops (HCS/RPT/SP/HDU/CLLG, Annual)

Identification of new germplasm sources for resistance to diseases: Two hundred and fifty new germplasm accessions were evaluated for resistance to ascochyta blight (AB), botrytis gray mold (BGM), dry root rot (DRR) and collar rot (CR) diseases under controlled environment conditions and for fusarium wilt (FW) under artificial epiphytotic conditions at ICRISAT-Patancheru. Standardized individual screening techniques were employed to evaluate these accessions for individual diseases. Severities of AB, BGM and DRR were scored on 1-9 rating scale and the incidence of FW and CR was presented as percentage of mortality.

Resistance to AB: High level of resistance to AB were not identified in any the accessions evaluated, however, eight accessions, ICCs 643, 1052, 1069, 1093, 1903, 1915, 2114 and 2142 were found moderately resistant (3.1 to 5 rating) to AB.

Resistance to BGM: Among these new accessions, 30 had moderately resistant (3.1 to 5 rating) reaction and rest were found susceptible.

Resistance to DRR: Of the 250 new accessions evaluated for DRR resistance, two accessions ICCs 562 and 1600 had resistant reaction (3 rating) and 76 accessions were found moderately resistant (3.1 to 5 rating).

Resistance to CR: Resistance to CR was not observed in any of the accessions evaluated. Hence our quest continued to identify resistant sources in the germplasm and breeding material.

Multiple disease resistance in new germplasm: No line was found resistant to all the four diseases tested. Only one accession ICC 1915 was found to be moderately resistant to AB and DRR. Ten accessions ICCs 153, 1229, 1360, 1419, 1422, 1560, 1837, 2113, 2118 and 2202 had moderate resistance (3.1to 5 rating on 1-9 rating scale) to BGM and DRR.

70 elite germplasm accessions are being screened in wilt sick-plot during 2006-2007. Data recording is in progress.

S Pande, HD Upadhyaya and CLL Gowda

Milestone: A.3.1.4: Vegetable type pigeonpea germplasm evaluated for agronomic performance (HDU/CLLG, 2008)

Identified 33 vegetable type pigeonpea accessions for further verification and evaluation. We are also testing a few more accessions for higher seed number per pod.

HD Upadhyaya and CLL Gowda

Output target A.4: Germplasm accessions regenerated for conservation and distribution (2009)

Activity A.4.1: Regenerate critical accessions of staple crops germplasm

Milestone: A.4.1.1: Germplasm accessions of staple crops germplasm with low seed stock/viability regenerated (HDU, Annual)

In the 2006 postrainy season, 2268 accessions of sorghum were grown for regeneration. This includes, 128 accessions for diversity, 632 critical accessions for conservation as active collection and distribution and 1466 accessions for long-term conservation. For pearl millet, a total of 887 accessions were planted in the post rainy season for long-term conservation (806 accessions) and medium-term conservation (81 accessions). In pigeonpea, planted a total of 922 accessions during the rainy season for long-term conservation (599 accessions) and medium-term conservation (323 accessions). In addition, we also planted a set of 146 mini core accessions of pigeonpea for conservation and utilization. In groundnut, during the rainy season, we planted for regeneration of global mini core (184 accessions), Asia mini core (60 accessions), and critical accessions of groundnut (209 accessions) for medium-term conservation and utilization. During the post-rainy season, a total of 1671 accessions of groundnut were planted for long-term conservation (1308 accessions) and medium-term conservation (363 accessions). In chickpea 2139 accessions were regenerated. A total of 374 critical accessions representing chickpea (68), pigeonpea (12), groundnut (283 including 110 wild relatives), and small millets (12) were regenerated in the glasshouse and special facilities.

HD Upadhyaya

The global pearl millet core collection (504 accessions and four control cultivars) was grown in a replicated trial at the ICRISAT Sahelian Center near Niamey, Niger, in the rainy Season 2006. Ms Jenny Coral Padilla, MSc student from University of Hohenheim, Stuttgart, Germany, was involved in the characterization. Data analysis and MSc thesis write-up are underway.

BIG Haussmann

Milestone: A.4.1.2: Seed viability and health of new and regenerated germplasm tested and viability of conserved germplasm monitored (HDU/RPT-PQL, Annual)

During 2006, we tested the seed viability of 7032 accessions. This included 2416 (pearl millet-981; chickpea-635; pigeonpea-376; and groundnut-424) accessions processed as active collection and 1452 (pearl millet-787; chickpea-418; and pigeonpea-247) accessions processed as base collection. Germplasm seed samples with viability above 85% were processed as base collection. The mean seed viability of germplasm ranged between 86.6 for groundnut and 99.0% for chickpea.

For safety back up, we prepared a set of 1470 groundnut accessions harvested from a special regeneration (2005 rainy season). The seed viability in this material ranged between 92-100% with mean 99.9 %. For monitoring seed viability of conserved germplasm, we tested the seed viability of a total of 3146 (sorghum-2264 and chickpea- 882) accessions conserved as active collection for different periods. The viability ranged between 56 and 100% with a mean of 95.6% in chickpea and between 71 and 100% with a mean of 95.7% in sorghum. Similarly, 1396 accessions of groundnut base collection for more than 10 years in storage were monitored and the viability ranged between 46 and 100% with a mean of 94.0 %. This exercise has resulted in the identification of 47 accessions of chickpea and 11 accessions of sorghum from active collection and 69 accessions of groundnut from base collection as critical for regeneration during 2007.

HD Upadhyaya and RP Thakur

Seed health testing of germplasm accessions for the medium- and long- term storage in the genebank: A systematic seed health testing of germplasm is critical for their medium- and long-term conservation without affecting seed viability by seedborne pathogens. A total of 646 germplasm accessions (sorghum 546, chickpea 100) from the medium term storage of the genebank were evaluated for their seed health status using the standard blotter method. Only 19 off 646 accessions were free from seedborne pathogens (sorghum 10 and chickpea 9). We detected 21 fungi in sorghum and 11 in chickpea. Major fungi detected both in sorghum and chickpea were species of *Cladosporium*, *Alternaria*, and *Fusarium,* while species of *Phoma*, *Curvularia* and *Bipolaris* were specific to sorghum. These seedborne fungi affected seed viability up to 5% in sorghum and 4.2% in chickpea.

RP Thakur and HD Upadhyaya

Milestone: A.4.1.3: Germplasm samples processed to for medium- and long-term conservation (HDU, Annual)

A total of 2746 freshly harvested germplasm seed samples of different crops have been transferred to the cold rooms following standard protocols. This included, 1597 (sorghum 330, pearl millet 194, chickpea 217, pigeonpea 129, groundnut 727) accessions as active collection and 1452 (pearl millet 787, chickpea 418 and pigeonpea 247) accessions as base collection. With this addition the total number of accessions as base collection increased to 99,927 accessions representing 85.6% of total collection. Processing of sorghum and groundnut germplasm sets from the 2005-2006 postrainy season is in progress.

HD Upadhyaya

Activity A.4.2: Establish safety back up collections of staple crops

Milestone: A.4.2.1: Facilities identified for back up safety storage of germplasm collections in collaboration with partners and samples processed for safety backup (HDU/CLLG, Annual)

During this year, 1800 chickpea germplasm accessions were transferred as 'Black box' collection for safety back up at ICARDA genebank, Syria. Seed samples of 1677 accessions consisting groundnut (1478 accessions) and pigeonpea (199 accessions) were prepared following standard protocols.

HD Upadhyaya and CLL Gowda

Output target A.5: Germplasm databases updated for utilization (2009)

Activity A.5.1: Update databases of staple crops germplasm

Milestone A.5.1.1: Gaps in germplasm characterization data filled for chickpea, pigeonpea and groundnut (HDU/CLLG/Scientists - Crop Improvement, 2008)

Gaps in germplasm characterization data was updated for 1233 accessions for days to 50% flowering and plant pigmentation in chickpea, for 261 to 6540 accessions for 37 traits in pigeonpea, and for 20 to 3509 accessions for 16 descriptors in groundnut.
Updated characterization databases for 1793 newly assembled accessions in chickpea, 296 accessions in pigeonpea, and 50 accessions in groundnut for the respective crop species descriptors.

HD Upadhyaya and CLL Gowda

Milestone A.5.1.2: Passport and characterization databases of sorghum and pearl millet germplasm updated (HDU, 2009)

Updated gaps in germplasm characterization databases for 10 to 3674 accessions in 24 traits in pearl millets, and for 47 to 614 accessions in 20 traits in sorghum
Updated characterization databases for 120 newly assembled accessions in sorghum, and 320 accessions in pearl millet for all crop species descriptors.
Updating of passport data of sorghum and germplasm databases is in progress.

HD Upadhyaya and CLL Gowda

Milestone A.5.1.3: Germplasm databases of staple crops updated to SINGER format (HDU, 2009)

Databases are being checked and updated for completeness of data for SINGER format

HD Upadhyaya

Output target A.6: Germplasm of staple crops assembled and conserved for utilization at Regional Genebanks in Africa (2010)

Activity A.6.1: Identify sorghum collection gaps, collect and conserve new germplasm from identified priority areas in the eastern and southern Africa (ESA)

Milestone A.6.1.1: Gaps in sorghum germplasm collection identified, germplasm collected from at least 3 ESA countries and conserved (MAM/SGM, 2008)

Milestone A.6.1.2: Wild and cultivated Sorghums collected in at least 40 locations across Kenya assembled. Collection report completed and delivered to NARS partners in Kenya (SdV, FS + NARS + University of Free State + University of Hohenheim, 2007)

Activity A.6.2: Safely conserve assembled germplasm for utilization

Milestone: A.6.2.1: Germination tested for groundnut, sorghum and millet germplasm accessions at Sadore, Niger (BH, Annual)

Tested 1786 accessions of groundnut, and 168 accessions of sorghum during 2006 (pearl millet germination tests had been completed in 2003-04, no critical accessions found).

BIG Haussmann

Milestone: A.6.2.2: Critical accessions of groundnut, sorghum and millet regenerated in glasshouses at Sadore, Niger (BH, Annual)

No critical accessions found.

BIG Haussmann

Milestone: A.6.2.3: Germplasm accessions of groundnut, sorghum and pearl millet regenerated in the field at Sadore, Niger (BH, Annual)

A total of 1500 groundnut accessions multiplied in 2005 and processed in early 2006 for medium and long-term storage; sample size enhanced from 40 to 200 seeds per accession In sorghum 200 accessions were multiplied in 2005 and processed in early 2006 for medium and long-term storage. No further regeneration activities in 2006.

BIG Haussmann

Activity A.6.3: Conserve safety copy of germplasm held at ICRISAT, Patancheru at Sadore, Niger

Milestone A.6.3.1: Safety copy of germplasm conserved at Niamey genebank (groundnut, finger millet and pearl millet) (BH, Annual)

A total of 11,971 accessions have been conserved as safety duplicate at the Niamey genebank in 2006 (2006 groundnut, 5205 pearl millet and 4580 finger millet accessions).

BIG Haussmann

Activity A.6.4: Upgrade an ESA regional short-term seed storage facility in Kiboko

Milestone A.6.4.1: Short-term seed storage facility renovated and modules purchased and installed (MAM/SGM, 2007)

A storage room for germplasm has been completed at Kiboko for processing germplasm for medium term storage. A seed drying room has also been completed and a dryer installed for drying seed for medium term storage. The medium term storage facility in Nairobi has been equipped with plastic trays (crates) for holding bottles containing germplasm accessions. This will facilitate easy arrangement and tracing of germplasm in the facility. Purchased a cooling system and installed at the Kiboko short-term storage facility and the following materials are currently stored. This was done with funds contributed by two special projects – The ABS and the SCOSA projects.

Regeneration of critical accessions for medium and long-term storage continues and new and additional germplasm identified for regeneration include the following:

- 529 African finger millet were conserved after first year of characterization
- 1081sorghum, 144 pigeonpea and 36 chickpea accessions rejuvenated in 2005 have been processed and stored
- Established 100 pigeonpea accessions as working collection
- 500 accessions of sorghum core collection evaluated
- 1060 sorghum accessions including those received form Zimbabwe and also photoperiod sensitive materials from Tanzania were rejuvenated and processed
- 382 pearl millet accessions were rejuvenated

MA Mgonja and SG Mwangi

Activity A.6.5: Develop strategy for better documentation of germplasm accessions at Sadore genebank

Milestone A.6.5.1: Strategic plan for documentation of germplasm developed and implemented (BH, 2008)

Output target A.7: Unrestricted access and movement for staple crops germplasm ensured (2009)

Activity A.7.1: Assure risk-free export and import of germplasm materials

Milestone A.7.1.1: Requested germplasm of staple crops distributed to bona fide users for utilization (RPT/HDU/NBPGR/BH/MGM, Annual)

During this year, we distributed a total of 8516 samples of staple crops germplasm (sorghum-1806; pearl millet-1261; chickpea-2847; pigeonpea-1308; and groundnut-1294) for utilization to scientists in 23 countries in 111 consignments following standard protocols. Some of the special requests include sets of germplasm for collaborative evaluation with NARS in India and other countries (Table 1). The details are as follows:

Table 1. Distribution of germplasm sets for utilization during the year 2006.

Crop	Nature of set	No. Locations	Countries
Chickpea	Mini core	4	India (3) and Mexico (1)
Groundnut	Mini core	6	India (4), China (1) and Malawi (1)
Pearl millet	Core	6	India (5) and Niamey (1)
Pigeonpea	Mini core	7	India (6) and UAE (1)

Additionally, we provided 14206 samples of germplasm for internal utilization. The total includes, sorghum-3520; pearl millet-1647; chickpea-2915; pigeonpea-1986; and groundnut-4138.

RP Thakur, HD Upadhyaya and NBPGR

A total of 429 samples of 203 sorghum varieties were provided to nine ESA countries namely Tanzania, Kenya, Uganda, Ethiopia, South Africa, Botswana, Zimbabwe, Mozambique and Malawi and also to Mali. For pearl millet a total of 192 samples of 48 varieties were provided to Kenya, South Africa and Sudan.

MA Mgonja, E Manyasa and E Muange

Milestone A.7.1.2: Requested germplasm of staple crops exported for utilization and new germplasm imported for conservation after seed health evaluation and clearance through NBPGR (RPT/HDU/NBPGR, Annual)

Export of germplasm of mandate crops: We processed and successfully exported 9077 seed samples (sorghum 2329, pearl millet 1462, chickpea 4042, pigeon pea 393 and groundnut 851) comprising of breeding lines and germplasm accessions to 40 countries under 141 phytosanitory certificates. Two hundred forty-one seed samples (sorghum 31, pearl millet 60, chickpea 117, pigeonpea 30, groundnut 2 and minor millet 1) were rejected due to poor germination and/or association of seedborne fungi (Exserohilum turcicum, Bipolaris setariae, Colletotrichum graminicola, Fusarium oxysporum f.sp. ciceri, Botryodiplodia theobromae, Rhizoctonia bataticola and R. solani), non-established FAO designated status of the samples, or bacterium-contaminated seed. During 2006, 85.8% more seed samples were exported than in 2005**.**

Bulk export of seed material: A bulk consignment of 435kg seed of four sweet sorghum varieties (SPV 422, ICSV 700, ICSV 93046 and ICSR 93046) was exported to Philippines and 7.5kg seed of Ber (*Ziziphus rotundifolia*) to Niger. The phytosanitory clearance for these consignments was obtained from the Directorate of Plant Protection, Quarantine and Storage (DPPQS), Hyderabad.

Import of germplasm of staple crops: We imported 2674 germplasm samples of: sorghum (483) from Niger, chickpea (1549) from Australia, Syria and USA, groundnut (634) from Japan, USA and Vietnam and Maize (8) from Italy. In addition, 324 maize accessions and 10 sorghum accessions imported by SM Sehgal Foundation from Egypt were planted in PEQIA for inspection and release by NBPGR.

Import of non-mandate crops' grain and plant samples: Through special import permit obtained from Directorate of Plant Protection, Quarantine and Storage (DPPQS), Faridabad we imported 800 dried and powdered samples of: maize stover (500) from Germany, wheat straw (100), wheat grains (100), Bersime (*Trifolium alexandrium*) (50) and Lucerne (50) from Pakistan for ILRI-ICRISAT, and 140 soil samples from Pakistan for IWMI. All theses samples were required for various nutritional and chemical analyses.

Detection of *Peanut strip virus* (*PStV*) in groundnut: *PstV* was detected in several groundnut accessions grown in the greenhouse. The presence of *PstV* in some accessions was confirmed by ELISA and the infected seedlings were incinerated. Since the virus is aphid transmitted prophylactic sprays of Rogor® were given to avoid spread of the disease.

RP Thakur, HD Upadhyaya and NBPGR

A total of 28 samples of groundnut were distributed by the Niamey genebank in 2006.

BIG Haussmann

Crop researchers need to have access to diverse germplasm to develop improved broad genetic based cultivars; and to sustain biodiversity for current and future generations. A number of our NARS partners regularly request for materials in form of repatriated landraces germplasm, new breeding materials, finished and semi-finished products as well as nucleus seed of released varieties and hybrid parents for different uses. A total of 429 samples of 203 sorghum varieties were provided to 9 ESA countries namely Tanzania, Kenya, Uganda, Ethiopia, South Africa, Botswana, Zimbabwe, Mozambique and Malawi and also to Mali. For pearl millet a total of 192 samples of 48 varieties were provided to Kenya, South Africa and Sudan. The recipient organizations signed MTAs and these included NARIs, Universities, Seed Companies and NGO for research as well as for further multiplication. On the other hand we received from ICRISAT Patancheru a total of 441 sorghum and 11 pearl millet accessions from Zimbabwe

MA Mgonja, E Manyasa and E Muange

Output B: Germplasm of six small millets assembled and conserved, and an additional 10% of germplasm characterized/evaluated annually for desirable traits and documented for utilization

MTP Output Targets 2006

- *500 accessions of small millets regenerated at Patancheru*
- *Unrestricted access to and movement of small millet germplasm ensured*
- *(Annual activity)*
- *Safety copy of germplasm at Niamey genebank conserved and regenerated as appropriate (finger millet 4,580 accessions)*
- *Germplasm accessions regenerated for conservation and distribution (Annual activity)*

Output target B.1: New germplasm of small millets assembled for conservation and utilization (2009)

Activity B.1.1: Identify gaps and priority areas for germplasm of six small millets

Milestone B.1.1.1: Global databases of finger millet compared to identify missing unique germplasm, and priority areas identified for finger millet for collection/assembly in collaboration with NARS (CLLG/HDU/NARS scientists, 2007)

Milestone B.1.1.2: Global databases of foxtail millet, little millet, kodo millet, proso millet and barnyard millet compared to identify unique germplasm (CLLG/HDU/NARS scientists, 2008)

The foxtail millet germplasm database of Chinese Academy of Agricultural Sciences (CAAS), China was compared with ICRISAT database and identified foxtail millet – race: Moharia and subrace: Glabra (40 accessions), race: Maxima and subrace: Compacta (50 accessions), subrace: Spongiosa (50 accessions), race: Indica and subrace: Glabra (60 accessions) as unique for ICRISAT genebank.

CLL Gowda and HD Upadhyaya

Milestone B.1.1.3: Priorities areas identified for foxtail millet, little millet, kodo millet, proso millet and barnyard millet for collection/assembly in collaboration with NARS (CLLG/HDU, 2009)

Output target B.2: Assembled germplasm characterized and evaluated for economic traits for utilization (2009)

Activity B.2.1: Characterize new germplasm/data missing accessions of six small millets for morpho-agronomic traits

Milestone B.2.2.1: New germplasm of finger millet characterized for economic traits (CLLG/HDU, 2008)

Data of 20 finger millet accessions and four control cultivars evaluated in a replicated trial during 2005-2006 were analyzed. IEs 2340, 3194, 3790, 4974, and 6142 (116.7 – 188.3 cm) were significantly taller than the tallest control PR 2 and can be a source for feed and fodder. IEs 2498 and 4974 (56.7 – 61.7 mm) had wider inflorescence than the widest control RAU 8 ((55.0 mm). IEs 2498, 2683, and 2983 (10.7 – 11.3) have greater width of the longest finger than the best control PR 202 (10.00 mm). IEs 2498, 2578, 2887, 2903, and 4974 (11.1 – 13.4 g 1000 seed weight) had significantly greater seed size than the large seeded control RAU 8 (8.6 g). IEs 94, 2578, 3790, 3802, 4974, and 6236 (2.01 – 2.61 t ha^{-1}) were good for grain yield. To update characterization database 208 accessions were characterized.
C.L.L. Gowda and H.D. Upadhyaya

In a collaborative trail with the MPKV, Kolhapur, Maharashtra, India, 65 accessions of finger millet and five control cultivars were evaluated. IE 2957 (66 days) flowered earlier than all the five controls. 27 accessions (86-99 cm) were taller than the tallest control (85cm). Panicle exertion was significantly greater in IEs 2034, 2572, 3077, 3317,3945, 3952, 4734, and 6337 (15-88 cm) than all the five controls ((5-13 cm). IE 3475 (13) had significantly greater number of basal tillers than all the five controls (3-6). IEs 3045 and 4491 (10-11) had significantly greater inflorescence length than all the five controls (5-6 cm). IEs 2437, 2589, 3945, 3952, 5367, 6154, and 6421 (6-7 cm) had significantly greater inflorescence width than all the controls 9(4 cm). IE 3077 (5.03 t ha^{-1}) had greater grain yield than all the controls (2.64 – 4.82 t ha^{-1}).

1000 accessions of finger millet are being evaluated during 2006. Data recording is in progress.
CLLGowda, DD Kadam and HD Upadhyaya

Milestone B.2.2.2: Germplasm of foxtail millet, little millet, kodo millet, proso millet and barnyard millet characterized (CLLG/HDU, 2009)

Data of 20 foxtail millet accessions and four control cultivars evaluated in a replicated trial during 2005-2006 were analyzed. Eleven accessions (38–45 days) flowered significantly earlier than the earliest control ISe 375 (56 days). ISe 1258 and ISe 1658 (38 days) were the earliest accessions. ISe 769 and ISe 1434 (159.3 – 161.3 cm) were significantly taller than the tallest control ISe 1541 (146.7 cm). ISe 1433 and ISe 1434 (3.0) had significantly greater number of basal tillers than all the four controls (1.0-2.0).). ISe 1433 and ISe 1434 (234.7 – 239.3 mm) had significantly greater inflorescence length than all the four controls (131.7 – 198.7 mm). ISe 1434 (2.06 t ha^{-1}) had significantly greater grain yields than all the four controls (0.81 – 1.53 t ha^{-1}).

One hundred and forty eight accessions of foxtail millet, three accessions each of barnyard and little millet, two accessions of proso millet, and ten accessions of kodo millet were characterized to update databases during 2005-2006. Four accessions of barnyard millet, one accession each of foxtail millet and kodo millet, and five accessions of little millet are being characterized for updating databases during 2006-2007. Data recording is in progress.
CLL Gowda and HD Upadhyaya

Of the 159 accessions and four control cultivars of foxtail millet evaluated in an augmented design trial, 21 accessions (25-40 days) flowered significantly earlier than the earliest control ISe 1541 (47 days). ISe 1161, ISe 1227, ISe 1234, ISe 1254, and ISe 1320 (25 – 33 days) were the five earliest flowering accessions. The 21 accessions (5-8) had significantly greater number of basal tillers than all the controls (1-2). ISe 796, ISe 1009, ISe 1026, ISe 1134, ISe 1408, and ISe 1892 (6-8) were the best tillering accessions. ISes 719, 827, 1151, 1161, 1163, 1227, 1286, 1312, 1320, 1547, 1563, 1593, and 1655 (200.3 – 274.7 mm) had significantly greater Panicle exertion than all the four controls (120.7 – 196.0 mm). ISe 785 (257.3 mm) had significantly greater inflorescence length than all the control cultivars (120.3 196.0 mm). ISe 1780 (40.4 mm) had significantly greater inflorescence length than all the control cultivars (18.1 – 30.7 mm) and ISe 1593 (13.2 g) had significantly greater panicle weight than all the control cultivars (7.4 – 9.9 g).

500 accessions are being evaluated during 2006. Data recording is in progress.
CLL Gowda and HD Upadhyaya

Output target B.3: Germplasm accessions regenerated for conservation and distribution (2009)

Activity B.3.1: Regenerate critical accessions of small millets germplasm

Milestone B.3.1.1: Germplasm accessions of small millets with limited seed stock/viability regenerated and ***seed samples processed to for medium- and long-term conservation (****HDU, Annual)*

During the year, we regenerated 127 critical accessions of finger millet (15), foxtail millet (46), barnyard millet (42), little millet (2), proso millet (17), and kodo millet (5) for conservation and distribution. We also regenerated 20 accessions of six small millets in which seeds stocks are below critical levels under glasshouse conditions. 1500 accessions of two small millet crops (finger millet and foxtail millet) while grown essentially for evaluation were also used for replenishing stocks of the active collections.

HD Upadhyaya

Milestone B.3.1.2: Seed viability and health of new and regenerated small millets germplasm tested and viability of conserved germplasm monitored (HDU/RPT, Annual)

Milestone B.3.1.3: Small millets germplasm processed for safety back up (CLLG/HDU/BH, Annual)

Output target B.4: Unrestricted access and movement for small millets germplasm ensured (2009)

Activity B.4.1: Assure risk-free export and import of small millets germplasm materials

Milestone B.4.1.1: Requested germplasm of small millets distributed to bona fide users for utilization (RPT/HDU/NBPGR, Annual)

During 2006 we distributed a total of 1125 samples of small millets germplasm (finger millet-1038; foxtail millet-25; proso millet-17; little millet-15; kodo millet-15; and barnyard millet-15) for utilization to scientists in three countries in 15 consignments. Some of the special requests include sets of germplasm for collaborative evaluation with NARS in India and other countries. The finger millet germplasm distribution includes 1000 accessions of composite set along with three controls for evaluation at Tamil Nadu Agricultural University, Coimbatore, India.

Additionally, we provided 127 samples of germplasm for internal utilization. The total includes, finger millet-15; foxtail millet-46; proso millet-17; little millet-2; kodo millet-5; and barnyard millet-42.

RP Thakur, HD Upadhyaya and NBPGR

Unrestricted access to and movement of staple crop germplasm ensured: Crop researchers need to have access to diverse germplasm to develop improved broad genetic based cultivars; and to sustain biodiversity for current and future generations. A number of our NARS partners regularly request for materials in the form of repatriated landraces germplasm, new breeding materials, and finished and semi-finished products as well as nucleus seed of released varieties and hybrid parents for different uses. A total of 553 samples of 369 finger millet accessions were provided to Tanzania and Malawi and Sudan. The recipient organizations signed MTAs and these included NARIs, Universities, Seed Companies and NGOs for research as well as for further multiplication. On the other hand we received from ICRISAT Patancheru, a total of 441 sorghum and 11 pearl millet accessions from Zimbabwe

MA Mgonja, E Manyasa and E Muange

Milestone B.4.1.2: Requested germplasm of small millets exported for utilization and new germplasm imported for conservation after seed health evaluation and clearance through NBPGR (RPT/HDU/NBPGR, Annual)

Export of small millets: A total of 17 small millets (foxtail millet 8 and finger millet 9) were exported to Botswana (7 samples) and Sudan (10 samples).

RP Thakur, HD Upadhyaya and NBPGR

Output target B.5: Germplasm of small millets assembled and conserved for utilization at Regional Genebanks in Africa (2009)

Activity B.5.1: Identify collection gaps for finger millet in ESA and conduct collection mission to fill gaps

Milestone B.5.1.1: Gaps in finger millet collection identified and filled in at least 2 countries in ESA (MAM/SGM, 2009)

Activity B.5.2: Facilities improved at ICRISAT Niamey genebank for safety back up collections of small millets

Milestone B.5.2.1: Storage facilities for the safety back up of small millet collections improved at Niamey, Niger (BH/HDU/CLLG, 2007)

Upgrading storage modules: 8 new deep freezers received in 2006 and installed (total of 20 deep freezers now functioning at Niamey genebank).

BIG Haussmann

Output target B.6: Databases of small millets germplasm updated for utilization (2010)

Activity B.6.1: Update germplasm databases of small millets

Milestone B.6.1.1: Passport, characterization and evaluation data of small millets germplasm documented (HDU, 2007)

Documented the characterization data of 1000 finger millet composite collection and 155 accessions of foxtail millet core collection for important morpho-agronomic characters. Additionally, data on grain characters for 224 accessions of little millet was documented.

HD Upadhyaya

Milestone B.6.1.2: Gaps in germplasm characterization data filled for small millets (HDU, 2008)

Gaps were updated in characterization databases for 1000 accessions in finger millet, 155 accessions in foxtail millet, and 224 accessions of little millet.

HD Upadhyaya

Milestone B.6.1.3: Germplasm databases of small millets updated to SINGER format (HDU/CLLG, 2009)

Databases of small millets are being checked and updated for completeness of data for SINGER format.

HD Upadhyaya

Output C: Core and mini-core collections, and trait specific germplasm identified and evaluated for utilization; composite sets and reference collections established and genotyped to assess genetic diversity and population structure; and made available to partners annually on request; data capture, storage and analysis through appropriate management systems and dissemination through databases and web services

MTP Output Targets 2006

- *Trait specific germplasm of five mandate crops available for utilization (annual activity)*
- *Chickpea composite set (3000 accessions) established for utilization*
- *Chickpea composite set (3000 accessions) genotyped with SSR markers in collaboration with ICARDA*
- *Germplasm reference collection for chickpea (300 accessions) established*
- *Sorghum composite set (3000 accessions) genotyped with SSR markers in collaboration with CIRAD and CAAS*
- *Minicore of pigeonpea germplasm established*

Output target C.1: Core and mini core subsets of germplasm established for utilization (2010)

Activity C.1.1: Establish core and mini core collections of staple crops and small millets

Milestone C.1.1.1: Mini core sub set of pigeonpea germplasm established (HDU/CLLG, 2006)

A mini core of pigeon pea consisting of 146 accessions was constituted, by evaluating a core collection of 1256 accessions. Validation of mini core by examination of data for various morphological and agronomic traits indicated that almost the entire genetic variation and a majority of co adapted gene complexes present in core subset are preserved in the mini core collection. Due to its greatly reduced size, the mini core subset will provide a more economical starting point for proper exploitation of pigeonpea genetic resources for crop improvement for food, feed, fuel, and other agricultural and medicinal purposes. A journal article on the development of finger millet mini core subset has been published Crop Science (46: 2127-2132).

HD Upadhyaya and CLL Gowda

Milestone C.1.1.2: Mini core subset of sorghum established (HDU/CLLG, 2008)

Data analysis and assessment of diversity is in progress for establishment of sorghum mini core subset.

HD Upadhyaya and CLL Gowda

Milestone C.1.1.3: Mini core subset of finger millet established (HDU/CLLG, 2009)

Milestone C.1.1.4: Core subset of foxtail millet established (CLLG/HDU, 2010)

Output target C.2: Composite sets of germplasm established for utilization (2008)

Activity C.2.1: Establish germplasm composite sets of staple crops and small millets

Milestone C.2.1.1: Germplasm composite sets for groundnut, pigeonpea, and pearl millet (1000 accessions each) established (HDU/RB/CLLG/RKV/DH/CTH/SS/SC/KBS/RPT/KNR, 2007)

Groundnut: Composite collection comprising of 911 accessions from ICRISAT and 192 from EMBRAPA was developed using phenotypic characterization & evaluation data. The composite set included 184 accessions each of groundnut mini core collection (Upadhyaya et al, 2002) and mini core comparator. Among these 50 accessions are chilling tolerant at germination (Upadhyaya et al., 2001) and 18 accessions are drought tolerant (Upadhyaya, 2005a). We included 50 accessions from groundnut mini core for Asia region and 60 accessions having traits of economic importance from groundnut core collection for Asia region (Upadhyaya et al., 2005b). We included 36 elite/released varieties (27 from breeding and 9 from germplasm), and lines resistant to biotic stresses (5 leaf miner, 1 aphid, 10 jassid, 4 thrips, 7 termite, 2 PMV, and 12 rosette, 27 *A. flavus*/aflatoxin, 9 bacterial wilt, 7 bud necrosis, 7 early leaf spot, 14 late leaf spot, and 5 LLS & rust resistant interspecific derivates, 15 rust, 9 stem and pod rot, 41 multiple resistant). (Table 1). We also included 41 drought tolerant, 6 fresh seed dormancy lines, 4 high nitrogen fixing lines, 5 non-nodulating, 25 early maturing, 16 large seeded, 10 high shelling percentage, 9 high oil containing, 9 high protein containing, beside 26 accessions for morphological variants. To represent wild species of *Arachis* species, 52 accessions of 14 species were included (Table 2).

Table 2. Composite collection of groundnut

Character	Number of accessions	Character	Number of accessions
Mini core (50 chilling tolerant, 18 high SPAD)	184	Rust resistant	15
Mini core comparator	184	Stem & pod rot resistant	9
Asia mini core	50	Multiple resistant	41
Best accessions from Asia core	60	Drought tolerant	41
Released/elite cultivar	36	Fresh seed dormancy	6
Leaf miner	5	High biological nitrogen fixation	4
Aphid resistant	1	Non-nodulating	5
Jassid resistant	10	Early maturing	25

Character	Number of accessions	Character	Number of accessions
Thrip resistant	4	Large seeded	16
Termite tolerant	7	High Shelling turn over	10
Peanut mottle virus	2	High oil content	9
Rosette virus resistant	12	High protein content	9
A. flavus resistant	27	High SPAD	1
Bacteria wilt resistant	9	Interspecific derivates	5
Bud necrosis resistant	7	Morphological variance	26
Early leaf spot resistant	7	Used by Morag Ferguson	18
Late leaf spot resistant	14	14 wild *Arachis* species (2 *batizocoi*, 7 *cardenasii*, 1 *chiquitana*, 1 *diogoi* (*chacoensis*), 19 *duranensis*, 2 *hoehnei*, 1 *ipaensis*, 2 *kempff-mercadoi*, 3 *monticola*, 1 *paraguariensis*, 1 *stenophylla*, 9 *stenosperma*, 2 *villosa*, and 1 *villosulicarpa*)	52 (1 each resistant to thrips and rust, and ELS, 2 each resistant to LLS and root rots, 3 for high SPAD, and 9 multiple resistant accessions)

HD Upadhyaya, R Bhattacharjee, CLL Gowda, RK Varshney and DA Hoisington.

Pigeonpea: Pigeonpea composite collection consisting of 1000 accessions was constituted based on phenotypic, taxonomic and characterization/evaluation data The composite collection includes accessions – minicore collection (146), minicore comparator (146), from core collection (236), superior morpho-agronomic traits (301), resistant to biotic stresses (74), resistant to abiotic stresses (14), elite/released cultivars (20), and 65 accessions of 7 wild species (Table 3).

Table 3. Composite collection of pigeonpea

Type of material	No. of accessions	Type of material	No. of accessions
Mini core collection	146	Abiotic stresses	14
Comparator	146	Drought	7
Checks	4	Water logging	3
Resistant sources:		Salinity	4
Biotic stresses	75	*Trait specific selections*	306
Pod borer	20	High nodulation	2
Pod fly	5	Photoperiod insensitive	4
Pod borer and pod fly	4	Agroforestry	7
Wilt	6	Forage	6
Sterility mosaic	16	Vegetable	7
Alternaria blight	7	High protein	20
Phytophthora blight	6	Released cultivars	16
Stem canker	5	Morpho-agronomic traits	244
Nematodes	6	Wild species	65
		Others	244

HD Upadhyaya, R Bhattacharjee, CLL Gowda, RK Varshney, DA Hoisington and KB Saxena.

Pearl millet: We constituted a pearl millet composite collection of 1000 accessions and grown for characterization and regeneration during the rainy season (Table 4).

Table 4. Composition of pearl millet composite collection

Type of material	No. of accessions
Core collection	504
Tolerant to abiotic stresses	
Drought	6
Heat	3
Salinity	20
Resistant to biotic stresses	
Downy mildew	42
Ergot	20
Rust	23
Smut	15
Multiple disease resistant	8
High seed iron and zinc content (>42ppm)	4
High seed protein (>17%)	20
Yellow endosperm	2
Trait-specific selections	197
Sweet stalks	12
Forage type	8
Released cultivars	5
Gene pools	4
Wild relatives	
P. mollissimum	6
P. orientale	1
P. pedicellatum	15
P. polystachion	15
P. ramosum	2
P. schweinfurthii	1
P. violaceum	20
Contribution from crop improvement	47
Total	1000

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Milestone C.2.1.2: Germplasm composite sets for finger millet (1000 accessions) and foxtail millet (500 accessions) established (HDU/CTH/DH/CLLG/RKV/SC, 2008)

Output target C.3: Germplasm composite sets genotyped, diversity analysed, population structure assessed and reference sets of staple crops and small millets established (2010)

Activity C.3.1: Genotype composite collections for studying diversity and population structure and developing reference sets of staple crops and small millets

Milestone C.3.1.1: Chickpea composite set (3000 accessions) genotyped with SSR markers in collaboration with ICARDA (ICRISAT-HDU/SLD/DH/RKV/CLLG/SC; ICARDA- SMU, MB, BJF, 2006)

Composite collection of chickpea was developed based on available phenotypic, characterization, evaluation, geographic origin, and taxonomic data. It includes 2271 cultivated and three wild genotypes from ICRISAT and 726 cultivated and 13 wild accessions from ICARDA. This composite collection was genotyped using high throughput assay (ICRISAT: ABI3700 and ICARDA: ABI3100) and 50 SSR markers. ICRISAT generated data on 35 SSR loci and ICARDA on 15 SSR loci on 3000 accessions (Table 5).

Identification of Markers: Identified 50 polymorphic SSRs for genotyping the global composite collection from preliminary screening of 288 diverse chickpea germplasm accessions with 200 SSRs in 2004. Three di-and tri-nucleoid repeat motifs markers were of 174-241 (bp) allele size at annealing temperature of 60-65^{o}C (Huttel et al., 1999), 42 di-and tri-nucleoid repeat motifs markers were of 132-436 (bp) allele size at annealing temperature of 55- 65^{o}C (Winter et al., 1999), and 5 di- nucleoid repeat motifs markers were of 195-306 (bp) allele size at annealing temperature of 60-65^{o}C (Niroj et al., 2003).

Data Generated: ICRISAT and ICARDA were involved in genotyping of the composite collection using high throughput assay and 50 SSR markers. ICRISAT generated 35 SSR loci data and provided sufficient and good quality DNA for the 3000 accessions to ICARDA. ICARDA generated 15 SSR loci data.

All allelic data for 50 SSR primers on 3000 accessions resulted in less than 5% missing data (i.e., marker x genotype). The dataset was then analyzed using the allele-binning algorithm of Idury and Cardon (1997) called "Allelobin". The quality index of the markers was calculated based on missing data recorded for each of the markers and also to see if there was any allelic drift for these markers. Except for TA21, TA22, TA28, and TA58 (data not shown), all markers produced an allele size expected on the basis of SSR repeat motif.

Data templates consisting of 105000 (3000 x 35) data points generated at ICRISAT has been delivered to GCP repository. Data templates consisting of 45000 data points (3000 x 15) generated at ICARDA are currently being checked for completeness will be delivered to repository soon after verification by ICARDA

Table 5. Number of missing data, allele number and variation in allele size (bp) as detected in 3000 accessions genotyped for 50 SSR loci.

Locus	Missing data	No. of Alleles detected	Allele size range (bp)
TA206	224(7.5)	33	350-449
CaSTMS15	292(9.7)	30	209-368
CaSTMS2	332(11.1)	30	209-326
CaSTMS21	176(5.9)	21	150-210
NCPGR12	352(11.7)	28	206-272
NCPGR19	200(6.7)	29	288-464
NCPGR4	124(4.1)	16	149-203
NCPGR6	202(6.7)	24	213-361
NCPGR7	298(9.9)	15	201-243
TA113	258(8.6)	20	145-238
TA116	296(9.9)	34	159-276
TA117	714(23.8)	37	173-323
TA118	212(7.1)	43	116-263
TA130	360(12)	23	185-254
TA135	178(5.9)	21	125-221
TA14	132(4.4)	41	210-354
TA142	258(8.6)	23	113-197
TA200	132(4.4)	39	250-379
TA21	392(13.1)	42	275-422
TA22	344(11.5)	50	131-344
TA27	284(9.5)	32	192-306
TA28	252(8.4)	58	231-438
TA46	586(19.5)	24	126-195
TA64	182(6.1)	36	173-284
TA71	228(7.6)	41	139-277

Locus	Missing data	No. of Alleles detected	Allele size range (bp)
TA72	312(10.4)	50	159-375
TA76s	458(15.3)	34	149-314
TAA58	190(6.3)	45	205-349
TaaSH	106(3.5)	40	301-496
TR2	206(6.9)	53	161-326
TR29	208(6.9)	34	127-259
TR31	150(5)	16	165-234
TR43	256(8.5)	55	249-474
TR7	242(8.1)	27	146-248
TS84	210(7)	16	197-266
Ta2	180(6)	68	93-251
Ta80	156(5.2)	35	148-275
Ta203	240(8)	56	160-294
Ta5	128(4.3)	43	155-293
Ta96	224(7.5)	48	161-324
Tr1	216(7.2)	62	133-324
Ta3	236(7.9)	30	205-305
Ta8	50(1.7)	34	174-280
Ta144	156(5.2)	56	179-287
Ta42	104(3.5)	46	101-251
Ta176	134(4.5)	65	146-365
Ts45	142(4.7)	27	134-274
Ta11	256(8.5)	30	158-296
Ta78	88(2.9)	39	95-262
Ta194	54(1.8)	30	87-291

ICRISAT-HD Upadhyaya, SL Dwivedi, CLL Gowda, PM Gaur,
RK Varshney, DA Hoisington and S Chandra;
ICARDA- SM Udupa, M. Baum and BJ Furman

Milestone C.3.1.2: Sorghum composite set (3000 accessions) genotyped with SSR markers in collaboration with CIRAD and CAAS (ICRISAT- CTH/SS/HDU/PR/DH; CIRAD-CB/JFR/MD/LG/RR; CAAS-YL/TW/PL, 2006)

A total of 3393 sorghum lines, including 10 duplicates, have been genotyped at 43 SSR loci (marker data provided by CIRAD and ICRISAT; the SSR data provided by CAAS for an additional 5 SSR primer pairs has not been included in the final data set). Some 6% of the 145,889 potential data points were missing, so several SSRs and lines were removed from the analysis due to unacceptably high frequency of missing data, leaving 3365 sorghum lines and 39 SSR loci. This represents 87.5% of our original target of 3000 sorghum lines genotyped at 50 SSR loci. The composite set of sorghum lines for which adequate marker data are available for diversity analysis is comprised of 2.0% wild accessions, 8.6% breeding lines and released cultivars, and 98.4% landrace accessions representing all five major races of sorghum and their 10 intermediates, as well as materials originating from 13 sub-continents.

ICRISAT - CT Hash, HD Upadhyaya, Punna Ramu,and DA Hoisington;
CIRAD – C Billot, J-F Rami, L Gardes, R Rivalian,and M Deu;
CAAS – Y Li, T Wang and P Lu.

Milestone C.3.1.3: Reference collection of chickpea (300 accessions) established (HDU/ DH/RKV/CLLG/ PMG, 2006)

All allelic data for 50 SSR primers on 3000 accessions resulted in less than 5% missing data (i.e., marker x genotype). The accessions with maximum missing values were removed and based on analysis on 2915 accessions, we have selected a reference set consisting of 300 accessions. This reference set consists of 211 accessions of mini core (Upadhyaya and Ortiz, 2001), which has been phenotyped for several traits including drought tolerance traits, and another 89 accessions. The reference set captured 78% (1403 alleles) of the 1791

alleles detected in the 2915 accessions of composite collection. Biologically, the reference set consists of 267 landraces, 13 breeding lines/cultivars, 7 wild *Cicer* accessions, and 13 accessions with an unknown biological status. Geographically it consists of accessions from Asia (198), Africa (21), Europe (3), Mediterranean (56), Americas (10), Russian Federation (6), and 6 accessions whose geographical origin is not known. The reference set has 195 desi, 88 kabuli, and 10 intermediate type (pea-shaped), and 7 wild *Cicer* accessions. The reference set is diverse and can be used for extensive phenotyping for traits of interest and genotyping for association studies and eventually for efficient allele mining.

HD Upadhyaya, DA Hoisington, RK Varshney, CLL Gowda and PM Gaur

Milestone C.3.1.4: Four hundred wild and cultivated Sorghums from 60 villages in Mali genotyped for 15 SSR. Write up report (FS + NARS, 2006)

Sorghum diversity in Mali assessed by microsatellite markers: We collected wild, weedy (60 types) and cultivated Sorghums (340 varieties) in 60 villages from all Sorghum agro-ecological regions in Mali (except the "décrûe" Sorghums from Gao region). Sorghum seeds were germinated in a greenhouse. DNA was isolated from fresh leaves collected on 1-3 week old seedling per variety following a modified CTAB protocol at the University of Bamako (Mali). Fifteen SSR markers were chosen among those selected for their reliability and scoring accuracy among laboratories, their level of polymorphism and genome coverage, for the Challenge Program Generation. The M13-tails added to forward primers for each SSR were labeled with IRD700 or IRD800 Xuorochromes. Plants were genotyped at the Montpellier Languedoc-Roussillon Genopole (France) platform using Li-Cor automated sequencers. Saga GT v. 2.2 (Li-Cor) was used to determine allele sizes. First analyses showed a strong genetic structuration between Sorghum botanical races in Mali with wild/weedy Sorghums genetically closer to the guinea margaritiferum cluster.

F Sagnard in collaboration with CIRAD and University of Bamako, Mali

Milestone C.3.1.5: Diversity of wild and cultivated Pearl Millet in Niger published (FS, JN, BG, IRD, CIRAD, NARS, 2006)

Diversity analysis of Pearl Millet to identify priorities for conservation programs: In Niger, pearl millet covers more than 65% of the total cultivated area. Analyzing pearl millet genetic diversity, its origin and its dynamics is important for *in situ* and ex situ germplasm conservation and to increase knowledge useful for breeding programs. Using 25 microsatellite markers, the genetic diversity of 46 wild and 421 cultivated accessions showed a significantly lower number of alleles and lower gene diversity in cultivated pearl millet accessions than in wild accessions. A strong differentiation between the cultivated and wild groups in Niger was observed. Wild accessions in the central region of Niger showed introgressions of cultivated alleles. Accessions of cultivated pearl millet showed introgressions of wild alleles in the western, central, and eastern parts of Niger. A journal article has been published in the Theoretical and Applied Genetics (114: 49–58).

F Signard, B Gerard and J Ndejeunga in collaboration with CIRAD and University of Bamako, Mali

Milestone C.3.1.6: Diversity of sorghum composite collection analyzed and reference set (300 accessions) established (ICRISAT--CTH/HDU/SS/RKV/DH/CLLG/SC/JB; CIRAD--CB/JFR/MD/LG/RR; CAAS-YL/TW/PL, 2007)

Diversity analysis of the sorghum composite collection was initiated in 2006 using a subset of 3365 sorghum lines genotyped at 39 single-copy SSR loci. The marker data clearly demonstrate the tremendous range of genetic diversity available in this sorghum composite collection. We detected an average of 19 alleles per SSR locus and the vast majority of the alleles detected are rare (>75% of alleles having frequencies <5%, and >50% of alleles detected having frequencies <1%).

The data set was analyzed using a factorial analysis of the simple matching dissimilarity index implemented in the Darwin diversity analysis software package. The overall picture emerging from this analysis is one of complex genetic structure within sorghum germplasm—as expected from previous studies of sorghum genetic diversity based on passport information, morphological characters, and RFLP marker data. Several distinctly different groups of wild accessions were detected. Among landrace accessions, race bicolor lacked structure and was found scattered across the first two axes of the factorial analysis. Race kafir accessions formed a single group, as expected from their origin from a relatively compact region in southern Africa. Race durra accessions formed four groups, each associated with a specific geographic region (eastern Africa, southern Africa, the Indian sub-continent, and eastern Asia), and these were congruent with four groups of caudatum accessions (Fig 1). This finding is in agreement with earlier studies that have found greater similarities in molecular marker

genotype between landrace accessions of difference races but originating from a common region than between landrace accessions of a common race originating from different regions. The guinea race accessions also exhibited considerable structure, with separate groups of materials originating from eastern and southern Africa, western and central Africa, and the Indian subcontinent, as well as a clearly defined subgroup comprised of the margaritiferums that may represent an independent domestication event. Intermediate-race landrace accessions also formed distinct clusters (by hybrid race and geographic region). We are now attempting to use this information to choose a representative "core" or "reference" subset of 300 to 500 accessions for more detailed study.

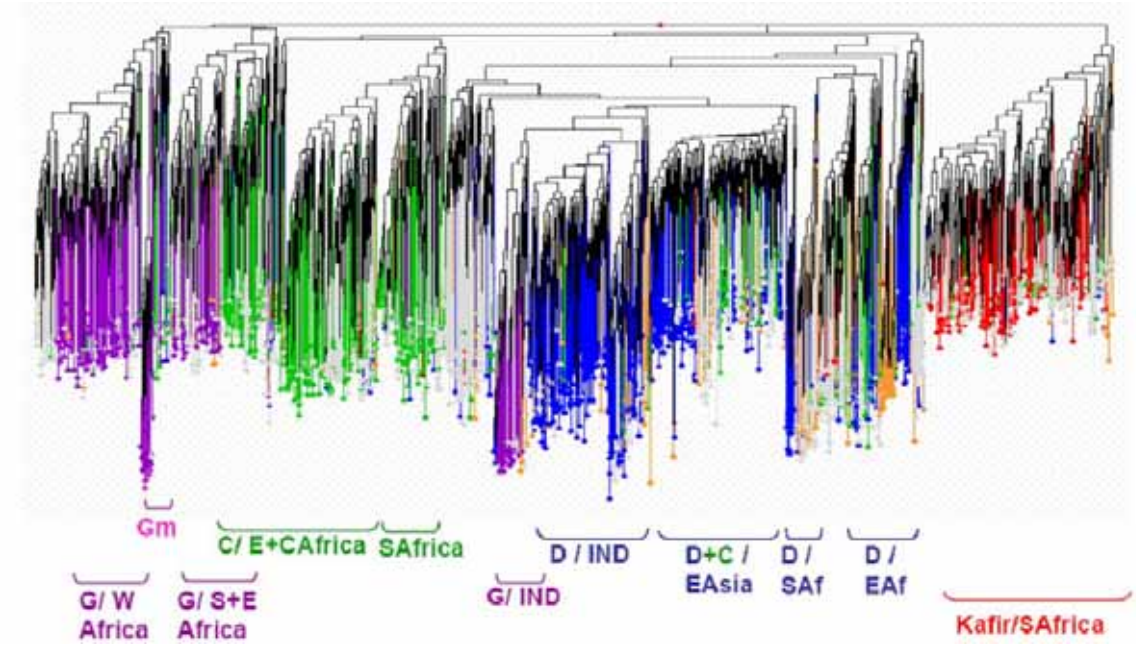


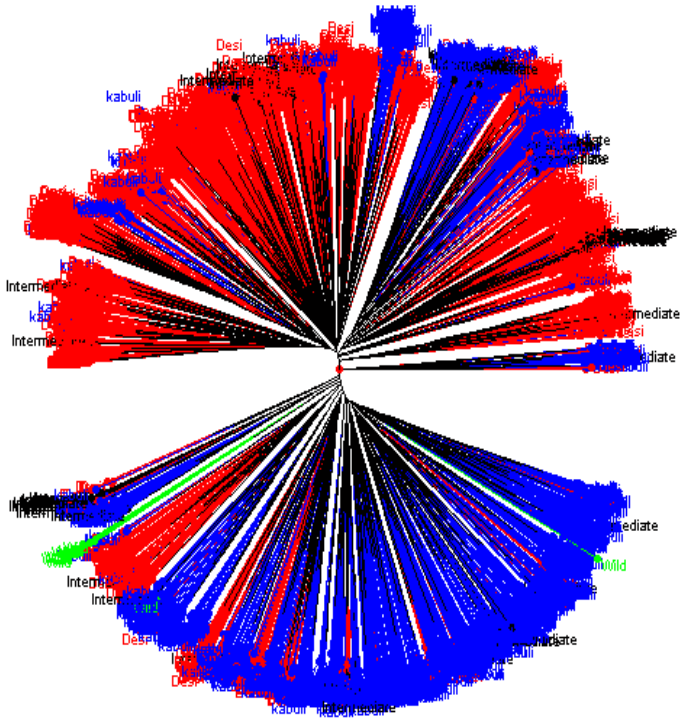
Fig 1. Relationships among 3008 sorghum landrace accessions in the Generation Challenge Program composite germplasm collection for sorghum, based on allelic variation at 39 SSR loci distributed across all 10 sorghum linage groups. In this figure landraces representing the five major sorghum races are colored: race bicolor accessions are orange, race kafir are red, race durra (D) are blue, race caudatum (C) are green, and race guinea (G) are purple (including the margaritiferum group, Gm). Geographic regions from which the different groups of accessions originated are: W Africa = Western Africa; S Africa = Southern Africa; E Africa = Eastern Africa; C Africa = Central Africa; IND = Indian subcontinent; and, E Asia = Eastern Asia (Figure courtesy of Claire Billot).

ICRISAT - CT Hash, HD Upadhyaya, Punna Ramu and DA Hoisington;
CIRAD – C Billot, J-F Rami, L Gardes, R Rivalian and M Deu;
CAAS – Y Li, T Wang and P Lu.

Milestone C.3.1.7: Genetic diversity and population structure of chickpea and sorghum assessed (HDU/CTH/RKV/PR/CLLG/SC/DH/PMG/MB/SMU/CB/JB/SC, 2007)

Chickpea:
The composite collection was genotyped using high throughput assay (ICRISAT: ABI3700 and ICARDA: ABI3100) and 50 SSR markers. ICRISAT generated data on 35 SSR loci and ICARDA on 15 SSR loci on 3000 accessions. Except for TA21, TA22, TA28, and TA58, all markers produced an allele size expected on the basis of SSR repeat motif). Preliminary data analysis detected 1829 alleles, ranging from 15 to 68 alleles with an average of 36.6 alleles per SSR locus. Mean PIC (Polymorphism Information Content) value of 0.858 (ranging from 0.471 to 0.974) and gene diversity 0.873 (ranging 0.536 – 0.975) were observed in the entire composite collection (Table 6). Kabuli types were more genetically diverse than the other three types. Gene diversity ranged from 0.253 to 0.965 in kabuli types, 0.419 to 0.974 in desi, 0.479 to 0.955 in pea-shaped, and 0.560 to 0.928 in wild types. Accessions from the West Asia region revealed high gene diversity (0.871) while those from Oceania revealed lowest gene diversity (0.504) (Table 5). Detected 1539 rare and 290 common alleles at 5% and 1137 rare and 692 common alleles at 1% in the entire composite collection. There were 252 alleles that

were detected in all the four biological groups (desi, kabuli, pea-shaped, and wild relatives). Accessions from the Mediterranean region had the largest number of region-specific alleles (137) and 69 alleles were common across all the seven geographical regions. Principal coordinate analysis delineated the accessions in two clusters (Fig 2). The desi and kabuli chickpeas each formed two distinct clusters; however, a number of desi chickpeas also grouped into kabuli cluster indicating progressive evolution of kabuli traits from the desi chickpeas.



Desi (1668); Kabuli (1167); Intermediate (70); Wild (10)

Fig. 2. Genetic structure of the global composite collection (50 SSR loci and 2915 accessions) of chickpea as revealed by the Principal Coordinate Analysis (PCoA) using DARwin 5.0 Structure program.

Table 6. Range and average PIC values and gene diversity in the global composite collection of chickpea.

Category	No. of accessions	PIC		Gene Diversity	
		Average	Range	Average	Range
Composite collection	3000	0.858	0.471-0.974	0.873	0.536 - 0.975
Composite collection	2915	0.858	0.468 –0.974	0.872	0.534 - 0.975
Reference sample	300 (211+89)	0.872	0.488 –0.964	0.884	0.540 - 0.965
Biological classification					
Desi chickpea	1712	0.836	0.382-0.973	0.851	0.419 - 0.974
Kabuli chickpea	1197	0.835	0.243-0.963	0.849	0.253 - 0.965
Pea-shaped chickpea	71	0.823	0.443-0.953	0.842	0.479 - 0.955
Wild species	20	0.834	0.499-0.923	0.850	0.560 - 0.928
Geographical classification					
Africa	153	0.791	0.297-0.946	0.806	0.308 - 0.949
North and Central America	95	0.815	0.431-0.944	0.834	0.478 - 0.946
South America	50	0.757	0.113-0.938	0.779	0.117 - 0.941
South and southeast Asia	1163	0.81	0.322-0.968	0.826	0.348 - 0.969

Category	No. of accessions	PIC		Gene Diversity	
		Average	Range	Average	Range
West Asia	740	0.858	0.451-0.967	0.871	0.478 - 0.968
Russian Federation	45	0.781	0.338-0.937	0.800	0.369 - 0.940
Mediterranean	650	0.824	0.260-0.964	0.839	0.272 - 0.839
Europe	66	0.794	0.167-0.932	0.809	0.173 - 0.936
Oceania	3	0.428	0.000-0.810	0.504	0.000 - 0.833
Unknown	35	0.837	0.469-0.935	0.854	0.545 - 0.939

ICRISAT- HD Upadhyaya, SL Dwivedi, CLL Gowda,
RK Varshney and DA Hoisington;
ICARDA- SM Udupa, M Baum,and BJ Furman.

Milestone C.3.1.8: Four hundred wild and cultivated Sorghum accessions from Kenya genotyped for 30 SSR markers (DK, SdV, FS + NARS, 2007)

Milestone C.3.1.9: Paper accepted on the in situ diversity of 472 Sorghum varieties collected in 79 villages in Niger (FS, BG, JN + NARS + CIRAD + IRD, 2007)

Milestone C.3.1.10: Comparative phylogeography of wild, weedy and cultivated Sorghums in Mali published (FS, PST, NARS + CIRAD, 2008)

Milestone C.3.1.11: Comparative phylogeography of wild, weedy and cultivated Sorghums in Kenya published (FS, SdV, DK, NARS + University of Free State, 2008)

Milestone C.3.1.12: Diversity assessment of sorghum and chickpea published (HDU/CTH/RKV/CLLG/SS/DH/SC/JB, 2009)

A journal article "Development of a composite collection for mining germplasm possessing allelic variation for beneficial traits in chickpea' published in Plant Genet. Resources 4: 13-19.

A poster "Genotypic and phenotypic variation in the global collection of chickpea (*Cicer aritienum* L.)" presented at the Third International Conference on Legume Genomics and Genetics, 9-13 April 2006, Brisbane, Queensland, Australia.

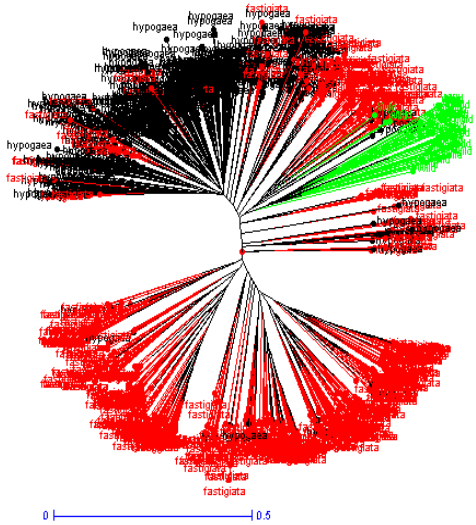
HD Upadhyaya, SL Dwivedi, CLL Gowda, RK Varshney,
DA Hoisington,and S Chandra

Milestone C.3.1.13: Data sets for sorghum and chickpea composite set made available globally via Internet (JB/HDU/CTH/RKV/CLLG/SS/DH/SC, 2008)

Milestone C.3.1.14: Diversity and population structure of groundnut composite collection analyzed and reference set (300 accessions) established (HDU/RB/RKV/CLLG/DH/JB, 2008)

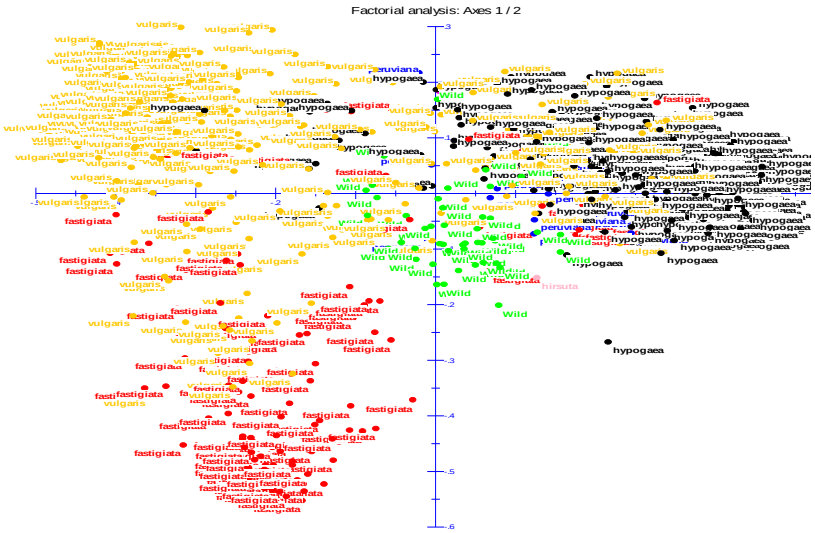
A preliminary analysis of data on 916 groundnut accessions and 21 SSR markers was carried out using DARwin 5.0 Structure program to determine the population structure of the composite collection. The software removes all those accessions/markers that have high missing values and finally 900 accessions were considered for principal coordinate analysis considering the taxonomical classification of *Arachis*, i.e. at the level of two subspecies and six botanical varieties. The analysis detected a total of 506 alleles, ranging from 6 (7H6) to 47 (5D5) with a mean of 24.1 alleles per locus and mean PIC value of 0.797 (ranging from 0.483 to 0.923).

Principal coordinate analysis (PCoA) using DARwin 5.0 on subspecies revealed that both *hypogaea* and *fastigiata* formed distinct clusters however, a number of *fastigiata* accessions also grouped with *hypogaea* types (Fig. 3), which maybe attributed to the geographic origin of these accessions. This is also confirmed when PCoA was performed considering botanical varieties (Fig. 3). The wild accessions formed a different cluster in both the cases and grouped with *hypogaea* types, indicating a close relation between them (Fig. 3 & 4).



fastigiata: Red; *hypogaea:* Black; Wild: Light Green

Fig. 3. Tree diagram of 900 accessions with 21 SSR markers at subspecies level.



fastigiata: Red; *hypogaea:* Black; *peruviana:* Blue; *vulgaris:* Orange; *hirsuta:* Pink; *aequatoriana:* Magenta; Wild: Light green

Fig. 4. Factorial analysis at the level of botanical varieties

To ascertain the quality and position of the SSR markers, these will be checked on 15-20 plants in each of three F_2 populations, whose parents have been included in the composite collection. Only those SSR markers that showed polymorphism on the parents will be checked on the F_2 population. Three crosses involving five parents have been selected for this purpose and 14 out of 21 markers have been found to be polymorphic between the parental combinations. The F_2 populations are being genotyped presently and the results will confirm the location/position of these SSR markers and would ensure appropriate peak calling. Data generated from the fingerprinting will be then subjected to statistical analysis using different computational tools.

HD Upadhyaya, R Bhattacharjee, CLL Gowda, RK Varshney and DA Hoisington.

Milestone C.3.1.15: Diversity and population structure of pigeonpea composite collection analyzed and reference set (300 accessions) established (HDU/RB/RKV/DH/SC/JB, 2009)

At ICRISAT, genotyping of 1000 accessions (composite collection) was carried out. DNA was extracted from 12 plants per accession and a series of artificial pools having different proportions of two genotypes showing polymorphism for a given SSR marker were developed and screened with the corresponding polymorphic SSR marker. The coefficients of correlations were analyzed between different proportion of alleles recorded (corresponding to 12 plants per accession) and proportion of genomic DNA used for the corresponding accession. As a result, 20 SSR markers with highly significant correlations (r^2> 0.9) were identified.

The selected 20 polymorphic SSR markers were optimized for PCR reactions following Taguchi method (Taguchi, 1986) as described in Cobb and Clarkson (1994). A fluorescent-based multiplex genotyping system was then used to generate different multiplexes, which were used to fingerprint the composite collection. The amplified PCR products were separated by capillary electrophoresis in an automated system using ABI 3700. SSR fragment sizes were called to two decimal places using the Genotyper v 3.7 software. The allelic data was analyzed following allele binning algorithm, written in a C program at ICRISAT called as "Allelobin", based on the least squares algorithm of Idury and Cardon (1997). Less than 5% missing data (i.e. marker x genotype) was recorded in the dataset and all the markers produced allele size that was expected on the basis of repeat motif of each of the SSR markers. Preliminary analysis has been completed on 17 out of 20 markers and 3 markers showed poor quality index (Table 6), which may be due to high missing vales recorded for these markers. Preliminary analysis detected a total of 184 alleles, ranging from 2 (PKS18) to 24 (CCB8) with a mean of 10.8 alleles per locus and mean PIC value of 0.31 (ranging from 0.02 to 0.0.59) (Table 7).

Table 7. SSR primers used in the study with information on their repeat units, quality index, number of alleles and PIC values.

Primer	Repeat Unit	Quality Index	No. of Accessions Genotyped	No. of Alleles	PIC Values
CCB1	(CA)10	0.26	1000	20	0.46
CCB9	(CT)22	0.46	951	18	0.53
PGM3	(GAA)5G(GAA)5	0.42	976	11	0.50
PGM101	(AC)7	0.14	984	8	0.43
CCB7	(CT)16	0.24	1000	17	0.47
PGM106	(AAG)13	0.31	1000	11	0.33
PGM109	(CTT)8	0.18	1000	5	0.44
CCB8	(CT)30	0.27	961	24	0.47
PKS21	(CT)6TT(CT)2	0.23	997	8	0.13
PGM5	(GAA)6	0.23	948	8	0.13
PGM10	(AGA)5	0.22	986	7	0.09
PGM16	(TC)8	0.26	962	7	0.17
PGM82	(AC)6AAG(CTAA)3	0.30	995	7	0.32
PKS18	(GGT)4	0.11	999	2	0.02
PKS25	(TTAT)4	0.40	967	10	0.50
PKS26	(TTA)4	0.17	931	7	0.07
CCB10	(CA)15	0.32	898	14	0.59

Principal co-ordinate analysis on 1000 accessions and 20 SSR markers was carried out using DARwin 5.0 Structure program to determine the population structure of the composite collection. The software removes all those accessions/markers that have high missing values and finally 970 accessions were considered for the analysis. Principal co-ordinate analysis grouped the accessions into two distinct clusters separating the cultivated accessions from the wild one's (Fig.5).

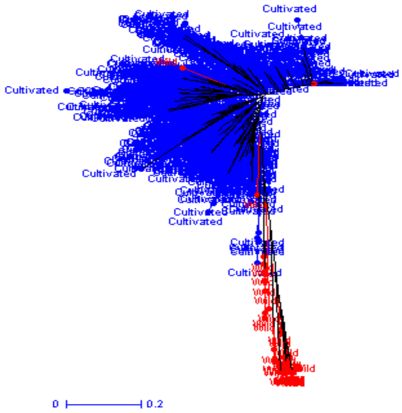


Fig. 5. Factorial analysis of 970 accessions with 17 SSR markers.

HD Upadhyaya, R Bhattacharjee, RK Varshney,
DA Hoisington, CLL Gowda, and S Chandra

Milestone C.3.1.16: Data sets for groundnut and pigeonpea composite set made available globally via Internet (JB/HDU/RKV/DH/RB/CLLG/SC/KBS, 2010)

Milestone C.3.1.17: Diversity and population structure of finger millet and foxtail millet composite collections analyzed and reference sets (300 accessions finger millet, 200 foxtail millet) established (HDU/CTH/ RKV/SS/DH/CLLG/SC/JB, 2009)

*Milestone C.3.1.18: Diversity and population structure of pearl millet composite collection analysed and reference set (300 accessions) established (HDU/CTH/ RKV/SS/DH/CLLG/SC/JB, 2010)***)**

Pearl Millet: A set of 1000 accessions of pearl millet composite collection were planted in the field. DNA was extracted at 15th day from leaf tissues of 15 plants by using high – through put method and quality checked by using agarose gel electrophoresis. Quantification of DNA concentration has been done with fluorescence detector and diluted to 5 ng/ul as working concentration in ABI 3700. Twenty polymorphic SSR were selected for genotyping. PCR conditions optimized. Finger printing is in progress.

HD Upadhyaya, CT Hash, RK Varshney, S Senthelvel,
DA Hoisington, CLL Gowda, S.Chandra and B Jayashree

Milestone C.3.1.19: Diversity assessment of pearl millet published (HDU/CTH/SS/RKV/DH/SC/JB, 2010)

Milestone C.3.1.20: Data sets for pearl millet, finger millet, and foxtail millet composite sets made available globally via Internet (JB/HDU/CTH/SS/RKV/CLLG/DH/SC, 2010)

Activity C.3.2: Diversity assessment in groundnut rosette virus resistant germplasm

Milestone C.3.2.1: The diversity of the sources of resistance to the groundnut rosette virus in groundnut assessed and documented (ESM/HDU/Others, 2009)

Activity C.3.3: Phenotypic and genotypic diversity assessment of sorghum germplasm in eastern and southern Africa

Milestone C.3.3.1: Phenotyping and genotyping of germplasm held by NARS (2006)

This study aims at characterizing and assessing diversity at the phenotypic and genotypic levels in sorghum germplasm currently used in NARS breeding programs in east and central Africa. The germplasm includes resources within national gene banks, international nurseries and important breeder germplasm. We have adopted a standardized approach to documentation, phenotyping and also in molecular characterization of the germplasm using 24 SSRs as part of the GCP set of high quality microsatellite markers that are being used for the survey of global composite set of sorghum germplasm. A project planning meeting was held in April 2006 in Nairobi, followed by a phenotyping workshop. Project partners agreed to work on a regional composite of 1720

accessions representing landraces, farmer varieties and breeders' lines. Seed multiplication and phenotyping of 1260 Sorghum accessions has started in Eritrea, Kenya, Uganda, Tanzania and Sudan. 27 consensus descriptors representing all the sorghum developmental stages are being used for morphological characterization. These are partially derived from the IPGRI descriptor lists for Sorghum. Characterization data is being generated and documented in the Access Database developed during the phenotyping workshop. Genotyping activities have been initiated, starting with the extraction, quantification and quality checks of DNA from 196 sorghum samples from Tanzania. The DNA will be used both for genotyping and repository as part of the GCP Sorghum global collection. Initial genotyping has started with the screening of 196 accessions using 6 polymorphic SSR markers. All the 24 SSR markers that will be used for genotyping have been optimized and screened for polymorphism. A database with an initial entry of 1260 germplasm accessions that are being phenotyped and genotyped by the participating NARS has been developed. The information on the accessions mainly comprises passport data and will be used to identify duplicates through the unique identifiers. Two PhD students have been registered at the University of Wad Medani and University of Free State, South Africa to phenotype and genotype 400 samples from Sudan. 4 MSc students have been registered in national universities in Ethiopia, Kenya, Uganda and Tanzania to advance the phenotyping and genotyping activities.

D Kiambi, DA Hoisington, CT Hash and S de Villiers

Milestone C.3.3.2: Phenotypic characterization of sorghum germplasm held by ESA NARS completed (DK, 2007)

Knowledge of the extent, nature and structure of genetic diversity in crop germplasm accessions is important for defining strategies for conservation and utilization. This project aims at characterizing and assessing diversity at the phenotypic and genotypic levels in sorghum germplasm currently used in NARS breeding programs in east and central Africa. The germplasm includes resources within national gene banks, international nurseries and important breeder germplasm. The project has adopted a standardized approach to documentation, phenotyping and also in molecular characterization of the germplasm using 24 SSRs as part of the GCP set of high quality microsatellite markers that are being used for the survey of global composite set of sorghum germplasm. A project planning meeting was held in April 2006 in Nairobi, followed by a phenotyping workshop. Project partners agreed to work on a regional composite of 1720 accessions representing landraces, farmer varieties and breeders' lines. Seed multiplication and phenotyping of 1260 Sorghum accessions has started in Eritrea, Kenya, Uganda, Tanzania and Sudan. 27 consensus descriptors representing all the sorghum developmental stages are being used for morphological characterization. These are partially derived from the IPGRI descriptor lists for Sorghum. Characterization data is being generated and documented in the Access Database developed during the phenotyping workshop. Genotyping activities have been initiated, starting with the extraction, quantification and quality checks of DNA from 196 sorghum samples from Tanzania. The DNA will be used both for genotyping and repository as part of the GCP Sorghum global collection. Initial genotyping has started with the screening of 196 accessions using 6 polymorphic SSR markers. All the 24 SSR markers that will be used for genotyping have been optimized and screened for polymorphism. A database with an initial entry of 1260 germplasm accessions that are being phenotyped and genotyped by the participating NARS has been developed. The information on the accessions mainly comprises passport data and will be used to identify duplicates through the unique identifiers. Two PhD students have been registered at the University of Wad Medani and University of Free State, South Africa to phenotype and genotype 400 samples from Sudan. 4 MSc students have been registered in national universities in Ethiopia, Kenya, Uganda and Tanzania to advance the phenotyping and genotyping activities.

D Kiambi, DA Hoisington, CT Hash and S de Villiers

Milestone C.3.3.3: Molecular characterization of germplasm held by ESA NARS completed (DK, 2007)

Milestone C.3.3.4: Phenotypic and molecular data for sorghum standardized and analyzed (DK, 2008)

Activity C.3.4: Development of a database for documentation and retrieval of morphological and molecular data

Milestone C.3.4.1: Database of passport information, farmer-knowledge, pedigrees, phenotyping and genotyping data of sorghum accessions held in ESA national genebanks and international nurseries developed (DK, 2008)

Output target C.4: Core, mini core, and or reference sets of germplasm evaluated for utilization in Asia (2009)

Activity C.4.1: Evaluate core/mini core/reference sets of staple crops and small millets for agronomic traits

Milestone C.4.1.1: Mini core collections of chickpea, groundnut, and pigeonpea evaluated in multilocations in Asia (HDU/CLLG/NARS, 2008)

Chickpea: 211 accessions of chickpea mini core collection with five control cultivars (Annigeri, ICCV 2, ICCV 10, L 550, KAK 2, G 130) were evaluated in augmented designed trials at Patancheru, Palampur, Chandigarh, and Bangalore in India, and at Culiacan in Mexico. Data is awaited for out stations. At ICRISAT, Patancheru ICCs 708, 3631, 6816, 8950, and 16903 (2.9-3.3 t ha $^{-1}$) were top five accessions with greater seed yield than all the five control cultivars (1.2-2.8 t ha $^{-1}$). ICC 16309 (42 days to 50% flowering and 105 days to maturity) was early flowering and early maturing than high yielding controls ICCV 10 (45 and 110 days) and Annigeri (45 and 108 days)

Pigeonpea: Pigeonpea mini core collection consisting of 146 accessions was evaluated with four control cultivars (ICP 11543, ICP 6971, ICP 8863, ICP 7221) in augmented designed trials at Patancheru, Dholi, Bangalore, Kanpur, Khaegone, Sardar Krishinagar, and Badnapur in India, and at Dubai in UAE. Data is awaited for out stations. At ICRISAT, Patancheru, ICPs 3451, 4167, 6123, 8255, and 14722 (2.6 – 2.8 t ha $^{-1}$) produced greater seed yield than the best control cultivar ICP 8863 (2.0 t ha $^{-1}$).

Groundnut: 184 accessions of groundnut mini core collection were evaluated with four control cultivars (Gangapuri, M 13, ICGS 44, ICGS 76) in augmented designed trials at Patancheru, Jalgaon, Raichur, Durgapura, and Aliyanagar in India, Qingdao in China, and two sets at Lilongwe in Malawi. Data is awaited for out stations. At ICRISAT, Patancheru ICGs 3992, 10185, 12625, and 14482 (4.3 – 4.6 t ha $^{-1}$) had greater pod yield than the high yielding control cultivar M 13 (4.2 t ha $^{-1}$). ICGs 115, 397, 3746, 10890, and 15042 (>75%) had greater shelling turn over, and ICGs 4746, 5662, 6993, and 9905 had greater seed size (100-110 g).

HD Upadhyaya and CLL Gowda

Milestone C.4.1.2: Core collection of pearl millet evaluated in multilocations in India (HDU/CLLG/NARS, 2008)

Sets of pearl millet core collection consisting of 504 accessions and four control cultivars were sent to All India Pearl millet improvement Program, Mandor for evaluation at five locations in India. Data is awaited from these locations. The preliminary analysis at ICRISAT, Patancheru revealed that IPs 9496, 11584, 15010, 17554, and 17566 (60-62 days) were early flowering, IPs 8130, 10401, and 12650 (30-56 cm) were short statured good for mechanical management and IP 12570 (210 cm) was good for fodder. IPs 5207, 5447, 10290, 11457, and 12338 (56 – 64 cm) had long spike length. IPs 7440, 8000, 10456, 10471, and 17753 (35-40 mm) had thick spikes.

HD Upadhyaya and CLL Gowda

Milestone C.4.1.3: Reference sets of chickpea and sorghum phenotyped for agronomic traits (HDU/CLLG/CTH/BVSR, 2008)

300 accessions of chickpea reference set are being phenotyped for yield and other important agronomic and morphological descriptors along with five control cultivars. Data recording is in progress.

HD Upadhyaya, N Lalitha and CLL Gowda

Output target C.5: Mini core and or reference collections of staple crops and small millets evaluated to identify trait specific germplasm (2012)

Activity C.5.1: Evaluate mini core and reference collections for resistance to important biotic stresses

Milestone C.5.1.1: Mini core and reference collections of chickpea germplasm evaluated for resistance to AB, BGM, wilt, collar rot and dry root rot under controlled environment and field (SP/HDU/PMG/RKV/CLLG, 2008)

Chickpea mini core evaluated for diseases resistance: Chickpea mini core subset consisting of 211 accessions were reevaluated to confirm their resistance to ascochyta blight (AB), botrytis gray mold (BGM), fusarium wilt

(FW), collar rot (CR) and dry root rot (DRR) diseases under controlled environment conditions at ICRISAT-Patancheru. Standardized optimum conditions for individual diseases were used for evaluation. Accessions were categorized and grouped based on their reaction to each disease in both tests.

Resistance to AB: High levels of resistance to AB were not found in mini core subset. However, three desi type accessions ICC 1915, ICC 6306, ICC 11284 were identified as moderately resistant (3.1 to 5 rating) to AB. Among these three accessions, ICC 6306 had a 100 seed mass of 25 g while the rest had <20 g.

Resistance to BGM: Absolute resistance to BGM was not found in mini-core subset. But, 55 accessions had moderately resistant reaction (3.1 to 5 rating). Of these moderately resistant accessions, 33 were Kabuli type, 17 were desi and remaining five of intermediate type. Of these Kabuli accessions, ICC 8151 and ICC 14199 were very bold with a 100 seed mass of around 58 g. Among desi types, one accession ICC 13124 had highest 100 seed mass of 35.4 g.

Resistance to FW: Several accessions had high levels of resistance to FW. Twenty one accessions were found asymptomatic and 25 accessions were identified as resistant (<10% mortality) to FW infection. Among asymptomatic accessions, one accession ICC 8058 was Kabuli type with 100 seed mass of 33.8 g. Two kabuli accessions, ICC 13816 and ICC 13441 with 100 seed mass of 29 g and 16.7g respectively were identified among resistant group.

Resistance to DRR: Only six accessions, ICC 1710, ICC 2242 (desi type), ICC 2277, ICC 11764, ICC 12328, and ICC 13441 (Kabuli type) were found moderately resistant (3.1 to 5 rating on 1-9 rating scale) to DRR. Of the promising Kabulis, ICC 11764 and ICC 12328 had a 100 seed mass of 28.8 g and 27.5 g respectively.

Resistance to CR: All the accessions of the mini core sub set were found susceptible to collar rot.

Multiple disease resistance: Among the mini core accessions, no line was found resistant or moderately resistant to more than two diseases in both tests. ICC 11284 (desi type) was the only accession with moderate level of resistance (3.1 to 5 rating on 1-9 rating scale) to both AB and BGM diseases. Combined resistance to AB and soil borne diseases was not observed in any of the accessions tested. Two accessions, ICC 11764 and ICC 12328 had a moderate resistance (3.1 to 5 rating on 1-9 rating scale) to both BGM and DRR. Combined resistance to FW (<10% incidence) and BGM (3.1 to 5 rating) was found in 11 accessions. Four accessions, ICC 1710, ICC 2242, ICC 2277 and ICC 13441 had a combined resistance to both FW (11 to 20% incidences) and DRR (3.1 to 5 rating on 1-9 scale).

S Pande, HD Upadhyaya and CLL Gowda

Milestone C.5.1.2: Mini core and reference collections of pigeonpea germplasm evaluated for resistance to wilt and sterility mosaic diseases under controlled environment and field conditions (SP/PLK/FW/HDU/KBS, 2008)

Fusarium wilt and SM resistance in mini core accessions: One hundred and forty six mini core accessions were evaluated for combined resistance to wilt and sterility mosaic disease (SMD) under artificial epiphytotic conditions using standard field evaluation techniques at ICRISAT-Patancheru. Threshold level of wilt fungus, Fusarium udum was maintained by incorporating chopped wilted pigeonpea plants in the sick plot every year. Each and every plant of test entries planted in wilt sick plot, were leaf inoculated with SM infested leaves using leaf staple technique at two-leaf stage for successful SM infection. Susceptible cultivars ICP 2376 for wilt (resistant to SM) and ICP 8863 for SM (resistant to wilt) were planted along with test material, after every ten test rows as indicator rows. Additionally, natural incidence of Phytophthora blight (PB) was also recorded at seedling stage during the current season as there were frequent and continuous rains.

Only two mini core accessions, ICP 11015 and ICP 14819 had a combined resistance (< 10%) to both wilt and SMD. Nine accessions, ICPs 7869, 9045, 11015, 11059, 11230, 11281, 11910, 14819 and 14976 were asymptomatic and 19 accessions were resistant to SM. Eight accessions, ICPs 4903, 6739, 6815, 7057, 10559, 11015, 14638 and 14819 were asymptomatic and 70 were found resistant (< 10% disease) to natural incidence of PB.

S Pande and HD Upadhyaya

Evaluation of pigeonpea mini-core collection for resistance to *Pigeonpea sterility mosaic virus* (PPSMV) under glasshouse conditions: During 2006, 120 of the 146-pigeonpea mini core accessions were evaluated under greenhouse conditions against PPSMV-P isolate at ICRISAT, Patancheru. Thirty seed of each accession was sown in plastic pots in three replications and maintained in a greenhouse at ICRISAT, Patancheru, India.

Plants were inoculated with PPSMV at 2-leaf stage with the viruliferous mites, and they were monitored for symptom type and percent incidence at 2 weekly intervals. Percent infection was estimated based on visual symptoms and the virus detection by double antibody sandwich ELISA using PPSMV-P polyclonal antibodies, at 20, 40 and 60 days post inoculation (dpi). Pigeonpea cvs. ICP8863 and ICP7035 were used as susceptible and resistant controls, which showed 100 and 0 percent infection at 60 dpi, respectively. Of 120 mini-core accessions, 9 accessions (ICP# 14976, 16264, 7869, 9045, 11910, 11015, 14801, 13579 and 15049) showed only ringspot symptoms (localized reaction, no systemic spread of the virus and no sterility) in 5 to 16% of the plants. Systemic leaves of the infected plants tested negative to virus in ELISA, indicating that virus multiplication is restricted to the inoculated leaves. Remaining accessions showed 22 to 100% infection and the plants developed severe mosaic symptoms at 60 dpi. This study indicates narrow base of resistance to PPSMV in the pigeonpea mini-core. This study is the second year evaluation of pigeonpea mini-core for PPSMV resistance. The resistant accessions provide entry point for further evaluation of genotypes for SMD resistance. All the resistant/tolerant accession will be tested against various PPSMV isolates in India.

PL Kumar, HD Upadhyaya and F Waliyar

Milestone C.5.1.3: Mini core collection of pigeonpea germplasm evaluated for resistance to Helicoverpa (HCS/HDU/KBS, 2009)

Milestone C.5.1.4: Mini core collection of groundnut evaluated for resistance to seed infection by Aspergillus flavus and aflatoxin contamination (FW/PLK/HDU, 2009)

Milestone C.5.1.5: Sorghum mini core set evaluated for resistance to grain mold and anthracnose (RPT/RS/HDU, 2010)

Milestone C.5.1.6: Pearl millet core set evaluated for resistance to downy mildew and rust (RPT/RS/HDU, 2011)

Milestone C.5.1.7: Finger millet core set evaluated for resistance to foliar and neck blast (RPT/RS/HDU/CLLG, 2010)

Milestone C.5.1.8: Foxtail core set evaluated for resistance to blast disease (RPT/RS/HDU/CLLG, 2012)

Milestone C.5.1.9: Identification and evaluation of trait specific germplasm in finger millet and foxtail millet core collections (CLLG/HDU, 2008)

Finger millet: We evaluated 20 finger millet accessions with four control cultivars in a replicated trial. IEs 2340, 3194, 3790, 4974, and 6142 (116.7 – 188.3 cm) were significantly taller than the tallest control PR 2 and can be a source for feed and fodder. IEs 2498 and 4974 (56.7 – 61.7 mm) had wider inflorescence than the widest control RAU 8 ((55.0 mm). IEs 2498, 2683, and 2983 (10.7 – 11.3) have greater width of the longest finger than the best control PR 202 (10.00 mm). IEs 2498, 2578, 2887, 2903, and 4974 (11.1 – 13.4 g 1000 seed weight) had significantly greater seed size than the large seeded control RAU 8 (8.6 g). IEs 94, 2578, 3790, 3802, 4974, and 6236 (2.01 – 2.61 t ha^{-1}) were good for grain yield.

Foxtail millet: We evaluated 20 foxtail millet accessions with four control cultivars in a replicated trial. 11 accessions (38 – 45 days) flowered significantly earlier than the earliest control ISe 375 (56 days). ISes 1258 and 1658 (38 days) were the earliest accessions. ISes 769 and 1434 (159.3 – 161.3 cm) were significantly taller than the tallest control Ise 1541 (146.7 cm). ISes 1433 and 1434 (3.0) had significantly greater number of basal tillers than all the four controls (1.0-2.0).). ISes 1433 and 1434 (234.7 – 239.3 mm) had significantly greater inflorescence length than all the four controls (131.7 – 198.7 mm). ISe 1434 (2.06 t ha^{-1})) had significantly greater grain yield than all the four controls (0.81 – 1.53 t ha^{-1}).

CLL Gowda and HD Upadhyaya

Activity C.5.2: Evaluate mini core and/or reference sets for important abiotic stresses

Milestone C.5.2.1: Groundnut, pigeonpea and chickpea mini-core sets screened for salinity tolerance (VV/LK/HDU/CLLG/PMG/RKV/KBS, 2007)

Chickpea screening: The screening that was performed in 2004-05 was repeated in 2005-06. We had a good agreement between the yield under salinity data of the two years ($R^2 = 0.40$). There was over a 6-fold range of variations in seed yield under salinity, indicating that this range of variations would be more suitable for

breeding salinity tolerant lines. From the 2 consecutive trials, the most contrasting genotypes were selected and used to develop crosses, in relation with the chickpea breeding group. To select the contrasting genotypes, we also tried to match the phenology of the parents used. We tried also to select parents with good yield potential under control conditions, knowing that yield potential explains a part of the performances under salinity. From the two years of trial, about 55 entries were dispatched to UWA (Australia) in the frame of the COGGO-funded project. Purpose is to test them for salinity in the Australian conditions. The mini-core collection of chickpea has also been sent to 2 locations in India (PAU, Ludhiana, Punjab and CSSRI, Karnal, Haryana), in collaboration with Dr Sandhu (PAU) and Dr RK Gautham (CSSRI), for field testing. Data from the 2004-05 were presented at the Australian Society of Agronomy meeting (Perth 10-14 Sept 06) and are being published in Field Crop Research.

V Vadez, L Krishnamurthy, HD Upadhyaya, CLL Gowda, PM Gaur and RK Varshney

Groundnut screening: The groundnut screening for salinity tolerance from 2005 was repeated at same time in 2006 (Sowing in April) and under the same conditions. This time, we cultivated plants under saline and control conditions until maturity and could therefore assess the yield. The range of variation for pod yield under salinity was about 6-7-fold between the most and the least tolerant genotypes. A list of tolerant and sensitive genotype from that screening is available on request. From that screening, we would be able to select the most promising for field-testing in Orissa in 2007-08. Unlike previous finding in chickpea, we found no relation between pod yield under salinity and pod yield under control, meaning that salinity tolerance for pod yield production under salinity was not related to the yield potential of groundnut. We found a modest relation between the ratio of pod yield (pod yield salinity / pod yield control) and the ratio of shoot biomass at maturity. This indicated that, although genotypes able to develop relatively more biomass under salinity were somewhat more likely to achieve a better relative pod yield, the screening for salinity tolerance is more reliable if based on the assessment of pod yield under salinity. Compare to control, the number of pod per plant was reduced by about 50%. Pod weight under salinity was further reduced under salinity, being only 30% of that under control. This result showed that not only the success of the reproductive process under salinity (proxied by the number of pods) was a key to high yield, but also the ability of plant to fill up these pods. A repeat of that screening is on going; from which a robust set of tolerant/sensitive genotypes will be identified for further testing. Crosses are also being made to develop segregating populations.

V Vadez, HD Upadhyaya, SN Nigam and RK Varshney

Pigeonpea screening: Data from the 2005 experiment were processed and analyzed. Here, we have assessed the morphological and physiological variation in pigeonpea for salinity tolerance in 300 genotypes, including the mini core collection of ICRISAT, wild accessions and land races from putatively saline prone areas worldwide. There was a large variation in the salinity susceptibility index (SSI) and the percent relative reduction (RR %) in both cultivated and wild accessions. The amount of Na^+ accumulation in shoot showed that more tolerant cultivated materials accumulated less Na in shoot (Fig 6). Such relation was not true for wild species. Wild species *C. acutifolius, C. cajanifolius* and *C. lineatus* were mostly sensitive, whereas *C. platycarpus, C. scarabaeoides* and *C. sericeus* provided good sources of tolerance. It was interesting to notice that *C. scarabaeoides* also provided a large range of sensitive materials. The minicore collection of pigeonpea provided a large range of variation for salinity tolerance. Among the tolerant genotypes, there were a large number of tolerant accessions originating from Bangladesh. A repeat of the pigeonpea screening done in 2005 was performed. Unfortunately, the trial failed because of soil quality used for that experiment (we noticed too late the poor fertility of the soil lot that was allotted to us and saline treated plant failed to make it to harvest).

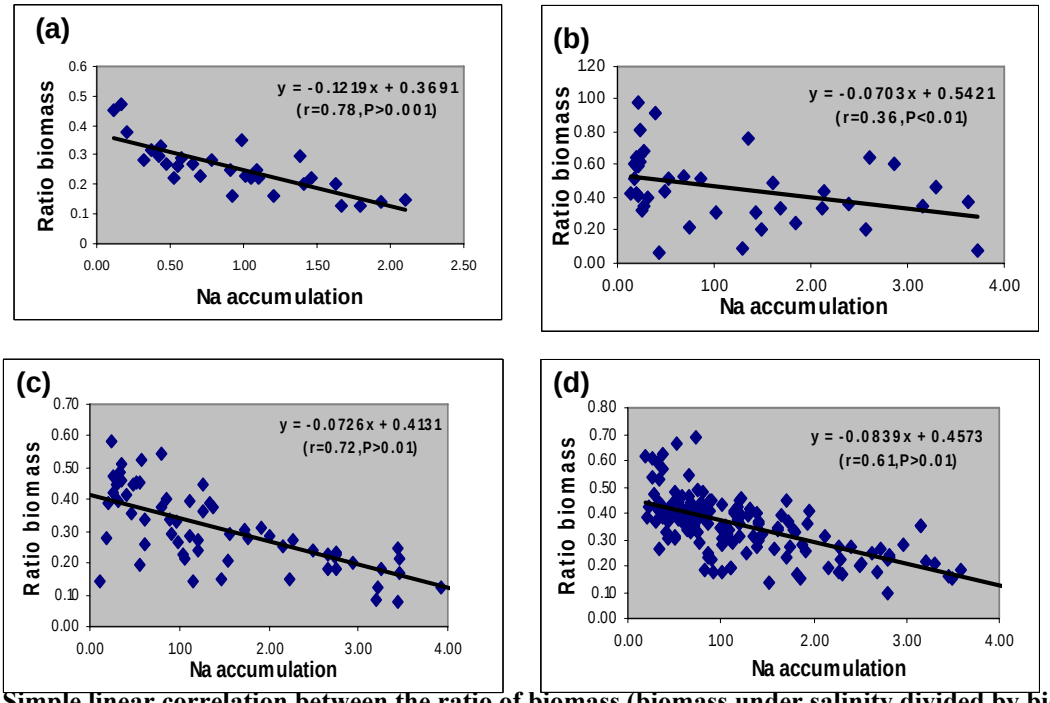


Fig 6. Simple linear correlation between the ratio of biomass (biomass under salinity divided by biomass under control) and Na^+ accumulation in shoot: (a) with a treatment of 1.34 g NaCl kg^{-1} soil in six genotypes of different maturity group, (b) in wild species (c) in selected landraces from saline areas (d) in the minicore collection. Data are the mean of 5, 3, 3, and 3 replicated data of each genotype, for (a), (b), (c) and (d) respectively

V Vadez, HD Upadhyaya, KB Saxena, CLL Gowda and RK Varshney

Milestone C.5.2.2: Chickpea mini core salinity evaluation data analyzed (VV/LK/HDU, 2008)

Salinity is an ever-increasing problem in agriculture worldwide, especially in South Asia (India, Pakistan) and Australia. A screening of 263 accessions of chickpea, including 211 accessions from ICRISAT's mini-core collection (10% of the core collection and 1% of the entire collection), was performed and showed, overall, that the large variation in salinity tolerance in chickpea was explained by differences in sensitivity at reproductive stages. Data showed a six-fold range of variation for seed yield under salinity, with several genotypes yielding 20% more than a previously released salinity tolerant cultivar. The range of variation in yields under salinity was similar in both kabuli and desi chickpeas, indicating that breeding for salinity tolerance can be undertaken in both groups. Among the genotypes evaluated, desi genotypes had higher salinity tolerance than kabuli genotypes. A strong relationship was found between the seed yield under salinity and the seed yield under a non-saline control treatment, indicating that the seed yield under salinity was explained in part by a yield potential component and in part by salinity tolerance *per se*. Seed yields under salinity were therefore computed to separate the yield potential component from the residuals that accounted for salinity tolerance *per se*. The residuals were highly correlated to the ratio of seed yield under salinity to that of the control, indicating that both parameters can be used to assess salinity tolerance. A similar ratio was calculated for shoot dry weight at 50 days after sowing. However, no significant correlation was found between the shoot dry weight ratio and the yield ratio, indicating that differences in salinity tolerance among genotypes could not be inferred from measurements in the vegetative stage. The major trait related to salinity tolerance was the ability to maintain a large number of filled pods, whereas seed size was similar in tolerant and sensitive genotypes. Salinity tolerance was also not related to the Na^+ or K^+ concentrations in the shoot. A journal article has been submitted to the field crops Research.

V Vadez, L Krishnamurthy and HD Upadhyaya

Milestone C.5.2.3: Sorghum mini core and pearl millet reference set screened for salinity tolerance (VV/LK/HDU/CTH/KNR, 2008)

First screening of a part of the mini-core collection of sorghum has been initiated (October 2006) and data will be available in 2007. The full collection should be screened in 2007-08.

V Vadez, L Krishnamurthy, HD Upadhyaya, CT Hash and KN Rai

Milestone C.5.2.4: Reference sets of chickpea, pigeonpea, and groundnut evaluated for salinity (VV/LK/HDU/RKV, 2010)

Activity C.5.3: Evaluate groundnut and sorghum mini core and/or reference sets for transpiration efficiency (TE) and root traits

Milestone C.5.3.1: Groundnut mini-core set screened for TE (VV/HDU/RKV/CLLG, 2008)

We have screened 440 groundnut genotypes for TE, under progressive soil drying conditions, during the Feb-April 2006 period. These 440 genotypes included the mini core collection of groundnut (184 accessions), other germplasm lines, control cultivars, and elite breeding lines. The data show an impressive range of variation between the top and the bottom ranked groundnut genotypes for transpiration efficiency (TE, g kg^{-1} water transpired). In that experiment, TE varied between about 0.3 g kg^{-1} water transpired and 3.6 g kg^{-1} water transpired. About 90% of the genotypes (excluding the bottom 40 and top 10 in the ranking of TE) were in a range of TE between 0.5 and 2.5 g kg^{-1} water transpired, i.e. still a 5-fold range of variation. Ten genotypes had TE higher than 2.5 g kg^{-1} water transpired and one genotype reached 3.6 g kg^{-1} water transpired. A repeat of that experiment will be carried out in 2007. TE was also assessed under well-watered (WW) conditions in the mini core collection of groundnut, plus with controls (200 accessions). It has been reported in the literature that there is a good relation between TE measured under water stress and TE measured under well-watered conditions, from which it has been inferred that mesophyll efficiency differences were the major reason for differences in TE. However, this relation has been worked out with a very narrow range of genotypes and fully overlooks the potential role of stomatal regulation in the determination of TE differences. Relation between TE under water stress (WS) and under well-watered conditions with a larger set of genotype was also assessed. TE was overall higher under WW conditions (1.92 g kg^{-1} water transpired) than under WS (1.44 g kg^{-1} water transpired). We found significant relation between TE under water stress and TE under well-watered conditions. However, the correlation coefficient was 0.32 only, indicating that it would be advisable to consider TE under WS and WW conditions separately. Analysis needs to be done to test the significance of genotype x moisture interaction.

V Vadez, L Krishnamurthy, HD Upadhyaya, RK Varshney, and, CL.L Gowda

Milestone C.5.3.2: Sorghum mini-core (or reference) set screened for TE (VV/HDU/CTH/BVSR, 2009)

Milestone C.5.3.3: Groundnut mini-core screened for root traits (VV/HDU/RKV/CLLG, 2010)

The protocol to screen groundnut for root traits is being worked out with a small set of genotypes to optimize the cylinder system that will be used. Screening for root traits in groundnut will be based both on transpiration values (using a lysimetric system) and on root parameters (max depth, length density at different depth).

V Vadez, HD Upadhyaya, RK Varshney and CLL Gowda

Milestone C.5.3.4: Sorghum mini-core (or reference set) screened for root traits (VV/HDU/CTH/BVSR, 2011)

The protocol to assess root traits in sorghum is also being worked out. Cylinders currently used to assess root traits in sorghum are 16 cm in diameters and very likely 25 cm diameter cylinder will be used. As for groundnut (Milestone C.5.3.3), the assessment would be a combination of lysimetric measurements and root characteristics.

V Vadez, HD Upadhyaya, CT Hash and BVS Reddy

Milestone C.5.3.5: C ¹³ in chickpea analyzed at JIRCAS (JK/HDU/LK/PMG/IIPR-Kanpur/JIRCAS-Japan, Annual)

Analysis of $\delta^{13}C$ was performed in Japan International Research Center for Agricultural Sciences (JIRCAS), Tsukuba, Japan with use of an isotope ratio mass spectrometer (IRMS), ThermoFinnigan Delta XP^{plus}, Hamburg, Germany, connected with an element analyzer, Carlo Erba EA Flash 1112, Milan, Italy. Total carbon in leaf samples were incinerated in a furnace of EA and separated as to be pure CO_2 gas. A small quantity of the gas was introduced to IRMS to measure the ratio of $^{13}CO_2/^{12}CO_2$ as the different mass weight of 45/44 to obtain $\delta^{13}C$ (‰). There was a significant difference in $\delta^{13}C$ among the 10 chickpea genotypes, and the $\delta^{13}C$ in stress condition was a significantly higher than it in the control. A genotype of ICC 5337 showed the highest $\delta^{13}C$ (-26.0‰) in the stress condition. ICC 4958, which is well known as drought resistance variety, showed superior

$\delta^{13}C$ than the other genotypes at the 2nd (-27.2‰) and highest (-28.4‰) in the stress and control condition, respectively.

J Kashiwagi, HD Upadhyaya, L Krishnamurthy, PM Gaur,
IIPR-Kanpur and JIRCAS-Japan

Milestone C.5.3.6: Ten chickpea lines identified, which showed steady high water use efficiency (WUE) as well as high yielding in two locations (JK/HDU/LK/PMG/IIPR-Kanpur/JIRCAS-Japan, 2009)

The genotype by irrigation (G x I) interaction was significant, which indicates that the each genotype had different reaction on $\delta^{13}C$ between the well watered and drought environments. A significant positive correlation between $\delta^{13}C$ and TE was observed ($r = 0.857$, $p<0.01$) under the stress condition. This relationship agrees with the theoretical relationship between $\delta^{13}C$ and TE as observed in several other legumes. However, no significant correlation was observed between them when the plants were grown under well watered condition. This would indicate that the ^{13}C discrimination is occurred in the chickpea when they have been subjected to soil moisture stress.

J Kashiwagi, HD Upadhyaya, L Krishnamurthy, PM Gaur,
IIPR-Kanpur and JIRCAS-Japan

Activity C.5.4: Investigation of genetic diversity of chickpea and groundnut reference sets and assessing its relevance with drought avoidance root traits

Milestone C.5.4.1: Chickpea reference set phenotyped for root traits in PVC cylinders (120cm height) (JK/HDU/LK/RKV/NL, 2008)

Milestone C.5.4.2: Chickpea reference set field evaluated for drought response (JK/LK/ HDU/NL, 2008)

Milestone C.5.4.3: Chickpea reference set genotyped with 100 SSR markers (HDU/NL/RKV/JK/LK/SC, 2008)

A set of more than 300 (TA, TS, TR, TAA, GA, GAA, STMS, AGL series) SSR markers, well distributed over the chickpea genome have been screened initially on two diverse genotypes of chickpea (Annigeri, ICCV2). Subsequently, sets of polymorphic and high quality markers were identified. In order to improve the efficiency of genotyping multiplexes for all markers are being optimized

The reference collection consisting of 300 accessions has planted in field on 1st November 2006 with 20 seeds per accession. The wild accessions were first sown in incubator and transplanted in the field. DNA was extracted at 22nd day from leaf tissue by using high – through put method and quality checked by using agarose gel electrophoresis. Quantification of DNA concentration has been done with fluorescence detector and diluted to 5 ng/ul as working sample.

The genotyping of reference collections of chickpea accessions will be started with optimized multiplexes by using Genetic Analyzer (ABI 3100).

HD Upadhyaya, N Lalitha, RK. Varshney, J Kashiwagi,
L Krishnamurthy and S Chandra

Milestone C.5.4.4: Groundnut reference set phenotyped for traits associated with drought resistance (VV/HDU/RKV, 2009)

Milestone C.5.4.5: Groundnut reference set genotyped with 100 SSR markers (HDU/RKV/DH, 2010)

Milestone C.5.4.6: Reference set of chickpea (300 accessions) utilized for candidate gene diversity for mining the drought tolerant alleles (RKV/HDU/JK/DH/PMG, 2010)

Under Allelic Diversity on Orthologous Candidate genes (ADOC), identification of drought responsive genes is underway. Currently, DREB1a and DREB2a genes are being identified in chickpea by using *in silico* analysis (see Output Target D.3).

RK Varshney, HD Upadhyaya, B Jayashree and DA Hoisington

Milestone C.5.4.7: Diversity analyzed for the molecular markers and markers associated with root traits identified (HDU/JK/RKV/LK/SC/NL, 2011)

Milestone C.5.4.8: Diversity analyzed for the molecular markers and markers associated with drought traits identified (HDU/RKV/DH, 2012)

Output target C.6: Germplasm sets evaluated for utilization in Africa (2009)

Activity C.6.1: Evaluation (field test) of the global pearl millet core set at Sadore, Niger

Milestone C.6.1.1: M Sc thesis on evaluation of the pearl millet core set at Sadore, Niger, completed and data available in Excel format (BH/HDU, 2007)

The global pearl millet core collection was grown in a replicated trial at the ICRISAT Sahelian Center near Niamey, Niger, in the Rainy Season 2006. Ms Jenny Coral Padilla, MSc student from University of Hohenheim, Stuttgart, Germany, was involved in the characterization. Data analysis and MSc thesis write-up are underway.

BIG Haussmann and HD Upadhyaya

Activity C.6.2: Characterize core collection of finger millet and identify materials for regional evaluation

Milestone C.6.2.1: Core collection of finger millet characterized for morpho-agronomic and end use traits (MAM/SGM/HDU, 2007)

Finger Millet: 506 finger millet germplasm accessions of core collection from India were planted and characterized at Kiboko (2005/06) and Alupe (April-Sept 2006). Preliminary principal component analysis results from Kiboko based on 15 quantitative traits showed 57% of the variability in the germplasm accounted for by the first two principal components (PCs). High positive loadings on PC1 were contributed by days to 50% flowering, plant heights, leaves per plant, leaf lengths, leaf widths, neck lengths, peduncle length, stem diameter and high negative loadings from heads per plant, number of nodal tillers, and number of productive tillers. Though generally 6 cluster groups were observed, 3 distinct clusters were evident with no distinct pattern of grouping based on country of origin as accessions from different countries were found in almost all clusters. Further analysis including all traits (both qualitative and quantitative) from Kiboko and Alupe will done and reported fully in the 2007 archival report

MA Mgonja, E Manyasa, E Muange and HD Upadhyaya

Milestone C.6.2.2: Promising and adaptable materials identified and distributed and evaluated in regional finger millet trials the ESA (MAM/SGM, 2008)

Activity C.6.3: Characterize a sorghum core collection from five African Bio-fortified Sorghum target for diversity in micro nutritional traits

Milestone C.6.3.1: Diversity for micronutrients contents in a sorghum core collection from at least 2 ABS target countries established (MAM/SGM/HDU, 2008)

The African Bio-fortified Sorghum Project intends to develop transgenic sorghum varieties that will deliver essential amino acids; lysine, threonine, methionine and tryptophan; vitamins A and E; iron; and zinc which are deficient in sorghum to African populations in the arid and semi-arid tropics. Field work has been initiated to document diversity of the above traits in sorghum germplasm and close gaps in sorghum germplasm from the at least two ESA countries. In 2006, using GIS we mapped sorghum collection sites in order to identify collection gaps. Further we examined ABS target countries in order to identify the major collection gaps within those countries targeting planned collection missions. We used data available at ICRISAT genebank and in addition we sought for information on collections that have been done by the national gene banks to update the database. The National Gene bank of Kenya provided us with 994 data sets while the Genetic Resources Unit of the National Department of Agriculture of South Africa provided us with 257 data sets. A core collection of sorghum from 5 ABS target countries has been formed and it contains 426 accessions. This collection was planted at Kiboko, Kenya in November 2006 for agronomic characterization. Additional accessions that are known to have great variation in the ABS traits of interest were included for characterization. The National Gene bank of Kenya, KARI and ICRISAT carried out a joint collection mission in July-August 2006 and a total of 154 accessions were collected from Western, Rift Valley, Eastern and Coastal provinces of Kenya. The

collections have been incorporated in the GIS map of Kenya and have been planted for morphological characterization at KARI-Embu. Saturated GIS maps for Kenya sorghum collections are therefore available

MA Mgonja, SG Mwangi and HD Upadhyaya

Activity C.6.4: Evaluate groundnut mini core collection/wild species in ESA

Milestone C.6.4.1: Mini core collection of groundnut evaluated for agronomic traits at different locations in ESA (ESM/HDU 2008)

Milestone C.6.4.2: Wild Arachis evaluated for target traits (GRD, ELS, aflatoxin resistance) at hotspot locations in ESA (ESM/HDU etc. 2009)

Milestone C.6.4.3: Gene introgression carried out for foliar and viral disease resistance from wild Arachis germplasm into cultivated varieties (ESM/HDU etc. 2010)

Output target C.7: Trait specific germplasm of staple crops and small millets available for utilization (2009)

Activity C.7.1: Ensure availability of germplasm accessions for selected traits of staple crops and small millets to partners

Milestone C.7.1.1: Trait specific germplasm regenerated/multiplied for distribution to partners on request (HDU/CLLG/RPT-PQL/NBPGR, Annual)

Groundnut: Multiplied the seed of 21 early maturing, 18 drought tolerant and 60 high yielding combined with other traits of economic importance groundnut accessions for distribution to partners.

Chickpea: Multiplied the seed of 28 early maturing and 18 droughts tolerant, 16 large-seeded kabuli, 39 high yielding combined with other traits of economic importance, 29 salinity tolerant chickpea accessions for distribution to partners.

Mini core: Multiplied the seed of chickpea, pigeonpea, and groundnut mini core collections for distribution to partners.

HD Upadhyaya, CLL Gowda, RP Thakur and NBPGR

Output target C.8: Germplasm reference collections available for utilization (2009)

Activity C.8.1: Ensure availability of reference collections of staple crops and small millets to partners

Milestone C.8.1.1: Germplasm accessions of reference collections regenerated/multiplied for distribution to partners on request (HDU/RPT/CLLG/NBPGR, Annual)

Output target C.9: Broadening the genetic base of legumes through wide crosses (2011)

Activity C.9.1: Broadening the genetic base of groundnut by creating tetraploid groundnut using wild *Arachis*, synthetic amphidiploids and/or other diverse germplasm.

Milestone C.9.1.1: Hybrids between A and B genome species made available (NM/HDU/DH, 2007)

We produced 18 F_1 diploid hybrids between different accessions of five A genome and five B genome Arachis species. Some of the hybrids did not set seeds in spite of profuse peg formation and only five hybrids produced seeds (F_2 seeds). Attempts are being made to obtain seeds from other crosses too.

N Mallikarjuna, HD Upadhyaya and DA Hoisington

Milestone C.9.1.2: Tetraploid hybrids between different genomes generated and skeletal map constructed (NM/RKV/HDU/DH, 2008)

Milestone C.9.1.3: Hybrids between cultivated groundnut and synthetic amphidiploids created, variation for different traits analyzed and molecular map constructed (NM/RKV/HDU/DH/FW/PLK, 2009)

Milestone C.9.1.4: Develop hybrids between section Arachis and section Procumbentes and generate fertile backcross population and screen for desirable traits (NM/DH/HDU/FW/PLK/EM, 2009)

F_2 plants from *A. hypogaea* x *A. chiquitana* ICG 11560 (section Procumbentes) and *A. hypogaea* x *A. kretschmeri* IG 8191 (section Procumbentes) crosses were used to produce back cross progenies with the cultivated species. Most of the pods on progenies were single seeded. Efforts will be directed to generate large numbers of seeds.

N Mallikarjuna, DA Hoisington and HD Upadhyaya

Milestone C.9.1.5: Tetraploid molecular map available for use in breeding program (NM/RV/HDU/DH/FW/PLK/CLLG, 2010)

Activity C.9.2: Broaden the genetic base of pigeonpea using *Cajanus platycarpus*, a tertiary gene pool species of *Cajanus*

Milestones C.9.2.1: Generate fertile hybrids between Cajanus platycarpus and C. cajan (NM/DH/HDU, 2007)

BC_3 plants (*Cajanus platycarpus* x *C. cajan*) are being crossed with recurrent cultivated parent in the glasshouse, as BC_3 plants do not set seeds from self-pollinations. Twenty three lines of fertile backcross progeny (BC_4) between *C. platycarpus* x *C. cajan* were generated and planted in the field for seed increase. BC_4 have set seeds from self-pollinations.

N Malikarjuna, DA Hoisington and HD Upadhyaya

Milestone C.9.2.2: Generate variation for desirable characters using Cajanus platycarpus (NM/HDU/RKV/DH/KBS, 2009)

BC_4 plants from the cross *Cajanus platycarpus* x *C. cajan* were screened for variability under field conditions and variability was observed for plant type, number of secondary branches, vegetable type of pod characteristics, days to flowering, profuse pod set, male sterility, and plant height. The experiment will be repeated to verify if these characters are heritable.

N Malikarjuna, HD Upadhyaya, RK Varshney and DA Hoisington

Output target C.10: Data management infrastructure development (2010)

Activity C.10.1: Data capture instruments expanded and functionality increased.

Milestone C.10.1.1: Beta testing of the Laboratory information management system and improvement (JB/DAH/SS/RKV/DK/SV, 2007).

The beta testing of the LIMS system is in progress at ICRISAT. The respective data producers have uploaded genotyping data for the composite core collections of chickpea and sorghum into the system. The application functionality and user interfaces continue to be modified to suit user requirements. The LIMS application has also been extended and adapted for use at ICRISAT/IITA/ILRI - Nairobi and IITA-Ibadan. A workshop was conducted at ICRISAT-Nairobi to demonstrate the software to potential users and obtain feedback. The software has also been shared with several universities and private partners under the terms of the GNU general public license.

B Jayashree, DA Hoisington, S Senthilvel, RK Varshney, DKiambi and S de Villiers

Activity C.10.2: Integrated database development with web interfaces and interoperability requirements.

Milestone C.10.2.1: Development of database and middleware with GUI (JB/DH/ and others, 2007)

The LIMS –ICRIS adaptor allows the flow of genotyping data from the LIMS application into the ICRIS database. The database is expected to integrate genotyping information with genetic resource and phenotype information. Progress was made this year in coding for generic middleware to this database to bring it in compliance with the GCP(generation challenge program) platform. This will allow for the database to become interoperable with other databases and repositories available in the public domain. A GCP platform compliant

data source API (application program interface) is also being developed that will allow data consumers such as publicly available analytical or visualization tools to access data within the database.

B Jayashree and DA Hoisington

Milestone C.10.2.2: Alpha and beta testing of database by users (CTH/SS/RKV/HDU/RB/VV/and others)

Milestone C.10.2.3: Curation of data with involvement of data providers.

Output target C.11: Development of data analysis tools

Activity C.11.1: iMAS a decision support system for marker aided breeding.

Milestone C.11.1.1: Further development and testing of application (SC/DH/JB, 2007).

The goal of this two-year project (2005-06) was to develop an integrated decision support system, called **iMAS**, to seamlessly facilitate marker-assisted plant breeding by integrating freely available quality software involved in the journey from phenotyping-and-genotyping of genetic entities to the identification and application of trait-linked markers, and providing simple-to-understand-and-use online decision guidelines to correctly use these software, interpret and use their outputs. The project was structured into nine activities, namely: **A1**: Analyze potentially useful free software, **A2**: Select software for inclusion in iMAS, **A3**: Develop iMAS system, **A4**: Develop & incorporate online decision guidelines, **A5**: Test iMAS system, **A6**: Refine iMAS system, **A7**: Develop iMAS user manual/tutorial, **A8**: Release of and Training in iMAS, **A9**: Consultation and support.

Five different software have been integrated in this pipeline along with online decision support and the integration of two more software is in progress in 2006. Integrated software include IRRISTAT, Gmendel, PlabQTL, POPMIN and GGT while the integration of WinQTLCartographer and TASSEL is in progress. The pipeline has been modified to incorporate user comments, two major workshops were conducted accommodating over 40 scientists from the NARS besides ICRISAT staff and interested private partners to test the application. Besides, project partners reviewed the application and their suggestions are being incorporated.

S Chandra, DA Hoisington and B Jayashree

Activity C.11.2: High performance computing toolbox.

Milestone C.11.2.1: Extension of existing tools with user friendly interfaces (JB/RKV/DH/SC and others).

The pipelines and standalone software available within the comparative genomics and population genetics toolboxes on the high performance computer have been extended through 2006. The comparative genomics toolbox now includes a suite of parallelized programs for marker mining and detection from large public datasets and open source software for comparative sequence analysis and phylogeny. The population genetics toolkit includes a parallelized version of the program 'structure' with user interfaces and visualization software along with format conversion tools for a variety of popularly used analysis software.

B Jayashree, RK Varshney, DA Hoisington, AG Sylvester,
S Chandra, MS Hanspal,and VT Jagdesh

Activity C.11.3: Comparative genomics tools to aid marker development. (JB/SS/CTH/RKV and others)

Milestone C. 11.3.1: Development of appropriate software pipelines for mining of markers from public data (2008, Annual)

Pipelines for the SNP, from public EST data are now available on HPC (http://hpc.icrisat.cgiar.org/pbsweb).

Output D: RILs of staple crops and small millets developed/assembled and DNA extracts conserved and distributed

MTP Output Targets 2006
Genetic diversity of chickpea composite collection determined

RILs of groundnut and chickpea (WUE, diseases) assembled

Output target D.1: RILs of staple crops assembled (2009)

Activity D.1.1: Assemble RILs of staple crops

Milestone D.1.1.1: RILs of chickpea (root traits, resistance to Helicoverpa, fusarium wilt, BGM, and salinity tolerance) assembled (PMG/HDU/CLLG, 2006)

ICRISAT has developed and is maintaining three RIL populations (Annigeri × ICC 4958, ICCV 4958 × ICC 1882, ICCV 283 × ICC 8261) for root traits, two for fusarium wilt resistance (ICCV 2 × JG 62, WR 315 × C 104) and one for *Helicoverpa* resistance (ICC 505 EB x Vijay). We assembled three RIL populations for root traits and fusarium wilt resistance from other institutes during 2006. These include two intraspecicfic RIL populations, JG 62 x ICC 4958 (fusarium wilt resistance and root traits) and Vijay x ICC 4958 (root traits) from National Chemical Laboratory, Pune, India and one interspecific RIL population, *Cicer arietinum* (ICC 4958) *x C. reticulatum* (PI 489777) (root traits) from Washington State University, Pullman, USA. The seed of these RILs are being multiplied during the 2006/07 crop season and would be submitted to the genebank.

Milestone D.1.1.2: RILs of groundnut (WUE - 4, rust – 2, and LLS – 2) assembled (SNN/HDU, 2006)

Seed of two RIL populations (ICGV 99001 x TMV 2 and ICGV99004 x TMV 2) for late leaf spot and other two populations (ICGV 99003 x TMV 2 and ICGV99005 x TMV 2) for rust assembled in ICRISAT genebank.

Milestone D.1.1.3: RILs of pigeonpea (one population of wilt resistance) assembled (KBS/HDU, 2008)

Milestone D.1.1.4: RILs of pearl millet (3 populations) assembled (CTH/HDU, 2008)

Milestone D.1.1.5: RILs of sorghum (grain mold – 2, stem borer –3, and shoot fly -2) assembled (BVSR/HCS, 2008)

Milestone D.1.1.6: TILLING population of pearl millet developed (RKV/CTH/DH, 2009)

To generate the TILLING population in pearl millet, the inbred line "P1449-2-P1" has been chosen, as it is one of the parental genotype of a mapping population (PT 732B x P1449-2-P1) developed and maintained at ICRISAT and segregates for plant height (*d2*), downy mildew resistance and stover grain quality. It is the tall parent, witch serves as a good source of downy mildew resistance for the improvement of local cultivars. The chemical mutagen ethyl methane sulfonate (EMS) was selected to develop the TILLING population, as it generates mostly single nucleotide polymorphisms or SNPs (in genes) and can be controlled to produce a high density of point mutations causing a variety of lesions including nonsense and missense mutations. After treating the seeds of the selected pearl millet inbred line with several concentrations (5 mM, 10 mM, 15 mM, 20 mM, 25 mM, 30 mM, 35 mM, 40 mM, 45 mM, 50 mM, 55 mM and 60mM), recommended in literature our results suggested that all the mutagen concentrations above 5 mM are lethal to the pearl millet genome. Subsequently a lower range of concentration of mutagen (1 mM, 2 mM, 3 mM, 4 mM, 5 mM, 6 mM, 7 mM, 8 mM, 9 mM, and 10 mM) for different treatment time (4 hrs, 8 hrs, 12 hrs) was used. As a result, finally about 2000 seeds each were treated with three mutagen concentrations i.e. 5 mM, 9 mM and 10 mM (each with 4 hrs) that should provide 60% 50% and 40% plants survived. The treated seeds were sown in 12" diameter pots and after about two weeks time, these plants were transplanted into field. In fields all plants were selfed and finally a total of 2,581 M1 lines have been harvested. These include 1169 lines from 5 mM, 737 from 9 mM and 675 from 10 mM. These lines will be advanced to M2 generation and another set of M1 lines will be generated.

RK Varshney, T Mahender, CT Hash and DA Hoisington

Activity D.1.2. Develop suitable contrasting parental lines for salinity tolerance in staple crops

Milestone D.1.2.1: Suitable contrasting parental lines for salinity tolerance in chickpea for the development of RILs provided (VV/RKV/HDU/PMG, 2007)

A set of contrasting genotypes for salinity tolerance identified and three crosses have been performed:

ICC 6263 (sensitive, DF 70) x ICC 1431 (tolerant, DF 69)
ICC 15802 (sensitive, DF 66) x ICC 9942 (tolerant, DF 63)
ICCV 2 (sensitive, DF 39) x JG 11 (tolerant, DF 40)

Parents used for these 3 crosses have been chosen based on the similarity in phenology (DF = days to flowering), since it was found that the number of days to flowering has some influence on the level of salinity tolerance in the conditions where we assess the materials. Diversity analysis using SSR markers is in progress.

V Vadez, RK Varshney, HD Upadhyaya and PM Gaur

Milestone D.1.2.2: Suitable contrasting parental lines for salinity tolerance in groundnut for the development of RILs provided (VV/SNN/AR/RKV/HDU, 2008)

A group of genotypes showing good contrast for salinity tolerance has been identified for use in groundnut breeding. The choice of tolerant and sensitive materials has been based on a high and low pod yield under saline conditions, with all genotypes obtaining a good yield under control conditions.

V Vadez, SN Nigam, R.Aruna, RK Varshney and HD Upadhyaya

Activity D.1.3: Assemble and make available for distribution the existent RILs of staple crops and small millets that are in the public domain in seed and DNA form

Milestone D.1.3.1: Seed multiplied for RIL populations for different crops (HDU/DH/CLLG/CTH/RKV/SS/RB/SNN/AR/PMG/KPS/KNR/BVSR, and others, 2009)

Milestone D.1.3.2: DNA of different RIL populations isolated (HDU/DH/CLLG/CTH/RKV/SS/RB/SNN/AR/PMG/KPS/KNR/BVSR, and others, 2010)

Milestone D.1.3.3: Marker and phenotype databases for the available RIL mapping populations curated (HDU/JB/DH/CTH/CLLG/RKV/SS/RB/SNN/AR/PMG/KPS/KNR/ BVSR, and others, 2010)

Output target D.2: DNA extracts of sub sets of germplasm conserved for utilization (2011)

Activity D.2.1: Conserve DNA extracts of sub sets of germplasm for utilization

Milestone D.2.1.1: DNA extracts of mini core and reference sets of chickpea conserved (HDU/RKV/DH/CLLG, 2008)

DNA was extracted from the chickpea reference collection consisting of the mini core collection.

Milestone D.2.1.2: DNA extracts of mini core and reference sets of groundnut and sorghum conserved (HDU/RKV/CTH/DH/CLLG, 2009)

Milestone D.2.1.3: DNA extracts of mini core set of pigeonpea conserved (HDU/RKV/CTH/DH/CLLG/KBS, 2010)

Milestone D.2.1.4: DNA extracts of mini core of finger millet and reference sets of pigeonpea, pearl millet and finger millet conserved (HDU/RKV/CTH/DH/CLLG/KBS, 2011)

Milestone D.2.1.5: Requested DNA samples of specific accessions distributed for utilization (HDU/RPT/RKV/DH/NBPGR, Annual)

Output target D.3: Allele specific sequence diversity in the reference sets staple crops studied (2011)

Activity D.3.1: Study allele specific sequence diversity in the reference sets of staple crops

Milestone D.3.1.1: Allele specific sequence diversity in the reference set of chickpea studied (RKV/HDU/DH, 2010)

To identify the candidate genes for analyzing the sequence diversity, bioinformatics analyses in the first instance, a total of 20 DREB1A and 34 DREB2A non-redundant protein sequences were obtained through an in depth analysis of databases. The output alignment files from both ClustalW and MUSCLE were used for phylogenetic analysis and consensus primer design. The primers could be designed by CODEHOP for five blocks (A, B, C, D and E) in case of DREB1A and only for one block (A) in case of DREB2A. A total of 22 (9 forward and 13 reverse) primers for DREB1A and 13 (5 forward and 8 reverse) primers for DREB2A were designed and synthesized. All possible combinations (90 combinations in DREB1A and 40 combinations of

DREB2A) were checked for amplification using 55-50^{0}C touch down PCR profile. In terms of getting homologous sequences in both species, single 'putative' amplicon was obtained with only 5 primer combinations; however, all these amplicons were smaller (<300 bp) in size. Therefore, good sequence quality data could not be obtained for any species by using these 5 primer combinations.

Besides degenerate primers, species-specific primers were designed using sequence information available in the public data domain. These species included *Oryza sativa*, *Arabidopsis thaliana*, *Medicago truncatula*, *Glycine max, Zea mays, Sorghum bicolor and Hordeum.* The specific primers for rice and soybean were designed according to the publications of Dubouzet et al. (2003) and Li et al. (2005) respectively.

The specific primers for *Arabidopsis*, Medicago*, Zea, Sorghum* and *Hordeum* were designed using PRIMER3 programme. However, the prominent fragments, to be used in sequenced, could not be obtained for any primer combination. Efforts are underway to identify the candidate DREB genes by using some other strategies.

RK Varshney, S Nayak, B.Jayashree,
HD Upadhyaya and Dave Hoisington

Milestone D.3.1.2: Allele specific sequence diversity in the reference set of sorghum studied (CTH/RKV/HDU/DH, 2011)

Use of above mentioned species-specific primers provided 'putative' DREB homologs in sorghum with groundnut DREB primer pairs (GmDREBb). Sequencing data of the prominent amplicons in seven diverse sorghum genotype however could not be matched with the known DREB genes with any species. Efforts are underway to identify the candidate DREB genes by using some other strategies.

RK Varshney, S Nayak, B Jayashree, HD Upadhyaya,
CT Hash and DA Hoisington

Output target D.4: Development of genomic resources for SAT crops (2011)

Activity D.4.1: Development of molecular markers

Milestone D.4.1.1: Novel set of microsatellite markers developed and characterized for chickpea, pigeonpea and groundnut (RKV/DAH, 2009)

A microsatellite or SSR (simple sequence repeat) enriched genomic DNA library of chickpea (ICC4958) has been constructed using pGEM-3Zf (+) vector in collaboration with the University of Frankfurt, Germany (Dr Peter Winter). The library has been enriched for (GA)n and (TAA)n microsatellites and a total of 359 clones have been collected. The plasmid DNA, however could be isolated for about 300 clones. These clones were sequenced for both strands, using T7 promater primer and SP6 primer of the pGEM-3Zf (+) vector. Analysis of sequence data with the *MI*cros*SA*tellite (MISA) search module provided about 220 clones containing at least one SSR. Based on the conserved flanking regions of the SSRs, the primer pairs are being designed and optimized.

RK Varshney, P Winter and DA Hoisington

In the case of pigeonpea, efforts have been initiated to generate the SSR enriched library from the pigeonpea variety "Asha". The genomic DNA library is being enriched for (CT)n and (TCG)n SSRs.

RK Varshney, C Prathima and DA Hoisington

Milestone D.4.1.2: Novel set of microsatellite markers developed and characterized for pearl millet (RKV/CTH/SS/DAH, 2008)

Development of microsatellite markers for pearl millet is in progress in collaboration with Centre for Cellular and Molecular Biology (CCMB), Hyderabad (Dr Ramesh Aggarwal). By using an SSR enrichment method, developed at CCMB, >1000 genomic DNA clones enriched for SSRs have been developed. The isolation of plasmid DNA and sequencing is in progress. Sequencing of about 120 clones has already been completed and about 80 clones containing at least one SSR have been identified. The primer pairs have been designed for 64 SSRs based on the conserved flanking regions and are being optimized for amplification of SSR loci in pearl millet.

RK Varshney, R Aggarwal, T Mahender and DA Hoisington

Activity D.4.2: Development of molecular genetic maps

Milestone D.4.2.1: Molecular genetic maps and consensus maps based on SSRs, DArTs and EST-based markers developed for chickpea, pigeonpea and groundnut (RKV/DAH/PMG/KBS/SNN/HDU, 2010)

For developing the genetic maps in chickpea, currently three recombinant inbred line (RIL) mapping populations i.e. one interspecific (*C. arietinum* ICC4958 x *C. reticulatum* PI 489777) and two intraspecific (ICC4958 x ICC1882; ICC283 x ICC8261) are being used with newly developed SSR or existing SSR markers for identification of the polymorphic markers. The polymorphic markers will be used for genotyping the respective mapping populations.

RK Varshney, PM Gaur, J Kashiwagi and DA Hoisington

In pigeonpea, an interspecific F_2 population developed after crossing *C. cajan* (ICP28) x *C. scarabaeoides* (ICPW94) has been screened with the SSR markers developed at ICRISAT. About 30 polymorphic SSR markers have been identified between the parental genotypes of the mapping population. These genotypes are being screened with DArT (Diversity Array Technology) markers in collaboration with Dr Andrzej Killian (Australia) for identification of larger number of polymorphic loci.

RK Varshney, HD Upadhyaya and DA Hoisington

The RIL mapping population of groundnut (ICGV86031 x TAG24) is being used for developing the genetic map for cultivated groundnut. All the existing SSR markers for groundnut as well as unpublished markers from some research groups e.g. David Bertioli (Catholic University/EMBRAPA, Brazil) and Steve Knapp (University of Georgia, USA) have been screened on the parental genotypes of the mapping population. However, so far about 120 polymorphic SSR markers. These polymorphic markers are being used for genotyping of the mapping population.

RK Varshney, V Vadez, SN Nigam, R Aruna and DA Hoisington

Milestone D.4.2.2: Molecular genetic maps and consensus maps based on SSRs, DArTs and EST-based markers developed for pearl millet (RKV/CTH/SS/DAH, 2010)

A total of 627 markers (100 genomic SSRs, 60 EST-SSRs, 100 pearl millet SSCP-SNPs, 57 wheat SSCP-SNPs, 310 CISP-SNPs) were screened on 24 genotypes including parental genotypes of 11 mapping populations (H 77/833-2 x PRLT 2/89-33; ICMB 841-P2 x 863-P3; Tift 23D2B1-P5 x WSIL-P8; PT 732B-P2 x P1449-2-P1; LGD 1-B-10 x ICMP 85410-P7; 81B-P6 x ICMP 451-P8; 81B-P6 x ICMP 451-P8; ICMP 451-P6 x H 77/833-2-P5 (OT); W 504-1-P1 x P310-17; IP 18293-P152 x Tift 238D1-P158; ICMB 89111-P6 x ICMB 90111-P6 and IPC 804 x 81B) and two tester lines (Tift 383 and Tift 186). Overall a total of 336 markers displayed polymorphism in at least one of 11 mapping populations. The polymorphic markers in different mapping populations ranged from 113 (Tift 23DB-1-P5 x WSIL-P8) to 151 (ICMB 841-P2 x 863-P3) with an average of 115 markers per population. Two mapping populations (i.e. ICMB 841-P2 x 863-P3 and 81B-P6 x ICMP 451-P8) are being advanced to RILs (at present F6 lines) and therefore in the first instance, these mapping populations will be genotyped with the polymorphic markers.

RK Varshney, T Mahender, CT Hash, S Senthilvel and DA Hoisington

Output target D.5: Agriculturally beneficial microorganisms assembled for utilization (2010)

Activity D.5.1: Assemble and conserve agriculturally beneficial microorganisms for utilization and distribution

Milestone D.5.1.1: Agriculturally beneficial microorganisms from diverse environments accessed and characterized for 6 different traits – P- solubilization, antagonism to disease-causing fungi, pathogenicity to insect-pest, siderophore production (OPR, Annual)

A total of 523 isolates involving bacteria, fungi, and actinomycetes were added to the short to medium term storage, for further studies. All these were picked to represent diversity in a soil sample from a given crop-production system and had at least one agriculturally beneficial trait such as ability to solubilize rock phosphate, plant growth promotion, antagonism to plant pathogenic fungi, pathogenicity to insect-pests. Most of these were bacteria and were characterized further for confirmation of the traits they were picked for and to learn those with more than one beneficial traits. The traits studied were P-solubilization, antagonism to Macrophomina phaseolina (a soil-borne root rot fungus), siderophore production (indicator of plant growth promotion) and

presence crystalline protein. Eight were positive for at least two traits and four (SRI77, EB24, SB26, HIB67) had crystal proteins of the type found in *Bacillus thuringiencis* (Table 8).

Table 8. Characterization of bacterial isolates for multiple beneficial traits

S. No	Isolate number	P-solubilization	Antagonistic to *M. phaseolina*	Siderphore production	Crystalline protein
1	SRI 77	-	-	-	+
2	SRI 151	+	+	+	-
3	SRI 156	+	+	+	-
4	SRI 158	+	+	+	-
5	SRI 178	+	+	+	-
6	SRI 229	+	+	+	-
7	SRI 305	+	+	+	-
8	EB24	-	+	ND	+
9	SB26	+	+	ND	+
10	HIB67	-	-	ND	+

ND = Not Determined

OP Rupela

Milestone D.5.1.2: Existing collection of agriculturally beneficial microorganisms conserved for medium and long-term storage system and annually 20% germplasm attended (OPR, Annual)

No long-term storage could be undertaken due to malfunctioning of the Freeze-drier, an equipment needed for the purpose.

OP Rupela

Milestone D.5.1.3: Requested agriculturally beneficial microorganisms distributed to bonafide users for utilization (OPR, Annual)

Requests received were for Rhizobia, the nodule forming bacteria, and for those microorganisms with ability to help manage insect-pests. Twenty five vials of rhizobial cultures and 24 units (one unit is sufficient to cover one acre of land) of carrier based inoculants of different rhizobial strains were provided. Three vials and 75 units of lignite-based entomopathogens were also provided. The requests were from NARS scientists, partners of Biopesticides Research Consortium (BRC) and peer scientists at ICRISAT.

OP Rupela

Project 3

Producing more and better food of the staple cereals and legumes of the west and central African (WCA) SAT (sorghum, pearl millet and groundnut) through genetic improvement

Output 3A. Heterotic relationships identified and utilized within sorghum and pearl millet germplasm adapted to WCA conditions and appropriate broad-based breeding populations and hybrid parents and made available to NARS and other partners annually to maximise genetic gain from selection

MTP Output Targets 2006
Multi-location testing of 100 pearl millet factorial crosses and parental populations completed at 6 locations to determine heterotic patterns

Multiplication and advance to BC5 generation of short statured Guinea-race A/B pairs of sorghum for the first time in west Africa

Impact from varietal improvement of the local dryland cereals of West and Central Africa (WCA) has seemingly lagged behind that of other crops and other regions. In recent years the ICRISAT team in WCA has made systematic efforts to improve this situation, by reorienting its breeding efforts towards farmers' expressed needs and preferences, by integrating varietal improvement efforts with natural resource management research, enhancing local cereal seed systems efficiently; and by starting to exploit heterosis within the well adapted locally widely grown guinea race sorghums. During 2006 we have been able to identify a set of hybrids based on guinea race sorghum landrace parents, which have potential for adoption by farmers, as they provide stable and significant heterosis across a range of test locations, on-farm and on-station. Producing seed of these hybrids appears to be feasible after our first experiences.

We have indications for consistent patterns of heterosis between different groups of guinea race sorghums from West-Africa. Sets of accessions representing these different groups of germplasm are presently under going SSR based marker analysis.

ICRISAT strengthened its capacities in Pearl Millet Breeding in WCA by recruiting a Senior Scientist in 2005. The program has gained strength and momentum by attracting a range of special donor funded projects. Thus research on heterotic grouping, developing potential hybrid parental lines based on West-African pearl millet germplasm is well under way.

Regional collaboration among the cereal breeders of the main cereal producing countries has been strengthened. Planning workshops, joint trials, joint monitoring tours of specific trials sites, and a regular information exchange are creating new dynamics for reviving hybrid breeding efforts among our partners.

Output target 3A1: Access of NARS to pearl millet and sorghum diversity enhanced

For pearl millet 360 early to medium-maturing pearl millet accessions originating from West-Africa were evaluated in the rainy season 2006 at six locations across WCA (Senegal, Mali, Burkina Faso, Niger, Nigeria). Similarly, 64 long-duration accessions were evaluated in the same season at 3 locations in the Sudanian zone, or with supplemental irrigation (Senegal, Mali, Niger). Breeders and farmers in the different countries and stations evaluated and selected germplam lines for different agronomic and traits that could be important for processing or commercialization. While the network of breeders intends to select materials for studies on heterotic groups, and for the creation of new broader based populations, each breeder can select materials for their own program, or for direct use. Patterns of adaptation among the WCA landraces are being examined carefully. An initial analysis was published in International Sorghum and Millets Newsletter (ISMN).

Sorghum breeders in the region have received locally adapted breeding materials and newly characterized accessions of guinea race sorghums on request, for specific zones of adaptation, and for specific uses. New groups of germplasm are being evaluated for use in the regional selection program, as well as for direct use by national programs. At present a set of earlier flowering materials from northern Nigeria and Cameroon is being assessed for widening the diversity of available breeding materials.

Activity 3A1.1: Evaluate early-medium and late pearl millet and sorghum germplasm from various locations in WCA (in Niger, Nigeria, Burkina Faso, Mali and Senegal for adaptation to specific agro-ecological zone in WCA.

Milestones: At least 10 useful accessions or population crosses identified and used for creating broad-based pearl millet breeding-populations for recurrent selection programs (BIGH 2007)

360 early to medium maturity pearl millet accessions evaluated in the rainy season 2006 (RS 2006) at 6 locations all over WCA (Senegal, Mali, Burkina Faso, Niger, Nigeria); 64 late-maturing accessions evaluated in RS 2006 at 3 locations (Senegal, Mali, Niger); 100 factorial crosses among diverse pearl millet populations evaluated in the RS 2006 at 7 locations (in Senegal, Mali, Burkina Faso, Niger, Nigeria). Data entry and analysis is underway. Each country will identify 5-10 superior accessions/population crosses from their own and the partner's trials; these will then enter recurrent selection programs targeting specific rainfall zones.

BIG Haussmann

Adaptation patterns of pearl millet germplasm in WCA published (2008)
First year multi-location field trials were completed in RS 2006. Preliminary characterization data and geographic differentiation patterns for 280 pearl millet landraces from WCA were evaluated at Sadore (Niger) in RS 2005 and published in the ISMN.

BIG Haussmann

Activity 3A1.2: Regional nurseries of sorghum and pearl millet germplasm assembled and made available to WCA NARS.

Milestones: Results from initial regional germplasm evaluation compiled and made available to partners (2007)

The multi-character evaluation of the guinea-race sorghum core collection includes a range of yield component traits, traits related to adaptation, fertility restoration on the A1 cytoplasm for male-sterility as well as standard morphological characters. The results are presently being compiled for publication and dissemination. In addition to the core collection evaluation, we conducted an evaluation of predominantly earlier maturing guinea or intermediates race germplasm from Nigeria, Chad and Cameroon during the 2006 rainy season. Individual panicles were selfed, and selected from accessions with interesting trait combinations. The Nigerian accessions were also sent to Zaria for evaluation in Nigeria. The sorghum core collection established by CIRAD was evaluated for growth rate during the 2004 and 2005 rainy seasons. The data was analysed, and is being prepared for publication.

HFW Rattunde, B Clerget and E Weltzien with CIRAD

Seed of at least 10 selected pearl millet accessions multiplied and made available to partners (2008-11)
Germplasm accessions from across West-Africa were evaluated in 5 countries (see above). Scientists from partner NARS, and farmers from areas near the research station evaluated these trials, and the selections were communicated to the ICRISAT breeder, who is planning to multiply seeds by sib-mating.

BIG Haussmann with IER, INERA, INRAN, ISRA and LCRI

Seed of at least 10 preferred new sorghum materials multiplied and supplied to at least 3 partners annually (2007-11)
The Nigerian germplasm evaluation, as well as the evaluations of early generation breeding materials on-farm by farmers and on-station evaluations served as opportunity for selections by partners through direct visual evaluations. The diffusion of analysed data will further serve to create demand by other researchers during the comings season.

HFW Rattunde and E Weltzien

Output target 3A2: Heterotic patterns in WCA pearl millet and sorghum germplasm understood.

A set of factorial crosses among a total of 20 pearl millet landraces from the five WCA countries was evaluated by all the contributors at 7 locations during the rainy season of 2006, enhancing everyone's access and insights into useful variability and potentially promising combinations of different types of pearl millet landraces from WCA. The first evaluation of a set of diallel crosses among contrasting pearl millet germplasm groups has been initiated

during the ongoing off-season. Genotyping of representative sets of pearl millet and sorghum germplasm has started in collaboration with the BECA in Nairobi, Kenya.

For the first time in 2005 and again in 2006 all interested partners in the different WCA countries have been able to test a range of the new guinea race sorghum hybrids. Results confirm consistently high levels of heterosis, especially for the Sudanian zone. Specific parents with high combining ability are starting to be become apparent, as well some patterns for high heterosis for grain productivity.

More detailed testing of grain yield heterosis is well underway, with the second season of data presently being analysed. In addition, 5 new guinea race hybrids and a local guinea race control, CSM 335, are being evaluated at 3 plant densities (67, 133 and 200,000 plants/ha) to confirm the results of yield potentials and grain yield components obtained in 2005. A sufficiently high rate of mineral fertilizers was applied to assess biomass and grain yield potentials of these hybrids.

The development of new hybrid parents of guinea race sorghum is advancing as planned. Thirteen new lines of diverse backgrounds and grain yield components have reached the BC6 stage of transfer into the male-sterility inducing cytoplasm. Seed of the lines developed earlier have been multiplied, and been made available to partners on request. For pearl millet a set of inbred lines representing the entire range of diversity from WCA is under development to initiate the process of parental line development parallel to the identification of heterotic groupings.

Activity 3A2.1: Multi-location evaluation of 100 pearl millet factorial crosses and their parental populations at 6 locations across WCA to study the effect of geographic and/or morphological distances of the parental populations on heterosis in pearl millet population crosses.

Milestones: At least 2 superior population crosses per partner country and agro-ecological zone identified and entering participatory recurrent selection programs (2007)

Field trials of a factorial design crossing involving 20 different parental populations from the five countries were evaluated at 7 locations (1 Senegal, 2 Mali, 1 Burkina Faso, 2 Niger, 1 Nigeria) during the RS 2006; and data entry and analysis are underway. Farmer and breeders performed visual selections that will enter into the overall analysis.

BIG Haussmann

Published article on combining ability patterns of WCA pearl millet landraces (2008)
The data from above trials will serve as a basis for this publication.

BIG Haussmann

Activity 3A2.2: SSR Genotyping of 250 pearl millet and 210 sorghum accessions for heterotic grouping

Milestone: Results on use of markers for assessing heterotic groups published (2008)

The sorghum and pearl millet accessions were selected from the germplasm collections evaluated for use in WCA, and for sorghum, including accessions and varieties in use in current hybrid breeding. Seed of selected accessions was sent for marker analysis.

BIG Haussmann and HFW Rattunde
with University of Hohenheim

Activity 3A2.4: Heterosis in Guinea-race sorghum hybrids assessed

Milestone: 2006: Regional Hybrid trials provided to WCA collaborators

A total of 91 experimental sorghum hybrids (based Guinea or inter-racial A lines crossed with Guinea male-parents) were produced and distributed as observation nurseries in 2006 for evaluating fertility restoration and agronomic desirability of hybrids *per se* and relative to the adjacent male-parent. These nurseries were provided to IER-Sotuba and INERA-Saria.

Regional Hybrid trials for collaborative yield testing of hybrids were prepared and dispatched to Mali, Burkina Faso, Senegal, and Nigeria (Table 1). Trials consist of 7 to 13 hybrids, based on adaptation zone and seed availability.

Table 1. Regional Hyrid Trials 2006

Zone	**N. Sudanian**		**S. Sudanian**					**Guinean**
Site (Country)	Bambey (Senegal)	Saria (Burkina Faso)	Bengou (Niger)	Samanko 1st Date (Mali)	Samanko 2nd Date (Mali)	Sotuba, Cinzana (Mali)	Sinthiou (Senegal)	Samaru (Nigeria)
# Entries	12	12	16	18	55	55	16	18

The Regional Sorghum Hybrid Trials in 2005 were conducted with NARS collaborators in the Northern Sudanian zone (ISRA, Bambey, Senegal; INERA, Saria Burkina Faso), the Southern Sudanian zone (INRAN, Bengou Niger; ISRA, Sinthiou Senegal; IER-Sotuba, Mali; and ICRISAT-Samanko, Mali (at two dates of sowing)), and the Northern Guinea zone (IAR-Samaru, Nigeria).

All trials had significant genetic differences for grain yields, with acceptable heritabilities and standard errors (Table 2). The grain yield superiority of hybrids over the local check is significant in the Southern Sudanian zone, with the best hybrid producing at least double the yield of the local check variety. The mean yield of all hybrids tested showed a 17 to 58% superiority over the local check, equivalent to 0.3 to 0.9 t more yield. In contrast, the superiority of these hybrids in the Northern Sudanian and Guinean zones were considerably less, indicating the need to develop other hybrids for these regions.

Table 2. Mean grain yields (t/ha) of all hybrids, best hybrid, open pollinated variety and local check variety in Collaborative Sorghum Hybrid Trials in the Northern Sudan (2) Southern Sudan (5) and Guinean (1) zones 2005

Zone	**N. Sudanian**		**S. Sudanian**					**Guinean**
Location	Bambey Senegal	Saria Burkina Faso	Bengou Niger	Samanko 1st Date Mali	Samanko 2nd Date Mali	Sotuba Mali	Sinthiou Senegal	Samaru Nigeria
Yield Best Hybrid	1.86	2.93	3.70	3.69	3.11	3.55	2.73	1.88
Mean (all Hybrids)	1.09	1.50	2.03	2.66	2.00	1.97	1.98	1.55
Best Variety	1.83	1.94	2.64	3.30	2.46	2.00	1.91	1.92
Mean (all Varieties)	1.15	1.12	1.45	1.81	1.59	1.21	1.21	1.20
Local Check	93B1062	Local 1	Sinthiou Local	CSM 388	CSM 388	CSM 388	Guinea Local	Fara Fara 27
Yield Local Check	1.48	1.75	1.73	1.79	1.58	1.25	1.37	1.92
Stand. Error	0.25	0.28	0.30	0.27	0.27	0.30	0.16	0.26
Heritability	0.79	0.77	0.85	0.87	0.83	0.87	0.93	0.63

HFW Rattunde, E Weltzien with IER, IAR, INERA, ISRA

Examination of the five highest yielding hybrids in each trial reveals the prevalence (and thus high combining ability) of a few male parents. Late maturing male parents from Humid West Africa; Nigeria (Fara Fara and IS 7978) and Cameroon (IS 30804 and IS 15629) occurred very frequently in the top yielding hybrids. The commercial feasibility of these hybrids is still unclear until off-season seed production techniques can be perfected. A male parent of the bicolor race (CSM 52-114) also consistently produced high yielding hybrids, although the tighter

glumes make threshing difficult. The potential of inter-racial hybrids was exhibited in Samanko late-sown environment. However, due to seed limitations, these hybrids were not tested in other environments and their response across variable sowing dates still needs to be assessed.

Results of multi-location guinea race sorghum heterosis trials analysed (2007)
Sets of hybrids made on a wide range of new guinea race A-lines, representing West, Central, and North-, as well South-Eastern African origins with a wide rage of restorer lines from WCA were evaluated at 4 locations during 2005 and 2006. Data analysis is underway, and shall results in a PhD thesis.

S Dagnoko, HFW Rattunde with IER, INRAN

Activity 3A2.5: Physiological characterization of heterosis in Guinea-race hybrids

Milestones: The contribution of individual sorghum yield components and panicle traits to grain yield heterosis quantified (2008)

As part of the heterosis trials mentioned above a wide range of panicle traits have been assessed. These assessments will be used to gain insights into the role of specific yield components for increasing grain yield in superior hybrids.

S Dagnoko, HFW Rattunde, B Clerget, E Weltzien with IER, INRAN

Adaptation of first generation guinea hybrids to different agronomic conditions assessed (2008)
Five superior hybrids were tested at three different stand densities and higher fertility, to test whether the yield potential of these materials is higher than that of parental varieties.

B Clerget and HFW Rattunde

Output target 3A3: Sorghum and pearl hybrid parents made available to NARS

Activity 3A3.1: Develop diversified Guinea-race sorghum maintainer and restorer lines

Milestones: Ten A/B pairs of diverse Guinea Landrace varieties advanced to BC5 generation (2006)

A diverse set of Guinea landrace maintainer lines were identified from the Guinea-race Core Collection, which samples the 3,900 Guinea-race accessions in the ICRISAT World Sorghum collection. A total of 13 maintainer lines were sterilized to produce A/B pairs. The maintainer lines were crossed to CK60A, and subsequently backcrossed to the recurrent parent, with the BC4 generation completed in the 2005/2006 off-season and the BC5 generation being completed in the 2006 rainy season (Table 3). These 13 A/B pairs extend the diversity of landrace-based Guinea-race A-lines beyond the initial three A-lines (Fambe A, IPS0001 A, and CSM 219A, all of Malian origin) previously developed, both for geographical origin, grain size and maturity. All of these A lines, being based on landrace germplasm, are of tall height.

A dwarf, photoperiod sensitive Guinea-race A-line (GPN 271-20 A) was produced from a derivative of the random-mating Dwarf Guinea Population. This population derivative also contains the A1 cytoplasm, from CK60A. This is truly dwarf, with short internodes and photoperiod sensitive, flowering at the beginning of October with late June sowing date. This A-line was multiplied in isolation in 2006.

Table 3. Genotypes and number of BC4 lines and corresponding B lines sown 2006 per recurrent parent and backcrossed to BC5

Landrace Parent	Snowden race	Origin	No. of Lines
IS 27580A/B	Guiniense	Burkina Faso	7
IS 6749A/B	Guiniense	Burkina Faso	5
IS 6781A/B	Guiniense	Burkina Faso	5
IS IS27494A/B	Gambicum	Burkina Faso	4
IS 19970A/B	Guiniense	Senegal	7
IS 20064A/B	Margaritiferum	Senegal	4
IS 20114A/B	Gambicum	Senegal	2
IS 22677A/B	Guiniense	Mali	16

Landrace Parent	Snowden race	Origin	No. of Lines
IS 23645A/B	Margaritiferum	Gambia	3
IS 27013A/B	durra-caudatum	Sudan	3
IS 3534A/B	Conspicuum	Sudan	5
IS 9220A/B	Margaritiferum	Uganda	3
IS 14414A/B	Conspicuum	Malawi	3

HFW Rattunde and E Weltzien

WCA partners trained in breeding techniques for creating Guinea-race sorghum hybrids adapted to WCA (2007)
The training course (to be held in 2007) will address the specific need to train sorghum breeder and their technicians in methods necessary for successful hybrid breeding, as well as the specificities of working with photoperiod-sensitive materials of the guinea race.

HFW Rattunde, E Weltzien, B Clerget with ROCAR*S*

At least 2 diversified guinea-race maintainer and restorer lines shared with at least three NARS in WCA (2008-11)
NARS have started to request hybrid parents for guinea races sorghums. Seed of FambeA and/or IS3534A was sent to Nigeria, Niger, Burkina Faso and Mali for initiating production of experimental hybrids. The training course may increase this trend.

HFW Rattunde and E Weltzien

Activity 3A3.2: Development of pearl millet inbred lines in West African background, and test of their general combining ability (GCA)

Milestones: At least 2 new pearl millet inbred lines with good GCA available to NARS/year (2009-2011)

We evaluated 432 testcrosses between WCA pearl millet landraces and 3 different male sterile lines at Sadore in RS 2006, to characterize fertility restoration/maintenance and to gain preliminary information about general combining ability. Data entry and analysis is underway. Lines are being developed out of the WCA pearl millet landraces and improved cultivars, these materials are currently at an inbreeding stage between S2 and S4 (depending on when inbreeding was started). A total number of 411 lines is currently under development.

BIG Haussmann

Output 3B. Improved breeding populations and open-pollinated varieties of groundnut, sorghum and pearl millet with adaptive advantages and/or improved yields under defined environmental and management conditions (including resistance to *Striga*) developed annually and made available to partners through regionally coordinated, largely participatory breeding efforts, integrated with natural resource management research, to meet the needs of poor farmers in each of the major agro-ecological zones of WCA

MTP Output Targets 2006

Partners from 4 countries trained in tools for participatory recurrent selection

700 F2-F7 groundnut breeding populations and lines screened for enhanced multiple disease resistance including rosette virus

While we expect hybrid breeding to open new perspectives for cereal improvement and seed system development for West-Africa, breeding open-pollinated varieties of sorghum and pearl millet will continue, and will be increasingly integrated with the hybrid breeding efforts. Breeding groundnut varieties for the needs of poor farmers, specifically women farmers shall continue.

The variety breeding and general crop improvement efforts are increasingly targeting specific needs of farmers. Thus the crop improvement efforts are addressing specific agro-ecological zones in WCA, as well as the evolving needs of intensifying production systems. Increasing the specificity of our plant breeding and variety development efforts is possible only through a close partnership with national researchers in the region, as well as farmers, farmer

organizations, and development projects targeting rural area development. We are placing high emphasis on the development and application of participatory breeding tools to improve the effectiveness and efficiency of achieving outcomes for farmers in WCA.

Improving grain yield in specific WCA growing and production conditions is of particular concern, specifically for pearl millet and sorghum, which have very high biomass yields, good adaptation to climate variability and change, and are relatively well protected against the common diseases and pests. Efforts to increase *Striga* resistance in farmer preferred cultivars are being pursued, as well as options tested to integrate *Striga* control options successfully. Searching for options of increasing the partitioning of photosynthate to high quality grains is thus necessary for WCA.

Nutrition and health concerns as targets for crop improvement research in WCA are gaining importance, and breakthroughs are being achieved. New groundnut varieties can resist the early invasion by the aflatoxin producing fungi (*Aspergillus flavus*). Combined with improved post-harvest techniques farmers now have the option to grow groundnuts that will not have negative health effects on those who consume them. Tools for monitoring the contamination of harvested or processed products are now available, and could be used for consumer protection.

Output target 3B1: Availability of pearl millet and sorghum cultivars with high and stable mineral (Fe, Zn) content in whole and decorticated grain

With the addition of a nutritionist to our team, our chances to effectively address micro-nutrient nutrition through crop improvement innovations have been significantly improved. We have been able to augment reliable information and research results on causes for mal-nutrition of young children and their mothers in regions that depend on sorghum and pearl millet as their staples. We are thus contributing to in-depth baseline studies of mineral mal-nutrition in targeted project areas.

For the short term, we have targeted to quantify losses of mineral content, and possibly changes in availability during key stages of processing, which have not received much research attention to date. Testing of varietal differences for these key traits require methodology development, which has been launched in recent years. The methods will include women farmers involved in processing whenever appropriate in a participatory manner.

To study genetic variation and environmental stability of mineral contents in improved cereal cultivars and landraces with contrasting grain characteristics, replicated experiments involving 40-60 pearl millet and sorghum cultivars from the WCA region, were conducted in both 2005 and 2006 in Niger and Mali. The results from 2005 revealed highly significant differences between the entries and estimates of heritability on a plot basis of 61% for Fe and 83% for Zn contents in the whole grain. The fertilizer treatments did not have any effect on the grain mineral contents. Methods for better quantifying and possibly predicting decortication losses are presently being tested.

Activity 3B1.1: Screening diverse pearl millet and sorghum cultivars for mineral (Fe, Zn) content of the grains, determine environmental stability and decortication losses

Milestones: Report on and seed of contrasting pearl millet varieties for Fe & Zn content available for distribution (2007)

Experiments involving 40 pearl millet cultivars (with two fertilization levels and 3 replications) were conducted in both 2005 and 2006 at Sadore, Niger, to study genetic variation and environmental stability of mineral contents in improved pearl millet cultivars and landraces with contrasting grain characteristics. The results from 2005 revealed highly significant differences between the entries and estimates of heritability on a plot basis of 61% for Fe and 83% for Zn contents in the whole grain. The fertilizer treatments did not have any effect on the grain mineral contents. Grain analyses of the 2006 trial are underway.

BIG Haussmann

Report and seed of contrasting sorghum varieties for Fe & Zn content available for distribution (2007)
Experiments involving 90 and 70 sorghum varieties of different origins and backgrounds were evaluated for Fe&Zn contents during 2005 and 2006, respectively. The trials are also being used to study relationships between ease of

decortication, other physical grain characteristics and the degree and amount of Fe and Zn losses in processed grain, and possibly foods.

HFW Rattunde, M Smit, E Weltzien with IER and HKI

Activity 3.B1.2 Assess opportunities for enhancing human nutrition in sorghum and pearl millet based cropping systems in WCA

Key sources of variation for micronutrient densities in sorghum and pearl millet identified (2006)
To identify sorghum varieties with high grain iron and/or zinc contents, a set of 90 sorghum varieties adapted to the Sudanian and Guinean-zones of West Africa were evaluated. The varieties tested included landrace varieties from Mali and Burkina Faso, accessions from the Guinea-race Core Collection of world-wide origins, and breeding lines and varieties from ICRISAT and Institut d'Economie Rural (IER) programs in Mali. Three replicate field trials were conducted on both red (P3b) and black soil (CSd) fields at ICRISAT-Mali in 2005. Although the early end of the rains resulted in severe drought stress in the black soil field, with later maturing entries producing no grain, acceptable yields were obtained from the red-soil environment.

Selfed, decorticated grain from each plot of the red-soil environment were analysed for iron and zinc contents at ICRISAT-India. Decorticated grain was analysed since previous analyses showed decortication reduces contamination, and, as most grain in West Africa is decorticated for food preparation, it corresponds most closely with the consumed product. Additional grain characteristics were observed such as pericarp thickness, decortication yield (10g samples in triplicate in TADD for 3minutes), ease of decortication (visual observation of extent of decortication with TADD) were also observed.

Varieties showed highly significant variation for iron and zinc contents of decorticated grains (Table 4). The varieties with the highest iron and zinc contents surpassed the mean by 19%, and 31%, respectively. The entry-mean heritability estimates for iron and zinc are above 0.70, indicating that genetically superior varieties can be bred. The superiority of these varieties needs to be confirmed however through testing in additional environments.

All of the grain characteristics studied showed highly significant genetic variation, with large genotypic ranges and high estimates of heritability. However, most of these characteristics showed no correlation with iron or zinc contents of the decorticated grain. Only decortication yield showed a moderate positive relationship with iron (r=0.52) and zinc (r=0.58). The high correlation between iron and zinc contents (r=0.84) suggests that the more highly heritable zinc content may be used for preliminary, indirect selection for iron contents.

Table 4. Mean, range, standard errors and entry-mean heritability estimates for 90 Sorghum varieties tested in Mali, 2005

Trait	Units	Mean	Minimum	Maximum	Std Err	Heritability
Iron	Ppm	25.2	19.7	30.0	1.56	0.72
Zinc	Ppm	15.7	10.5	20.6	1.20	0.80
Pericarp Thickness	Score 0(thin)-1(thick)	0.2	0.0	0.8	0.08	0.89
Ease of Decortication	Score 1(easy)-5(most difficult)	1.9	1.0	4.4	0.22	0.91
Decortication Yield	%	81.6	63.9	88.2	2.30	0.81
Vitreousness	Score 1(floury) to 9(vitreous)	3.4	1.6	6.7	0.31	0.90
Seed Weight	g per 100 seeds	2.4	1.4	3.4	0.11	0.92

HFW Rattunde, EWeltzien and M Smit

Literature review and survey published on importance of sorghum and pearl millet in West-African diets, with specific reference to micronutrient nutrition (2008).
An internal report and literature review was completed and is being shared with partners before finalizing for a publication. It is entitled: 'Evaluation of bio-fortification strategies and introduction of the best means to enhance the diets of nutritionally disadvantaged populations in developing countries.'

To help guide ICRISAT and West-African NARS partners to appropriately include nutritional enhancement in ongoing sorghum and millet variety development activities, a thorough review of literature was conducted. This review compiled and synthesized the information for Mali, Niger and Burkina Faso on millet and sorghum consumption, their contribution to overall diet and nutritional role, with special emphasis on micronutrients (iron and zinc).

In Mali 85% children (6 to 59 months age) suffer from anaemia, 92% in Burkina Faso and the figures for Niger, although not available, can not be any better. The deficiency of iron and zinc results in weak immune systems, reduces growth and cognitive development, thus contributing to the alarming child mortality rates of around 25% as well as restricting the potential of many of those who survive.

Micronutrient deficiencies may also be serious in adults as well, especially women. High rates of anaemia in pregnant women in Burkina Faso (68%) and Mali (63%) may place women's lives at risk during childbirth.

Millet and sorghum are confirmed to be the staple foods in the three countries, although rice is also important along the Niger river in Mali. In Niger millet (70%) and sorghum (20%) account for 90% of total cereal consumption. Their contributions, together with some maize, are similar in Burkina Faso (90%) and Mali (75%). These cereals have tremendous dietary importance, as diets are primarily based on plant-derived products with relatively little animal products. Sorghum and pearl millet contribute 67% to 90% to the total consumed energy (kilo calories). Millet and sorghum are rich sources of the micronutrients iron and zinc and of B-vitamins. Calculations based on mean iron and zinc levels from ICRISAT trials (pearl millet at Sadore, sorghum at Samanko) suggest that these cereals provide 29-51% of iron and zinc required by children, the group suffering most serious levels of micronutrient deficiencies and whose diets are based predominantly on millet and sorghum.

The review concludes that there needs to be both a) increase of bio-availability of existing sources of micronutrients through better transformation and food preparation practices, including the combination with other foods to increase absorption of iron and zinc, and b) exploiting the genetic diversity for iron and zinc contents in the staple cereals.

Marjolein Smit, H Fred W Rattunde and Eva Weltzien

Output target 3B2: Availability of genetically broad-based pearl millet and sorghum gene pools

To use the enormous variability among pearl millet and sorghum accessions from the WCA region for effective, and sustainable varietal improvement we have started to develop broad based populations. These populations tend to be well adapted to specific zones of adaptations, and harbour genetic variability for key traits targeted for improvement. Some examples are: the development of a pearl millet population combining available sources of *Striga* resistance, the guinea race sorghum population with reduced height due to reduced internode length, and thus improved stover digestibility.

Activity 3B2.1: Population diversification and recurrent selection for farmer-preferred traits including *Striga hermonthica* resistance in pearl millet

Milestone: At least three broad-based pearl millet populations developed and seed available for specific agro-ecologies of WCA and available for distribution (2008)

Farmer-preferred parental materials for the diversified populations are being identified from the 2006 rainy season trials (both on-station and on-farm) in Senegal, Mali, Burkina Faso, Niger and Nigeria. These will be crossed and recombined in the off-season and rainy season 2007, to be made available in 2008.

BIG Haussmann

One highly diversified pearl millet genepool with improved resistance to *Striga hermonthica* developed for use in West African breeding programs (2009)

A first screening of 64 pearl millet landraces from Niger was conducted in RS 2005 at Sadore under artificial striga infestation. Out of the least sensitive populations, both S1 and Full-sib families were developed in the off-season 2005-06. These were then evaluated in artificially infested field trials at Sadore, and partially also in two on-farm trials at Toroid and Falwel (both Niger) in the RS 2006. Data are being compiled.

BIG Haussmann

Activity 3B2.2: Creation of diversified Dwarf Guinea Sorghum Populations

Milestones: Partners from 4 WCA countries trained in tools for participatory recurrent selection (2006)

A training course involving pearl millet breeders and sorghum breeders, their technicians, and partners from farmer organizations as well as development agents from large-scale development projects from Mali, Burkina Faso, Niger and Nigeria was conducted from 9-18 September 2006 in Segou, Mali. In addition to the plant breeders the course participants include plant breeding technicians, farmers chosen from farmer organizations active in seed and variety identification activities, and development workers involved in activities related to variety testing and seed production. The course covered three main topics: a) an introduction to recurrent selection methods and factors that contribute to successful selection gains; b) Options for farmer participation in the different stages of a recurrent selection program for variety development and c) Organizational and institutional issues to consider in setting up and implementing participatory variety development programs. The last day of the workshop was used to plan activities for future collaboration on sorghum and pearl millet breeding.

E Weltzien

One diversified dwarf Guinea – race Sorghum population with farmer preferred traits available for at least two different agro-ecologies of WCA (2009)

One trait that attracts farmers' attention and strong expressions of preference is the stover quality of guinea race sorghums. Diversifying and improving these locally well adapted sorghums for stover quality may contribute significantly towards improving the economic value of the sorghum crop in WCA. We thus characterized key parameters for stover quality in novel dwarf- and landrace-sorghum varieties adapted to WCA and their relationship with agronomic traits.

Assessment of stover quality parameters of stem samples from 70 novel dwarf-stature Guinea-race sorghum lines and landrace check varieties was conducted to a) quantify the genetic variability for stover quality provided by our new dwarf Guinea-race sorghums, and b) assess relationships between stover quality and major agronomic traits. This first systematic assessment of stover quality of these materials will provide guidance on the opportunities and directions for developing novel dual-purpose sorghum varieties for West Africa.

The stem samples and agronomic measurements were made at ICRISAT-Samanko in 2004, in a four replicate trial. Key agronomic characteristics such as grain yield, maturity, stem length (distance from ground to base of peduncle), stem diameter, and internode lengths were measured. Stem quality parameters were estimated for dried, ground (1mm sieve) stem samples using NIRS at CIRAD, Montpellier. The parameters estimated were total minerals (MM), crude protein (CP), crude fiber (CBW), Neutral detergent fiber (NDF), Acid Detergent Fibre (ADF), Acid Detergent Lignin (ADL), In-vitro digestibility of dry-matter (IVD-DM) and In-vitro digestibility of organic matter (IVD-OM).

Highly significant genetic variation was observed for all quality parameters.
Considerable similarity among sister lines, and considerable differences between families of progenies and high entry-mean heritability estimates confirms the importance and repeatability of genetic differences for all quality traits (Table 5).

Table 5. Variation and entry-mean heritability for key stem-quality and agronomic traits among 70 new dwarf Guinea-race sorghum breeding lines and landrace checks

Trait	Units	Minimum	Maximum	Mean	Standard Error	Heritability
In vitro Organic Matter Digestibility	%	16	45	28	2.3	0.89
In vitro Dry Matter Digestibility	%	17	48	32	2.3	0.89
Acid Detergent Fiber	%	35	52	44	1.6	0.87
Neutral Detergent Fiber	%	60	86	76	2.2	0.88
Crude Fiber	%	33	50	42	1.5	0.89
Acid Detergent Lignin	%	5.0	8.6	6.8	0.3	0.91
Crude Protein	%	1.0	4.6	2.6	0.3	0.82
Total Minerals	%	2.1	6.4	3.9	0.6	0.63

Trait	Units	Minimum	Maximum	Mean	Standard Error	Heritability
Stem Diameter	Cm	1.4	2.5	1.9	0.1	0.71
Stem Internode Length Observed	Cm	4.5	23.8	12.1	0.8	0.97
Stem Length	Cm	65	252	137	8.9	0.95
Leaf Percent	%	26	58	41	2.2	0.88
Heading date	joulian d	260	287	275	1.5	0.92
Grain yield	g m2	22	249	129	23.0	0.74
Harvest Index	%	13	43	28	2.3	0.88

Shorter stem internode length was correlated with higher stem organic-matter digestibility (r= -0.64), indicating that approximately 40% of variation for stem digestibility due simply to stem internode lengths. Entries with very short internode (<12cm) show on average considerably higher nutritional values. Breeding lines with internodes less than 12cm, derived both from inter-racial pedigree breeding and from a Dwarf Guinea Population, showed higher organic-matter digestibility (adjusted by covariance for grain yield differences) and crude protein and lower NDF, ADF, and ADL mean values (Table 6). The 40 progenies with length of stem internode of less than 12cm had mean IVD-OM of 32.1, with a range from 20.1 to 44.8, as compared to traditional landrace varieties with IVD-OM ranging from 17 to 26. Also, short internode length showed strong association with higher leaf percent (r=-0.80).

Table 6. Mean stover quality parameters for Inter-racial and Dwarf Guinea lines with stem internodes 12cm or less as compared with landrace varieties

	IVD-OM	IVD-DM	ADF	ADL	NDF	CBW	CP	MM
Inter-racial	29	34	42	6.0	73	40	2.8	4.3
Dwarf Guinea	31	36	42	6.3	72	40	3.0	3.8
Landrace Varieties	*26*	29	47	7.2	78	45	1.8	2.7

Varieties with short stem internodes, however, do not necessarily exhibit higher stover quality. Certain varieties with intermediate to short stem internodes showed stem IVD-OM values that were no better than those of traditional landrace varieties (Table 7).

Table 7. Stem organic-matter digestibility, yield and internode lengths of dwarf- and traditional landrace varieties of sorghum

Variety	IVD-OM	Grain Yield t/ha	Internode (cm)
Short Internode varieties			
01-CZ-F5P-169	35	0.3	13
GPN01 266-1-2	34	1.1	8
Grinkan	31	1.4	8
Nafalen	31	1.5	13
Malisor 92-1	26	0.6	8
CGM 19/9-1-1	21	1.5	15
Weli	20	1.8	14
Mean 40 varieties internodes<12cm	32	1.2	9
Landrace Varieties			
Bobodje	34	0.6	17
Sakoykaba	26	1.3	21
CSM 335	25	1.4	20
CSM388	19	1.5	24

Very strong correlations (r= –0.98 to –0.99) were observed between digestibility estimates (IVD-OM, IVD-DM) with NDF and CBW; indicating that these estimates are not independent and do not need to be examined separately.

The relationship of IVD-OM with grain yield was weak (r= -0.44), with combination of higher yield and digestibility being possible. Surprisingly, high stem digestibility was association with later flowering (r=0.78) (all average to late heading (270-285 days) entries had IVD-OM >30%). This may be due in part to later flowering being associated with lower harvest index (r=-0.74), and low harvest index being associated with higher digestibility (r=-0.62).

The major conclusions that can be drawn from these results are 1) new dual-purpose sorghum varieties with substantially superior stover quality and acceptable grain yield can be developed for West Africa, 2) the dwarf (short stem internode) phenotype is associated with improved stover quality, although considerable variation for quality exists within dwarf phenotypes and thus, 3) *in vitro* and/or *in vivo* assessment of stover quality of promising varieties are necessary to develop superior dual-purpose types that maximize total productivity and value of the crop/livestock systems.

HFW Rattunde, Eva Weltzien and D Bastinell*i*

Activity 3B2.4: Develop farmer preferred *Striga* resistant varieties of sorghum and options for integrated *Striga* management.

Milestone: Transfer of Striga resistance QTL's into at least on guinea race variety adapted to the Sudanian zone of WCA (2009)

BC2F1 were developed at IER, and are presently being analysed for specific markers for positive selection for he presence of he desired QTL's as well as for background selection for the target genotype.

D Kiambi with IER, BeCA

Approach tested for adapting integrated *Striga* control options to specific cropping systems in collaboration with farmers (2008)

A farmer field school methodology was adapted to work with pearl millet farmers on integrated *Striga* management options in Sahelian cropping systems. The approach was tested in two clusters of 6 villages each. Results indicate that the chosen combination of treatments resulted in a reduction of the amount of *Striga* seed in the soil.

T van Mourik and E Weltzien

Quantification of interaction between different *Striga* management options on *Striga* seed bank dynamics in the soil (2008)

In both sorghum and pearl millet systems the factorial combinations of three control options, varietal resistance, intercropping, and organic amendments were evaluated in on-station trials at Samanko and Sadore.

T van Mourik, E Weltzien and BIG Haussmann

Output target 3B3: New farmer-preferred pearl millet and sorghum cultivars with improved yields

The development of superior finished varieties is necessary in WCA, because many national programs have poorly supported breeding programs. They are also necessary as a proof of concept in this region, where farmer managed yield improvements have been rarely manifested for newly developed varieties of local dryland cereals. For pearl millet our efforts at variety improvement have re-started this year in several target production systems across the WCA region.

New guinea race sorghum varieties (tall and dwarf) are showing consistent yield improvements in farmer manage trails, primarily in the Soudanian zone. We have initiated efforts to increasingly target the southern Sahelian zone, as well as the northern Guinean zone.

An initial effort at mapping the adaptation of specific guinea race sorghum varieties, based on their photoperiod response, and thus time of maturation, has resulted in products that give some broad indications, but lend themselves to explaining issues of varietal adaptation to those involved in developing seed sector innovations, regionalizing seed policies, and preparing for seed distribution or commercialization.

Activity 3B3.1: Farmer-participatory recurrent selection for head/grain yield in diversified pearl millet populations

Milestones: Four improved, farmer-preferred pearl millet OPVs available to farmers in Niger and Mali (2009)

Farmers conducted full-sib progeny trials in population crosses at three locations in northern Nigeria, targeting the Sahelian zone. Farmers made visual selections, and researcher conducted agronomic evaluations. Joint selections are being recombined for further recurrent selection.

BIG Haussmann and S Boureima

Activity 3B3.2: Farmer-participatory sorghum variety development and testing

Milestones: Farmer preferred varieties from on-going trials registered in Malian Variety catalog (2007)

A set of 15 tall guinea race lines and varieties, and 15 short line and varieties were evaluated during 2005 and 2006 in multi-location yield trials in two areas of Mali, in the Sudanian zone. The trials were conducted at two research stations, and in 10 villages in 2005, and 9 villages in 2006. The varieties were assessed by researchers for agronomic performance traits (grain yield, panicle yield, panicle number, and 1000 grain mass). Farmers who grew the trials, evaluated the plots at regular intervals visually. Other farmers from the vicinity visited the trials before harvest, and scored each plot for its desirability. After harvest, and analysis of the agronomic and preference data, farmers and researchers selected together the four best entries in each village. These four varieties were evaluated for their processing characteristics for local dishes, as well as for culinary traits of the main dish commonly prepared from sorghum in this region. The data is presently being analyzed for preparing release proposals, and other publications.

E Weltzien, HFW Rattunde, with IER, ULPC and AOPP

Tools for farmer participatory early generation testing of sorghum genotypes published (2008)
Specific tools for facilitating farmers' input into the evaluation and selection of sorghum varieties under testing have been tested over the past years in Mali. During 2006 these tools were disseminated through a training program for breeders. The tools are now being used in four countries in West-Africa. Based on feed-back from these users a publication shall be prepared.

E Weltzien, HFW Rattunde with IER

Activity 3B3.3: Information on variety adaptation made widely available

Milestones: Regional validation of the variety adaptation maps reported with the breeders of WCA countries (2008)

In collaboration with Agryhmet and using the regional data sets of weather observations, and a simplified water balance model, the dates for the end and the beginning of the rainy season across different ecologies in West Africa were estimated. Based on these estimates, and the known dates of flowering of widely tested photoperiod sensitive sorghum and pearl millet varieties, zones of adaptation were proposed, and mapped. Further refinement of these maps is under discussion.

PCS Traore, B Clerget, BIG Haussmann, E Weltzien, HFW Rattunde with IER

Publication of results, based on field studies and modeling, on the effect of *Striga resistance* of the host crop varieties on the effectiveness of different combinations of *Striga* control techniques (2008)

Trials observing *Striga* seed bank dynamics were conducted during 2005 and 2006 at Samanko, and Sadore estimating factors contributing *Striga* both seed production as well loss of viable seed in the soil. The preparation of a PhD thesis is underway.

T van Mourik

Output target 3B4: Availability of allele-specific molecular markers for genes controlling photoperiod sensitivity of flowering time in pearl millet and sorghum

As photoperiod sensitivity is key for varietal adaptation of sorghum and pearl millet in WCA, we are investigating options for increasing our efficiency in handling this character in a breeding program through the application of

molecular markers. Material for this research has largely been assembled, tools and methods are being finalized. Protocols for phenotyping are available now, but need to be published.

Activity 3B4.1: Phenotyping and genotyping sorghum and pearl millet lines and accessions

Milestones: Paper published on effect of photoperiod on growth and development of West-African sorghum and pearl millet varieties (2007)

The 24th of the two year series of monthly sowings of 12 pearl millet varieties was completed in June 2006. An additional sowing has been done in November 2006 to compensate for the failure of the sowing of November 2004. A publication is under preparation combining the results from the sorghum observations, with these new pearl millet results.

B Clerget, HFW Rattunde, BIG Haussmann, E Weltzien and S Boureima

Output Target 3B5: Improving plant growth model for highly photoperiod-sensitive sorghum and pearl millet

Understanding the biology of photoperiod sensitivity, and modelling plant growth and development of photoperiod sensitive sorghum and pearl millet are instrumental for devising avenues for further improvements of productivity, as well as for predicting responses to existing and future climate variability. A key issue for predicting varietal adaptation is a better understanding of the relationships between latitude and photoperiod-sensitivity. Trials to arrive at a better understanding of this situation have put in place over a large range of latitudes in WCA and ESA.

Similarly crucial for the adaptation and stability of productivity of the local cereals is a better understanding of the relationship between root, shoot growth and grain yield potential of photoperiod sensitive varieties. Studies have been initiated but require confirmation across years with different rainfall regimes.

Activity 3B5.1: Study relationships between latitude and photoperiod-sensitivity in sorghum and pearl millet

Milestones: Multi-latitudinal trials from equator to temperate latitudes conducted, data assembled (2008)

Five sorghum and five pearl millet West African varieties have been sown in May, June and July 2006 at the CIRAD station, Lavalette, Montpellier, France (43° 37'N). Leaf appearance and panicle initiation have been recorded. An international experiment on the effect of the latitude on the photoperiod-sensitivity of sorghum has been planned. From December 2006 to December 2007, 10 sorghum varieties from West, East and South Africa, will be monthly sown in Kenya, Tanzania, Mozambique, Zimbabwe and Mali.

B Clerget and M Mgonja

Output target 3B6: Groundnut breeding lines with resistance to groundnut rosette disease, early and late leaf spots and aflatoxin contamination available for distribution

In groundnut genetic enhancement, there has been a shift in emphasis from single stress factor resistance to multiple stress factors resistance to ensure wide adaptability of newly developed genotypes in the semi-arid tropics. Our collaborators have access to early-generation breeding populations, advanced breeding populations, and near-finished genotypes to enrich and diversify their national programs. ICRISAT is continuing to make new segregating lines, and stable varieties available to NARS partners for testing and release. Most of this work is being conducted in a collaborative manner, and involves farmers in the variety evaluation phase. Variety releases for specific countries are imminent, and farmers are continuing to adopt the new varieties from the trials plots. Increasing the availability of breeder's seed is increasing the potential for adoption of these varieties.

Activity 3B6.1: Screening groundnut breeding populations for multiple resistances

Milestone: About 700 F2-F7 breeding populations and lines with enhanced resistance to groundnut rosette disease and aphids screened for resistance to early and late leaf spots at Samanko, Mali (2006)

Groundnut rosette is the most destructive disease of groundnut in sub-Saharan Africa. Improved rosette resistant varieties of short-, medium- and long-duration have been developed but many are highly susceptible to the most important foliar diseases (early and late leaf spots and rust). The resistant genotypes will form a strong component of overall integrated disease management strategies along with cultural and biological management options. A total of 844 diverse breeding populations and advanced breeding lines were screened for early leaf spot, the most prevalent disease at Samanko (Table 8). Segregating populations were grown essentially for generation advance and further selection.

Table 8. Breeding populations and lines screened for early leaf spot (ELS), Samanko , 2006

Objective	Generation	No. entries	Source	Number resistant to ELS (score ≤ 5)
Rosette resistance and earliness	F3	70	WCA	14
Rosette resistance and earliness	F4	64	WCA	9
Aphid resistance and confectionery (Virginia)	F4	55	ESA	8
Aphid resistance x ELS resistance	F4	124	ESA	23
Aphid resistance x rosette virus resistance	F5	33	ESA	0
GRV resistance	F6	36	ESA	5
GRV and dormancy	F6	63	ESA	4
Aphid and GRV resistance	F6	49	ESA	0
Aphid resistance inheritance	F6	27	ESA	1
Aphid resistance backcrosses	F6	8	ESA	0
GRV inheritance- aphid x GRV	F6	5	ESA	0
Dormancy and rosette resistance	F6	27	ESA	0
Aphid and ELS resistance	F6	16	ESA	1
Aphid and ELS resistance	F7	92	ESA	7
Rosette resistance	F7	5	ESA	0
High oleic acid and rosette resistance	F7	21	ESA	0
Aphid and rust resistance	F7	22	ESA	1
Aphid inheritance	F7	127	ESA	2
Total		**844**		

Generation advance: We grew 70 F3 and 63 F4 bulk populations at Samanko for generation advance. These populations are derived from crosses involving early maturing susceptible lines and sources of resistance to groundnut rosette disease. The aim of screening these for early leaf spots is to eliminate the super susceptible ones. Among the F3 populations 14 recorded a score of ≤ 5 (on a scale of 1-9, where 1 is resistant and 9 highly susceptible) while 9 of the F4 populations were rated as resistant. The most susceptible population had a score of ≥ 7. Single pod bulks were made from the most productive plants with tolerance to ELS. These will be tested in rosette disease nursery at Samaru, Nigeria at an appropriate time.

Evaluation of diverse breeding lines for early leaf spot: Breeding lines derived from various population for incorporating resistance to both Groundnut Rosette Virus and aphid into different backgrounds were grown for seed increase and screened for early leaf spot. Selected lines based on pod load will be evaluated for yield and other desirable traits in initial variety trials in the 2007 crop season.

Advanced breeding nursery: At the request of the groundnut breeder at the Savanna Agricultural Research Institute, Tamale Ghana, an observation nursery consisting of 12 rosette resistant early maturing (90 days) lines were dispatched. Breeder seed of these lines was produced at Samanko.

BR Ntare and AT Diallo

Breeder seed production: Breeder and foundation seed production is an essential component of the breeding program. Breeder seed (30-135 kg) of six promising early maturing and rosette resistant varieties were produced. In addition foundation seed of the currently popular varieties in Mali (ICG 7878, ICGV 86124, ICGV 86024, ICG (FDRS) 4, Fleur 11 and JL 24) was produced and quantities range from 225-450 Kg (Table 9). This seed will be sold to replenish the revolving fund established in 2005.

Table 9. Breeder and foundation seed production (kg) produced at Samanko, 2006

Variety	Breeder	Foundation
ICG 7878	-	102
ICG 6222	-	150
ICG(FDRS) 4	-	465
ICGV 86124	-	225
ICGV 86024	-	388
ICGV-IS 01836	65	
ICIAR 6 AT	135	
ICIAR 19 BT	-	165
ICIAR 7B	60	
ICGV-IS 01835	30	
JL 24	-	295
Fleur 11	-	378
ICGV 86124	15	
Fleur11	2	
ICGV 86024	12	
ICG 7878	2	
ICG 7	12	
JL 24	2	
ICG 6222	2	
ICGV-IS 01859	5	
ICIAR 19 BT	5	
ICGV 92093	2	
ICGV-IS 01851	3	
ICGV-IS 01850	4	
ICGV 97188	4	
ICIAR 6 AT	1	
ICG(FDRS)4	4	
ICIAR : Varieties jointly developed by ICRISAT and with IAR, Nigeria		

BR Ntare and AT Diallo

Activity 3B 6.2: Evaluating advanced breeding lines with multiple resistances for yield and other performance trait

Milestone: Two groundnut varieties released in Senegal and Mali (2006)

Farmers' access to improved groundnut varieties is fundamental to achieve the desired the impact. Farmers select varieties based on their own preference criteria for variety use and cultivation. This enhances the rate of adoption.

Participatory variety selection (PVS) trials were initiated in the main groundnut growing areas regions in Senegal (11 varieties) and Mali (13 varieties) in 2003. During the three-year testing farmers in Senegal selected ICGV 86124 and ICGV 89063, while in Mali they have selected ICGV 86124. These varieties also showed wide adaptation in earlier regional variety trials conducted across West Africa. Both varieties are early maturing and high yielding (2.0-2.5 t/ha under good management on-farm). In addition, ICGV 89063 is tolerant to Aflatoxin contamination while ICGV 86124 is tolerant to drought. Based on these attributes and farmers' preference they have been recommended for release, and the relevant documents have been submitted to the variety release committees in the

two countries. Formal release has been delayed as none of the release committees in both countries has met to approve the varieties for registration.

BR Ntare, A Dasylva and O Kodio

Lines with combined resistance to groundnut rosette disease, aphids, early and late leaf spots evaluated for yield performance (2007)
During the 2006 crop season, a number of lines with desirable agronomic and quality characteristics (plant type, pod shape, seed size and color) will be entered into preliminary on station trials to evaluate their yield potential.

B Ntare

A report on groundnut participatory variety selection published (2007)
A draft synthesis report on the participatory variety selection (PVS) process was prepared, summarizing groundnut variety selection trials conducted in Mali, Niger, Nigeria and Senegal. It documents the pathways to adoption of improved groundnut varieties, the lessons learned and the perspectives for enhancing variety adoption. Surveys were initiated in Dec 2006 for additional information to strengthen the report.

B Ntare and J Ndjeunga

10-25 breeding lines with multiple resistance to groundnut rosette and foliar diseases made available on request to NARS in WCA (2008)
Promising lines from the preliminary evaluation trials will be made available to NARS on request.

B Ntare

Activity 3B6.3: Conduct adoption and impact assessment of groundnut improvement research in WCA

Survey on the adoption and impact of improved groundnut varieties in four countries of WCA completed (2008).
Surveys on the adoption of improved varieties in Mali, Niger and Nigeria were initiated in December 2006.

J Ndjeunga and B Ntare

Output 3C: Crop management, *Aspergillus flavus* resistant groundnut varieties and post-harvest technologies available to reduce aflatoxin contamination in food and feed products in the SAT of WCA

MTP Output Target 2006: Analysis of results from integrated aflatoxin management options trials undertaken

The main health related research area the WCA is pursuing concerns of aflatoxin contamination in groundnut. Aflatoxin is carcinogenic and have a broad range of negative side effects on the immune system and growth and development, especially of children. While it would be useful to understand the medical ramifications better, we focus on options to reduce contamination of food and feed products. The field research conducted with farmers confirms on-station results that aflatoxin contamination can be reduced significantly, down to levels that are safe for consumption for young children. Monitoring of marketed products also showed that quality of available products covers an enormous range, including samples that are safe for consumption. Producer and consumer awareness of the existing problem, as well as options for its improvement are extremely low, but we are starting to tackle them.

Output target 3C1: Post-harvest technologies, and resistant groundnut varieties tested with producers, and contamination levels monitored at the consumer end.

Research results from on-farm trails testing different options for reducing aflatoxin contamination in a range of locations are all very positive, and showing the feasibility of control options. These results need to be published, and undergo economic feasibility analysis.

Activity 3C 1.1. Develop breeding populations and lines with enhanced resistance to aflatoxin contamination

Milestones: A synthesis report on Integrated Aflatoxin management published (2007)

Data from verification and demonstration trials of technologies to minimize aflatoxin contamination were analyzed. Preparation of an information bulletin is in progress.

B Ntare and F Waliyar

At least 10 promising breeding lines resistant to foliar diseases and aflatoxin contamination available to NARS in WCA (2008)
Promising lines from the preliminary evaluation trials (see above) will be available for distribution to NARS on request.

B Ntare and F Waliyar

Activity 3.C.1.2 Test crop management options to reduce Aflatoxin contamination in groundnuts

Milestone: Technologies to minimize aflatoxin contamination scaled-out to 3 countries in WCA (2007)

On-farm demonstration of best-bet harvesting techniques and tolerant varieties: We have successfully developed and tested integrated management technologies to prevent aflatoxin contamination at the farmer level in Mali. However large scale dissemination of these technical packages, along with intensive sensitization campaigns across the commodity chain remains a major challenge. Awareness about aflatoxin contamination is improving and efforts were made to continue dissemination technology packages on the control of aflatoxin contamination at production level.

On-farm trials/demonstrations of the best-bet harvesting and drying techniques were conducted in Nigeria for a second year and in Senegal for the first time. In Nigeria, improved method of drying the pods facing the sun reduced Aflatoxin contamination by as high as 97% compared to the farmers' method of windrowing. Aflatoxin content in seed ranged from 3.73 to 9.00 ppb under the improved method compared to 6.00 to 337.00 ppb under the traditional method. These results are consistent with those obtained in the previous crop season. This simple management technique can significantly contribute to healthy groundnut products and needs to be promoted widely.

Raising awareness through training workshops and other information dissemination pathways: ICRISAT provided technical and partial financial support for a 2-day workshop on best-bet harvesting and drying techniques in Niger. Projet Intrants of FAO Niger organized the workshop. Two representatives from each of the four regions of Niger attended the workshop. These would in turn impart the knowledge to a large audience in their respective areas.

In Mali, a two-day workshop on crop management practices and quality control was held for a group of farmers in Sanakoroba district. KILABO, a local NGO facilitated the workshop. Two extension agents and 15 women attended. At the request of Women Association of Wakoro in Doila, ICRISAT organized a 4-day training workshop in crop management. During these workshops, the participants were sensitized on how to minimize the aflatoxin contaminataion.

Group discussions were regularly held with farmers, traders and processors to assess their awareness. The interaction revealed that many farmers were ignorant of the aflatoxin problem. They opined that since there is no visible indications of aflatoxin on the seed they considered end-of season drought to be responsible for low yields and bitterness of seeds. It is known that any delay in harvesting the crop under end-of season drought could severely reduce yield and increased aflatoxin contamination.

Information dissemination: Under the CFC supported project, information leaflets on how to minimize aflatoxin contamination (English and Hausa languages) were prepared in Nigeria and will be widely distributed. In Mali 1000 flyers (in Bambara and French) have been distributed to raise awareness about Aflatoxin contamination.

Information was also disseminated using posters. Two posters on integrated management of Aflatoxin and ICRISAT strategy to control Aflatoxin contamination were displayed at the Malian National Agricultural Research week. More than 500 persons including policy makers, NGOs, etc., visited the stand.

BR Ntare, AT Diallo, HY Bissala, F Waliyar,
C Echekwu and A Da Sylva

Monitoring aflatoxin contamination in target areas in WCA: In addition to on-farm trials/demonstrations, ICRISAT has also been monitoring levels of Aflatoxin contamination from 20 farmers in the districts of Kolokani, Kayes and Kita in Mali. Some of the farmers have participated in participatory variety trials while others have visited some of the demonstration plots in the districts. From 21 samples in Kolokani, the Aflatoxin content ranged from 0.2 to 75 ppb, in Kayes (20 samples) the range was 6.39 to 1597 ppb, and in Kita (80 samples) the range was 4.2 to 1152 ppb. The alarming high levels of Aflatoxin contamination in Kayes and Kita are of concern and show that these areas are Aflatoxin risk prone. However, results from Kolokani showed a significant decline in the level of aflatoxin contamination. This is an indication of adoption of improved management practices.

Aflatoxin contamination was also determined in samples from pods, kernels and groundnut butter in several markets in Bamako city. The samples showed a wide range of aflatoxin content levels (Table 10). Over 80% of the samples had Aflatoxin content far beyond permissible (i.e 20ppb) levels for animal feeds. Permissible level for human consumption is < 5 ppb. These high levels in market samples arise from contamination through transportation, poor handling and storage conditions. Thus stakeholder sensitization about measures to control contamination further down the commodity chain is essential.

Groundnut is an important component of the diet of many groundnut producers in West Africa. Many people are not only malnourished but also chronically exposed to high levels of toxic fungal metabolites (mycotoxins). Aflatoxin contaminates staple foods, in West Africa, particularly maize and groundnuts, as a result of hot, humid storage conditions that promote fungal growth. Groundnut paste is used in nearly every household in Mali. High exposure to aflatoxins occurs throughout childhood in the region suggesting that growth and development could be critically affected. It would thus be important to assess the exposure of aflatoxins in relation to anthropometric measures (weight, height, age, malnutrition scores, weaning status and the economic status of the mother and family etc) in children in the target areas. Linking with surveys on malaria or other health related issues (level of malnutrition, types of diet etc) in target villages to assess aflatoxin exposure is vital. Elsewhere in West Africa (Benin and Togo) a striking association between exposure to aflatoxin in children and both stunting (a reflection of chronic malnutrition) and being underweight (an indicator of acute malnutrition) has been revealed. This emphasizes the need to investigate this question and develop strategies to minimize exposure to mycotoxins in staple foods.

Table 10. Means and ranges for Aflatoxin content from groundnut market samples in Mali, 2006

Market/Product	No. of Samples	Mean (ppb)	Range (ppb)
Groundnut paste			
Kita	22	203	30-1674
Bamako	69	260	5-2914
Kernels			
Bakodjikoroni	22	103	3-914
Banakabougou	72	155	7-2666
Banconi	25	222	4-1150
Daudabougou	24	179	5-2040
Dibida	12	20	5-46
Magnambougou	10	92	2-455
Medine	83	135	3-1800
N'Golonina	16	115	5-536
Sokorodji	21	145	6-1018

BR Ntare, AT Diallo and F Waliyar

Project 4

Producing more and better food from staple cereals (sorghum and millets) and legumes (groundnuts, chickpea and pigeon pea) at lower cost in eastern and southern African (ESA) SAT through genetic improvement

Output 4A: Sustainable regional breeding networks that integrate conventional and biotechnology tools established; and improved germplasm and parental lines of adaptable sorghum, pearl millet, pigeon pea, chickpea and groundnut that are resistant to biotic stresses and meet end user preferences developed and disseminated with associated capacity development annually in ESA

MTP Output Target 2006: At least 10 new groundnut lines with increased rosette virus resistance made available to partners

Output Targets A1: Task networks established to pursue regionalized breeding in response to determined sorghum and millet regional challenges [2011]

Activity A1.1 Enhance capacity on the use of GIS and establish recommendation domains for technology targeting by task networks for sorghum and millet improvement for food security and market needs

Milestone A1.1.1: At least 10 NARS collaborators trained on GIS and other data/information management tools by 2007

Capacity building on tools, methods and strategies to improve crop breeding efficiency: Resources available for agricultural research have fallen sharply in many NARS and in International Agricultural Research Centers. The Sub Regional Organizations, such as ASARECA, are pursuing a regionalized approach to technology development, dissemination, scaling out and information sharing with a major goal of attaining efficiency and cost effectiveness. In order to develop improved sorghum and millet cultivars, the strategy is to establish breeding networks that will address regional crop improvement challenges that cut across the region. This requires a good understanding of the relevant plant adaptations for sorghums and millets in ECA, tools and methods that will help in methodical delineation of recommendation and scaling out domains. The concept of agro-ecological characterization and mapping the crop adaptation areas in the region of interest is a priority that can also support integration of crop improvement and natural resource management issues (IGNRM). ECARSAM and ICRISAT organized a training course for its collaborators on GIS and data and information management. The training course was conducted in Nairobi, Kenya and attended by a total of 22 participants (14 men and 8 women) including breeders, GIS and data management specialists from Kenya, Uganda, Tanzania, Ethiopia, Eritrea, Sudan, Rwanda and Burundi. The main achievements included:

1. Articulation with scientists the benefits to be derived from pursuing a regionalized crop improvement program citing experiences from southern Africa,
2. Exposure of scientists to GIS, and other tools and methods useful in developing strategy for sustainable crop improvement programs,
3. Identification of main production systems for sorghum and millets in ECA in view of biophysical-climatic conditions as well as socio-economic factors such as end use quality requirements in terms of food and market needs, industry needs e.g. food, feed, fodder to determine crop improvement needs, and
4. Clear identification of the environmental factors to which specific adaptation is expressed e.g. highland sorghum, photoperiodic sensitivity and determine studies to inform the crop improvement programs.

This was followed up by a training and refresher course to 10 scientists on biometry and statistical methods, how to organize, manage and analyze biodiversity and molecular data. Emphasis was placed on using Genstat 9.0 and other software's such as Numerical Taxonomic multivariate Analysis System (NTSYS), Arlequin, R.2.2 and Popgene. This training was considered very useful and plans are underway to expand it and include a broader range of scientists in ESA.

MA Mgonja, B Mitaru and E Manyasa

Milestone A1.1.2: At least one new breeding target identified in collaboration with regional networks to address evolving challenges for either sorghum or millet by 2009

Sorghum for bio-ethanol: Fuel prices skyrocketing and concomitant shrinking supplies have triggered efforts in exploring the use of alternative and renewable fuel sources. . One such alternative is bio-ethanol and many developing countries are racing for bio-ethanol production. In recent years sweet sorghum (*Sorghum bicolor* L. Moench) stalks are emerging as viable alternative sources. Comparative advantages for the use of sweet sorghum, status of available technology, challenges and opportunities on the use of sweet sorghum based ethanol have been articulated. NARS partners of South Africa, Zimbabwe and Kenya have shown interest in this area and ICRISAT-ESA envisages working in collaboration with diversified range of partners from these countries. The main focus will be to establish a holistic and integrated system that will look into the sweet sorghum germplasm identification, adaptation testing, linking it with the seed systems and processing technologies. To kick off this new intervention, germplasm have been acquired from ICRISAT India (22 varieties and 45 hybrids) and also from the ESA region for preliminary evaluation for adaptability and also for identification of the most productive varieties. The evaluation has been initiated in 2006/07 season in Kiboko, Kenya and Matopos in Zimbabwe. The expected outcome will be expanded sweet sorghum utilization in the bio-ethanol production to expand livelihood opportunities and environment protection

MA Mgonja and E Manyasa

Milestone A1.1.4: At least one group of NARS working together for pearl millet improvement by 2007

Pearl millet breeders have released a number of improved varieties (11 in ECA and 19 in SADC) that are high yielding potentials and adaptable to the targeted environments. However, production and productivity has remained low due to the continued use of low yielding landraces, poor crop production practices under environments with declining fertility. A proposal was developed by ICRISAT in collaboration with NARS partners from Eritrea, Sudan, Tanzania and Kenya and was funded in 2006 by ECARSAM through the Competitive Grant System. The proposed project is IGNRM oriented and focuses on introducing genetically superior varieties and environmentally friendly integrated production practices to increase production and productivity. By integrating findings on soil and water management, variety improvement, crop protection, farmer seed knowledge, production and delivery systems and environment conservation awareness, the project will be putting to use cumulative findings from related scientific departments and disciplines in the region over periods of time for the benefit of the current pearl millet farming community.

Use of GIS will ensure effective environmental placement of integrated production packages during validation and evaluation to save in both time and resources. Pooling of review and validation findings across partner countries will enhance sharing of scientific findings from these. Inclusion of the farmers and extension personnel will ensure sense of ownership of the findings and sustainability of the technologies.

In preparation for effective implementation of the project, NARS collaborators from Zimbabwe, Sudan, Kenya and Tanzania participated in the International Pearl Millet Training course held in ICRISAT-India in May 2006 to sharpen skill on pearl millet improvement and seed production. The project's implementation period is 2007 to 2009

MA Mgonja and B Mitaru

Output Targets A2: Genetically diverse sorghum varieties tolerant to striga and midge developed in collaboration with NARS participating in breeding networks [2011]

Activity A2.1: Transfer *Striga* resistance in sorghum to elite African cultivars using marker-assisted selection

Milestone A2.1.1: Elite farmer varieties carrying 1 to 3 QTLs evaluated through a participatory approach (2007)

Flanking simple sequence repeats (SSR) markers to the QTLs in marker-assisted backcrossing is vital in transferring *Striga* resistance from the donor cultivar N13 to susceptible farmer preferred sorghum varieties (FPSVs). In this project entitled, "*Arresting the scourge of* Striga *on sorghum in Africa by combining the strengths of marker-assisted backcrossing and farmer-participatory selection*", NARS in the Kenya, Mali, Eritrea and Sudan are being assisted to strengthen *Striga* resistance of farmer-preferred sorghum varieties (FPSVs) through a combination of marker-assisted backcrossing (MAB) and farmer-participatory selection. Near-isogenic FPSVs carrying one to three *Striga*

resistance QTLs are being developed. Simultaneously, studies are being undertaken to enhance the understanding of sorghum seed supply systems and to ensure the effective integration of seven *Striga*-resistant FPSVs into farming systems in Eritrea, Kenya, Mali and Sudan. The extent of outcrossing rates and gene flow are being determined in five selected FPSVs.

So far, 712 plants were genotyped from two backcross generations (BC_1F_1 and BC_2F_1) at the BecA research platform (ILRI; Nairobi, Kenya) using a total of 10 foreground SSR markers and 16 background SSR markers. Genotyping revealed that 256 plants from the second backcross generation (BC_2F_1) were heterozygous for 1 to 3 QTLs.

In Kenya, 43 BC_2F_1 plants were selfed and genotyped to select for segregating homozygous BC_2S_1 plants that have been taken through another selfing generation (BC_2S_2) to fix the QTLs. Plans are now underway to genotype the BC_2S_2 plants, and those confirmed to contain one to three QTLs will then be multiplied and evaluated for *Striga* resistance in artificially infested fields.

The selfing of 73 BC_2F_1 plants in Mali and 103 BC_2F_1 plants in Sudan has been completed and genotyping of the BC_2S_1 is now planned. The segregating homozygous BC_2F_1 plants will be selfed to fix the QTLs. Preliminary studies have revealed some variability in FPSVs with outcrossing rates ranging between 3 and 5%. Initial gene flow studies have shown pollen dispersal for distances of up to 100 m in multiple directions, with a marked decrease after 40 m from the center.

Activity A2.2: Develop Sorghum varieties resistant to Midge through marker-assisted selection.

Milestone A2.2.1: Markers segregating with traits associated with resistance to midge identified and linkage map of the F2 population of AF28 Seredo generated by 2007 (DK, TH)

Sorghum midge (*Stenodiplosis sorghicola* Coquillet), spotted stem borer (*Chilo partellus* Swinhoe) and sorghum shoot fly (*Atherigona soccata* Rond.) are the three most destructive insect pests of sorghum in the ASARECA (Association for Strengthening Agricultural Research in East and Central Africa) region. Midge control in sorghum, therefore, is one of the research priorities in ECARSAM (the Eastern and Central Africa Regional Sorghum and Millet Network), the sorghum and millet network of ASARECA.
Midge resistance in sorghum is location specific and the identification of sorghum genotypes with stable resistance to sorghum midge is therefore hampered by genotype × environment interactions. Identifying DNA markers closely linked to midge resistance loci and using these markers in marker–assisted selection (MAS) will aid breeding for sorghum midge resistance. The objectives of this project are therefore to 1) identify SSR markers that segregate with traits associated with resistance to sorghum midge and 2) initiate MAS towards the development of midge resistant sorghum varieties to improve sorghum production in East Africa. The project aims to map midge resistance QTL in order to identify SSR markers closely linked to these QTL and to initiate the development of sorghum varieties with resistance to sorghum midge through MAS. So far the project, generated a segregating sorghum population for midge resistance using the highly resistant, locally adapted sorghum landrace from Africa AF 28 and Seredo, a high-yielding, drought tolerant, midge susceptible Kenyan sorghum cultivar. The resulting F_1's are now being genotyped in the ICRISAT/BecA lab using SSR markers to confirm their heterozygosity.

D Kiambi, D Hoisington, T Hash, S de Villiers

Output Target A3: Genetically diverse and regionally adapted germplasm and breeding populations of sorghum and millet developed and disseminated [2011]

Activity A3.1: Develop and evaluate traits and end use specific sorghum and millet populations and breeding lines for adaptation to specific environments and resistance to biotic stresses

Milestone A3.1.2: Conventionally bred midge, stem borer and leaf disease resistant lines for sorghum and millets evaluated in advanced trials for yield and adaptability by 2008

Sorghum midge [*Contarinia sorghicola*] is the most widely distributed of all sorghum insect pets. Host plant resistance and time of planting are some of the important components for the management of the pest. Several sources of resistance have been identified, however the levels of resistance vary from location to location. This calls

for localized breeding and testing for sorghum midge resistance. A total of 155 conventionally bred midge resistant lines plus 5 checks were evaluated in a preliminary trial at Alupe, Kenya in the long rainy season 2006 under natural infestation. These lines were crosses between MR No.s 2, 3, 4, 6, 7, 8, 10, 17, 21 22 from ICRISAT India with IS 8613, IS 21016 and AF 28. The trial had two plantings; one early and the other 2 weeks after normal planting to ensure adequate insect pressure for midge damage evaluation. The first planting had a mean score of 3.4 midge damage on a 1-9 scale. 91 entries had scores of 1.0-3.0 midge damage showing high levels of resistance. Eighty eight entries had grain yields between 1.3 and 4.6t/ha and these high yielding lines corresponded to the 91 lines with low midge damage, an indication that at this site midge resistance enhanced grain yield. The second planting had a mean score of 7.4 midge damage (1-9 scale) and 29 lines showed moderate levels of resistance to midge with scores of 4-6 (1-9 scale) and with a grain yield potential of over 1.0 t ha^{-1} (trial mean 0.981 t ha^{-1}). In the second planting, all check varieties except for IS 8613 had grain yield <1.0 t ha^{-1}. Cross MR # 7 x AF 28-2-5-1-1-1 showed the best performance against midge with score of 4.0. These selections will be put into an advanced sorghum midge resistance trial in 2007 and supplied to interested NARS. A small preliminary sorghum midge resistance trial (12 entries) was also sent to Uganda and Ethiopia in April 2006 on their request and results of their evaluation are yet to be received.

MA Mgonja, E Manyasa and J Kibuka

Milestone A3.1.3: Performance and adaptability of the Bristled pearl millet variety ICMV221 (ICMV221 Br) line established by 2010

Development of bristled pearl millet population was initiated in the quest to address the perennial problem of bird damage in pearl millet production. Three bristled pearl millet population cycles were developed from the released variety ICMV 221. The bulked selections from the three cycles (C1, C2 and C3) plus C0 and check varieties KAT PM 1, KAT PM2 and ICMV 221 were evaluated for their performance at Kiboko and Kampi ya Mawe (Kenya) in 2005/06. Results showed that bristle density and length increased with increase in cycle with cycles 2 and 3 having the highest percentage of medium and long bristles (54.8, 13.6 % and 55.1, 18.8% respectively). Grain yields for C0 to C3 ranged from 0.474 to 0.939 t ha-1 compared to 1.365 t ha- for ICMV 221 with a decrease in yield with increase in cycle. However, at Kampi ya Mawe, C1 gave better grain yields (0.570 t ha^{-1}) compared to C2 and C3. But ICMV 221 (0.715 t ha^{-1}) still out-yielded all the population cycles. This trial has been established at Kiboko and Kampi ya Mawe in 2006/07 rainy season to evaluate the cycles for bird damage and results will be reported in 2007.

MA Mgonja, E Manyasa and E Muange

Output target A4: Improved sorghum and millet varieties and hybrids with end use preferred plant and grain traits developed, evaluated and disseminated [2011]

Activity A4.1: Develop and evaluate a wide range of varieties and hybrids that are adaptable and meet end use requirements

Milestone A4.1.4: Regional sorghum hybrid / variety trials with brown/red and white seeds evaluated annually in the short/medium season environments

A regional sorghum hybrid trial comprising of 8 hybrids developed at ICRISAT-Nairobi and 4 from ICRISAT-Bulawayo plus 4 parental checks (KARI Mtama 1, Gadam Hamam, Macia and Kiboko local 2) were evaluated at Kiboko and Alupe (Kenya) in long rainy season of 2006. Both trials were under rainfed conditions. The hybrids performed better than the checks at Kiboko with the best hybrid IESH 22019 (2.987 t ha^{-1}) yielding 126% better than the best parental check Gadam Hamam (1.319 t ha^{-1}). Hybrids IESH 22019, IESH 22002, IESH 22010, IESH 22011, SDSH 409, IESH 22012 and IESH 22005 attained grain yields above 2.0 t ha^{-1} (trial mean 1.885 t ha^{-1}). Similarly at Alupe also, the hybrids out-yielded the parental check varieties. The best hybrid IESH 22021 yielded 1.716 t ha^{-1} compared to the best parental check (Gadam Hamam) which yielded 1.420 t ha^{-1}. The regional trial was also sent to Tanzania, Zimbabwe and Malawi in the rainy season 2006/07 and results will be reported in 2007. A new hybrids test cross evaluation with 203 entries was also carried at Kiboko and Alupe in the long rainy season 2006 using hybrids developed using A/B/R lines from ICRISAT-India. Out of the 203 test hybrids, 25 at Kiboko and 14 at Alupe with good fertility restoration and agronomic potential were selected. The 25 selected hybrids at Kiboko have been put into a preliminary hybrid trial in the 2006/07 rainy season whereas the 14 selections made at Alupe will be planted in long rainy season 2007.

Forty nine advanced and 50 preliminary sorghum lines developed from crosses between bold grain B lines from ICRISAT- Bulawayo and adapted local and improved varieties to improve grain quality of the adapted varieties were evaluated at Alupe in the long rainy season 2006. Yields of the 11 best test lines in the advanced trial ranged from 2.292 to 2.736 t ha^{-1} with 100 seed mass >2.9gm. The best check variety (IESV 93036 SH) yielded 2.222 t ha^{-1}. The best lines will be evaluated at more sites and supplied to interested NARS. In the Preliminary yield trial, yields in the best 8 test lines ranged from 1.896 to 2.469 t ha^{-1} and were above the best check variety IESV 93036 SH (1.854 t ha^{-1}).

MA Mgonja, E Manyasa, J Kibuka and E Muange

Activity A4.2: Facilitate information, knowledge and product sharing among NARS partners of available improved sorghum and millets populations and germplasm

Milestone A4.2.1: At least 6 NARS are annually provided with breeding materials for further evaluation

The ICRISAT breeding program provides breeding materials as semi-and-finished products for further selection and testing by NARS collaborators. In 2006, breeding materials were provided to six NARS partners at their request for inclusion in their respective evaluation programs.

Breeding Materials supplied to Collaborators in 2006 (from ESA, Kenya)

Country	Crop	Organization	No. of varieties	No. of samples	Category	Quantity (kg)
South Africa	Sorghum	CSIR	20	20	Varieties/Advanced breeding lines	2.0
Kenya	Sorghum	Moi University	12	12	,,	1.2
Kenya	Sorghum	KEPHIS	54	54	,,	0.54
Uganda	Sorghum	SAARI/NARO	12	36	,,	0.12
Ethiopia	Sorghum	IAR	12	36	,,	0.12
Kenya	,,	KARI-Embu	26	75	,,	1.8
Kenya	,,	Western Seed Company	7	14	A/B lines	0.4
Botswana	,,	MOA-Research	1	1	Breeder	0.1
Sudan	Finger millet	Research	337	409	Germplasm/varieties	2.6

The materials are shared under the Material Transfer Agreement.

MA Mgonja and E Manyasa

Milestone A4.2.2: At least one improved sorghum and /or pearl millet cultivar released every two years in any ESA country from 2007 to 2011

Improved sorghum and millet varieties identified for release in Malawi: NARS collaborators continue evaluation of improved germplasm to identify the most adaptable and acceptable materials for release and cultivation by farming communities. The Malawi Agricultural Technology Clearing Committee (ATCC) met at Chitedze Research Station and considered several technologies including sorghum and millet materials derived from ICRISAT germplasm. The National sorghum and millet breeder submitted for consideration for release two sorghum varieties, one pearl millet variety and two pearl millet hybrids. Two sorghum varieties (Sima and SV2) were given pre release status based on the station performance data. SV2 is released in Zimbabwe (1987) and Sima is also released in Zambia (1989). Initially thought to be restricted in adaptation to medium long season southern Zambia, analyses of available multi environment trial data for the SADC region indicated that Sima is one of the varieties that possessed characteristics that Zambian farmers prefer; expressed highest mean yields (327gm/m2) and positive responses to favorable environments (β=1.0) and was also found to be relatively biologically more stable with a CV=70.6%. Sima is one variety that based on its performance and stability stand even a higher chance for regional registration especially now that it has been pre-released in Malawi, and Zimbabwe is also considering its release.

The pearl millet variety SDMV 90031 was granted full release and was described to be superior in yield and an excellent complement to the other two released varieties (Nyankhombo and Tupatupa). The two pearl millet hybrids were not considered favorably due to the inadequacy of the seed system to handle hybrid seed production. Current efforts will target on farm testing to generate data to facilitate full release of the two sorghum varieties Sima and SV2

MA Mgonja and S Kudita

Activity A4.3: Develop and use alternative seed delivery models to efficiently disseminate improved sorghum and millet cultivars for adoption

Milestone A4.3.1: At least 3 NARS are annually availed with sorghum and millet basic seed for further multiplication 2007-2010

Sorghum and pearl millet basic seed: The pearl millet variety ICMV 221 has been released in three ECA countries-Kenya, Uganda and Eritrea. 20 kg breeder seed of pearl millet variety ICMV 221 were sent to Uganda through the DANIDA program for multiplication. The seeds were multiplied and distributed to farmers in northern Uganda. The NARS in Zimbabwe, South Africa and Mozambique have also been supplied with basic seed for sorghum and pearl millet to the tune of 10-100kgs for further multiplication and use in the Challenge Program on Water for Food Project 1 on integrated crop varieties with soil fertility and water management to increase productivity and profitability for the Limpopo basin. Seed production was done mainly in Zimbabwe and the land areas involved were for Sorghum variety Macia (2.2ha), Sima (0.32ha); and Pearl millet variety PMV3 (0.32). Others included sorghum varieties SV3, SV4 and Chokwe; pearl millet varieties OK 1, PMV2, Kuphanjala 1, Kuphanjala 2 and Changara in relatively smaller quantities.

MA Mgonja and Sakile Kudita

Milestone A4.3.2: At least 6 NARS are availed with improved germplasm and participate in regional cultivar evaluation annually from 2007

ICRISAT in collaboration with ECARSAM has re-instituted regional cultivar evaluation as a platform for germplasm exchange, improve efficiency in identification of suitable varieties with broader adaptability and sharing data to support cultivar releases. The regional trials are composed based on agreed breeding zones and end use targets.

Table: Sorghum and Millets Regional trials Composed and Distributed - 2006/07

Country	Crop	Organization	Trial name	No. of varieties	No. of samples	Category
Tanzania	**Sorghum**	DARD	Regional sorghum hybrid trial	26	106	Varieties/ Advanced breeding lines
			Sorghum Photoperiod Trial	10		
Mozambique	,,	Research	Sorghum Photoperiod Trial	10	20	Varieties/ landraces
Malawi	,,	ICRISAT	Regional sorghum hybrid trial	16	48	Breeder (hybrids)
Mali	,,	ICRISAT	Sorghum Photoperiod Trial	7	7	Varieties/ landraces
Tanzania	**Finger millet**	DARD	Regional finger millet trial	16	96	Germplasm/varieties
Malawi	,,	ICRISAT	Regional finger millet trial	16	48	Germplasm/varieties
Kenya	**Pearl millet**	KARI-Embu	Regional pearl millet trial	16	48	Varieties
South Africa	,,	Research	Regional pearl millet trial	16	48	Varieties
Tanzania	,,	DARD	Regional pearl millet trial	16	96	,,
Namibia	,,	Ministry of Agric., Water and Forestry	Regional pearl millet trial	16	48	,,

Sorghum and Millet Variety evaluation, release and seed multiplication in Eritrea: ICRISAT's research collaboration in Eritrea started in earnest in 1995 when over 600 sorghum and 98 pearl millet lines were introduced from ICRISAT-Nairobi and evaluated at Shambuko (western lowlands), Halhale (mid-highlands) and Shieb (eastern lowlands). On-farm testing of farmer selections from these trials began in 1997 and one pearl millet variety [ICMV 221(Kona)], 5 sorghum varieties [ICSV 210 (Bushuka) and PP 290 (Shambuko) for western lowlands; 89 MW 5003 (Laba) and 89 MW 89 5056 for eastern lowlands; IS 29415(Shiketi) for mid-highlands] were released in the year 2000.

During the 2004/05 season the Ministry of Agriculture and NARI produced: 163 t of PP 290, 63 t of ICSV 210, 119 t of Gadam Hamam, 15.8 t of ICMV 221 and 6.6 t of Hagaz. All of this seed was distributed to farmers in the western lowlands in 2005. In the western lowlands, 57 farmers had been contracted to produce seed and were growing over 110 ha of sorghum variety ICSV 111 IN. One farmer by the name Ato Mebrahtu Asfaha grows 2000 ha of both seed and grain sorghum under irrigation at Tesseney. Farmers in Eritrea have adopted released varieties and a number of new lines are being multiplied in readiness for release and distribution to farmers during the 2006/07 season. With these new varieties, farmers are now achieving grain yields of over 1.5 t/ha compared to 0.8 t/ha from local varieties

In the mid-highlands of Eritrea, IESV 92029 DL (bred at ICRISAT-Nairobi) has been earmarked for release and over 7 ha were under seed crop at Halhale. In the western lowlands, ICSV 111 IN has been earmarked for release and over 110 ha were under seed crop at Shambuko. The ICRISAT ESA breeding program have plans to carry out a Sorghum and millets adoption and impact study in Eritrea during the 2007 season.

MA Mgonja, B Mitaru and E Manyasa

Activity A4.3: Develop and use alternative seed delivery models to efficiently disseminate improved sorghum and millet cultivars for adoption

Milestone: A4.3.3: Business plans developed for establishment of Seed Enterprise Enhancement and Development Services [SEEDS] in at least three countries of ESA by 2007

Seed Enterprise Enhancement and Development Services: Farmers in many countries in Africa are unable to obtain high quality seed of improved publicly developed varieties due to a lack of effective commercial seed production distribution networks. There is a need to facilitate the introduction of both commercial and publicly developed varieties through distribution networks in a sustainable manner. This will expand farmers' selection and allow them to choose what best suits their requirements.

The African Seed Trade Association (AFSTA) and the ICRISAT program for the Sustainable Commercialization of Seeds in Africa (SCOSA) have proposed the establishment of Seed Enterprise Enhancement and Development Services (SEEDS) to support the development of small and medium-sized seed companies. These will include independently run and financially sustainable foundation seed enterprises (FSEs) that will market foundation seed of publicly developed varieties for use by seed companies without their own breeding programs. They will also provide access to seed storage and processing facilities to existing and emerging seed entrepreneurs. SEEDS will provide technical and business development support to seed entrepreneurs who will become the customers of the FSEs, ensuring their sustainability.

In 2006 national teams from Ethiopia, Kenya, Tanzania, Uganda, Malawi, Mozambique, Zambia, Zimbabwe, Botswana and Angola were trained in business plan development and then supported to hold national consultation meetings needed to solicit input into their respective business plans. An Excel template was developed to assist in the preparation of financial projections, and these will be used to solicit for funding from interested development investors in 2007.

RB Jones

Output Targets A6: High yielding farmer and market-acceptable groundnut varieties developed

Activity A6.1: Identify through PPB and introgress groundnut germplasm for yield components farmer/market preferences and adaptation with special reference to incorporation of drought tolerance in short duration Spanish types with fresh seed dormancy

Milestones A6.1.1: At least 3 farmer/market preferred varieties incorporating drought tolerance in short duration background? released in 2–3 ESA countries (2010)

Breeding material incorporating Adaptation Traits: Groundnut breeding in ESA is targeting the following traits for adaptation, farmer and market preferences

1. Fresh seed dormancy in short duration varieties
2. Large seeds in short duration varieties
3. Large seeds and high yield in medium to long duration varieties
4. Grading qualities as measured retention above sieve 19 compared to CG 7 and Chalimbana.

Fresh seed dormancy is essential among early maturing varieties of groundnut to avoid sprouting in field. Advance progenies with fresh seed dormancy from the segregating nurseries. Eight progenies from 4 rosette nurseries were identified. It was interesting to note that fresh seed dormancy was not found among the Dormancy and Rosette resistance - F7s in spite of the fact that the nursery originated from crosses involving parents with fresh seed dormancy. This finding will need further investigation.

Elite Short Duration Drought Tolerance and Dormancy Trial: the best two entries were ICGV-SM 98511 and ICGV-SM 86021 with grain yields 102 – 115% of JL 24 (1.7 – 2.0 t ha^{-1}). None of the entries were better than ICGV-SM 99568 (2.2 t ha^{-1}) which is both early maturing and resistant to rosette. However the rainfall in Southern Malawi (Drought screening site) was higher than normal during the season, providing a good opportunity for fresh seed dormancy screening. Sprouting was not a problem at this site – indicative of acceptable levels of fresh seed dormancy

Elite Drought Resistant Groundnut Variety Trial: ICGV-SM 03535, ICGV-SM 03521, ICGV-SM 03502 and ICGV-SM 03510 had grain yields ranging from 2.4 t ha^{-1} to 3.3 t ha^{-1} (153–208% superior in yield as compared to the standard check JL24).

Large seeded germplasm and grading qualities are highly sought after in the groundnut confectionary market. From the Short duration large seeded nursery, the best three lines were ICGV-SM 00528, ICGV 94536 and ICGV-SM 00503 with yields 111 – 148% of the standard check JL 24 but acceptable confectionary size nuts. From the Elite High Yield and Quality Groundnut Variety Trial (Spanish), four lines ICGV-SM 03552, ICGV-SM 03564, ICGV-SM 03573 and ICGV-SM 03576 had grading qualities and yield similar or better than the newly released variety ICGV-SM 99568. Yield was 2.6 t ha^{-1} as compared to 1.9 t ha^{-1} for ICGV-SM 99568 and are resistant to rosette disease.

From the Elite High Yield and Quality Groundnut Variety Trial (Virginia), ICGV-SM 01721 was identified with 100 seed grain weight significantly higher than CG 7 but the yield was only 93% that achieved from CG 7. Four lines with grading qualities and yield similar to CG 7 and Chalimbana with combined advantage of rosette resistance were identified. These are ICGV-SM 02707, ICGV-SM 02715, ICGV-SM 02724, and ICGV-SM 01731. All these lines had grain yields significantly higher than that of the check Chalimbana.

New Variety Release(s)
ICGV-SM 99568 for the rosette constraint in Malawi under the local name Chitala, and ICGV 94297 in Zimbabwe under the local name Ilanda.

ES Monyo

Activity A6.2: Develop and evaluate diverse groundnut breeding populations, lines and varieties while developing the capacity to screen for GRD, foliar disease resistances and aflatoxin contamination in the NARS

Milestone A6.2.1: Infector row technique for screening of GRD resistance established and operational with at least one NARS in ESA by 2008 (ESM)

2006 - Targeted milestones:

- Diverse populations of groundnut developed through conventional and marker assisted selections
- Local germplasm deployed in the breeding program for incorporating specified traits (e.g. adaptation, yield grain type and disease resistance)

Diverse breeding populations, lines and varieties and capacity to screen for GRD and foliar diseases (rosette, ELS, LLS, and rust). The infector row technique is currently in use at the ICRISAT Chitedze Research Centre with Malawi NARS.

The rosette disease pressure was excellent. There was 100% infection in the spreader rows and susceptible checks. The observed resistant progeny selections above were therefore a true reflection of genetic resistance in the selected progenies.

From 16 nurseries ranging from F4 – F6 in 1003 progeny rows, a total of 636 plants were identified for generation advance through single plant selection. Out of these we identified 58 single plant selections from 8 of the nurseries with 0% rosette incidence (9% of total) and an additional 66 plants with rosette incidence ranging 1 - ≤ 20%.

From seven F7 nurseries with 468 progeny rows, a total of 323 were selected for promotion to checkrow trials, out of which 91 had 0% rosette incidence.

Most of the inter-specific derivative progenies exhibited susceptible reaction to ELS. Even the 13 selected single plants had relatively higher ELS than many of the selections from other nurseries. This finding has implication on the nature of resistance and inheritance of ELS from wild *Arachis* that may need further investigation.

There are excellent progenies both in Check row trials and in F7 that combines ELS and rosette resistance. As seen in the table above, our efforts of pyramiding these two constraints are starting to pay off.

Good progress has been made in identification of ELS resistant progenies received from ICRISAT-Patancheru. From an initial nursery of 443 F2 planted in 2004, we have identified 80 progenies with very good levels of resistance to ELS for promotion to F4 families. In pyramiding ELS resistance genes, 240 F5 single plant family progenies have been selected for generation advance to F6 in ELS x ELS crosses and 48 single plant family progenies identified in F4 combining ELS resistance with confectionary market traits.

Achievements on single plant selection program for segregating breeding populations in the Rust and LLS nursery: There were three segregating population nurseries:

- From 104 F6 Rust and LLS Resistance screening nursery, 81 families were identified for generation advance
- From 1673 F7 Rust and Rosette Resistance nursery a total of 408 families were selected to make two sets of checkrow trials each comprising 204 entries
- From 216 entries of the checkrow trial, 62 lines were identified to develop Preliminary Rust and LLS Trial each consisting of 31 entries plus checks (36 entry trials)

The disease pressure for 2006 was low hence most of the selections were based on yield and adaptation to the Low altitude hot humid environment of the Malawi Lake Shore.

ES Monyo

Lead NARS crop improvement Networks for Groundnut Research in ESA:
Through the McKnight Foundation that funded a project on "Developing short-and medium-duration groundnut varieties with improved yield performance, acceptable market traits and resistance to foliar diseases". This project

will be implemented through research task networks in Tanzania and Malawi based on competitive advantage for the researchable constraints while building capacity to tackle these constraints at national level. In particular Tanzania's Naliendele Research Station will focus on Rust resistance screening while Chitedze in Malawi will focus on Rosette and ELS.

ES Monyo

Output Target A7: Adoption rates of improved farmer and market-acceptable groundnut varieties and production technologies enhanced

Activity A7.1: Enhance institutional innovations to improve access of the poor to good quality seeds of improved high yielding adapted groundnut varieties and conduct training of trainers program on seed production techniques

Milestone A7.1.2: At least 1 ton breeder seed of 3 released farmer/market preferred varieties in ESA produced annually as source for foundation seed for collaborating NARS and other Partners 2007-2011

Institutional Innovations to improve access of the poor to good quality seed: Efforts to promote adoption rates of farmer and market acceptable varieties were linked to institutional building through the Challenge Program on Water for Food. Seed for implementation of activities to initiate crop water productivity on-farm trials for the 2005-06 season was produced and supplied to partners as follows:

- Seed enough to implement 96 mother trials and over 600 baby trials in Zimbabwe; 9 mother trials and 19 baby trials in Mozambique – (groundnuts, sorghum and pearl millet).
- Varieties of the various crops important in the basin for use with the crop water productivity studies have been documented as follows: Sorghum (Macia, SV1, SV2, SV3, SV4, Sima and Chokwe), Pearl Millet (Okashana 1, PMV2, PMV3, Kuphanjala-1, Kuphanjala-2 and Changara), Maize (ZM 421, ZM 403, and ZM 521) and groundnuts (Jesa, Nyanda, Ilanda, Mwenje, JL 24, Sellie, Nematil). Concerted efforts for seed production were concentrated in a few preferred varieties from the list – particularly Macia for sorghum, PMV3 for pearl millet, Nyanda and Nematil for groundnuts, ZM 421 and ZM 521 for maize.
- During 2006–07 the following seed production activities were implemented by ICRISAT and NARS in Zimbabwe under off season for the 2006-07 trials: Sorghum variety Sima 0.32 ha, Macia 2.2 ha, Pearl Millet variety PMV 3, 0.32 ha and groundnut variety Mwenje 0.8 ha at Chiredzi Research Station.
- The following varieties and quantities of groundnut seed were produced for CPWF-CP1 on-farm trials and demonstrations: Nyanda; 650kg, ICGV-SM 01513 200kg, ICGV-S, 99541 97kg, ICG 12991, 900kgs. A total of 440 kgs was sent to Mozambique for trials.

Milestone A7.1.1: At least 5 kg nuclear seed of each of 15 varieties in Regional Trials produced annually as source for breeder seed and entries for collaborative trials with NARS in ESA 2007 – 2011

Other non CPWF-CP1 seed production efforts resulted in the following achievements;

- 2–5 kg nuclear seed for 129 varieties of groundnuts in Advanced and Elite Trials produced
- 2203 kg breeder seed of the Aphid resistant variety ICG 12991
- 3759 kg breeder seed of the groundnut rosette virus resistant variety ICGV-SM 90704

ES Monyo

Milestone: A7.1.3: One new institutional arrangement for supply of legume seed tested in Kenya in 2006

New institutional arrangement: Analysis of groundnut value-chains has identified market opportunities for farmers producing surplus groundnuts, but the variable quality and limited supply of marketable surpluses constrains their ability to attract premium prices. Traditional formal seed production schemes result in high seed costs resulting in limited seed demand through such channels rendering them unsustainable. In 2006 a new institutional arrangements was designed and tested whereby a private seed company marketed improved groundnut foundation seed to organized groups of seed farmers identified by the communities in which they live. These seed farmers were then lined to a farmer owned company to market seed to other farmers in these and other communities where these crops were being promoted. Preliminary results suggest that this institutional arrangement builds upon the strengths of informal seed supply systems that most smallholder farmers rely on, but enhances the choice of varieties available

to such farmers from research, and that this leads to increased productivity and improved quality on the part of buyers.

RB Jones

Output target A8: Adapted germplasm of pigeon pea with enhanced productivity and desirable traits [2010]

Activity A8.1: Develop genetically enhanced and regionally adapted pigeon pea germplasm

Milestone A8.1.1: At least 3 high-yielding medium-duration pigeonpea cultivars adapted to the ecological and cropping systems in southern Africa developed by 2009

Photoperiod sensitivity in both medium- and long-duration pigeonpea was one of the limitations for increasing productivity of pigeonpea particularly around equator (10^{o} to 20^{o} north and south) areas in ESA. In such areas, flowering and maturity are delayed thus rendering the crop prone to terminal drought stress and winter frost. Newly developed early to medium-duration (photoperiod-insensitive) cultivars developed for southern Africa were evaluated at Chitedze Research Station (13^{o}59' S and 33^{o}44' E), Malawi during the 2005/06 cropping season. Several traits including the number of days to 50% flowering (50%DF), number of days to 75% maturity (75%DM), grain size (100-GW) and yield were measured. There was variability in 50%DF among the cultivars ranging from 83 d (ICEAP 01514/15) to 112 d (ICEAP 01160/15). In comparison, the preferred and popular local check variety (Mutawajuni) and commercial cultivar (Royes) required 83 d and 119 d respectively to attain 50%DF. Cultivar ICEAP 01514/15 matured significantly ($P<0.05$) earlier (153 d) than both Royes (173 d) and Mutawajuni (172 d). Royes obtained the smallest (100-GW = 13.1 g). However, 100-GW of the improved cultivars ranged between 13.6 - 15.5 g. The highest grain yield (3.0 t/ha) was observed for the cultivar ICEAP 01480/32 compared with 1.0 t/ha attained by Royes. Similarly, cultivar ICEAP 01514/15 obtained 2.9 t/ha.

SN Silim and E Gwata

Milestone A8.1.2: At least 3 early short-duration cultivars that can escape terminal drought stress in ESA developed by 2009

Some parts of ESA, such as the eastern in Kenya, experience a bi-modal rainfall pattern with the first season lasting only three months (November to January). Short-duration pigeonpea varieties that provide flexible options for pigeonpea farmers, are ideal for these conditions. Evaluation of determinate short-duration types was conducted at Kiboko Research Station in eastern Kenya. Six lines averaged >1.0 t/ha compared with 0.5 t/ha obtained for the check cultivar ICPL 87091. On average, the cultivars required 87 d to achieve 50% flowering. ICEAP 01134 developed the largest grains (100-grain weight = 14.1 g) compared with the small (100-grain weight = 12.3 g) observed for the check cultivar. In the advanced variety trial, 14 experimental lines averaged 1.6 t/ha. The genotype ICEAP 01275 obtained the highest grain yield (1.97 t/ha) which was 25% more that the yield observed for the check cultivar ICPL 90050. In another field trial conducted at Ilonga (Tanzania), ICEAP 00624 attained the highest (1.24 t/ha) and required 57 d to achieve 50% flowering.

One of the major drawbacks in our short-duration germplasm is its susceptibility to insect pests particularly pod borers and pod suckers. Currently, pesticides are necessary in the management of short-duration cultivars in ESA.

SN Silim and E Gwata

Activity A8.2: Widen genetic base of pigeon pea with enhanced resistance to fusarium wilt

Milestone A8.2.1: Collection of pigeon pea from Tanzania and Mozambique screened for resistance to fusarium wilt by 2008

Preliminary evaluation of germplasm (previously collected from Tanzania and Mozambique) for resistance to *Fusarium* wilt was conducted in a wilt-sick plot at Kiboko (Kenya).There were 12 (one from Mozambique and 11 from Tanzania) accessions with medium to high resistance levels. Further evaluation of these accessions is in progress. Similarly, the elite medium duration cultivars developed for areas away from the equator were also included in the wilt-sick plot for the 2006/07 season. Although genetic material showing resistance in this wilt-sick plot at Kiboko is expected to show wide amplitude resistance elsewhere in ESA, these preliminary results should be

interpreted with caution. Differential host responses to the disease have been demonstrated in previous studies in ESA. Nevertheless, sources of resistance to the disease in locally adapted material will be useful in pigeonpea breeding programs in the region.

SN Silim and E Gwata

Milestone A8.2.2: Pigeonpea breeding populations derived from resistant x adapted germplasm developed by 2009

Crosses between the resistant germplasm (originating from both Mozambique and Tanzania) and adapted cultivars in both medium- and long-duration pigeonpea types (and their reciprocals) were initiated at both Kiboko and Kampi ya Mawe. The major target is to introgress the resistance to wilt particularly in the early medium-duration types developed for areas away from the equator. Likely, some of the germplasm lacks adequate levels of resistance to the disease since photoperiod insensitivity was the main focus of the initial improvement effort. F_2 populations derived from a landrace (popular in Malawi, Mutawajuni) x resistant cultivars (ICEAP 00040 and ICEAP 0576-1) were raised in the field at Kabete, Kenya. The segregating populations will be shared with the prospective national programs to facilitate selection in target production areas. The selection criteria from this batch of crosses will include resistance to the wilt and insect pests as well as white (cream) seed coat desirable in the market.

SN Silim and E Gwata

Milestone A8.2.3: Selection and evaluation of resistant pigeonpea germplasm initiated by 2011

Medium- and long-duration pigeonpea cultivars require more than three months to mature in ESA. This makes these types prone to the wilt disease. All newly developed germplasm in both maturity groups needs to be evaluated in high-disease pressure conditions such as provided by wilt-sick plots at Kiboko (Kenya) and Ilonga (Tanzania). The selection process was initiated at Kiboko where the improved resistant long-duration cultivars (ICEAP 00040, ICEAP 00020, ICEAP 0576-1 and ICEAP 00933) were identified originally. Moderate levels of wilt resistance were observed for the medium-duration cultivars ICEAP 00554 and ICEAP 00557. It is desirable to establish similar testing facilities in the other countries such as Mozambique where the disease is prevalent. In addition, research effort directed toward identification of pathogenic races of the wilt disease across the region would be merited.

SN Silim and E Gwata

Milestone A8.2.4: Report/article on performance of resistant pigeonpea germplasm in ESA published by 2011

A multi-location evaluation of elite pigeon pea cultivars previously identified as resistant to fusarium wilt was conducted in three countries (Kenya, Malawi and Tanzania). The agronomic performance of the germplasm was communicated (*Journal of Plant Pathology 154: 62-64)* in 2006. The long-duration cultivar ICEAP 00040 showed stable resistance across the region. It was released for commercial production in both Malawi and Tanzania.

SN Silim and E Gwata

Activity A8.3: Test and select improved pigeonpea varieties for provision to NARS and further dissemination

Milestone A8.3.1: Evaluate, promote and disseminate through participatory/ on-farm methods, at least 6 newly improved pigeonpea cultivars for production in ESA by 2009

Seed of elite pigeonpea cultivars was disseminated to partners in southern Africa for the 2006/07 cropping season. In Malawi, where the crop is grown mainly in the southern region, on-farm evaluation in both the central and northern regions was initiated for the 2006/07 cropping season. In Tanzania, seed was disseminated to new pigeonpea areas (Karatu and Mbulu districts). Our partners (Diocese of Mbulu-CRS and the Selian Agricultural Research Institute) facilitated the expansion of the area under pigeonpea in Tanzania. In addition, a group of 16 media practitioners representing various international agencies (such as BBC East Africa, Kenya TV Network, Tanzania Newspapers) participated in a three-day field visit to Babati, Karatu and Mbulu (Tanzania) aimed at high-lighting the impact of pigeon pea in ESA as well as the critical role of ICRISAT in farmer-driven research. The wilt resistant cultivars (particularly ICEAP 00040 and ICEAP 00053) developed by ICRISAT were adopted widely in the area. In Kenya, pigeonpea was promoted through field days held in Makueni district.

SN Silim and E Gwata

Milestone A8.3.2: Promote participatory community-based methods for seed production of farmer-preferred pigeonpea cultivars in at least 3 countries in ESA by 2009

Farmer-training in the agronomy of pigeonpea is a continuous process as new farmers adopt the crop. During the pre-season (in 2006), farmers in Makueni district (Kenya) were trained in various aspects of pigeonpea production including the methods and techniques for improving seed quality. In Malawi, the on-farm demonstration plots established for the 2006/07 season will be used also for training farmers in community based seed production. It is anticipated that the farmers (particularly those new to the crop in the central and northern areas) will be trained in basic seed production techniques such as rouging off-types using phenotypic characters at both vegetative and reproductive phases, scouting for pests, crop isolation, seed cleaning and storage.

SN Silim and E Gwata

Output Target A9: High yielding and adapted chickpea germplasm for small-holder farmers identified and disseminated in ESA [2011]

Activity A9.1: Identify genetically enhanced chickpea germplasm adapted for production in ESA

Milestone A9.1.1: At least 30-40 high-yielding advanced chickpea lines/breeding lines (from ICRISAT Patancheru) evaluated in regional field trials in eastern and southern Africa by 2008

Evaluation of chickpea lines for adaptation to prevailing agro-ecological conditions in ESA was conducted in Kenya and Mozambique. The field trials consisted of both *desi* and *kabuli* chickpea types. In Chokwe district (Mozambique) three *desi* genotypes obtained high (>3.0 t/ha) grain yield. The highest (3.6 t/ha) grain yield was observed for ICCV 97126. The control cultivar (Ngara Local) attained 2.5 t/ha. The grain size (as measured by 100-grain weight) of the *kabuli* types ranged between 29.1 – 43.6 g. In contrast, the maximum grain size among the *desi* types was 30.g. At Kabete Research Station (Kenya), the *kabuli* cultivar ICCV 92311 attained 4.2 t/ha compared with 2.6 t/ha for Ngara Local. Eight *desi* and 12 *kabuli* cultivars achieved >3.0t/ha. It is desirable to extend the field evaluation of chickpea to other countries in ESA (Malawi, Tanzania and Zimbabwe) partly because of the genotype x location interaction observed in the trials.

SN Silim and E Gwata

Milestone A9.1.2: 10-15 high-yielding desi and kabuli type breeding lines identified in at least 2 ESA countries (Kenya and Mozambique) by 2009

In Kenya, 5 *kabuli* cultivars showed consistently high (>3.0 t/ha) in the previous three consecutive cropping seasons. In comparison with *desi* types, the *kabuli* types are more lucrative on the international markets. Nevertheless, significantly more *desi* cultivars (in the current germplasm) performed equally well thus providing the local farmers wider options. Similarly, in Mozambique, 80% of the high-yielding elite cultivars were of the *desi* type. There were no significant differences in maturity duration between the two chickpea types at either location indicating that the *kabuli* germplasm evaluated in these trials is adapted to tropical conditions in ESA. Historically, the *kabuli* types are adapted to cool temperate conditions. Therefore *kabuli* types adapted to tropical conditions as represented by Kenya and Mozambique were identified successfully. Probably, the next phase of the research effort should evaluate these new cultivars on-farm in order to facilitate their adoption as well as seed dissemination.

SN Silim and E Gwata

Activity A9.2: Identify chickpea germplasm with drought avoidance traits

Milestone A9.2.1: A sub-set of chickpea reference collection (from ICRISAT Patancheru) with selected root traits evaluated in regional field trials in at least 2 countries in eastern and southern Africa by 2010

The work under this activity is anticipated to begin in 2007 pending availability of funding. Preparation for the work is in progress.

Activity A9.3: Share seed of improved pigeonpea and chickpea germplasm with drought tolerance with partners in ESA

Milestone A9.3.1: Nucleus/breeder seed of at least 10 pigeonpea and 20 chickpea cultivars/ advanced lines available for sharing with NARS multiplied by 2009

In 2006/07 cropping season, the production of breeder seed was increased to include two locations (Kampi ya Mawe and Kabete Research Stations) in Kenya. For pigeonpea, it is necessary to cover the crop with nets during the flowering period of the crop to avoid cross pollination. Production of breeder seed of five wilt resistant and eleven pigeopea cultivars is in progress at Kabete and Kampi yaMawe respectively. Breeder seed of 13 chickpea advanced lines was also produced in 2006 at Kabete. Samples of the breeder seed will be stored for medium-term in the mini-gene bank at ICRISAT-Nairobi and the remainder will be shared with partners to produce foundation or certified seed.

SN Silim and E Gwata

Milestone A9.3.2: Seed of improved pigeonpea and chickpea germplasm disseminated to at least 4 NARS/ countries in ESA by 2008

The high level of out-crossing (30%) in pigeonpea poses a challenge to farmers in terms of maintaining cultivar purity. Therefore it is necessary to disseminate high quality seed to national programs for multiplication and distribution to farmers. Samples of clean breeder or foundation seed were shared with partners in Malawi, South Africa, Sudan, Tanzania, Uganda and Zimbabwe.

For chickpeas, the national programs (in Eritrea, Kenya and South Africa) as well as other partners (Catholic Relief Services and Egerton University) received seed. Requests for seed have also been received from Rwanda and Zimbabwe. In Mozambique, adequate stocks of high quality seed are available to both the national program and NGOs. It is evident that the demand for seed of these two legumes in ESA has increased significantly necessitating the legume enhancement program (at ICRISAT-Nairobi) to respond positively and ensuring availability of the germplasm.

SN Silim and E Gwata

Milestone A9.3.3: Farmers' access to legume seed in at least two ESA countries enhanced through collaboration with at least one public/private partnership (NARS/NGO/Private Seed Company by 2010

Collaborative efforts in seed multiplication with the private sector were initiated in at least two countries in ESA. In Kenya, a private seed company (Leldet Ltd.), with assistance from the Kenya Plant Health Inspectorate Service, agreed to undertake multiplying seed of ICRISAT chickpea (four) and pigeon pea (three) cultivars. Similarly, in Tanzania a seed company (Rotian Seed Company) initiated seed multiplication jointly with ICRISAT. The company is multiplying two cultivars (ICEAP 00040 and ICEAP 0053) in particular which are in high demand in Tanzania. The seed requirement for the two cultivars already exceeds 6.0 t in Babati district.

The partnerships with private companies are expected to rise with the increase in demand for legume seed in the region. Apart from providing the seed, ICRISAT provides technical support to the seed companies. This model of seed production is still being developed with the hope that eventually, it can be scaled-up to include other countries in ESA.

SN Silim and E Gwata

Milestone: A9.3.4: New institutional models designed and tested to enhance the availability of improved groundnuts, pigeon pea and chickpea seed through commercial channels by 2009

New institutional models: In Mozambique foundation seed of improved pigeonpea varieties from a Foundation Seed Enterprise (FSE) was marketed to a farmer-owned company for further multiplication into certified seed. This was then sold to farmers linked to a pigeonpea processing factory producing *dhal* for export. Preliminary results suggest that this institutional arrangement builds upon the strengths of informal seed supply systems that most smallholder farmers rely on, but enhances the choice of varieties available to such farmers from research. The availability of uniform grain from a single variety reduces the losses associated with processing, and creates

incentives for agro-processors to introduce price premiums for quality that stimulate sustained investment in improved seed.

RB Jones

Output Target A10: Pigeonpea and groundnut transformation protocol developed in Asia applied to locally adapted varieties in ESA [2009].

Activity A10.1: Establish tissue culture protocol for various local pigeonpea and groundnut varieties

Milestone A10.1.1: Evaluate and establish seven locally adapted pigeonpea varieties in tissue culture (2006)

Seven pigeon pea varieties, adapted to the ESA region, as well as the Indian variety ICPL 88039 which was used as control, were introduced into tissue culture in the Kenya Agricultural Research Institute's Biotechnology laboratories. The varieties were from different duration types as well as those with resistance to *Fusarium* and included ICPL 86012, ICPL 87091, ICPV 00020, ICPV 00040, ICPV 00053, ICPV 00554 and ICPV 00447. The tissue culture responses of the different varieties were evaluated for suitability for subsequent transformation. All seven ESA adapted varieties responded well and it was possible to regenerate rooted plants from seed explants of all the varieties. However, the medium (ICPV 00554 and ICPV 00557) and short duration varieties (ICPL 88091 and ICPL 86012) reponded best and ICPV 00554 and ICPV 00557 will be used in transformation studies in 2007.

S de Villiers, D Hoisington, E Gwata, E Manyasa and S Silim

Output 4 B: New knowledge of the QTLs for the stay green trait confirmed, marker assisted selection efficiency improved, specific abiotic stress tolerant varieties and associated knowledge and capacity building measures for sorghum, pearl millet and groundnuts developed and disseminated annually in ESA from 2009 onwards

MTP Output Target: 3 partners received training in generation and interpretation of marker data for MAS in sorghum (Staygreen and Striga)

Output target: B1: 2 QTLs for stay-green introgressed into Sorghum farmer varieties (2008)

Activity B1.1: Develop marker assisted breeding of the stay- green trait of sorghum to enhance terminal drought tolerance in East African farmer-preferred varieties.

Milestone B1.1.1: Capacity of the NARS in marker assisted breeding technologies enhanced through MSc. training by 2007 (DK, TH)

Milestone B1.1.3: Efficiency and effectiveness of marker assisted breeding for stay-green trait in sorghum backgrounds determined by 2008

Attempts by plant breeders to exploit genetic variation for drought tolerance through conventional methods to improve grain yields have proven slow and ardous. However, molecular marker technology has provided powerful tools needed to dissect complex traits such as drought tolerance. A series of backcrosses has already been initiated to transfer the stay green trait from donor parents B35 and E 36-1 into a wide range of elite tropically adapted sorghum varieties including S35, Macia, ICSV 111 and ICSV 112 which are currently grown by resource-poor farmers in Africa. African adapted open pollinated varieties I ICSV 112, Macia and S35 have been advanced from BC1 to BC2 for stay green donor E-36 using SSR-based marker assisted selection targeting 6 stay green QTLs that were detected. During the process, foreground selection has been made to select QTLs of interest and background selection has been undertaken to select for all the loci of the recurrent parent. Near isogenic lines could then be developed for individual stay green QTLs so that the physiological responses associated with individual QTLs controlling various components of the stay green traits and interactions among these QTLs can be dissected. Consistently identified QTLs are therefore good candidates for marker assisted introgression into locally adapted varieties.

However, before marker assisted selection for stay green is applied widely in breeding programs that target extremely drought stressed environments, the benefit of stay green for yield performance and stability should be

proven in target areas. This project therefore aims to transfer the terminal drought tolerance genomic segments (QTLs) in donor parents B 35 and E 36-1 into five commercial, locally adapted, open pollinated farmer-preferred sorghum varieties in Kenya, Uganda, Eritrea, Ethiopia and Sudan. Subsequently, the efficiency and effectiveness of marker assisted breeding for stay green trait in sorghum backgrounds selected from these countries will be determined. An integral part of the project's objectives is to enhance the capacity of scientists in the participating countries in marker assisted breeding technologies. To this end, a one-week a training course for seven scientists, technicians and MSc students involved in the project was organized in November 2006 at the ICRISAT/BecA lab in ILRI,Nairobi. It was organized in collaboration with Nairobi University and the Kenya Agricultural Research Institute (KARI). Participants were trained in DNA extraction, quantification and quality checks using agarose gel electrophoresis and purity tests using nanodrop spectrophometer. They were also trained in PCR optimization and genotyping using capillary electrophoresis. The project teams will now generate the backcrosses and using the skills gained, come back to the ICRISAT/BecA lab for genotyping.

D Kiambi and T Hash

Output Target B2: A diversified set of sorghum varieties tested for sensitivity to photo-periodism by 2011

Activity B2.1: Develop marker assisted breeding program for the photoperiod sensitive sorghum to enhance adaptability and productivity

Milestone B2.1.1: Capability of a diversified set of sorghum varieties to sense the rate of change of photo-periodism clarified by 2008 (MAM, SGM)

Work starts in 2007 and no progress is due for 2006

Milestone B2.1.2: Segregating populations for photoperiod sensitivity and stay green evaluated using molecular markers from 2009 (MAM, SGM)

Work starts in 2007 and no progress is due for 2006

Activity B2.2: Integrate drought tolerant sorghum and millet varieties with water management to improve productivity for sorghum and millet

Milestone B2.2.1: Adaptable drought tolerant varieties for evaluation with water management technologies identified by 2007

Participatory approaches to identify crop water productivity enhancing technologies (IGNRM)
Total crop production needs to increase under increasing water scarcity. The Challenge Program Water for food (CPWF) seeks to identify and deploy interventions that can contribute to increasing food production and saving or using water more efficiently. One of the interventions can be improvement of genetic resources (germplasm) for drought resistance and integrate these with water management technologies. The Challenge Program Water for Food Project no 1 focuses on integrating improved crop varieties with soil fertility and water management to enhance crop water productivity and partnership linkages with markets to improve profitability. Participatory approaches were deployed with NARS partners of Mozambique, South Africa and Zimbabwe collaborating in the CPWFPN1 to identify adaptable crop varieties for evaluation with water management technologies. These include released crop technologies of sorghum, pearl millet, maize, legumes and groundnut varieties. Exploratory field trials comparing the productivity (crop productivity as a measure of water productivity in rainfed situations) of different crop species and varieties using different water conservation techniques and fertilizer use strategies were established in Mozambique and Zimbabwe. Trials mostly followed the Mother-Baby technique with one complete replication of a 2 x 2 x 2 factorial trial in a village and several partial replications of this trial established on fields of nearby farmers. Results from the first year of trials have helped refine the focus of future field work in the project for the 2006/07 season. The total numbers of the crop varieties x soil fertility x water management technologies that will be evaluated with farmers in the 2006/07 season are 485, 108 and 47 for Zimbabwe, South Africa and Mozambique respectively. Training on crop varieties, seed production and water management techniques was provided to 86 collaborators from extension and farming communities of the three target countries

MA Mgonja and Sakile Kudita

Output Target B4: Diverse array of farmer/market preferred varieties and germplasm lines to which breeders can efficiently introgress resistances through MAB

Activity B4.1: Evaluate with farmer participation released and farmer varieties to identify target groundnut varieties for Marker Assisted Backcross (MAB) improvement

Milestone B4.1.1: A groundnut working collection of at least 15 farmer- and market-preferred varieties, as the basis of a marker-assisted breeding program established by 2008 (ESM)

Evaluation of groundnut lines and Elite Varieties for Resistance to Rosette, ELS, Rust and Adaptability for different ESA agro-ecologies: Four nurseries were evaluated for different groundnut production constraints in ESA at the ICRISAT Chitedze Research Center in Malawi.

Details of performance of each set of trials for given constraints are reported. The following are top entries in the Elite Trials for the particular constraints tested:

- Under High Rosette disease pressure, the top three entries in the Elite (Spanish) Trial are: ICGV-SM 01515, ICGV-SM 99529 and ICGV-SM 01501 yield range 1.11 – 1.62 t ha^{-1} vs 0.51 t ha^{-1} for the check JL 24.
- Under High Rosette disease pressure, the top three entries in the Elite (Virginia) Trial are: ICGV-SM 88710, ICGV-SM 01710 and ICGV-SM 01708 yield range 1.22 – 1.78 t ha^{-1} vs 0.78 t ha^{-1} for the check CG 7.
- Under High ELS disease pressure, the top three entries in the Elite Trial are: ICGV-SM 95741, ICGV-SM 95713 and ICGV-SM 96678 yield range 0.76 – 1.18 t ha^{-1} vs 0.61 t ha^{-1} for the check JL 24.
- Rust and Late Leaf Spot were not in epidemic proportions – however the top three entries in the Elite (Spanish) were 86-87/175(b), 86-87/175-3, and 92R/70-4 yield range 3.00 – 3.39 t ha^{-1} vs 1.06 t ha^{-1} for the check JL 24.
- Similarly for the Elite Virginia Trial, top entries were ICGV 95346, RMP 12, and ICGV 90092 yield range 1.43 – 2.00 t ha^{-1} vs 1.24 t ha^{-1} for the check CG 7.
- Drought was not serious. Nevertheless the top three entries in the Elite (Spanish) were ICGV-SM 03535, ICGV-SM 03521, and ICGV-SM 03502 yield range 2.53 – 3.27 t ha^{-1} vs 1.60 t ha^{-1} for the check JL 24.

Three Regional Elite Nurseries (Spanish, Virginia and Valencia) were widely distributed across countries in ESA for evaluation and for selection of Elite varieties for national release in Malawi, Mozambique, Kenya, Tanzania, Zambia and Zimbabwe. The following are highlights of performance data for some of the varieties in reporting countries. Highlights of the Spanish type varietal trial results are reported here.

The Regional Elite Groundnut Variety Trial (Spanish):

- In Malawi, the top three varieties under high rosette disease pressure were ICGV-SM 01514, ICGV-SM 01506, ICGV-SM 01513, yielding 1.3 – 1.6 t ha-1 compared to the released resistant check ICG 12991 (0.7 t ha-1) or the standard check JL 24 (0.21 t ha-1), a yield advantage of over 600%. Under low disease pressure, the best three varieties were ICGV-SM 01513, ICGV-SM 01514, ICGV-SM 01502 yielding 1.8 – 2.5 t ha-1 this compared to the released resistant check ICG 12991 yield of 1.4 and the standard check JL 24 yield of 1.2 t ha-1 (a yield advantage 153 – 215%).
- In Tanzania the top three entries were ICGV-SM 01506, ICGV-SM 99574 and ICGV-SM 01504. However none of the entries were superior to the check JL 24. The apparent superiority was not expressed because there was no rosette disease pressure.
- In Zambia the top three entries were ICGV-SM 99589, ICGV-SM 96714 and ICGV-SM 99568 yielding 0.90 – 0.96 t ha-1 compared to the released check Katete (0.77 t ha-1).
- In Zimbabwe the top three entries were ICGV-SM 86068, ICGV-SM 01514 and ICGV-SM 96714 yielding 1.53 – 2.00 t ha-1 compared to the released check Nyanda (1.22 t ha-1)

With funding support obtained from the McKnight Foundation, the identified superior entries above will be subjected to farmer and market preference tests next season to further prioritize germplasm which will form the basis for a MAB for groundnut improvement in ESA region.

ES Monyo

Output Target B5: New backcross populations incorporating farmer/ market preferences and disease resistance and breeding populations with short duration for marginal drought prone areas [2010]

Activity B5.1: Develop groundnut mapping populations (F2/F3) incorporating farmer/market preferred varieties and known resistance sources for use with MAB

Milestone B5.1.1: At least one new breeding population each for GRD, ELS and rust resistance for ESA by 2009

Several seasons of evaluation of groundnut germplasm under artificial inoculations to create conditions conducive for resistance screening have identified several unique germplasm with enhanced levels of resistance. Simultaneous evaluation of the same under normal conditions have revealed few with high yield potential and preferred confectionary market traits.

The following are the proven sources for Groundnut Rosette Disease resistance: ICGV-SM 90704, ICGV-SM 94584, ICGV-SM 01501, proven sources for ELS; ICGV-SM 93555, ICGV-SM 95714 to be incorporated with farmer/market confectionary types – CG7, Chalimbana, ICGV-SM 87003, JL24, ICG 12991, ICGV 93437, Robut 33-1. Since the confectionary market is the fastest growing for groundnut demand from ESA, we have initiated a hybridization program that will combine resistance to the disease constraints with confectionary market traits. The F1s will be produced during the 2007 main season to generate F2/F3 mapping populations incorporating farmer/market preferred varieties and known resistance sources for use with MAB by 2009.

Activity B5.2: Phenotype segregating groundnut populations for GRD, ELS and/or rust with NARS in selected NARS hotspot locations

Milestone B5.2.1: At least 1 backcross population for each farmer preferred variety incorporating one or more sources of disease (GRD, ELS, rust) resistance or drought tolerance for use in marker assisted backcross improvement by 2009 (ESM, MO).

No report is due for 2006

Output 4C: Progress in knowledge and/or improved germplasm of nutritionally enhanced transgenic sorghum and bio-fortified transgenic events and non-transgenic germplasm with enhanced micronutrient levels available for evaluation with associated capacity building annually from 2009

Output Target C1: Transgenic sorghum breeding lines with increased levels of micronutrients to deliver Recommended Dietary Allowances (RDAs) of vitamins and amino acids developed for use in backcrossing to adapted varieties

Activity C1.1: Screen a core collection of sorghum germplasm to determine variability in morpho-agronomic and micronutrients traits

Milestone C.1.1: Variability for grain densities of Fe, Zn and β carotene determined in at least 200 accessions from at least two ESA countries by 2009

The World Health Organization (WHO) has widely recognized three micronutrients, Fe, Zn and beta carotene as the most limiting for human health. Essential amino acids such as lysine and methionine are also limiting in the diets of most people in ESA especially those whose diets are cereals such as sorghum. Commercial varieties that are available are very low in amino acids and vitamins. Bio-fortification is the process of breeding food crops that are rich in bio-available micronutrients. A number of initiatives by global alliances are in progress to bio-fortify the staple food crops. One such is the Grand Challenge for Global Health (GCGH) initiative that is supporting the project on African Bio-fortified Sorghum. One of the tasks in this project is to assess the natural variability for grain densities of Fe, Zn and β carotene among 450 landraces composed from the five ABS target countries. The landraces and varieties have been planted at Kiboko -Kenya during the 2006/07 season and after harvesting grain

samples will be subjected to micronutrient compositional analyses to establish a baseline for the bio-fortification process.

MA Mgonja and SG Mwangi

Milestone C.1.2: Variability for morpho- agronomic traits determined by 2009

The 450 landraces and varieties composed from the five ABS target countries and being evaluated in milestone C1.1 will be assessed for variability of morpho-agronomic traits.

MA Mgonja and SG Mwangi

Milestone C.1.3: Heritability and correlations among micronutrient traits determined by 2009

The 450 materials planted at Kiboko-Kenya during the 2006/07 season under milestone C1.1 and assessed for micronutrients variability will provide data for assessing broad sense heritabilities and correlations among micronutrients to give a surrogate for the relationships of the micronutrients in the bio-fortified materials.

MA Mgonja and SG Mwangi

Milestone C.1.4: Correlations between morpho-agronomic and micronutrient traits determined by 2010

The 450 materials planted at Kiboko-Kenya during the 2006/07 season will provide data to allow establishment of correlations between morpho–agronomic and micronutrient traits.

MA Mgonja and SG Mwangi

Output Target C2. Regulatory and Biosafety aspects for approval of transgenic sorghum with enhanced micronutrient levels [2011]

Activity C2.1: Conduct non- transgenic baseline environmental and socio ecological research prior to introduction of transgenic micronutrient enhanced sorghum products for regulatory approval and deployment

Milestone C.2.1.1: Determinants of sorghum seed systems, variety information pathways and farmers' maintenance of variety purity established by 2009 in at least two ESA countries

A quantitative survey (structured & semi-questionnaire 200hh/district) was done in 2006 in three districts in Kenya where sorghum is an important crop and where a broad range of weedy/wild and cultivated sorghums co-exist. The objectives were:

- To determine biophysical, socio-economic and cultural factors influencing gene flow (pollen and seed mediated).
- Understand farmers' perception, knowledge and information pathways on sorghum varieties, seed systems, and agronomic practices
- Understand and get baseline information on human factors that influence on-farm gene flow and coping strategies e.g. variety purity maintenance

Data have been collected in the three districts of Western and Nyanza province and is in the process of analysis, and a report will be available in the 2007 archival report for project 4.

MA Mgonja and SG Mwangi

Milestone C.2.1.2: Farmers' knowledge on wild and weedy sorghum and implications on cultivated sorghum documented for at least 2 ESA countries by 2008

The analysis of the socio economic and cultural data collected from the three districts in Kenya and that which will be collected in South Africa will provide information on farmers' knowledge on wild and weedy sorghums and implications on cultivated sorghums and aspects of seed quality and variety identity or no identity preservation.

MA Mgonja and SG Mwangi

Activity C2.2: Understand the in situ dynamics of crop-to-wild and crop-to-weed genetic introgression in Kenya at the country scale

Milestones 2.2.1: Paper accepted on Sorghum crop-to-wild introgression rates in contrasted Kenyan agro-ecological regions (FS, SdV, DK, KARI, University of Free State, University of Hohenheim) 2008

During 2006 an extensive germplasm collection of cultivated landraces, cultivated-wild hybrids and wild sorghums was conducted in the Turkana, Western, Coastal and parts of Eastern provinces of Kenya. A total of 218 samples were collected. DNA, suitable for genotyping, has been isolated from all of these samples. 30 SSR markers were identified for diversity assessment and to date all samples have been genotyped with 12 of these markers. Additional collection trips are planned for 2007 to collect sorghum that mature in different seasons and to fill gaps that were not covered in the first collection. All additional samples will be added to the current 218 for genotyping to estimate the crop-wild geneflow.

F Sagnard, S de Villiers and DKiambi

Activity C2.3: Determine gene flow and outcrossing rates between cultivated, wild and weedy sorghum types and assess hybrid fitness in diverse ecologies using conventional and molecular markers

Milestone C.2.3.1: Molecular analytical laboratory equipped and 10 molecular markers identified for gene flow studies by 2007

The BioScience for Eastern and Central Africa (BECA) facilities were identified for the bulk of our studies. Other activities will be conducted in the Biotechnology laboratory, ICRISAT-India. There are over 100 SSR markers for sorghum that are publicly available and many more are being developed every month. SSR markers will be picked from the sorghum linkage map. SSR markers that are present/frequent in leading sorghum varieties in ESA but absent/rare in wild sorghums will be identified. Work on genotyping of leading sorghum varieties in ESA (22 varieties sent to CSIR) and wild sorghums was started at BECA, Nairobi in November 2006. Seeds from all the genotypes were germinated in trays and DNA extracted from10-day old seedlings using the CTAB method (Doyle and Doyle, 1987). A total of 24 sorghum SSR markers showing high polymorphism under the Generation Challenge Program Project are being evaluated. SSR markers that will show polymorphism between cultivated and wild/weedy sorghum will be identified. Other SSR markers will be tested later.

S Mwangi and MA Mgonja

Milestone C.2.3.2: Agro-ecosystem characterization and genetic sampling on the Intensive Study Site (South Meru District) completed and reported by 2007

During 2006, a Ph.D student was recruited in this project. He conducted a preliminary assessment of the chosen 8 x 8 km intensive study site to develop an appropriate sampling methodology for the further collections of cultivated and wild sorghum planned in this project. The aim was to identify the optimum number of farms that have to be sampled in order to acquire all the sorghum varieties grown in a particular target region and also to obtain some preliminary information on crop diversity and wild sorghum ecological distribution and identification criteria. This was done along an altitudinal gradient of 750 to 1200 masl and across 4 different language groups and it was concluded that at least 27 farms/sites at an interval of 20 m altitudinal difference need to be sampled in order to include all the varieties grown in the survey area. The presence of wild/weedy sorghums in close proximity to the cultivated counterpart seems to point to the occurrence of crop-wild geneflow. The planned studies on mating systems and geneflow will confirm both the historical and present occurrences. A more detailed survey is planned for 2007.

F Sagnard, S de Villiers and D Kiambi

Milestone C.2.3.3: Out crossing and gene flow between at least 5 cultivated and wild or weedy sorghum determined and reported by 2009

In situ introgression in natural habitat:
Seed samples of cultivated and weedy sorghums were collected from three districts (Busia, Teso, and Siaya) located in Western and Nyanza Provinces of Kenya in July 2006. The samples were planted at Kiboko, Kenya in November 2006 for morphological characterization. The samples will further be evaluated in the laboratory in 2007 using 10

SSR markers that are polymorphic between cultivated and wild sorghums. The frequencies of the SSR markers will be used as a measure of gene flow.

Determine the distance of pollen flow:
This activity is being conducted at two locations, Kenya and South Africa in 2006 and 2007. A well adapted variety will be planted in the center of the field in a 20 x 20 m plot as a source of pollen. From the center of the field in eight directions four rows of A-lines with similar maturity to the adapted variety will be sown. Seed set on the A-line was/will be recorded in a one-meter quadrant covering all the four rows at 10 meter-intervals up to a maximum of 100m. The presence of plants with seeds will be used to detect distance of pollen flow. The frequency of outcross seed will be used to determine the outcrossing percentage at various distances.

A trial was established at the Dominion farm in Yala, Nyanza Province, Kenya in April 2006. The pollen source was a locally adapted sorghum variety (Nahandavo) while the female rows were ms lines (ATX 623 and ICSA88006). Preliminary results indicate that mean outcrossing rates were high at short distance from the pollen source and the rate decreased as we move further from the pollen source; beyond 40 m, outcrossing rates were below 1%; outcrossing rates and pollen flow distance were high downwind than upwind. This trial will be repeated at the same site next season. A similar trial was planted at the University of Limpopo farm, South Africa on 28-29 November, 2006. The trial is being conducted in collaboration with the Department of Agriculture, Limpopo Province and the University of Limpopo. The pollen source was a locally adapted variety (SDS 6013) while the female rows are ms lines (A150, A8607, SDSA 27). Data is being collected during this 2007 by a University of Limpopo graduate student who intends to write a MSc thesis on this information

S Mwangi and MA Mgonja

Milestone C.2.3.4: Hybrid fitness determined between cultivated/wild and weedy sorghum types from at least two ESA countries by 2009

Hybrids produced from crop-wild sorghum crosses must persist in the wild if there is to be continued gene exchange in future. The possible generation of "superweeds" after fitness enhancing genes escape from transgenic crops to wild populations is a risk that is often discussed, but rarely studied (Halfhill et al, 2002). This study aims to evaluate the fitness of sorghum crop-wild hybrids for fitness parameters under field conditions.

This activity will be conducted at two sites in Kenya to evaluate the performance of cultivated, wild/weedy and crop-wild/weedy hybrid sorghums under field conditions.

Four cultivated varieties (IS 8593, KARI mtama 1, Gadam Hamam, Seredo) and two wild/weedy sorghum were planted in the crossing block at Kiboko in November 2006. Crossing between cultivated and wild/weedy sorghum will be made. The hybrids and parents will be evaluated for fitness and agronomic performance.

MA Mgonja and SG Mwangi

Milestone C.2.3.5: Fitness of F1, F2, BC1F1 crop-wild hybrids and their wild and cultivated parents evaluated in 2 experimental stations (Eastern and Coastal provinces) by 2008 (FS, SdV, DK + KARI + University of Hohenheim

Artificial, reciprocal crosses are planned between cultivated and wild sorghum varieties to assess the rate of crop-to-weed genetic introgression (and vice versa) and to determine the hybrid fitness resulting from such crosses. Two agro-ecological zones, low- and mid-elevation, have been selected and seed from the collection conducted in mid 2006 was planted at both locations for this purpose. A total of 64 crosses will be made (16 crosses in each of the 4 Sorghum growing areas) and these will be advanced to the F2 and BC1F1 stages in 2007.

F Sagnard, S de Villiers and D Kiambi

Milestone C.2.3.6: Documentation for the risk/safety assessments on the environment needed when actual GM will be presented for regulatory approval initiated by 2009

A document is being prepared on: The significance of gene-flow through pollen transfer in sorghum and implications at centre of diversity using other crops as examples. The document will be prepared in collaboration with the ABS Project regulatory affairs consultant, Dr Willy de Greef.

MA Mgonja and SG Mwangi

Output 4D: Technological options and knowledge to reduce aflatoxin contamination at different stages of the groundnut crop cycle developed and with associated capacity building measures disseminated annually to partner NARES, traders and processors in ESA for enhanced food and feed quality

MTP Output Target: Survey instrument for isolation of atoxigenic strains of Aspergillus flavus developed for ESA

Output Target D.1: Groundnut productivity, sale and income increased

Activity D1.1: Conduct participatory adaptive trials and demonstrations including promotion of systems for control and management of aflatoxin at different stages of the crop cycle
Milestone D1.1.2: Trainers available in quality on-farm seed production and maintenance, and pre-harvest and post harvest aflatoxin control measures implemented in at least 2 ESA NARS on an annual basis 2007- 2011

Training Programme for Field Officers on crop water productivity including soil/water conservation, seed production and harvesting technologies. Since aflatoxin contamination is conditioned by drought stress and poor post harvest handling of produce, the major tenets of this technology were emphasized in this training program offered through the Challenge Program Water for Food during 4-6 October 2006 at Polokwane, South Africa.

On-job training of field staff on field layout, data collection and data processing; and on-job training for front line staff in the Ministry of Agriculture from Ngabu (Malawi) on disease scoring and data collection were conducted.

Training of front line staff from Salima and Blantyre (Malawi) was organized on groundnut diseases and drought constraints affecting groundnut production.

ES Monyo

Activity D1.2: Conduct participatory adaptive trials and demonstrations including promotion of systems for control and management of aflatoxin at different stages of the crop cycle

Milestone D1.1.2: Trainers available in quality on-farm seed production and maintenance, and pre-harvest and post harvest aflatoxin control measures implemented in at least 2 ESA NARS in an annual basis 2007- 2011

Activity D1.2: Conduct field days, agricultural shows, & rural seed fairs with farmers, researchers, market players and processors to promote proven aflatoxin control practices

Milestone D1.2.1: Alternative seed production and distribution system implemented and documented by 2011

Programs for the dissemination of improved seed of ICRISAT mandate crops through promotion of alternative seed supply systems: Participated in developing training materials for on-farm seed production course held for field officers implementing CPWF-CP1 in Zimbabwe, 25–30 September and South Africa, 4–6 October. Forty trainees implementing program activities gained knowledge on seed production linked to on-farm demonstrations of crop water productivity technologies.

Discussions on basic revolving fund initiative was completed with Zimbabwe, South Africa and Mozambique. Agreements were reached to link CPWF-CP1 seed revolving fund with similar activities already under implementation through support from CIMMYT in Zimbabwe, and through USEBA in Mozambique. This activity will be linked with the community seed provision efforts through the Madzivandila Agricultural College which is operating with support from the Agricultural Research Council (ARC) of South Africa.

ES Monyo

Activity D2.1: Isolate atoxigenic strains of *Aspergillus flavus* from soils and develop protocols for multiplication

Milestone: D2.1.1: Survey of groundnut farms in Kenya and Malawi completed by 2007

Survey of groundnut farms: A survey of major peanut growing areas of Malawi and Kenya was carried out in 2005. A total of 162 (84 from Malawi and 82 from Kenya) soil samples were obtained for laboratory isolation and screening of *A. flavus* isolates for aflatoxin production. During this survey, a pod rot complex with obvious symptoms of *Sclerotium rolfsii* was identified as a major constraint to peanut production in both countries. Disease was most severe in fields around Karonga and Salima (eastern plains next to Lake Malawi) in Malawi and in Busia and Siaya districts in Kenya. Severity exceeded 35% of all pods in some fields in Karonga area while a field incidence >25% was recorded in one field in Busia district. Of the farmers interviewed, 59% and 65% in Malawi and Kenya, respectively, were not aware of 'mold' damage or aflatoxin problems of peanuts. A protocol for soil isolation and quantification of *Aspergillus* spp. section *Flavi* was developed and tested in Nairobi. Screening of isolates of *A. flavus* for aflatoxin production was completed and at least six atoxigenic strains identified.

RB Jones

Project 5

Producing more and better food at lower cost of staple cereal and legume hybrids in the Asian SAT (sorghum, pearl millet and pigeonpea) through genetic improvement

Output 5A: Hybrid parents and breeding lines of sorghum, pearl millet and pigeonpea with high yield potential and pro-poor traits in diverse and elite backgrounds, for specific target markets, production environments and research application made available biennially (from 2008) to defined partners with associated knowledge and capacity building in the Asian SAT

MTP Output Targets 2006

Sorghum
New genetic variability introgressed and new derivates less susceptible to shoot fly and grain mold hybrid parents available to partners

Pearl Millet
At least 9 each of male-sterile and restorer lines and more than 800 trait-specific and downy mildew (DM) resistant improved breeding lines developed and disseminated

Pigeonpea
Knowledge on hybrid seed production in pigeonpea published and disseminated globally for the first time

I. Sorghum

Output target 5A.1: More than 35 parental lines of potential sorghum hybrids with high grain yield, and improved agronomic traits and biotic resistance developed (2007-2009)

Activity 5A.1.1: Develop and characterize a diverse range of improved parental lines

Milestone 5A.1.1.1: Ten male-sterile lines and five restorer lines with high yield and large grain developed (BVSR, 2007)

Race-specific trait-based B-lines: To diversify hybrid parents for high yield, large grain and other traits, diverse parents consisting of high-yielding B-lines and germplasm lines that belong to different races possessing useful traits were crossed. The resulting progenies were advanced with selection for different traits while maintaining desired maturity and grain yield. Promising F_4 progenies with maintainer reaction are being converted into A-lines with A_1 and A_2 CMS systems. They are in the various stages (BC_2 to BC_9) of conversion. The B-lines that were completely converted into male-sterile lines and stabilized were evaluated in replicated yield trials. The results are as under.

Preliminary B-line trial (PBT): Maintainers (B-lines) of five newly developed male-sterile lines with A_1 CMS system and 17 newly developed lines with A_2 CMS system were evaluated along with two checks in preliminary B-line trial during the 2006 rainy season. For grain yield, two B-lines of the A_1 CMS system – SP 18847 (4.6 t ha^{-1}), SP 18845 (4.5 t ha^{-1}) and 5 B-lines of the A_2 CMS system – SP 19255 (5.2 t ha^{-1}), SP 19207-1 (4.9 t ha^{-1}), SP 19257 (4.9 t ha^{-1}), SP 19213 (4.6 t ha^{-1}), SP 19251 (4.6 t ha^{-1})] were significantly superior to the check 296 B (3.6 t ha^{-1}). The panicle grain mold rating showed that one B-line of the A_2 CMS system SP 19257 (1.7) and two B-lines of the A_1 CMS system SP 18847 (2.0) and SP 18845 (2.0) were superior (where 1 = no grain mold infection and 5 = >75% infection) compared to the susceptible check 296B (3.0).

Advanced B-line trial (ABT): An ABT consisting of 21 high-yielding B-lines selected from the evaluation of Preliminary B-line trial during the 2005 postrainy season was conducted during the 2006 rainy season. Fifteen of the test B-lines with grain yield ranging from 3.8 to 5.0 t ha^{-1} were on par with the check 296B (4.7 t ha^{-1}). The most promising among them include SP 2863, SP 2305, SP 2781 and SP 2385.

BVS Reddy

Milestone 5A.1.1.2: Five male-sterile lines resistant each to grain mold and shoot fly developed (BVSR/RPT/HCS/RS, 2008)

Grain mold resistance: Grain mold is one of the major biotic constraints in grain sorghum during the rainy season. Efforts have been underway to develop new hybrid seed parents for grain mold resistance (GMR) as the available hybrid seed parents only possess moderate resistance levels.

1. A total of 26 F_6 progenies developed from the crosses involving grain mold resistant B-lines, grain mold resistant landraces and high-yielding B-lines were evaluated in a preliminary screening trial for GMR and also testcrossed to identify maintainer lines for conversion into male-sterile lines during the 2006 rainy season. The data from the screening nursery is awaited. From the breeding nursery, 35 F_7s tolerant to grain mold (PGMR ≤5) and the desirable agronomic traits were produced and harvested along with their testcrosses for assessing their maintainer/restorer reaction

2. In an advanced screening trial for GMR, a total of 72 F_6 progenies developed from the crosses of grain mold resistant landraces, high-yielding B-lines and grain mold resistant breeding lines were evaluated during the 2006 rainy season. These progenies along with their testcrosses on known sources of A_1 and A_2 CMS systems-based male-sterile lines were also evaluated in a separate nursery in the breeding block during the 2006 rainy season. The results from screening trial are awaited. A total of nine lines with GMR (PGMR ≤5) and maintainer reaction were selected from the breeding block nursery. They included two white grain lines and seven red grain lines.

Shoot fly resistance: Shoot fly is one of the major biotic constraints in both rainy and postrainy seasons. Considering that the available seed parents bred for shoot fly resistance (SFR) possess only moderate resistance levels and grain yield potential, efforts are underway to develop and diversify seed parents for SFR.

1. A total of 110 rainy season-adapted F_7 progenies (which included 55 progenies with testcrosses and the remaining 55 yet to be testcrossed) derived from crosses involving shoot fly resistant B-lines and high-yielding B-lines were advanced with selection in 2005 postrainy season. Based on maintainer/restorer reaction, 36 testcrosses and parents were selected for backcrossing. However, backcrossing could be carried out only on 32 parents (BC_1s). The remaining four testcrosses and parents will be advanced and testcrossed along with the evaluation and continuation of backcrossing of 32 BC_1s (as set 1) in the 2006 rainy season. The 55 F_7 progenies (which were yet to be testcrossed) were testcrossed onto known A_1 and A_2 CMS systems-based male-sterile lines in 2005 postrainy season. Based on visual assessment of agronomic aspects, 39 testcrosses and parents were selected for assessing their maintainer/restorer reaction (as set 2) in the 2006 rainy season.
    - The set 1 (32 BC_1s and 4 testcrosses parents) and set 2 (39 testcrosses parents) were screened for SFR in a screening block and also evaluated in a breeding block for a conversion program in the 2006 rainy season. In set 1, the deadhearts ranged from 40 to 69%. The resistant control (IS 18551) had 59% deadhearts and susceptible control (296 B) had 80% deadhearts. Corresponding BC_1s in breeding block which showed below 60% deadhearts in screening block were further backcrossed and a total of 20 BC_2s (8 on A_1, 8 on A_2 and 4 A_1 and A_2) were selected.

    - In set 2, 35 testcrosses and parents and four parents to be testcrossed were screened for SFR in screening block and also evaluated in breeding block for conversion program in the 2006 rainy season. Due to very poor germination and crop establishment, data on deadhearts could not be recorded in the screening block. However, in the breeding block, based on agronomic performance, 10 testcross progenies with maintainer reaction (BC_1s) (6 on A_1 and 4 on A_1 and A_2) were selected for screening for shoot fly resistance.

BVS Reddy, RPThakur and HCSharma

Advanced B-lines resistant to sorghum shoot fly: In an advanced B-line trial, 21 lines along with three checks viz., 296B, ICSB 52, and IS 14384 were evaluated for resistance to shoot fly, *Atherigona soccata,* during the 2006 postrainy season in a randomized complete block design with three replications. Data were recorded on leaf glossiness score (1 = highly glossy, and 5 = non-glossy) and shoot fly deadhearts at 14 and 18 days after seedling emergence. The leaf glossy score ranged from 4.7 to 5.0 in the advanced B-lines compared to 5.0 in 296B and 2.7 in

IS 14384. At 18 days after seedling emergence, deadheart incidence ranged from 50.3 to 85.9% in the seed parents compared to 33.7% in IS 14384 and 75.7% in 296B.

HC Sharma and BVS Reddy

Milestone 5A.1.1.3: Five new high-yielding and large grain male-sterile lines in diverse backgrounds developed (BVSR/HDU, 2009)

High-yielding, large seeded male-sterile lines: A total of 631 F_3s derived from high-yielding B-line × B-line crosses were evaluated during the 2006 rainy season and 509 F_4s were produced. Similarly, from the evaluation of 104 F_3s derived from high-yielding B-line × B-line crosses during the 2005 postrainy season, a total of 20 F_4s were produced. These were evaluated during the 2006 rainy season and simultaneously testcrossed onto A_1-based male-sterile line. From 20 F_4s, 47 F_5s were produced.

A total of 896 F_5 progenies derived from the crosses involving high-yielding B-lines, brown-midrib lines and sweet sorghum B-lines were advanced with selection and testcrossed onto A_1-based male-sterile line, and the resulting 654 F_6 seed and the testcrosses were harvested.

From 46 F_2s (derived from crosses involving postrainy season-adapted varieties, landraces and high-yielding B-lines) evaluated during the 2005 postrainy season, 123 F_3s were produced. Similarly, from 81 F_3s (derived from crosses involving postrainy season-adapted varieties, landraces and high-yielding B-lines) evaluated during the 2005 postrainy season, 62 F_4s were produced during the 2006 postrainy season.

Large grain postrainy season-adapted germplasm lines: A total of 98 highly lustrous and 128 medium-lustrous germplasm lines were planted in two separate trials during the 2005 postrainy season. Due to severe midge attack, the data on grain traits could not be recorded. Hence, during the late 2005 postrainy season, 98 highly lustrous germplasm lines were re-planted for use in a crossing program. Depending on the synchrony of flowering, 67 germplasm lines were crossed with elite advanced breeding progenies with maintainer reaction and established high-yielding B-lines. The 129 F_1s, so generated were planted in the 2006 postrainy season.

In order to diversify the hybrid parental lines adapted to the postrainy season for grain yield and grain size, 11 germplasm lines with large grain (>4.5 g 100^{-1}) IS 30654, IS 30684, IS 30651, IS 30719, IS 19928, IS 19938, IS 35050, IS 36554, IS 36557, IS 30683 and IS 30678 were crossed with elite advanced breeding lines having maintainer reaction. A total of 57 F_1s were made and advanced during the 2006 rainy season. The F_2s are being evaluated during the 2006 postrainy season.

BVS Reddy and HD Upadhyaya

Milestone 5A.1.1.4: Four new male-sterile lines resistant each to shoot fly and grain mold in diverse backgrounds developed (BVSR/RPT/HCS, 2010)

Shoot fly resistance: In a program to develop new male-sterile lines resistant to shoot fly, several shoot fly-resistant germplasm lines (new lines that were not used before) were crossed with high-yielding established B-lines. The 126 F_2s derived from these crosses were evaluated during the 2006 rainy season and 240 F_3s were produced which are being evaluated in the 2006 postrainy season.

In another program to develop shoot fly resistant male-sterile lines in diverse genetic backgrounds, 31 F_1s were made between shoot fly resistant RILs derived from the cross 296B × IS 18551 and high-yielding B-lines, shoot fly resistant B-lines, postrainy season varieties, shoot fly resistant germplasm line and stem borer resistant line. The F_1s are being advanced.

BVS Reddy and HC Sharma

Milestone 5A.1.1.5: New male-sterile lines (4) introgressed with three selected shoot fly resistance QTLs in 296B and BTx 623 backgrounds completed (CTH/BVSR/ HCS/SS, 2008)

While several single-QTL introgression lines were generated in 2007 in these two recurrent parent genetic backgrounds, and screening to assess the efficacy of the introgressed shoot fly resistance QTLs was initiated (see Milestone 5A.2.1.6), no further funds are available to continue application of this work [pyramiding the effective

shoot fly resistance QTLs (once these had been confirmed) in these genetic backgrounds and then developing male-sterile lines from the QTL introgression lines].

Output target 5A.2: A diverse range of trait-specific sorghum breeding lines and populations with morphological diversity and resistance to shoot fly, stem borer and grain mold (2011)

Activity 5A.2.1: Generating new breeding lines with resistance to disease and insect pest resistance, and mapping of QTL and assessment of their effects on resistance levels for these traits

Milestone 5A.2.1.1: Forty F_4 lines developed for resistance to each of grain mold and shoot fly (BVSR/RPT/HCS/RS, 2008)

Grain mold resistance: A total of 200 F_4s derived from the crosses between grain mold resistant B-lines and high-yielding lines were screened for GMR [panicle grain mold rating (PGMR) score taken on 1 to 9 scale where 1 = no mold or <10% and 9 = >90%] in a nursery during the 2006 rainy season. These were also evaluated and testcrossed onto A_1 and A_2 CMS systems in breeding nursery during the 2006 rainy season. The data from the screening nursery are awaited. From the breeding nursery, 51 F_5s tolerant to grain mold (PGMR ≤5) and desirable agronomic traits were produced and harvested along with their testcrosses for assessing their maintainer/restorer reaction.

BVS Reddy and RP Thakur

Shoot fly resistance

Shoot fly resistant maintainers: 12 F_5s derived from the crosses between shoot fly resistant maintainer lines and high-yielding breeding lines (as set 3) were screened for shoot fly resistance in screening block and also evaluated in breeding block for testcrossing in the 2006 rainy season. The deadhearts ranged from 41 to 88%. The resistant control – IS 18551 showed 20% and susceptible control – 296 B showed 87% deadhearts. Corresponding 47 progenies with testcrosses (22 on A_1, 25 on A_2) in the breeding block which showed below 65% deadhearts were selected for continuing the conversion program.

Shoot fly resistant maintainers (postrainy season): In the 2005 postrainy season, 80 F_4s, 174 F_5s, and 17 F_6s with postrainy season adaptation were screened for SFR in a screening block and also advanced in breeding block for testcrossing onto A_1 and A_2 CMS systems-based male-sterile lines. Based on deadheart % (below 57%) in the screening block, 203 F_5 progenies (with 28 testcrosses on A_2, 118 testcrosses on A_1 and A_2 and 57 yet to be testcrossed) from 80 F_4s; 166 F_6 progenies (14 testcrosses on A_2, 101 testcrosses on A_1, A_2 and 51 to be testcrossed) from 174 F_5s; and 20 F_7 progenies (11 testcrosses on A_1 and A_2 and 9 to be testcrossed) from 17 F_6s were produced. These testcross parents along with 27 stabilized B-lines are being evaluated as four separate sets for SFR in screening and breeding blocks in the 2006 postrainy season.

BVS Reddy and HC Sharma

Restorer lines evaluated for resistance to sorghum shoot fly, *Atherigona soccata:* Over 50 F_6 lines were screened for resistance to sorghum shoot fly, *A. soccata* during the 2006 rainy season along with resistant (IS 18551) and susceptible (296B) checks in a randomised complete block design with three replications. Deadheart formation ranged from 10.7 to 58.4%, and 11 lines suffered <25% deadheart incidence compared to 14.1% in the resistant check, IS 18551, and 43.9% in the susceptible check, 296B. The lines 31249, 31242, 31274, and 31292-1 suffered 10.7 to 12.4% deadhearts, and were the most promising.

HC Sharma and BVS Reddy

Pest-resistant nurseries supplied to NARS: A set of 30 lines identified to be resistant to insects (shoot fly, stem borer, aphids, midge and head bugs) were assembled in the form of international pest resistance screening nursery, and ten sets of it were distributed to NARS on request. The material was planted in a randomized complete block design, and there were three replications. Data were recorded on leaf feeding and deadheart formation for stem borer under artificial infestation. Lines SBRIL 66175, SFRIL 65111, SFRIL 65136, SFRIL 65146, SFRIL 65153, SFRIL 65222, SFRIL 65273, SFRIL 65278, and ICSV 700 suffered <20% deadhearts and a leaf damage rating of <5.0 compared to 8.8% in IS 18551, 2.7% in IS 2205, 43.0% in Malisor 84-7, and 17.8% in Swarna. The same material was also screened for resistance to shoot fly, *A. soccata* under natural infestation. Data were recorded on leaf glossiness and deadheart incidence at 14 days after seedling emergence. The lines SFRIL 65108, SFRIL 65136,

SFRIL 651151, SFRIL 65278, IS 2205, ICSV 705, and PS 30710 showed <40% deadhearts and a glossy score of <1.7 compared to 23.9% deadhearts in IS 18551 (glossy score 1.0) and 66.8% deadhearts in Swarna (glossy score 5.0).

HC Sharma and BVS Reddy

Milestone 5A.2.1.2: Two F_6 RIL populations (300 lines each) developed for mapping grain mold resistance (CTH/BVSR/SPD/RPT/RS, 2008)

No progress yet

Milestone 5A.2.1.3: RIL for grain mold resistance from two mapping populations phenotyped and genotyped, and QTL maps developed using 300 markers (BVSR/CTH/SPD/RPT/RS, 2010)

Two RIL F_5 populations (i) IS 23599 × AKMS 14B (350 RILs) and (ii) IS 25017 × KR 188 (350 RILs) and their parents are being advanced to next generation (F_6) during the 2006 postrainy season.

BVS Reddy

Milestone 5A.2.1.4: Putative QTL for stem borer resistance and its components based on RIL from two crosses identified (HCS/CTH/SPD, 2009)

Mapping population evaluated for resistance to stem borer: To identify molecular markers associated with resistance to the spotted stem borer, *Chilo partellus*, the mapping population of 270 lines from the cross IS 18551 × 296B was evaluated for resistance under artificial infestation in a randomized complete block design with three replications. Data were recorded on leaf feeding, deadheart formation, leaf glossiness, days to panicle initiation, recovery resistance, agronomic score, and grain yield. Leaf damage rating (DR) varied from 4.5 to 7.8. Deadheart formation ranged from 31.3 to 97.9% in the mapping population, 44.2% in the resistant check - IS 2205, and 56.7% in the susceptible check ICSV 1. The 296B showed 68.2% deadheart formation compared to 63.2% in IS 18551. Leaf glossiness score varied from 1.0 to 5.0 in the mapping population as compared to 1.3 in IS 2205 – the resistant check, and 4.7 in the susceptible check - ICSV 1. Leaf glossiness score was 5.0 in 296B and 1.0 in IS 18551.

The mapping population, PB 15881-3 × ICSV 745, was evaluated for resistance to sorghum midge during the 2005/06 postrainy season. There were three replications in a balanced alpha design. Data were recorded on midge damage (1 = <10% spikelets with midge damage, and 9 = >80% spikelets with midge damage). Midge damage in the mapping population ranged from 2.0 to 9.0 compared to 1.0 in the resistance check, ICSV 197, and 7.7 in the susceptible check, Swarna. The resistant (ICSV 745) and the susceptible parents (PB 15881-3) suffered a midge DR of 2.0 and 6.3, respectively. Data analysis to identify QTLs associated with resistance to these insects is in progress.

HC Sharma, BVS Reddy and CT Hash

Milestone 5A.2.1.5: Comparative mapping of QTL for stem borer resistance in sorghum and maize completed (HCS/SPD/DH/CTH, 2009)

A total of 272 and 363 progenies (along with their parents) of sorghum $F_{6:10}$ RIL mapping populations derived from crosses [ICSV 745 (susceptible to stem borer) × PB 15220-1 (resistant to stem borer)] and [ICSV 745 × PB 15881-3 (resistant to stem borer)], respectively, were sown during the 2006/07 postrainy season for seed multiplication. These populations will be screened for stem borer resistance during the 2007 and 2008 rainy seasons. DNA extraction for these populations has been completed.

HC Sharma, SP Deshpande, D Hoisington and CT Hash

Milestone 5A.2.1.6: Effectiveness of two best QTL for resistance to shoot fly in two genetic backgrounds demonstrated (CTH/SPD/SS/HCS/BVSR, 2007)

To be reported in 2007

Milestone 5A.2.1.7: Comparisons of lines with single-QTL introgressions and QTL pyramided in two genetic backgrounds for shoot fly resistance completed (HCS/BVSR/CTH/SPD, 2010)

To be reported subsequently

Output target 5A.3: Variation in sorghum grain mold pathogens and mycotoxin contamination risk assessed, insect–host genotype-natural enemy interactions studied, and mechanisms of resistance to insect pests identified (2010)

Activity 5A.3.1: Understanding host-pathogen-environment interaction in grain mold complex.

Milestone 5A.3.1.1: Major grain mold pathogens in sorghum growing states in India identified and their distribution in relation to weather factors determined (RPT/RS, 2007)

Weather variables and grain mold pathogens: We conducted a Sorghum Grain Mold Resistance Stability Nursery (SGMRSN) under the ICAR (AICSIP)-ICRISAT partnership project at five AICSIP centers Coimbatore, Dharwad, Parbhani, Palem and Patancheru. The SGMRSN 2006 consisted of 50 entries (14 from NRCS and 36 from ICRISAT). In addition to grain mold severity, data on weather variables (temperature, relative humidity and rainfall) were also collected from these centers. Molded grain samples from these locations will be assayed for the presence of various mold fungi and their frequency determined. From these and earlier data sets of the past four years, the relationship between weather variables and frequency of mold pathogens at different locations will be determined.

RP Thakur and Rajan Sharma

Milestone 5A.3.1.2: Mycotoxin-producing isolates of Fusarium species associated with grain mold identified and characterized and genetic resistance in relation to other major pathogens determined (2009)

Genetic resistance to sorghum grain mold: Identification of advanced sorghum breeding lines and germplasm accessions with improved level of grain mold resistance has been a major focus towards developing grain mold-resistant hybrids. During 2006, a number of sorghum lines were screened in the glasshouse against individual pathogens and in the grain mold nursery at ICRISAT, Patancheru, against general mold fungi.

Grain mold resistance in hybrid parental lines: Twenty sorghum genotypes [12 B-lines, 6 R-lines and 2 susceptible checks (CSH 9 and SPV 104)] were screened against three prominent pathogenic fungi, *Fusarium verticillioides* (high fumonisins-producing strain), *Curvularia lunata* and *Alternaria alternata* under glasshouse conditions. The experiment was conducted in a randomized complete block design (RCBD) with 20 genotypes × 3 fungi × 3 replications with 10 plants per replication. Panicles were inoculated at >80% flowering stage with conidial suspensions of individual fungi and exposed to wetness for 48 h after inoculation. As the visual mold infection was not clearly evident, we measured grain mold colonization at hard dough (HD), physiological maturity (PM) and on threshed grain (TG) using the blotter method and 50 grains per replication at each grain development stage. In general grain colonization by different fungi at HD was low and varied from 0 to 38%. However, grain colonization at PM and of TG was quite variable for sorghum genotypes × pathogen combinations.

Grain colonization by *F. verticillioides* varied from 1 to 40% on test genotypes compared to 8–32% on susceptible controls. Similarly, grain colonization by *C. lunata* varied from 11 to 92% on test genotypes compared to 51–95% on susceptible controls, while that by *A. alternata* varied from 0 to 85% on test genotypes compared to 1–6% on susceptible controls. Sorghum genotypes with resistance to single and multiple pathogens were identified. Four genotypes (ICSB 352-5, ICSB 402-3, ICSB 370-2 and ICSR 89013-2) were resistant to *F. verticillioides* (0–9% colonization); three genotypes (ICSB 402-3, ICSB 402-1-2 and SGMR 40-1-2-3) to *C. lunata* (7–17% colonization) and five genotypes (ICSB 402-3, ICSB 402-1-2, IS 41397-3, SPV 462-3, SP 72519-1-3) to *A. alternata* (0–5% colonization). Of these, only one genotype (ICSB 402-3) was resistant to all three pathogens; four (ICSB 370-1-5, ICSV 96094-2, ICSR 89013-2 and ICSB 379-2) to both *F. verticillioides* and *A. alternata* and two (ICSB 402-1-2 and SGMR 40-1-2-3) to both *C. lunata and A. alternata*. This information would be useful for breeding grain mold resistant hybrids and for studies on genetics and mechanism of resistance.

Grain mold resistance in selections from B- and R-lines: Ninety-seven grain mold-resistant single plant selections (33 from 14 B-lines and 64 from 24 R-lines from the 2005 screen) along with 4 resistant and 4 susceptible checks were evaluated to confirm their resistance. The experiment was conducted in a RCBD with 2 replications, 1 row of 2 m long/replication. Sprinkler irrigation was provided twice a day for 30 min. each on rain-free days from flowering to physiological maturity to provide high humidity (>90% RH) essential for mold development. The grain mold scores were recorded at physiological maturity (PM) using a 1 to 9 scale, where 1 = no mold infection and 9 >75% molded grains on a panicle. The mean grain mold scores on test genotypes ranged from 2.0 to 7.8 compared to 1.0–2.0 score on resistant checks (IS 14384, IS 8545 and IS 25017) and 8.8–9.0 score on the susceptible checks (CSH 9, CSH 16, Bulk Y and SPV 104). Results indicated that 50 selections (23 from 8 B-lines and 27 from 10 R-lines) were resistant (≤3.0 score). Some of these selections have been utilized in developing test hybrids.

Grain mold resistance in sorghum germplasm: One hundred fifty-six germplasm lines reported as resistant to grain mold during 1985–87 along with two resistant and one susceptible checks were screened to find the stability of resistance under field conditions. These were evaluated unreplicated with 2 rows of 2 m per entry. Sprinkler irrigation was provided twice a day for 30 min. each on rain-free days from flowering to physiological maturity. The visual grain mold scores were recorded at physiological maturity (PM). The grain mold scores of the test lines varied from 1 to 7 compared to 1.0 to 2.0 on resistant checks (IS 14384 and IS 25017) and 9.0 on the susceptible check (SPV 104). Of the 156 lines, 19 (IS 3413, IS 8848, IS 13885, IS 14375, IS 14380, IS 14384, IS 14385, IS 14387, IS 14390, IS 13756, IS 21599, IS 24995, is 24989, IS 24996, IS 25038, IS 25075, IS 25084, IS 25100 and IS 25105) were highly resistant (1.0 score); 134 resistant (1.1 to 3.0 score) and 3 moderately resistant (3.1–5.0 score). Some of the highly resistant germplasm lines from the 19 identified above having desirable agronomic traits would be useful in breeding program to develop grain mold resistant hybrid parents.

Grain mold resistance in *zerazera* selections: Thirty-two selections from two *zerazera* conversion lines (IS 18758C and IS 30469C) and two susceptible checks were evaluated for grain mold resistance. The 34 entries were grown in a RCBD with 2 replications, 1 row of 2 m per replication. The sprinkler irrigation was provided twice a day for 30 min. each on rain-free days from flowering to physiological maturity. The grain mold scores were recorded at physiological maturity (PM). Five (from IS 18758C) of the 32 selections were moderately resistant (3.4 to 4.3 score), while the remaining were susceptible (>6.0 scores). These five resistant selections are early maturing with medium height and white grain and thus could be utilized in grain mold resistance breeding.

RP Thakur and Rajan Sharma

Milestone 5A.3.1.3: Relative contributions of host and environmental factors in mold development assessed (RPT/RS/BVSR, 2010)

Host and environmental factors in relation to mold development: In the 2006 grain mold nursery, we evaluated 156 germplasm lines reported as resistant (during 1985–87) to confirm their resistance under changed environment and screening procedure. These lines originate from different countries of Africa and possess diverse morphological traits (such as panicle shape and size, grain color, glumes color and glumes coverage of grains) and agronomic traits (plant height and days to flowering). In addition to grain mold severity, we obtained data on the above morphological and agronomic traits. From these and other data sets from diverse breeding lines collected during the past years we plan to determine the relative contributions of host and environmental factors to grain mold development.

RP Thakur, Rajan Sharma and BVS Reddy

Activity 5A.3.2: Develop screening techniques and investigate host genotype - natural enemy interactions, resistance mechanisms and genetics of insect pest resistance

Milestone 5A.3.2.1: Insect–host genotype - natural enemy interactions, and mechanisms of resistance and their inheritance studied in sorghum (HCS/BVSR, 2009)

Physico-chemical mechanisms of resistance to sorghum shoot fly: Fifteen lines comprising of shoot fly-resistant and -susceptible types were evaluated for resistance to *Atherigona soccata* under no-choice, dual-choice and multi-choice conditions in the field and glasshouse. There were three replications in a randomized complete block design. Data were recorded on shoot fly oviposition, deadheart formation, recovery resistance, leaf glossiness score, trichome density, insect survival and development, and fecundity. There were 5.11 eggs per 10 plants in IS 2146 to

19.55 eggs on ICSV 112, and 16.49 in case of Swarna. Deadheart incidence ranged from 55.85 in IS 2312 to 99.2% in Swarna, compared to 78.3% deadhearts in the resistant check, IS 18551. Under dual-choice conditions, the genotypes IS 1054, IS 2146, IS 18551, IS 4664, and SFCR 125 showed non-preference for oviposition as compared to the susceptible check, Swarna. Antibiosis in terms of success of the neonate larvae to establish on the plants and cause a deadheart was observed in case of IS 1054, IS 1057, IS 18551, IS 2312, IS 4664, IS 2205, SFCR 125, SFCR 151, and ICSV 700. The leaf glossiness score varied from 1.0 to 5.0. Genotypes showing less susceptibility to shoot fly were trichomed and had a leaf glossiness score of <3.0. Genotypes IS 18551, IS 4664, IS 2312, SFCR 151, and ICSV 700 showed some adverse effects on the survival and development of *A. soccata*.

Transplanting and clipping of leaves at 15 days after seedling emergence resulted in a significant reduction in deadheart formation in the shoot-fly resistant genotypes, but did not result in better recovery or overall resistance and agronomic performance at maturity. Some of these lines also showed resistance to stem borer, *Chilo partellus* and the sugarcane aphid, *Melanaphis sacchari* during the postrainy season. Biochemical analysis of the plant samples for essential minerals, nutritional quality, and secondary metabolites in relation to expression of resistance to sorghum shoot fly is in progress.

HC Sharma

Milestone 5A.3.2.2: Techniques to evaluate sorghums for resistance to sugarcane aphid and shoot bug developed, and sources of resistance identified in sorghum (HCS, 2009)

Improved breeding lines resistant to sugarcane aphids: Thirty-five sorghum lines comprising of improved breeding lines and germplasm accessions were screened for resistance to sugarcane aphid, *Melanaphis sacchari* during the 2006 rainy season. There were three replications in a RCBD, and observations were recorded at physiological maturity on aphid damage (1 = <10% leaf area damaged, and 9 = >80% leaf area with aphid damage) and agronomic performance (1 = good, and 5 = poor). The lines 61011, 61523, 61588, 61592, 61596, DJ 6514, ICSB 12, ICSB 88017, ICSV 197, ICSV 700, ICSV 745, and IS 40620 showed an aphid damage rating of <2.5 compared to 3.5 in the resistant check TAM 428, 5.5 in CK 60B, and 6.5 in Swarna. Of these, the lines 61011, 61523, 61588, ICSB 88017, and ICSV 745 were also desirable agronomically, and can be used in sorghum improvement program to develop cultivars for resistance to *M. sacchari.*

HC Sharma and BVS Reddy

Output target 5A.4: Information on association between CMS and agronomic traits, and between molecular diversity and yield heterosis in sorghum (2009)

Activity 5A.4.1: Evaluation of iso-cytoplasmic hybrids for grain yield and agronomic traits

Milestone 5A.4.1.1: Twelve hybrids with four diverse CMS systems compared for agronomic traits and resistance to shoot fly and grain mold (BVSR/RPT/RS/HCS, 2009)

CMS effect on grain yield and resistance to grain mold and shoot fly: The need for cytoplasmic diversification of A-lines (and hybrids) to avert the potential risk of unforeseen disease and insect pest outbreaks associated with cytoplasmic uniformity is a common knowledge. Cytoplasmic diversification also enhances the opportunities for diversifying the nuclear genetic base of A-lines as some of the outstanding restorers on one cytoplasm are found to be maintainers of other cytoplasms. However, in pursuit of diversifying the CMS base of hybrid seed parents and hence the hybrids, the performance of hybrid seed parents and the hybrids based on alternative CMS systems for grain yield and other agronomic traits and plant defensive traits of economic importance cannot be compromised. An investigation was, therefore, carried out to assess the efficiency of A_2 A_3 and A_4 CMS system in comparison to the widely used A_1 CMS system.

Isonuclear alloplasmic A-lines with A_1, A_2, A_3 and A_4 (G), A_4 (M) and A_4 (VZM) cytoplasm each in six nuclear genetic backgrounds (ICSB 11, ICSB 37, ICSB 38, ICSB 42, ICSBA 88001 and ICSB 88004) were crossed with two common R-lines (IS 33844-5 and M 35-1-19) to generate 72 hybrids in 2005 postrainy season. These 72 hybrids were evaluated for grain yield and other traits during the 2006 rainy season in split-split-plot design with three replications using R-lines as main plots, genetic backgrounds of A-lines as sub-plots and cytoplasm in sub-sub-plots. The 6 B-lines and 4 checks (296B, RS 29, CSH 16 and CSV 15) were evaluated in a separate trial using randomized

complete block design with three replications. Same set of hybrids and their parents were screened for grain mold and shoot fly in screening blocks.

Grain yield and other traits: There were significant differences amongst female nuclear genotypes for days to 50% flowering, plant height and grain yield but the differences were non-significant amongst R-lines. Similarly, non-significant mean squares due to A × R-line interaction indicated that hybrids do not differ significantly for their *sca* effects for grain yield. Cytoplasm *per se* appeared to have significant influence on the expression of hybrids for plant height and grain yield, as evident from significant mean squares due to cytoplasm. It is important to note that first-order interaction of cytoplasm with nuclear genetic background of A-lines (for all traits) or R-lines (for grain yield) and second-order interaction with A-line and R-lines (for all traits) towards variation of iso-nuclear hybrids was significant, suggesting significant but variable influence of cytoplasm for grain yield, depending on the genetic background.

The comparison of A_1, A_2, A_3, A_4 (M), A_4 (G) and A_4 (VZM) cytoplasms-based hybrids indicated that A_4 (M) cytoplasm-based crosses were significantly earlier to flower compared to those based on A_3 and A_4 (VZM) cytoplasms (though only by a day, which has no practical significance). A_2 cytoplasm-based hybrids were significantly taller compared to A_3, A_4 (M), A_4 (G) and A_4 (VZM) cytoplasm-based hybrids by 6 to 9 cm, (which again has no practical significance). A_1, A_2, A_3, A_4 (M), A_4 (G) cytoplasm based hybrids were significantly superior to A_4 (VZM) cytoplasm-based hybrids (by 1.0 t ha^{-1}) and were comparable among themselves.

Thus, the comparable grain yield potential of A_2, A_3, A_4 (M), A_4 (G) cytoplasms-based hybrids in similar maturity and plant height backgrounds suggests the usefulness of A_2, A_3, A_4 (M), A_4 (G) cytoplasms for diversifying the cytoplasmic and nuclear genetic base of sorghum hybrid parents.

BVS Reddy

Grain mold resistance: There were significant differences amongst A-lines (nuclear genotype) and R-lines for PGMR score, suggesting considerable differences among the parents for responses to grain mold. Significant mean squares due to cytoplasm *per se* as for PGMR indicated that cytoplasms showed differential responses to grain mold. The significant mean squares due to interaction of cytoplasms with A-lines and R-lines and A × R-lines interaction suggested that the effect of cytoplasms was significantly influenced by the genetic backgrounds of both female and male parents.

The comparison of A_1, A_2, A_3, A_4 (M), A_4 (G) and A_4 (VZM) cytoplasms-based hybrids indicated that A_1, A_2, A_3, and A_4 (VZM) cytoplasms-based hybrids were relatively less susceptible to grain mold than those based on A_4 (M) and A_4 (G) cytoplasms. However, when mean PGMR scores of different cytoplasms-based hybrids were examined, there were seldom any differences in their responses to grain mold.

BVS Reddy and RP Thakur

Male-sterility-inducing cytoplasm *vs* normal fertile cytoplasm: Two sets of 36 (A × R) hybrids in iso-nuclear alloplasmic backgrounds (36 in A_1 and 36 in A_2) were made by crossing iso-nuclear, alloplasmic A_1 and A_2 system A-lines in 12 nuclear genetic backgrounds with three dual R-lines. The male-fertile counterparts of the 12 male-sterile lines (B-lines) were emasculated and crossed with the same three dual R-lines and obtained 36 B × R crosses. The two sets of 36 A × R and one set of 36 B × R crosses were evaluated at ICRISAT, Patancheru during the 2006 rainy season in split-split-plot design using three replications by using R-lines in the main plots, A-lines as sub-plots and cytoplasms as sub-sub-plots. The 12 A-lines and their B-lines were evaluated in a separate trial using randomized complete block design with three replications. Sufficient care was taken for adequate supply of pollen grains to A-lines for meaningful comparison of yield performance of A-lines *vs.* B-lines.

There were significant differences amongst A-/B-lines (nuclear genotype) and R-lines for plant height and grain yield justifying the selection of the hybrid parents (A/B- and R-lines). Similarly, significant mean squares due to A-/B- × R-lines interaction indicate that hybrids differed significantly for their *sca* effects for grain yield. Cytoplasm *per se* appeared to have significant influence on the expression of hybrids for plant height as evident from significant mean squares due to cytoplasm. It is important to note that first-order interaction of cytoplasm with nuclear genetic background of A-lines (for grain yield) or R-lines (for all the traits) and second-order interaction with A-line and R-lines (for grain yield) towards variation of iso-nuclear hybrids were also significant, suggesting that the cytoplasm was influenced by the genetic backgrounds of both male and female parents.

The comparison of A × R and B × R crosses (in both A_1 and A_2 backgrounds) indicated that while A × R (both A_1 and A_2) crosses were significantly early (by 1 day in both A_1 and A_2 backgrounds for days to 50% flowering) and significantly taller (by 0.2 m in A_1 and by 0.2 m in A_2 backgrounds), B × R crosses manifested higher grain yield (by 0.1 t ha^{-1} than A_1 and by 0.2 t ha^{-1} than A_2 cytoplasm) when average performance of A_1 and A_2-based A × R and B × R hybrids as separate groups was considered.

Significant cytoplasmic effects were observed for all the traits when individual nuclear genetic backgrounds of A × R (both A_1 and A_2) and B × R crosses were examined. Significant cytoplasm effects were detected in some of the nuclear genetic backgrounds for all the traits. While A × R crosses, besides being early, were taller compared to those of B × R crosses in a majority of nuclear genetic backgrounds, the reverse was true for grain yield. These results were not in conformity with those reported earlier, wherein A × R crosses were significantly superior to B × R crosses for grain yield suggesting the need for repetition of the trials for onfirming these results.

BVS Reddy

Shoot fly resistance: The F_1 hybrids based on different male-sterile cytoplasms were tested for resistance to sorghum shoot fly, *A. soccata* under field conditions using interlard fishmeal technique. Seventy-two hybrids and 12 restorers were tested for resistance to shoot fly under natural infestation. There were three replications in a randomized complete block design. Data on shoot fly deadhearts were recorded at 18 days after seedling emergence, when the differences between the resistant and susceptible checks were maximum. Shoot fly deadhearts ranged from 40.0 to 100.0% in the hybrids, and it was 28.8% in IS 18551, 84.8% in 296B, and 81.2% in CSH 16. Hybrids based on $ICSA_4M$ were less susceptible than those based on other cytoplasms.

In another trial, 26 hybrids, 10 B-lines, and 10 restorers along with resistant, IS 18551 and susceptible, CSH 16 checks were evaluated for resistance to shoot fly, *A. soccata*. There were three replications in a randomized complete block design. Data on shoot fly deadhearts was recorded at 18 days after seedling emergence, when the differences between the resistant and susceptible checks were maximum. Shoot fly deadhearts ranged from 32.2 to 78.4% in the hybrids, and it was 46.1% in IS 18551, and 91.1% in CSH 16. Hybrids based on ICSA 425 and ICSA 452 were less susceptible than those based on ICSA 455, indicating the potential of using these lines for producing shoot fly resistant hybrids.

HC Sharma and BVS Reddy

Activity 5A.4.2: Assessing the relationship between molecular diversity of parental and yield heterosis

Milestone 5A.4.2.1: Relationship between parental molecular diversity and hybrid heterosis assessed (CTH/SPD/BVSR, 2009)

To be reported subsequently

Output target 5A.5: High-yielding and good combining sorghum hybrid parents developed for postrainy season adaptation (2009)

Activity 5A.5.1: Developing high-yielding and good combining sorghum hybrid parents for postrainy season adaptation

Milestone 5A.5.1.1: Five each of high-yielding and good combining sorghum male-sterile lines and restorer lines for postrainy season developed (BVSR, 2009)

Several early-maturing advanced generation progenies derived from the crosses between postrainy season-adapted varieties, high-yielding B-lines and landraces were evaluated during the 2005 postrainy season.

A total of 44 postrainy season-adapted F_6 progenies were evaluated and testcrossed onto A_1–male-sterile line. From these, 34 F_7 progenies were produced and their testcrosses were harvested. The selected progenies flowered in 76–86 days and had a plant height range of 1.3 to 2.1 m.

In another nursery, 384 F_5s derived form B × B crosses were evaluated and testcrossed onto A_1–male-sterile line producing 359 F_6 seed and harvested their testcrosses. The selected progenies flowered in 75–100 days and had a plant height range of 1.1 to 2.0 m.

In yet another nursery 93 F_5 progenies were evaluated producing 78 F_6 progenies and their testcrosses onto A_1–male-sterile line during the 2005 postrainy season. The selected progenies flowered in 75–95 days and had plant height ranging from 1.5 to 2.5 m. From 140 postrainy season-adapted F_4 progenies evaluated, 241 F_5s were produced. The selected progenies flowered in 74–97 days and had a plant height range of 1.1 to 2.5 m.

Combining ability of hybrid parents: To identify the best combiners among the postrainy season-adapted B-lines and restorers and to identify good hybrid combinations, eight postrainy season-adapted B-lines (ICSB 6, ICSB 14, ICSB 51, ICSB 52, ICSB 215, ICSB 297, ICSB 592, ICSB 675) were crossed to eight postrainy season -based R-lines (IS 4504-1, M 35-1-19-1, M 35-1-69-1, M 35-1-16-1, M 35-1-25-1, BP Ent 14, SP 7521, SP 7523) in a Line × Tester mating design to produce 64 hybrids. These 64 hybrids were evaluated along with a hybrid check CSH 15R in a RCBD design during the 2005 postrainy season.

Significant differences were observed among the lines and testers for days to 50% flowering, plant height, stay-green score, lodging score, grain yield and 100-grain weight. Significant line × tester interaction mean squares for grain yield, stay-green score and lodging score suggested the involvement of dominance and/or epistatic gene action. While ICSB 297 and ICSB 592 among the A-lines and M 35-1-19-1 and M 35-1-69-1 among the R-lines were good general combiners for grain yield, ICSB 6 and ICSB 52 among the A-lines and IS 4504-1 and Ent 14 among the R-lines were good general combiners for grain size. The hybrids, ICSA 215 × M 35-1-19-1 and ICSA 51 × IS 4504-1 were good specific combiners for grain yield.

R-line development: From the seed parent development program for postrainy season adaptation, the advanced progenies with R-reaction were evaluated.

A total of 605 F_3s derived from high-yielding R × R crosses were evaluated, and 140 F_4 seeds were produced during the 2005 postrainy season, and evaluated during the 2006 rainy season to produce 36 F_5s. Days to 50% flowering in the selected progenies ranged from 69 to 75 days and plant height ranged from 1.3 to 2.7m.

From the 53 postrainy season-adapted F_5 progenies (variety × variety crosses) evaluated during the 2005 postrainy season, 31 F_6 progenies were produced and their testcrosses were harvested. The selected progenies flowered in 77 - 87 days and had a plant height range of 1.0 to 1.8 m.

From the 63 (F_7) advanced selections, derived from postrainy season-adapted large- grain and high-yielding variety × variety crosses, 25 advanced progenies were produced and evaluated in a trial during the 2006 postrainy season.

Restorer lines trial (RLT-2005R): A trial was constituted with 40 advanced breeding lines showing restorer reaction. The material in the trial includes the lines developed from the crosses involving postrainy season-adapted varieties (M 35-1, SPV 1359) and advanced breeding lines and the selections from M 35-1 bulk for evaluation for grain yield and other desirable traits in a randomized complete block design with three replications along with the checks Moulee and M 35-1. Compared to the check M 35-1 (2.1 t ha^{-1}), 31 R-lines with a grain yield range of 3.1 to 4.7 t ha^{-1} performed significantly better for grain yield and seven of these were significantly superior to the check Moulee (3.2 t ha^{-1}). The luster score among these 31 lines varied from 1.0 to 2.7 (M 35-1: 1.0, Moulee: 1.0), while the grain size varied from 1.7 to 2.6 g 100^{-1} grains (M 35-1: 2.1g 100^{-1} grains, Moulee: 2.4g 100^{-1} grains).

BVS Reddy

Output target 5A.6: High-yielding dual-purpose foliar disease resistant forage/sweet sorghum hybrid parents (2009)

Activity 5A.6.1: Developing dual-purpose foliar disease resistant forage/sweet sorghum hybrid parents

Milestone 5A.6.1.1: Six new dual-purpose foliar disease resistant forage/sweet sorghum hybrid parents developed (BVSR, 2009)

High-yielding designated hybrid parents with sweet stalk, and varieties and hybrids developed by crossing promising sweet sorghum A- and R-lines were evaluated in replicated trials during the 2005 postrainy season. Results of these trials are given below.

Sweet sorghum B-line trial: Based on the performance of sweet sorghum B-lines evaluated during the 2005 rainy season, a total of 30 B-lines were selected and evaluated along with the checks NSSH 104 and SSV 84 in the 2005 postrainy season. ICSB 73 with 0.9 t ha^{-1} sugar yield performed significantly better than the best check NSSH 104 (0.74 t ha^{-1}) for sugar yield based on Brix reading and juice yield, while ICSB 324 (0.7 t ha^{-1}), ICSB 652 & 401 (0.6 t ha^{-1}) and ICSB 24001 (0.5 t ha^{-1}) were significantly better than SSV 84 (0.3 t ha^{-1}). The performance of the lines for other traits is presented in Table 1.

Sweet sorghum advanced B-line trial (SSABLT, 2006K): Based on the performance of B-lines in sweet sorghum B-line trial in 2004 postrainy season, 2005 rainy and postrainy seasons, PPV (Protection of Plant Varieties) trials in 2004 rainy and postrainy seasons, PPV trials in 2005 rainy and postrainy seasons, 75 B-lines were selected and evaluated during the 2006 rainy season along with the checks 296B and SSV 84. Three B-lines, ICSB 729 (3.3 t ha^{-1}), ICSB 722 (3.1 t ha^{-1}), ICSB 321 (3.0 t ha^{-1}) were on par with the check SSV 84 (2.7 t ha^{-1}) for sugar yield. Among these, ICSB 722 (14 t ha^{-1}) was significantly better than the check 296 B (10.9 t ha^{-1}) for grain yield, while the rest of them were on par with 296 B, except ICSB 321. The performance of these lines for other traits is given in Table 2.

Table 1. Performance of selected sweet sorghum B-lines (at maturity stage) - 2005 postrainy season at ICRISAT, Patancheru

B-line	Days to 50% flowering	Plant height (m)	Cane yield (t ha^{-1})	Juice yield (t ha^{-1})	Brix reading at maturity	Sugar yield based on Brix reading and juice yield (t ha^{-1})
ICSB 73	75	1.5	13.8	5.7	16.7	0.9
ICSB 324	75	1.5	12.0	3.7	19.0	0.7
ICSB 652	75	1.3	11.1	3.3	16.7	0.6
ICSB 401	75	1.4	12.9	4.2	13.0	0.6
ICSB 24001	75	1.5	13.2	4.4	11.7	0.5
NSSH 104 (Check)	73	1.5	13.3	5.3	13.3	0.7
SSV 84 (Check)	70	1.4	9.0	2.7	16.3	0.4
Mean	75	1.2	6.9	2.1	12.0	0.3
CV (%)	3.9	15.0	21.9	22.3	22.4	29.4
CD (5%)	3.96	0.30	2.5	0.74	4.36	0.13

Table 2. Performance of selected sweet sorghum B-lines (at maturity stage) - 2006 rainy season at ICRISAT, Patancheru

B-line	Days to 50% flowering	Plant height (m)	Cane yield (t ha^{-1})	Juice yield (t ha^{-1})	Brix reading at maturity	Sugar yield based on Brix reading and juice yield (t ha^{-1})	Grain yield (t ha^{-1})
ICSB 729	77	2.2	49.5	23.4	14.7	3.3	10.9
ICSB 722	75	2.2	41.5	20.5	15.2	3.1	14.0
ICSB 321	78	2.3	40.6	18.0	17.5	3.0	7.9
SSV 84 (Check)	82	2.9	45.7	18.6	18.2	3.3	3.0
296 B (Check)	69	1.5	12.1	2.7	8.3	0.5	10.9
Mean	67	1.6	21.5	9.2	12.9	1.2	11.4
CV (%)	1.69	8.20	12.5	21.11	9.33	30.3	11.84
CD (5%)	1.82	0.22	4.36	3.13	1.94	0.41	2.17

Sweet sorghum male-sterile line development: A total of 10 crosses were made involving lines with high Brix (SSV 84, ICSV 700) and low Brix (ISIAP DORADO, BTx 623), and high-yielding lines (ICSB 52 and ICSB 101). The 10 F_1s are planted in 2006 postrainy season for advancement.

Milestone: 5A.6.1.2: Six new high-yielding sweet sorghum restorers identified (BVSR/HDU, 2008)

Sweet sorghum varietal and restorers trial: Based on the performance of sweet sorghum varieties and restorers in the trial during the 2005 rainy season, 45 lines were selected and evaluated along with the checks SSV 74, SSV 84 and NSSH 104. Compared to the sugar yield in checks (SSV 74: 1.2 t ha^{-1}, SSV 84: 0.5 t ha^{-1} and NSSH 104: 1.1 t ha^{-1}), 14 varieties were significantly superior (1.67 to 3.0 t ha^{-1}). Some of the lines had high brix reading but poor juice yield. They include IS 21991 (21.2), SP 4511-3 (21.0) and SP 4511-2 (20.0). These lines will be used in crossing program. The performance of the top five high-yielding lines for sugar yield is given in Table 3.

BVS Reddy

Table 3. Performance of selected sweet sorghum varieties/restorers (at maturity stage) - 2005 postrainy season at ICRISAT, Patancheru

Variety/restorer	Days to 50% flowering	Plant height (m)	Cane yield (t ha^{-1})	Juice yield (t ha^{-1})	Brix reading at maturity	Sugar yield based on Brix reading and juice yield (t ha^{-1})
SP 4484-2	94	2.0	36.3	18.08	16.0	3.0
SP 4487-3	95	2.1	35.3	14.0	17.7	2.4
SP 4504-2	95	2.2	33.3	14.0	17.3	2.3
SP 4482-2	94	2.2	29.8	13.2	18.7	2.3
SP 4482-1	95	2.0	33.4	13.4	17.0	2.2
SSV 84 (Check)	86	1.5	10.7	2.7	17.3	0.5
NSSH 104 (Check)	86	2.0	19.5	7.6	14.2	1.1
SSV 74 (Check)	88	2.1	18.9	6.5	18.7	1.2
Mean	94	1.9	20.6	7.6	16.2	1.2
CV (%)	1.9	7.9	27.8	23.5	12.0	28.9
CD (5%)	2.85	0.24	9.28	2.90	3.15	0.57

Output target 5A.7: Stay-green QTLs associated with improved fodder quality introgressed into elite sorghum hybrid parents and their potential utility assessed (2010)

Activity 5A.7.1: Mapping and introgression of stay-green QTL into elite parental lines, and assessment of their effects on hybrid performance

Milestone 5A.7.1.1: Assessment of near-isogenic BC_3F_3 and BC_4F_3 stay-green QTL introgression lines completed in R 16 and ISIAP Dorado backgrounds (2010)

Evaluation of early-generation R 16 stay-green QTL introgression lines: We completed two years of field evaluation of the initial (BC_1 and BC_2) stay-green (non-senescent) introgression lines with 1–4 QTL in the background of the Indian postrainy season sorghum variety R 16. The primary objectives of the evaluation were to (1) assess the phenotypic expression of various stay-green QTL under postrainy season conditions, when available (stored) soil moisture is limited, and stress-induced crop senescence is the norm, and (2) assess the effects of stay-green on the ability to maintain grain yield in moisture-deficit environments and on the ruminant nutritional quality of stover. There were differences in the senescence patterns of the various QTL introgression lines, but in general, across all test environments, the average percent green leaf area (% GLA) of all the introgression lines fell between the values of the two parents. Even though a number of the introgression lines contained many of the putative stay-green QTL from B 35, none demonstrated the same degree of stay-green as did B 35, especially in the latter part of the grain-filling period. Nevertheless, the data indicated that the transfer of various stay-green QTL from B 35 to R 16 was successful in improving the stay-green character of the latter, in both stress and non-stress conditions.

There were significant, linear and positive (although somewhat variable) relationships of the ability to maintain normal green leaf area, and normal grain filling with 100-grain mass (r^2 = 0.32 and 0.56) and grain yield (r^2 = 0.34 and 0.76) in two of the three dryland environments. Only in the most severely stressed environment, where virtually all of the lines senesced, were the relationships non-significant. Thus, backcrossing the non-senescence trait into a generally senescent lines should result in improved grain filling and, therefore, improved grain yield in most dryland, postrainy season environments, at least when introgressed into genetic background as senescent as R 16. An attempt to explain differences in various ruminant nutritional quality traits of the stover of the R 16 QTL introgression lines by differences in stay-green indicated no quantitative relationship in four of the five environments. Only in the 2005–2006 supplementally irrigated environment, where the leaf senescence was generally less than that in the other environments, was there a modest relationship between % GLA and stover N% and % GLA and stover digestibility. This was despite six of the introgression lines having a significantly higher mean N% than R 16 across all environments. It may be that a stronger expression of stay-green will be needed in the introgression lines to significantly improved the stover quality.

FR Bidinger, CT Hash and M Blümmel

Milestone 5A.7.1.2: Stay-green QTL mapping of E 36-1 confirmed based on phenotypic assessment of two F_6 RIL populations genotyped with DArT, SSR, and CISP-SNP markers (CTH/SS/SPD/VV, 2010)

Progress to be reported in subsequent years

Milestone 5A.7.1.3: Stay-green QTL introgression sorghum lines based on donor parent E 36-1 available for phenotypic evaluation in two diverse genetic backgrounds (CTH/SPD, 2011)

Progress to be reported in subsequent years

Milestone 5A.7.1.4: Initial evaluation of animal performance on near-isogenic hybrids differing in allelic composition at two stay-green QTLs completed (CTH/SPD/MB/BVSR, 2010)

Progress to be reported in subsequent years

Output target 5A.8: Commercialization of sorghum grains and impact of improved germplasm enhanced

Activity 5A.8.1: Strengthen research and development partnerships, and technology exchange

Milestone 5A.8.1.1: Hybrid parents (>50) and other breeding materials (>100) supplied to NARS and their impact assessed (BVSR/Sorghum Team—annual)

Seed producers sorghum hybrid trial (SPSHT): Public and private sector scientists utilize ICRISAT-bred hybrid parents for developing commercial hybrids. To assess the performance of sorghum hybrids produced by different private and public sector organizations, "Seed producers' sorghum hybrid trials" are constituted and coordinated in rainy and postrainy seasons by ICRISAT as one of the activities of the ICRISAT-Private sector Sorghum Hybrids Parents Research Consortium. Under this activity, SPSHT during the 2006 rainy season with 14 entries was conducted at five consortium members' locations (Biostadt, Aurangabad; MHseeds, Jalna; Emergent Genetics, Hyderabad; Kanchan Ganga and Nuziveedu Seeds (2 locations), Hyderabad; and Tulasi Seeds, Guntur) who volunteered to conduct the trial as well as at ICRISAT, Patancheru. Due to incessant rains at crop maturity stage in Guntur, the grain yields were so low that the trial was lost. Data from the other five locations were analyzed and reports distributed to all consortium members who contributed the hybrids. The results showed that the mean grain yield of MLSH 60 was on par with the check SPH 1342, with 10% larger grains. Among the others, three hybrids viz., BSH 10, BSH 33, and BSH 31 were comparable to the check CSH 16 for grain size.

Sorghum scientists' field days: Field visits were arranged for public and private sector scientists. Sorghum scientists' field day was organized at ICRISAT, Patancheru on 28-29 September 2006 for public and private sector scientists for selection of the breeding materials and to get feedback on the ICRISAT-supplied breeding material. A total of 46 scientists (28 public sector and 18 private sector scientists) participated in the field day and selected 971 distinct lines. A total of 2460 sorghum seed samples (1377 samples including 459 samples of designated hybrid seed

parents selected by 13 public sector scientists and 1083 samples including 365 samples of designated hybrid seed parents selected by 11 private sector scientists) were supplied. The selected progenies/lines (where sufficient seed is not available) are being multiplied in 2006–07 postrainy season to supply them in February 2007.
Seed supplies: A total of 1768 seed samples of hybrid parents/breeding lines were sent to 16 countries. India received 1375 samples followed by Mexico 95 samples. Of the 1375 seed samples to India, 651 were sent to public sector scientists, and 682 to private sector scientists. Seed in bulk quantities (278 kg) of six high-yielding/released cultivars was supplied to 42 farmers. We also supplied 150 kg seed of NTJ 2 and 15 kg seed of SSV 84 to Rusni Distilleries, Hyderabad, for seed multiplication in farmers' fields.

Partnerships with NARS, networks and regional fora strengthened: A salinity-tolerant hybrid trial (consisting of 30 hybrids) and a salinity-tolerant varietal trial (consisting of 29 varieties) were sent for evaluation at ARS, Gangavathi, Karnataka in saline soils. Seed of 12 salinity-tolerant varieties was multiplied and sent to Central Rice Research Institute, Cuttack, India for evaluation in saline soils in farmers' fields.

Seed of sweet sorghum varieties and hybrids was multiplied and sent to National Research Centre for Sorghum (NRCS), Hyderabad, India, for testing at the All India Co-ordinated Sorghum Improvement Project (AICSIP) locations in the 2006 rainy season. Three varieties were tested in advanced sweet sorghum varietal trial (ASSVT); one variety in initial sweet sorghum varietal trial (ISSVT); one hybrid in advanced sweet sorghum hybrid trial (ASSHT) and four hybrids in initial sweet sorghum hybrid trial (ISSHT).

A sorghum grain mold resistance stability nursery (SGMRSN) consisting of 33 ICRISAT-bred grain mold resistant hybrid parents and 22 NARS-bred hybrid parents in elite genetic backgrounds along with the five checks was constituted and sent for evaluation at Akola, Dharwad, Parbhani, Palem and Coimbatore for GMR in the 2006 rainy season. This enabled NARS scientists to select grain mold-resistant hybrid parents suitable for their locations for further use in grain mold resistance improvement programs or for direct utilization in hybrid development.

The use of the breeding materials by private sector scientists: The utilization of ICRISAT-bred sorghum hybrid parents by private sector scientists was assessed through a good mix of formal (structured questionnaires and one-to-one dialogue) and informal means. A social scientist and a sorghum breeder from ICRISAT met the representatives of 16 private sector organizations who are members of sorghum hybrid parents' research consortium.

The preliminary findings indicated that a total of 15 hybrids are being marketed by 10 private sector organizations, with each having marketed at least one hybrid. All of these 15 hybrids involve various proportions of ICRISAT-bred germplasm in at least one of the hybrid parents. Two hybrids have been developed by directly using ICRISAT-bred A- and R-lines; two hybrids have been developed by directly using ICRISAT-bred R-lines; four hybrids involve both parents with 25–50% ICRISAT-bred improved germplasm content; six hybrids involve both the parents with 50–75% ICRISAT-bred hybrid parents' content. One hybrid has been developed by directly using ICRISAT-bred A-line and R-line with 50% ICRISAT-bred improved germplasm. These preliminary results clearly indicate that sorghum hybrids being marketed by private sector organizations are largely based on ICRISAT-bred improved hybrid parents, particularly the A-lines.

Technical information and documents developed and dissemination: The results of SPSHT-2005 rainy season trial were summarized, report prepared and distributed to all the members of sorghum hybrid parents research consortium who contributed hybrids for the trial. Prepared the reports of (a) ICAR-ICRISAT partnership projects and distributed to all the concerned at All India Coordinated Sorghum Improvement Project (AICSIP) group meeting held at Marathwada Agricultural University (MAU), Parbhani during May 2006. Further, two information bulletins on 1) Sorghum grain mold and 2) population improvement in sorghum, book chapter on "Sorghum Hybrid Parents Research at ICRISAT–Strategies and Impacts" were prepared and distributed to all sorghum scientists. Also, some of the ICRISAT-bred sorghum hybrid parents were characterized as per DUS test guidelines of Indian Council of Agricultural Research and published as "Characterization of ICRISAT-bred Sorghum Hybrid Parents" in a special issue of International Sorghum and Millets Newsletter in 2006.

Farmers' preferred cultivars identified: The seed of eight sorghum cultivars preferred by farmers (CSV 15, PVK 801, ICSV 93046, ICSR 93034, SPV 422, NTJ 2, SPV 1411 and SPV 1359) in IGNRM system was multiplied and distributed for evaluation in the watersheds (Sujala and TATA projects) for assessing farmer-preferred varieties of

various crops. The most preferred varieties by farmers were PVK 801 for rainy season and SPV 1411 for the postrainy season.

BVS Reddy

Milestone 5A.8.1.2: Ten sorghum scientists trained biannually (BVSR/Sorghum Team—alternate year)

A learning program on "Sorghum Hybrid Parents and Hybrid Research and Development" is planned to be conducted from 6 to 17 February 2007. About 25 sorghum scientists from both public and private sector organizations from Asia and Africa and Latin America are expected to participate in the program.

BVS Reddy

Milestone 5A.8.1.3: Two thousand farmers adopt improved sorghum cultivars and crop production practices in India, China and Thailand (ASA/ChRR/BVSR/PPR/ CLLG/FW, 2007)

The project titled "Enhancing the utilization of sorghum and pearl millet grains in poultry feed to improve the livelihoods of the small-scale farmers in Asia" funded by CFC–FAO is operational since 2005. The project aims at (i) development of effective coalition of all stakeholders (groups of small-scale sorghum and pearl millet farmers, poultry and feed production farmers, private sector, NGOs, etc.) in order to improve crop productivity and enhance skills in harvesting, bulking, storage and handling practices of grain, (ii) identifying the constraints in sorghum and pearl millet production and to provide information on improved production (aims at packages and seeds of improved cultivars by involving private seed companies), (iii) strengthening input supply chain system (fertilizers, pesticides, seeds of improved varieties, credit facility etc.) for sorghum and pearl millet production and output supply chains to stimulate the use of these crops as raw material for commercial poultry feed production, (iv) develop linkages with other input dealers for credit, seeds, fertilizer, etc., by organizing farmers into groups for effective input delivery mechanisms, and (v) to link farmer groups with poultry feed manufacturing companies and poultry producers to enable the farmers to sell the grain to feed manufacturers for use in manufacturing poultry feed.

As a part of basic requirement for implementation of the project, two clusters in Andhra Pradesh, three clusters in Maharashtra, and one cluster each in China and Thailand have been identified. A total of 10 partners were identified as coalition partners to support the project activities that includes research institutes, state agricultural universities, Krishi Vignana Kendras (KVKs), and private sector companies. The farmers' associations have been formed and necessary support has been extended to these partners. The farmers were trained in the use of improved production technologies, grain bulking, grading and improved grain storage technologies for sorghum and pearl millet.

The project interventions enabled farmers to realize enhanced sorghum productivity in Andhra Pradesh clusters by 70–90% and in Maharashtra clusters by 20–30%; and pearl millet productivity by 90–110% in Andhra Pradesh and Maharashtra clusters through improved production practices. Storage structures have been constructed in the clusters

in India through farmers' participatory approach and being used by cluster farmers for bulking their sorghum and pearl millet grains.

BVS Reddy, AS Alur, CR Reddy, P Parthasarathy Rao,
CLL Gowda and F Waliyar

Milestone 5A.8.1.4: Market linkages for the sale of sorghum grain (100 t) to poultry feed manufacturers by sorghum farmers in India, China and Thailand established (ASA/ChRR/BVSR/PPR/CLLG/FW, 2008)

One of the major activities of the CFC-FAO-ICRISAT project is to link the farmers cultivating sorghum and pearl millet with the processors. Thus, the reasons for distress sale of the produce immediately after the crop harvest to the commission agents/ middlemen in the local market or to the money lenders were studied. The study of the marketing systems in the project area indicated that this distress sale of the farm produce is due to one or more of the following reasons:

- Immediate cash needs of the farmers for family expenses such as education/health/ family functions
- Repayment of loans borrowed from private money lenders
- Lack of storage space for keeping the produce without deterioration in grain quality
- Lack of market intelligence/non-availability of market information

- Difficulties of transport of small quantum of produce to market

The project is promoting the utilization of sorghum and pearl millet for poultry feed in India, China and Thailand. Efforts to link small and marginal farmers to the processors (poultry feed manufacturers and other industrial users) through bulk storage and marketing is being promoted. To overcome the above constraints, the project has facilitated the farmers to get organized into Farmers Associations though participatory approaches and helped them in constructing the storage structures and drier sheds. Panicle driers were also installed in all the cluster villages which were helpful in drying the produce to the required moisture levels.

The farmers have been trained in various aspects of bulk marketing such as aggregation of the produce, scientific storage, godown management, grading of the produce, negotiating the sale price with feed manufacturers and poultry producers, etc.

Bulking and bulk marketing aims at storing the farmers produce in a godown over a period for gaining better market price. Grains were dried to established moisture levels, which could extend their shelf life and prevent the losses from pest and diseases. The drier installed in the clusters were useful in ensuring the drying of the produce. The farmers were able to realize a better price, through enhanced bargaining power, minimization of middlemen charges/exploitation, increased involvement of the farmers and improved market intelligence that helped in expanded market.

In 2006, farmers (association) were linked with the poultry feed manufacturers. During the current season farmers bulked more than 225 t of sorghum and stored in the godowns constructed under the project. The stored produce was sold to M/s Janaki Feeds Private Limited @ Rs 6000–6250 per t from the project villages in the clusters of Maharashtra and Andhra Pradesh. Thus, farmers realized an additional income of Rs 750–1250 per t as compared to the market prices, which were about Rs 5250 per t.

II. Pearl millet

Output target 5A.1: Genetically diverse, high-yielding and downy mildew (DM) resistant pearl millet parental lines of potential grain hybrids (at least 9 each of seed parents and restorer parents) developed annually during 2006-2011

Activity 5A.1.1:Develop and characterize regionally adapted high-yielding and DM resistant hybrid parents

Milestone 5A.1.1.1: Diverse range of high-yielding and DM resistant seed parents and restorer parents developed (KNR/RB/RPT/RS, annual)

Male-sterile line (A-line) development: Development and dissemination of up to nine morphologically diverse A-lines with high grain yield potential and downy mildew resistance annually continues to be making a direct contribution to strengthening the hybrid development programs in the public and the private sector. In 2006, nine 2006-series A-lines (2 A_1 cytoplasm and 7 A_4 cytoplasm) with 40–54 days to flowering, 13–23 cm panicle length and 7.4–13.8 g 1000-seed mass were developed for dissemination. Under high disease pressure in the glasshouse (more than 95% DM incidence in susceptible check 843B), four of these were highly resistant (0–10% DM incidence) to all the five diverse pathotypes (Jodhpur, Jalna, Jamnagar, Durgapura and Patancheru), another four were resistant to at least four of the five pathotypes, and one was resistant to three pathotypes (Jodhpur, Durgapura and Jamnagar). Conversion of 34 elite maintainers of A_1 CMS system into A_4-system A-lines was completed. The sixth and final backcross of these maintainers was completed to convert them into A_5-system A-lines. Seed of these A_4- and A_5-system A-lines is now available for dissemination. About 210 B-lines and their counterpart backcross (BC) progenies were evaluated and backcrossed for further advancement for A-line breeding. Of these, 172 B-lines and their BC_5–BC_{10} progenies (67 A_1, 97 A_4 and 48 A_5) were selected for further advancement. Another set of 165 B-lines and their early generation BC progenies (61A_1, 92A_4 and 84A_5) were evaluated of which 120 B-lines were selected for advancing to BC_2-BC_4. First backcross was made with 11 B-lines, of which 5 B-lines and their backcross progenies (1 A_1, 5 A_4 and 5 A_5) were selected for further backcrossing.

In a continuing effort of pyramiding DM resistance genes into ICMB 89111, all the 260 BC_5F_4 progenies were screened against Patancheru pathotype under heavy disease pressure (>95% DM incidence in susceptible check 81B) under glasshouse condition. About 70 resistant progenies (d_2 dwarf) with <5% DM incidence were evaluated during the 2006 rainy season, of which 12 were selected based on visual assessment for phenotypic resemblance to

ICMB 89111. Seven of the selected progenies flowered in 54 days and 2 in 55 days, while 3 flowered in 56 days (ICMB 89111-P_2 flowered in 54 days and ICMB 89111 flowered in 52 days).

KN Rai and RP Thakur

Restorer line (R-line) development: A restorer development and dissemination strategy, similar to that followed for A-lines, was initiated in 2006. Nine 2006-series R-lines (5 A_1 cytoplasm and 4 A_4 cytoplasm) with 46–60 days to flowering and 11–30 cm panicle length were developed for dissemination. Under high disease pressure in the green house (>95% DM incidence in susceptible check 843B), eight of these were highly resistant (0–10% DM incidence) to four of the five diverse pathotypes (Durgapura, Jodhpur, Jalna, Jamnagar and Patancheru) and one was resistant to two pathotypes (Durgapura and Jodhpur). About 60 A_4 restorer progenies (S_7-S_{12}) were evaluated during the rainy season, of which 42 were selected with 33 of these flowering in 46–60 days (checks ICMR 356 flowered in 49 days and ICMP 451 in 52 days).

In 2005 rainy season, 62 potential A_4 R-lines had been identified based on the limited testcross evaluation of their fertility restoration. These were crossed on 2 A_1 and 6 A_4 male-sterile lines. Results showed that 44 of these were A_1 restorers, 32 were A_4 restorers (fertility restoration proven on at least on 4–6 A-lines), and 24 were dual-restorers. Of these, 25 lines flowered in 49–55 days, and 37 lines flowered in 56–62 days (ICMP 451 flowered in 52 days). All the 62 potential restorers were tested for resistance to Durgapura and Jalna pathotypes, of which 34% were highly resistant (<10% incidence) to Durgapura pathotype and 16% were highly resistant to Jalna pathotype under high disease pressure (susceptible checks ICMP 451 and 834B had >95% DM incidence against both pathotypes). About 80 additional potential restorer progenies were crossed on both 81A_1 and 81A_4 during the postrainy season and the resultant testcross nursery was evaluated for fertility restoration during the rainy season. Of these, 50 showed fertility reaction on 81A_1, 29 on 81A_4, and 11 on both 81A_1 and 81A_4.

Restorer line yield trial: Two dual-purpose restorer progenies with large morphological differences for panicle length/girth and seed size, but expected to have high gain yield were compared with three commercial varieties (ICTP 8203, WC-C 75 and Raj 171) and a commercial hybrid (ICMH 356) for grain yield in a 5-replication yield trial at Patancheru during the 2006 rainy season. Raj 171 was the highest-yielding variety (1533 kg ha^{-1} grain yield) with 49 days to flower, 170 cm plant height, 22 cm panicle length, 1.5 panicles $plant^{-1}$ and 8.4 g of 1000 seed mass. Both inbred lines had 70% of the grain yield of Raj 171, flowered in 52 days, with tillering ability similar to Raj 171, and 30–35 cm shorter height. While the MC 94-derived inbred line had 2 cm longer panicle than Raj 171 and 9.3 g of 1000-seed mass, the long panicle line had 39 cm long panicle and smaller seed (6.7 g 1000-seed mass).

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Milestone 5A.1.1.2: Seed parents and restorer parents adapted to arid zone developed (KNR/RB/FRB/RPT/RS, 2010)

Hybrid parents adapted to western Rajasthan: The Sehgal Foundation provided 3-year funding support in the year 2002 to increase the emphasis on breeding seed parents specifically adapted to arid Rajasthan. During the third year of the project, restorer line breeding was also included in this project. Although the project ended in 2004, the materials generated from this project have been carried forward.

Seed parent progenies: Two types of materials were generated that have been further evaluated and advanced: (i) progenies derived from ICRISAT-CAZRI B-composite (ICCZBC) that was constituted from a set of diallel crosses, and (ii) progenies derived by pedigree breeding in some of the most promising B × B crosses in the above diallel. About 275 S_4 progenies derived from ICCZBC were evaluated during the 2006 rainy season, of which 86 were selected, with 34 of these flowering in 41–50 days and 48 flowering in 51–55 days (checks 843B flowered in 44 days and 81B in 56 days). Another 268 S_3 progenies were evaluated in the summer drought nursery, of which 152

were selected based on the visual agronomic evaluation. Of these, 21 progenies flowered in 46–50 days and 91 flowered in 51–55 days (checks 843B flowered in 47 days and 842B in 55 days).

We also evaluated 191 F_7/F_8 progenies derived from B × B crosses during the 2006 rainy season, of which 90 were selected, with 23 of these flowering in 41–50 days and 53 flowering in 51–55 days (checks ICMB 94555 flowered in 48 days, 842B in 46 days and 843B in 44 days). About 100 F_5 progenies derived from these crosses were also evaluated during the summer season in drought nursery, of which 54 were selected. Of these, 20 flowered in 51–55 days and 34 flowered in 56–60 days (checks ICMB 93111 flowered in 58 days, ICMB 95444 in 55 days and ICMB 93333 in 57 days).

Restorer parent progenies: About 100 progenies (S_6–S_9) were evaluated in the drought nursery during the summer season, of which 40 were selected based on visual assessment for agronomic traits. Of these, 18 flowered in 46–50 days (checks RIB 3135-18 flowered 55 days and H 77/833-2 in 52 days). In addition, 13 progenies (S_8–S_{10}) were evaluated during rainy season and all flowered in 46–55 days (check H 77/833-2 flowered in 44 days).

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Evaluation of hybrid parents progenies: We continued the evaluation in western Rajasthan of four sets of breeding materials bred specifically for adaptation to the arid zone. These included 48 lines from the Mandor Restorer Composite, 46 lines from the ICRISAT-CAZRI Maintainer Composite, 24 crosses of adapted B-lines and 11 restorer populations. We evaluated testcrosses of these lines (made with arid zone-adapted testers) to assess their hybrid potential for grain and stover yields under arid zone conditions. Because of the high degree of inherent variability in rainfall in the arid zone, years differ significantly in the timing and severity of the inevitable periods of drought stress, making multi-year evaluations necessary to separate true genotype differences from genotype × environment interaction effects. The 2006 rainy season at Jodhpur faced a very late arrival of rains (early August) but a normal ending of the rainy season (late September), with a total rainfall of about 200 mm. Flowering in all trials was early (40 days from emergence) and most trials were subject to post-flowering moisture shortage, resulting in grain yields in the range of 600 to 800 kg ha^{-1}. Despite this, there were significant differences among the hybrids for grain and stover yields, and among lines for the general combining ability.

The ICRISAT-CAZRI B-composite was bred by intercrossing maintainer lines thought to be adapted to the arid zone, and is intended to serve as a source of new, higher yielding seed parents for this zone. Ten lines from this composite along with controls ICMA 95111 and HMS 7A had a significant general combining ability (GCA) effect for earliness which is very encouraging. Three of the B-lines, plus control HMS 7A had significant positive GCA for grain yield. Similarly, three B-lines and two controls (ICMA 93333 and HMS 7A), had significant positive GCA for stover yield. The basic limitation of many of the lines was their non-significant GCA for biomass in this very short season environment, which is a common finding in the case of conventionally bred parental lines that have been selected for high grain yield through high harvest index (HI). In this trial, the relationship of GCA for biomass with grain ($r = 0.78$, $P<.001$) and stover ($r = 0.91$, $P <.001$) yield was very high, underlining the requirement for a positive GCA for biomass under arid zone conditions if new parental lines are to produce hybrids with improved grain and improved stover yields. Two of the 46 lines from this composite and the two controls (ICMA 93333 and HMS 7A), met this requirement.

The Mandor Restorer Composite was bred in a similar fashion as the CAZRI-ICRISAT B-composite, as a source of high-yielding restorers for the arid zone. This composite appears somewhat better adapted to the arid zone, judging by the GCAs of the lines extracted from it. Twelve of the lines had significant GCA for early flowering, as did three of the five controls, and between 5 and 7 of the lines had a significant GCA for grain, stover and total biomass yields. The correlation of GCA for grain yield was high both with GCA for total biomass ($r = 0.84$, $P<.001$) and GCA for HI ($r = 0.82$, $P<.001$). GCA for total biomass was the only determinant of GCA for stover yield ($r = 0.78$, $P<.001$). There was one outstanding line (MRC $S_1$107-1-3-B-B-B) that had significant ($P<.01$) GCAs for early flowering, biomass, harvest index and grain and stover yields. None of the standard inbred controls (which included ICMR 356 and ICMR 01004), apart from the Early Rajasthan Restorer Population, came even close in performance.

The B-line cross trial is specifically designed to identify crosses with a positive GCA for total biomass, from which to select new seed parents to make hybrids with improved biomass productivity in the arid zone. The short rainy season constrained the expression of differences in biomass productivity; only two of the 23 crosses (ICMB 93111 ×

ICMB 91444 and ICMB 93333 × ICMB 95111) had a significant (P<0.05) GCA for total biomass. In both cases the crosses also had a significant (P<0.05) GCA for stover yield and positive, if not significant (P<0.20) GCA for grain yield. The only cross with a significant positive GCA for grain yield (ICMB 97555 × ICMB 97111) appeared to achieve this by partially escaping the late season drought, as it had a highly significant (P<0.001) GCA for early flowering. In such a short season, GCA for early flowering was more closely related (r = 0.65, P<.004) to GCA for grain yield than was GCA for biomass (r = 0.54, P<.005) in these materials, which were originally selected for biomass productivity. The data set will be very useful in the ultimate multi-environment analysis, to assess how crosses doing well in more favorable seasons manage a very dry one.

The arid zone restorer populations were bred from diverse arid zone-adapted materials, primarily arid zone landraces, with the intention of providing different source populations from which to breed restorer lines for the arid zone. Because of their landrace backgrounds, these materials are generally good biomass producers; and GCA for grain yield was better related to GCA for harvest index (r = 0.85, P<.001) than to GCA for biomass (r = 0.41, P = .13). GCA for grain yield was also related to GCA for early flowering, and two of the three restorers with a significant GCA for grain yield were early flowering - the Early Rajasthan Restorer Population and the control ICMR 01004 (a DM resistant version of H 77/833-2). However both had a negative GCA for stover yield, which approached significance (P<0.10) in the case of ICMR 01004. The best overall entry was the Jakharana Restorer Population with positive GCAs for biomass (P<0.10), stover (P<.05) and grain yields (P<0.15), despite its hybrids flowering slightly later than the mean of the trial.

FR Bidinger

Output target 5A.2: More than 500 trait-specific and DM resistant improved breeding lines of pearl millet developed and disseminated alternate years (2006, 2008, 2010 and 2012) for use in breeding parental lines of grain hybrids

Activity 5A.2.1: Develop a diverse range of high-yielding and DM resistant trait-specific breeding lines

Milestone 5A 2.1.1: Germplasm with large seed, large panicle and white grain color identified and introgressed (KNR/RB/HDU/RPT/RS, 2009)

Germplasm sources: In search of new germplasm sources for long and compact panicle traits, 25 S_1 progenies (compactness score 7–9 as per the genetic resources characterization data) derived from long panicle germplasm accessions were evaluated during the 2006 postrainy season, of which 18 were selected, producing 76 S_2 progenies for further evaluation during the 2006 rainy season. Of these, 18 progenies were selected, of which 10 flowered in 56–65 days and 2 flowered in 51–55 days (check NCd_2 flowered in 53 days), producing 49 S_3 progenies for further evaluation.

Introgression of panicle and grain traits

Grain size: Large grain size is an important grain yield component and a farmer-preferred trait, especially in Maharashtra state of India. Most of the commercial hybrids and OPVs have 10–12 g of 1000-grain mass, with very few having 15 g of 1000-grain mass. Availability of gemplasm with 19–20 g of 1000-grain mass provides opportunity to develop OPVs and hybrids with 1000-grain mass in excess of 16 g. This germplasm source, however, is highly photosensitive. Recent attempts to introgress this large grain size in the adapted and elite genetic backgrounds have made considerable progress. Based on the visual assessment of 480 F_5 progenies for gain size and other agronomic traits, 45 were selected (producing 95 F_6 progenies) of which 37 flowered in 56–65 days (47 days for the control ICMB 00444). The remaining 8 were late. There were 24 progenies that had more than 15 g of the 1000-grain mass, with 5 of these having 18–20 g of 1000-grain mass. In another large-seeded seed parent nursery, 196 three-way F_3 progenies had been evaluated. Based on grain size and other agronomic traits, 101 progenies were selected, of which 73 flowered in 51–60 days. There were four progenies that flowered in 46–50 days. Others were late. There were 48 progenies that had more than 15 g of 1000-grain mass, with 6 of these having 18–20 g of 1000-grain mass.

Panicle length: The panicle length of most of the commercial hybrids is 20–25 cm and it rarely exceeds 30 cm. A large number of improved breeding lines developed at ICRISAT have 30–40 cm of panicle length. With panicles as

long as 140 cm available in the germplasm, opportunities exist to develop improved breeding lines with 60–80 cm of panicle length. These germplasm accessions, however, are late maturing (70–80 days to flowering) and tall (>250 cm), and have sparse spikelet density in the basal portion of the panicles. Introgression of this long panicle from the germplasm into elite and adaptive backgrounds has met with considerable success. For instance, in restorer parents research, about 645 long spike progenies (F_3-F_8) were evaluated during the postrainy season, of which 164 were selected based on visual assessment for agronomic performance to generate 352 progenies (F_4-F_9) for further advancement. These 352 progenies along with 90 progenies (F_4) coming from rainy season 2005 evaluation were evaluated during the 2006 rainy season. Of these, 105 were selected based on the visual assessment of spike length and agronomic score for further evaluation. About 90 of these flowered in 56–65 days with panicle length ranging between 24 and 83 cm, while 11 progenies flowered in 51–55 days with panicle length ranging from 32 to 48 cm (check NCd_2 flowered in 54 days and had panicle length of 41 cm). There were 13 progenies that had 60–83 cm panicle length.

With the objective of getting good tillering, compact panicles with good exsertion and large seed in long panicle progenies, a crossing program was undertaken during the 2005 summer season. Nine long panicle dwarf progenies (F_4-F_6) were used as female parents and crossed with 17 restorer progenies (F_4-F_9) possessing desirable traits with respect to panicle compactness, good exsertion and tillering. The resulting 124 crosses were evaluated in hybrid observation nursery during the 2005 rainy season, of which 74 were selected based on visual assessment for agronomic traits. Out of 74 F_2s produced, five F_2s derived from F_1s with compact panicles were evaluated in 50-row plots each during the 2005 summer season (all flowering in 55–60 days). Based on visual assessment for agronomic performance, single plant selections were made in four F_2s to generate 260 F_3 progenies for further advancement. Apart from these, another 12 F_2s derived from F_1s with compact panicles were evaluated in 10-row plot each during the 2006 rainy season, of which four flowered in 51–55 days and eight in 56–60 days (check NCd_2 flowered in 53 days). Single plant selections were made in all F_2s to generate 170 F_3 progenies for further advancement.

Grain color: The consumer preference for white grain color in pearl millet stills remains to be ascertained, though the flour of such grains is presumed to be more acceptable for blending with wheat flour. Excellent sources of white seed color are available in the germplasm, but these are highly photosensitive and have small grains. A recent low-key initiative to introgress this color in the elite and adapted genetic backgrounds led to the production and evaluation of 110 progenies, most of which were late, flowering in more than 65 days. Sixteen progenies were selected (two of these flowering in 51–60 days), primarily based on white grain color, that produced 32 F_4 progenies, which will be further evaluated for selecting those stable for enhanced levels of white grain color.

KN Rai and HD Upadhyaya

Milestone 5A.2.1.2: Genetically diverse trait-specific (eg., large seed, large panicle size, diverse maturity and height) advanced breeding lines developed and disseminated (KNR/RB/RPT/RS, 2006, 2008, 2010, 2012)

Trait-specific seed parent progenies: High grain yield and high levels of DM resistance are common denominators for developing improved breeding lines of pearl millet at ICRISAT. Apart from this, users of this improved germplasm both in the public and the private sector hybrid breeding programs lay considerable emphasis in selecting lines with specific traits, including plant types of the parental lines of some of the commercial hybrids. Thus, improved breeding lines, mostly those at the later stages of the inbreeding, are classified and evaluated in trait-specific nurseries for easy and more focused evaluation, and selection for further utilization. These trait-specific nurseries are continually updated with the new materials generated almost every year.

Early-maturing progenies: We evaluated 430 advanced generation progenies ($S_8/F_8/F_{10}$), of which 226 were selected based on visual assessment of agronomic traits and yield potential. Of these, 72 progenies flowered in 41–45 days and 145 progenies flowered in 46–55 days (checks 843B flowered in 45 days and 842B in 46 days). About 45 progenies were screened against Durgapura pathotype under high disease pressure in the glasshouse (susceptible checks 842B and 843B had 100% DM incidence), of which 17 progenies had <10% DM incidence. In addition, we also evaluated 1140 F_3 progenies, of which 664 progenies were selected based on earliness and visual assessment of yield potential and agronomic traits. Of these, 95 progenies flowered within 40 days and 475 progenies flowered in 41–50 days (checks 843B flowered in 40 days and 842B in 46 days). More than 2100 F_4 progenies produced from the selected F_3s were evaluated for resistance to Durgapura pathotype under high disease pressure in the glasshouse.

Of these, 1170 progenies were highly resistant (<10% DM incidence), and another 300 progenies had 11–20% DM incidence.

Large-seeded progenies: About 140 advanced generation (F_{10}-F_{12}) progenies were evaluated, of which 87 were selected based on visual assessment for large seed and agronomic traits. Of these, 64 progenies flowered in 46–55 days (ICMB 00444 flowered in 47 days). There were two lines that flowered in 40 days. The selected progenies were screened for DM incidence against Durgapura and Jalna pathotypes under high disease pressure (>90% incidence in susceptible checks) in glasshouse condition. Out of 87 progenies, 52 were highly resistant (0–10% DM incidence) to Durgapura pathotype and 56 were highly resistant to Jalna pathotype.

Long panicle progenies: We evaluated 240 advanced generation progenies ($S_8/F_8/F_{12}$), of which 107 were selected based on visual assessment for panicle length and agronomic performance. Of these, 88 progenies flowered in 56–65 days. There were 11 progenies that flowered in 46–55 days (checks 81B and ICMB 04111 flowered in 55 and 59 days, respectively). Out of 107 selected progenies, 45 were screened against Durgapura and Jalna pathotypes under high disease pressure (susceptible check ICMP 451 had >95% DM incidence), of which 15 were resistant (<20% DM incidence) to Durgapura pathotype, 19 were resistant to Jalna pathotype, and they flowered in 56–65 days. In addition, we evaluated 550 progenies ($S_3/F_5/F_6$), of which 150 progenies were selected. Of these, 128 progenies flowered in 56–65 days (check 81B and ICMB 04111 flowered in 55 and 59 days, respectively). There were 6 progenies that flowered in 51–55 days. Of these selected progenies, 40 had been evaluated against Durgapura pathotype under high disease pressure. Interestingly, all the progenies were highly resistant (<10% DM incidence) and most of them flowered in 51–65 days. We also evaluated 1100 early generation progenies (F_3), of which 320 progenies with 45–60 cm panicle length were advanced based on visual assessment of grain yield potential and morphological characters, producing 950 F_4 progenies.

Thick panicle progenies: We evaluated 386 advanced generation progenies ($S_6/S_7/F_{10}$ onward), of which 160 were selected based on visual assessment for panicle thickness and agronomic performance. Of these, 96 progenies flowered in 51–60 days and 3 flowered in 46–50 days (HHVDBC, the dwarf population, flowered in 55 days).

Other traits: Besides the above traits of primary breeding significance, there are other traits, which are important in breeding and should be combined with the primary traits to the extent possible. These so-to-speak secondary traits initially are useful genetic resource. Thus, improved breeding lines with very dwarf plant height (45–80 cm tall, but 30–50% of this being the panicle length), compact panicles, and erect growth habit have been developed.

We evaluated 52 advanced generation d_2 dwarf progenies (F_{10} and beyond), of which 33 were selected based on agronomic traits and yield potential, with 24 of these flowering in 46–55 days (ICMB 96555 flowered in 52 days). Similarly, we evaluated 27 advanced generation progenies ($S_5/F_7/F_{10}$), of which 19 were selected based on the panicle compactness and visual assessment of agronomic traits with 14 of these flowering in 46-55 days (check 841B flowered in 51 days). We also evaluated 28 advanced generation progenies (F_9 and beyond), of which 21 were selected for sturdy and erect plant type with 12 of these flowering in 51–60 days (checks 81B flowered in 56 days and ICMB 97111in 48 days).

Specific seed parent-type progenies: We evaluated 142 advanced generation progenies (F_9 and beyond) resembling the broad morphological frames of some of the most popular commercial seed parents (eg., 843A and ICMA 89111), of which 110 progenies were selected with 84 of these flowering in 46–55 days and 6 flowering in 41–45 days (checks 843B flowered in 39 days, ICMB 89111 in 50 days, 81B in 56 days and ICMB 01222 in 55 days).

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Trait-specific restorer progenies: Trait-based seed parent breeding has proved useful in comparative evaluation of breeding lines and their utilization in breeding programs both at ICRISAT and in the NARS and the private sector breeding programs. Thus, a similar approach of trait-based breeding is being followed in restorer development program as well. The trait-based approach is also being followed to develop breeding lines perceived to be adapted to very contrasting agro-ecoregions.

Large-seeded progenies: We evaluated 5 progenies (S_8-S_9) during the 2006 postrainy season. Based on visual assessment for agronomic performance, 4 progenies were selected. This nursery was further upgraded by adding 41 progenies (S_4-S_{10}) from various other restorer parent nurseries. Thus, a total of about 45 progenies (S_5-S_{11}) were evaluated during the 2006 rainy season, of which 34 were selected with 7 of these flowering in 40–50 days and 20 flowering in 51–55 days.

High-tillering progenies: We evaluated 30 progenies (S_9-S_{11}) during the 2006 postrainy season. These were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451 and 834B had 95–98% DM incidence against Durgapura and Jalna pathotypes), of which 16 were highly resistant to Durgapura pathotype, 8 were resistant to Jalna pathotype, and 5 were resistant to both pathotypes (<10% DM incidence). Based on the visual assessment for agronomic performance, and DM incidence, 22 progenies were selected to generate 26 progenies (S_{10}-S_{12}). This nursery was further upgraded by adding 191 progenies (S_3-S_{11}) from various other restorer parent nurseries. Thus, a total of about 215 progenies (S_4-S_{12}) were evaluated during the rainy season, of which 145 were selected with 75 of these flowering in 51–55 days and 20 flowering in 46–50 days (checks ICMR 356 flowered in 50 days and IPC 1268 in 60days).

Thick panicle progenies: We evaluated 12 progenies (S_9-S_{10}) during the 2006 postrainy season. These were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451, and 834B had 95 and 98% DM incidence against Durgapura and Jalna pathotypes respectively) in glasshouse. Of these, 5 were highly resistant to Durgapura pathotype, 4 were resistant to Jalna pathotype and 3 were resistant to both pathotypes (<10% DM incidence). Based on visual assessment for agronomic performance and DM incidence, 7 progenies were selected to generate 9 progenies (S_{10}-S_{11}). This nursery was further upgraded by adding 51 progenies (S_3-S_{10}) from various other restorer parent nurseries. Thus, a total of about 60 progenies (S_4-S_{11}) were evaluated in rainy season 2006, of which 40 were selected with 27 of these flowering in 51–55 days and 4 flowering in 46–50 days (check IPC 1518 flowered in 52 days).

Compact panicle progenies: We evaluated 25 progenies (S_9-S_{11}) during the 2006 postrainy season. These progenies were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451, and 834B had 95–98% DM incidence against Durgapura and Jalna pathotypes) in glasshouse. Of these, 9 were highly resistant to Durgapura pathotype, 4 to Jalna pathotype and 2 were resistant to both pathotypes (<10% DM incidence). Based on visual assessment for agronomic performance and DM incidence, 10 progenies were selected to generate 13 S_{10}-S_{12} progenies. This nursery was upgraded by adding 143 progenies (S_3-S_{11}) from various other restorer parent nurseries. Thus, a total of about 155 progenies (S_4-S_{12}) were evaluated during the 2006 rainy season, of which 100 were selected, with 73 of these flowering in 51–60 days and 14 flowering in 40–50 days (checks ICMP 451 and IPC 1518 flowered in 52 days

Early-maturing progenies: Most of the restorer parent progenies are of medium to mid-late maturity as these are largely derived from improved populations and open-pollinated varieties, which were developed for these maturities. Although these are useful for breeding restorers of dual-purpose hybrids, there has been considerable demand from user programs both in the public and the private sector for early-maturing restorers. Thus, a major initiative has recently been undertaken to breed early-maturing restorer lines.

We evaluated 70 progenies (S_4-S_{10}) during the 2006 postrainy season. These were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451, and 834B had 95-98% DM incidence against Durgapura and Jalna pathotypes) in glasshouse. Of these, 27 were highly resistant to Durgapura pathotype, 19 were resistant to Jalna pathotype, and 8 were resistant to both pathotypes (<10% DM incidence). Based on earliness, visual assessment for agronomic performance, and DM incidence, 44 progenies were selected to generate 61 progenies (S_5-S_{11}). This nursery was further upgraded by adding 186 progenies (S_3-S_{12}) from various other restorer parent nurseries. Thus, a total of about 250 progenies (S_4-S_{13}) were evaluated during the 2006 rainy season, of which 180 were selected, with 63 of these flowering in 46–50 days and 94 flowering in 51–55 days (checks EEBC 407 flowered in 42 days and ICMR 356 in 49 days). Additionally, 39 progenies (all flowering in 46–50 days) were added to further upgrade this nursery.

In another effort, we evaluated 50 progenies (S_3-S_{12}) selected from various trait-specific groups during the 2006 postrainy season to constitute an early-maturing restorer composite. These progenies were also screened against

Durgapura and Jalna pathotypes under high disease pressure (ICMP 451 and 834B had >95% DM incidence against Durgapura and Jalna pathotypes). Of these, 13 progenies were highly resistant to Durgapura pathotype and 5 to Jalna pathotype. These progenies were random mated using bulk pollen from all the entries. Based on agronomic scores and DM incidence, crossed seeds from 42 entries were harvested and the hybrids (equivalent to topcross) were evaluated during the 2006 rainy season in unreplicated 4-row plots. Of these, 38 plots were selected, of which 32 flowered in 40–45 days and six in 46–50 days (checks EEBC 407 flowered in 42 days and ICMR 356 in 49 days). Selfing was done in these plots up to 46 days after sowing to select early-maturing plants and produce F_2 seed.

Dual-purpose progenies: We evaluated 140 progenies (S_4-S_{11}) during the 2006 postrainy season. These progenies were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451 and 834B had 95–98% DM incidence against Durgapura and Jalna pathotypes) in glasshouse. Of these, 78 progenies were highly resistant to Durgapura pathotype, 41 were resistant to Jalna pathotype, and 30 were resistant to both pathotypes (<10% DM incidence). Based on visual assessment for agronomic performance and DM incidence, 37 progenies were selected to generate 47 progenies (S_5-S_{12}). This nursery was further upgraded by adding 54 progenies (S_4-S_{11}) from various other restore parent nurseries. Thus, a total of about 100 progenies (S_5-S_{12}) were evaluated during the 2006 rainy season, of which 55 were selected, with 45 of these flowering in 56–65 days and 11 flowering in 46–55 days (checks ICMP 451 flowered in 53 days and HTP 94/54 in 54 days). This nursery was further refined during the 2006 rainy season by constituting three sub-groups (medium-height, medium-tall and tall). Based on visual assessment for height and other agronomic traits, 630 dual-purpose progenies were selected from various other restorer nurseries (including this one). Of these, 350 belonged to medium-height, 200 to medium-tall and 80 to tall groups. In the medium-height group, 255 progenies flowered in 51–60 days and about 60 flowered in 46–50 days. In the medium-tall group, 148 flowered in 51–58 days; and in the tall group, 46 progenies flowered in 51–60 days. The remaining progenies in all the groups flowered in more than 60 days.

Other traits: Besides the above traits of primary importance, there are other traits that add value to the breeding materials, and they also serve as sources of germplasm in elite genetic backgrounds for use in breeding programs. Some of these are: lodging resistance, stay-green and erect growth habit. Thus, about 40 progenies (S_8-S_{10}) were evaluated during the 2006 postrainy season for agronomic traits and more specifically for lodging resistance. These progenies were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451 and 834B had 95–98% DM incidence against Durgapura and Jalna pathotypes) in glasshouse. Of these, 13 were highly resistant to Durgapura pathotype and 3 to both pathotypes (<10% DM incidence). Based on visual assessment for agronomic performance and DM incidence, 21 progenies were selected to generate 24 progenies (S_9-S_{11}). This nursery was further upgraded by adding 18 progenies (S_3-S_{11}) from various other restorer parent nurseries. Thus, a total of about 42 progenies (S_4-S_{12}) were evaluated in 2006 rainy season, of which 23 were selected, with 16 of these flowering in 51–60 days and 4 flowering in 46–50 days (checks ICMR 356 flowered in 50 days and ICMP 451 in 52 days). Similarly, 100 progenies (S_5-S_{12}) were tested in the postrainy season drought nursery for stay-green trait, of which 44 progenies were selected based on visual assessment for stay-green and agronomic performance, with 33 of these flowering in 56–60 days and 3 flowering in 46–55 days (check ICMR 356 and ICMP 451 flowered in 56 days). These were also evaluated in the breeding nursery, of which 70 progenies were selected and further evaluated during the 2006 rainy season. Of these, 46 were selected, with 32 flowering in 51–60 days and 2 progenies flowering in 46–50 days (checks ICMR 356 flowered in 50 days and ICMP 451 in 51 days). We also evaluated 82 advanced generation progenies (S_5-S_{11}) for erect growth habit, of which 58 were selected, with 39 of these flowering in 51–60 days and 12 progenies flowering in 46–50 days (check IPC 804 flowered in 50 days).

Specific restorer-type progenies: Parental lines of commercial hybrids or lines identified as good general combiner and with farmer-preferred traits occasionally become reference lines with users of ICRISAT-bred lines requesting seed of breeding lines depicting traits or trait combinations of the reference lines. ICMR 356 (restorer of a commercial hybrid) with short height, high tillering; and IPC 804 with short height, long panicles, and erect growth habit are two such reference lines that were used for grouping of restorer parent progenies. We evaluated about 50 ICMR 356 type progenies (S_3-S_{11}) during the 2006 postrainy season These progenies were also screened against both Durgapura and Jalna pathotypes under high disease pressure (susceptible checks ICMP 451 and 834B had 95–98% DM incidence against Durgapura and Jalna pathotypes). Of these, 15 were highly resistant to Durgapura pathotype, 10 to Jalna pathotype, and 4 to both pathotypes. Based on visual assessment for agronomic performance and DM incidence, 24 progenies were selected to generate 35 progenies (S_4-S_{12}). This nursery was further upgraded by adding 66 progenies (S_3-S_{11}) from various other restorer parent nurseries. Thus, a total of about 100 progenies

(S_4-S_{12}) were evaluated in rainy season 2006, of which 56 were selected, with 23 of these flowering in 46–50 days and 28 flowering in 51–55 days (check ICMR 356 flowered in 49 days). Similarly, we evaluated 85 IPC 804 type progenies (S_4-S_{11}) during the postrainy season. These progenies were also screened against both Durgapura and Jalna pathotypes under high disease pressure. Of these, 37 were highly resistant to Durgapura pathotype, 13 to Jalna pathotype, and 9 to both pathotypes (<10% DM incidence). Based on visual assessment for agronomic performance and DM incidence, 40 progenies were selected to generate 50 progenies (S_5-S_{12}). This nursery was further upgraded by adding 48 progenies (S_3-S_{11}) from various other restorer parent nurseries. Thus, a total of about 100 progenies (S_4-S_{12}) were evaluated in rainy season 2006, of which 62 were selected, with 44 of these flowering in 46-55 days (check IPC 804 flowered in 50 days). Another 190 progenies, of which 142 flowered in 46–55 days, were identified during the 2006 rainy season to further upgrade this group.

Restorer parent progenies with specific agro-ecological adaptation: The two most contrasting agro-ecoregions in India that require different plant types and seed traits are Rajasthan (specifically western Rajasthan) and Maharashtra. Thus, for Rajastahn adaptation (largely high-tillering type with small to medium seed size), we evaluated 87 (S_7-S_{12}) progenies, of which 60 were selected, with 47 of these flowering in 46–55 days and one progeny flowering in 40 days (checks H 77/833-2 flowered in 44 days and ICMR 356 in 47 days). About 300 additional progenies were identified as Rajasthan-adapted type and added in the group for further evaluation, of which 150 flowered in 51–55 days and 95 in 46–50 days. For Maharashtra adaptation (mostly *iniari* type with dark gray color and medium to large grains), we evaluated 26 advanced generation progenies (S_7-S_{12}), of which 14 were selected, with 13 of these flowering in 46–55 days (check ICTP 8203 flowered in 47 days). During the rainy season, two sub-groups within the *iniari* type (early and medium height) comprising of 320 progenies were constituted based on days to 50% flowering and visual assessment for plant height and agronomic performance. Out of 320 progenies, 22 belonged to early group and 300 to medium-height group. Among early-maturing progenies, all flowered in 40–50 days, while of those in the medium-height group, 50 flowered in 40–50 days and 165 flowered in 51–55 days.

Genetic diversification of restorer lines: As in the seed parent breeding program, evaluation of restorer parent progenies is also done by planting them according to their parentage during the initial stage of inbreeding and selection. Though these materials are carried forward in this structure, some of the most promising ones, mostly at F_5 and beyond, are also included in the trait-specific groups until all those surviving selections up to the final stage are grouped into trait-specific classes or rejected in the final evaluation.

Early-generation progenies (S_1-S_3): This consisted of *iniari* type progenies (S_1/S_2) from ICTP 8202 and LaGrap, and dual-purpose progenies from seven populations. We evaluated 107 progenies from ICTP 8202 in the postrainy season drought nursery, of which 56 were selected based on agronomic performance, with 33 of these flowering in 51–55 days and 16 flowering in 40–50 days (check ICMR 356 flowered in 56 days). About 120 S_2s were generated for further evaluation. In LaGrap, we evaluated 106 progenies (S_1/S_2) during the rainy season, of which 67 were selected with 11 of these flowering in 40–45 days and 35 flowering in 46–50 days (check ICMP 451 flowered in 52 days). About 95 (S_1) progenies were screened against Durgapura pathotype in glasshouse under high disease pressure (susceptible check ICMP 451 had >95% DM incidence), of which 65% progenies had <10% DM incidence. In the dual-purpose group, we evaluated 852 S_1/S_3 progenies derived from seven populations (including OPVs), of which 238 were selected, with 27 of these flowering in 46–50 days and 181 flowering in 51–60 days (checks ICMP 451 flowered in 52 days and HTP 94/54 flowered in 53 days).

Advanced generation progenies (S_5-S_{11}): This also consisted of both *iniari* type progenies derived from GB 8735 and non-*iniari* type progenies from four OPVs. Of the 67 progenies derived from GB 8735 and evaluated in 2006 rainy season**,** 41 were selected with 18 of these flowering in 51–60 days and 9 flowering in 46–50 days (checks ICMR 356 flowered in 50 days and IPC 1518 in 53 days). In the dual-purpose group, we evaluated 770 S_5/S_{11} progenies, of which 248 progenies were selected, with 52 of these flowering in 46–50 days and 174 flowering in 51–60 days (checks ICMP 451 flowered in 51 days and HTP 94/54 in 53 days). The DM resistance level of the material in this nursery seems quite high for Durgapura pathotype as the evaluation of a subset of 196 progenies belonging to JBV 2 and JBV 3 during summer season against this pathotype under high disease pressure (check ICMP 451 had >95% DM incidence) showed that 69% of it were highly resistant (<10% DM incidence).

A_5 restorer development: About 120 F_4 progenies derived from 6 F_2 populations in the cytoplasmic background of the A_5 and having the A_5 restorer gene(s) were evaluated, of which 63 were selected based on the visual assessment of agronomic traits. In addition, 65 F_2 populations were generated from fertile F_1 plants that had been produced by crossing elite inbred lines on fertile plants of eight initial F_2 populations during the 2005 summer season.

Two F_2 populations (IPC 1617 × SDMV 90031-S_1-84-1-1-1-1 and IPC107 × ICMV 91059 S_1-14-2-1-1-2) with A_5 cytoplasmic background and having A_5 restorer gene(s) were screened against Durgapura pathotype under high disease pressure during the postrainy season. These had 13 and 51% DM incidence, respectively. About 600–700 DM-free F_2 plants from these populations were transplanted, selecting 104 plants (66 and 38 plants, respectively) that were evaluated during the rainy season. Of these, 51 progenies were selected (39 and 12 progenies, respectively), based on the visual assessment for agronomic performance. Of these, 45 progenies flowered in 51–60 days and 4 in 46–50 days (checks ICMR 356 flowered in 51 days and ICMP 451 in 52 days).

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Genetics of panicle and grain size: Panicle length and girth (determinants of the seed number per panicle) and grain mass are the two of the important yield components of grain yield. Numerous studies have investigated the genetics of these three traits, but never before with as large parental contrasts as those now available in the ICRISAT-bred lines. Thus, a renewed effort is underway to investigate the genetics of these three traits using lines that have 70 cm of panicle length, >45 mm of panicle diameter and >16 g of 1000-grain mass. During the 2005 postrainy season, four parents of almost similar maturity but with large contrasts for these traits were crossed to produce 6 F_1s (2 for each trait). During the rainy season, all the parental lines along with their F_1s, F_2s and backcrosses (seed produced in the glasshouse) were evaluated in a randomized complete block design with three replications. Observations on quantitative traits like plant height, number of productive tillers, panicle length and diameter were recorded on all the plants in F_2s and backcross progenies, while these observations were recorded on 30 randomly selected plants in parental lines and F_1s. The data on 1000-grain mass is yet to be recorded. Additionally, seeds of triple testcross (TTC) progenies have been produced during rainy season by crossing more than 60 F_2 plants as male parent onto respective parents and their F_1s, which will be evaluated during the 2007 postrainy season to provide the additional information on the inheritance pattern of the targeted traits.

KN Rai

Milestone 5A.2.1.3: An elite B-composite and an elite R-composite with resistance to multiple pathotypes of downy mildew populations developed (KNR/RB/RPT/RS, 2009)

Output target 5A.3: Morphological and molecular diversity of more than 150 elite inbred lines of pearl millet assessed and the relationship between diversity and yield heterosis demonstrated (2009)

Activity 5A.3.1: Evaluate parental lines, advanced breeding lines and their hybrids for grain yield, and morphological and molecular diversity

Milestone 5A.3.1.1: Designated seed parents and restorer lines characterized for DUS traits and molecular diversity (KNR/RB/RV, 2008)

Two-season characterization of 108 A/B pairs and 88 restorer lines for 26 characters from replicated trials was completed. The data were computerized in a catalogue format and field photographs have been taken for the A/B lines. The data computerization for the R-lines and their field photographs are yet to be completed.

KN Rai, Ranjana Bhattacharjee and Rajeev Varshney

Milestone 5A.3.1.2: Selected hybrid parents and advanced breeding lines characterized for morphological and molecular diversity, and yield heterosis (KNR/RB/RV, 2008)

This research is to start in 2007.

KN Rai, Ranjana Bhattacharjee and Rajeev Varshney

Milestone 5A.3.1.3: Two medium-maturity heterotic gene pools based on molecular marker diversity constituted (KNR/RB/SC/RV, 2012)

This project is to start in 2007.

KN Rai, Ranjana Bhattacharjee, S Chandra and Rajeev Varshney

Output target 5A.4: QTLs for downy mildew resistance in pearl millet identified, compared to those previously detected, and their effect on DM resistance assessed (2008)

Activity 5A.4.1: Development of mapping populations and QTL mapping of DM resistance

Milestone 5A.4.1.1: QTL mapping based on F_6 RILs and $F_{2:4}$ progenies from two crosses completed and results compared (CTH/SS/RPT/RS, 2008)

To be reported in 2008

*Milestone 5A.4.1.2: Genetically diverse parents of mapping populations identified and crossed to generate F_6 RILs (CTH/KNR/RPT/RS, 2007***)**

To be reported in 2007

Activity 5A.4.2: Map-directed conventional backcrossing and marker-assisted backcrossing of DM resistance QTLs into parental lines of hybrids

Milestone 5A.4.2.1: Ten major QTL imparting resistance against specific DM pathtypes identified (CTH/RPT/RS, 2007)

To be reported in 2007

Milestone 5A.4.2.2: Near-isogenic lines containing different DM resistance genes (QTL) developed (RPT/RS/CTH, 2010)

Near-isogenic lines of pearl millet possessing resistance genes effective against different pathotypes of DM can be used as a set of host differentials for monitoring virulence shift in the pathogen isolates. A number of DM resistance QTLs effective against different pathotypes have been mapped from different resistance sources and introgression of these QTLs into elite B-lines is in progress. During 2006, we evaluated in greenhouse about 560 backcross progenies in different generations ($BC_{3F1.F2}$ to BC_{6F1}) against two pathotypes Sg 298 (New Delhi) and Sg 409 (Patancheru). These progenies carried the DM resistance QTLs from IP 18293, P 1449-2 and 863B-P_2 that were introgressed into three elite hybrid parents 843B, 81B and 841B. About 15 to 30% of the progenies in the phenotypic background of the above three B-lines were found resistant (≤10% incidence) compared to 84–99% incidence on the susceptible checks. Several other DM resistance QTLs have been introgressed into 843B and these are at different stages of development. Detailed data analysis is in progress.

RP Thakur and CT Hash

Milestone 5A.4.2.3: QTL with known effects against diverse pathotypes pyramided in 843B and other parental lines and their resistance levels determined (CTH/RPT/RS, 2010)

QTL with known effects against diverse pathotypes pyramided in 843B and other parental lines and their resistance levels determined (CTH/RPT/RS, 2010) Single QTL introgression continued (see Milestone 5A.4.2.4 below) to maximize recovery of hybrid parental line recurrent parent genetic backgrounds, which must be completed before initiation of a crossing and marker-assisted selection program to pyramid QTLs from different sources in common recurrent parental line backgrounds in a species such as pearl millet where marker distribution and density is less than optimal.

Milestone 5A.4.2.4: Several different single-QTL introgression homozygotes available in genetic backgrounds of two elite seed parents (CTH/RPT/RS, 2007)

A major QTL for downy mildew resistance, from linkage group 4 of 863B-P2, was advanced from BC6F1 to BC6F2/BC7F1 pairs and from BC5F2 to BC5F3 progenies in the genetic background of ICMB 841. Backcrossing advanced by two generations to BC4F2/BC5F1 pairs for several downy mildew resistance QTLs from donor P1449-2-P1 in the genetic background of 843B. The resistant allele for one of these QTLs is linked to the tall height allele at the d2 dwarfing gene locus on pearl millet linkage group 4. Similarly, backcrossing advanced by one generation to BC3F2/BC4F1 pairs for several downy mildew resistance QTLs from donor IP 18293 in the genetic background of elite seed parent maintainer lines 81B and 843B.

Milestone 5A.4.2.5: Several different multiple-QTL introgression homozygotes available in genetic backgrounds of an elite restorer line and three diverse elite seed parents (CTH/TN/SS/RPT/RS, 2009)

Crossing was initiated for marker-assisted backcrossing programs to introgress downy mildew resistance QTLs from several donor parents (including P1449-2, P310-17, ICMB 90111-P6, PRLT 2/89-33 and 863B-P2) into the genetic backgrounds of several elite hybrid parental lines chosen by national program partners in India (including restorer lines J 2340, H 77/29-2 and ICMR 01004 as well as seed parent maintainer lines HMS 7B, ICMB 93333, and ICMB 95444).

Output target 5A.5: Virulence changes in pearl millet DM pathogen populations determined (2009)
Activity 5A.5.1: Conduct field and laboratory studies to monitor the nature of virulence change in DM pathogen populations

Milestone 5A.5.1.1: Ten to fifteen DM isolates each from Gujarat, Rajasthan, Maharashtra and Uttar Pradesh characterized for pathogenicity and virulence (RPT/RS/KNR/RB, 2008)

Characterization of isolates of *Sclerospora graminicola* for pathogenicity and virulence: Isolates collected from highly susceptible pearl millet cultivars during on-farm survey are characterized for pathogenicity and virulence. Highly virulent isolates thus identified are used for screening breeding lines for developing downy mildew resistant hybrid parental lines.

Pathogenic variation: Eleven isolates collected from five districts (Jamnagar, Anand, Kheda, Banaskantha and Gandhinagar) of Gujarat during the on-farm survey of 1998–2005 were established on potted pearl millet seedlings of 7042S in a glasshouse. The pathogenicity and virulence diversity of these isolates were determined by inoculating the pot-grown seedlings of a set of eight host differential lines (P 7-4, P 310-17, 700651, 7042R, IP 18292, IP 18293, 852B and ICMP 451) and a susceptible check 7042S. The experiment was conducted with 11 isolates, 9 host differentials and 3 replications with 30–35 seedlings/replication in a completely randomized design (CRD). The latent $period_{50\%}$ was recorded from 4th day onwards after inoculation and disease incidence 2 weeks after inoculation. Based on latent $period_{50\%}$ and disease incidence, virulence index (disease incidence × latent $period^{-1}$) was calculated to measure the quantitative virulence of the isolates. The results indicated that of the 11 isolates Sg 441 and Sg 445 (from Banaskantha) were the more virulent with mean incidence of 78–84% across 9 host differentials, and Sg 348 (from Anand) was the least virulent (26% incidence). The dendrogram based on the virulence index using the Euclidean square distances (the average linkage cluster analysis) classified these isolates (at 90% similarity coefficient) into four groups- Sg 200, Sg 348 and Sg 442 in Gr 1 (less virulent); Sg 432, Sg 438, Sg 439, and Sg 440 in Gr 2 (moderately virulent); Sg 435 and Sg 437 in Gr 3 (virulent) and Sg 441 and Sg 445 in Gr 4 (highly virulent). The isolate Sg 445 is currently being used to screen breeding lines targeted for Gujarat.

RP Thakur

Effectiveness of pathotypes mixture for screening breeding lines: Development of improved breeding lines as potential parental lines (A-, B- and R-lines) of hybrids with high level of DM resistance has been the major research focus at ICRISAT. These breeding lines are routinely screened against individual pathotypes in succession to identify those with resistance to single or multiple pathotypes. Many times, a question is asked what if the pearl millet lines could be evaluated against a mixture of pathotypes instead against individual pathotypes in order to reduce time and save resources. To address this issue, an experiment was conducted to test the relative efficacy mixture of two pathotypes - Sg 150 from Jalna, and Sg 212 from Durgapura along with the individual pathotypes

for screening pearl millet lines. Twelve pearl millet lines (P 7-4, P 310-17, 700651, 7042R, 852B, 834B, 843B, IP 18292, IP 18293, 863B, ICMB 03999 and S 2003-188) known for their differential reactions to these pathotypes and two susceptible checks (7042S and ICMP 451) were used. Experiment was conducted in CRD with three treatments of pathogen and 14 genotypes in 6 replications (one pot/replication with 35–40 seedlings). The experiment was repeated to confirm the results. The results indicated that the pearl millet lines that were highly susceptible to either or both pathotypes were also highly susceptible to pathotype mixture, and the lines that were not highly susceptible to either of the pathotype had average susceptibility. Thus, the use of pathotype mixture would serve the purpose of discarding the highly susceptible lines. However, screening against specific pathotypes may provide more reliable data on resistance levels of the genotypes.

RP Thakur

Milestone 5A.5.1.2: Spatial and temporal virulence pattern of downy mildew pathogens assessed through virulence nursery and on-farm survey results (RPT/RS/KNR/RB/CTH, 2009)

Virulence monitoring in pearl millet downy mildew: The downy mildew pathogen, *Sclerospora graminicola*, evolves fast in response to selection pressure imposed by host resistance gene(s). Occurrence of new virulence and virulence shift is monitored through on-farm surveys and multilocation virulence nursery.

On-farm downy mildew survey: Under the ICAR-ICRISAT partnership project, roving surveys were conducted in Gujarat, Maharashtra and Rajasthan in collaboration with AICPMIP pathologists of the respective states during the summer and rainy seasons. In Gujarat, during summer, a total of 70 pearl millet fields were surveyed in six talukas of two districts (Mehasana and Banaskantha) where summer pearl millet is important. Of the 70 fields surveyed, 41 (58%) had DM incidence ranging from 1 to 93%. Of the three public sector hybrids observed, HHB 67 had 11% incidence and the other two hybrids, GHB 557 and GHB 558 were DM-free. Of the 23 private sector hybrids, 11 (Bioseed, HY 555, M-50, Nandi 5, Nandi 52, PAC 982, Pioneer 86M34, Pioneer 86M52, Proagro 9444, Raasi 2246 and Ritu) were disease-free, six (Chandini 511, Nandi 35, Nirmal 1651, NK 1616, Paras Sarpanch and Proagro 9555) were resistant (1–10% incidence) and the remaining six (Ajay VBBH 334, Nandi 32, Paras Ganesh, Pioneer 7777, Rani, and Seedtek) were susceptible (30–61% incidence).

In Maharashtra, we surveyed a total of 99 pearl millet fields in 18 talukas of 7 districts, (Ahmadnagar, Aurangabad, Beed, Dhule, Jalgaon, Jalna and Nashik) during the rainy season. There were 15 private sector hybrids (Amar Ratna 6, Anuja 6, Hy. 555, MBH 163, MDBH 318, MLBH 308, MLBH 367, MLBH 504, MRB 204, MRB 212, MRB 2210, Nirmal 1651, Pioneer 86M34, Proagro 9330 and Tulja) and all were disease free.

In Rajasthan, a total of 123 pearl millet fields in 20 talukas of 7 districts (Alwar, Dousa, Jaipur, Karouli, Sawai Madhopur, Sikar and Tonk) were surveyed during the rainy season. Of the 123 fields surveyed 93 (76%) showed DM incidence ranging from of 1–96%. Of the 24 private sector hybrids observed, 3 (JKBH 598, Kanchan and MRB 2210) were disease free, 4 (JKBH 26, Nirmal 1651, Proagro 9444 and Pioneer 86M52) were resistant (1–5% incidence) and the remaining 17 were susceptible (Akash, Ankur 2226, Anmol, Arjun, Aswani, Bioseed 8510, Sri Hari Krishna 1044, Kaveri 456, MLBH 75, MLBH 308, PAC 982, PG 5850, Pioneer 7688, Pioneer 86M32, Pioneer 86M34, Rani and RBH 36) were susceptible (37–96% incidence). One public sector hybrid, ICMH 356 found in just 2 fields was also disease free. Most of the seed companies supplied the metalaxyl-treated seeds to the farmers.

We believe that the absence of DM in several hybrids and high levels of resistance in some other hybrids may require confirmation of their genetic resistance as most of the seed were treated with metalaxyl. In addition, lack of disease in Maharashtra could be due to the initial dry spell of about 20 days after planting and the practice of different cropping sequences, such as soybean-cotton-millet; wheat-cotton-millet, sorghum-cotton-millet; maize-cotton-millet; cotton-castor-millet and onion-cotton-millet that affect initial soil inoculum level. This needs further investigation.

RP Thakur and Rajan Sharma

Output target 5A.6: At least two improved populations and experimental hybrids of pearl millet with high forage yield potential developed (2009)

Activity 5A.6.1: Develop and evaluate improved open-pollinated varieties and hybrids for their forage yield potential

Milestone 5A.6.1.1: Additional germplasm sources with high biomass yield identified (KNR/RB/HDU/MB, 2009)

Germplasm sources: Over the last few years, there has been an increased emphasis on developing hybrid parents or varieties with high forage yield potential. This has necessitated exploiting germplasm accessions of diverse origin with high biomass yield. Ten germplasm accessions visually selected for high forage yield during the 2005 rainy season were planted for producing S_1 progenies. In addition, three of the most promising accessions were random mated to develop an open-pollinated forage variety ICMV 06111. An experimental forage hybrid (ICMA 00999 × IP 17315) earlier identified as the most promising, and consistently giving 15–30% more dry forage yield over the released forage hybrid Proagro 1, is now routinely used as a control in forage trials. In a preliminary yield trial conducted during the 2006 rainy season, ICMV 06111 gave 9.1 t ha^{-1} of dry forage yield at 80-d harvest, which was 90% of the forage yield of ICMA 00999 × IP 17315 and comparable to that of Proagro 1 (8.9 t ha^{-1}). ICMV 06111 flowered in 87 days compared to 69 days for ICMA 00999 × IP 17315 and 54 days for Proagro 1.

KN Rai and HD Upadhyaya

Milestone 5A.6.1.2: Improved populations and experimental hybrids with high forage yield potential developed (KNR/RB/ HDU/MB, 2009)

Open-pollinated varieties hybrids with high forage yield: Nine OPVs developed for forage purposes have now been evaluated for two years (rainy season of 2005 and 2006) at Patancheru. Based on the mean performance across the two years, ICMV 05555 was found to be the highest-yielding, giving 14.8 t ha^{-1} of dry forage yield at 80-d harvest, which was 14% more than that of the control (ICMA 00999 × IP 17315). Another variety ICMV 05777 gave 13.1 t ha^{-1} of the dry forage yield, which was comparable to that of the control (13.0 t ha^{-1}). ICMV 05555 flowered in 70 days and ICMV 05777 in 74 days compared to 69 days for the control. There were five additional varieties that had 10.3–11.9 t ha^{-1} of the dry forage yield. Two of these flowered in 60 and 65 days, one in 78 days, while the other two flowered in 94 days. The yield potential of the latter two is likely to be higher if harvested at 90-95d harvest.

Twenty-seven topcross hybrids produced by crossing three forage type male-sterile lines (ICMA 89111, ICMA 00999 and ICMA 03222) with each of the nine forage populations were evaluated during the 2005 and 2006 rainy seasons for forage yield at 50 and 80-day harvest. Based on the mean performance over the two seasons, seven hybrids (3 on ICMA 89111 and 4 on ICMA 00999) produced 12.5-13.8 t ha^{-1} of dry forage yield (11.8 t ha^{-1} for control hybrid, ICMA 00999 × IP 17315) at the 80-d harvest. The highest yield was obtained for hybrid ICMA 00999 × ICMV 05222 (13.8 t ha^{-1}). All the selected hybrids flowered in 65–75 days (72 days for the check hybrid).

KN Rai, HD Upadhyaya and Michael Blümmel

Milestone 5A.6.1.3: Diverse seed parents with high forage yield potential developed and characterized (KNR/RB/MB, 2012)

Hybrid parents progenies: We evaluated 24 advanced generation progenies (F_6/F_9), of which 18 were selected based on the visual assessment of forage yield. Of these, 17 flowered in 51–60 days (checks ICMB 00999 flowered in 47 days and ICMB 98777 in 58 days). Sixteen potential forage type seed-parental lines with 50–60 days of flowering were intercrossed during the postrainy season. The resulting 16 F_1's were evaluated during the 2006 rainy season for biomass, of which 10 were selected based on the visual assessment of forage yield, with their flowering ranging between 50 and 55 days. We also evaluated 155 S_2/S_6 population progenies (earlier selected for high biomass yield) during the 2006 rainy season, of which 60 were visually selected for high biomass yield, producing 120 progenies. Most of the progenies flowered in 50–60 days (check ICMP 451 flowered in 50 days).

KN Rai and Michael Blümmel

Output target 5A.7: Information on breeding efficiency and genetics of three diverse cytoplasmic-nuclear male-sterility (CMS) systems in pearl millet documented (2009)

Activity 5A.7.1: Documentation of research results related to CMS genetics and breeding efficiency in pearl millet

Milestone 5A.7.1.1: Comparative studies on efficiency of three diverse CMS systems completed (KNR, 2008)

The field trial data from two years and 2–3 locations have been analyzed, showing that the hybrids based on the A_4 cytoplasm give about 5% less grain yield than those based on the A_1 cytoplasm, although there is large cytoplasm × genotype interaction. These results will be written up for formal publication in 2007/2008**.**

KN Rai

*Milestone 5A.7.1.2: Genetical studies of diverse CMS systems completed (KNR/RB, 2009***)**

This research was a part of a PhD thesis that was completed in 2005. Drafts of the genetics of two CMS systems have been prepared. The publication of this work will start in 2007 and will be completed by 2009.

KN Rai and Ranjana Bhattacharjee

Output target 5A.8: Pearl millet technology exchange, capacity building and impact assessment undertaken and documented (2009).

Activity 5A.8.1: Enhance technology exchange and partnership building, and assess its impact

Milestone 5A.8.1.1: Seed of hybrid parents and breeding lines multiplied and distributed (KNR/RB, annual)

In response to seed requests, 1200 seed samples were supplied to public and private organizations (263 public and 937 private) within India, and about 730 samples of breeding materials supplied to about 10 countries abroad (Africa, USA and UAE). We produced 400 kg breeder seed of ICTP 8203 and 228 kg was supplied from the carry-over stock. Also, we supplied 320 kg breeder seed of seven hybrid parental lines. In addition, about 300 kg breeder seed of two seed parental lines (ICMA/B 89111 and ICMA/B 93333) was multiplied.

KN Rai

Milestone 5A.8.1.2: ICRISAT's research partnerships with NARS, networks and regional fora strengthened ((KNR/CLLG/Pearl Millet Team, annual)

Under the ICAR-ICRISAT partnership project, 8 trials (6 hybrid trials, and one each of biofortification and salinity trials) and 11 nurseries (7 maintainer and 4 restorer lines) were constituted and sent to the AICPMIP coordinator for coordinating the evaluation at 14 locations. NARS and the private sector scientists were involved in planning for an international course on Pearl Millet Improvement and Seed Production and also serving as resource persons. NARS scientists were increasingly involved in project development, joint publications, and facilitation of a research grant from ICAR for DM resistance marker selection work.

During a brief visit to two research centers in Mexico, presentations were made on adaptation and yield potential of pearl millet for grain and fodder production to assess the prospects of pearl millet and develop possible research collaboration for introducing this crop for crop diversification in that country. Similar presentations were made and pearl millet and sorghum trials evaluated during a 2-week visit to several research stations and universities in Central Asia (Uzbekistan, Turkmenistan and Kazakhstan) to assess the prospects of pearl millet in this region. There is keen interest among NARS to introduce pearl millet for crop diversification and farmers livelihood improvement in Central Asia and Mexico. Also, linkages were developed for testing the prospects of pearl millet in Morocco and China.

ICRISAT-Private Sector partnership was further strengthened with 20 new seed companies joining the Pearl Millet Hybrid Parents Consortium in 2006, taking it to a total of 34 consortium members. Maharashtra State Seed Corporation, the first public-sector seed producing agency coordinated a 21-entry trial of consortium hybrids from 10 seed companies at 9 locations, including Patancheru. Analysis of Patancheru data showed that none of the hybrids produced higher grain yield over the high-yielding check 7688 (4330 kg ha^{-1}). However, for dry fodder

yield, the hybrids of GK 1059 and KH 325 were 35 and 9% superior over the check 7688 (4480 kg ha^{-1}), and the flowering of all the entries ranged between 46 and 52 days, while 7688 flowered in 48 days and HHB 67-2 in 38 days

KN Rai, CLL Gowda and Pearl Millet Team

Milestone 5A.8.1.3: International Pearl Millet Training Course (2009) and Field Day (2006, 2008, 2010, 2012) conducted (KNR/CLL/Pearl Millet Team)

Pearl millet scientists field day: Pearl Millet Scientists Field Day held on 14–15 September 2006 at ICRISAT, Patancheru had 60 scientists from 12 public and 32 private organizations. The participants selected breeding lines and potential parents (A-, B- and R-lines) of hybrids for utilization in their programs. All the private sector requests have been obtained and consolidated which shows that 1382 breeding lines/parental lines of potential hybrids were selected by these companies with a total of 5420 seed samples as some of the lines were selected by more than one seed company (more than 7 companies requested a few lines).

International training course on pearl millet improvement and seed production: An international training course on pearl millet improvement and seed production was held on 2–15 May 2006 at ICRISAT, Patancheru. The course was initially co-sponsored by the FAO. Subsequently, INTSORMIL, USA; ICBA, Dubai, The Sehgal Foundation, India; and Syngenta Foundation, Switzerland also co-sponsored this course. Besides ICRISAT scientists (11), resource scientists from national (3) and private sector (8) in India, along with one scientist from USA, were included as resource persons for the training. About 40 trainees (including 5 women) from 15 countries participated in the training, of which 19 participants were from private seed companies and 9 from public sector institutions within India; 7 from Africa; 4 from Middle-East; and one each from USA, Uzbekistan, Pakistan and Myanmar. The training course consisted of several lectures, practical classes, visits to field and private seed companies and group discussions. The success of the training course was evaluated by providing questionnaires to the participants. In a group exercise, they also identified 11 major constraints in pearl millet production. Six participants also took the opportunity to select more than 500 breeding lines (including hybrid parental lines, improved populations and germplasm accessions) and also requested for ICRISAT's assistance in supplying breeding materials, organizing drought and salinity nurseries, and guiding their breeding programs. The general opinion was to conduct this training course once in 3 years. Participants also suggested for conducting similar training courses in biotechnology and data analyses.

KN Rai, CLL Gowda and Pearl Millet Team

Milestone 5A.8.1.4: Technical information and public awareness documents developed and disseminated (KNR/CLLG/Pearl Millet Team, annual)

Two flyers were prepared in collaboration with the International Center for Biosaline Agriculture (ICBA). One of this deals with salinity tolerance of sorghum and pearl millet and the other one deals with the prospects of these crops in Central Asia. A booklet describing pearl millet cultivars for dry environments was brought out as a joint publication of ICRISAT and ICAR. Pearl millet as a highly nutritious cereal for grain iron and zinc was publicized through a poster at the International Plant Breeding Symposium in Mexico.

Milestone 5A.8.1.5: Commercialization of pearl millet grains strengthened through researcher-farmer-industry alliances (KNR/CLLG/Pearl Millet Team, annual)

Plans were developed under the CFC-funded project for on-farm trials of pearl millet in Andhra Pradesh (AP) and Maharashtra village clusters to enhance pearl millet productivity through cultivar replacement and micronutrient applications. Based on the results during 2005, the popular pearl millet variety ICTP 8203 in the AP village clusters was almost completely replaced with a pearl millet hybrid (MLBH 308) that showed substantial yield advantage. The data on the grain and fodder yield of new cultivars over those grown by the farmers (both with and without micronutrient applications) were collected to examine the income generation values of the recommended practices.

III. Pigeonpea

Output Target 5A.1: About 15 high-yielding pigeonpea hybrids made available for cultivation in different environments (2006-2009).
Activity 5A.1.1: Development of widely adapted high-yielding hybrids for different environments.

Milestone 5A.1.1.1: At least 100 new hybrid combinations evaluated to identify new fertility restorers/male sterility maintainers (KBS/KML, 2006-09)

During 2006 rainy season, 227 short-duration and 234 medium-duration new hybrids were evaluated for fertility restoration. Of these, 298 hybrids (64.6%) had full pollen fertility and 33 (7.2%) were male-sterile, while the remaining segregated for male-fertility and sterility in various proportions. All the male-sterile F_1s were backcrossed to their respective male-parents to produce BC_1F_1 seed for the diversification of A-lines. Among the

fertile hybrids, the promising combinations will be selected to produce hybrid seed again for confirming their fertility restoration and agronomic performance.

KB Saxena and K Madhavi Latha

Milestone 5A.1.1.2: At least five high yielding hybrids each in early and medium maturity duration identified for multi-location testing (KBS/KML, 2007)

A total of 405 new experimental hybrids were evaluated for their agronomic performance in 20 station trials at Patancheru. Due to frequent rains during early growth stages, the short-duration experiments were damaged by intermittent water-logging and phytophthora blight and eight trials were abandoned. In the remaining four trials, the yield levels were low when compared with the previous season. Among these hybrids, ICPH 3538 (2014 kg ha^{-1}) was found to be the best with 92% superiority over the control ICPL 88034 (1027 kg ha^{-1}). The other promising hybrid in this group was ICPH 3548 (1832 kg ha^{-1}, 78% superiority). The data from medium-duration hybrid trials are awaited.

KB Saxena and K Madhavi Latha

Milestone 5A.1.1.3: At least five pigeonpea hybrids identified for on-farm testing (KBS/KML, 2008)

Based on the performance of hybrids in multilocation trials conducted in 2005 rainy season and the availability of seed, one short-duration hybrid (ICPH 2438) and one medium-duration hybrid (ICPH 2671) were evaluated at 20–25 farmers' fields in Maharashtra, Karnataka and Andhra Pradesh by cooperating NARS partners and seed companies. The data are awaited.

In order to identify new hybrids for on-farm testing in 2007 rainy season, 24 short-duration hybrids are being evaluated at 10 locations. Similarly, in the medium-duration group 32 hybrids are being evaluated at 15 locations. The parental seeds of these hybrids are being multiplied either in isolation or under insect-proof cages.

KB Saxena and K Madhavi Latha

Milestones 5A.1.1.4: Elite pigeonpea hybrids evaluated for their resistance to major insects and diseases (2009)

Four hundred and eighty-one hybrids and their parents were evaluated for wilt and SM resistance under artificial epiphytotic conditions following standard field evaluation techniques. Wilt-susceptible ICP 2376 and SM-susceptible 8863 were sown after every 10 test entries as infector-cum-indicator rows. Of these, 11 hybrids, ICPHs 2324, 2373, 3183 (ICPL 20106), 3224 (ICPL 87051), 3363, 3450, 3476, 3477, ICPR 2337 (ICPL 20106), ICPLs 87119, ICPL 20110, were found asymptomatic, while 21 were found resistant with <10% disease incidence of both wilt and SM.

Suresh Pande and KB Saxena

Output Target 5A.2: Genetically diverse pigeonpea hybrid parents (about 5-10 A lines and 10-15 R lines) with resistance to major biotic stresses developed (2009)

Activity 5A.2.1: Development of high-yielding pigeonpea hybrid parents with resistance to major biotic stresses

Milestone 5A.2.1.1: At least six A_4 male-sterile and 15 fertility restorer lines with resistance to wilt and sterility mosaic disease developed (2007)

Three hundred and eleven A_4 male-sterile lines and their restorers were evaluated under artificial epiphytotic conditions following standard field evaluation techniques for combined resistance to wilt and SM. Four lines, ICPB 2092 (ICPL 96058), ICPR 2926 (ICPL 20129), ICPR 2352 (ICPL 94068) and ICPL 99044 were found asymptomatic to both wilt and SM diseases, while 29 lines had combined resistance (<10% wilt and SM diseases). Two lines, ICPA 2048 and ICPA 2087 were asymptomatic for wilt and 63 lines for SM.

Suresh Pande and KB Saxena

Milestone 5A.2.1.2: At least six promising maintainers of A_4 cytoplasm improved for agronomic traits (seed and pod size and disease resistance) through backcrossing (KBS/KML, 2009)

In any dynamic hybrid breeding program the genetic enhancement of female parents for desired agronomic traits is essential to develop promising hybrid combinations. To increase the pod size of promising A-lines, ICPA 2039 and ICPA 2089 were crossed with a white seeded variety 'Kanchan'. Similarly, crosses are being initiated to incorporate sterility mosaic and wilt resistance in ICPA 2044 and ICPA 2045.

KB Saxena and K Madhavi Latha

Output target 5A.3: Pigeonpea hybrid parents (25–30 A-lines and 50–55 R-lines) characterized for important agronomic traits and molecular diversity (2009)

Activity 5A.3.1:Assessing the agronomic and molecular diversity of pigeonpea hybrid parental lines

Milestone 5A.3.1.1: A/B- and R- lines characterized for important agronomic traits (KBS/KML/HCS/SP, 2008)

During the 2006 rainy season, 26 maintainer lines and 22 restorer lines were evaluated in separate replicated trials at Patancheru. The data are being compiled on a number of qualitative and quantitative traits. We propose to repeat this trial in 2007 rainy season also. This information will be shared with the germplasm unit and also will be used for registration and selection of hybrid parents in the future.

KB Saxena, K Madhavi Latha, HC Sharma and S Pande

Relative susceptibility of maintainer and restorer lines to *Helicoverpa armiger:* Fifteen restorers and 13 maintainer lines along with resistant (ICPL 187-1, ICPL 332) and susceptible (ICPL 87 and ICPL 87119) checks were evaluated for resistance to the pod borer, *H. armigera*. There were three replications in a randomized complete block design for each set. Data were recorded on pod damage and overall resistance scores at maturity. The overall resistance scores ranged from 4.5 to 9.0. The maintainer lines ICPB 2032, ICPB 2049, ICPB 2050, and ICPB 2051 showed a damage rating of 5.5 to 6.0 compared to 8.2 in ICPL 87 and 4.5 in ICPL 332. Percentage pod damage ranged from 75.7 to 86.4%. Most of the lines (except ICPB 2032, ICPB 2043, ICPB 2046, ICPB 2046, and ICPB 2048) were highly susceptible to wilt (over 80% mortality).

Among the restorer lines, the genotypes ICPR 2913, ICPR 2920, ICPR 2922 showed a damage rating of 4.7 to 6.0 compared to 4.5 in ICPL 332 and 8.2 in ICPL 87. Pod damage was 56.6 to 70.0% in ICPR 2336 and ICPR 2904 compared to 64.8% in ICPL 332 and 86.0% in ICPL 87. The lines ICPR 2904, ICPR 2905, and ICPL 187-1 showed high susceptibility to wilt (>80% plant mortality) compared to 7.8% in improved version of ICPL 332 and 8.5% in ICPL 87.

HC Sharma and KB Saxena

Milestone 5A.3.1.2: Available male sterile (A/B) and fertility restorer (R) lines characterized using molecular markers (RKV/KML/KBS/DAH, 2009)

Output Target 5A.4: **Seed production technology for pigeonpea hybrids and their parents improved (2009)**

Activity 5A.4.1: Developing an efficient seed production technology for pigeonpea hybrids and their parents.

Milestone 5A.4.1.1: Improved seed production technology for pigeonpea hybrids and their parents developed (KBS/KML, 2009)

Hybrid pigeonpea technology is new and the efficiency of commercial hybrid production depends on the improved seed production technology. To reduce the cost of seed production of A-lines, an experiment was carried out by sowing ICPA 2048 at three spacings (75 × 30, 75 × 100, 75 × 150 cm). The results are awaited.

KB Saxena and K Madhavi Latha

Milestone 5A.4.1.2: A hybrid seed production manual published (KBS, 2006)

To guide scientists and seed producers involved in hybrid pigeonpea research and development a manual entitled "Hybrid Pigeonpea Seed Production Manual" was published in 2006. This manual contains the detailed information on various aspects of seed production technology of pigeonpea hybrids and their parents.

KB Saxena

Output Target 5A.5: Efficiency of hybrid pigeonpea breeding improved through strategic research (2010)

Activity 5A.5.1: Conduct strategic research to improve the efficiency of hybrid breeding

Milestone 5A.5.1.1: Cytology and genetics of A_4 CMS system and its fertility restorers investigated (VD/KBS, 2007)

Transverse sections of young anthers from male-sterile and fertile plants showed no differences in the development of sporogenous tissues. The process of microsporogenesis was similar in both up to differentiation of PMCs into tetrad formation and differed thereafter. The PMCs of the male-sterile plants became shriveled due to breakdown of the tapetum layer, which not only gives support but also provides nutrition to PMCs. In the case of fertile plants, the process of microsporogenesis proceeded normally and all the layers of anther wall had developed by the time PMCs had formed. Unlike the male-sterile plants, the tapetum layer in the fertile plants was also found to be persistent till the development of pollen grains and subsequently ruptured to release the pollen grains.

Vijay Dalvi and KB Saxena

Milestone 5A.5.1.2: Genotype - environment interaction for the expression of male-sterility and fertility restoration assessed (KBS/VD/KML, 2009)

Stability of CMS lines: For commercial production of high-yielding hybrids, it is essential to identify stable CMS lines. Three CMS lines with diverse cytoplasm were selected for this study. These include ICPA 2067 (A_1 cytoplasm), ICPA 2052 (A_2 cytoplasm), and ICPA 2039 (A_4 cytoplasm). These lines were evaluated at Parbhani and Patancheru during 2004 and 2005 rainy seasons. The observations on pollen staining showed that ICPA 2039 was the most stable CMS line at both locations. In case of ICPA 2052, five out of 49 plants showed fertile pollen grains (5–30%) and the remaining plants were male-sterile at Patancheru. More or less similar results were observed at Parbhani. ICPA 2067 was found to be the most sensitive to the environmental changes, as 24 out of 28 male-sterile plants, reverted to male-fertility in winter months at Patancheru. At Parbhani also, 24 out of 36 male-sterile plants reverted to male-fertility.

Stability of fertility restoration: In general, the testers performed differently with regards to the fertility restoration of the three CMS lines. At Latur (18°N), three testers (ICPL 129-3, BWR 23, and Nirmal 2) restored the male fertility of CMS line ICPA 2067, while BSMR 175 showed only 35% fertility restoration. The testers BDN 2, BSMR 736, and BSMR 853 showed >70% fertility restoration with ICPA 2067. These three testers can be purified to get 100% fertility restoration with ICPA 2067. Out of seven F_1 hybrids developed on ICPA 2052, three testers (ICPL 129-3, Nirmal 2, and BSMR 175) maintained complete male-sterility and four testers (BDN 2, BWR 23, BSMR 736, and BSMR 853) showed partial (60–80%) fertility restoration. The F_1 hybrids developed on ICPA 2039 showed that only ICPL 129-3 maintained complete male-sterility, while BSMR 736 restored complete fertility. The other five testers segregated for fertility restoration (66–85%). These testers can be purified to get 100% fertility restoration for development of hybrids. The results of the present study revealed that although there were

differences among testers for fertility restoration of different cytoplasm, the genotype × environment interactions were not strong enough to influence selection and breeding of hybrids.

KB Saxena, Vijay Dalvi and K Madhavi Latha

Milestone 5A.5.1.3: New sources of cytoplasm identified and diverse CMS systems developed (KBS/NM/KML, 2010)

Crosses between diverse pigeonpea cultivars and wild species *Cajanus acutifolius* gave rise to F_1 hybrids. It was consistently observed that hybrids between pigeonpea cultivar ICPL 85010 and *C. acutifolius* ICCW 15613 gave rise to hybrids with high degree of male sterility. These hybrids were crossed with diverse pigeonpea cultivars to identify maintainers and restorers of male sterility. Cultivar 85010 consistently gave rise to progeny with high level of sterility (60–100 %).

Crosses between *Cajanus platycarpus* and *C. cajan* have given rise to progeny with variation for many desirable characters. One such character is a high degree of male sterility in the progeny when crossed with cultivar ICPL 85010. To test the proof of concept if the progeny belonged to the class of CMS, they were crossed with diverse parents including wild species *C. platycarpus*. The progeny from crosses with *C. platycarpus* gave rise to the progeny, all with 100% male sterility. This shows that *C. platycarpus* maintains male sterility in the progeny.

Nalini Mallikarjuna and KB Saxena

Output target 5A.6: Trait-based breeding populations developed for selecting elite hybrid pigeonpea parental lines (2011)

Activity 5A.6.1: Development of trait specific (diverse maturity, disease resistance, seed and pod size) breeding populations for selecting new maintainers and restorers

Milestone 5A.6.1.1: For each trait, about 10–12 genetically diverse lines will be identified and crossed in a half-diallel mating scheme to generate B and R breeding populations for selection (KML/KBS/SP, 2011)

The main objective of breeding parental lines is to broaden genetic base of the restorers and maintainers. Such genetic diversification of hybrid parents will help in increasing the magnitude of heterosis. During 2006, a program on the diversification of parental line was initiated by making crosses among B-lines in a diallel mating scheme. The similar approach is being followed to improve restorer lines.

K Madhavi Latha, KB Saxena and Suresh Pande

Output Target 5A.7: Hybrid pigeonpea technology exchange, capacity building of partners and documentation (2010)

Activity 5A.7.1: Exchange improved technologies and new knowledge with ARIs, NARS, NGOs, private sector, and farmers' groups

Milestone 5A.7.1.1: ICRISAT partnerships with NARS and Hybrid Parents Research Consortium Partners strengthened (KBS/CLLG/SP, annual)

Concerted efforts were made to enhance the partnership base for hybrid pigeonpea research and development. During 2006, the number of members in pigeonpea hybrid parents' research consortium increased from 9 to 14. In addition, links were strengthened with NARS partners in India and China.

KB Saxena, CLL Gowda and S Pande

Milestone 5A.7.1.2: Seeds of elite parental lines, and hybrids multiplied and distributed to NARS and seed companies (KBS, annual)

In 2006, seed of six A-lines and three hybrids were multiplied at Patancheru. Among the A-lines, ICPA 2043 (0.91 ha), ICPA 2047 (0.50 ha), ICPA 2048 (0.030 ha) and ICPA 2092 (0.16 ha) were multiplied in isolation using 1 male: 4 female ratio. Similarly, hybrids ICPH 2438 (0.30 ha), ICPH 2671 (0.30 ha) and ICPH 2741 (0.32 ha) were multiplied in isolation using 1 male: 4 female rows.

During 2006, a total of 2114 hybrid pigeonpea seed samples were supplied to private seed companies, public seed sector and NARS in India and other countries. This includes 1069 samples of hybrids, 164 A/B-lines, and 881 fertility restorer lines.

KB Saxena

Milestone 5A.7.1.3: Technical information and public awareness literature developed and disseminated (KBS/HCS/SP, 2007)

A public awareness flyer 'Hybrid Pigeonpea–Seeds of Excellence' was prepared and widely distributed among researchers and seed producers. This flyer contains information on hybrid technology, characteristics of A/B and R-lines, performance of some heterotic hybrids, and training opportunities.

KB Saxena, HC Sharma and S Pande

Milestone 5A.7.1.4: Capacity of NARS and seed sector scientists/technicians in hybrid breeding strengthened (KBS/KML, annual)

A total of 35 persons from NARS and the private seed sector were trained at Patancheru in hybrid pigeonpea breeding and production technology.

KB Saxena and K Madhavi Latha

Milestone 5A.7.1.5: Molecular markers and genetic maps developed and exchanged with the scientific community (RV/DAH/ KML/HDU/NM/KBS, 2010)

Efforts have been initiated for molecular characterization of 25 male-sterile and 30 restorer lines through micro satellite and DarT markers. The leaf samples of these lines have been collected and DNA extracted. The molecular characterization work is in progress.

Rajeev Varshney, DA Hoisington, K Madhavi Latha, HD Upadhyaya, N Mallikarjuna and KB Saxena

Output 5B: Enhanced molecular genetic and phenotyping platforms for drought and salinity screening and parental lines of hybrid sorghum, pearl millet and pigeonpea with improved tolerance to abiotic stresses, made available to partners with associated knowledge and capacity building in SAT Asia

Sorghum and Pearl Millet

MTP Output Targets 2006:
Three sorghum varieties with greater tolerance to salinity identified and made available to partners

Six pearl millet hybrid parental lines and populations with superior tolerance to salinity identified and made available to partners

New pearl millet hybrid parents selected for salinity tolerance in pot culture available for field testing

New genetic variability in sorghum introgressed and new derivates less sensitive to terminal drought stress available to partners along with grain mold resistant hybrid parents

I. Sorghum

Output target 5B.1: At least five salinity-tolerant sorghum breeding lines/populations and a mapping population developed (2009)

Activity 5B.1.1: Developing/identifying salinity-tolerant improved breeding lines/populations and associated QTL

Milestone 5B.1.1.1: Five salinity-tolerant breeding lines/populations developed/ identified (BVSR/VV, 2009)

Introgression of salinity-tolerance into high-yielding backgrounds: A total of 405 F_3s were produced from 139 F_2s derived from the crosses between salinity-tolerant breeding lines and high-yielding B-lines during the 2006 rainy season. Similarly, a total of 139 F_3s were produced from 54 F_2s derived from the crosses between salinity-tolerant breeding lines and high-yielding R-lines during the 2006 rainy season. These are being further advanced.

Comparative performance of hybrids for salinity tolerance: A trial of 30 hybrids along with three checks, viz, CSH 16, SP 40646 and ICSB 406 was conducted during 2006 rainy season at the Agricultural Research Station (ARS), Gangavathi to ascertain the salinity tolerance and grain yield in large grain backgrounds. Among them, 16 hybrids with a grain yield range of 4.8 to 6.0 t ha^{-1} were on par with the hybrid check CSH 16 (6.6 t ha^{-1}). For grain size, 25 hybrids with a range of 2.76 to 3.56 g 100^{-1} were comparable with the best check SP 40646 (3.22 g 100^{-1}) in the saline soil (10 dS m^{-1}) at ARS, Gangavathi.

BVS Reddy

Pot-culture trial of parental lines and hybrids: A total of 38 parental lines of sorghum were tested for salinity tolerance, along with 34 hybrids. There was a large variation among the lines for the grain yield ratio (grain yield salinity/grain yield control) ranging from 0.022 to 0.536, which is a proxy for salinity tolerance. Most of the accessions tested appeared to have low ratio, indicating that they were susceptible to salinity. Genotypes ICSB 405, ICSR 170, ICSR 93034, ICVS 93046, ICSV 745, GD 65115, SP 47513, SP 47519, SP 39105, SP 39053 had high ratios and were, therefore, tolerant to salinity. Regarding the stover yield ratio (stover yield salinity/stover yield control), most of the genotypes were in the range 0.40–0.80, showing much less contrast than for yield. There was no advantage of hybrids over parents in terms of stover yield ratio, which was slightly higher for inbreds than for hybrids (0.75). The grain yield ratio was also higher for inbred parents (0.20) than for hybrids (0.13). The same was true for the absolute values of yield under salinity conditions, with average across parents being higher (8.4 g $plant^{-1}$) than the hybrids (7.4 g $plant^{-1}$). Therefore, it was concluded that sorghum hybrids had no significant advantage compared to inbreds under saline conditions.

I. Salinity field trial of varieties: We tested promising salinity-tolerant sorghum breeding lines in coastal Orissa. There was a large range in adaptation of these genotypes to the area, and some varieties such as S 35, ICSV 112, ICSV 406, ICSR 170, ICSV 93034 were able to reach over 10 t/ha of green fodder yield in saline field over two cuttings. These data are very promising because it provides potential to have a crop grown after rice in a cropping system that usually leaves lands fallow after rice harvest, mostly because salinity levels rise during the postrainy season, which is too high for most crops. Repeat of this testing is planned for 2006–07.

Vincent Vadez, L Krishnamurthy and Belum VS Reddy

Milestone 5B.1.1.2: New F_6 RIL mapping populations for salinity tolerance available for phenotyping and genotyping (CTH/BVSR/VV, 2009)

From the screening in milestone of breeding lines and parental lines, several suitable parents have been selected and crosses will be made to develop new RIL for QTL mapping of salinity tolerance.

These crosses are:
BTx 623 (tolerant) × ICSR 93024-1 (sensitive)
ICSV 93046 (tolerant) × S 35 (sensitive)
BTx 623 (tolerant) × S 35 (sensitive)
SP 39105 (tolerant) × ICSR 93024-1 (sensitive).

Vincent Vadez and L Krishnamurthy

Milestone 5B.1.1.3: QTLs for salinity tolerance identified (VV/CTH, 2010)

Salinity screening of parents of existing mapping populations of sorghum: Ten parents (BTx 623, IS 18551, 296B, ICSV 745, PB 15881-3, PB 15520, B 35, E 36-1, N 13, IS 9830) were tested in 2005–06 for salinity tolerance to examine whether any of these could be used for QTL mapping of salinity tolerance. We found a good contrast for the grain yield ratio between E 36-1 (0.39) and N 13 (0.13), and also between PB 15220 (0.45) and ICSV 745 (0.10).

However, we did not detect any significant contrasts for the stover yield ratios among the available pairs of parental lines.

V Vadez

Output target 5B.2: Identification of sorghum genotypes with contrasting root traits (2009)

Activity 5B.2.1: Stay-green QTL introgression lines tested for root traits (VV/CTH, 2009)

Milestone 5B.2.1: Putative relation between stay-green QTL introgression lines and root traits identified (VV/CTH, 2009)

Root traits of stay-green lines: A repeat trial with long (2.40 m) PVC pipes was done to evaluate root growth at different stages and under different timing of imposed stress, using 2 stay-green genotypes (B 35, E 36-1) and 2 senescent genotypes (R 16 and ISIAP Dorado). The purpose of the experiment was to determine the most suitable timing of stress imposition to identify differences, if any, in the rooting pattern. Stress was imposed at 21, 35, 49 and 63 days after sowing. For each date, fifteen plants per genotype were grown under well-watered conditions until the seedset date. At each date, 5 plants were used to assess their root characteristics at that date. Then 5 plants were exposed to water stress and the remaining 5 plants were kept under well-watered conditions. Water-stressed and well-watered plants were used at 42, 40, 37, and 26 days after stress imposition, respectively, for sets of plants treated at 21, 35, 49, and 63 days after sowing.

In the sets of plants that were stressed at 21 and 35 DAS, we did not find any root depth differences between stay-green and senescent genotypes at harvest (respectively at 63 and 75 DAS). By contrast, we found that when stressed at 49 DAS, both stay-green genotypes had deeper rooting systems than senescent R 16 and ISIAP Dorado (15–35 cm deeper). Also, the two stay-green genotypes had a larger proportion of roots in the deeper layers of soil than R 16. Overall, this experiment confirmed the earlier finding of a deeper rooting and more profuse rooting in deeper layers of stay-green genotypes compared to senescent materials, under conditions of water deficit. What appeared also from this experiment is that compared to control, there was little difference in the root distribution in the layers below 90 cm in water-stressed and well-watered R 16, whereas B 35 was able to allocate a much larger proportion of roots in deeper layers under water stress than under well-watered conditions. In this experiment, the shoot dry weight under water stress, expressed as a percentage of control well-watered plants, was also higher in B 35 and E 36-1 than in R 16.

Root traits of stay-green introgressed lines: RSG 04001 and RSG 04005 are BC_1F_3 and BC_2F_3 derivatives from B 35 in the background of recurrent parent R 16, whereas IDSG 04211 is a BC_2F_3 derivative from B 35 in the background of recurrent parent ISIAP Dorado. Fifteen plants of these genotypes were grown in PVC pipes as above and stress was applied from 21 days onwards. The plants were harvested at 60 DAS. In this experiment, similar to the one reported above, there was no difference in root depth between B 35 and R 16. By contrast, two of the promising derivatives (RSG 04001 and RSG 04005) had deeper rooting and more profuse rooting in the 120–150 cm layer than R16. In this experiment also, it was evident that, compared to well-watered plants, B 35, RSG 04001 and RSG 04005 were able to increase their deep rooting relatively more than R 16. Also, the shoot dry weight under water stress, expressed as a percentage of control well-watered plants, was higher in B 35 and RSG 04001 than in R 16.

Several conclusions and future orientations can be drawn from the different sets of experiments that have been carried out to investigate root traits in sorghum: (i) Stay-green donor parent B 35 and in some cases E 36-1, and derivatives from B 35 appear to have deeper rooting and more profuse rooting in deeper layers than R 16. However, these differences seem to be conditioned by the timing of stress imposition and duration of stress. Major differences between senescent and stay-green materials are found when stress is applied close to the reproductive period and/or when plants are exposed to water stress for a sufficient time; (ii) Though we found some differences, which tend to indicate a role of roots in the stay-green material, we have had difficulties to obtain significant results in all experiments, because of plant to plant variations and because of the fairly small differences in rooting traits; (iii) We have mostly assessed, so far, the parents of the stay-green trait, and have only recently started working with derivatives, which are showing promising results. There is a need to move forward and study the advanced backcross generations of these derivatives that have been produced from RSG 04001 and RSG 04005, the two promising introgression lines mentioned before; (iv) So far, we have assessed rooting characteristics. However, this

does not give any indication on whether roots do contribute to a better water uptake. We observed that B 35 tended to senesce and dry later than R 16 under water stress, which would indicate that water uptake occurs for a longer period of time. Therefore, we are currently designing a lysimetric system, by which we could assess plant water uptake from the time of stress imposition. We believe that putative differences in the rooting characteristics would get integrated over time in larger differences for water uptake (easier to measure). If successful, such assay could eventually replace the time-consuming process of root extraction and its characterization.

Vincent Vadez and CT Hash

II. Pearl millet

Output target 5B.1: At least five salinity-tolerant improved breeding lines/ populations of pearl millet identified and feasibility of breeding salinity tolerant hybrids assessed (VV/KNR/RB) (2009)

Activity 5B.1.1: Develop salinity-tolerant lines and populations in pearl millet and assess their hybrid potential under saline conditions

Milestone 5B.1.1.1: Inbred lines and populations identified as salinity-tolerant in preliminary evaluations re-evaluated for their salinity tolerance and yield potential (KNR/RB/VV, 2008)

Four yield trials consisting of 5–70 entries were evaluated at the Agricultural Research Station of the University of Agricultural Sciences at Gangavathi under saline field condition (10 dSm^{-1}). The data are awaited. A trial of 13 entries consisting of five parental lines (3 salinity tolerant and 2 susceptible) and 6 hybrids derived from them, along with two salinity-tolerant populations, was conducted under non-saline conditions at Patancheru to assess their yield potential. A hybrid (ICMA 01222 × ICMP 451) based on salinity-tolerant male and female lines had the highest grain yield (3400 kg ha^{-1}) and it flowered in 47 days, followed by another hybrid also based on salinity-tolerant parental lines (female parent ICMB 95333 and male parent ICMP 451) that had 3355 kg ha^{-1} of grain yield and flowered in 48 days. In comparison, the dual-purpose commercial hybrid ICMH 451 (based on salinity-tolerant male parent ICMP 451 and susceptible female parent 81B) had 3260 kg ha^{-1} of grain yield and flowered in 49 days. These two new hybrid, however had lower dry stover yield (3370 and 3320 kg ha^{-1}) compared to ICMH 451 (3850 kg ha^{-1}). Among the dwarf parental lines, ICMB 01222 had the highest grain yield (1970 kg ha^{-1}) and it flowered in 51 days compared to 1420 kg ha^{-1} of grain yield for 81B, which flowered in 54 days. However, 81B had 2490 kg ha^{-1} of dry stover yield compared to 1900 kg ha^{-1} for ICMB 01222.

KN Rai, Ranjana Bhattacherjee and Vincent Vadez

Pearl millet breeding line testing in farmer's field in Orissa: We have tested promising salinity-tolerant pearl millet parental breeding lines in coastal Orissa. There is a large range of adaptation of these genotypes to the area, and some accessions such as HHVBC Tall, Raj 171, IP 6105, IP 6106, IP 19586 were able to produce over 10 t ha^{-1} of dry fodder yield in saline field over two cuttings. These results are promising because they provide the potential to have pearl millet successfully grown in that transiently salinity-affected rice-based cropping system.

Vincent Vadez, L Krishnamurthy and KN Rai

Output target 5B.2: Putative QTLs for salinity tolerance of grain and stover yield identified in pearl millet (2009)

Activity 5B.2.1: Genotyping and phenotyping of mapping populations for salinity tolerance

Milestone 5B.2.1.1: Putative QTL for salinity tolerance based on 160 RILs from one mapping population identified (CTH/SS/VV, 2009)

Confirmation of the contrast for salinity tolerance between parents of pearl millet populations: Twenty parental lines of ten pearl millet mapping populations were tested for salinity tolerance, using the grain yield ratio as a selection criterion. This was a repeat experiment of the previous year, where 6 pairs of parents showed good contrast for the grain yield ratio. In the current trial, 4 pairs out of these 6 previously identified confirmed that trend.

Among these, pairs 1 (LGD 1-B-10 and ICMP 85410-P7) and 3 (863B-P2 and 841B-P3) appear more suitable for molecular marker identification, because of the large phenotypic differences and differences in marker polymorphism. However, based on the similarity in grain yield under control conditions (43.4 and 47.9 g plant^{-1} for 841B-P3 and 863B-P2, respectively), and the large differences in their grain yields under salinity, the pair 841B-P3 and 863B-P2 appeared to be the most suitable. We also found two other pairs that showed contrasts for grain yield ratio.

The phenotyping of F_6 inbred progenies of the mapping population developed between 841B (tolerant) and 863B (sensitive) has been initiated and the harvesting is underway.

Vincent Vadez and CT Hash

Milestone 5B.2.1.2: Putative QTLs for salinity tolerance based on 35 BC_6F_3 contiguous segment introgression lines identified (CTH/SS/VV, 2010)

Funding to complete development of the contiguous segment introgression line set, and initiate its assessment, was received from DBT in December 2006, so work towards this milestone on this will recommence in 2007.

Milestone 5B.2.1.3: New F_6 RIL mapping populations for salinity tolerance available in pearl millet for phenotyping and genotyping (CTH/BVSR/KNR/VV, 2009)

The existing (ICMB 841-P3 x 863B-P2)-derived pearl millet mapping population was advanced to F6 RILs. The new RILs are being skeleton mapped as part of an exercise to create a more informative pearl millet consensus linkage map. Seed of approximately 150 progenies of this RIL mapping population are now available for salinity tolerance screening while new pearl millet RIL mapping populations are being generated for this target trait. Crosses were made between several selected pairs of sorghum parental lines exhibiting substantial pair-wise dissimilarity (>70%) at 67 SSR loci distributed across all 10 sorghum linkage groups, as well as substantial phenotypic differences for salinity tolerance based on pot screens of 30 candidate parental lines. F1 hybrids from several such crosses were sown for advance to the F2 generation by selfing during the 2006/07 postrainy season.

Output target 5B.3: Breeding value of putative terminal drought tolerance QTLs in pearl millet documented (2009)

Activity 5B.3.1: Publication of earlier results on drought tolerance QTL and gene pyramiding

Milestone 5B.3.1.1: Publication of results from marker-assisted selection for the linkage group 2 drought tolerance QTL into the genetic background of two parental lines (CTH/FRB, 2008)

QTL for improved grain yield across variable grain-filling moisture environments: Previous molecular work on drought tolerance in pearl millet has focused on QTL for the ability to maintain yield under post-flowering drought stress, which can be easily transferred to otherwise well-adapted hybrid parents to improve the performance of their hybrids under this type of stress. Pearl millet breeding programs targeting adaptation to variable post-flowering moisture environments would benefit from QTLs that improve grain yield across the full range of post-flowering moisture conditions, rather than in just some specific drought-stressed environments. We reanalyzed an extensive (12 environment) phenotyping data set that included both stressed and non-stressed post-flowering environments, to identify QTLs for improved yield over the whole range of moisture environments. Genetic materials were testcrosses of 79 F_2-derived F_4 progenies from a mapping population based on a widely adapted maintainer line (ICMB 841) × a post-flowering drought tolerant maintainer (863B). Three QTLs (on LG 2, LG 3, LG 4) were identified as primary candidates for MAS for improved grain yield across variable post-flowering moisture environments. QTLs on LG 2 and LG 3 (the most promising) explained a useful proportion (13 to 25%) of phenotypic variance for grain yield across environments. They also co-mapped with QTLs for harvest index across environments, and with QTLs for both grain number and individual grain mass under severe terminal stress. Neither had a significant QTL × environment interaction, indicating their predicted effects should occur across a broad range of available moisture environments. Finally, both are linked to SSR markers so they are amenable to efficient MAS. The remaining QTL (LG 4) is of secondary interest as it has a less consistent performance across individual moisture environments and a less clear effect on secondary traits. Responses to MAS predicted for each of the

identified grain yield QTLs ranged from 70 to 100 kg ha^{-1} across environments and as much 160 kg ha^{-1} in individual moisture environments.

FR Bidinger, T Nepoleon and CT Hash

Improvement of the post-flowering drought tolerance of ICMB 841 by MABC: We completed the evaluation of 13 BC_5F_3 segmental introgression lines targeting the LG 2 QTL (for post-flowering drought tolerance) from 863B in the background of widely adapted seed parent ICMB 841. These segmentally near-isogenic lines were evaluated in testcross hybrid form (with both drought-tolerant and drought-susceptible testers) in multiple years in both the Patancheru dry season drought nursery (where the LG 2 QTL was identified) and in natural drought-prone locations. B-line tolerance/sensitivity to post-flowering terminal drought stress was assessed by comparing B-line general combining abilities (GCAs) for grain yield in the stress and non-stress environments. The data set included three non-stress and seven post-flowering stress environments in the drought nursery and four naturally-stressed environments in western Rajasthan. The original recurrent parent ICMB 841 had a significant ($P<0.01$) negative grain yield GCA in the drought nursery stress environments and a non-significant GCA for grain yield in the non-stressed drought nursery environments, confirming its sensitivity to post-flowering stress, but not necessarily a lack of adaptation to the dry season environment *per se*. The QTL donor parent 863B had highly significant ($P<0.001$) GCAs for grain yield in both the stressed and non-stressed drought nursery environments, indicating that it is both adapted to the dry season environment and is tolerant to post-flowering drought stress. The best of the QTL introgression lines (ICML 03056) had a significant positive GCA under stress in the drought nursery and five others had non-significant positive GCAs for grain yield, indicating their superiority over their recurrent parent under stress. The remaining lines were similar to ICMB 841 (ie, sensitive to the stress). In the non-stress drought nursery environment, these lines had grain yield GCA values similar to ICMB 841, indicating that the effect of the LG 2 QTL was specific to the stress environment. None of the introgression lines had grain yield GCA values as high as the QTL donor parent 863B in either of the drought nursery environments. This was expected, however, as the original mapping exercise identified additional QTLs for post-flowering drought tolerance (with the positive allele from 863B) that were not transferred to the ICMB 841 background in this set of introgression lines. The significant improvement in the grain yield GCA of a number of the ICMB 841 QTL introgression lines, in the same stress environment in which the QTL was originally identified, provides a solid proof-of-concept of the effectiveness of MABC transfer of the LG 2 drought tolerance QTL to a drought-sensitive genetic background.

Of equal importance is the effectiveness of the LG 2 drought-tolerance QTL across a range of post-flowering stress environments, rather than just in the environment in which the QTL was identified. The testcross trials in the Rajasthan environments generally did experience post-flowering stress (often severe and early onset), but they were more variable than the managed drought nursery post-flowering stress environments. In addition, the donor and recurrent parents responded differently to the Rajasthan environments than they did to the drought nursery stress environments, as the grain yield GCA of 863B was negative ($P<0.01$) and that of 841B was not significant. Part of this was due to the 863B testcrosses flowering later than those of 841B, which placed them at a significant yield disadvantage, as there were very strong negative correlations between time to flowering and grain yield in all of the Rajasthan test environments ($r = -0.73$ to -0.98, $P<0.01$). We adjusted testcross grain yield (by covariance) for time to flowering, in order to separate the effects of drought escape (early flowering) and drought tolerance/sensitivity, and recalculated grain yield GCA on the adjusted values. Despite this, the GCA for grain yield of the individual QTL introgression lines varied considerably among the individual Rajasthan environments, with the result that mean GCA values for grain yield across environments were not significantly different from zero for either ICMB 841 or any of the QTL introgression lines (although the mean grain yield GCAs of several lines were positive and numerically superior to that of ICMB 841). The drought-tolerance QTL on LG 2 was detected based on the measurement of grain yield under stress in the drought nursery. Grain yield is typically subject to significant GE interaction, so the discrepancy in the effect of the QTL in the drought nursery and the arid zone evaluation environments, while disappointing, is not an anomaly. Yield-based drought tolerance QTL will clearly need to be identified either in the target environment, or in an environment that is sufficiently similar to the target environment so that the GE interaction between the two is minimal.

P Sathish Kumar, FR Bidinger and CT Hash

Milestone 5B.3.1.2: DM resistance and terminal drought tolerance QTLs pyramided in the genetic background of elite pollinator H 77/833-2 and QTL introgression homozygote product lines available for testing (CTH/PSK/SS/VV/ RPT/RS/KNR, 2007)

To be reported in 2007

Output target 5B.4: Pearl millet germplasm with superior P-acquisition identified (2009)

Activity 5B 4.1: Development of an effective protocol and identifica-tion of germplasm with enhanced P-acquisition ability

Milestone 5B.4.1.1: An effective P-acquisition protocol applicable for large-scale screening developed (VV/HDU, 2007)

Large efforts have been dedicated in 2005 and 2006 to develop an efficient and reproducible protocol to test the response of pearl millet genotypes to low P availability under controlled conditions. This protocol has been developed based on the fact that pearl millet seeds are small and that plant establishment is a key factor to a successful performance under low P conditions. To get uniform sowing depth, a template making holes of a standard depth is used, in which seeds are placed. Several seeds were usually sown per hill, and then thinned to one seedling per hill. It was observed that thinning was affecting the development of the remaining seedling (probably by disturbing roots of the remaining seedling). Therefore, 9 seeds are now planted per pot, one each into an individual hole made with the template, and most uniform 3 seedlings are retained. The protocol is also based on observations that growing 3 plants per replicate pot helps in decreasing the replicate-to-replicate variation. We are also aware of a genotype by nitrogen interaction effect under low P soil, which has to be taken into account at the time of screening for low-P tolerance (choice has to be made and repeat experiment should use the same N source). Finally, we have found out that small differences in soil Olsen P value could bring about large differences in the response of pearl millet plants. Therefore, the same soil lot is used for each experiment, and this lot is first homogenized with a concrete mixer prior to preparing the soil:sand mix (1:1 v/v).

Vincent Vadez

Milestone 5B.4.1.2: Pearl millet germplasm with superior P-acquisition from low-P sources identified (VV, 2009)

We have initiated the assessment of two pearl millet open-pollinated varieties (OPVs) (ICMV IS 92222 and ICMV IS 89305) for their ability to acquire P from low-P soils. In a controlled pot experiment using the protocol described above, we have found a 2-fold variation for biomass under low-P in 184 S_1 progenies. The top and bottom 25 S_1s for shoot biomass under low-P conditions have been planted to generate full-sibs of the top and bottom ranked. These will be selfed for one generation and S_1s will be evaluated in 2007.

Vincent Vadez and KV Padmaja

Milestone 5B.4.1.3: QTL for P acquisition from low-P sources identified in pearl millet (VV/CTH, 2011)

The parents of existing mapping populations of pearl millet have been tested for their ability to acquire P from low-P sources. Consistent contrast has been found between 2–3 pairs of parents. We have found that the shoot biomass under low-P conditions was well correlated to the shoot biomass under controlled conditions, meaning that low-P tolerance may be better assessed by the ratio biomass under low-P / biomass under control. However, differences

found with in mapping population parents were relatively small. Larger number of inbred parents will be tested to identify the most contrasting pairs for developing new RIL populations.

Vincent Vadez

Output target 5B.5: At least five pearl millet breeding lines with tolerance to high air temperatures (>45°C) during reproductive stage developed (2009)

Activity 5B 5.1: Evaluate a diverse range of parental lines, advanced breeding lines and populations for high temperature tolerance during flowering and grain- filling period; and identify major QTL associated with this trait

Milestone 5B.5.1.1: Breeding lines with >70% seedset under field conditions at high temperatures identified/ developed and their tolerance under glasshouse conditions validated (KNR/RB/VV, 2009)

Heat-tolerant hybrid parents: Most of the hybrids bred for rainy-season adaptation have been found to become male-sterile, or set very poor seed during the summer season if flowering takes place at a time when the air temperatures could be as high as 46-48°C. A few of the hybrids, however, set excellent seeds, indicating genetic variability for thermo-tolerance during the reproductive stage. With the assistance of MAHYCO, 92 maintainer lines (B-lines) and 7 restorer lines (R-lines) were evaluated for seed set under open pollination in a summer planting at Jodhpur. Most of the lines either did not set, or had <1% seed set under open-pollination. However, 30–60% seed set was observed in at least one plant in six B-lines (ICMB 96222, ICMB 00333, ICMB 01555, ICMB 03555 ICMB 04333, and ICMB 05333) when the air temperature was 46–48^{0}C at the time of stigma emergence, indicating high levels of thermo-tolerance and variability for this trait within these lines. Ten plants were selfed within these lines during the 2006 rainy season to derive single plant progenies that will be evaluated during the 2007 summer season at Jodhpur to confirm the within-line variability for this trait and to make selection for higher levels of thermo-tolerance.

Milestone 5B.5.1.2: Relationship between hybrids and their parental lines for tolerance to high temperatures during reproductive stage quantified (KNR/RB/VV, 2010)

In the first evaluation attempt, six lines with 30–60% seedset at 46–48°C air temperatures have been identified, with indication of within-line variability for this trait. The purification of these lines for this trait was initiated, and plans were developed to screen more lines for thermo-tolerance. Lines with confirmed high levels of tolerance will have been identified by 2008, and the research on the relationship between parental lines and their hybrids will start in 2009.

KN Rai, Ranjana Bhattacharjee and V Vadez

Salinity trial: A trial was conducted with hybrids made utilizing salinity tolerant and susceptible male sterile lines and restorer lines, including their parents along with two salinity tolerant populations as checks (GB 8735 and HHVBC Tall) during rainy season at Patancheru location. All the hybrids flowered in 47–49 days (checks GB 8735 flowered in 45 days and HHVBC Tall in 51 days). None of the hybrids produced significantly superior grain and dry fodder yields over the checks. However, two hybrids, ICMB 95333 × ICMB 94111 and ICMA 01222 × ICMP 451 gave comparable grain yields of 3413 kg ha^{-1} and 3400 kg ha^{-1} respectively, as that of check HHVBC Tall (2958 kg ha^{-1}). Only one hybrid ICMH 451 gave comparable dry fodder yield of 3850 kg ha^{-1} as that of check GB 8735 (3270 kg ha^{-1}). This indicates that hybrids with either one both the parents, tolerant to salinity will give higher grain yields when compared to hybrids produced by involving susceptible parents.

Hybrid and parental lines of pearl millet (Exp1-2-3-4-5)

A set of tolerant and sensitive inbred parents, along with their respective hybrids were tested under saline controlled conditions. The large pot system allowed us to evaluate grain and stover yield at maturity. There was no clear relation between the performance of the hybrids and the performance of its parents. Overall, hybrids performed better under salinity, reaching an average grain yield of 19.8 g plant^{-1}, higher than an average 12.2 g for the inbreds/populations. In fact, we found a very close correlation ($r^2 = 0.69$) between the grain yield under control and the grain yield under salinity. This meant that a lager part of the grain yield under salinity was explained by yield potential. The yield data were then separated into yield as a component and a residual (the latter accounting for the salinity tolerance *per se* plus error. These residuals were well related to the grain yield ratio (grain yield under salinity/grain yield under control). So, both grain yield ratio and residual were used to assess salinity tolerance. The yield ratio was not significantly different for the hybrids and the inbreds/populations (0.38 and 0.36, respectively). There were no differences in residuals of hybrids and inbreds/populations either, indicating that hybrid performance under salinity was likely to be largely due to their yield potential, but partly due to salinity tolerance as well, the latter not being any greater in hybrids than in the parental lines.

Vincent Vadez, L Krishnamurthy and KN Rai

Milestone 5B.5.1.3: QTL for high temperature tolerance from two diverse mapping populations identified (KNR/RB/VV, 2012)

Based on the results of activities under above milestone 5B.5.1.2, parental lines of mapping populations will have been identified by 2009.

KN Rai, Ranjana Bhattacharjee and Vincent Vazed

Output 5C: Germplasm and improved breeding lines with high and stable grain Fe and Zn density in sorghum and pearl millet made available to specific partners biennially (from 2008) with associated knowledge and capacity building

MTP Output Target 2006: Existing hybrid parents in sorghum having grain density of Zn above 25 ppm and Fe above 30ppm available to partners

I. Sorghum

Output target 5C.1: Sorghum germplasm lines/breeding lines with stable and high grain Fe (40–50 ppm) and Zn (30–40 ppm) contents identified and their character association, and inheritance studied (2009)

Activity 5C.1.1: Screening of germplasm and breeding lines for grain Fe and Zn and evaluating for grain yield and agronomic traits

Milestone 5C.1.1.1: Five each of germplasm lines/breeding lines with stable and high grain Fe (40–50 ppm) and Zn (30–40 ppm) contents identified (BVSR/HDU, 2008)

Core collection evaluation for micronutrient density

A large number of germplasm accessions (2974) from the core collection of sorghum were evaluated for accessing genetic variability for grain Fe and Zn contents. As core germplasm captures most of the variability present in world collection (>37000) maintained at ICRISAT, the information on the genetic variability would enable identifying micronutrient-rich lines for use in crossing with agronomically elite lines to generate exploitable variability to develop micronutrient-dense cultivars and hybrid parents. It would also enable identifying contrasting parents for effecting crosses to identify transgressive segregants and to develop mapping populations for identifying molecular markers linked to loci controlling grain Fe and Zn contents.

The core collection along with four control lines known for their Fe and Zn contents were evaluated in an augmented design at ICRISAT, Patancheru in 2005 postrainy season. For the sake of convenience, the accessions were evaluated (in contiguous blocks) as three separate groups classified based on days to 50% flowering (early: ≤65 days; medium: 66 to 80 days; late: >80 days). The early group comprising 1095 accessions along with 4 checks (repeated 11 times); the medium group comprising 1128 accessions along with 4 checks (repeated 12 times); and the late group comprising 751 accessions along with 4 checks (repeated 8 times). Each accession was sown in one row of 2 m length. In each accession, the border plants were left for open-pollination and all others were selfed. The data were collected on days to 50% flowering, plant height, grain yield, 100-grain weight, grain color, plant agronomic aspect, panicle shape, panicle compactness, glume color, glume coverage, and presence/absence of seed sub-coat. Grain samples from some of the accessions could not be collected due to severe bird damage. In a few accessions, the available grains were not sufficient for estimation of Fe and Zn contents. The grain samples harvested from selfed panicles of 702 accessions of early maturity, 461 accessions of medium maturity, and 238 accessions of late maturity (making a total of 1401 accessions), and the grain samples harvested from open-pollinated panicles of 118 accessions of early maturity, 69 accessions of medium maturity, and 21 accessions of late maturity (making a total of 208 accessions) where sufficient quantities were available processed and sent to ICRISAT's analytical services laboratory for grain Fe and Zn contents estimation.

A large variability for grain Fe (7.7 ppm to 132.6 ppm) and Zn (15.1 ppm to 91.3 ppm) was observed among the 1401 accessions. The variability observed in core collection is much higher than that reported earlier, based on screening of 86 genotypes consisting of germplasm lines, hybrid parents of released/marketed hybrids and popular varieties. Interestingly, white grain accessions used for human consumption in India had on an average 43.6 ppm Fe and 35.1 ppm Zn which was marginally higher than those with colored grains (40.4 to 42.6 ppm Fe and 30.9 to 34.0

ppm Zn). The average Fe and Zn contents of accessions with testa (42.5 ppm Fe and 34.2 ppm Zn) and without testa (42.9 ppm Fe and 33.2 ppm Zn) were comparable. However, endosperm texture and grain size have significant effects on grain Fe and Zn contents. While the accessions with higher than 75% corneous endosperm had 56.2 ppm Fe and 44.3 ppm Zn contents, those with 0% to 75% corneous endosperm had less than 44.8 ppm Fe and 35.0 ppm Zn. The texture of endosperm has significance in food preparations. While grains with 50% corneous endosperm are useful for preparation of *'roti'* or *'chapati'* (unleavened bread), the most popular food forms of sorghum grains in India; and for 'ingera' (leavened bread), the most popular food forms of sorghum grains in some parts of Africa, those with more than 75% corneous endosperm are useful for the preparation of '*to*' the most popular food form of sorghum grains in some parts of Africa. Accessions with small grains (with <2.5 g 100-grain weight) had significantly higher grain Fe (44.4 ppm) and Zn (35.9 ppm) contents than those with medium (with <2.5 g to 3.5 g 100-grain weight) (42.6 ppm Fe and 33.5 ppm Zn) to large (with >3.5 g 100-grain weight) grains (40.8 ppm Fe and 32.2 ppm Zn).

BVS Reddy, HD Upadhyaya and KL Sahrawat

Milestone 5C.1.1.2: Correlations of grain Fe and Zn contents with grain yield and size and agronomic traits estimated (BVSR, 2008)

Correlations of grain Fe and Zn contents with agronomic and grain traits in 1394 core germplasm lines were estimated. Fairly higher correlation (r = 0.6) between grain Fe and Zn contents suggests ample scope for simultaneous improvement of both the micronutrients in sorghum. Though correlation of grain Fe and Zn contents with days to 50% flowering (0.1 for Fe and 0.2 for Zn), plant height (0.2 for Fe and 0.4 for Zn) is significant and positive, the lower magnitude of the correlation suggests near independence of the crop growth traits and grain micronutrients contents. The results indicate that sorghum grain Fe and Zn contents can be improved in different maturity and plant stature backgrounds. Similarly, significant negative but lower magnitudes of correlation of grain Fe and Zn contents with grain yield (-0.2 Fe and -0.2 Zn) and grain size (-0.1 Fe and -0.1 Zn) suggest that it is possible to enhance grain Fe and Zn contents in high-yielding backgrounds with large grains.

BVS Reddy

Milestone 5C.1.1.3: G × E interactions for grain Fe and Zn contents assessed (BVSR/HDU, 2008)

With a view to assess the effect of soil micronutrient fertilization on sorghum grain micronutrient contents, a set of selected 12 sorghum lines (including hybrid seed parents, restorer lines and popular varieties) contrasting (high and low) for grain Fe and Zn contents were grown in vertisols (medium black soil) and alfisols (red sandy loam soils) with a combination of five different levels of micronutrients in 2005 postrainy season. To rule out the possible confounding effect of deficiency (as is true in experimental field soils of ICRISAT) of other micronutrients such as boron (B) and sulphur (S), Fe and Zn fertilization was combined with recommended levels of boron and sulphur. The five fertilization levels were-first level (T_1): recommended NPK + zero micronutrients; second level (T_2): recommended NPK + recommended Fe @ 10 kg ha^{-1}; (T_3): recommended NPK + recommended Fe @ 10 kg ha^{-1} + recommended S @ 30 kg ha^{-1} + recommended B @ 0.5 kg ha^{-1}; fourth level (T_4): recommended NPK + Zn @ 10 kg ha^{-1} + recommended S @ 30 kg ha^{-1} + recommended B @ 0.5 kg ha^{-1}; and fifth level (T_5): recommended NPK + Zn @ 10 kg ha^{-1} (Table 3). The experiment was laid out in strip-plot design with three replications. The data were collected on plant growth traits such as days to 50% flowering, plant height, 100-grain weight, grain color, panicle shape, panicle compactness, glume color, glume coverage, grain color, presence/absence of seed sub coat and grain yield. Hand threshed selfed seed samples from each entry grown at each fertilizer level and each replication were analyzed for grain Fe and Zn contents in soil chemistry laboratory at ICRISAT, Patancheru.

The analysis of variance (ANOVA) indicated significant mean squares due to genotype and first-order interaction of genotype with soil type and second-order interaction of genotype with soil type and different micronutrient fertilization levels for grain Fe and Zn contents as expected. While soil type did not have significant effect on grain Zn content of the test sorghum lines, it did have significant effect on grain Fe content. Interestingly, non-significant variance due to micronutrient fertilization levels per se suggested poor evidence on the influence of soil micronutrient fertilization on grain Fe and Zn contents in any particular soil type. However, significant mean squares due to interaction of micronutrient fertilization levels with soil type indicated that grain Zn content (but not Fe content) of the genotypes varied with a given combination of micronutrient level and soil type.

The ANOVA indicates only overall trend in variation of lines for grain micronutrient contents as influenced by micronutrient fertilization, and it does not reveal pattern of variation when pair-wise fertilization level effects are examined. For example, mean performance of the lines at different fertilization levels indicated that grain Fe and Zn contents were significantly higher without micronutrient fertilization compared those with both Fe and Zn fertilization per se or in combination with boron and sulphur. However, there were no differences in grain Fe and Zn contents of the lines grown in any of the micronutrient levels. It appears that micronutrient fertilization does not influence the grain micronutrient contents. The results are based on single year and single location data with rather smaller plot size. Further experimentation is needed to confirm these results.

BVS Reddy, HD Upadhyaya and KL Sahrawat

Activity 5C.1.2: Conduct inheritance studies and develop mapping populations for Fe and Zn

Milestone 5C.1.2.1: Genetics of grain Fe and Zn established (BVSR, 2009)

Selfed seed of a total of 12 lines contrasting for grain Fe content consisting of six core germplasm lines (>74 ppm) and six breeding lines (<25 ppm); and 12 lines contrasting for grain Zn content comprising of six core germplasm lines (>57 ppm) and six breeding lines (<15 ppm) were selected and planted in 2006 postrainy season for effecting crosses in half-diallel mating design. The F_1s derived from half-diallel crosses along with parents will be evaluated for investigating the inheritance of grain Fe and Zn contents.

BVS Reddy

Milestone 5C.1.2.2: F_6 RILs from at least one cross developed (BVSR/CTH, 2009)

Two best half-diallel crosses involving the most contrasting parents for Fe and Zn contents will be used to derive recombinant inbred lines (RILs).

BVS Reddy

II. Pearl millet

Output target 5C.1: Magnitude of variability for grain iron (Fe) and zinc (Zn) in more than 300 inbred lines, 50 improved populations, 400 germplasm accessions, and 40 commercial hybrids of pearl millet quantified, and at least three lines and three populations with high levels of Fe (65–75 mg kg^{-1}) and Zn (45–55 mg kg^{-1}) identified (2009)

Activity 5C.1.1: Evaluation of germplasm, breeding lines and improved populations for grain Fe and Zn contents

Milestone 5C.1.1.1: Variability for Fe and Zn in designated hybrid parents, population progenies and improved populations developed in Asia and African region quantified (KNR/RB/KLS, 2007)

Grain Fe and Zn content of designated hybrid parents: Evaluation of a limited number of designated seed parents in an earlier study showed some of these having high levels of grain Fe and Zn contents. Thus, we screened 99 seed parents and 93 restorer parents for Fe content, using the Perls Prussian Blue staining method to get a preliminary assessment of the variability for Fe content. Open-pollinated grains, produced for 1000 grain-mass data that were collected as a part of their characterization for DUS (Distinctness, Uniformity, Stability) database, were used for Fe analysis. Of the 99 seed parents, 29 seemed to have high Fe content (18 with deep blue and 11 with medium blue stain). Of the 93 restorer lines, 23 lines seemed to have high Fe content (10 with deep blue color and 13 with medium blue color). Since there is large variability among the lines both within the B-line and R-line sets, these will be further subjected to laboratory analysis to obtained quantitative estimates of the Fe and Zn contents.

Intra-population variability for Fe and Zn content: A total of 64 S_3 progenies previously derived from a released variety in India, AIMP 92901, and 68 S_2 progenies from another released variety in Africa, GB 8735, that had been identified for high Fe and Zn from the set 1 trial, were field tested during the 2005 rainy season and 2006 postrainy season to determine the intra-population variability. The correlation between two seasons for Fe ($r = 0.75$ in AIMP 92901 and $r = 0.60$ in GB 8735) and Zn content ($r = 0.80$ in AIMP 92901 and $r = 0.64$ in GB 8735) was highly significant ($P<0.01$) and positive. Hence, the mean over the seasons were considered for analysis. Highly significant differences were observed among the progenies for both micronutrients, indicating the possibility of exploitation of

intra-population variability for enhancing the levels of these micronutrients by recurrent selection, and deriving inbred lines with high Fe and Zn to be used as hybrid parents. Based on two-season data, approximately three-fold variation for grain Fe content in AIMP 92901 (40.9 to 118.9 mg kg^{-1}) and in GB 8735 (45.5 to 108.3 mg kg^{-1}), and two-fold variation for grain Zn content (31.8 to 82.7 mg kg^{-1} in AIMP 92901 and 33.8–70.5 mg kg^{-1} in GB 8735) was observed (Figs. 1 and 2). There was highly significant ($P<0.01$) and positive correlation between Fe and Zn in progenies of AIMP 92901 ($r = 0.75$) and GB 8735 ($r = 0.77$).

Fifty progenies, derived from each of the four additional populations (CGP, GGP bulk, ICTP 8203 and PVGGP 6) that were identified for high Fe content in a previous trial in 2004 rainy season were evaluated during the summer and rainy seasons 2006. Results of the summer season grain analysis showed significant differences among the S_1 progenies of ICTP 8203 and CGP. The remaining two populations (GGP Bulk and PVGGP 6) are yet to be analyzed. Approximately two-fold variation was observed for both grain Fe and Zn content in both ICTP 8203 (55.3–138.2 mg kg^{-1} Fe and 38.9–106.7 mg kg^{-1} Zn) and CGP (64.4–150.7 mg kg^{-1} Fe and 57.9–99.6 mg kg^{-1} Zn) (Figs. 3 and 4). Nine progenies of ICTP 8203 and 23 of CGP population had grain Fe content in excess of 100 mg kg^{-1} (100.3–150.7 mg kg^{-1}); and 10 progenies in each of the two populations had grain Zn content in excess of 80 mg kg^{-1} (80.5–106.7 mg kg^{-1}). There was highly significant ($P<0.01$) positive correlation between Fe and Zn content ($r = 0.78$ in ICTP 8203 and $r = 0.74$ in CGP).

About 100 S_1 progenies have been produced from each of the four populations (CGP, GGP bulk, ICTP 8203 and PVGGP 6). Based on the laboratory results available for all the populations, progenies of the two populations having the highest levels of Fe and Zn will be field tested and the grains produced will be analyzed for the Fe and Zn content. About 100 S_1 progenies were also produced from another five populations (Ugandi, Higrop, PVGGT-5, IAC-ISC-TCP 4 and ICMV-IS-94206) for Fe and Zn analysis.

KN Rai and KL Sahrawat

Milestone 5C.1.1.2: Variability for Fe and Zn in iniari germplasm, core collection and commercial hybrids assessed (KNR/RB/HDU/KLS, 2008)

Search for high grain Fe and Zn content in *iniari* germplasm

Grains of 313 *iniari* germplasm accessions along with two checks (ICTP 8203 and ICMR 356) were produced during the 2006 postrainy season for grain Fe determination using Pearls Prussian Blue staining method. Of these, 99 germplasm accessions had deep blue color and 62 had medium blue color. The accessions showing medium to deep blue color flowered in 37–58 days. Based on visual assessment of agronomic traits, 60 accessions were selected, producing 76 S_1 progenies. These were evaluated during the 2006 rainy season, and 50 progenies were selected, of which 4 flowered in 41–45 days and 39 flowered in 46–55 days and (check ICTP 8203 flowered in 46 days). More than 100 S_2 progenies were generated from the selected progenies for further evaluation and utilization in breeding large-seeded hybrid parents.

Evaluation of core collection for grain Fe and Zn content: Since iniari germplasm is genetically more related, the likelihood of finding different genes in this group for either Fe or Zn content is not very high. Thus, attempts are underway to explore new sources of germplasm accessions for higher micronutrient content in the non-iniari groups of materials. A core collection of 504 accessions from 25 countries was screened using the Perls Prussian Blue staining technique on a 1–4 scale (1 = no color; 2 = less intense blue color; 3 = medium blue color and 4 = deep blue color) to identify those with scores 3 and 4. There were 117 accessions each in the deep and medium-blue color stain, indicating that about 46% of accessions in core collection have medium to high Fe content. These will be field grown for grain production and laboratory analysis of the grain Fe and Zn content.

KN Rai, HD Upadhyaya and KL Sahrawat

Milestone 5C.1.1.3: G × E interaction for Fe and Zn assessed and lines stable for >70 mg kg^{-1} Fe and >50 mg kg^{-1} Zn identified (KNR/RB/KLS, 2009)

Stability of grain Fe and Zn content: Twenty-eight entries representing a wide range of Fe and Zn content and selected from a 120-entries trial conducted for two seasons along with a check OPV (WC-C 75) were further evaluated during the summer and rainy season of 2005 and the summer season of 2006 for stability of these

micronutrients. Grain Fe and Zn data from these five trials were subjected to stability analysis following Eberhart and Russell model. Highly significant (P<0.01) differences were observed among entries, among environments, and for entries × environment interaction, both for the Fe and Zn content. However, entries × environment (linear) was significant for only grain Zn content, implying that, in general, linear sensitivity of the different entries was variable with respect to environment, which was not so with grain Fe content. The correlation among the environments was highly significant (r = 0.73 to 0.96; P<0.01 for Fe and r = 0.74 to 0.96; P< 0.01), suggesting, in general, a relatively consistent ranking of genotypes for both grain Fe and Zn content across the environments.
The mean grain Fe content of lines varied from 33.3 to 82.6 mg kg^{-1} and that for Zn it varied from 28.6 to 64.3 mg kg^{-1}. All except three entries were stable both for Fe and Zn. Incidentally, a S_4 progeny from AIMP 92901 had the highest levels of both Fe and Zn, with unit regressions and non-significant deviations from regression both for Fe and Zn., indicating it the most stable line. Seed parent 863B, found to be drought-tolerant, high general combiner for grain yield, and highly resistant to multiple pathotypes of DM, had the next highest level of Fe (71.9 mg kg^{-1}) and had 53.1 mg kg^{-1} of Zn content. This line was also highly stable for both Fe and Zn content.

This stability trial was sent to nine diverse locations in different agro-ecological conditions in India under the ICAR-ICRISAT partnership project to further investigate the stability of these lines for grain Fe and Zn content. Also, based on two seasons' data of a 69-population trial, 18 populations with diverse grain Fe and Zn density were identified for their stability evaluation.

KN Rai

Output target 5C.2: Information on genetics and recurrent selection efficiency for grain Fe and Zn available (2009)

Activity 5C2.1: Conduct genetical studies and recurrent selection for grain Fe and Zn contents and develop mapping populations

Milestone 5C.2.1.1: Inheritance of Fe and Zn and relationship between the parental lines and hybrids for these traits determined (KNR/RB/KLS, 2009)

Genetics of grain Fe and Zn content: Recent research has identified several pearl millet breeding lines with high levels of gain iron (Fe) and zinc (Zn) content, and large variability for these traits in the improved breeding lines and populations. Utilization of this variability in breeding hybrid parents and populations with enhanced levels of these micronutrients can be more effective following efficient breeding methodologies based on the nature of inheritance. Genetics of these traits has not been reported so far in pearl millet. In an initial attempt in this direction, 10 inbred lines with a wide range of Fe and Zn contents were crossed in a diallel fashion (including reciprocals). The resulting 90 F_1s and 10 parents were evaluated during 2005 rainy season and 2006 summer seasons. Sib-mated grains were used for laboratory analysis of grain Fe and Zn content using dry ashing and Atomic Absorption Spectrophotometry method at the National Institute of Nutrition, Hyderabad.

Entry × season interaction was significant, but it was of negligible order. Reciprocal effect was non-significant for both grain Fe and Zn content. Therefore, means over the reciprocal crosses and the seasons were subjected to half-diallel analysis. The genetic component analysis indicated absence of epistasis for both traits, the Wr-Vr graph revealed presence of partial dominance for grain Fe and Zn content. The predictability ratio measured by $2\sigma^2gca/(2\sigma^2gca + \sigma^2sca)$ was around unity for both grain Fe (0.86) and Zn content (0.88), implying preponderance of additive gene action. Also, there was highly significant positive correlation between the mid-parental values and mid-parent heterosis (r = 0.79; P<0.01 for Fe and r = 0.81; P<0.01 for Zn), which was an additional indication of the predominant role of additive gene action for these traits. The grain Fe and Zn content in parents were highly correlated with their *gca* effects (r = 0.92; P<0.01 for Fe and r = 0.94; P<0.01 for Zn). The average heterosis was negative and negligible both for grain Fe (-6.1%) grain Zn content (-2.1%). In general, the high grain Fe and Zn contents in parents were governed by recessive alleles with increasing effects and the low contents were due to excess of dominant alleles with decreasing effects. The correlations between the performance *per se* of the inbred lines and their general combining ability (GCA) were positive and highly significant both for Fe (r=0.79) and Zn (r=0.81), indicating that selection of lines with high levels of these micronutrients will be highly effective in selecting lines with high GCA for these traits.

KN Rai

Milestone 5C.2.1.2: Effectiveness of S_1 recurrent selection for Fe and Zn, and its effect on grain yield and other agronomic traits in four populations quantified (KNR/RB/KLS, 2012)

Recurrent selection for high Fe and Zn content: Nine progenies with high grain Fe and Zn content were selected separately from AIMP 92901 (81.5–104.0 mg kg-1 Fe and 57.0–68.0 mg kg-1 Zn) and GB 8735 (78.5–104.5 mg kg-1 Fe and 57.0–59.5 mg kg-1 Zn), and random mated in 2006 summer to initiate recurrent selection. The C1 cycle bulks and original bulks of both the populations were evaluated in four replications during the 2006 rainy season and grains were produced for the laboratory analysis.

KN Rai and KL Sahrawat

Milestone 5C.2.1.3: QTL for high grain Fe and Zn identified based on F_6 RIL mapping populations from two crosses (CTH/SS/KNR, 2010)

Plant x plant crossing of five pairs of candidate parental lines, was followed by head-to-row advance of the F1 hybrids and their parents. Phenotypic assessment of Fe and Zn grain density of the parental lines, accompanied SSR marker polymorphism assessment of the parents, and visual assessment of the F1 progenies and selfed progenies of their parental plants, resulted in identification of four candidate populations from which F6 RIL populations can be derived to map these traits in pearl millet, and two of these have been chosen for generation advance during 2007

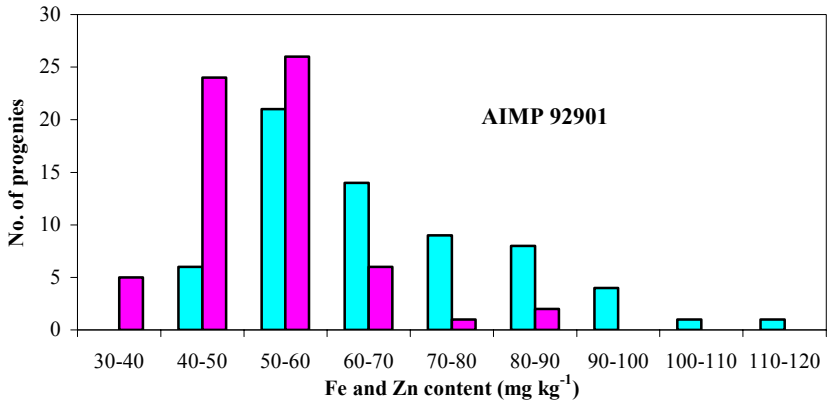

Figure 1. Frequency distribution of AIMP 92901 (S_3) progenies for grain Fe and Zn content, 2005 rainy and 2006 summer season, ICRISAT, Patancheru.

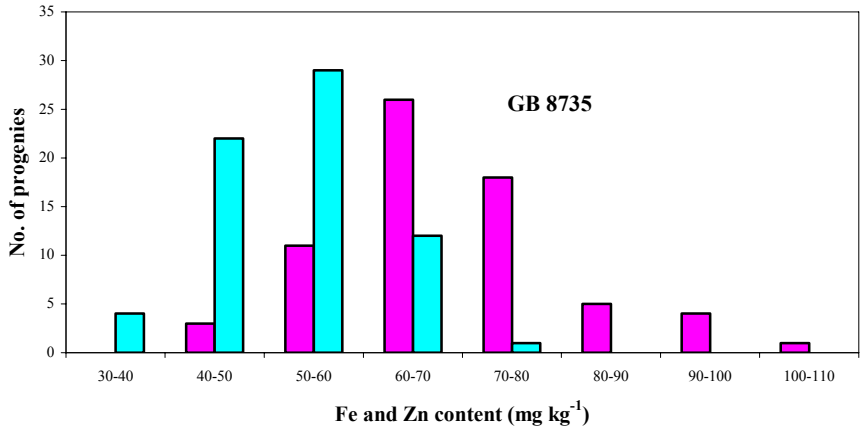

Figure 2. Frequency distribution of GB 8735 (S_2) progenies for grain Fe and Zn content, 2005 rainy and 2006 summer season, ICRISAT, Patancheru.

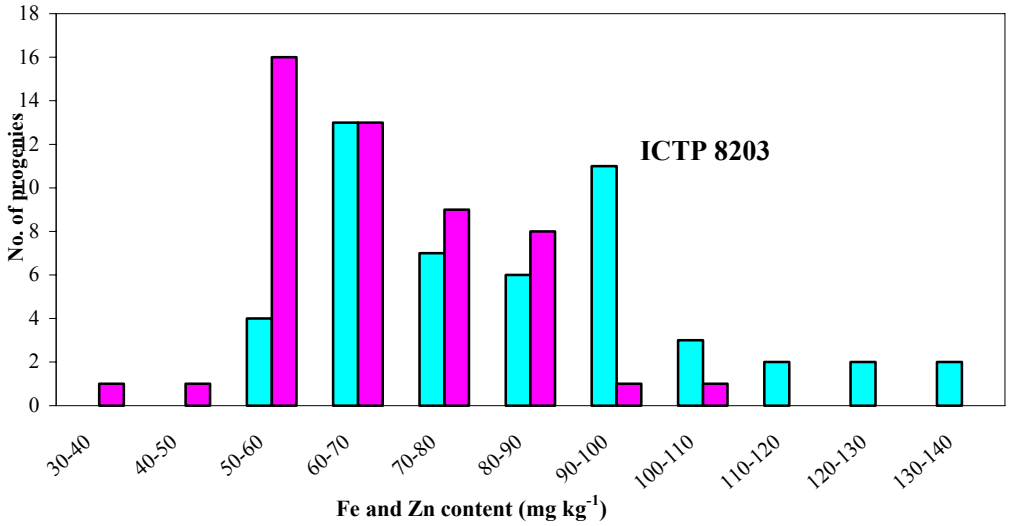

Figure 3. Frequency distribution of ICTP 8203 (S_1) progenies for grain Fe and Zn content, 2006 summer season, ICRISAT, Patancheru

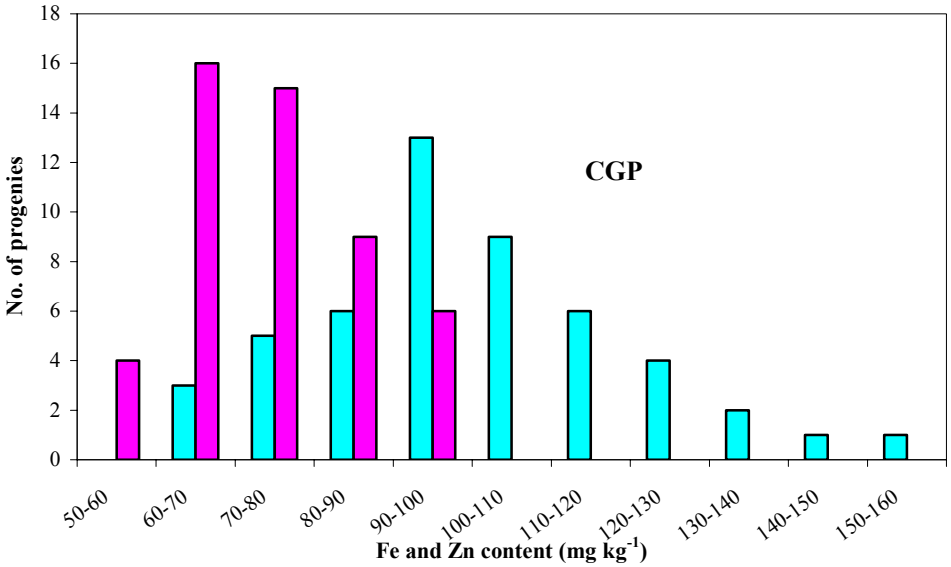

Figure 4. Frequency distribution of CGP (S_1) progenies for grain Fe and Zn content, 2006 summer season, ICRISAT, Patancheru.

Evaluation of sorghum and pearl millet for mycotoxins

Sorghum and pearl millet are staple food for millions of poor people in India, and they are important raw material in food and feed processing industry. These crops are affected by several fungi, causing grain molds (GM) that deteriorate grain quality. Some GM fungi can produce toxic metabolites, termed as mycotoxins, in grains that are hazardous to human and animal health. Of various grain molds, aflatoxins produced by *Aspergillus* species and fumonisins produced by *Fusarium* species are most frequent contaminants in sorghum and pearl millet. To enhance the quality of the grain by mitigating mycotoxin contamination through improved production technologies, participatory on-farm trials were conducted in Maharastra and Andhra Pradesh states, India, and grain samples from these trials were collected for mycotoxin analysis to assess the effect of improved varieties and cultural practices on mycotoxin contamination. A total of 440 sorghum grain samples were collected from 88 farmer fields from 5 villages (Udityal, Kakarjala, Veerannapalli, Rangampalli and Bandapalli) and they were analyzed for aflatoxins and fumonisin B1 by enzyme-linked immunosorbent assay (ELISA).

Aflatoxin contamination in sorghum samples collected from Udityal ranged from 0 to 561 μg kg^{-1}. Samples from 12% of the fields contained high levels of aflatoxins; lowest toxin levels (0–4.2 □g kg^{-1}) were found in samples from Rangampalli. In remaining villages, aflatoxin ranged between 0 and 48 □g kg^{-1}. Majority of the samples tested had toxins under permissible levels of 30 □g kg^{-1}; 28.4% samples had no detectable levels of aflatoxins; in 50.7% it was between 1 and 5 □g kg^{-1}; in 14.1% samples it ranged between 5 and 30 □g kg^{-1}; and 3.2% and 3.6% of the samples had 30.1–100 and 100.1–561 □g kg^{-1} aflatoxins. Fumonisin contamination in sorghum samples ranged from 0 to 1356 μg kg^{-1} with samples from 8% of the fields containing >100 μg kg^{-1}. Eighty-one percent of the individual sorghum samples were free from fumonisin contamination and 7% contained >100 μg kg^{-1} fumonisin.

Pearl millet samples (505) collected from 101 farmers' fields from six villages (Palavai, Peddapalli, Pavanampalli, Parmal, Kuruvupalli, Kondapalli) were analyzed for aflatoxins and fumonisin. Aflatoxin contamination ranged from 0 to 1047 μg kg^{-1}. Samples from 16% of the fields contained >30 μg kg^{-1}. In Pavanampalli, Parmal, Kondapalli villages, 29–38% fields contained >30 μg kg^{-1} aflatoxin. To our knowledge, this is the first record of high levels of aflatoxin contamination in pearl millet. Fumonisin contamination in pearl millet ranged from 0 to 186 μg kg^{-1} and only 1% of the samples collected from various farmers fields contained >100 μg kg^{-1}.

A field day was organized at Palavai village (Mahbubnagar district, Andhra Pradesh, India) to disseminate the technologies for improved productivity of crops and mitigate grain mold contamination in sorghum and millet. More than 200 participants (farmers and NGOs) participated in the field day.

Farid Waliyar

Project 6

Producing more and better food at lower cost of staple open-pollinated cereals and legumes (sorghum, pearl millet, pigeonpea, chickpea and groundnut) through genetic improvement and crop management in the Asian SAT

Groundnut

Output A: Improved germplasm and varieties of sorghum, pearl millet, pigeonpea, chickpea, and groundnut with pro-poor traits and advanced knowledge of selection tools and breeding methods made available to partners internationally

MTP Output Targets 2006:
At least 15 new varieties with resistance to late leaf spot and rust available and shared with partners

Farmer preferred varieties in India, Vietnam and China identified and disseminated amongst partners

Activity 6A.1.1: Evaluate and introgress new germplasm sources (cultivated and wild *Arachis* species) of variability for yield components, resistance to rust, LLS, and other emerging diseases, crop duration, and food and fodder quality traits

Milestone: At least 100 crosses involving diverse germplasm and breeding lines for aforementioned traits effected (SNN/RA/FW/PLK) 2009

Ninety-seven crosses (42 for foliar diseases, 22 for medium-duration, 10 for short-duration, and 23 for confectionery traits) were made during the 2005/06 post-rainy and the 2006 rainy seasons to generate populations for selection for high yield, diseases resistance, desired crop duration, and confectionery traits in desirable agronomic backgrounds. New parents used in hybridization included high-yielding foliar diseases tolerant breeding lines (ICGV 04060, ICGV 04055, ICGV 04078, and ICGV 04093); germplasm lines (ICG 7340, ICG 6843, ICG 7621, and ICG 6330); high-yielding and medium-duration advanced breeding lines (ICGV 04112, ICGV 04124, ICGV 04149, ICGV 99159, and ICGV 95069); short-duration advanced breeding lines (ICGV 00308, ICGV 93392, ICGV 00290, and Nyanda); high-yielding, advanced breeding lines with confectionery traits, (ICGV 99083, ICGV 00350, ICGV 00451, and ICGV 00440), and germplasm lines (ICG 6767, ICG 6670, and ICG 1651).

SN Nigam and R Aruna

Milestone: Evaluation of groundnut lines for resistance to late leaf spot (LLS) and rust during under field conditions:

Late leaf spot (LLS) (*Phaeoisariopsis personata*) and rust (*Puccinia arachidis*) are the most serious fungal diseases of groundnut, particularly in the rainy season. Systematic screening of groundnut germplasm and breeding lines was initiated in the field and laboratory to incorporate resistance into high yielding cultivars with agronomic and quality characters suited to different environments. Ten groundnut breeding lines (ICGV 37, ICGV 00005, ICGV 00064, ICGV 01270, ICGV 01276, ICGV 92267, ICGV 86590, ICGV 87846, ICGV 99029, and ICGV 00068) along with the susceptible cultivar TMV 2, and a resistant control ICG 13919, were evaluated against late leaf spot and rust at 61, 74, 92, 106, and 123 days after sowing (DAS) at ICRISAT, Patancheru, India, during the 2006 rainy season. Highly significant differences were observed among the genotypes in both the trials for disease score (LLS and rust) and leaf area damage (LAD).

Late leaf spot (LLS): At 92 days after sowing (DAS), of the ten advanced groundnut breeding lines evaluated, ICGV 00068 (LLS score = 2.7; LAD = 7.0) was highly resistant. Six lines showed a disease score between 3.0 – 5.0 and LAD = 11.0 - 26.0 as compared to the resistant check ICG 13919 (LLS score = 2.7 and LAD = 6.3) and susceptible check TMV 2 (LLS score = 7.0 and LAD = 60.0) (Fig. 1).

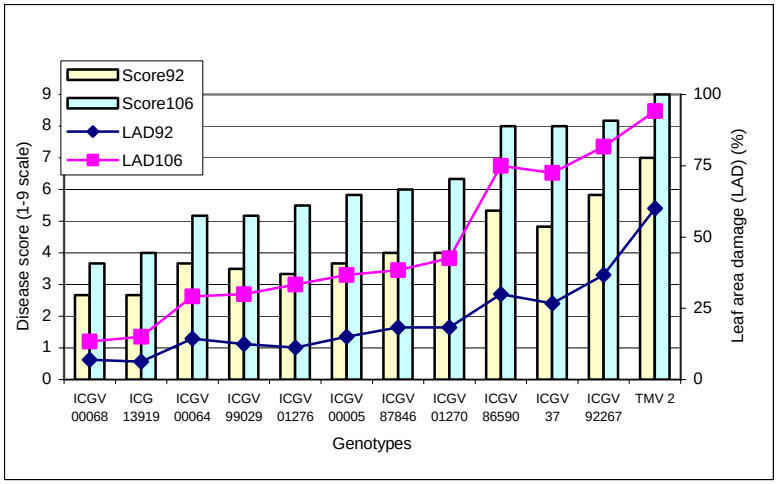

Fig. 1. Evaluation of advanced groundnut breeding lines for resistance to late leaf spot (ICRISAT, Patancheru, 2006 rainy season)

Rust: At 92 DAS, of the 10 advanced groundnut lines tested, eight lines showed a disease score of 1 – 2 and LAD = 0.3-1.7, and two lines (ICGV 92267 and ICGS 37) showed moderate levels of resistance (rust score = 3.8 -4.2, and LAD = 18.3 – 20.0) as compared to the resistant check ICGV 86699 (rust score = 1.8 and LAD = 0.8) and susceptible check TMV 2 (rust score = 6.0 and LAD = 40.0) (Fig. 2). ICGV 00064 showed resistance to both rust and LLS.

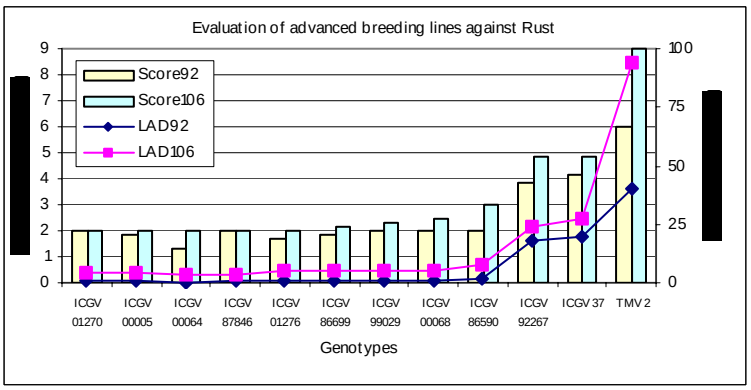

Fig. 2. Evaluation of advanced groundnut breeding lines for rust resistance (ICRISAT, Patancheru, 2006 rainy season).

Evaluation of advanced breeding lines for resistance to LLS and rust: Six replicated yield trials (elite foliar diseases resistant groundnut varietal trial (Spanish bunch) (EFDRGVT - SB), elite foliar diseases resistant groundnut varietal trial (Virginia bunch) (EFDRGVT - VB), advanced foliar diseases resistant groundnut varietal trial (Spanish bunch) (AFDRGVT - SB), advanced foliar diseases resistant groundnut varietal trial (Virginia bunch) (AFDRGVT - VB), preliminary foliar diseases resistant groundnut varietal trial (Spanish bunch) (PFDRGVT - SB), and preliminary foliar diseases resistant groundnut varietal trial (Virginia bunch) (PFDRGVT -VB); consisting of 101 advance breeding lines were screened for resistance to rust and LLS in an experimental sick-plot containing inoculum bearing infector-rows. Experiment was laid out in a broad-bed-and-furrow (BBF) system with two replications. Size of each plot was 1.5 x 4 m, with inter-row spacing of 30 cm, and plant to plant spacing of 10 cm within a row. Chemical sprays were used to control insect pests. At 45 days after sowing, plots were inoculated by spraying the infected and test rows with mixed conidial suspension of *P. personata* and *P. arachidis* urediniospores. After inoculation, perfo-irrigation was provided daily for 30 min in the evening for 30 days to increase humidity

required for disease development. Diseases (LLS and rust) were scored on a 1 - 9 rating scale (1 = highly resistant, and 9 = highly susceptible) at 89 and 105 days after sowing.

Development of LLS and rust was uniform throughout the infector rows, and 100% infection was observed in the susceptible controls. Highly significant differences were observed among the genotypes in all the trials against LLS and rust. For all the trials, coefficient of variation was in the range of 4 to 22% for LLS, and 5 to 15% for rust . Of 101 breeding lines tested, none of the test lines was resistant to LLS. Twenty-four lines showed an overall disease severity score of 4.0. Another 16 lines had a score of 4.5, 29 lines were scored 5.0, and 15 lines rated as moderately resistant (score 5.5) (Table 1). Eighty-eight of these lines showed good resistance to rust (disease score of 2); 11 lines scored 2.5 to 3; and the remaining two lines ICGV 05092 recorded 8.0 and ICGV 06160 scored as 6.5 at 105 days after sowing (Table 1).

Table 1: Evaluation of advanced breeding lines for late leaf spot and rust resistance (ICRISAT, Patancheru, 2006 rainy season)

Trial	No. of lines tested	Disease severity rating					
		LLS			Rust		
		1-3*	4-6*	7-9*	1-3*	4-6*	7-9*
EFDRGVT (SB)	11	Nil	10	1	11	Nil	Nil
EFDRGVT (VB)	5	Nil	5	Nil	5	Nil	Nil
AFDRGVT (SB)	15	Nil	14	1	14	Nil	1
AFDRGVT (VB)	16	Nil	16	Nil	16	Nil	Nil
PFDRGVT (SB)	28	Nil	24	4	27	Nil	1
PFDRGVT (VB)	26	Nil	25	1	26	Nil	Nil
Total	**101**	**Nil**	**94**	**7**	**99**	**Nil**	**2**

*Disease score on a 1 - 9 scale, where 1 = highly resistant, and 9 = highly susceptible for LLS and rust.

Preliminary evaluation for resistance to foliar diseases: Two foliar disease-resistant groundnut variety (PFDRG - VT) trials consisting of 164 lines were screened against LLS and rust. Six lines showed resistance to LLS with a score of 3.0, while 121 had a score of 4 - 6. For rust, 137 lines showed high levels of resistance (disease score 2.0). The remaining lines had a score of >3.0 on a 1 - 9 scale.

Farid Waliyar, Lava Kumar, SN Nigam and R Aruna

Performance of F_2 to F_6 populations for resistance to LLS and rust under field conditions: Five breeding populations (F_2 to F_6, 118 lines) were evaluated for resistance to LLS and rust under field conditions during the 2006 rainy season in an un-replicated, 9 m row plot, with 60 cm inter-row spacing, and 10 cm distance between the plants. TMV 2, a highly susceptible cultivar to both LLS and rust, was sown in infector rows after every five test rows. Forty-five days after sowing, plots were inoculated by spraying 30 L of mixed suspension of *P. personata* conidia and *P. arachidis* urediniospores. One day after inoculation, perfo-irrigation was provided daily for 30 minutes in the evening up to 30 days to favor disease development. All the populations were evaluated on a 1 - 9 rating scale for reaction to foliar disease at harvest. The development of LLS was uniform throughout the infector rows. The breeding populations were categorized as resistant (disease score 1 - 3), moderately resistant (disease score 4 - 6) and susceptible (disease score 7 - 9). A few single plants from F_6 population showed moderate levels of resistance to LLS.

Farid Waliyar, Lava Kumar, SN Nigam and R Aruna

Screening of advanced breeding lines for resistance to *Aspergillus flavus* infection and aflatoxin contamination: During the 2005 post-rainy season, 106 advanced breeding lines were evaluated for resistance to *Aspergillus flavus* seed infection and aflatoxin contamination. The breeding lines were planted in five separate trials in the *A. flavus* sick plot. The fungal inoculum multiplied on sorghum and maize grains was applied in the soil at two-weekly intervals during the crop growth period, and end of season drought stress was imposed 30 days before harvest to facilitate seed infection. Harvesting was done manually and pods were sun dried for 2 - 3 days. Pods from each plot were collected separately, shelled manually and kernel sub-samples were analyzed for *A. flavus* seed infection by

blotter plate method and aflatoxin contamination by indirect competitive ELISA. The seed infection in the test lines ranged from 0 to 40% and aflatoxin contamination was between 0 to 6,668 µg kg^{-1}, respectively. Five of the 30 elite aflatoxin-resistant lines (ICGV 01002, ICGV 01149, ICGV 02148, ICGV 02189, and ICGV 02191) showed <1% *A. flavus* seed infection and <2 µg kg^{-1} aflatoxin. All the 18 advanced Virginia bunch lines were susceptible (aflatoxin >20 µg kg^{-1}) to aflatoxin contamination. Among 21 advanced Spanish bunch varieties, eight lines (ICGV 03300, ICGV 03308, ICGV 03328, ICGV 03331, ICGV 03332, ICGV 03337, ICGV 03344, ICGV 03346, and ICGV 03352) had less than 5% seed infection and less than 5 □g kg^{-1} aflatoxin. One hundred thirty advanced breeding lines in four replications were screened in the sick plot for resistance to *A. flavus* infection and aflatoxins contamination during the 2006 rainy season. Kernels were harvested, sun dried, and pods were stripped manually. The samples are being analyzed for *A. flavus* infection and aflatoxin contamination.

Farid Waliyar, SN Nigam and R Aruna

Milestone: Screening groundnut germplasm and breeding material for resistance to foliar pests organized (GVRR/SNN/HDU/RA) 2009

Consolidation of information on resistant sources for various insect pests on groundnut has been completed. Screening of elite breeding material against insect vectors and defoliators under field condition is in progress during the post-rainy 2006-07.

Milestone: 15 - 20 new high yielding lines with resistance to biotic stresses and quality, and adaptation traits identified and made available to NARS (SNN/RA/FW/PLK) - Annual

The trait-specific international trials are made available on request to collaborators in NARS. The promising lines from on-station elite trials feed into international trials, are organized every two years.

Foliar diseases resistance: We evaluated 369 advanced breeding lines (including controls) in 14 replicated trials, and 108 advanced breeding lines in 2 augmented trials during the 2005/06 post-rainy and the 2006 rainy seasons. The disease scores were recorded on a 1 - 9 scale (where 1 = no disease, and 9 = >80% foliage damaged) at 105 DAS.

In the elite variety trial, Spanish types, ICGV 02410 was the best performer (4.7 t ha^{-1} pod yield; 63% shelling outturn) during the 2005 rainy season. The highest yielding control in the trial was ICGV 98374 (4.2 ± 0.33 t ha^{-1}). ICGV 02410 performed equally well during the 2005 rainy season (3.4 ± 0.21 t ha^{-1} pod yield). In the elite variety trial, Virginia types, ICGV 02434 (4.9 ± 0.46 t ha^{-1} pod yield) outperformed the best control ICGV 98373 (3.2 ± 0.45 t ha^{-1} pod yield**).**

In Spanish elite variety trial, nine lines significantly out yielded (3.0 - 2.0 ± 0.21 t pod yield ha^{-1}) the highest yielding control, ICGV 98374 (1.6 t ha^{-1} pod yield). In this trial, ICGV 04060 (3.0 t pod yield ha^{-1}, 64% shelling out-turn, LLS score = 5.0, and rust score = 2.0) was the best line. In the Virginia elite variety trial, three entries ICGV 04087, ICGV 04091, and ICGV 04094 out-yielded the best control ICGV 86699 (1.7 t ha^{-1}, 60%, 4.0 and 2.0). Five lines (ICGV 02410 and ICGV 02411 in the Spanish group, ICGV 02429, ICGV 02434, and ICGV 02446 in the Virginia group) were selected for inclusion in international trials.

High oil content: Forty-six breeding lines were evaluated during the 2005/2006 post-rainy season for oil content and pod yield in a replicated trial. ICGV 01274 (5.5 t ha^{-1} pod yield, 49.0% oil content, and 1.9 t ha^{-1} oil yield) and ICGV 00009 (5.4 t ha^{-1}, 50.5%, 1.8 t ha^{-1}), produced significantly higher pod yield than the highest yielding control, ICGV 86564 (4.6 t ha^{-1}, 47.0%, 1.5 t ha^{-1}). Both of these lines also had higher oil yield. But for oil content *per se*, ICGV 00351 (4.3 t ha^{-1}, 51.0 %, 1.6 t ha^{-1}) and ICGV 00171 (4.4 t ha^{-1}, 51.5%, 1.5 t ha^{-1}) were quite good. The oil content of entries in this trial ranged from 51.5 - 44.7%, and the protein content from 26.9% - 18.4%. Twelve lines had an oil content ≥50%. In the 2006 rainy season, ICGV 01274 (4.3 t ha^{-1} pod yield) was the top performer under irrigated conditions.

Medium-duration: Seven hundred and ninety advanced breeding lines (including controls) in 16 replicated trials, and 76 lines in 2 augmented trials were evaluated in the 2005/2006 post-rainy and 2006 rainy seasons for pod yield and other agronomic traits.

In the advanced Spanish type trial in 2005/06, 12 lines significantly out-yielded (pod yield 4.6 - 5.6 ± 0.52 t ha^{-1}) the best control ICGS 11 (4.0 t ha^{-1} pod yield). The best entry in the trial was ICGV 04115 (5.6 t ha^{-1}pod yield, 68% shelling outturn, and 50.4% oil), followed by ICGV 04124 (5.5 t ha^{-1}, 69%, and 47.6%). In the advanced Virginia type trial 14 entries produced significantly higher pod yield (4.3 - 5.5 ± 0.60 t ha^{-1}) than the best control ICGS 76 (3.7 t ha^{-1}). ICGV 04140 (5.5 t ha^{-1} pod yield) and ICGV 04139 (5.5 t ha^{-1}) were the best entries in this trial. However, the former had higher 100-seed weight, and the latter higher oil content.

In the Spanish elite variety trial, all 7 test lines outperformed (3.7 - 4.8 ± 0.18 t ha^{-1} pod yield) the highest yielding control ICGV 95058 (2.5 t ha^{-1} pod yield) during the 2006 rainy season. The best entry ICGV 04124 (4.8 t ha^{-1}) originated from the cross (ICGV 92069 x ICGV 93184) x ICGV 98105. In the Virginia elite variety trial, three lines; ICGV 04149 (4.9 t ha^{-1}), ICGV 04140 (4.6 t ha^{-1}), and ICGV 04139 (4.6 t ha^{-1}), out-yielded the control, ICGV 86325 (2.7 ± 0.43 t ha^{-1}). The best entry came from (ICGV 92069 x ICGV 93184) x ICGV 99171 cross. In another Virginia elite variety trial (conducted with the material transferred from short-duration trials), all 22 lines significantly out-yielded (3.8-3.1 ± 0.43 t ha^{-1}) the best control ICGS 76 (2.5 t ha^{-1}). Seven medium-duration lines were selected in the 2005/2006 post-rainy season for inclusion in international trials.

Confectionery types: One hundred and sixty-two advanced breeding lines (including controls) were evaluated in nine replicated yield trials under high input conditions during the 2005/2006 post-rainy and the 2006 rainy seasons. In the 2005/06 post-rainy season, none of the test entries out-yielded the best control in any trial. In the 2006 rainy season, 6 lines (3.5 - 2.7 ± 0.16 t ha^{-1} pod yield; 57 - 66% shelling outturn; and 48 - 56 g 100-seed weight) out-yielded the best control ICGV 99085 (2.3 t ha^{-1}; 65% shelling out-turn; and 42 g 100 seed weight). The best entry ICGV 05176 (3.5 t ha^{-1}; 57% shelling outturn; 49 g 100 seed weight) came from ((ICGV 88414 x USA 63) x ICGV 95172) x ICGV 00440) cross. In another advanced trial (Virginia), 2 lines, ICGV 05198 (3.1 t ha^{-1}) and ICGV 05200 (3.1 t ha^{-1}) outperformed the highest yielding control ICGV 00440 (2.6t ha^{-1}). The best entry came from ((BPI Pn 9 x ICGV 95172) x ICGV 88414) x (USA 63 x ICGV 95172)).

Short-duration: We evaluated 385 lines (including controls) in 14 replicated trials and 192 advanced breeding lines in 4 augmented trials for their yield and other agronomic traits under irrigated conditions in the 2005 rainy and the 2005/2006 post-rainy seasons. The Spanish trials were harvested when the crop accumulated 1470 °Cd (equivalent to 90 DAS in the rainy season) and the Virginia and large-seeded trials when the crop accumulated 1705 °Cd (equivalent to 105 DAS in the rainy season) at ICRISAT Center, Patancheru, India.

None of the new breeding lines out-yielded the best control in all the eight replicated yield trials in 2005/2006 post-rainy season. In another trial, 45 elite entries, originating from different series of international short-duration groundnut varietal trial (ISGVT) in the past 10 years, and other elite trials, were evaluated with 4 controls in a 7 x 7 lattice design. Thirteen entries out-yielded (3.4-3.9 ± 0.36 t ha^{-1} pod yield) the highest yielding early-maturing control J 11 (3.0 t ha^{-1} pod yield). ICGV 00308 (3.9 t ha^{-1} pod yield), ICGV 97243 (3.8 t ha^{-1}), and ICGV 00338 (3.7 t ha^{-1}) were the most promising. ICGV 97243 belonged to the IX series, and ICGV 00308 to the X series of ISGVT and ICGV 00338 to elite trial.

In the Spanish elite varietal trial, 6 lines significantly out-yielded (1.8 - 1.4 ± 0.12 t ha^{-1} pod yield) the highest yielding control, Chico (1.1 t ha^{-1} pod yield) in 2006 rainy season. ICGV 03211 (1.8 t ha^{-1}), the best entry among the 6 lines, came from (ICGV 98191 x ICGV 93382) cross. In another Spanish elite varietal trial, 6 lines (2.4 - 2.0 ± 0.13 t ha^{-1} pod yield) produced significantly higher pod yield than the highest yielding control ICGV 91114 (1.8 t ha^{-1}). ICGV 02038 (2.4 t ha^{-1}), the best entry among the 6 lines, came from (ICGV 95244 x ICGV 92206) cross. Two lines (ICGV 02022 and ICGV 02144) were identified for inclusion in the international trials during the 2005/2006 post-rainy season.

SN Nigam and R Aruna

On station evaluation of groundnut genotypes for LLS: Ten advanced breeding lines were supplied to scientists at the University of Agriculture Sciences, Bangalore, Karnataka, India. These lines were planted during the 2005 rainy season at UAS, Hebbal Campus, Bangalore, along with the susceptible check TMV 2 to evaluate their resistance against LLS. The test genotypes were evaluated for disease severity at 15 day intervals and the final scores are presented in the Table 2. Six genotypes (ICGV 01270, ICGV 00005, ICGV 00064, ICGV 01276, ICGV 87846, and ICGV 00068) showed good level of resistance. All the ten genotypes are being evaluated during the 2006 rainy

season to select promising lines for multilocation on-farm evaluation in southern Karnataka during the 2007 rainy season.

Table 2. On station evaluation of groundnut genotypes for LLS (UAS, Bangalore, 2005 rainy season).

Genotype	Percent disease index (PDI) as on 2/10/2005	Percent disease index (PDI) as on 15/10/2005	Percent disease index (PDI) as on 26/10/2005
ICGV 01270	16.66	23.33	32.21
ICGV 92267	46.66	79.99	94.99
ICGV 00005	15.55	21.10	33.33
ICGV 00064	15.55	26.66	33.88
ICGV 01276	14.99	23.33	31.66
ICGV 86590	44.44	72.21	78.32
ICGV 87846	19.99	28.33	39.44
ICGV 37	38.32	74.43	92.76
ICGV 99029	14.99	19.99	34.44
ICGV 00068	13.88	17.22	27.77
TMV 2	61.66	87.76	96.10

Farid Waliyar, Lava Kumar, SN Nigam and R Aruna

On-farm evaluation of *Aspergillus flavus* and aflatoxin resistant varieties in Andhra Pradesh, India: On-farm participatory varietal selection trials and the participatory evaluations of improved varieties have been carried out with four (ICGV 91278, ICGV 91328, ICGV 94379, and ICGV 94434) varieties in Anantapur; and five (ICGV 91114, ICGV 91341, ICGV 93305, ICGV 94379, and ICGV 94434) varieties in Pileru area, in Andhra Pradesh, India. The trials were planted in 18 farmers' fields in six villages of the each of the two districts. Performance of the 4 selected groundnut improved varieties was better in all the 18 farmers' fields in six villages in Anantapur district and produced higher pod and haulm yield than the control TMV 2. Highest pod yield 1029 kg ha^{-1} was obtained with ICGV 94434 in Cherlopalli village. The variety ICGV 94434 produced 40 - 43% higher pod yield in three villages, and in the remaining 3 villages it produced 23 - 34% higher pod yield than the control TMV 2. From each plot, about one kg pod sample was drawn (after recording the bulk pod yield). The damaged pod sub-samples and the remaining pods were shelled and sorted in large and small kernel sub-samples. Large seeds, which are mostly used for confectionery purpose, showed 0.4 - 32% *A. flavus* infection and 0 -897 µg kg^{-1} aflatoxin across the villages and varieties. Overall, improved varieties showed reduction in *A. flavus* infection (31 - 50%) and aflatoxin contamination (56 - 93%) over the controls. Aflatoxin levels in three varieties (ICGV 91328, ICGV 94379, and ICGV 94434) was <20 µg kg^{-1} against 128 µg kg^{-1} in control TMV 2. Analysis of immature small sized kernels showed *A. flavus* infection and aflatoxin contamination in the range of 0 - 22% and 0 - 679 µg kg^{-1} g, respectively. Three improved varieties showed 82 - 93% reduction (<10 µg kg^{-1}) in aflatoxin against 58 µg kg^{-1} in control, and these three varieties were promising in large seed category also. Irrespective of variety, kernels from damaged pods contained high levels of aflatoxin (range 88 - 393 µg kg^{-1}) because damaged pods are vulnerable to fungal infection and subsequent aflatoxin production. Considering the complex nature of the aflatoxins problem in groundnut, the overall mean of the six villages for large and small sized kernels of improved varieties showed good tolerance to aflatoxins contamination. In addition, these lines produced 15 - 34% higher pod and haulm yields than the local control TMV 2.

Farid Waliyar, Lava Kumar and SN Nigam

Milestone: Wild Arachis species evaluated for Tobacco streak virus (TSV) resistance and durable resistant genotypes identified (PLK/FW/SNN/RA) 2007

Evaluation of wild *Arachis* for resistance to *Tobacco streak virus*: Stem necrosis disease caused by *Tobacco streak virus* (TSV) has emerged as a potential threat to groundnut in southern states of India. All the 189 groundnut cultivars currently grown in India and over 400 cultivated genotypes tested at ICRISAT, Patancheru, India are highly susceptible to the virus. Therefore, wild relatives of peanut were evaluated to identify potential sources of resistance to TSV infection. Fifty-six germplasm accessions from twenty wild *Arachis* species of four sections (*Arachis, Erectoides, Procumbente,* and *Rhizomatosae*) along with the susceptible peanut cultivars (JL 24 and K 1375) were evaluated for resistance to TSV under greenhouse conditions by mechanical sap inoculations. Percent

TSV infection was recorded both on inoculated and subsequently emerged leaflets based on visual symptoms and by enzyme-linked immunosorbent assay (ELISA). Systemic virus infection in the test accessions ranged between 0 and 100%. Twenty-four of these accessions in section *Arachis* that had 0 to 35% systemically infected plants were re-tested, and eight of these accessions did not show systemic infection in repeated trials in the greenhouse. These were ICG 8139, ICG 8195, ICG 8200, ICG 8203, ICG 8205, and ICG 11550 (*A. duranensis*), ICG 8144 (*A. villosa*), and ICG 13210 (*A. stenosperma*). Although, these accessions showed TSV infection in inoculated leaves ranging between 0 and 100%, the virus was not detected in the subsequently emerged leaves, indicating block in systemic spread of virus amounting to the disease resistance. The eight TSV resistant accessions are cross compatible with *A. hypogaea* for utilization in breeding for stem necrosis disease resistance. The resistant accessions, ICG 8139 and ICG 11550 also possess high levels of resistance to rust (*P. arachidis*) and late leaf spot (*P. personata*), and ICG 8144 to Peanut bud necrosis virus, and thus have multiple resistance to virus and pathogens, and can be used to develop multiple disease resistant peanut varieties through inter-specific breeding.

Lava Kumar, Farid Waliyar, SN Nigam and R Aruna

Milestone: 50 lines of advanced generation interspecific derivatives of groundnut evaluated for LLS disease and promising lines identified (FW/NM/PLK) 2008

Screening of advanced interspecific derivatives against late leaf spot resistance: About 84 selections from 4 wide crosses (Arachis kempf-mercadoi, A. glabrata, A. batizocoi, and A. duranensis) were screened for resistance to late leaf spot (LLS) under field conditions during 2006 rainy season. Field trials were laid out in a broad-bed-and-furrow (BBF) system with three replications. The size of each plot was 1.5 x 4 m, with inter-row spacing of 30 cm, and plant to plant spacing of 10 cm. TMV 2, a highly susceptible cultivar to LLS was used as an infector row after every five-test rows. Chemical sprays were used to control insect pests. At 45 days after sowing, plots were inoculated by spraying the conidial suspension of P. personata urediniospores. After inoculation, perfo-irrigation was provided daily for 30 min in the evening for 30 days to create high humidity required for disease development. The LLS incidence and severity was scored on a 1 - 9 rating scale at 91 and 106 days after sowing. The development of LLS was uniform throughout the infector rows. Among the test lines, highly significant differences were observed and coefficient of variation was in the range of 14.4 to 15.1%. Out of 84 interspecific derivatives, twenty-two lines from A. hypogaea X A. cardenasii showed high level of resistance to LLS with a score of 2, Fifty-one lines had a score of 2 to 3, nine had a score of 3 to 4, and the remaining two lines were rated >5 at 106 days after sowing (Fig. 3). The disease severity in the susceptible check was 9.0. Promising lines with a disease rating of 3 and less will be advanced.

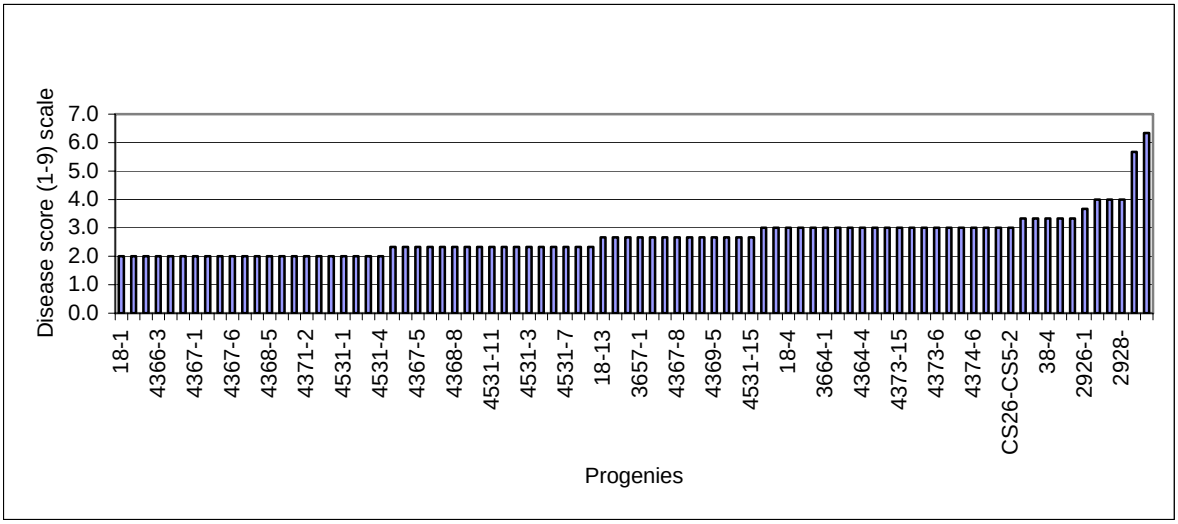

Fig. 3. Screening of advanced interspecific derivatives against late leaf spot at 106 days after crop emergence (ICRISAT, Patancheru, 2006 rainy season).

Farid Waliyar, Lava Kumar and Nalini Mallikarjuna

Evaluation of progenies from crosses of wild *Arachis* for resistance to *Aspergillus flavus*: Sixty progenies from wide hybridization crosses were evaluated for resistance to *A. flavus* seed colonization by *in vitro* assay. From each progeny, 60 seeds were surface sterilized with sodium hypochlorite and inoculated with *A. flavus* spores and kept in a moist blotter paper in a petridish in a humid box and incubated at 28°C for 6 days. Percent seed infection at the end of the 6th day was estimated. The seed colonization ranged between 7 to 52% in all the progenies evaluated. Only 5 progenies (numbers. 2929-6, 2929-15, 4367-3, 4368-7, and 4373-15) showed less than 10% seed infection, while the rest showed a susceptible reaction. Uninfected seed from all the sixty progenies were retrieved and planted in the *A. flavus* sick plot at ICRISAT-Patancheru, for resistance evaluation under field conditions during the 2006 rainy season. Field screened samples are being processed for *A. flavus* seed infection and aflatoxin contamination and results are awaited.

Farid Waliyar, Lava Kumar and Nalini Mallikarjuna

Evaluation of interspecific derivatives for resistance to LLS: Eighty-four advanced interspecific derivatives lines were screened for LLS in 2005, which had a score of 2 to 3 on a 1 - 9 scale (1 = immune, and 9 = highly susceptible). These were screened for late leaf spot (LLS) in rainy season 2006. The lines which had a score of less than 3 in 2006, are presented in Table 3.

Table 3. Groundnut interspecific derivatives with stable resistance to LLS (ICRISAT, Patancheru, 2006 rainy season).

Genotype No:	Generation	*Wild species used in the cross*	2005	2006
LCBG-1	BC2F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-2	BC5F6	*A .hypogaea* x *A. cardenasii*	2	2.0
LCBG-3	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-4	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBG-5	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBG-6	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-7	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBG-8	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBG-9	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-10	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.7
LCBG-11	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-12	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBG-13	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-14	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.7
LCBG-15	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBG-16	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-17	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBG-18	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBP-19	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBP-20	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.7
LCBG-21	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBG-22	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-23	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBG-24	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.7
LCBG-25	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.7
LCBG-26	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBG-27	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-28	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBG-29	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBG-30	BC3F5	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBG-31	BC3F5	*A. hypogaea* x *A. cardenasii*	3	2.7
LCBG-32	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.0
LCBG-33	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0

Genotype No:	Generation	*Wild species used in the cross*	2005	2006
LCBG-34	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBG-35	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBG-36	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBG-37	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.7
LCBG-38	BC5F6	*A. hypogaea* x A. cardenasii	3	2.0
LCBG-39	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBG-40	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.0
LCBP-41	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3
LCBP-42	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBP-43	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBP-44	BC5F6	*A. hypogaea* x *A. cardenasii*	3	2.3
LCBP-45	BC5F6	*A. hypogaea* x *A. cardenasii*	2	2.3

Farid Waliyar, Nalini Mallikarjuna and Lava Kumar

Milestone: 8 - 10 selected advanced breeding lines in each country evaluated for local adaptation and farmer-preferred traits in farmers' fields in SAT Asia (Special Projects) (SNN/RA/FW/PLK) 2009

With the aim of selecting farmer-preferred varieties under moisture stress and popularize them, an Integrated Scheme of Oilseeds, Pulses, Oilpalm and Maize (ISOPOM)-funded project on "Farmers participatory groundnut improvement in rainfed cropping systems" was launched in 2006. The project is being implemented by ICRISAT and National Research Centre for Groundnut (NRCG) in Andhra Pradesh, Orissa, and Gujarat. Anantapur and Chittoor districts were selected for project implementation in Andhra Pradesh. A farmer participatory varietal selection (FPVS) trial consisting of 9 varieties (K 1271, K 1375, TCGS 888, TPT 25, ICGV 00350, ICGV 00308, ICGV 86015, ICGV 91114, and a local control) was conducted in five villages (Varigireddipalli in Kadiri, Potharajukalava and Medhapuram in Anantapur, and Seenapagaripalli and Marikuntapalli in Chittoor), and also at all the three research stations (Agricultural Research Station, Kadiri and Anantapur; and Regional Agricultural Research Station, Tirupati) in Andhra Pradesh . Results of the FPVS Mother-baby trials would be available in the near future.

SN Nigam and R Aruna

Progress reported towards the achievement of milestone for 2007 above will contribute towards achievement of the milestones listed below.

Milestone: Five interspecific derivatives of groundnut evaluated for TSV and peanut bud necrosis virus (PBNV) diseases and promising lines identified (NM/FW/PLK/SNN/RA) 2010

Milestone: Field trials of five stable, promising interspecific derivatives conducted in target location for LLS resistance and yield (NM/FW/PLK/SNN/RA) 2011

Milestone: Field trials of three stable and promising derivatives for TSV and peanut bud necrosis diseases conducted in Anantapur and other target locations (NM/FW/PLK/SNN/RA) 2012

Activity 6A.1.2: Develop a better understanding of inheritance of components of resistance to late leaf spot (LLS), confectionery traits, and traits associated with drought tolerance (specific leaf area (SLA), and SPAD chlorophyll meter reading (SCMR)

Milestone: Knowledge of inheritance of components of resistance to LLS in three crosses gained and appropriate breeding strategy devised (SNN/RA/FW) 2008

Inheritance of components of resistance to rust and LLS: Two LLS resistant germplasm lines (ICG 11337 and ICG 13919) and a susceptible variety JL 24 were used as parents in straight and reciprocal crosses for developing the materials for studying the inheritance of components of LLS resistance. Under controlled environmental

conditions, fully expanded quadrifoliate leaves of 45 days old plants (third or fourth from the top) of each line were excised and planted in sand cultures (roughly 1.5 cm thick) in plastic trays (39.5 × 29 × 7 cm). In each tray, 20 leaflets were planted and the trays were covered with plastic bags and incubated in the growth chamber at 24 ºC and 85% relative humidity. The LLS inoculum (30000 spores ml^{-1}) was sprayed on both the surfaces of each leaflet. Some plants showed latent period (LP) of 30 - 36 days after inoculation and 2 - 5 lesions with minimum lesion diameter (<0.5 mm). Detailed observations on incubation period, latent period, lesion number, % leaf area damage (LAD), and lesion diameter were recorded. In the field, plants were inoculated with conidial suspension of *P. personata* at 50 DAS. After inoculation, perfo-irrigation was provided daily for 15 min in the evening for 30 days. The field observations included % defoliation at 75, 90, and 105 DAS, and the disease scores on a 1- 9 scale. The data are being processed.

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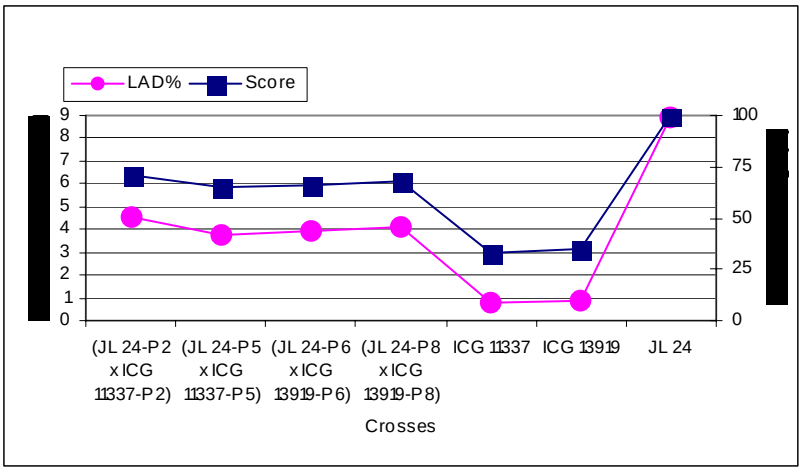
Inheritance of resistance to late leaf spot (LLS) under field and controlled environment conditions: F_3 progenies (439) of two crosses (ICGV 11337 X JL 24 and ICGV 13919 X JL 24) along with parental lines (total 2556 single plants) were planted in an unreplicated trial in 60 cm inter-row spacing, and 10 cm between the plants, and screened for resistance to LLS under field conditions during the 2006 rainy season. TMV 2, a highly susceptible cultivar, was used as an infector row after every five-test rows. Plants in field trials were inoculated with conidial suspension of *P. personata* at 45 days after sowing. After inoculation, perfo-irrigation was provided daily for 30 min in the evening hours for 30 days. Subsequently, leaf area damage (%) and percentage of defoliation was measured in each plant. Plant reaction to LLS was recorded on a 1 - 9 rating scale at 82, 97, and 110 days after sowing (DAS). Of the two crosses and three parents, only two plants from JL 24-P6 x ICG 13919-P6, and one plant from JL 24-P2 x ICG 11337-P2 showed high levels of resistance to LLS with a score of 2.0; 42 progenies had a score of 3.0, 218 had a score of 4.0, and 337 recorded a score of 5.0 at 110 days after sowing. Remaining plants were scored >6.0. Data showed that the segregation is very high in the crosses, and there were no differences in the parent line plants (Table 4).

Table 4. Area under the disease progress curve (AUDPC), disease score, and leaf area damage (LAD) in F_3 progenies (ICRISAT, Patancheru, 2006 rainy season).

Crosses	No of Progenies	AUDPC			Score *			LAD (%) *		
		Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
(JL 24-P2 x ICG 11337-P2)	166	1567	500	2581	6	2	9	51	5	95
(JL 24-P5 x ICG 11337-P5)	18	1687	816	2485	6	3	8	41	10	80
(JL 24-P6 x ICG 13919-P6)	134	1411	568	2265	6	2	9	43	5	95
(JL 24-P8 x ICG 13919-P8)	118	1534	511	2459	6	4	9	46	10	95
ICG 11337	1	752	647	935	3	3	3	9	6	10
ICG 13919	1	719	593	813	3	3	4	10	6	12
JL 24	1	1779	1553	1987	9	9	9	100	95	100

* Score = Late leaf spot disease score on 1 - 9 scale; LAD = Leaf area damage (%) at 110 days after sowing.

Based on mean data of all the plants in each progeny, four progenies and two parent lines showed resistance to LLS with score of 3.0, 38 progenies showed a disease score of 4.0, 79 had score of 5.0, and the remaining progenies recorded a score of 6 - 9 on 1 - 9 scale. Leaf area damage was 7.5 - 95.5%. Area under the disease progress curve (AUDPC) was observed in a range from 615 to 2285 among the progenies at 110 days after sowing. Taking the mean data of all the plants and progenies in each cross, it was observed that the AUDPC, disease score and leaf area damage were more than double in JL 24 and in crosses, compared to ICG 11337 and ICG 13919 parent lines (Fig. 4a, b).



Crosses are mixed – Needs clarity (Spread)

Fig. 4a. Disease score and leaf area damage at 110 days after sowing in F_3 crosses (mean of all progenies) (ICRISAT, Patancheru, 2006 rainy season).

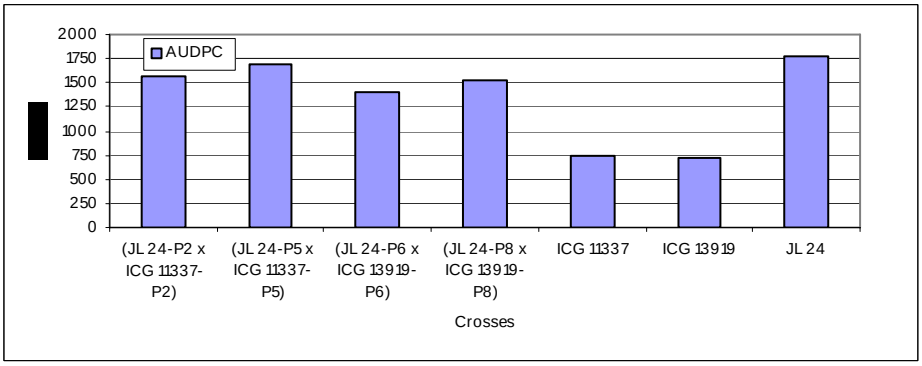


Fig. 4b. Area under the disease progress curve (AUDPC) of F_3 crosses (mean of all progenies of each cross) (ICRISAT, Patancheru, 2006 rainy season).

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Milestone: Knowledge of inheritance of traits associated with drought tolerance in three crosses gained and appropriate breeding strategy devised (SNN/RA/VV) 2009

To study the inheritance of traits associated with drought tolerance, four crosses (ICGS 76 x ICGV 93291, ICGV 99029 x ICGV 91284, JL 24 x ICGV 86031, and ICR (Is this ICR or ICG?) 48 x ICGV 99029) and their reciprocals were made involving diverse parents with high and low values of SCMR (SPAD Chlorophyll Meter Reading) and SLA (Specific Leaf Area). The F_1 plants have been produced and backcrosses would be attempted in the 2006/2007 post-rainy season.

SN Nigam and R Aruna

Milestone: Knowledge of inheritance of confectionery traits in two crosses gained and appropriate breeding strategy devised (SNN/RA) 2009

To study the inheritance of confectionery and quality traits (seed characteristics, O/L ratio, oil and protein content, and blanching ability), parents, F_{1s}, F_{2s} and backcrosses of two crosses (Chico x ICGV 01393 and Chico x ICGV 02251) along with their reciprocals have been sown in a replicated trial during the 2006/2007 post-rainy season.

SN Nigam and R Aruna

Milestone: Three mapping populations for LLS and two for confectionery traits developed (RA/SNN) 2009

Three mapping populations (ICG 11337 x JL 24, ICG 13919 x JL 24, and ICG 11337 x ICG 13919) involving diverse parents for reaction to LLS have been developed and the material is in F_4 stage. Two populations (ICGV 01393 x Chico and ICGV 02251 x Chico) for confectionery traits have also been developed and the material is in F_3 stage.

SN Nigam and R Aruna

Output target 6A.2: Promising transgenic events of groundnut for resistance to TSV and PBNV available for commercialization and introgression in locally adapted germplasm

Activity 6A.2.1: Develop transgenic events of groundnut for resistance to TSV and evaluate their performance under contained greenhouse and field conditions

Milestone: 100 transgenic events of groundnut with TSVcp gene developed and screened in contained greenhouse (KKS/PLK/SNN) 2007

Stem necrosis disease caused by the *Tobacco streak virus* (TSV) has emerged as a serious problem on groundnut in Andhra Pradesh, and Karnataka, India. All the currently grown groundnut varieties are susceptible to the virus. Research has been initiated to incorporate resistance to TSV in groundnut by using TSV coat protein (*TSVcp*) gene through *Agrobacterium tumefaciens*-mediated genetic transformation of popular groundnut cultivars JL 24, TMV 2, and ICGV 91114. Twenty events, with six plants per event, were planted in the P_2 greenhouse. A cotyledonous leaflet was collected from 8 - 10 day old plants and analyzed for transgene by PCR assay, which revealed *TSVcp* transgene in 68 of 118 plants tested (58% transformation rate).

At the 3-leaf plant growth stage (8 - 10 day old plants), all the 118 test plants [alongwith susceptible control (JL 24)] were inoculated by standard mechanical sap inoculation procedure using 1: 30 (w/v) TSV-affected French bean leaf sap extracts. All the inoculated plants developed necrotic symptoms on the inoculated leaves 7-days post inoculation and tested positive to TSV with ELISA. These plants were monitored at weekly intervals for systemic virus infection by testing newly emerged apical leaves by ELISA, and symptoms were recorded. All the non-transgenic plants (*TSVcp* gene negative in PCR assay) were susceptible to TSV and showed symptoms typical of TSV infection within 2-weeks post inoculation. Of the 68 transgenic plants (*TSVcp* gene positive), 24 plants (35.2%) showed negative reaction to TSV (no symptoms) or delayed expression of symptoms compared to controls. Delayed symptom expression (by 2 - 3 weeks compared to controls) was also observed in 42 plants (61.7%). After appearance of initial symptoms, disease progress was rapid leading to premature death of the plants. It is likely that these plants may have some tolerance at early growth stages offering resistance to virus spread, but this mechanism is not sufficient to protect plants at later stages. All the symptomatic plants, either transgenic or controls, tested positive to TSV with ELISA, and all asymptomatic plants were negative, indicating a correlation between virus presence and the stem necrosis disease.

Eight plants from six events (1B, 1F, 3E, 4B, and 9C) did not show any systemic symptoms and non-inoculated leaves were TSV negative, indicating putative TSV resistance in these plants. Considering the fact that TSV was detected in the inoculated leaves of these plants and the lack of virus in the subsequently emerged leaves suggests a blockage in the systemic spread of virus, which seems to be responsible for the virus resistance. Delayed symptom expression (at the time of flowering) was observed on one or two branches in events 9B, 19B, and 22B. These plants apparently had normal growth pattern. It is likely that these plants may also have some resistance amounting to the protection. Further evaluation of these events is being continued. Recently, genetic transformation of groundnut cv. ICGV 91114 has also been undertaken to incorporate transgenic resistance to TSV using *TSVcp* gene.

KK Sharma, Lava Kumar and Farid Waliyar

Progress reported towards the achievement of milestone for 2007 above will contribute towards achievement of the milestones listed below.

Milestone: At least 10 promising TSVcp transgenic events identified and the disease resistance characterized under contained green house conditions (KKS/PLK/SNN) 2008

Milestone: Five promising TSVcp transgenic events identified and the disease resistance characterized under contained field conditions (KKS/PLK/SNN) 2009

Milestone: Two best transgenic events with resistance to TSV used for introgression into locally adapted groundnut genotypes and their evaluation (KKS/PLK/SNN/RA) 2010

Milestone: Commercialization package for groundnut with transgenic resistance to TSV available for deployment (KKS/PLK/SNN/RA) 2011

Activity 6A.2.2: Develop transgenic events of groundnut for resistance to PBNV and evaluate their performance under contained greenhouse and field conditions

Milestone: 100 transgenic events of groundnut with PBNVnp gene or alternative gene developed and screened in contained greenhouse (KKS/PLK/SNN/RA) 2008

Peanut bud necrosis disease (PBND) is an economically important virus disease of groundnut caused by *Peanut bud necrosis virus* (PBNV), transmitted by thrips, for which no durable resistance is available in the groundnut germplasm. Transgenic approach was undertaken to engineer resistance to PBNV using nucleoprotein (*PBNVnp*) gene in groundnut cultivar JL 24. Forty-eight independent transgenic events were produced by using two binary vectors encoding for *PBNVnp* gene through *Agrobacterium tumefaciens* and micro-projectile mediated genetic transformation. The progeny of the transgenic plants were mechanically inoculated with PBNV at two concentrations, 1: 50 and 1: 100 (w/v) under P_2 greenhouse conditions. In the T_1 generation, at 1: 100 concentration of leaf sap inoculum, 24 of the 36 events tested did not show any symptoms and virus was not detected in these plants with ELISA. However, at higher concentration [1: 50], all the 24 events were infected by the virus, but the symptoms were delayed by 40 to 60 days in six events, when compared with the untransformed controls, which showed nearly 100% mortality within two weeks after inoculation. The 24 events were evaluated in a contained on-station trial at ICRISAT, Patancheru farm, during 2005. Planting was done with wide spacing to attract thrips vector for the virus inoculation. Most of the 24 events were affected by PBND, however, symptom appearance was delayed by 2 to 3 weeks in the transgenic plants compared to the controls. Although all the infected transgenic groundnut plants showed severe PBND symptoms, eight plants from the events B 8, B 9, B 11, B 13, B 15, B 19, B 20, and B 22 showed recovery, suggesting some tolerance to PBND. Apparent lack of resistance to PBNV in transgenic plants could be attributable to the presence of RNA silencing suppressor gene, NSs, in the PBNV genome, which could be rendering *PBNVnp* gene ineffective. Further studies have been planned to develop RNAi construct to counter the effect of NSs gene for transformation either by genetic engineering or conventional crossing into *PBNVnp* transgenic events. In addition, strategies are being developed to use RNAi constructs for conserved domains of PBNV replicase, nucleoprotein or movement genes, combined with RNAi constructs to counter and NSs gene for inducing RNAi-mediated resistance to PBNV in groundnut and other crops susceptible to this virus.

KK Sharma, Lava Kumar and Farid Waliyar

Progress reported towards the achievement of milestone for 2008 will contribute towards achievement of the milestones listed below.

Milestone: At least 10 promising transgenic events identified and resistance to PBNV characterized under contained greenhouse conditions (KKS/PLK/SNN/RA) 2009

Milestone: Five promising PBNVcp or alternative gene transgenic events identified and the disease resistance characterized under contained field conditions (KKS/PLK/SNN/RA) 2010

Milestone: Two best transgenic events with resistance to PBNV used for introgression into locally adapted groundnut genotypes and their evaluation (KKS/PLK/SNN/RA) 2011

Chickpea

Output A: Improved germplasm and varieties of sorghum, pearl millet, pigeonpea, chickpea, and groundnut with pro-poor traits and advanced knowledge of selection tools and breeding methods made available to partners internationally

MTP Output Target 2006: 20 new high yielding fusarium wilt resistant kabuli and desi chickpea breeding lines made available to NARS

Chickpea: Six lines (ICC 14194, ICC 14206, ICC 14215, ICC 17109, WR 315, and KAK 2) have been found to be asymptomatic to Fusarium wilt in chickpea. Three lines (ICCX-990026-F3-BP-25-BP, ICCX-990026-F3-BP-34-BP, and ICCX-990026-F3-BP-40-BP) were also superior to KAK 2 in seed yield, 100-seed weight, and flowering. Five lines (ICCV 05525, ICCV 05526, ICCV 05534, ICCV 97207, and ICCV 98501) have been identified to be resistant to BGM and FW, while ICCV 05523 showed resistance to AB and FW.

Large-seeded Kabuli chickpea germplasm lines ICC 14194 and ICC 14198 have been found to be very early (days to maturity <100 days) and had 50 to 53 g 100 seeds^{-1}, suggesting that it is possible to breed early-maturing Kabuli varieties with extra-large seed with FW resistance. Analysis of eight Indian isolates of Ascochyta using 20 RAPD primers (decamers) placed the isolates into two groups.

The 2-day-old eggs of *Helicoverpa armigera* can be stored at 10ºC for 10 days for bioassays at convenient times on insect host plant interactions, biological control, and toxicology. The genotypes RIL 27, RIL 51, RIL 81, RIL 83, ICC 10613, ICCL 87315, ICCL 87322, ICCL 87315, and ICCV 10 suffer less damage by *Helicoverpa* and have a yield potential comparable to commercial cultivars, and these can used in chickpea improvement for resistance to *H. armigera.*

High levels of antibiosis to *H. armigera* have been recorded in wild relatives of chickpea belonging to the tertiary gene pool (*Cicer judaicum*, *C. bijugum*, *C. cuneatum*, and *C. microphyllum*), and moderate levels of resistance in the secondary gene pool (*C. reticulatum*).

Bacillus thuringiensis (*Bt*) sprays on chickpea reduced the cocoon formation of *Campoletis chlorideae* reared on *Bt* intoxicated larvae of *H. armigera* (18.7 - 38.7% compared to 69.3 to 77.3% on untreated controls). The ELISA test indicated the presence of *Bt* protein in the *H. armigera* larvae fed on *Bt* treated chickpeas, while no *Bt* protein was detected in the cocoons and adults of the parasitoid, *C. chlorideae*, suggesting that lower survival of the parasitoid was due to the poor quality of the host.

Output target 6A.1: Nearly 50 - 100 chickpea breeding lines with high yield, improved seed quality traits, and resistance to one or more biotic stresses [Fusarium wilt (FW), Ascochyta blight (AB), Botrytis gray mold (BGM) and *Helicoverpa*] developed and disseminated to the NARS

Activity 6A.1.1: Develop chickpea breeding lines (Desi and Kabuli) with enhanced resistance to AB, BGM, and FW

Milestone: 15 - 20 high yielding FW resistant Desi and Kabuli chickpea breeding lines made available to NARS (SP/PMG) Annual

Evaluation of advanced Desi and Kabuli chickpea breeding lines and populations for resistance to Fusarium wilt: We evaluated 162 entries (143 + 19 checks), 244 crossing block entries/parents, 47 F_2 and three F_4 populations for FW resistance under artificial epiphytotic conditions. In addition, preliminary yield trial (PYT)-Desi (23 + 3), PYT-Kabuli (52 + 8), advanced yield trial (AYT)-desi (18 + 2), AYT-Kabuli (14 + 2), international chickpea screening nursery (ICSN)- Desi (18+2), and ICSN-Kabuli (18+2) were also evaluated for resistance to wilt. Wilt incidence in early-wilting cultivar, ICC 4951 was 100% in 30 days after sowing (DAS) and same in 90 DAS in the late wilting cultivar, K850 across the wilt-sick field. In the PYT-Desi, nine entries (ICCX 990009-F_3- BP-9-BP, ICCX 990009-F_3- BP-12-BP, ICCX 990022-F_3- BP-2-BP, ICCX 990009-F_3-BP-13-BP, ICCX 990009-F_3- BP-19-

BP, ICCX 990009-F_3- BP-20-BP, ICCX 990011-F_3- BP-12-BP, ICCX 990026-F_3- BP-6-BP, and ICCX 970010-F_3-BP-P13-BP-BP) showed a resistant (<10% FWI incidence) reaction. In PYT-Kabuli, 17 were asymptomatic and 20 were resistant (<10% FW incidence). In AYT-Desi, ICCX 970047-BP-BP-P24-BP-BP-BP, and ICCX 970047-BP-BP-P52-BP-BP-BP were asymptomatic, and eight lines showed a resistant reaction. In AYT-Kabuli, four were (ICCX 980068-F_4-P10-BP-BP, ICCX 980068-F_4-P13-BP-BP, ICCX 980068-F_4-P23-BP-BP, and ICCX 970075-BP-BPP43-BP-BP-BP) asymptomatic and three were resistant. In ICSN-Desi, ICCV 04103 and ICCV 05114 were asymptomatic and 11 lines were resistant to wilt. Among the ICSN-Kabuli entries, seven genotypes (ICCV 03407, ICCV 04305, ICCV 04308, ICCV 04309, ICCV 05306, ICCV 05308, and ICCV 05312) were free from wilt, and eight were resistant (10%). In 244 crossing block entries, six lines (ICC 14194, ICC 14206, ICC 14215, ICC 17109, WR 315, and KAK 2) were asymptomatic and 10 lines (Vijay, Jumbo 2, JG 11, JGK 1, ICCV 89302, ICCV 98502, ICC 7032, ICC 13053, ICC 14248, and ICC 16340) were resistant. Individual healthy plants were selected from F_2 and F_4 populations.

Suresh Pande and PM Gaur

International Chickpea Screening Nurseries (ICSNs) of Desi and Kabuli chickpea: A total of 61 sets of two ICSNs (ICSN-Desi and ICSN-Kabuli) were supplied to NARS in eight countries (Australia, Bangladesh, Ethiopia, India, Jordan, Myanmar, Nepal, and Pakistan) during 2005/06. Each nursery consisted of 18 entries and two checks - one common check (ICCC 37 in ICSN-Desi and KAK 2 in ICSN-Kabuli) and one local check. The entries were evaluated in a randomized complete block design (RCBD) with two replications. Each plot had four rows of 4.0 m each. One set of each nursery was evaluated at ICRISAT-Patancheru. The results from India were compiled in a report, which was distributed to Indian NARS during the annual meeting of All India Coordinated Research Project on Chickpea at Mahatma Phule Krishi Vidyapeeth (MPKV), Rahuri, Maharashtra, India. The most promising breeding lines were ICCV 03203, ICCV 05113, and ICCV 04111 in ICSN-Desi; and ICCV 04311, ICCV 05308, and ICCV 97306 in ICSN-Kabuli. During the 2006 cropping season, 67 sets of ICSN-Desi and ICSN-Kabuli were supplied to NARS in 9 countries (Brazil, China, India, Iraq, Myanmar, Nepal, Pakistan, Portugal, and South Africa). In addition to these, 812 samples of advanced breeding lines, 191 of released varieties, and 52 samples of segregating populations were supplied to 11 countries (Australia, Canada, England, Eritrea, India, Iraq, Japan, Mali, Morocco, Nigeria, and Vietnam).

PM Gaur

Evaluation of advanced breeding lines of Desi and Kabuli chickpeas: Advanced breeding lines of Desi (41 lines) and Kabuli (56 lines) chickpeas were evaluated in two advanced yield trials (AYTs) and four preliminary yield trials (PYTs) during the 2005/06 post rainy season. Three most promising lines identified in AYT-Kabuli included ICCX-980061-F4-P15-BP-BP, ICCX-980068-F4-P10-BP-BP, and ICCX-980068-F4-P13-BP-BP. These lines outperformed the best check, KAK 2 with respect to seed yield, 100-seed weight, and earliness, and had high level of FW resistance. In AYT-Desi, ICCX-980055-F4-P53-BP-BP and ICCX-970038-BP-BP-P12-BP-BP-BP were promising for yield, ICCX-970047-BP-BP-P24-BP-BP-BP and ICCX-970047-BP-BP-P52-BP-BP-BP for seed size and FW resistance, and ICCX-970077-BP-BP-P32-BP-2BP-BP for earliness and seed size. Three lines (ICCX-990026-F3-BP-25-BP, ICCX-990026-F3-BP-34-BP, and ICCX-990026-F3-BP-40-BP) were superior to KAK 2 in seed yield, 100-seed weight, and flowering. ICCX-990006-F3-BP-10-BP showed >12% seed yield increase over KAK 2. In the PYT-Desi, ICCX-990023-F3-BP-3-BP had more 100-seed weight, and higher level of FW resistance as compared to check variety ICCC 37.

PM Gaur and Suresh Pande

Milestone: 20 - 30 sources of resistance to FW, BGM, and AB tested for stability across locations and pathotypes in Asia (SP/PMG) Annual

Around 30 Fusarium wilt, Ascochyta blight, and Botrytis gray mold resistant/moderately resistant cultivars were evaluated at different locations and pathotypes to identify stable and broad-based resistance to these diseases.

Chickpea wilt and root rot nursery (CWRRN): Chickpea wilt and root rot nursery consisted of 30 entries (28 wilt resistant + 2 wilt susceptible cultivars) and was evaluated at 22 locations in India during 2005/06 season. Each entry was planted in one row, 4 m long, and there were two replications. Data on wilt was recorded at the flowering and at maturity stages of the crop. Data from fourteen locations (Akola, Badnapur, Bangalore, Berhampore, Dhaulakuan, Dholi, Hazaribagh, Hisar, ICRISAT, Patancheru, Jabalpur, Junagadh, Ludhiana, Rahuri, and Sehore) were received and compiled. Incidence of FW was very high in both susceptible cultivars at all the locations, except at Hazaribagh,

where the nursery was planted in a normal field. ICC 12467 and ICC 14433 in eight locations, ICCX 950106-F4-66P-BP in seven locations, and ICC 14344, ICC 14391, ICC 14432, ICC 14436, and ICC ICCX 950110-F4-26P-BP-BP in five locations were found to be resistant (<10% incidence) to wilt. Preliminary results indicated considerable diversity in the population of *F. oxysporum f.sp. ciceris.*

International Ascochyta blight nursery (IABN): Thirty- nine AB promising entries and one susceptible check were included in IABN and evaluated under field conditions at two locations (Islamabad and Attok) in Pakistan, and five locations (Dhaulakuan, Gurdaspur, Ludhiana, Hisar, and Patancheru) in India. Each entry was planted in two replications with one row of 2 - 4 m long. Artificial inoculation with conidial suspension was done at flowering and pod initiation stages of the crop at all the locations. The IABN was evaluated under controlled environment conditions at ICRISAT following standardized evaluation technique. Data were received from all the five Indian locations, but not from Pakistan. Susceptible cultivar Pb 7 showed a susceptible reaction at all the five locations in India. Eight lines; ICC 1069, ICC 1400, ICC 12952, ICC 15978, ICC 1700, ICCX 810800, ICCX 900221-31-PABR, and ICCX 910028-46 PABR-BP-1PABR-L were showed resistant (1.1 to 3 rating on 1 - 9 rating scale) to moderately resistant (3.1 to 5 rating) reaction to AB at all the five locations in India. ICC 4991 and ICC 14344 were found to be susceptible at all the five locations.

International Botrytis gray mold nursery (IBGMN): The IBGMN consisted of 30 entries (29 moderately resistant and one susceptible check), and was evaluated under field conditions at two locations in Nepal (Rampur and Tarahara), two locations in Bangladesh (Ishrudi and Jessore), and four locations in India (Pantnagar, Gurdaspur, Ludhiana, and ICRISAT-Patancheru). At ICRISAT- Patancheru, the nursery was evaluated under controlled environment conditions. All the entries were artificially inoculated with conidial suspension of the local isolate at the flowering and pod initiation stages of the crop at all the locations, except at Tarahara, Nepal; where the nursery was evaluated under natural epiphytotic conditions. Data was received from Ludhiana, Pantnagar, and ICRISAT-Patancheru. Susceptible cultivar showed a highly susceptible reaction at all the three locations. ICC 1069 and ICCX 850498-3PN-17H-BH-BH in three locations; and ICC 12512, ICC 12952, ICC 14344, ICC 14559, ICC 14824, ICCV 89302, ICCV 89303, ICCV 98505, ICCL 86242, ICCL 87322, and ICCX 860029-BH-1PN-BPN-B in two locations (ICRISAT, Patancheru and Pantnagar) were found to be moderately resistant (3.1 to 5 rating on a 1 - 9 rating sale).

Suresh Pande and NARS Collaborators

Milestone: 5 - 10 new sources of resistance to AB and BGM identified (SP/PMG) 2009

Identification of new sources of resistance to Ascochyta blight (AB) in advanced breeding and germplasm lines: One hundred and ninety seven AB promising advanced breeding and germplasm selections (36 ICRISAT bred lines, 23 germplasm lines, 42 lines from ICARDA, and 96 lines from Australia) were evaluated for AB resistance under controlled environment conditions. Eighty-five lines (18 ICRISAT bred lines, 7 germplasm lines, 24 ICARDA lines, and 36 lines from Australia) were found to be moderately resistant (3.1 to 5 rating on 1 - 9 scale) to AB. Susceptible cultivar Pb 7 showed had a damage rating of 9.0.

Suresh Pande and PM Gaur

ICAR-ICRISAT collaborative research on AB resistance: In collaboration with Indian Institute of Pulses Research (IIPR), Kanpur, 171 lines (101 IVT entries, 27 AVT entries, 20 entries form AB nursery, and 23 from BGM nursery) were evaluated for AB resistance under controlled environment conditions. Among the IVTs, IGLS 9, IGEB 6 and IGEB 7 were moderately resistant to AB (<5 rating on 1 - 9 rating scale). In the AVT group, AGK 4 (AVT 1 Kabuli) was found to be moderately resistant (3.1 to 5 rating). None of the entries in BGM and AB nurseries was found to be resistant to AB.

Suresh Pande and /Collaborators

Australia-ICRISAT collaborative research for AB resistance: In collaboration with the Center for Legumes in Mediterranean Areas? (CLIMA) and the Council of Grain Growers Organization (COGGO), Australia, 113 chickpea advanced breeding lines were evaluated for AB resistance following standardized screening technique using controlled environment facility at ICRISAT, Patancheru, India. Among the advanced breeding entries, ICGV 05558, ICGV 05562, ICGV 05563, ICGV 05564, ICGV 05565, and FLIP 94508C) were found to be resistant (<3 rating on a 1 - 9 rating scale), and 86 entries showed a moderately resistant reaction (3.1 to 5 rating). Segregating F_3 population involving 38 crosses (a total of 9225 plants) and F_4 generation involving 10 crosses (2430 plants) were

also evaluated for AB resistance under controlled environment conditions. A total of 3248 single plants from the F_3 generation and 427 plants from the F_4 generation were free from disease, and were advanced in the field. Single plants from eighth crosses (1066 plants from F_3 generation) of the above material, which were planted in the field, were also evaluated for resistance to AB. Data on severity of AB was recorded at 10 days after inoculation (DAI) on a 1 - 9 rating scale. No resistance (<3 rating) was observed in any of the plants tested. However, out of 1066 plants tested, 196 single-plants showed a moderately resistant reaction.

Suresh Pande, PM Gaur and KHM Siddique

Identification of new sources of resistance to BGM in advanced breeding and germplasm selections: Sixty advanced breeding lines, 124 entries from preliminary and advanced yield trials, 40 entries from international screening nurseries, 58 Ascochyta blight promising entries, seven BGM promising selections, 131 crossing block entries, 332 BGM promising lines (83 ICRISAT bred lines, 154 germplasm lines, and 95 lines from Australia) were evaluated for resistance to BGM under controlled environment conditions. Susceptible cultivar JG 62 was found to be highly susceptible, and had a rating of 9 on a 1 - 9 rating scale.

Only ICCV 97653 was found to be resistant (3 rating). Of the 60 advanced breeding lines, 36 were moderately resistant to BGM. Seventy entries from yield trials, 20 from international screening nurseries, 32 from Ascochyta blight selections, five from BGM selections, and 78 from the crossing block entries were found to be moderately resistant to BGM. Of the 332 promising selections, 139 lines (26 from breeding lines, 58 from germplasm lines, and 55 lines from Australia) were moderately resistant to BGM.

Suresh Pande and PM Gaur

ICAR-ICRISAT collaborative research on BGM resistance: Twenty-three entries received from IIPR, Kanpur, Uttar Pradesh, India, were evaluated for resistance to BGM using standardized protocols in controlled environment facility. NBG 4, NBG 8, NBG 9, NBG 10, and NBG 21 were found to be moderately resistant (DR 3.1 to 5 on a 1 - 9 rating scale) to BGM.

Suresh Pande and NARS Collaborators

Australia-ICRISAT collaborative research on BGM resistance: Sixty of the 113 advanced breeding entries, which were evaluated for AB, were also evaluated for BGM resistance under controlled conditions, of which 36 were found to be moderately resistant to BGM.

Suresh Pande, PM Gaur and KHM Siddique

Milestone: 10 - 15 Kabuli chickpea breeding lines with extra large seed (>50 g 100 seeds^{-1}) and high resistance to FW developed (PMG/SP/HDU) 2009

Identification of FW resistant extra-large seeded Kabuli chickpea germplasm: There are many sources with high levels of resistance to FW in Desi chickpea, while resistance in Kabuli types is limited. Desi x Kabuli crosses have been widely used at ICRISAT for enhancing FW resistance in Kabuli chickpeas. However, most Kabuli varieties that involved one or more Desi parents in the pedigree have a brown tinge in seed color, e.g., Swetha (ICCV 2), KAK 2 (ICCV 92311), JGK 1 (ICCV 92337), and Vihar (ICCV 95311), while the market prefers cream to white (zero tannin) seed color in Kabuli chickpea. Thus, it is important to identify additional sources of FW resistance in Kabuli chickpea, particularly in the large-seeded category, so that large-seeded Kabuli varieties with resistance to FW and typical Kabuli type seed (ram's head shape and white seed color) can be developed from Kabuli x Kabuli crosses.

We selected 50 large-seeded Kabuli chickpea germplasm lines from ICRISAT's gene bank and evaluated them for agronomic traits at ICRISAT-Patancheru during 2004/05 post-rainy season. From these, 12 accessions having seed size of more than 50 g 100-seeds^{-1} were selected for further evaluation. During the 2005/06 post-rainy season, one set of these genotypes was grown in wilt sick plot for screening against FW, and another set in wilt free area for evaluation of agronomic traits. Two accessions, ICC 14194 and ICC 17109, originating from Mexico, showed complete resistance (0% plant mortality) to FW, whereas other lines showed 11 to 100 % plant mortality (Table 1). The resistant control (WR 315) had no plant mortality, whereas the early-wilt susceptible check (JG 62) had 100%, and the late-wilt susceptible check (K 850) had 87% mortality. Both the resistant accessions had pinnate (fern) leaf, which is common leaf type in chickpea. ICC 14194 was very early (97 days), while ICC 17109 was medium

maturity (115 days). Two accessions (ICC 14194 and ICC 14198) were very early (days to maturity <100 days) and had 50 to 53 g 100 seeds^{-1}, suggesting that it is possible to breed early maturing Kabuli varieties with extra-large seed with FW resistance and typical Kabuli type seed.

Table 1. Morphological and agronomic characteristics of twelve extra-large Kabuli chickpea germplasm lines (ICRISAT, Patancheru, 2005/06 post-rainy season).

Accession	Origin	Leaf type	Days to flower[1]	Days to mature[1]	100-seed mass (g)[1]	Wilt reaction (%)[2]
ICC 7344	Mexico	Pinnate	38	100	50.2	95.2
ICC 8155	USA	Simple	45	112	62.2	100.0
ICC 11742	Chile	Pinnate	64	130	51.9	86.4
ICC 11883	Spain	Pinnate	56	130	58.7	90.9
ICC 13821	Ethiopia	Simple	50	118	51.0	92.0
ICC 14194	Mexico	Pinnate	38	97	52.9	0.0
ICC 14195	Mexico	Simple	50	109	60.2	52.2
ICC 14198	Mexico	Pinnate	42	94	50.2	70.8
ICC 14202	Mexico	Pinnate	46	118	58.1	75.0
ICC 15576	Mexico	Pinnate	52	120	55.6	81.0
ICC 16670	USA	Simple	45	110	50.1	11.1
ICC 17109	Mexico	Pinnate	46	115	63.2	0.0
WR 315 (Resistant check)	India	Pinnate	44	102	13.5	0.0
K 850 (Late wilting susceptible check)	India	Pinnate	56	109	28.9	87.0
JG 62 (Early wilting susceptible check)	India	Pinnate	42	103	15.8	100.0

[1]Data from crop grown in wilt-free field.
[2]Data on resistance to race 1 of *Fusarium oxysporum* f. sp *ciceris* from wilt screening nursery.

PM Gaur, S Pande and HD Upadhyaya

Development of extra-large seeded Kabuli chickpea breeding lines: A total of 88 Kabuli x Kabuli crosses were advanced by two generations (F_2 and F_3) for development of extra-large seeded (>50 g 100 seeds^{-1}) Kabuli breeding lines with resistance to FW. One of the parents involved in the crosses was a released cultivar (ICCV 2, KAK 2, JGK 1, or Vihar) with a seed size between 25 - 40 g 100 seeds^{-1} and the other parent was a extra-large seeded (>50 g 100 seeds^{-1}) Kabuli germplasm line, including the FW resistant lines ICC 14194 and ICC 17109. The F_2s were grown during the 2005/06 cropping season, and F_3s as a second crop during February to April 2006. The F_4 seed of eight of these crosses, which involved ICC 14194 and ICC 17109 as one of the parents, were supplied to four collaborating centers (Mahatma Phule Krishi Vidyapeeth, Rahuri; Indian Institute of Pulses Research, Kanpur; Indian Agricultural Research Institute, New Delhi; and Punjab Agricultural University, Ludhiana, India) for screening against FW races prevalent in those regions, and selection of single plants for development of new progenies.

PM Gaur, S Pande and CLL Gowda

Association of leaf type, seed size, and seed yield in Kabuli chickpeas: As several of the extra-large seeded Kabuli germplasm lines have simple (unifoliate) leaf, we conducted an experiment to study the relationship of pinnate (fern) and simple leaf traits with seed yield and seed size. Three crosses; ICCV 2 × ICC 14195, ICCV 2 × ICC 14215, and ICC 16644 × ICC 16670 were selected in which the parents differed for leaf type and seed size. ICCV 2 and ICC 16644 have pinnate leaf and medium seed size (23 - 25 g 100 seeds^{-1}), while ICC 14195, ICC 14215, and ICC 16670 have simple leaf and large seed size (50 - 59 g 100 seeds^{-1}). The F_2 populations from these crosses were grown during the post-rainy season 2005/06. There were 226 plants in ICCV 2 × ICC 14195, 247 plants in ICCV 2 × ICC 14215, and 244 plants in ICC 16644 × ICC 16670 cross. Observations were recorded on leaf type, number of pods per plant, number of seeds per plant, 100 seed weight, and seed yield per plant. In each

cross, the F_2 plants were classified into two groups based on leaf type (pinnate-leaved and simple-leaved) and then mean value of each trait was calculated for each group.

The pinnate-leaved plants gave significantly higher seed yield (53% in ICCV 2 x ICC 14215, 59% in ICCV 2 x ICC 14195, and 74% in ICC 16644 x ICC 16670) than the simple-leaved plants, mainly because of higher number of pods per plant. On an average, the pinnate-leaved plants produced 23 - 31 pods per plant, whereas simple-leaved plants produced 14 - 19 pods per plant. The increased number of pods per plant in pinnate-leaved plants resulted in increased number of seeds per plant and ultimately increased yield per plant. Seed size of pinnate-leaved plants and simple-leaved plants did not differ significantly in any of the crosses. The results clearly established negative effect of simple leaf traits on seed yield. Thus, it is recommended that selections should be practiced for pinnate-leaved plants in crosses involving simple-leaved and pinnate-leaved types.
PM Gaur/S Srinivasan

Milestone: 15 - 20 Desi and Kabuli chickpea breeding lines with combined resistances to FW, AB, and BGM developed (PMG/SP) 2010

Development of Desi chickpea breeding lines with combined resistance to FW, AB, and BGM: One hundred advanced breeding lines, selected primarily based on resistance to AB, BGM, and seed size were evaluated in two sets (Set A and Set B) of 50 each. In Set B, seed was not enough for a replicated trial. The 50 lines in Set-A were grown in a replicated trial along with 10 controls, which included the promising cultivars/breeding lines from Western Australia (Sona, Moti, Sonali, Rupali, WACPE 2078, WACPE 2098, WACPE 2099, and ICCV 96836) and India (JG 11 and ICCV 10). The trial was sown in a RCBD with 3 replications. Each plot consisted of 4 rows, 4 m long, and the plants were 30 cm apart. Central two rows were used for recording grain yield and plant biomass. The entries were screened against FW in a wilt-sick nursery, and for AB and BGM under controlled environment conditions. Nine breeding lines (ICCV 04512, ICCV 04513, ICCV 05527, ICCV 05528, ICCV 05529, ICCV 05530, ICCV 05531, ICCV 05532, and ICCV 05533) were resistant to FW (<10% plant mortality), AB (score between 3.0 - 4.0), and BGM (score between 4.0 - 5.0) (Table 2). Most breeding lines had a good seed size, but were late in maturity. These lines may perform well in long-season environments. Efforts are being made to improve these lines for earliness.

Table 2: Desi chickpea breeding lines with combined resistance to FW, AB, and BGM (ICRISAT, Patancheru, 2005/06 post-rainy season).

Entry	Days to maturity	100 seed wt. (g)	Seed yield (kg ha^{-1})	FW mortality (%) (2005)	AB score (mean of 2005 & 2006)	BGM score (mean of 2005 & 2006)
ICCV 05532	114	21.5	1732	6.8	3.50	4.3
ICCV 04513	121	19.1	1850	7.3	3.50	4.9
ICCV 05529	112	21.6	1914	3.6	3.58	4.2
ICCV 05531	112	21.2	2128	5.3	3.58	4.3
ICCV 04512	119	19.1	1829	7.8	3.58	4.9
ICCV 05530	114	22.1	1811	0.0	3.67	3.8
ICCV 05533	113	20.1	1863	0.0	3.75	4.7
ICCV 05528	114	22.4	1867	1.1	3.83	4.8
ICCV 05527	114	20.9	1768	5.9	3.83	4.7
Australian Checks						
Sonali	100	13.4	1906	90.3	4.67	7.2
Rupali	103	11.1	1336	67.5	4.58	8.2
WACPE 2078	103	15.9	1281	50.3	5.25	5.7
WACPE 2098	-	-	-	59.3	4.67	7.2
WACPE 2099	-	-	-	57.8	3.83	5.8
Moti	-	-	-	69.6	5.33	5.0
Sona	100	15.3	1232	84.0	7.00	5.4

Entry	Days to maturity	100 seed wt. (g)	Seed yield (kg ha^{-1})	FW mortality (%) (2005)	AB score (mean of 2005 & 2006)	BGM score (mean of 2005 & 2006)
ICCV 96836	110	13.2	942	55.3	5.00	5.8
Indian Checks						
ICCV 10	108	15.4	1963	15.5	7.58	6.0
JG 11	100	21.5	2756	5.2	5.50	5.2
SE ±	1.5	0.44	157.5			
CV (%)	2.4	3.83	16.9			

New breeding materials are being generated to develop breeding lines with resistance to multiple diseases. During the post-rainy season 2005/06, 60 F_2, 28 F_3, and 10 F_4 populations were grown. The F_3 and F_4 populations were screened for AB resistance under controlled conditions and the resistant plants were transplanted in the field. The resistant plants from F_4 populations of 12 crosses and F_3 populations of 33 crosses were screened for AB resistance and the resistant plants were transplanted in the glasshouse. Over 1,000 single plants were harvested and examined for seed size, shape, and color. A total of 110 plants were selected for further evaluation and also shared with Department of Agriculture and Food, Western Australia and Punjab Agricultural University, Ludhiana, India. A total of 37 new crosses were made during 2006 for development of breeding lines with resistance to multiple diseases. This included 28 intercrosses among the 10 selected F_4/F_5 progenies and nine single crosses. The single crosses included JG 11, Sona, Sonali, WACPE 2078, and WACPE 2099 as one parent, and ICCV 04512 or ICCV 05529 as the other parents.
PM Gaur/S Pande/CLL Gowda

Milestone: About 100 advanced germplasm and advanced breeding lines (Desi and Kabuli) screened for FW resistance (SP/PMG/HDU) 2009

Evaluation of advanced breeding and germplasm selections for resistance to FW: Eighty nine advanced breeding and germplasm lines were evaluated for resistance to wilt and root rots in a multiple-disease-sick-plot (MDSP) during the 2005/06 cropping season under artificial epiphytotic conditions. Susceptible early wilting cultivar ICC 4951 had 100% incidence within 30 DAS and the late wilting cultivar ICC 5003 showed 100% incidence within 90 DAS. Of the 89 advanced germplasm and breeding lines, KAK 2, ICCX 950106-F_4-40P-BP, ICCX 950106-F_4-43P-BP, ARFG 8, NNW 7, NNW 8, ICCX-970047-BP-BP-P27-BP, and ICCX 980068-F_4-P10-BP were asymptomatic (0%), while 80 lines showed a resistant reaction (<10% wilt incidence). Seven entries, KAK 2, NNW 19, ICCX 950151-F_4-50P-BP-BP, ICCX 950151-F_4-58P-BP-BP, ICCX 950151-F_4-31P-BP-BP, ICCX 950151-F_4-32P-BP-BP, and ICCX 950151-F_4-61P-BP-BP were free from collar rot and 69 entries had <10% collar rot incidence. PG 9421-1 was free from DRR, while 72 entries showed a resistant reaction (<10% incidence) to DRR. Resistance to collar rot and DRR will be confirmed under controlled environment conditions.

Suresh Pande and PM Gaur

ICAR-ICRISAT collaborative research on FW resistance in chickpea: In collaboration with scientists from Indian Institute of Pulses Resarch (IIPR), Kanpur, India 101 entries from IVT trials (26 entries from IVT-Desi, 28 from IVT-late sown, 20 from IVT-Kabuli, 20 from IVT rainfed, and 7 from IVT-extra bold), 48 from AVT trials (3 from AVT 1-Desi, 7 from AVT 1-late sown, 9 from AVT 1-Kabuli, 21 AVT 1- bold seed, 2 from AVT 1-rainfed, and 6 from AVT 1-high input), 10 donor lines, 9 chickpea wilt differential lines, and 46 from the national were evaluated for FW resistance under field conditions, and AB and BGM under controlled environment conditions. Of the 101 IVT entries, 23 were free from FW, while 30 were resistant (<10% incidence). Among the AVT entries, four (AIGB 15, AIGB 17, AIGB 19, and AIGB 21) were asymptomatic and six (AIGB 4, AIGB 5, AIGB 6, AIGB 12, AIGB 18, and AIGB 20 (showed a resistant reaction (<10% incidence). Of the 10 donors, D8 was resistant to FW. Among the wilt differential lines, D 05 was free from FW. NNW 5 and NNW 36 were asymptomatic and 10 lines were resistant.

Suresh Pande and NARS Collaborators

UAS Dharwad-ICRISAT collaborative research on wilt resistance in chickpea: Ninety-six single plant progenies (SPP) of the cross F_3 Bheema x ICCV 10, 106 SPPs of the cross F_3 M_2 Bheema x ICCV 10, 113 SPP of

M_3 Bhima, five susceptible populations of the cross Bheema x ICCV 10, five resistant populations of Bheema x ICCV 10, five heterozygous populations of the cross Bheema x ICCV 10, and both the parents (Bheema and ICCV 10) were evaluated for wilt resistance under controlled environment conditions in the greenhouse. Susceptible cultivar JG 62 was included for comparison. High levels of resistance to wilt were observed in F_3 and M_3 populations, while no resistance was observed in susceptible, resistant, and heterozygous populations of the cross Bhima x ICCV 10.

Suresh Pande and PM Salimath

Milestone: Identification, characterization and resistance screening of potentially important diseases (DRR, BRR, CR) of chickpea (SP) 2008

Standardization of screening technique for collar rot (CR) resistance: Pure culture of collar rot fungus, Sclerotium rolfsii, was multiplied on three media; sterilized groundnut shells, sterilized sorghum straw (2 - 3 cm long pieces), and potato dextrose broth, and incubated at 25^0C for 20 days. Sclerotial production was estimated from 100 g ml^{-1} from each medium. About 10,245 sclerotia from groundnut shells, 5,074 from sorghum straw, and 1,853 from potato dextrose broth were recorded. Since the sclerotia are the main disease producing bodies in CR, groundnut shell medium was selected for further standardization of the screening technique. Thirty grams of inoculum (multiplied on groundnut shells) was mixed with 1 kg sterilized top 15 cm soil and filled in metal trays (70 x 30 x 16 cm). Light irrigation was given to make the soil moist and was left undisturbed for three days for the establishment of the fungus. Susceptible chickpea cultivar Annigeri was planted in rows in the trays and observed for 21 days for seed and seedling (collar) rot symptoms. About 21% of seed rotting and 79% collar rot was recorded at 21 days after sowing. Since the seed and seedling rots were consistent in three screenings in Annigeri, these trays were used for routine evaluation for collar rot resistance.

Evaluation of wilt promising selections for resistance to CR: Eighty-nine wilt promising advanced breeding and germplasm selections were evaluated for CR resistance following standardized sick tray technique under controlled environment conditions. None of the lines tested was found to be resistant CR. Therefore, we initiated systematic evaluation of germplasm accessions for CR resistance.

Suresh Pande

Milestone: Basic and strategic research on succession of Fusarium wilt and root rots of chickpea (SP) 2009

Threshold and quantification of fungal pathogens of chickpea in wilt and multiple disease sick plots: Quantification of fungal pathogens of chickpea wilt complex was carried out for the third year in the wilt sick plot (BIL 3 C), and the multiple disease sick plot [(MDSP) (BIL 1)] at ICRISAT, Patancheru, India. Collection of soil samples before planting and after harvesting of the crop and processing of soil was done as in the previous year. Number of fungal colonies g^{-1} soil was estimated using synthetic media (PDA and CDA) at 48 to 96 h after incubation. Number of sclerotia of *S. rolfsii* was quantified by using the rapid floatation technique.

Unlike previous years, the number of colonies of all the wilt complex pathogens was greater in the samples taken before sowing than after harvesting of the crop in both the fields. *Fusarium oxysporum* f.sp. *ciceris* (FOC) colonies were around 1,700 g^{-1} soil before planting and up to 4,150 g^{-1} soil after harvest of the crop in both fields. FOC colonies were recovered up to 80 cm depth before planting and up to 100 cm depth after harvest. In the wilt sick plot, in addition to FOC, other root rot causing pathogens (*Fusarium solani, Rhizoctinia bataticola,* and *Sclerotium rolfsii*) were negligible. This observation was in agreement with that of previous season.

Additionally, soil collected from MDSP was also assessed for root rot pathogens, *Fusarium solani* (black root rot), *Rhizoctonia bataticola* (dry root rot), and *Sclerotium rolfsii* (collar rot). About 475 colonies of *F. solani* and 220 colonies of *R. bataticola* g^{-1} soil, and four sclerotia of *S. rolfsii* 10 g^{-1} of soil were recovered from surface soil, which was collected before sowing of chickpea crop. These root rot pathogens multiplied during the cropping period and their population was almost double after crop harvest. Further, *F. solani* and *R. bataticola* were recovered up to 50 cm depth before planting and up to 65 cm depth after harvesting of the crop, while *S. rolfsii* was recovered up to 20 cm depth before sowing and 25 cm after harvest of the crop. Number of colonies of all these pathogens decreased as the depth increased. Lower number of propagules of the pathogens before sowing of the crop (October) may be

due to absence of chickpea crop for about eight months in these fields. The phenomenal increase of the propagules at crop harvest (February) suggested that chickpea supports the multiplication of these pathogens in the soil.

Suresh Pande and Mamta Sharma

Succession of fungal pathogens of chickpea in wilt and multiple disease sick plots: Succession of occurrence of fungal pathogens in chickpea wilt and multiple diseases sick plots was studied during the season to confirm the results obtained in the previous year. Methodology including check cultivars (early wilting JG 62, late wilting L 550, and FW resistant WR 315), sampling and isolations on potato dextrose agar and czepek dox agar media were carried out as in the previous year. Isolations were made from root tip, root hair, epidermis and cortex, vascular bundles and collar region of apparently healthy looking plants at 10-day intervals from 10 DAS in both fields.

All the plants of cultivar JG 62 wilted within 30 DAS and that of L 550 in 80 DAS, while those of WR 315 remained healthy till maturity. FOC was found in all the root parts of healthy looking early wilting and late wilting cultivars from 20 DAS till the death of the plants. FOC was found only in root tip and root hairs in the resistant cultivar WR 315 at 50 DAS till maturity. This late infection and restriction of the fungus at the root tips in this cultivar may be due to its resistance to wilt pathogen. Black root rot pathogen, *Fusarium solani* and collar rot pathogen, *Sclerotium rolfsii* attacked and killed the plants in all the three cultivars at the seedling stage (up to 30 DAS) when the soil moisture was high. Warm temperature and soil moisture stress encouraged dry root rot fungus, *Rhizoctonia bataticola* (RB) to cause rotting of the roots. During the season, dry root rot appeared from 50 DAS till maturity in both late wilting and resistant cultivars. In both these cultivars, a mixture of FOC and RB was observed from 50 DAS till maturity indicating that interaction of both these pathogens may be responsible for death of the plants.

The FOC was dominant in the wilt sick plot throughout the cropping season, and recovered from all the root parts of apparently looking healthy plants of both JG 62 and L 550 from 20 DAS, and were present till the death of the plants. Susceptible cultivar JG 62 wilted in 30 DAS and L 550 in 80 DAS as in MDSP. Resistant cultivar, WR 315 yielded FOC only from root tips and root hairs from 60 DAS as in multiple disease sick plot. Additionally, a mixture of FOC and RB was also recovered from root tips of late wilting and resistant cultivars from 60 DAS till maturity. These results indicated that the death of the plants may be due to interaction of both these pathogens. Though wilt fungus is predominant in wilt sick plot, low intensities of *F. solani* and *S. rolfsii* at the seedling stage (up to 30 DAS), and *R. bataticola* during later stages were recorded in both L 550 and WR 315.

Suresh Pande and Mamta Sharma

Activity 6A.1.2: Develop chickpea breeding lines with, resistance to *Helicoverpa.*

Milestone: 5 - 10 resistance sources and advanced breeding lines tested for stability of resistance (HCS/CLLG/PMG) 2009

Breeding chickpeas for resistance to pod borer, *Helicoverpa armigera*: We evaluated 1,586 segregating progenies of chickpea during the 2005/06 season, and selected 1,161 progenies for further testing. The selection of progenies was based on visual scoring for pod damage and yield. Subsequently, progenies having <9% pod borer damage and high yield than the check varieties were selected for evaluation in the 2006/07 season. Eight hundred and fifty-five progenies (342 F_8 progenies and 315 F_9 progenies from single-crosses, and 198 F_8 progenies from four-way crosses) were sown to select lines for resistance to *Helicoverpa armigera* under natural infestation. Apart from this, we have sown F_2 diallel (71 entries, F_1's with parents having 64 entries), crossing block (54 entries) to develop new lines for resistance to this pest.

CLL Gowda, HC Sharma and P M Gaur

International Helicoverpa Screening Nursery: Using reliable field screening techniques developed at ICRISAT, several lines with resistance to *H. armigera* have been identified. The resistant (less susceptible) sources identified in field screening were used in crosses to transfer resistance into high-yielding varieties. A new set of lines was assembled for the international chickpea screening nursery *for H. armigera* (ICSN-HR) in 2006, including a few lines found to be less susceptible to *H. armigera.* Most of the lines are of short- to medium-duration, adapted to environments similar to southern and central India (16 to 22°N latitudes). The objective is to evaluate promising *Helicoverpa*-resistant selections in varying environments and to provide an opportunity to NARS partners for

selection of material for use as parents or as end products suitable for local conditions. The trial with 25 chickpea genotypes, including two checks, was sent to six collaborators in India, and one to Myanmar.

CLL Gowda and HC Sharma

Stability of resistance to *Helicoverpa armigera* in chickpea: Twenty-five chickpea lines belonging to short- and medium-duration group (international *Helicoverpa* resistance screening nursery) were evaluated for resistance to *H. armigera* at different locations in India. There were three replications in a randomized complete block design. Data were recorded on leaf and pod damage rating and overall resistance score (1 = highly resistant, and 9 = highly susceptible), percentage pod damage, and grain yield. In the short-duration nursery, the lines RIL 115, ICC 14402, ICC 14559, ICCL 79037, and ICCX 73008 during the flowering stage, and RIL 115 and ICCL 79037 during the podding stage recorded less oviposition. Larval numbers were <8.3 in nine lines as compared to 54.0 larvae on the susceptible check, ICC 3137. The lines RIL 83, RIL 115, RIL 126, ICC 14402, ICL 86111, ICCX 73008, ICC 5383, and ICC 506 recorded an overall resistance score of <1.8. Grain yield of RIL 27, RIL 83, RIL 1, RIL 180, ICC 14559, ICCL 87322, and ICCC 37 was significantly greater than the susceptible or commercial checks in 4 or 5 locations. Based on insect damage parameters and grain yield, the lines RIL 27, RIL 81, RIL 83, and ICC 10613 may be used in chickpea improvement for resistance to *H. armigera*.

In the medium-duration nursery, low oviposition was recorded in RIL 51, RIL 85, RIL 169, ICCL 87314, ICCL 87315, ICC 67, Annigeri, and ICCV 10 in two or more locations, while the lines RIL 85, ICC 14559, ICC 162612, ICCL 87315, ICCL 87315, ICC 67, and Annigeri recorded low larval density in three or more locations. Low pod damage was recorded in RIL 51, ICC 14559, ICCL 87314, ICCL 87322, C 235, Annigeri, ICCV 10, and ICC 506. Grain yield was high in case of RIL 51, ICCL 87314, ICCL 87315, ICCL 87322, ICCV 10, and ICCC 37 in four or more locations. Principal component analysis placed the test genotypes into four groups, suggesting that there was a considerable diversity in chickpea genotypes to damage by *H. armigera*. Based on insect damage and grain yield, the lines RIL 51, ICCL 87315, ICCL 87322, ICCL 87315, and ICCV 10 may be used in chickpea improvement for resistance to *H. armigera.*

HC Sharma, CLL Gowda, PM Gaur and NARS partners

Milestone: Physico-chemical mechanisms of resistance to Helicoverpa identified and nature of inheritance studied (CLLG/HCS) 2012

Evaluation of protocols to screen chickpeas for resistance to pod borer, *Helicoverpa armigera*: A collaborative trial was conducted to evaluate the efficiency of screening for resistance to *H. armigera* using detached leaf assay (at the flowering stage), comparing pod damage and grain yield under protected and unprotected conditions under natural infestation, and caging the test entries with 20 pairs of adults at the flowering stage. Each entry was sown in 2 rows, 2 m long, and there were three replications in a RCBD. The crop was protected at the flowering and podding stages with methomyl (0.02%) in the protected plots. The genotypes ICC 10613, ICC 506 EB, and ICC 86111 showed antibiosis under lab conditions (detached leaf assay). DCP 92-3 and ICC 506EB showed antixenosis for oviposition under field conditions. The lines showing slow larval growth in the detached leaf assay also suffered low pod damage under natural infestation in the field (except PDE 2, which showed moderate antibiosis in the detached leaf assay). The genotypes ICC 506EB, ICCL 90034, JG 130, and PDE 2 yielded higher (1175 – 1307 kg ha^{-1}) than ICC 10613 (779 kg ha^{-1}) under protected conditions, of which ICC 506EB, ICC 90034, JG 130, and PDE 2 also performed well under unprotected conditions. Infestation of the test genotypes with adults in the cage resulted in poor infestation. Most of the eggs were laid on the nylon cloth. The results indicated that detached leaf assay provides a fairly good indication of expression of resistance to *H. armigera* in chickpea genotypes under field conditions.

Effect of egg age, storage duration, and temperature on egg hatch and incubation period of *H. armigera*: *Helicoverpa armigera* is used routinely as a tool for research on insect host plant interactions, biological, physiological, behavioral, and toxicity studies. Hence, manipulation of its life cycle can be used as a tool to have adequate numbers of insects at the appropriate stage of development for experimental purposes. Therefore, studies were undertaken to determine the effect of storage temperature on the duration and viability of eggs. Percentage egg hatch and incubation period were significantly influenced by egg age, storage temperature, and storage duration. Average egg hatch ranged from 0.0 to 96.8% across temperatures (-20 to 35^0C) and storage durations. None of the eggs hatched at -20 and 0ºC. Day degrees required for egg hatching decreased with an increase in temperature from 10 to 27ºC, and egg age from 0 to 3 days. The day degree requirements were highest for 0 day-old eggs at 10ºC, and

lowest at 27ºC. It is safer to store *H. armigera* eggs at 10ºC for 10 days, with a hatchability of >75.0%, and an incubation period of <2 days.

HC Sharma and MK Dhillon

Physico-chemical mechanisms of resistance in chickpea to *Helicoverpa armigera*: Fifty morphologically diverse lines of chickpea were evaluated for resistance to the pod borer, *H. armigera* under field conditions. The material was sown in two sets of early- (30 lines), and medium-late-maturity (20 lines) along with appropriate resistance and susceptible controls. There were three replications for each trial in a randomized complete block design. Data were recorded on leaf damage rating at the vegetative stage, egg and larval numbers during the vegetative and flowering stages, overall resistance score (including recovery resistance), pod damage, and grain yield. In the early-maturity group, six lines showed a over all resistance score (1 = highly resistant, and 9 = highly susceptible) of <3.0 compared to 7.5 in JGM, 3.2 in ICCV 10, 3.0 in Vijay, and 5.0 in Vishwas. Of these, ICC 12475, ICC 4934, ICC 5434, and JGM 2 also had lower larval numbers during the vegetative, flowering, and podding stages, and showed an overall resistance rating of <3.0 compared to 6.0 in ICC 3137 and 1.0 in ICC 506. In the medium maturity group, the overall resistance scores ranged from 1.2 to 6.0, and the lines ICC 506, ICC 4934, ICC 5783, and JGM 4 showed resistance to *H. armigera* (damage score <2 compared to 6.0 in ICC 5002). Data have also been collected on the morphological traits, and the grain has been analyzed biochemically for protease inhibitor activity, protein profiles, nutritional composition, and acid exudates in leaves to identify physico-chemical factors associated with resistance to *H. armigera* in chickpea.

HC Sharma

Output target 6A.2: Molecular markers for AB and BGM resistance validated, and for Helicoverpa resistance identified in chickpea

Activity 6A.2.1: Mapping and marker-assisted breeding for diseases and insect resistance in chickpea

Milestone: One intra-specific RIL population for mapping AB resistance QTLs developed using contrasting parents (PMG/SP/RV) 2008

One intraspecific RIL mapping population is being developed from a cross between ICCV 04516 (resistant) and Pb 7 (susceptible). ICCV 04516 has a AB disease score of 3 - 4 (1 = immune, and 9 = highly susceptible), while Pb 7 is highly susceptible (diseases score of 9). About 300 F_2s of ICCV 04516 x Pb 7 were grown during the post-rainy season 2005/06. The F_3 progenies were grown during the off-season in greenhouse using single seed descent (SSD) method. Several plants died due to root rot and it was discovered that the pathogen-infected soil used in greenhouse was the cause of this disease. Hence, the population was discarded and the F_3 were grown from the remnant seeds.

PM Gaur and S Pande

Progress reported towards the achievement of milestone for 2008 will contribute towards achievement of the milestones listed below.

Milestone: The reported markers for AB and BGM resistance QTLs validated in other populations (PMG/SP/RKV) 2009

The work for this milestone will start after the development of mapping populations in 2008.

Milestone: QTLs for AB and BGM resistance introgressed in 3 - 4 farmer-preferred and locally adapted cultivars (PMG/SP/RKV) 2011

The work towards introgression of AB and BGM resistance QTLs will start after validation of these QTLs in 2009.

Milestone: Intra-specific and interspecific (C. arietinum x C. reticulatum) RIL populations for mapping Helicoverpa resistance developed and phenotyped (PMG/HCS) 2009

Evaluation of mapping population (ICC 506 x Vijay) for resistance to *Helicoverpa armigera*: The mapping population ICC 506 x Vijay (200 lines) was evaluated for resistance to *H. armigera* under natural infestation in the field. There were three replications in a randomized complete block design. Observations were recorded on leaf

damage, numbers of eggs laid, larval density, number of pods, pods damaged, and grain yield. The overall resistance score (1 = highly resistant, and 9 = highly susceptible) ranged from 1.5 to 6.0 in the mapping population, 3.0 in Vijay, 1.3 in ICC 506, and 3.0 in ICCC 37. Percentage pod damage ranged from 5.2 to 29.8% in the mapping population, 10.5% in Vijay, 7.0% in ICC 506, and 17.4% in ICCC 37. The numbers of larvae at the reproductive stage were 1.3 to 13.3 in the mapping population, 6.7 in Vijay, 1.3 in ICC 506, and 7.3 in ICCC 37, indicating considerable variation in the susceptibility of the population and the parents to *H. armigera*. This population needs to be genotyped to identify molecular markers for resistance to *H. armigera.*

HC Sharma and PM Gaur

Milestone: One inter-specific (C. arietinum x C. reticulatum) RIL populations for mapping Helicoverpa resistance QTLs developed (PMG/HCS) 2009

RILs are being developed from an interspecific cross between ICC 3137 (*C. arietinum* - susceptible) and IG 72953 (*C. reticulatum* – resistant). The population was advanced by two generations (F_2 and F_3) in the greenhouse during 2006. Some plants died due to root rot disease as the soil used in the greenhouse was infected with the disease. New crosses were made between these parents and the F_1s were grown during the off-season in the greenhouse. The F_2 population from the new cross and F_4 progenies from earlier cross are being grown during the 2006/07 cropping season.

PM Gaur and HC Sharma

Milestone: QTLs for Helicoverpa resistance identified from C. arietinum x C. reticulatum RIL population (HCS/RKV/PMG) 2010

Newly developed SSR markers at ICRISAT are being screened on the parental genotypes of the mapping population to identify the polymorphic markers and integrate these into the genetic map.

RK Varshney

Milestone: Characterization of pathotypes of Fusarium oxysporum f. sp. ciceris, Ascochyta rabiei, and Botrytis cenerea in chickpea and Fusarium udum in pigeonpea (SP) 2009

Characterization of races of *Fusarium oxysporum* f.sp. *ciceris*: Chickpea lines identified to be resistant to wilt at one location often show a susceptible reaction at other locations mainly due to the existence of different pathotypes/races of the pathogen, *F. oxysporum* f. sp. *ciceris* (FOC). Our recent research on races of FOC indicated that the earlier (early 1980s) race scenario has changed. Hence, we initiated studies to collect races of FOC from major chickpea growing areas in India. A total of 38 virulent single spore isolates collected from 18 locations in 12 states in India were used for morphological, cultural, and pathogenic characterization of FOC. Of the 38 isolates, seven were collected from ICRISAT, Patancheru, two from Kurnool (Andhra Pradesh), four from Hisar (Haryana), one each from Dholi (Bihar), Delhi, Dhaulakuan (Himachal Pradesh), and Gulbarga (Karnataka), two from Junagadh (Gujarat), one from Sehore, four from Jabalpur (Madhya Pradesh), one from Akola, two from Badnapur, two from Rahuri (Maharashtra), two from Ludhiana, one from Gurdaspur (Punjab), three from Kanpur, one from Ghaziabad (Uttar Pradesh), and two from Pantnagar (Uttaranchal) (all in India). Four races, which were reported during 1982 from India were also included in this study.

Morphological variation: Five mm discs of actively growing five-day old culture of each isolate were transferred separately onto potato dextrose agar (PDA) medium in 90 mm plastic plates and incubated at 25^0 C for 7 days. Data on colony color, colony type, zonation, and colony growth (total growth and growth per day) were recorded at 7days after incubation. Based on type of mycelium, color, and colony growth; all the isolates were categorized into nine groups: FOCs 4, 8, 10, 16, 27, 32, 37, 38, and R4. Most of the isolates had cottony type of mycelium with erumpent growth. Isolate FOC 8 had submerged type of mycelium, FOC 16 showed effused growth, and isolate R4 had a pinkish mycelium.

Cultural variation: The nine isolates, which differed in apparent morphological characters, were used to study cultural characters. All the nine isolates were multiplied separately on potato dextrose broth medium and incubated at 25^0 C for 7 days. Data on percent micro and macro conidia, conidial size, and number of cells in macro conidium were recorded at 7 days after incubation. Considerable variation existed among the isolates in all the parameters

studied. Percentage of micro-conidia was significantly more than macro-conidia in all isolates. Highest percentage of micro conidia (81.4%) in FOC 4 and lowest (51.6) in R4 was recorded. Largest micro-conidia (10.1x 5.5 µ m) were observed in FOC 38, and smallest (7.7 x 4.8 µ m) in FOC 8. Macro-conidia were larger (22.1 x 5.8 µ m) in the isolate R4, while they were smaller (14.9 x 5.5 µ m) in FOC 32. Mean number of cells in macro-conidia were 3.1 in FOC 10 and 4.6 in FOC 41.

Characterization of variability of *Ascochyta rabiei* : Differential reaction of cultivars to AB at different locations in India indicated the probable existence of races/pathotypes in the pathogen, *A. rabiei.* Hence, we initiated a systematic research to characterize races/pathotypes in *A. rabiei.* The *A. rabiei* infected chickpea plants were collected from 14 locations in different agroclimatic regions in India. Isolations were made from these infected plants and pure cultures of single spore isolates of *A. rabie* were obtained.

Morphological and cultural variation: Seven single spore isolates representing different agroclimatic zones were selected for studying morphological, cultural, and pathogenic characters. Five mm diameter discs from actively growing cultures of *A. rabiei* were placed at the center of 90 mm diameter petri plates containing CDA, and inoculated at 20^0 C. Colony color, intensity of the mycelium, colony diameter, number of conidiomata, and conidia were recorded. The *A. rabiei* isolates differed in morphology, colony color, colony size, pycnidial color (brown to slate grey), number of conidioamata (42.3 to 90.7 cm-^{2}), number of conidia (0.55 to 3.01 conidia 10^3 cm-^{2}), and conidial size (10.7 x 4.6 to 14 x 6.2 µm).

Pathogenic variation: All the 14 isolates were used for inoculation on 180 germplasm and advanced breeding lines under controlled environment conditions. All the 14 isolates of *A. rabiei* differed in their virulence pattern against 180 lines with AB 13 having maximum virulence index of 7.9 and AB 6 with a minimum index of 5.4 (Fig. 1). None of the lines were asymptomatic to any of the test isolates. Of the 180 lines, 10 lines to AB 6 and seven lines to AB 27 showed a resistant reaction. None of the entries were resistant to five isolates (AB 4, AB 17, AB 26, AB 1, and AB 13), whereas only one entry showed resistant reaction to four isolates (AB 8, AB 15, AB 3, and AB 18). Of the 180 entries, 15 entries (ICC 12, ICC 607, ICC 2165, ICC 3918, ICC 4200, ICC 4475, ICC 5124, ICC 6306, ICC 7002, ICC 13754, ICC 14911, ICCX 810800, ICCX 910028-39ABR-BP-10, ABR-BR, ILC 3870, and FLIP 82-258) differed significantly in their reaction to different isolates of *A. rabiei,* and hence were selected as differentials.

Characterization of variability of *Botrytis cinerea*: Thirty-two isolates of *B. cinerea* infecting chickpea, lentil, and marigold were collected from different locations to determine the variability of the fungus. Of the 32 isolates, 23 were collected from chickpea, 5 from lentil, 2 from marigold, 1 from Dahlia and 1 from grasspea. All the isolates were purified using BGM specific medium containing tannic acid. Preliminary analysis of eight Indian isolates using 20 RAPD primers (decamers) categorized them into two distinct groups. Detailed molecular analysis will be followed during 2007-08 season.

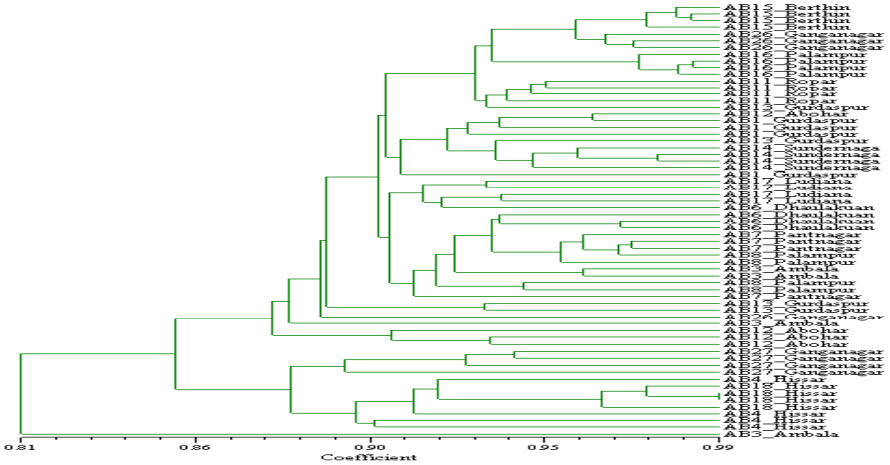

Fig. 1. Dendrogram showing the clustering of the single spore isolates of *A. rabiei* based on AFLP analysis;

Characterization of variability in *Fusarium udum* : We also initiated work on characterization of variability in *F. udum*. Eleven isolates of *F. udum* from nine locations in six states [Akola, Badnapur (Maharashtra); Bangalore, Gulbarga (Karnataka); Khargone (Madhya Pardesh), Muradnagar (Meerut), Varanasi (UP), Warangal and ICRISAT-Patancheru (Andhra Pradesh) all in India] were collected and single spore cultures were obtained using standard mycological techniques. All the single spore isolates once again were passed through common wilt susceptible cultivar ICP 2376 by following root dip inoculation technique, and aggressive isolates were stored at 4^0 C in the laboratory. Since the collection of the isolates did not represent the entire country, we continued to collect the isolates from pigeonpea growing areas. During the current crop season, wilt infected samples were collected from 9 locations (Yenagandla, Kulcharam, Jogipet, Andole, and Shivampet in Andhra Pradesh; Aurad in Karnataka; and Dhuki, Osmanabad, and Parbhani in Maharashtra, India). Isolations were made and *F. udum* was obtained from all the locations. All the cultures were purified and single spore isolates were obtained following standard mycological techniques. All these isolates were tested for virulence using a common susceptible cultivar ICP 2376 following root dip technique. All the virulent cultures were stored at 4^0 C in the laboratory.

Suresh Pande and Mamta Sharma

Milestone: Mapping of one intra-specific RIL population for dry root resistance conduced (PMG/SP) 2008

Mapping of RILs populations to DRR resistance under controlled environment: One hundred and twenty-six RILs and parents (JG 62 and ICCV 2) were evaluated for DRR resistance using paper towel technique under controlled environment conditions. Each entry was replicated thrice. Of the 126 RILs evaluated, ICCX 930111- 57-1-1-1-1SP-1-1-1-BP-1BP showed a resistant reaction (3 rating on 1 - 9 scale), while 15 entries were moderately resistant (3.1 to 5 rating). Both parents showed a susceptible reaction.
Suresh Pande/PM Gaur

Activity 6A.3.1: Advanced generation inter-specific derivatives with resistance to *Helicoverpa* and BGM generated using wild *Cicer* from different gene pools

Milestone: Strategies to cross wild Cicer between primary, secondary and tertiary gene pools developed (NM) 2008

In collaboration with Washington State University, Pullman, USA crossing program was initiated to cross perennial *Cicer* with annual as well as cultivated *Cicer*. Pollen germination was observed between intra-perennial *Cicer* species. It is possible to have successful pollination and fertilization between annual species *C. reticulatum* and perennial species *C. oxyodon* and *C nurstichacum,* as well as between cultivated chickpea and *C. oxyodon* and *C. nursticum*, but mature seeds were not observed. Since perennial *Cicer* does not set flowers at ICRISAT Patancheru, techniques were developed to preserve pollen of perennial *Cicer* for use at ICRISAT at a later date. Germination medium for annual and perennial *Cicer* pollen has also been developed.

Nalini Mallikarjuna and WSU partners

Milestone: Fifteen stable inter-specific derivatives using resistant wild Cicer from secondary gene pool generated and screened for Helicoverpa, AB, BGM, and good agronomic characters under field conditions (NM/HCS/SP/PMG) 2010

Cicer accessions ICC 17159, IG 73074, IG 72937, IG 72933, and IG 72934, which have shown resistance to Botrytis grey mold were used in the crossing program. Seed set varied depending upon the pollen parent used in the crossing program. The F_2 progenies from the crosses involving wild Cicer species IG 73074 and ICC 17159 are listed in Table 3. Chi square analysis of the progenies showed that the ratio of resistant to susceptible plants was 1: 3, and hence BGM resistance introgressed from wild Cicer was monogenic and recessive.

Table 3. Segregation of F_2 population for Botrytis gray mold resistance in interspecific derivatives of Cicer (ICRISAT, Patancheru, 2005/06 post-rainy season)

Cross	Resistant	Susceptible	Ratio	χ^2	P
ICC 4951 x IG 73074	3	30	1:3	4.45	(0.05-0.01)
ICC 4954 x ICC 17159	1	38	1:3	10.5	(0.01-0.001)
ICC 4954 x IG 73074	12	51	1:3	1.19	(0.3-0.2)
ICC 10136 x ICC 17159	4	10	1:3	0.09	(0.8-0.7)
ICC 10136 x IG 73074	7	23	1:3	0.04	(0.9-0.8)

Nalini Mallikarjuna and Suresh Pande

Progress reported towards the achievement of milestone for 2010 will contribute towards achievement of the milestones listed below.

Milestone: Ten advanced generation interspecific derivatives screened for Helicoverpa, AB and BGM in target locations in India (NM/HCS/SP/PMG) 2012

Milestone: Wild relatives with diverse mechanisms of resistance to Helicoverpa, AB, and BGM identified (HCS/SP/NM) 2009

Evaluation of wild relatives of chickpea for resistance to *Helicoverpa armigera:* We evaluated seven accessions of wild relatives of chickpea, along with resistant (ICC 506) susceptible (ICCC 37, ICC 3137, and L 550) lines of the cultivated chickpea for resistance to pod borer, *H. armigera* using detached leaf assay. Ten neonate larvae of *H. armigera* were released on fresh terminal branches of the test genotypes embedded in 3% agar-agar in 250 ml plastic cups. There were five replications in a completely randomized design. Observations were recorded on weights of the larvae on 10th day after initiating the experiment. The larvae weighed 41.3 to 61.6 mg per larva when reared on IG 70034 (*Cicer judaicum*), ICC 17148 (*C. microphyllum*), IG 70006 (*C. bijugum*), and ICC 69979 (*C. cuneatum*) compared to 74.2 to 79.6 mg on *C. reticulatum* (IG 72933 and IG 72953), 141.5 mg on Annigeri, 188.6 mg on ICC 506, 225.2 mg on L 550, and 222.3 mg on ICC 3137. The results suggested high levels of antibiosis to *H. armigera* in the wild relatives of chickpea belonging to the tertiary gene pool (*C. judaicum*, *C. bijugum*, *C. cuneatum*, and *C. microphyllum*), and moderate levels of resistance in the secondary gene pool (*C. reticulatum*), and low levels in the cultigen.

In another experiment, 12 accessions of wild relatives were tested along with resistant (ICC 506) and susceptible (ICC 3137) checks under greenhouse and field conditions. The terminal branches were infested with 10 neonate larvae of *H. armigera* using detached leaf assy. At five days after initiating the experiment, the larval weights were lower on ICC 69979, IG 70032, IG 70034, and ICC 17248 (<3.7 mg per larva under greenhouse conditions, and <4.98 mg under field conditions) as compared to the resistant and the susceptible checks (6.72 and 5.91 mg in ICC 506, and 12.89 and 13.84 mg in ICC 3137 under greenhouse and field conditions, respectively).

HC Sharma, SL Clement and TJ Ridsdill-Smith

Multiple disease resistance in wild *Cicer* accessions: One hundred and forty eight wild accessions belonging to seven *Cicer* species (30 accessions of *C. bijugum,* 3 accessions of *C. cuneatum,* 4 accessions of *C. echinospermum,* 47 accessions of *C. judaicum,* 27 accessions of *C. pinnatifidum,* 31 accessions of *C. reticulatum,* and 6 accessions of *C. yamashitae*) were evaluated individually for AB and BGM under controlled environment conditions. Of the 148 accessions, five accessions of *C. judaicum* (ICC 17211, IG 69986, IG 70030, IG 70037, and IG 70038) were found resistant (<3 rating on 1 - 9 rating scale), while 55 accessions were moderately resistant (3.1 to 5 rating on 1-9 rating scale) to AB. Two accessions IG 70037 and IG 70038 belonging to *C. judaicum* had a combined resistance (<3 rating on 1 - 9 scale), while three accessions of the same species (ICC 17211, IG 69986, and IG 70030) had a moderate resistance (3.1 to 5.0 rating) to AB and BGM.

Suresh Pande and Nalini Mallikarjuna

Output target 6A.4: Promising transgenic events of chickpea with proven resistance to *Helicoverpa* available for commercialization and introgression in locally adapted germplasm

Activity 6A.4.1: Develop transgenic events of chickpea for resistance to *Helicoverpa armigera* and evaluate their performance under contained greenhouse and field conditions

Milestone: 50 transgenic events of chickpea with Bt genes developed and screened in contained greenhouse (KKS/HCS/PMG) 2008

By using the axillary meristems as the ex-plant, *Agrobacterium* mediated genetic transformation of chickpea using *cry1Ac* and *cry1Ab* genes for resistance to *H. armigera* is being carried out by using the construct PBS 2310Ac carrying the *cry1Ac* gene. Thirty-five independent transgenic events have been produced and transferred to the greenhouse. Molecular characterization of these plants is underway. Chickpea transgenics carrying the *cry1Ab* gene are also being produced using the construct PHS 723 Bt. The construct contained *cry1Ab* gene under CAMV 35 promoter and *npt*II under *nos* promoter and polyadenylation sequence. About 40 independent transgenic events were transferred to greenhouse and the molecular characterization of these are underway. More than 30 are under the process of regeneration.

KK Sharma

Putative transgenic chickpea plants carrying *cry1Ab*, and *cry1Ac* genes evaluated for resistance to *Helicoverpa armigera* under greenhouse and field conditions: Six selections from over 50 transgenic events with *cry1Ac* gene produced at ICRISAT-Patancheru and Bose Institute, Kolkata, India (Under the IndoSwiss Project on Biotechnology) were bio-assayed for resistance to *H. armigera.* The plants were tested using the detached leaf assay in the laboratory. There were five replications in a completely randomized design. The detached terminal branches embedded in 3% agar-agar were infested with 10 neonate larvae in 250 ml plastic cups. Observations were recorded on leaf feeding (1 = <10% leaf area damaged, and 9 = >80% leaf area damaged), larval survival, and larval weights at 5 days after initiating the experiment. In the transgenic plants, leaf feeding scores ranged from 7.0 in ICCL 89314 Bt 2-4 to 8.8 in C 235 X 9-2 compared to 8.5 on the non-transgenic plants of ICCL 89314 and 7.8 on C 235. The larval weights ranged from 6.46 mg on C 235 X 6-5 to 8.37 mg on ICCL 89314 *Bt* 2-4 in the transgenic plants. The larval weights on the non-transgenic control were 7.99 mg on ICCL 89314 and 7.39 mg on C 235. The larval weights on the transgenic cotton were 1.99 mg compared to 9.6 mg on the non-transgenic control plants. The levels of expression in the transgenic chickpea plants appeared to be low, and we need to test new events to identify lines comparable to transgenic cotton in biological activity against *H. armigera*.

HC Sharma and KK Sharma

Progress reported towards the achievement of milestone for 2008 will contribute towards achievement of the milestones listed below.

Milestone: At least 8 promising Bt transgenic events of chickpea identified and insect resistance characterized under contained greenhouse conditions (KKS/HCS/PMG) 2009

Milestone: Three promising Bt transgenic events of chickpea identified and insect resistance characterized under contained field conditions (KKS/HCS/PMG) 2010

Milestone: One or two transgenic events of chickpea used for introgression into locally adapted genotypes and the progeny characterized and evaluated (KKS/HCS/PMG) 2011

Milestone: Biosafety of transgenic plants to non-target organisms assessed (HCS/KKS) 2010

Effect of *Bt* toxins and transgenic plants on the survival and development of the parasitoid, *Campoletis chlorideae*, and the coccinellid predator, *Cheilomenes sexmaculatus*: We studied the influence of *Bacillus thuringiensis* (*Bt*) toxins and the *Helicoverpa*-resistant genotypes of chickpea on the survival and development of the pod borer, *Helicoverpa armigera*, and their indirect effects on the host specific parasitoid, *Campoletis chlorideae*. The foliar damage by *H. armigera*, larval survival, and larval weights were significantly lower on *Bt* sprayed

chickpeas as compared to that on the untreated controls. The larval and pupal periods of the parasitoid, *C. chlorideae* reared on *H. armigera* larvae fed on different chickpea genotypes treated with *Bt* increased by 0.37 to 1.29 days and 0.19 to 0.58 days, respectively, as compared to those fed on untreated controls. The parasitoid cocoon formation was 18.7 to 38.7% on *H. armigera* larvae fed on *Bt* sprayed chickpeas compared to 69.3 to 77.3% on the untreated controls. Adult emergence was reduced by 61.1 to 83.8% over the untreated controls. There were no significant effects of *Helicoverpa*-resistant chickpea genotypes on the development and survival of *C. chlorideae*, suggesting that the *Helicoverpa*-resistant chickpea genotypes are compatible with this parasitoid. Larval weight of *H. armigera* showed a significant and positive association with *C. chlorideae* cocoon formation and adult emergence, and weights and size of male and female parasitoids (r = 74* to 99**), while significant and negative association was observed with larval period of *C. chlorideae* (r = -0.70*). The ELISA test indicated the presence of *Bt* protein in the *H. armigera* larvae fed on *Bt* treated chickpeas, while no *Bt* protein was detected in the larvae, cocoons, and adults of the parasitoid, *C. chlorideae* reared on *Bt* intoxicated *H. armigera* larvae, suggesting that lower survival of the parasitoid was due to poor quality of the host.

In another experiment, we studied the effects of cotton aphid, *Aphis gossypii* fed on *Bt*-transgenic and non-transgenic cottons on development and survival of the coccinellid, *Cheilomenes sexmaculatus*. The larval period of *C. sexmaculatus* reared on *A. gossypii* fed on *Bt*-transgenic cotton was prolonged by 1.5 to 2.0 days. The weight of *C. sexmaculatus* larvae was reduced by 3 to 4 mg when fed on *A. gossypii* reared in *Bt*-transgenic cotton plants. There was no effect of *Bt*-transgenic cotton fed aphids on larval survival, pupal period, and sex ratio of *C. sexmaculatus*. However, adult emergence decreased by 20 to 30% in coccinellids fed on *Bt*-RCH 2 and *Bt*-Mech 12 cotton hybrids as compared to those reared on non-transformed controls. The ELISA test indicated the presence of *Bt* protein in aphids, and the larvae and adults of the *C. sexmaculatus* fed on aphids obtained from *Bt*-transgenic cotton.

HC Sharma and MK Dhillon

Pigeonpea

Output 6A: Improved germplasm and varieties of sorghum, pearl millet, pigeonpea, chickpea and groundnut with pro-poor traits and associated advanced knowledge of selection tools and breeding methods made available to partners internationally

MTP Output Target 2006: New knowledge synthesized on seed production systems of improved lines, wild relatives and putative transgenics of pigeonpea, published and disseminated globally

Output target 6A.1: About 5 - 6 pigeonpea varieties with stable resistance to *Fusarium* wilt, sterility mosaic and *Helicoverpa* made available to NARS

Activity 6A.1.1: About 15 new genetically diverse germplasm sources/ breeding lines resistant to wilt and sterility mosaic diseases identified

Milestone: 25 - 30 pigeonpea lines tested multilocationally for their stability to wilt and sterility mosaic resistance in India (Annual) (SP)

Identification of stable sources of resistance to Fusarium wilt and sterility mosaic (SM): Pigeonpea lines found resistant to wilt and SM at ICRISAT, Patancheru were tested for wilt and SM resistance at different locations in India through the pigeonpea wilt and sterility mosaic disease nursery (PWSMDN) to identify stable and broad-based resistance to both these diseases. Thirty entries (28 wilt and SM resistant lines, and a susceptible check for each of the two diseases) were evaluated in 20 locations in India(Akola, Badnapur, Bangalore, Berhampore, Coimbatore, Dholi, Faizabad, Gulbarga, Hazaribagh, Hisar, ICRISAT, Patancheru, Kanpur, Khargone, Rahuri, Raipur, Sehore, SK Nagar, Pudukottai, Varanasi, and Warangal) during the 2005/06 cropping seasons. The experiment was planted in a wilt sick plot and inoculated with SM infested pigeonpea leaves using the leaf staple technique at two leaf stage or spreading SM infested twigs on test entries wherever possible. Data on wilt and SM was recorded twice, at flowering and at maturity stages of the crop.

Data was received from 13 locations. Wilt incidence in the susceptible check, ICP 2376 was high (> 60%) at Akola, Badnapur, Gulbarga, ICRISAT-Patancheru, Kanpur, Rahuri, and Sehore; while it was moderate at Bangalore and

Warangal (~ 38%); and low in Khargoan (13%). There was no incidence at Berhampur, Hazaribagh, and Pudukottai (Vamban). Incidence of wilt in the local check was 83% (TTB 7) at Bangalore, 54% (LRG 30) at Warangal, 50% at Pudukottai, 10% at Hazaribagh, and no infection at Berhampur. The differential reaction of susceptible checks at different locations indicated the possible presence/existence of races/pathotypes of the wilt pathogen. This phenomenon needs detailed investigations to resolve the race scenario of *Fusarium udum* in India.

Of the 28 entries tested, 27 entries at Berhampur, 26 at Bangalore, 25 at Sehore, 24 at Akola, 23 each at Badnapur, ICRISAT, Patancheru, and Pudukottai, and <20 entries in rest of the locations were resistant to wilt. Four entries, ICP 7870, ICP 9174, ICP 9576, and KPBR 80-2-2-1 at 11 locations, six entries (ICP 8610, ICP 12749, ICP 12755, PR 5149, ICPL 87119, and V 71 A) in 10 locations, eight entries (ICP 12751, ICPL 93179, ICPL 96053, ICPL 96058, ICPL 96061, ICPL 99044, IPA 40, and KPBR 80-2-1) in 9 locations were resistant to wilt (< 10% wilt incidence).

Though data books were received from 13 locations, SM data was recorded only in nine locations. Susceptible cultivar ICP 8863 had >75% SM incidence at Bangalore, Badnapur, ICRISAT, Patancheru, and Rahuri; while it was moderate (31%) at Pudukottai, and low (10%) at Coimbatore. The SM was completely absent at Berhampur, Hazaribagh, and Sehore. Incidence of SM in local susceptible check was highest at Bangalore (100%), Badnapur (65%), ICRISAT-Patancheru (100%), and Rahuri (100%); moderate at Coimbatore (45%) and Pudukottai (46%), and low at Berhampore (4%). The differential reaction of susceptible checks at different locations indicated the possible presence/existence of races/pathotypes in the SM pathogen.

ICPL 99044 showed resistance at seven locations. Ten entries, ICPL 7870, ICPL 8610, ICPL 9576, ICPL 12957, ICPL 12759, ICPL 94062, KPBR 80-2-1, KPBR 80-2-2-1, V 71A, and V71B at six locations, 12 entries at five locations, and five lines at four locations were resistant.

Suresh Pande and Collaborators

Milestone: About 100 germplasm/advanced breeding lines screened for wilt and sterility mosaic disease resistance using different isolates and characterized for agronomic traits (SP 2009)

Fusarium wilt and SM resistance in advanced germplasm lines: Fifty-six lines (eight lines from advanced selections in 2003/04, four from advanced selections in 2004/05, and 44 selections from breeders material in 2005-06) were evaluated for combined resistance to FW and SM under artificial epiphytotic conditions at ICRISAT-Patancheru. Additionally, data on natural incidence of *Phytophthora* blight (PB), which occurred during the current season due to heavy rains in the months of July and August, were also recorded.

Both FW and SM susceptible cultivars showed >85% disease incidence. Of the eight advanced selections, six lines (BDN 2010, BSMR 846, MAL 3, PT 1037, PT 2033, and PT 2035) had a combined resistance (< 10%) to both FW and SM. Additionally, all these six lines had a very low (~ 1%) natural incidence of PB, and PT 1037 was free from FW. KPL 96053 and MA-S-DEO-74 were asymptomatic to both FW and SM, MAL 13 and MAL 23 were resistant to both diseases. These lines were also resistant to PB.

Among the 44 promising breeding lines tested, ICPL 20098, ICPL 20106, ICPL20116, and ICP 11376-5 were asymptomatic, and 26 lines resistant to both FW and SM. Additionally, eight lines were asymptomatic and 25 were resistant (<10%) to FW, while 14 were asymptomatic and 27 were resistant to SM.

Fusarium wilt and SM resistance in advanced breeding inbred lines: Seventy-four advanced breeding inbred lines were evaluated for resistance to FW and SM following standard field screening technique. Among the 74 entries, ICPL 99044 was asymptomatic and 41 lines were resistant (< 10%) to both FW and SM. ICPL 20127 and ICPL 20108 showed a resistant reaction to FW, while 14 lines were asymptomatic and 5 were resistant to SM.

Fusarium wilt and SM resistance in breeding populations: One hundred and forty breeding populations were evaluated for resistance to FW and SM under artificial epiphytotic conditions following standard field screening technique. Neither combined resistance to FW and SM nor resistance to FW was recorded in any of the lines tested. However, five lines were asymptomatic and 23 were resistant (< 0%) to SM.

Suresh Pande and KB Saxena

Fusarium wilt and SM reaction of *Helicoverpa* resistant lines: Thirty-five *Helicoverpa*-resistant lines were also evaluated for combined resistance to FW and SM under artificial epiphytotic conditions using the standard field screening technique. Natural incidence of PB was also recorded during this season. Among these lines, neither combined resistance to both diseases nor to FW alone was observed in any of these lines. However, two lines ICP 4983-4 and ICPL 20040 were resistant to SM (< 0% disease). Twelve lines (selections 4977-16-2, 4978-4, 4978-5, 4982-2, and 4983-11, ICPL 20042, ICPL 20045, ICPL 20058, ICPL 20060, ICPL 97249, ICPL 97250, and ICPL 97253) showed resistance (<10% incidence) to natural incidence of PB.

Additionally, *Helicoverpa* promising selections of the genotype ICPL 332 from 2002 to 2004 were also evaluated to FW and SM. All the selections showed a susceptible reaction to both FW and SM.

Suresh Pande and HC Sharma

ICAR-ICRISAT collaborative research on pigeonpea Fusarium wilt and SM: Under the ICAR-ICRISAT collaboration, 82 entries (65 entries from national nursery for AVT and 17 entries from national nursery of disease-resistant genetic stocks) from IIPR, Kanpur, were evaluated for FW and SM resistance under artificial epiphytotic conditions following standard field screening technique. Natural incidence of PB was also recorded on these lines during this season.

In the AVT, combined resistance to FW and SM was not found in any of the lines tested. However, combined resistance to SM and PB was identified in eight lines (JKM 208, JKM 186, IPA 15F, KAWR 91, KAWR 11, ICP 7035, BRG 3, and BRG 2-5). Seven entries (PT 8208-1, IPA 16 F, IPA 15 F, Bahar, ICP 7035, NTL 30, and IPA 8F) were asymptomatic, while 10 entries were resistant (<10%) to SM.

Among the entries in the national nursery for disease-resistant genetic stocks, one entry MAL 13 had a combined resistance to FW, SM, and PB. Nine entries (ICP 8862, ICP 9174, Azad, KPBR 80-2-1, DA 11, KPL 43, KPL 44, MAL 3, and MAL 6) were resistant (<10%) to SM and PB. Two entries (Azad and DA 11) to SM, and five entries (ICP 8862, ICP 9174, KPL 43, MAL 3, and MAL 13) to PB were asymptomatic.

Suresh Pande and NARS Collaborators

ANGRAU-ICRISAT collaborative research on pigeonpea FW and SM resistance: In collaboration with breeders and pathologists from ARS, Warangal (ANGRAU), F_2 to F_6 breeding populations and progenies belonging to11 crosses and 25 advanced lines were evaluated for FW and SM resistance under artificial epiphytotic conditions following standard field screening techniques. Among these lines, two progenies (entry numbers 129, 130) showed combined resistance to FW and SM. Several progenies of all 11 crosses were found resistant to SM. Of the 25 advanced lines, no line had a combined resistance to FW and SM, but 10 lines were found asymptomatic to SM.

Suresh Pande and S Vanisri

MAU-ICRISAT collaborative research on pigeonpea wilt and SM resistance: Nine entries received from Badnapur, Maharashtra, were evaluated for combined resistance to FW and SM under artificial epiphytotic conditions. Additionally, all these entries were also scored for natural incidence of PB. Of these, none was resistant to FW. However, BSMR 198 and BSMR 853 were asymptomatic, while BWR 2 and BSMR 846 showed a resistant reaction to SM. Additionally, five entries (BDN 708, BDN 2009, BSMR 198, BSMR 846, and BSMR 853) were also resistant to natural incidence of PB.

Suresh Pande and OD Kohire

Milestone: Molecular characterization of wilt/sterility mosaic resistant and susceptible germplasm/breeding lines for developing mapping populations with diverse genetic background (RKV/KML/KBS/DAH) 2009

A set of 32 pigeonpea genotypes with known resistance and susceptibility to several diseases (e.g. sterility mosaic, wilt, etc.) has been selected. DNA from these genotypes has been isolated. A set of 30 polymorphic SSR markers has been selected and optimized for screening 32 genotypes. The genotyping with the SSR markers is in progress. Subsequently, the genotyping data will be analyzed for understanding the genetic relationships among 32 lines and for selecting the best parental genotype combinations for generating the mapping populations with diverse genetic background, segregating for wilt, and sterility mosaic diseases.

RK Varshney, R Saxena, K Madhavi Latha, K B Saxena and D A Hoisington

Activity 6A.1.2: Genetically diverse germplasm/breeding lines with resistance to *Helicoverpa* identified

Milestone: About 100 germplasm/advanced breeding lines screened for resistance to Helicoverpa under field and/or laboratory conditions (HCS/KBS/HDU) 2009

Advanced pigeonpea-breeding lines evaluated for resistance to *Helicoverpa armigera*: We evaluated selections from mini-core collection and purple pod lines of pigeonpea germplasm along with resistant and susceptible checks for resistance to *H. armigera*. There were three replications in a randomized complete block design. Fifteen lines showed a DR of <4.0 compared to a DR of 2.8 in ICPL 332, 5.0 in ICPL 87119, and 8.7 in ICPL 87.

In another trial, we evaluated 8 newly developed varieties of pigeonpea along with ICPL 332, ICPL 84060, and ICPL 87 for resistance to *H. armigera* under protected and unprotected conditions. Data were recorded on pod damage by pod borer, pod fly, and pod wasp, and grain yield. Percentage pod damage was 46.8 to 62.1% in ICPL 97249, ICPL 97250, and selection numbers 4985-10, 4985-11, and 4989-7 compared to 60.7% in ICPL 332 and 100.0% in ICPL 87. The overall damage rating was 1.9 to 3.0 in ICPL 97249, ICPL 97250, and selection numbers 4985-4, and 4985-7 compared to 4.0 in ICPL 332 and 7.7 in ICPL 87, indicating improved levels of resistance to *H. armigera* in some of the newly developed lines.

HC Sharma and KB Saxena

Activities 6A.1.3: Advanced generation interspecific derivatives with resistance to *Helicoverpa* using wild species from different gene pools developed

Milestone: Physico-chemical mechanisms of resistance to Helicoverpa in pigeonpea and its wild relatives identified (HCS) 2009

Physico-chemical mechanisms of resistance to *Helicoverpa* in pigeonpea: Fourteen morphologically diverse lines of pigeonpea were studied for their interaction with *H. armigera* in the field. The genotypes ICP 12476, ICP 8102, and ICP 9879 suffered <40.5% pod damage by *H. armigera* compared to 40.8% in ICPL 332, 75.6% in ICPL 87, and 73.9% in ICPL 87119. These genotypes also had 49.6 to 58.4% healthy pods per plant compared to 66.7% in ICPL 332, 11.5% in ICPL 87, and 48.9% in ICPL 87119. Under laboratory conditions, weight gain by the larva at 7 days after initiating the experiment was 41.5 mg on ICP 15632, 128.4 mg on ICP 5529, 155.3 mg on ICP 13555, and 159.3 mg on ICP 11975 as compared 287.3 mg on ICPL 332 and 252.5 mg on ICPL 87. The mechanisms of resistance in these pigeonpea genotypes are being studied in greater detail to identify morphological and biochemical markers for use in crop improvement.

HC Sharma

Milestone: Gene introgression from wild Cajanus into cultivated pigeonpea studied (NM/HDU/KBS) 2010

Cajanus acutifolius and *C. lineatus* from secondary gene pool of *Cajanus,* and *C. platycarpus* from tertiary gene pool were used in the crossing program. When used as a female parent, mature seeds were not obtained in case of *C. lineatus* and *C. acutifolius*, but the reciprocal of the cross was successful. It was possible to introgress male-sterility genes from *C. acutifolius*. Experiments are underway to introgress genes for resistance to *H. armigera*. The F_1 hybrid from the cross *C. lineatus* x ICPL 85010 is being grown in the glasshouse.

Nalini Mallikarjuna

Milestone: Ten interspecific derivatives from different Cajanus species belonging to different gene pools with resistance to Helicoverpa identified for use in pigeonpea improvement (NM/HCS/HDU) 2011

A total of 11,565 plants from 2,067 selections made in 2005/06 were screened for resistance to *H. armigera* under unprotected field conditions. Twenty-five percent of the selections showed 20% or less damage, and 45% of the selections had 30% or less damage by *H. armigera* (Fig. 1). Progenies of the plant 8329 selected in 2003 continued to segregate for *H. armigera* resistance with 0 to 100% damage. Nearly 60 % of the progenies had <50% damage (Fig. 2). Another selection, 6028 had some progenies with little damage, and 50% of the progenies had less than 30% damage by *H. armigera* (Fig. 3).

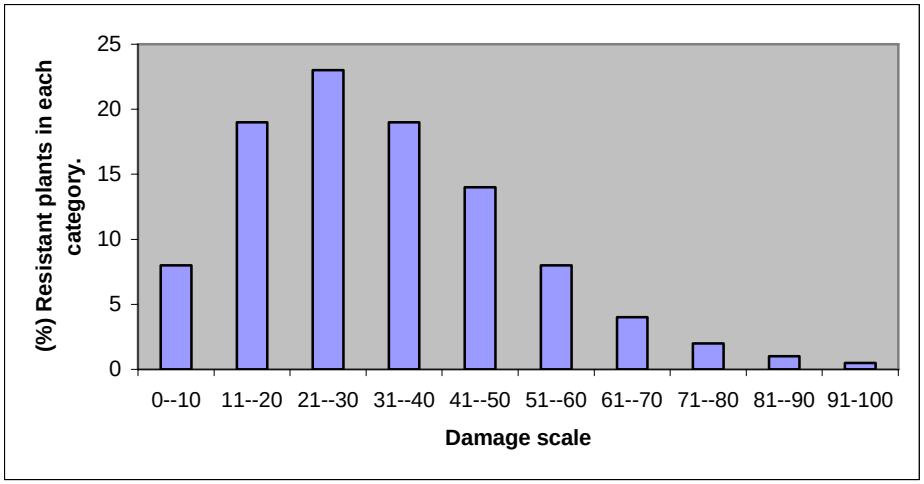

Fig. 1. Interspecific derivatives of pigeonpea (a total of 2067) evaluated for resistance to *H. armigera*

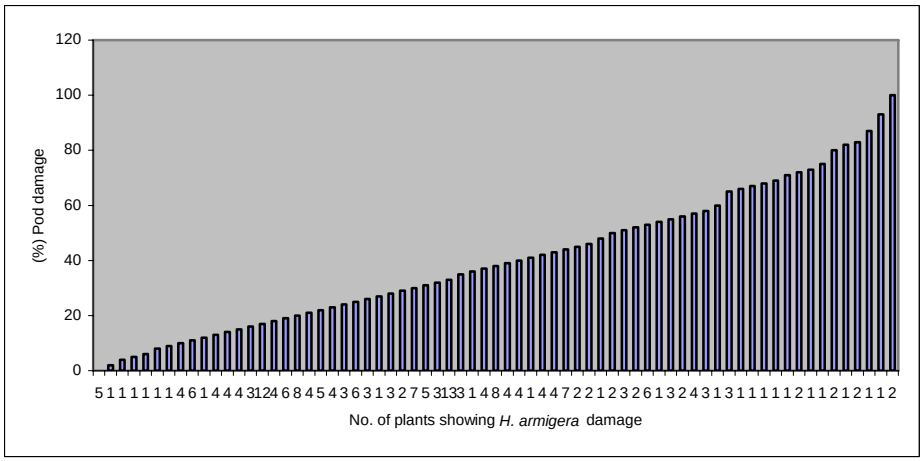

Fig. 2. Interspecific derivatives of pigeonpea (a total of 8329) evaluated for resistance to *H. armigera* (plant no. 8329)

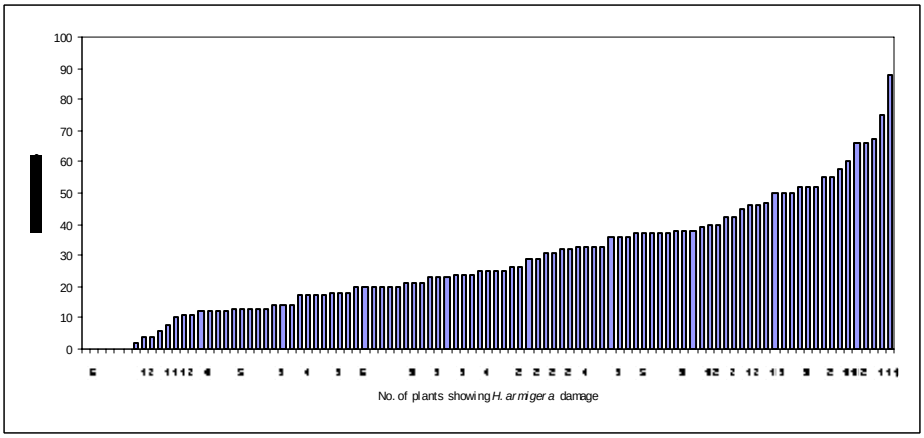

Fig. 3. Interspecific derivatives of pigeonpea (a total of 99) evaluated for resistance to *H. armigera* (plant no 6028)

Nalini Mallikarjuna and HC Sharma

Wide crosses for pod borer resistance in pigeonpea: To transfer the pod borer resistant gene(s) from *C. scarabaeoides* to cultigens, biparental mating between 38 *Helicoverpa*-resistant F_6 interspecific single plant progenies were attempted, and 17 F_1's were advanced to F_2 generation. A total of 78 F_2 single plant progenies of the cross ICPW 94-P1 x ICP 28-P1 were evaluated under controlled environment facilities.

HD Upadhyaya

Output target 6A.2: Promising transgenic events of pigeonpea with proven resistance to *Helicoverpa* available for commercialization and introgression in locally adapted germplasm

Activity 6A.2.1: Develop transgenic events of pigeonpea for resistance to *Helicoverpa armigera* and evaluate their performance under contained greenhouse and field conditions

Milestone: 80 transgenic events of pigeonpea with Bt genes developed and screened in contained greenhouse (KKS/HCS/KBS) 2008

Transgenic pigeonpea plants were developed through *Agrobacterium tumefaciens*-mediated transfer of the Bt *cry1Ac* gene, and were evaluated for resistance to *H. armigera* under contained greenhouse and field conditions. Using seedling leaf petiole as the explant, a total of 50 independent transgenic events; 20 events of ICPL 88039, 25 of ICPL 87, and 5 of LRG 41 were produced and successfully transferred to the greenhouse. All these plants were characterized for the presence, integration, and expression of the *cry1Ac* gene by PCR, RT-PCR, Southern, IPCR and ELISA. A total of 35 (16 of ICPL 88039, 14 of ICPL 87, and 5 of LRG 41), and 20 (12 of ICPL 88039 and 8 of ICPL 87) events were advanced to T_1 and T_2 generations, respectively. These transgenic plants were evaluated for resistance to *H. armigera* in contained greenhouse conditions using leaf and pod bioassays. Larval survival and weights were found to be significantly less on 18 independent transgenic events in leaf bioassays. A contained field trial was conducted by using nine events of ICPL 88039 and nine events of ICPL 87 during the 2005 rainy season. In the leaf, pod, and inflorescence bioassays of the field grown transgenics, 5 events showed significant effect on weight gain by *H. armigera* larvae, but no significant effects on mortality were observed.

More events of pigeonpea transgenics with *cry1Ab* and *cry1Ac* genes are being produced using *Agrobacterium*-mediated transformation. Ten independent transgenic events with the *cry1Ab* gene have been transferred to greenhouse and are being characterized for the integration and expression of the transgenes. Over the course of next year, the development of at least 75 transgenic events with each gene are planned.

KK Sharma and HC Sharma

Milestone: Three promising Bt-transgenic events of pigeonpea identified and insect resistance characterized under contained field conditions (KKS/HCS/KBS) 2009

Putative transgenic pigeonpea plants carrying *cry1Ab*, and *cry1Ac* genes evaluated for resistance to *Helicoverpa armigera* under greenhouse conditions: Transgenic pigeonpea plants developed by introducing the synthetic *cry1Ac* gene through *Agrobacterium tumefaciens*-mediated genetic transformation were bioassayed for resistance to *H. armigera* using detached leaf assay. Each fully expanded leaf embedded in 3% agar-agar was infested with 10 neonate larvae of *H. armigera*. Data were recorded on leaf damage, larval survival, and weight gain at 5 days after initiating the experiment. The leaf damage rating ranged from 5.5 to 8.4, and larval survival from 70 to 86%. The weight gain by the larvae was 0.96 mg to 1.71 mg on 723 *Bt* 1-2-1-2, 723 *Bt* 2-1-1-1, 723 *Bt* 11-19, 723 *Bt* 1-2-1-2, 2310 Ac-0-3, 2310Ac-10-3, 2310Ac-20-1, and 2310Ac-15-4 compared to 2.41 mg on the non-transformed control plants of ICPL 88039. In case of transgenic plants derived from ICPL 87 using cry1Ac gene, the larval weights were 8.99 to 9.76 mg on the ICPL 87-12-1, ICPL 87-14-3, ICPL 87-23-3, ICPL 87-25-5, ICPL 87-12-1, and ICPL 87-3-2 transgenic plants compared to 9.48 mg on the non-transgenic ICPL 87 control plants and 13.69 mg on ICPL 87-5-1, suggesting that the effect of *Bt* protein on *H. armigera* was lower in plants derived from the susceptible cultivar ICPL 87 than the ones derived from the relatively less susceptible cultivar, ICPL 88039.

HC Sharma and KK Sharma

Progress reported towards the achievement of milestone for 2008 will contribute towards achievement of the milestones listed below.

Milestone: One to two Bt-transgenic events of pigeonpea used for introgression into locally adapted genotypes (KKS/HCS/KML) 2010

Milestone: Commercialization package for the introduction of pigeonpea with transgenic resistance to Helicoverpa armigera available for deployment (KKS/HCS/KML) 2011

Milestone: Biosafety of transgene products/transgenic events to non-target organisms investigated (HCS/KKS) 2010

Movement of *Bt* toxins through different trophic levels in the field: We monitored the movement of *Bt* toxin, Cry1Ac through different trophic levels in the field. A total of 40 insect specimens (25 from transgenic and 15 from non-transgenic cotton) were collected, and tested for the presence of *Bt*-toxin using qualitative ELISA at different intervals during the crop-growing season. Of the 25 insect species colleted from transgenic plots, 7 showed high levels, 9 showed low levels, and the remaining 9 had no Bt-toxin. Further studies on the sensitivity of different insect species to *Bt*-toxins and their uptake by the natural enemies are in progress.

HC Sharma and MK Dhillon

Output Target 6A.3: Twenty medium-long duration vegetable type pigeonpea germplasm/breeding lines made available

Activity 6A.3.1: Evaluation and selection of large pod medium – long duration germplasm and breeding lines for use as vegetable

Milestones: Research Bulletin on vegetable pigeonpea published (KBS) 2007

A Research bulletin on various aspects of vegetable pigeonpea including breeding and quality will be published in 2007.

KB Saxena

Milestones: At least 5-10 large seeded high-yielding vegetable type breeding lines and germplasm identified (KML/KBS/HDU) 2008

A number of large seeded vegetable types pigeonpea breeding and germplasm lines will be evaluated in vertisols for pod yield and seed of the selections will be made available for multilocation testing.

K Madhavi Lata, KB Saxena and HD Upadhyaya

Milestones: Genetically diverse large seeded vegetable type 10-15 breeding populations for further selection developed (KBS/KML) 2011

Genetically diverse vegetable lines will be crossed in 2007 rainy season to develop breeding populations for further selection and seed supply to NARS.

K Madhavi Lata and KB Saxena

<u>Sorghum</u>

Output A: Improved germplasm and varieties of sorghum, pearl millet, pigeonpea, chickpea, and groundnut with pro-poor traits and advanced knowledge of selection tools and breeding methods made available to partners internationally

MTP Output Target 2006: Sweet sorghum germplasm (500 lines) screened for Brix percentage and selected material available to NARS

Output target 6A.1: High biomass forage/sweet sorghum lines with tolerance to stem borer, bold grain lines with resistance to shoot fly for post-rainy season, and high-yielding sorghum lines with resistance to grain mold for rainy season developed

Activity 6A.1.1: Selecting high biomass forage and sweet sorghum lines with tolerance to insect and foliar diseases, and grain sorghum with tolerance to grain mold

Milestone: Ten sweet-sorghum lines with high biomass and tolerance to stem borer and foliar diseases developed (BVSR/HCS/RPT/RS) 2007

A trial involving 30 varieties, including two checks, was conducted during the 2006 rainy season at the Agricultural Research Station (ARS), Gangavathi, Karnataka, India to ascertain the salinity tolerance and grain yield in large grain backgrounds. Eight varieties out-yielded the best check, ICSV 112 for grain yield and were comparable for grain size in the saline soils (10 dSm^{-1}) of ARS, Gangavathi. They will be evaluated for stalk yield, sugar content, tolerance to stem borer, and foliar diseases in the 2007 rainy season.

To improve the stalk and stalk sugar yield, the high tillering population (ICSP-HT) was mass selected for tillering ability, biomass, and height during the 2006 rainy season. From the population bulk, 220 male-steriles and 130 male-fertiles were selected. From SSV 74 F_3 crosses bulk, 38 male-steriles and 60 male-fertiles were selected, and from the SSV 84 F_3 crosses, 27 male-steriles and 98 male-fertiles were selected. The C_{11} population bulk introgressed with F_3 bulks of SSV 74 and SSV 84 was constituted by mixing the seed of male-steriles and male-fertiles in a 3: 1 ratio. These will be evaluated and advanced with selection in the 2007 rainy season. Similarly, the tall lines among them will be evaluated for tolerance to stem borer, foliar diseases, and for stalk and grain yield to be used as sweet sorghum varieties.

Based on the evaluation of sweet sorghum restorer parents, varieties and landraces in the 2005 rainy season, 36 lines were selected and evaluated along with the check, SSV 84 in the 2006 rainy season. Compared to the check, SSV 84 (1.8 t ha^{-1}), 19 lines with a grain yield range of 4.6 to 7.1 t ha^{-1} (on par with the control for sugar yield) were advanced. The performance of the selected five lines is given in Table 1**.**

Table 1. Performance of selected sweet sorghum restorers/varieties (at maturity stage) (ICRISAT, Patancheru, 2006 rainy season)

R-line/Variety	Days to 50% flowering	Plant height (m)	Grain yield (t ha^{-1})	Cane yield (t ha^{-1})	Juice yield (t ha^{-1})	Brix reading at maturity	Sugar yield based on Brix reading and juice yield (t ha^{-1})
SP 4487-3	83	3.2	7.6	83.6	41.0	18.3	7.1
SPV 422	84	3.3	7.2	75.4	38.4	19.0	6.9
SP 4484-1	83	3.4	5.6	82.5	38.1	18.5	6.8
SSV 53	80	3.2	8.9	76.9	36.6	19.2	6.7
SP 4482-2	82	3.4	5.2	76.0	40.4	17.0	6.7
SSV 84 (Check)	83	3.0	1.8	66.4	27.6	21.0	5.3
Mean	77	3.1	6.0	58.4	27.3	17.2	4.6
CV (%)	2.33	7.78	13.24	19.40	23.10	8.25	22.33
LSD (5%)	2.88	0.38	1.26	18.17	10.11	2.28	1.63

BVS Reddy

Milestone: At least 5 sorghum lines with large grain, high grain yield, and tolerance to shoot fly for post-rainy season adaptation developed (BVSR/HCS) 2008

In order to assess the farmers' selections in relation to breeders selection, a trial consisting of 9 (4 farmers and 5 breeders) post-rainy season adapted sorghum lines selected from F_7 generation were evaluated at ICRISAT, Patancheru; UAS Regional Agricultural Research Station, Bijapur; and Punjabrao Krishi Vidyapeeth, Akola (India) during the 2005/06 post-rainy season. Data were recorded for days to 50% flowering, plant height (m), staygreen score on a 1 to 5 scale at harvest (where 1 = >75% green, 2 = up to 75%, 3 = 26 - 50%, 4 = 10 - 25%, and 5 = <10% green leaf area), lodging score (on a scale 1 to 5 scale; where 1 = no lodging, 2 = up to 25% lodging, 3 = 26 - 50%, 4 = 51 - 75%, and 5 = >75% lodging), grain size (g 100^{-1}), and grain yield (t ha^{-1}). The CV for grain yield from Akola was very high, and hence the data from Bijapur and ICRISAT were combined and presented here. For grain size, data recorded at ICRISAT is presented. The mean grain yield of breeders' selections (3.7 t ha^{-1}) was marginally superior to that of farmers' selections (3.4 t ha^{-1}). However, for other traits (grain size, days to 50% flowering, and

plant height) farmers' selections were similar to that of breeders' selections (grain size: 2.3 g 100^{-1}, plant height: 1.9m, and days to 50% flowering: 77 days). Breeders' selections were more tolerant to lodging (Table 2). From these results, it can be concluded that though farmers selections in the early generations had bolder grains (2005 archival report), with advancement in generations, breeders' selections had high grain yield with grain size similar to farmers' selections.

Table 2. Performance of sorghum varieties in Participatory Plant Breeding Trial (PPBT) (ICRISAT, Patancheru, and Bijapur, 2005 post-rainy season).

digree	Grain yield (t ha^{-1})	Days to 50% flowering	Plant height (m)	Stay green score	Lodging score	100-grain weight (g) at ICRISAT
Farmers' selections						
(M 35-1 × SPV 1359)-5-2-1-1-1-1	3.6	77	1.9	2.8	1.7	2.3
(M 35-1 × SPV 1359)-8-2-2-1-1-1	2.9	79	1.9	3.0	1.5	2.2
(M 35-1 × SPV 1380)-1-1-1-1-1-1	3.6	76	2.0	3.3	1.8	2.2
(M 35-1 × SPV 1380)-1-1-2-1-1-1	3.5	76	1.9	2.7	1.8	2.3
Mean	**3.4**	**77**	**1.9**	**3.0**	**1.7**	**2.3**
Breeders' selections						
(M 35-1 Bulk 2 × SPV 1359)-1-1-1-1-1-1	3.9	76	1.9	3.3	1.7	2.3
(M 35-1 Bulk 3 × SPV 1359)-4-1-3-1-1-1	3.1	77	1.9	3.0	1.0	2.6
(M 35-1 Bulk 3 × SPV 1359)-4-3-1-1-1-1	3.9	78	1.9	3.0	1.5	2.4
(NTJ 2 × SPV 1359)-5-2(Tan)-1-1-1-1	3.6	78	1.9	3.3	1.5	1.8
M 35-1-Bulk-3-29-3-1-1-1-1-1	4.3	76	1.9	3.0	1.3	2.4
Mean	**3.7**	**77**	**1.9**	**3.1**	**1.4**	**2.3**
Moulee (Check)	3.0	76	1.9	2.8	2.0	3.9
M 35-1 (Check)	3.0	76	1.8	2.5	1.2	3.7
CSH 15 R (Check)	4.4	76	1.9	3.0	1.3	3.4
Mean	**3.6**	**77**	**1.9**	**3.0**	**1.5**	**3.8**
SE ±	0.45	0.87	0.08	0.31	0.21	0.04
CV (%)	21.92	1.97	6.98	17.90	23.91	1.70

The advanced lines will be evaluated for shoot fly resistance. In another experiment, a total of 60 entries (selected based on their yield and large grain), including checks, were planted in RCBD with 3 replications for evaluation for grain yield and other agronomic traits during the post-rainy season 2006. The promising lines will be evaluated for shoot fly resistance in 2007.

BVS Reddy

Milestone: Ten high yielding grain mold tolerant sorghum lines developed for rainy season adaptation (BVSR/RS) 2010

New sources of resistance for grain mold will be obtained and selfed during 2006/07 post-rainy season. Crossing program will be initiated during 2007 rainy season using new sources of resistance to grain mold from germplasm and high yielding adapted B lines.

BVS Reddy

Activity 6A.1.2: Developing QTL mapping populations for economic yield components of sweet sorghum

Milestone: Two F_5 sorghum RIL mapping populations (300 lines each) available for genotyping and multilocational phenotyping for biomass yield, sugar content, and sugar extraction characteristics (CTH/BVSR) 2010

Contrasting parents, two sets each for biomass yield, sugar content, and sugar extraction characteristics have been identified. They will be used in crossing program in 2007 to derive the mapping populations (RILs).

BVS Reddy

Milestone: SSR-anchored DArT marker linkage maps (>250 marker data points per RIL) available for the two sorghum RIL populations (CTH/SPD) 2010

During the 2006 rainy season, plant x plant crosses were made, and F_{1s} and selfed parental plant seeds harvested. Pairs of genetically diverse (dissimilarity greater than 0.70 based on allelic variation detected at 67 polymorphic SSR loci distributed across 10 sorghum linkage groups), agronomically elite inbred parental lines differing in stem sweetness, salinity tolerance in both pot and field screens), and other agronomic characteristics were selected for making the crosses. Parental line pairs from which F_1 seed has been harvested included: BTx623 (salinity tolerant, non-sweet) x ICSR 93024-1 (salinity sensitive, sweet), BTx623 x S 35 (salinity sensitive, sweet), ICSV 93046 (salinity tolerant, sweet) x ICSR 93024-1, ICSV 93046 x SPV 1022 (salinity sensitive, somewhat sweet), ICSV 93046 x S 35, and SP 39105 (salinity tolerant) x ICSR 93024-1. The F_1 seeds have been sown in the 2006/07 post-rainy season.

SP Deshpande and CT Hash

Activity 6A.1.3: Selecting for high grain yield and large grain sorghum lines with resistance to shoot fly and adaptation to post-rainy season

Milestone: Sorghum lines (5) with large grain and high grain yield and less susceptible to shoot fly with post-rainy season adaptation developed (BVSR/HCS) 2008

During post-rainy season 2005-06, 13 lines adapted for post-rainy season (Moulee, M 35-1, IS 33844-5, M 35-1-19, M 35-1 bulk 1, 2, 3, 4, SPV-1359, SPV 1411, SPV 1380, Giddi Maldandi, and NTJ-2) were crossed to obtain the F_1s. The F_1s were grown in the 2006 rainy season and harvested separately. A total of 125 F_2s were planted during the post-rainy 2006 to undertake selections. The agronomically superior advanced breeding lines obtained will be screened for shoot fly resistance. In another experiment, 18 F_2s derived from crossing five dual season restorer lines (M 35-1-19, ICSR 93001, 92003, 93031, and IS 33844-5) were sown during the post-rainy 2006 to undertake selections. The advanced lines will be screened for SFR based on their agronomic performance.

BVS Reddy

Activity 6A.1.4: Selecting for high grain yield and grain mold tolerant sorghum lines

Milestone: Sorghum lines (5) with high grain yield and less susceptibility to grain mold developed for rainy season adaptation (BVSR/RPT) 2009

A total of 120 F_5 progenies selected from the crosses of grain mold resistant landraces, high yielding B-lines, and grain mold resistant breeding lines were evaluated in advanced screening trial for grain mold resistance (GMR) during the 2006 rainy season. These progenies along with their test-crosses on known sources of A_1 and A_2 cytoplasmic male-sterile (CMS) systems-based male-sterile lines were also evaluated in a separate nursery in breeding block during the 2006 rainy season. The results on screening trial are awaited. A total of 64 lines with GMR (PGMR $\leq$ 5) and maintainer reaction were selected from the breeding block nursery. These included 60 white grain lines and four red grain lines. The time to 50% the flowering in these 60 white grain lines ranged from 52 - 80 days and plant height ranged from 1.0 - to 2.0 m.

BVS Reddy and RP Thakur

Output B. Annually knowledge of the improvements of the biotechnological and conventional tools designed to facilitate drought and salinity tolerance breeding and germplasm of mandate crops and associated capacity building made available to partners internationally

Groundnut

MTP output Target 2006: At least 15 new high yielding drought tolerant breeding lines made available to partners

Output target 6B.1: Develop groundnut varieties with tolerance to drought using conventional and biotechnological approaches

Activity 6B.1.1: Develop high yielding groundnut varieties tolerant to drought

Milestone: 15 - 20 new high yielding drought tolerant breeding lines made available to NARS annually (SNN/RA/VV) Annual

During the 2005/2006 post-rainy and the 2006 rainy seasons, 37 new crosses were made to generate populations for selection for resistance to drought and high pod yield with desirable agronomic traits. ICGV 05151, ICGV 06423, ICGV 01265, ICGV 03115, ICGV 95386, ICGV 03057, and ICGV 06443 were involved in hybridization.

Milestone: 5 - 10 new sources of resistance to drought identified (SNN/RA/VV) 2009

During the 2005/2006 postrainy season and the 2006 rainy seasons, 386 lines (along with controls) were evaluated in nine replicated yield trials, and 192 lines in 4 augmented trials. In the post-rainy season, early generation breeding materials and advanced breeding lines were evaluated by subjecting them to moisture stress by withholding alternate irrigation in the normal irrigation schedule (12 day intervals) starting from 65 days after sowing until harvest. The elite and advanced trials weree also grown with full irrigation in the same field to assess the full yield potential of the breeding lines included in these trials.

2005/2006 post-rainy season: In an elite trial (Spanish), ICGV 03064 (under stress - 3.1 t ha^{-1} pod yield and 61% shelling outturn; irrigated - 4.9 t ha^{-1} pod yiels, and 67 % shelling outturn) outperformed the best control ICGS 76 (under stress- 2.7 ± 0.26 t ha^{-1} and 67%; irrigated - 3.9 ±0.63 t ha^{-1} and 76%) under irrigated and stress conditions. It also significantly outyielded (rainfed - 4.9 t ha^{-1} and 60%; irrigated - 5.0 t ha^{-1} and 70%) the best control (rainfed: ICGV 00350 – 2.8 ± 0.21 t ha^{-1} and 67%; irrigated : ICGV 86325 – 4.8 ± 0.32 t ha^{-1} and 71%) in the 2005 rainy season.

2006 rainy season: In the new elite trial (Spanish), none of the entries outperformed the highest yielding control under both irrigated and rainfed conditions. In the new advanced trial (Spanish) evaluated under rainfed conditions, ICGV 05151, CGV 05155, CGV 05162, and CGV 05158 outyielded (3.3-2.9 ± 0.18 t ha^{-1}) the highest yielding control ICGV 87846 (2.3 t ha^{-1}). When this trial was evaluated under irrigation, the same four lines (4.2 - 3.9 ± 0.27 t ha^{-1}) outyielded the highest yielding control ICGV 87846 (2.9 t ha^{-1}). The best line came from (ICGV 99160 x ICGV 99240) cross. In the new advanced trial (Virginia) under rainfed conditions, 14 lines (3.7 - 2.8 ± 0.23 t ha^{-1}) significantly outyielded the highest yielding control ICGV 00350 (2.3 t ha^{-1}). Under irrigation, only ICGV 06424 (4.1 t ha^{-1}) outperformed the highest yielding control ICGV 87846 (2.3 t ha^{-1}). In the new preliminary trial (Spanish), 23 lines (3.8 - 3.2 ± 0.21 t ha^{-1}) outyielded the best control ICGV 02266 (2.6 t ha^{-1}). Best performer ICGX 020048 came from ((ICGV 99069 x ICGV 93184) x (ICGS 44 x ICGS 76)) cross. In the new preliminary trial (Virginia), 6 lines (3.5 - 3.0 ± 0.27 t ha^{-1}) significantly outperformed the best control ICGV 87846 (2.9 t ha^{-1}). The top line ICGX 020054 came from (ICGV 92069 x ICGV 93184) cross. Four lines (ICGV 03056, ICGV 03057, ICGV 03109, and ICGV 03115) were selected for inclusion in the international trials in the 2005/06 post-rainy season.

Milestone: 15 - 20 advanced lines tested in Anantapur in India and other drought prone areas (SNN/RA/VV) 2009

A special trial was formulated to develop varieties specially suited for drought prone Anantapur district in Andhra Pradesh. Based on the results obtained from both Anantapur and ICRISAT during the last two years (details given in Archival report 2005), we selected 16 lines for further evaluation at ICRISAT and ARS, Anantapur. At Anantapur, the trial could be sown only on 12 September 2006 as the monsoon was delayed during the 2006 rainy season. At

ICRISAT, 8 lines (2.6-1.6 ± 0.13 t ha^{-1}) outperformed the best yielding control TAG 24 (1.3 t ha^{-}) and 13 lines (2.6 - 1.4 ± 0.13 t ha^{-1}) significantly outperformed ICGV 91114 (1.0 t ha^{-1}). When the same trial was evaluated under irrigation, 7 lines (3.2 - 2.4 ± 0.16 t ha^{-1}) outperformed the highest yielding control TAG 24 (2.1 t ha^{-1}). The top seven lines were common in both the environments. The best line ICGV 06436 came from (TAG 24 x ICGV 86300) cross. In another trial, 7 out of 16 lines (2.9 - 2.1 ± 0.16 t ha^{-1}) outperformed the best control ICGV 00350 (1.7 t ha^{-1}) under rainfed conditions at ICRISAT. Under irrigation, none of the entries significantly outyielded the best control. The best entry ICGV 06456 came from (AK 12-24 x ICGV 99032) cross.

Milestone: Physiological traits in 3 – 4 superior sources for drought tolerance dissected (VV/SNN/RA) 2010

An experiment was carried out to test the effect of a water deficit applied at different stages (early flowering, late-flowering, and pod-filling) on pod yield and pod number. We used standard dry-down technique to apply the stress at these stages. At each stage, stress was imposed according to dry-down technique, whereby the WS set was re-watered when the relative transpiration was between 10 - 20% of control. We based the analysis on pod number per plant relative to control. Water stress at mid-pod filling stage had no large influence on the pod number relative to the control (pod number was 90% of control across genotypes), although a few genotypes such as ICR 48 and JUG 26 had relatively lower pod number under stress. By contrast, the relative pod number of plants exposed to stress at early- and late-flowering was 63 and 76% of that of control across all genotypes tested, with a fairly large variation across genotypes (35 - 93% for early-flowering stage, and 49 - 98% for late-flowering stage). During the early-flowering stage, the drought tolerant genotype ICGV 91114 was able to maintain 93% of control pod number.

Vincent Vadez

Activity 6B.1.2: Mapping and marker-assisted breeding for drought tolerance in groundnut

Milestone: QTL mapping of component traits of drought tolerance (TE) using available and suitable populations (VV/RA/SNN/RKV) 2008

One population [ICGV 86031 (high TE) x TAG 24 (low TE)] has been phenotyped for two consecutive years for TE in pot experiments in outdoor conditions during the Feb - April. Transpiration efficiency (TE) data have shown a good agreement between years. Usual surrogates for TE [specific leaf area(SLA), and Spad chlorophyll meter reading (SCMR), and the carbon discrimination ratio, $\Delta^{13}C$] have shown poor relation with the TE values, indicating that, though the use of these surrogates remains valid to carry out large scale screening of germplasm, they have to be used carefully for phenotyping, and measurement of TE itself is advised.

The parental genotypes of the mapping population were screened with >500 microsatellite markers and the polymorphic markers were identified. Genotyping of the mapping population has been initiated. Based on genotyping data of >50 markers on the population, a preliminary map-free linkage analysis has been undertaken. The analysis revealed a few markers linked to TE, and to several surrogates (SCMR). However, these markers explained 10% of the phenotypic variation. A larger number of markers would be needed in groundnut, as well as a larger range of variation in TE, to find out robust QTLs for TE. In this direction screening of the parental genotypes with a larger number of markers and genotyping of the mapping populations with more polymorphic markers is in progress.

Vincent Vadez, Rajeev Varshney and L Krishnamurthy

Milestone: Mapping populations between contrasting parents developed to identify QTLs for component traits of drought tolerance (root traits) (VV/RA/SNN) 2009

Screening for TE was undertaken in 2006 on 440 genotypes, including the mini-core collection of groundnut, which showed over 5-fold variation for TE. This range is higher than the variation observed in 45 lines tested previously (mostly breeding lines) to identify contrasting parents for TE, from which 2 segregating populations for TE have been developed. Similarly the germplasm collection was genotyped with 21 microsatellite markers to assess the molecular diversity. In a subset of the germplasm (189 accessions), the microsatellite markers yielded 3 to 20 alleles with an average of 12.4 alleles per marker with an average PIC value of 0.84. A repeat of the TE screening will be

performed. Subsequently the molecular diversity data together with the trait diversity data will be analyzed and the candidate genotypes with a greater TE and genetic diversities will be identified to develop new populations for TE.

Vincent Vadez, Rajeev Varshney and L Krishnamurthy

Milestone: Range of variations for root traits assessed in groundnut germplasm (VV/SNN/RA/HDU) 2008

Measurement of root traits, though better and more easily done in a controlled cylinder system, remains a time consuming exercice, with large error component, using destructive sampling, and providing "static" data that does provide little information about the actual activity of roots and the kinetics of this activity. In the end, more than the roots, water uptake and the kinetics of water uptake to cope with terminal drought are important. We used 4 groundnut genotypes and grew them in 1.2 m PVC cylinders with 16 cm diameter. Plants were grown for 30 days. Fifteen plants per genotype were grown. At 30 DAS, 5 plants per genotype were harvested to assess root depth and root dry weight in 15-cm layers. The other 10 cylinders were saturated with water, and 5 plants maintained under well-watered conditions (WW) and the other 5 left with no further irrigation (water stressed, WS). Cylinder weight was recorded on a regular basis, usually every 3 days. Water loss in WW plants was adjusted to the cylinder weight 3 days after imposing the treatment. The process of weighing the cylinders was relatively simple and rapid. Data showed that 5 days after stress imposition, normalized TR (NTR) was about 50% of controls. Thereafter, and until harvest at 17 days after treatment imposition, NTR of TMV 2, a genotype known to be drought-susceptible, was 20 % lower than TAG 24 and ICGS 44 (these 2 genotypes are known for deep and profuse rooting). At 17 days after stress imposition, TMV 2 plants were permanently wilted whereas TAG 24 and ICGS 44 were not. Total transpiration values from the time of treatment imposition were similar in TAG 24 and TMV 2, but the total TR value from 5 to 17 days after stress imposition were about 20% higher in TAG 24 than in TMV 2, showing that TAG 44 was able to manage better its water uptake compared to TMV 2, which allowed it to maintain higher NTR later during the stress. At harvest, root depth was measured after washing the roots and stretching. The root were cut in 15 cm portions, which were dried and weighted. We found that there was no significant correlation between either root dry weights, or with root dry weight in each of the 15-cm portion and TR. By contrast, total TR between 5 and 17 days after stress imposition was related to root depth. These data show that there is variation for the pattern of water extraction, and that root dry weight, which is usually well correlated to root length density, appears to be a poor proxy for water uptake.

Vincent Vadez

Milestone: Molecular markers ready for validation and use in introgression studies for abiotic and biotic stresses (VV/RA/SNN/RKV) 2009

The set of polymorphic markers suitable to map QTLs in groundnut is being built up.

Rajeev Varshney and Vincent Vadez

Milestone: QTLs for root traits identified (VV/SNN/RA) 2011

In the experiment reported above, we showed that though root traits displayed variation between genotypes. Only the differences in root depth were related to differences in water uptake, whereas differences in dry weight at different soil depth were not. Furthermore, differences in water extraction in the last 10 days of the experiment were relatively larger than the differences in root depth. This shows that water uptake is probably a better estimator of "root" traits than root traits (such as root length density, depth, dW, etc.) traditionally measured.

Vincent Vadez

Output target 6B.2: High throughput molecular genetic and phenotyping platforms for drought and salinity stress and promising transgenic events of groundnut for tolerance to drought stress available for commercialization and introgression in locally adapted germplasm

Activity 6B.2.1: Develop groundnut transgenic events for enhanced tolerance to drought

Milestone: 50 transgenic events of groundnut with DREB1A gene screened for drought tolerance in the contained greenhouse (KKS/VV/RA) 2007

The transcription factor *DREB1A,* which regulates the gene expression via recognition of the DRE (Dehydration Responsive Element) sequence was placed under the control of the stress inducible *rd29A* promoter, both isolated from *Arabidopsis thaliana*, and introduced into the peanut variety, JL 24 through *Agrobacterium tumifaciens* mediated gene transfer. Over 50 transgenic events were produced and advanced to the T_4 generation. The transformants were screened by polymerase chain reaction (PCR), RT-PCR, and Southern analysis for the presence and expression of *DREB1A*. Initial assessment of 14 transgenic events carried out by using the soil drying experiments showed differences in their transpiration responses to soil drying. There was a significant variation amongst the tested events under drought stress for transpiration efficiency (TE). A significant positive correlation with SCMR($r = 0.7359$) and a negative correlation with specific leaf area (SLA) ($r = 0.8237$) were obtained. However, TE did not correlate significantly with $\Delta^{13}C$, thus suggesting a lack of relationship between TE and $\Delta^{13}C$. Two events, RD2 and RD11 appeared to have higher TE than original cultivar JL 24.

Comparison of promising events RD2 and RD with high TE (JUG 24), and low TE (TAG 24) materials: The purpose of this experiment was to confirm the superiority of these events and compare them to known germplasm for high (JUG 24) and low (TAG 24) TE level. JL 24 had TE at the level of TAG 24, which was expected, lower than TE in JUG 24. Results confirm that RD 2 has a higher TE than JL 24 under water deficit conditions. It also showed that RD 2 had higher TE than the best reported genotype (JUG 24) for TE under water deficit. Event RD 11 had a TE that was only slightly above that of JL 24 and TAG 24. Therefore, only RD 2 event appeared to confirm its previous superiority for TE.

Effect of DREB1A on the response to water deficit at different growth stages (early flowering, late-flowering, and pod-filling): This experiment was carried out to: i) measure TE at different stages in two transgenic events (RD 2 and RD 11), control JL 24, and few breeding lines; and ii) test the effect of a water deficit applied at different stages (early flowering, late-flowering, and pod-filling) on pod yield and pod number. We used the standard dry-down technique to apply the stress at different stages. At each stage, TE was measured so that sets of plants were harvested before imposing the stress, and others (well-watered and water stress) harvested after water stressed plants had depleted all the soil moisture. To measure and compare the effect of a drought spell at different stages on the final pod yield, we applied the treatment (WS and WW) to two more sets of plants at each stage. After treatment imposition, the WS were re-watered and kept until maturity along with the WW set. The WS set was re-watered when their relative transpiration was between 10 - 20% of that of control. We based our analysis on pod numbers per plants, rather than pod yield because of mite infection towards the end of crop maturation.

The TE measured at flowering stage was higher in RD 2 and RD 11 than in JL 24 under water stress conditions, but was similar in all the lines under WW conditions. At the late flowering stage, TE of RD was superior to that of JL 24 only under WW conditions, whereas TE of RD 11 was similar to JL 24. Under water stress, TE was similar in all three lines.

The effect of a water stress at mid-pod filling stage had no influence on the pod number relative to the control (pod number was 90% of control across all genotypes), although a few genotypes such as ICR 48 and JUG 26 had a relatively lower pod number under stress. By contrast, the relative pod number of plants exposed to stress at early- and late-flowering was 63 and 76 % of that of the control across all genotypes tested, with a fairly large variation across genotypes (35 - 93% for early-flowering stage, and 49 - 98% for late-flowering stage). During the early-flowering stage, the drought tolerant genotype ICGV 91114 was able to maintain 93% of control pod number. RD 2 and RD 11 maintained 82 and 56% of control pod number, respectively, whereas JL 24 maintained only 63 % of control pod number, indicating that RD 2 was able to maintain pod number when exposed to stress. When the stress was applied at the late-flowering stage, there were no differences in relative pod number between RD 2 and JL 24 (61% in both cases). The breeding lines ICGV 91114, TAG 24, and ICGV 86031 had a relative pod number at about 90% of the control. This experiment showed that flowering is an extremely sensitive stage to water deficit, where the differences between the genotypes were maximum.

Five more transgenic events of groundnut transformed with rd29::DREB1A were tested for TE. The positive plants were tested at T_2 stage. Using the regular dry-down technique, a large number of replicates were tested for each

genotype. We included also RD2, the event showing consistently higher TE in previous experiments. Under WW conditions, TE of RD was similar to that of JL 24. However, one event, RD33, had higher TE under WW than JL 24 (7.70 vs 4.80). Under water stress conditions, RD2 had higher TE than JL 24 (7.42 vs 4.89). Two events (RD33 and RD34) also had TE superior to JL 24. Event RD33 had higher TE than JL 24 across water regimes, and even higher TE than best lines identified so far RD2. We also recorded transpiration response as a function of FTSW (index for soil moisture content) and found that all DREB1A plants showed a decline in transpiration in dryer soil than JL 24. This pattern is fully consistent with previous experiments using DREB1A transformants. A repeat of that experiment is planned to confirm the differences.

The lysimetric system described above has been used to assess 5 DREB1A transgenics (RD2, RD11, RD12, RD19, and RD20) and their non-transformed parent. The experimental design was similar to that described above, except we had 6 replications for each set (pre-treatment harvest at 30 DAS, WW, and WS treatment). Shoot dry weight data are still pending. Normalized transpiration dropped to about 50 - 60% of control at 12 days after stress imposition. Thereafter, NTR of RD 11 was above that of control JL 24. The total transpiration was also greater in several transgenics than in JL 24. For instance, transpiration of RD 19 during the 12 - 42 days after stress imposition was about 600 times higher than that in JL 24 (1788 g vs 2375 g water) under WS conditions. Under WW conditions, RD 19 used about 600 g more water (5933 g vs 5333 g). This showed that under water stress, RD 19 was able to take up relatively more water than JL 24. This trend was true in all transgenic plants. In fact, a remarkable finding of that work was that root dry weight of all 6 genotypes was within a very narrow margin under WW conditions (1.48 – 1.63 g, with JL 24 having 1.61 g). By contrast, under WS conditions, root dry weight of JL 24 remained unchanged (1.73 g) whereas it increased in all transgenics dramatically and reached a range of 2.27 – 2.65 g (30% increase). Under WS conditions, all transgenics had more profuse rooting in deep layers compared to JL 24, whereas under WW, there were no differences. There was a good correlation between the root dry weight within the 40 - 120 cm depth and the total transpiration ($r^2 = 0.41$). It was rather unexpected that DREB1A transgenic could have such a root-related response. This trial has shown the importance of understanding the kinetics of water uptake, more than the importance of knowing about the roots.

Evaluation of transgenic plants in culture chambers: There is a convergence of evidence from the literature and from our own work, in particular in groundnut, that there may be genotypic differences in the sensitivity of stomata to the vapor pressure deficit (VPD). That fact is getting documented in a “slow-wilting” genotype of soybean (PI 416937) that shows no further increase in transpiration at VPD level above 2 kPa, whereas other genotypes of soybean maintain a linear increase in transpiration with VPD level increasing above 2 kPa. Practically, such behavior would shutdown stomata during the hours of the day reaching the highest VPD, trading-off some loss of carbon fixation for water saving, and therefore, contributing to higher transpiration efficiency. Here the purpose of the experiment was to test whether a transgenic line having consistent higher TE than wild type JL 24 differed in stomatal sensitivity to VPD.

The plants were grown in the P2 facilities until about 30 days, and then transferred to growth chamber having different combinations of temperature and humidity, resulting in VPD of 0.63, 1.01, and 1.66 kPa. Plants were left for acclimatizing for 2 days, after which they were submitted to a dry-down experiment. Under WS conditions, there were no differences in TE between JL 24 and RD 2. By contrast, under WW conditions, TE was higher in RD 2 than in JL 24, across the 3 VPD environments.

Several tests to measure response to stepwise increase in VPD were conducted. A ladder of VPD conditions was set up throughout the day, with 60 min for each VPD value. Prior to that, the transpiration was measured gravimetrically from 9.00 am to 16.00 pm (by weighing pots every hour) to check any possibility of a diurnal effect. Data showed that transpiration was virtually constant throughout the day. Therefore, the response of transpiration to stepwise increase in VPD could be assessed without any data adjustment. The first ramp of VPD increases (0.95, 1.13, 1.35, 1.6, 1.88, 2.21, and 2.58 kPa) gave a strict linear increase in transpiration at each VPD interval, with no significant difference between the slopes of the two genotypes. Since plants in the natural environment face higher VPD, a ramp exploring higher values of VPD was also tested (0.95, 1.51, 2.02, 2.31, 2.64, 3.01, and 3.43 kPa). This ramp also gave a linear increase in transpiration in both genotypes, with no significant differences in the slope between the two genotypes. The same ramp carried out 5 days later also gave a linear increase in transpiration, until VPD was 2.31 kPa. At VPD values above 2.31 kPa, there seemed to be no increase in transpiration.

Vincent Vadez and KK Sharma

Progress reported towards the achievement of milestone for 2007 will contribute towards achievement of the milestones listed below.

Milestone: At least 8 promising transgenic events of groundnut containing DREB1A gene identified and their drought tolerance characterized under contained field conditions (KKS/VV/RA) 2008

Milestone: Three promising transgenic events of groundnut identified for drought tolerance and characterized under contained field conditions (KKS/VV/RA) 2009

Milestone: One or two transgenic events of groundnut used for introgression into locally adapted genotypes with better adaptation and the progeny characterized and evaluated (KKS/VV/SNN/RA) 2010

Milestone: 15 - 20 introgressed transgenic lines of groundnut with improved tolerance to water-limiting conditions evaluated and development of commercialization package initiated (KKS/VV/SNN/RA) 2011

Activity 6B.2.2: Mapping and marker-assisted breeding for salinity tolerance in groundnut

Milestone: At least 10 genotypes with superior salinity tolerance identified (VV/RA/SNN/RKV) 2007

Screening of 288 groundnut genotypes has been carried out for salinity tolerance. Over 6-fold range of variation for salinity tolerance was observed in this material. High yielding varieties (ICGV 87187, ICGV 86155, ICGV 00309, ICGV 93382, ICGV 97245, ICG 3027, ICG 76, ICG 5195, ICG 6892, and ICG 11651) would be used to screen the molecular diversity and most diverse parental genotype combinations will be used to develop mapping populations. Confirmation of the tolerance is under way.

Vincent Vadez

Milestone: Mapping populations between contrasting parents for salinity tolerance developed to identify QTLs (VV/RA/SNN/RKV) 2008

A set of contrasting parents for salinity tolerance based on the screening for pod yield under salinity has been given to the breeding group. Contrasting parents will be also assessed for their level of polymorphism, using a number of SSR markers. After confirmation of the contrast in salinity tolerance between the parents, and after ensuring that parents have sufficient level of polymorphism at the DNA level, the F_2 populations will be advanced by single-seed descent.

Vincent Vadez and R Aruna

Milestones: QTLs for salinity tolerance identified (VV/SNN/RA) 2011

Tolerant and sensitive groundnut genotypes for salinity tolerance have been dentified and crosses are currently being made.

V Vadez, SN Nigam and Aruna R

Output B. Annually knowledge of the improvements of the biotechnological and conventional tools designed to facilitate drought and salinity tolerance breeding and germplasm of mandate crops and associated capacity building made available to partners internationally

<u>Chickpea</u>

MTP Output Target 200: New Kabuli and desi germplasm with early maturity to avoid drought developed and made available to partners

Output target 6B.1: Identification of QTLs for drought avoidance root traits and salinity tolerance in chickpea

Activity 6B.1.1: Mapping and marker-assisted breeding for drought tolerance in chickpea

Milestones: Molecular markers for additional QTLs for drought avoidance root traits identified (PMG/JK/LK/RKV) 2007

Evaluation of new RIL populations for phenology, yield, and yield contributing traits: Two new RIL populations (ICC 283 x ICC 8261 and ICC 4958 x ICC 1882) developed for mapping of additional QTLs for drought avoidance root traits were evaluated for phenology, yield, and yield traits. A total of 281 F_8 RILs of ICC 283 x ICC 8261 and 264 F_8 RILs of ICC 4958 x ICC 1882 were evaluated in an augmented design along with the parental lines. Both populations exhibited a wide range of variability for all traits studied (Table 3). Over 120 RILs of ICC 283 x ICC 8261 gave 10 to 96% higher yield than the drought tolerant parent ICC 8261, while 25 RILs of ICC 4958 x ICC 1882 gave 10 to 26% higher yield than the higher yielding drought tolerant parent ICC 4958. A RIL population of ICC 4958 x ICC 1882 has been phenotyped using tall cylinder (120cm height) culture systems in 3 replications in 2005 post-rainy season. The substantial variation was observed in root length density, rot dry weight and rooting depth between the parental lines, and the RILs showed normal distribution on these root traits. In ICC 283 x ICC 8231, the phenotyping has been completed in 2006 post-rainy season, and analysis is going on. These populations are being phenotyped for root traits and genotyped using polymorphic SSR markers.

Table 3. Variability for different traits in two new RIL mapping populations developed for mapping of drought avoidance root QTLs.

	ICC 283 x ICC 8261 RIL population			ICC 4958 x ICC 1882 RIL population		
	ICC 283	ICC 8261	ICC 283 x ICC 8261 RILs	ICC 4958	ICC 1882	ICC 4958 x ICC 1882 RILs
Days to 50% flowering	60	58	45-88	50	54	42-66
Days to maturity	125	126	108-140	116	110	105-119
Plant height (cm)	35.0	56.0	30.0-64.0	43.7	39.3	26.0-56.3
Plant width (cm)	12.7	18.3	9.0-53.7	18.0	26.7	13.3-44.3
No, of primary branches per plant	2.7	3.3	1.7-6.7	2.3	3.0	1.3-4.3
No. of secondary branches per plant	2.0	3.0	1.0-11.7	3.0	4.7	1.3-6.3
No. of pods per plant	92	37	24-220	41	135	26-238
100-seed weight (g)	16.2	32.2	10.3-33.1	39.4	13.7	13.4-35.3
Biomass (kg ha^{-1})	1996	3013	1146-4413	3158	2650	479-4443
Seed yield (kg ha^{-1})	1036	1095	356-2148	1820	1453	270-2476
Harvest index (%)	51.9	36.3	24.6-60.0	57.6	54.8	24.6-66.2

PM Gaur and J Kashiwagi

Milestones: QTLs for drought avoidance root traits validated (PMG/JK/LK/RKV) 2009

This milestone is linked to the progress reported above, and one population will be used for mapping, and another for validation of QTLs.

Milestones: MABC-derived drought tolerant lines available from 2-3 locally adapted cultivars (PMG/JK/LK/RKV) 2011

Initiation of MABC for introgression of drought avoidance root traits in farmer-preferred cultivars: We identified three farmer-preferred cultivars, JG 11 in Desi type and Chefe (ICCV 92318) and KAK 2 (ICCV 92311) in Kabuli type for introgression of large root traits (high root length and high root depth density) from ICC 8261 (Kabuli) and ICC 4958 (Desi) using marker-assisted backcrossing (MABC). Six crosses (JG 11 X ICC 8261, KAK 2 X ICC 8261, Chefe X ICC 8261, JG 11 X ICC 4958, KAK 2 X ICC 4958, Chefe X ICC 4958) were made during 2006 in the greenhouse. The F_1s from these crosses are being grown during the post-rainy season 2006/07 along with parents for making backcrosses with the parental cultivars.

PM Gaur, J Kashiwagi and Rajeev Varshney

Activity 6B.1.2: Mapping and marker-assisted breeding for salinity tolerance in chickpea

Milestones: Phenotyping of ICCV 2 x JG 62 mapping population for salinity tolerance completed and data analyzed with available data for QTL mapping (VV/LK/RKV/PMG) 2007

The phenotyping of 125 F_{13} progenies from ICCV 2 x JG 62 cross has been completed under controlled saline conditions (saline treatment corresponding to an application of 1.870 L of a 80 mM NaCl solution to 7.5 kg of black soil from ICRISAT farm). The seed yield under salinity ranged from 4.05 g pot^{-1} to 14.5 g pot^{-1} in the 126 RILs, showing a 3-fold range of variation, and there was a very good segregation pattern. Parents ICCV2 (sensitive) and JG62 (tolerant) were at the extremes of the ranking, with seed yield being 7.77 g pot^{-1} (23^{rd} from bottom) and 13.46 g pot^{-1} (6^{th} ranked among all), respectively. QTL analysis is yet to be performed.

Milestone: Mechanisms of tolerance to salinity characterized (VV/LK/NM) 2008

Screening of 263 accessions of chickpea, including 211 accessions from ICRISAT's mini-core collection (10% of the core collection, and 1% of the entire collection), showed a six-fold range of variation for seed yield under salinity, with several genotypes yielding 20% more than the previously released salinity tolerant cultivar. The range of variation in yield under salinity was similar in both Kabuli and Desi chickpeas, indicating that breeding for salinity tolerance can be undertaken in both groups. A strong relationship was found between the seed yield under salinity and the seed yield under a non-saline control treatment, indicating that the seed yield under salinity was explained in part by a yield potential component, and in part by salinity tolerance *per se*. Seed yields under salinity were therefore computed to separate the yield potential component from the residuals that accounted for salinity tolerance *per se*. Among the genotypes evaluated, Desi genotypes showed greater salinity tolerance than the Kabuli genotypes. The residuals were highly correlated to the ratio of seed yield under salinity to that of the control, indicating that both parameters can be used to assess salinity tolerance. A similar ratio was calculated for shoot dry weight at 50 days after sowing. However, no significant correlation was found between the shoot dry weight ratio and the grain yield ratio, indicating that differences in salinity tolerance among genotypes could not be inferred from measurements at the vegetative stage. The major trait related to salinity tolerance was the ability to maintain a large number of filled pods, whereas seed size was similar in tolerant and sensitive genotypes. Salinity tolerance was also not related to the Na^+ or K^+ concentrations in the shoot.

Vincent Vadez and L Krishnamurthy

Milestone: New RILs populations for mapping of salinity tolerance QTLs developed (PMG/VV/LK) 2009

A set of 10 tolerant and 10 susceptible lines with salinity tolerance have been provided to the chickpea breeding group. Three crosses have been developed: ICC 6263 (DF 70) x ICC 1431 (DF 69), ICC 15802 (DF 66) x ICC 9942 (DF 63), and ICCV 2 (DF 39) x JG 11 (DF 40). These genotypes will also be assessed for their range of polymorphism at the DNA level, using a set of SSR markers**.**

PM Gaur, Vincent Vadez and L Krishnamurthy

Milestone: QTLs for salinity tolerance identified (VV/LK/RKV/PMG) 2009

The phenotyping of 125 F_{13} progenies from ICCV 2 x JG 62 cross has been completed under controlled saline conditions (saline treatment corresponding to an application of 1.870 L of a 80 mM NaCl solution to 7.5 kg of black soil from ICRISAT farm). Grain yield has been recorded at maturity. The seed yield under salinity ranged from 4.05 g pot^{-1} to 14.5 g pot^{-1} in the 126 RILs, showing a 3-fold range of variation, and there was a very good segregation pattern. Parents ICCV2 (sensitive) and JG62 (tolerant) were at the extremes of the ranking, with seed yield being 7.77 g pot^{-1} (23^{rd} from bottom) and 13.46 g pot^{-1} (6^{th} ranked among all), respectively. QTL analysis is yet to be performed.

Vincent Vadez and L Krishnamurthy

Milestone: QTLs for salinity tolerance introgressed in farmer-preferred varieties (PMG/VV/LK/RKV) 2011

The work on this milestone will start when RIL populations are available from the above milestone in 2009.

Output target 6B.2: High throughput molecular genetic and phenotyping platforms for drought and salinity stress and promising transgenic events of chickpea for tolerance to drought stress available for commercialization and introgression in locally adapted germplasm

Activity 6B.2.1: Develop and evaluate chickpea transgenic events for enhanced tolerance to drought stress

Milestones: 50 transgenic events of chickpea with DREB1A and P5CSF genes developed and screened for drought tolerance in the contained greenhouse (KKS/VV/PMG) 2007

Genetic engineering of chickpea for enhanced tolerance to water stress is being carried out using the osmoregulatory *P5CSF129A* gene and DREB1A transcription factor that acts as a major "switch" that triggers a cascade of genes in response to a given stress. Forty-eight chickpea events with 35S: P5CSF129A and 18 events carrying rd29A:DREB1A were advanced to T_4 generation to maintain the homozygosity. Southern analysis of the tested events indicated a low copy number (1 - 2 copies) in the 35S:P5CSF129A transgenics, whereas most of the events carrying rd29A: DREB1A had only a single copy of the transgene. Inheritance studies carried out on the T_3 generation transgenic plants showed a 15: 1 segregation for both types.

Ten transgenic events each of rd29A:DREB1A and 35S: P5CSF129A in T_3 generation were evaluated in drydown experiments to study various physiological parameters including plant response to soil drying as measured by the fraction of transpirable soil water (FTSW), stomatal conductance, and transpiration efficiency (TE). The events exhibiting a diversity of stress response patterns, especially with respect to the NTR-FTSW relationship were selected from that ranking and comparative studies were carried out with these transgenics under optimized conditions for evaluating the water use efficiency of the selected events. All the selected transgenic events differed from the wild type parent in their NTR response to FTSW, showing a decline in transpiration at lower FTSW values (dryer soil). Several events had superior transpiration efficiency, photosynthetic activity, stomatal conductance, and total transpiration under water limited conditions in comparison to the control parent C 235. All the selected transgenic events had a transpiration decline in drier soil than in the untransformed parent. The total biomass produced during the dry down cycle (Δ biomass) showed differences amongst the transgenic events, thus indicating apparent differences in the biomass produced per unit of water used.

A repeat experiment was performed with 5 transgenic events of DREB1A and 4 events of P5CSF. We measured the response of transpiration to progressive drying and also measured TE using standard protocol. Among the different events tested, one DREB1A event (RD 2) had higher TE than wild type C 235 under water deficit. This event was also among the few having higher TE than control variety C 235 in the previous experiment, which confirms its superiority. One event of P5CSF (P8) also showed superior TE than control C 235, whereas TE in the other 3 events was similar or below C 235.

Vincent Vadez and KK Sharma

Progress reported towards the achievement of milestone for 2007 will contribute towards achievement of the milestones listed below.

Milestone: At least 8 promising transgenic events of chickpea containing DREB1A or P5CSF genes identified and their drought tolerance characterized under contained greenhouse conditions (KKS/VV/PMG) 2008

Milestone: Three promising transgenic events of chickpea identified for drought tolerance and characterized under contained field conditions (KKS/VV/PMG) 2009

Milestone: One or two transgenic events of chickpea used for introgression into locally adapted genotypes with better adaptation and the progeny characterized and evaluated (KKS/VV/PMG) 2010

Milestone: 10 - 15 introgressed transgenic lines of chickpea with improved tolerance to water-limiting conditions evaluated and development of commercialization package initiated (KKS/VV/PMG) 2011

Pigeonpea

Output B. Annually knowledge of the improvements of the biotechnological and conventional tools designed to facilitate drought and salinity tolerance breeding and germplasm of mandate crops and associated capacity building made available to partners internationally

MTP Output Target 2006: New germplasm with early maturity to avoid drought developed and made available to partners

Output target 6B.1: High throughput molecular genetic and phenotyping platforms and promising transgenic events for salinity tolerance in pigeonpea available for commercialization and introgression in locally adapted germplasm

Activity 6B.1.1: Identify superior pigeonpea genotypes for salinity tolerance

Milestone: A set of pigeonpea genotypes suitable for breeding salinity tolerant breeding lines identified (VV/KBS) 2009

Identify superior pigeonpea genotypes for salinity tolerance: We assessed the morphological and physiological variation in pigeonpea for salinity tolerance in 300 genotypes, including the minicore collection of pigeonpea, wild accessions, and landraces from putatively saline prone areas worldwide. There was a large variation in the salinity susceptibility index (SSI) and the percent relative reduction (RR %) in both cultivated and wild accessions. The amount of Na^+ accumulation in shoot showed that more tolerant cultivated materials accumulated less Na in shoot. Such relationship was not true for the wild species. Wild species such as *C. acutifolius, C. cajanifolius,* and *C. lineata* were highly sensitive, whereas *C. platycarpus, C. scarabaeoides,* and *C. sericeus* were tolerant. It was interesting to note that *C. scarabaeoides* also provided a large range of sensitive materials. The minicore collection of pigeonpea provided a large range of variation for salinity tolerance. Among the tolerant genotypes, there were a large number of tolerant accessions originating from Bangladesh. Data were published in the 2nd issue of the electronic Journal of the Semi Arid Tropic Agriculture Research, and is available online at: www.icrisat.org. The methods and traits used to assess salinity tolerance in groundnut and pigeonpea have been put together in a paper submitted to the Indian Journal of Crop Science.

Vincent Vadez and KB Saxena

Activity 6B.1.2: Develop intra- and inter-specific mapping population of pigeonpea between contrasting materials for salinity tolerance

Milestone: At least two mapping populations developed to map QTLs for salinity tolerance in pigeonpea (VV/KBS/NM) 2009

Cajanus platycarpus, C. scarabaeoides, and *C. sericeus* were found to be tolerant and will be used for development of mapping populations. The minicore collection of pigeonpea provided a large range of variation for salinity tolerance. Contrasting parents in minicore would also be available for crossing and development of mapping populations.

Vincent Vadez and KB Saxena

Milestones: QTLs for salinity tolerance identified in pigeonpea (VV/KBS) 2011

The screening of breeding lines has revealed a large range of variation for salinity tolerance, both in the wild and the cultivated pigeonpea. Given the low level of polymorphism within cultivated pigeonpea, accessions are available to develop both intra- and inter-specific RIL populations.

Vincent Vadez and KB Saxena

Output C: Annually knowledge of the improvements of the biotechnological and conventional tools designed to facilitate bio-fortification and bio-detoxification, breeding improved germplasm, and management strategies of mandate crops and associated capacity building made available to partners internationally

MTP Output Target 2006: Variability of Fe and Zn in 80 germplasm lines developed and quantified in sorghum

Output target 6C.1: High yielding and micronutrient dense hybrids/improved populations/varieties of sorghum and millet, and promising transgenic events of groundnut and pigeonpea with high beta-carotene content available for testing in national trials

Activity 6C1.1: Develop groundnut transgenic events for enhanced production of beta-carotene

Milestone: 75 transgenic events of groundnut with maize psy1 gene developed and screened for high ß-carotene production in the contained greenhouse (KKS/SNN) 2007

Development of groundnut transgenics for enhanced β-carotene: Binary gene constructs harboring the ß-carotene biosynthetic genes, phytoene synthases (*psy*1 and *crt*B) from maize and *Erwinia herbicola,* respectively, were developed. These genes are driven by oleosin promoter for oil body seed specific expression, and are being used in *Agrobacterium*-mediated genetic transformation for the enhancement of ß-carotene in groundnut seeds. Over 70 putative transgenic groundnut plants were transferred to the containment greenhouse for seed development. The T_0 putative transgenic plants were analyzed for the integration of transgenes by using PCR with gene-specific primers, and Southern hybridization, with gene specific probes. In molecular analysis, 70% of putative groundnut transgenic plants showed the integration of *psy*1 gene. The transgene expressions were observed in the developing pods of groundnut by RT- PCR analysis. The presence of mRNA transcripts of *psy*1 gene was found in 3 out 4 plants tested. The T_1 seeds from transgenic events of PSY1 are being collected for further generations to study gene stability, inheritance, and expression of transgene. The procedure for beta-carotene extraction and analysis in groundnut seeds was optimized. The quantification of beta-carotene in primary transgenic groundnut seeds is in progress for the selection of best events. In a preliminary analysis, the enhancement of ß-carotene was observed in 2 out of 10 events tested that showed the levels of 10 $\mu g\ g^{-1}$ of the seed. For the development of marker-free transgenic plants, maize *psy*1 gene driven by oleosin promoter was sub-cloned into binary vectors pCAMBIA 2300:φ*npt*II (minus kanamycin gene). Around 30 marker-free putative groundnut transgenic plants carrying the m*psy*1 genes were transferred to the containment greenhouse. Molecular analysis is being carried out for these transgenic plants. Similarly, for the development of antibodies against phytoene synthase, the *psy*1 gene was cloned into pET expression vector. The over expression of PSY1 protein in *E. coli* is being analyzed.

A Vanamala, KK Sharma and SN Nigam

<u>Progress reported towards the achievement of milestone for 2007 will contribute towards achievement of the milestones listed below.</u>

Milestone: At least 8 promising transgenic events of groundnut containing maize psy1 gene identified and their stability characterized under contained greenhouse conditions (KKS/SNN) 2008

Milestone: Three promising transgenic events of groundnut identified for high ß-carotene production and characterized under contained field conditions (KKS/SNN) 2009

Milestone: One or two transgenic events of groundnut with high beta-carotene content used for introgression into locally adapted genotypes and the progeny characterized and evaluated (KKS/SNN/RA) 2010

Milestone: 5 - 7 introgressed transgenic lines of groundnut with improved beta-carotene content evaluated and development of commercialization package initiated (KKS/SNN/RA) 2011

Activity 6C1.2: Develop pigeonpea transgenic events for enhanced production of beta-carotene

Milestone: 50 transgenic events of pigeonpea with maize psy1 gene developed and screened for high ß-carotene production in the contained greenhouse (KKS/KBS/KML) 2007

Development of pigeonpea transgenic events for enhanced level of ß-carotene: *Agrobacterium*-mediated genetic transformation is being carried out regularly using the binary vectors containing maize *psy*1 gene driven by oleosin promoter for generating pigeonpea transgenic events with enhanced level of ß-carotene. The putative transgenic pigeonpea shoots obtained under antibiotic selection pressure are being elongated for rooting. About 40 putative transgenic plants with maize *psy*1 have been transferred to the containment greenhouse for further analysis. The primers specific to the coding sequence of *psy*1 genes were designed and conditions for PCR amplification were optimized. The primary T_0 putative pigeonpea plants are being molecularly analyzed using PCR for the presence of *psy*1 gene.

Development of pigeonpea transgenic events for enhanced level of methionine: In separate studies, binary gene constructs containing *SSA* gene driven by vicillin promoter (pHS723:SSA) in *Agrobacterium tumefaciens* strain C 58 is being used for the development of pigeonpea transgenics for enhanced level of seed methionine content. Around 20 putative transgenic plants were transferred to the containment greenhouse for further analysis. The primers specific to the coding sequence of *SSA* genes were designed and conditions for PCR amplification were optimized. The primary putative transgenic pigeonpea plants obtained under antibiotic selection pressure are being analyzed using PCR for the presence of *SSA* gene.

A Vanamala, KK Sharma and KB Saxena

Progress reported towards the achievement of milestone for 2007 will contribute towards achievement of the milestones listed below.

Milestone: At least 8 promising transgenic events of pigeonpea containing maize psy1 gene identified and their stability characterized under contained greenhouse conditions (KKS/KBS/KML) 2008

Milestone: Three promising transgenic events of pigeonpea identified for high ß-carotene production and characterized under contained field conditions (KKS/KBS/KML) 2009

Milestone: One or two transgenic events of pigeonpea with high beta-carotene content used for introgression into locally adapted genotypes and the progeny characterized and evaluated (KKS/KML) 2010

Milestone: 5 - 7 introgressed transgenic lines of pigeonpea with enhanced beta-carotene content evaluated and development of commercialization package initiated (KKS/KML) 2011

MTP Output Target 2006: 10 new groundnut varieties with resistance to aflatoxin contamination made available to partners

Output target 6C.2: Transgenic groundnut with enhanced resistance to *Aspergillus flavus* and aflatoxin production identified and available for introgression into regionally adapted germplasm

Activity 6C.2.1: Develop and evaluate groundnut transgenic plants for enhanced resistance to *Aspergillus flavus*

Milestone: Performance of the nine promising groundnut transgenic events expressing RChit gene for A. flavus resistance evaluated in contained on-station trials at ICRISAT, and best performing events identified (KKS/FW/PLK/SNN) 2007

Aspergillus flavus and *A. parasiticus*, with the ability to produce aflatoxins in groundnut, present a great human and animal health hazard globally. Extensive efforts for developing resistance to *A. flavus/A. parasiticus* infection and aflatoxin contamination in cultivated groundnut have resulted in the identification of partially resistant genotypes. Genetic engineering approach was therefore initiated to develop groundnut germplasm with durable resistance to *A.*

flavus/A. parasiticus invasion in groundnut. The rice chitinase (*RChi*) gene under the control of the *CaMV 35S* promoter was introduced into a popular groundnut variety JL 24, using *Agrobacterium tumefaciens*-mediated genetic transformation by using the cotyledon explants from mature seeds. Thirty transgenic events were selected that were positive for the *RChi* gene integration. These events were assessed for resistance against *A. flavus* seed colonization by *in vitro* seed inoculation with *A. flavus* spores. Fifteen *RChit* transgenic events that showed promising resistance during T_2 generation were evaluated for post-harvest seed resistance to *A. flavus* by *in vitro* seed inoculation assay by blotter plate method. Seeds (10 - 15% moisture) were surface sterilized with 0.1% (v/v) mercuric chloride and inoculated with *A. flavus* (strain AF 11-4) spore suspension (1×10^6 spores ml^{-1}) and placed on a moist blotter paper in a petridish, and incubated in a humid chamber maintained at 80% relative humidity and 28°C for six days. The percent seed infection was recorded at the end of 6^{th} day. Seeds of progenies with less than 10% infection were advanced to next generation by planting uninfected seed only. This procedure was continued up to T_5 generation. Post-harvest infection in the transgenic groundnut seed ranged between 0 to 100%, compared to 60 to 100% in controls. A few progenies of nine events numbers 12, 23, 24, 27, 29, 30, 31, 36, and 44 showed consistently low seed infection (<10%). Although progenies with less than 10% seed infection were advanced, seed from subsequent generation had infection between 0 to 100%. This variability could be due to the levels of *Rchit* expression and seed moisture at the time of infection. Seed from T_5 progenies of the nine selected events were bulked and these will be evaluated in an *A. flavus* sick plot in a contained on-station trial in 2007.

To assess resistance to *A. flavus* in germinating seed, three transgenic events (# 12, 23, and 24) and control (JL 24) seeds were pre-soaked for 24 h and 48 h in moist petridishes, and then inoculated with *A. flavus* spores. Observations were recorded after 6 days of inoculation. Infection in germinating seed was lower than un-germinated transgenic seed and controls. However, low-seed infection was also observed in germinating control seed, which may be due to the activation of native seed defense mechanism. Overall, the percent seed infection in germinating transgenic seed was always lower than germinating control seed, indicating likely action of *Rchit* gene product and native seed defense contributing to high resistance.

Further work is in progress to understand the gene expression, *Rchit* accumulation in seed, and *Rchit* activity in the seed at various moisture levels and its effect on *A. flavus* inhibition. The transgenic events will be evaluated for pre-harvest *A. flavus* infection following the regulatory approval for on-farm trials in the sick plot established at ICRISAT-Patancheru farm.

KK Sharma, Lava Kumar and Farid Waliyar

Progress reported towards the achievement of milestone for 2007 will contribute towards achievement of the milestones listed below.

Milestone: At least 75 transgenic events of groundnut containing the peanut lipoxygenase (PNLOX13S) gene developed and characterized for gene integration and expression (KKS/FW/PLK/SNN) 2007

Milestone: Ten promising transgenic events of groundnut with PNLOX13S identified for enhanced resistance to aflatoxin production under contained greenhouse conditions (KKS/FW/PLK/SNN) 2009

Milestone: Five promising transgenic groundnut events with PNLOX13S identified and disease resistance characterized under contained field conditions (KKS/FW/PLK/SNN) 2010

Milestone: Two best transgenic groundnut events with resistance to A. flavus used for introgression into locally adapted groundnut genotypes and their evaluation (KKS/FW/PLK/SNN) 2011

Output target 6C.3: Simple and cost-effect test for the estimation of mycotoxins (Aflatoxins, Fumonisins, and Ochratoxin-A) in crops and commodities, and aflatoxin-adducts in human serum developed and validated

Activity 6C.3.1: Develop a diagnostic test to determine the human exposure to aflatoxins

Milestone: Enzyme-linked immunosorbent assay (ELISA) assay developed for the estimation of aflatoxin adducts in human serum (FW/PLK) 2007

Development of ELISA for determining human exposure to aflatoxins: Aflatoxins are naturally produced food-borne metabolites of *Aspergillus flavus* and related fungi found in most of the food crops grown in tropics and sub-tropics. Aflatoxins constitute a group of four compounds, aflatoxin B1 (AfB1), B2, G1, and G2. Severe intoxication due to consumption of highly contaminated food results in acute liver damage and even death. Frequent exposure to sub-lethal doses leads to several nutritional and immunological consequences and greatly increases the risk of liver cancer. Moreover, research during the past two decades has established a synergistic interaction between Hepatitis B virus infection and AfB1 in causing liver cancer. Various studies suggest that the chronic type of aflatoxin poisoning is common in many parts of developing countries in Asia and Africa, and usually goes unnoticed. In order to assess the risk of human exposure to aflatoxins, a simple competitive enzyme-linked immunosorbent assay (ELISA) was developed. This test is based on the estimation of AfB_1-lysine, a metabolite of AB1, whose concentration in the blood albumin fraction has been shown to correlate with dietary aflatoxin intake over the previous 2 - 3 months and the level of DNA damage in the liver. The test involves isolation of the albumin fraction from blood, followed by digestion of albumin and estimation of AfB_1-lysine content by ELISA using AfB1-lysine polyclonal antibodies produced at ICRISAT. These polyclonal antibodies were raised against AFB1-lys-BSA adducts in a New Zealand White rabbit. In the ELISA test, polyclonal antibodies are first bound to the AfB_1-lysine present in the extracted albumin. Then, the antibody- AfB_1-lysine complex is detected using an alkaline phosphatase enzyme-labeled reporter antibody. Finally, the enzyme-labeled reporter antibody is detected using a colorimetric reaction that provides an estimate of the original AfB_1-lysine concentration. The current test can detect levels of AfB_1-lysine in blood as low as 5picogram per milligram (pg mg^{-1}) albumin. The test is simple to perform, inexpensive, and is effective for routine monitoring of human as well as animal samples for aflatoxin exposure.

The assay was used in a survey of 80 HBV positive blood samples from the Hyderabad region in India. The results revealed that almost 20% of the samples contained AfB_1-lysine in the range of 5 - 30 picogram mg^{-1} albumin, indicating that there is a high risk for liver cancer in Hepatitis B positive individuals. This simple test can be used to screen large numbers of samples and provides scope for preventive interventions in individuals at high risk of liver cancer. This test compliments commercial HBV testing, and the ELISA test we developed earlier for the detection of aflatoxins in foodstuffs. Both tests allow field studies to identify aflatoxin-exposed populations, determine the source of contaminated food, and initiate management approaches to limit dietary-aflatoxin exposure, thereby enhancing the food and human health safety and reducing the risk of hepatocellular carcinoma.
Lava Kumar/Farid Waliyar/CN Reddy

Activity 6C.3.2: Develop simple and cost-effective assays for the detection of mycotoxins in crops and commodities

Milestones: Simplified ELISA-based assay for the detection of mycotoxins developed (FW/PLK) 2007

Detection of food-borne mycotoxin contaminants is essential to ensure food safety. A simple and cost-effective competitive ELISA technique has been developed at ICRISAT for the detection of aflatoxins (AfB1, AfB2, AfM1, and total toxins), ochratoxin A ,and fumonisins; which are being widely used. However, these laboratory-based techniques require skilled technicians, and samples need to be sent to the laboratory for testing purpose. However, simple on-site mycotoxin detection methods would aid in testing large numbers of samples by non-experienced technicians in the fields, and rapid identification of contaminated lots, which can be segregated and sent for laboratory analysis for quantitative estimation of mycotoxins. For this purpose, we plan to develop plate-based, tube-based, or filter paper-based (lateral-flow) assays that are suitable for on-site testing. Diagnostic reagents (poly- and monoclonal antibodies, mycotoxin standards, and enzyme conjugates) already developed for the mycotoxin detection at ICRISAT will be used for the development of these tests. To achieve this objective, a simple ELISA

method is being developed to distinguish the variable concentration of aflatoxins (0, 2, 5, 10, and 25 ng ml^{-1}) in the samples based on color intensity (chromogenic reaction). ELISA plates were coated with 75 ng ml^{-1} of AFB1-BSA and the AFB1 standard was added at 2, 5, 10, and 25 ng ml^{-1}, and the anti-aflatoxin rabbit antisera was added at 1: 25 K and 1: 50 K, and the alkalinephosphatase enzyme-conjugated anti-rabbit antibody was used at 1:4 000 and the p-nitrophenylphosphate substrate was used at 0.5 mg ml^{-1}. Based on the color intensity in the wells of ELISA plate, samples with more than 5 ng ml^{-1} toxin can be distinguished in 30 min. This qualitative ELISA test will aid in rapid screening of samples to identify lots with more than 5 ng ml^{-1} toxin**.** This procedure is being refined further to develop tube-based and filter paper-based assays.

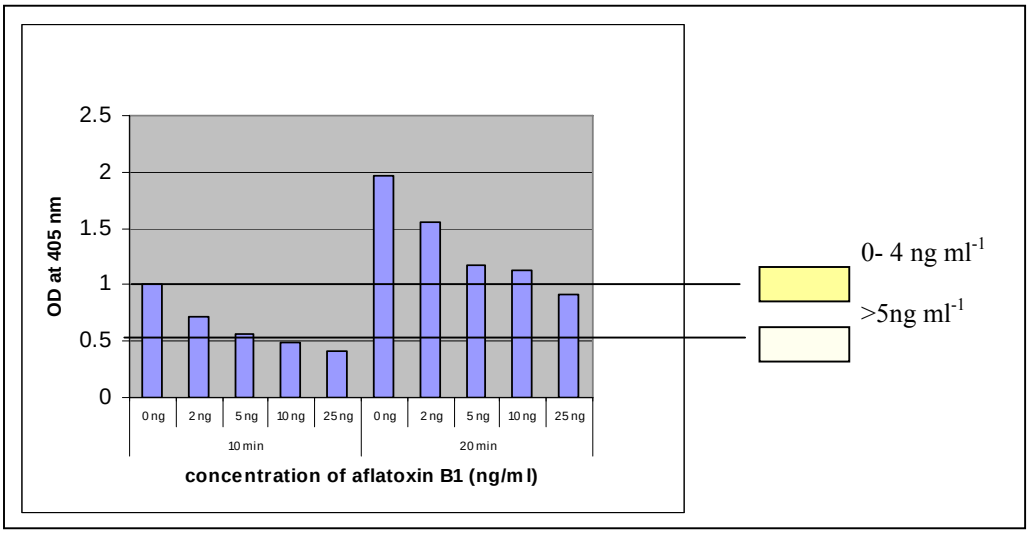

Fig. 1. Differentiation of aflatoxin contaminated lots based on color intensity in ELISA plate

Progress reported for 2007 for this activity will contribute to the milestones listed below.

Milestones: Tube/filter paper based semi-quantitative immuno assay developed for the on-site detection of aflatoxins (FW/PLK) 2008

Milestones: Multiplex filter paper immuno assay developed for the rapid estimation of aflatoxins and fumonisins (FW/PLK) 2009

Output target 6C 4: Aflatoxin resistant/tolerant groundnut genotypes identified

Activity 6C.4.1: Evaluate groundnut varieties for resistance to *Aspergillus flavus* and aflatoxin production by *in vitro* inoculation studies and on-station testing in sick fields

Milestone: At least 10 - 15 crosses involving diverse germplasm and breeding lines for aflatoxin resistant traits effected (SNN/RA/FW/PLK) Annual

Fifteen new crosses were made in the 2005/2006 post-rainy and the 2006 rainy seasons to develop aflatoxin tolerant breeding lines. The new germplasm lines used in the crossing are ICG 1859, ICG 1326, ICG 3267, ICG 10097, and ICG 3241.

SN Nigam and R Aruna

Milestone: 10 - 15 new high yielding, aflatoxin resistant lines identified and made available to NARS (RA/SNN/FW/PLK) Annual

We evaluated 349 advanced breeding lines (including controls) in 12 replicated trials, and 308 advanced breeding lines in 3 augmented trials for agronomic performance in the 2005/06 post-rainy and the 2006 rainy seasons under normal conditions. All the entries in replicated trials were also grown in an *A. flavus* sick plot to record observations on seed infection by *A. flavus* and aflatoxin production.

2005/2006 post-rainy season: In elite and advanced (Spanish) trials, none of the entries produced significantly higher pod yield than the highest yielding controls. In the elite trial (Virginia type), ICGV 01001 (2.5 t ha^{-1} pod yield, 0.3 % *A. flavus* seed infection, and 0.0 µg kg^{-1} aflatoxin production), ICGV 01002 (2.8 t ha^{-1}, 0.0 %, and 1.1 µg kg^{-1}), ICGV 01120 (3.2 t ha^{-1}, 0.3 %, and 0.0 µg kg^{-1}), and ICGV 02191 (3.0 t ha^{-1}, 0.0 %, and 0.0 µg kg^{-1}) were resistant to infection by *A. flavus* and aflatoxin contamination compared to the resistant control J 11 (2.9 ± 0.13 t ha^{-1}, 0.8 %, and 1.1 µg kg^{-1}). In another advanced trial (Spanish), ICGV 03301 (2.9 t ha^{-1}, 6.3 %, and 1.2 µg kg^{-1}) and ICGV 03328 (2.8 t ha^{-1}, 4.8 %, and 0.5 µg kg^{-1}) were more resistant than the resistant control J 11 (2.4 ± 0.13 t ha^{-1}, 2.5 %, and 222 µg kg^{-1}). In advanced trial involving Virginia types, the performance of ICGV 03363, ICGV 03372, and ICGV 03373 for pod yield and % infection by *A. flavus* (4.7 t ha^{-1}, 0.3 %, and 733.5 µg kg^{-1}; 4.4 t ha^{-1}, 3.0 %, and 21.3 µg kg^{-1}; and 4.9 t ha^{-1}, 0.8 %, and 148.0 µg kg^{-1}) was superior to both resistant J 11 (3.9 ± 0.13 t ha^{-1}, 1.8 %, and 4.3 µg kg^{-1}) and susceptible control, JL 24 (3.7 ± 0.13 t ha^{-1}, 1.0 %, and 5.2 µg kg^{-1}). However, none of these entries were significantly superior to the highest yielding control ICGS 76 (4.3 ± 0.13 t ha^{-1}, 1.8 %, and 9.7 µg kg^{-1}) for pod yield. In a preliminary aflatoxin trial, five lines (4.0 - 4.7 ± 0.68 t ha^{-1}) out-yielded the highest-yielding control ICGS 11 (3.1 t ha^{-1}). In augmented trial-1, ICGX 000109 (adjusted pod yield 4.9 t ha^{-1}) gave significantly higher yield than the highest yielding control ICGS 76 (4.9 t ha^{-1}). In augmented trial - 3, ICGX 000021 (5.2 t ha^{-1}) significantly out-yielded the highest yielding control ICGS 76 (5.0 t ha^{-1}).

2006 rainy season: None of the entries significantly out-yielded the best control in the elite trials (Spanish and Virginia). In the advanced trial - Spanish types, ICGV 04044 (3.9 t ha^{-1} pod yield, 64% shelling outturn, and 51 g 100-seed weight) and ICGV 04040 (3.9 t ha^{-1}, 65%, and 38 g) gave significantly higher pod yield than the highest yielding and resistant control ICGS 11 (2.3 t ha^{-1}, 66%, and 35 g). The best entry, ICGV 04044 came from (ICGV 97077 x ICGV 91284) cross. In the advanced trial - Virginia types, ICGV 06348, ICGV 06344, and ICGV 06345 (3.6 - 3.3 ± 0.33 t ha^{-1}) significantly out-yielded the highest yielding control ICGS 11 (2.3 t ha^{-1}, 60%, and 31 g). The best entry ICGV 06348 came from (ICGV 98077 x ICGV 91284) cross. In the Augmented trial, three entries significantly out yielded (adjusted pod yield 3.8 - 3.2 t ha^{-1}) the best check ICGS 76 (3.1 t ha^{-1}). The best entry in the trial was ICGVX 000109 (3.8 t ha^{-1}). Results of pre-harvest aflatoxin contamination and aflatoxin content are awaited. Six aflatoxin tolerant high yielding lines (ICGV 01060, ICGV 01094, ICGV 02171, ICGV 02184, 02206, and ICGV 02207) were identified during the 2005/2006 post-rainy season for inclusion in international trials.
SN Nigam/R Aruna

Milestone: Preliminary, advanced and elite foliar disease resistant breeding lines evaluated for resistance A. flavus and aflatoxin production under artificial inoculation conditions in the field and at least 5 resistant varieties identified for commercialization (FW/SNN/PLK/RA) 2009

Milestone: Ten interspecific derivatives of groundnut evaluated for A. flavus and aflatoxin resistance and promising lines identified (FW/NM/PLK) 2010

Sixty lines of advanced generation interspecific derivatives of groundnut generated using *Arachis duranensis*, *A. cardenasii* and *A. batizocoi* are being screened for *Aspergillus flavus* colonization and aflatoxin production. The results of the screening experiments are awaited.

Nalini Mallikarjuna

Milestone: Multilocational trials of five A. flavus resistant/low aflatoxin producing interspecific derivatives conducted in target locations in India (FW/NM/RA/PLK) 2012

Activity 6C.4.2: Evaluate various soil amendments and biocontrol agents for reducing pre-harvest *A. flavus*/ aflatoxin contamination in groundnut

Milestones: Efficacy of pseudomonas and actinomycets in preventing pre-harvest aflatoxin contamination determined (FW) 2008

Milestones: Integrated management package using various soil amendments and biocontrol agents, for preventing pre-harvest aflatoxin contamination developed (FW) 2009

Output targets 6C.5: Effective and eco- friendly IPM technologies designed, evaluated , and shared for the management of insect pests in legumes

Activity 6C.5.1: Impact of the village level bio-pesticide production on the effective implementation of IPM

Milestone: Impact of village level bio-pesticide production and utilization documented (GVRR) 2007

Production and utilization of HaNPV at village level: During 2006, emphasis was placed on establishing bio-pesticide units and imparting training on HaNPV production to farmers and extension officers. In this process, 27 HaNPV production units in India and 10 in Nepal have been established after extensive training of farmers and extension staff from each location. Through these interactions (on site training and village wide interactions), this project has influenced the farmers in judicious use of pesticides in plant protection, and the importance of use of protective clothing while carrying out the spraying. Farmers in these villages have adopted the concept of integrated pest management (IPM) and initiated the bio-pesticide production (range 1000 - 10,000 LE among the units), and using the product on different crops (chickpea, cotton, and vegetables) covering an area of about 10 – 50 ha under each unit.

GV Ranga Rao

Milestone: Rural stakeholders trained in biopesticide production and utilization and relevant rural enterprise initiated (OPR) 2009

Activity 6C.5.2: Develop technologies for production, storage, and utilization of entomopathogenic strains of NPV, bacteria, fungi and botanicals with insecticidal properties

Milestone: Virulent strains of entomopathogenic NPV, bacteria, fungi, and botanicals with insecticidal properties identified (GVRR/OPR/HCS) 2007

Evaluation of botanicals with insecticidal properties: Eleven indigenous plant materials (*Cleistanthus collinus*, *Calotropis gigantea*, *Pongamia glabra*, *Artemisia dubia*, *Sphaeranthus indicus*, *Cassia occidentalis*, *Chloroxylon swietensia*, *Vitex negundo*, *Madhuca indica*, *Strychnos nuxvomica*, and *Strychnos pototorum*) known for insecticidal properties, were collected from Andhra Pradesh and Chhattisgarh (India),and evaluated against tobacco caterpillar, *Spodoptera litura* larvae. The water extracts of these products tested against second/fourth instar larvae clearly indicated the superiority of *Cleistanthus collinus*, *Calotropis gigantia* (leaf extracts), and *Pongamia glabra* (seed extract) in suppressing the growth and development of *S. litura.* Though none of these plant extracts showed larval mortality, significant reduction in larval growth rate was observed, indicating the antifeedant/antibiotic properties of the extracts. Among the 11 plant materials tested, *Cleistanthus collinus*, *Pongamia glabra,* and *Calotropis gigantia* resulted in slower growth rates of 1.78, 1.96, and 2.07, respectively, compared to 2.74 in untreated control. This resulted in a reduction of 35, 28, and 24% in larval growth rate in comparison to the untreated larvae. Further studies will be carried out to make use of these products in future IPM programs.

GV Ranga Rao

Milestone: Botanicals with ability to kill insects having compatibility with entomopathogenic microorganisms identified and appropriate delivery systems developed (OPR/GVRR) 2008

Characterization of bacterial isolates for multiple traits: Ensuring a healthy crop is a first step towards protecting it from insect pests and diseases. Microorganisms can play a vital role in promoting plant growth, managing insect-pests, and maintaining soil health. Keeping this in view, bacteria from habitats such as composts and soil, having at least one beneficial trait and found highly promising in previous studies, were evaluated again to see if a given strain had more than one beneficial trait. Of the 14 isolates studied, four (HIB 67, SB 24, SB 26, and SRI 77) produced crystalline parasporal bodies, three (EB 13, BWB 21, and SB 21) were positive for siderophore production, two (EB 13 and SB 21) solubilized Rock Phosphate, eight (BCB19, BWB21, EB 13, HIB67, SB9, SB21, SB24, and SB26) were antagonistic to *Macrophomina phaseolina,* and two (BWB 21 and SRI 360) were compatible with *Metarrhizium anisopliae* - a fungus pathogenic to *Helicoverpa armigera*. In antagonistic studies with *M. phaseolina*, maximum zone of inhibition was recorded with EB 13 (17 mm diameter), followed by BCB 19 (12 mm). Range of inhibition zone was 5 - 17 mm diameter on culture medium. Insect killing ability of these isolates was studied by releasing *H. armigera* neonates on sprouted chickpea seeds inoculated with the bacterial isolates. Of the nine

isolates, maximum percent mortality was observed with SB 26 (66%), followed by BWB 21, SB 9, SB, 21, and BCB 19. Market sample of a Bt product from USA showed 74% kill. The plant growth promoting property of these isolates was studied on pearl millet variety ICM 155 by paper towel method. Four strains (SB9, CP8-3, HIB67, and SB21) enhanced plant growth at least on par with the reference strain of *Azotobacter* (HT54), which was 12.6% superior over the control. Based on the presence of multiple traits and their potential value in crop production, four bacterial isolates (SB9, SB21, BCB19, and BWB21) were selected for field studies for crop protection in 2006-07. Strain SB 26 did not get selected despite showing high mortality because it reduced plant growth over the untreated control.

OP Rupela

Microbial properties of cattle excrement and their fermentation products: Visits to fields of practitioners of certified organic farming (OF) reporting high yield and apparent high population of natural enemies of insect-pests prompted this study. Apparent concerns of policy makers and research managers on increased cost of crop production was to be addressed through the low-cost and biological inputs widely used by OF practitioners. A fermented broth called Amrit Paani (AP) or Jeevamrut was one such input. Microbial properties of AP and of cow dung, its major ingredients are presented here. Buffalo dung was included in the studies to learn differences in excrements of the two bovines. Fresh samples of excrement were collected aseptically and subjected to counting population of total bacteria, total fungi, total actinomycetes (indicators of soil health), P-solubilizers, *Pseudomonas fluorescens* (manager of soil borne diseases), siderophore producers (chellate iron and promote plant growth), and *Escheritia coli* (indicator of threat to human health). Data in Table 1 suggest that both cow and buffalo dung had similar population of total bacteria, total fungi, total actinomycetes, and *P. fluorescens*. Population size of P-solubilizers was undetectable in cow dung, but very high in buffalo dung ($\log_{10}$ 6.00 g^{-1} dry mass) and siderophore producers were absent in buffalo dung, but high in cow dung ($\log_{10}$ 4.99 g^{-1} dry mass). Microbiology of AP (applied to soils along with irrigation water and reported by farmers to improve crop growth) using excrements from the different bovines was quite revealing. We studied the population of different microorganisms at day 0 and day 3 under two fermentation conditions: a) flasks placed on shaker, and b) stationery culture. Data of day 3 indicated that microbial population, except actinomycetes and *P. fluorescens,* grew well in shake culture (Table 1). The counts were similar irrespective of the source of excrement. P-solubilizers in shake culture of AP of cow dung were about 10 times greater than that of buffalo dung. Noticeably, siderophore population was not detected even at the lowest dilution (Table 1) suggesting that this group of bacteria do not like aeration. Population of different microorganisms in stationary cultures was similar in the AP prepared using cow or buffalo dung. The striking difference was in the population of siderophores, which was high in the AP prepared using cow dung ($\log_{10}$ 3.73 mL^{-1}), while it was absent even at the lowest dilution in buffalo dung (Table 1).

Population of *E. coli* (which is considered a human health risk if present in consumables) was high in excrements of both cow and buffalo, and in the AP prepared using these. Since use of cow dung has been widely practiced in India for centuries for plastering floors of kitchens daily (when kitchens used to be of mud floor, practiced in rural areas even today), it is likely that Indian population is adapted to *E. coli*. Presence of high population of *E. coli* in human intestines (about one million per g excrement) is a normal phenomenon, where they are an important source of Vitamin K (absorbed through intestines). Rarely a strain of this bacterium is pathogenic to humans. However, their presence in food products is an indicator of human hygiene because disease-causing bacteria pass through excrements when a person is sick, and therefore, presence of *E. coli*, a safe bacterium – even for lab culture, suggests contamination from intestinal matter. Bacteria such as *P. fluorescens* have been reported inside plant tissue and provide induced systemic resistance (ISR) to plants in managing pests.

Overall, preparation and use of AP is a low-cost source of consortia of several agriculturally beneficial microorganisms. As there is no apparent health risk, its use can be promoted. This should reduce dependence of farmers on purchased inoculants. Previous studies have indicated that population of some beneficial microorganisms such as *Rhizobium* can be low in fields practicing OF, and therefore be purchased from market. The studies suggested that cow dung slurry if used directly will also provide different types of bacteria. But its use as AP allowed 10 times increase in total population of some key beneficial bacteria in the 3-day fermentation. A cow provides 8 to 10 kg dung (wet mass) per day, and if used as a source of beneficial microorganisms, one animal would be adequate (as a source of microorganisms through availability of about 1000 L Amrit Paani per day, round the year) for one ha. While if perceived as source of nutrients (N, P, and K) needed for crop growth without chemical fertilizers, it will be highly inadequate.

Table 1. Microbial population in cattle dung (log_{10} g^{-1} dry mass) and its fermented slurry (log_{10} mL^{-1}) (ICRISAT, Patancheru, 2006)

Microbial group	Dung		AP (shake)		AP (stationary)		SE
	C	B	C	B	C	B	
Bacteria	**6.78**	**6.94**	**7.44**	**7.60**	**7.28**	**7.44**	**0.146***
Fungi	**4.90**	**5.09**	**3.87**	**3.98**	**3.64**	**3.20**	**0.122*****
Actinomycetes	**5.99**	**5.65**	**<2.00**	**<2.00**	**3.43**	**4.10**	**0.460****
P. fluroscens	**<3.00**	**<3.00**	**<3.00**	**<3.00**	**4.67**	**4.67**	**0.139*****
P-solubilizers	**<3.00**	**6.00**	**5.87**	**4.77**	**6.07**	**6.02**	**0.557***
Siderophore+	**4.99**	**<2.00**	**<1.00**	**<1.00**	**3.73**	**<1.00**	**0.120*****
E. *coli*	**4.63**	**5.23**	**5.38**	**4.95**	**4.73**	**5.77**	**0.334NS**

AP = Amrit Paani or Jeevamrut - a fermented cow dung slurry widely used by practitioners of organic farming; C = Cow; B = Buffalo; *, **, * = Differences are statistically significant at *P* 0.05, 0.01, and 0.001, respectively.**

OP Rupela

Bioefficacy of selected botanicals and their compatibility: Botanicals and microorganisms are widely accepted as eco-friendly managers of insect-pests and diseases. Focus of this study was on materials readily available to farmers and at low cost, e.g., use of neem foliage instead of neem oil. Identification of botanicals that may enhance bioefficacy of microbial pathogens of insect-pests was another focus of the study. Fruits of neem, *Anona*, *Pongamia*, and *Jatropha* have been used for insect control. In previous studies, we noted that even foliage of neem and *Parthenium* had the ability to kill larvae of *Helicoverpa armigera.* A bioproduct involving stinging nettle (*Urtica diocia*) - a weed, has been used by farmers since 1924. In previous studies, *Bacillus subtilis* strain BCB 19 and *Metarrhizium anisopliae* have shown promise for pest management, including some disease-causing fungi. Individually, both the microorganisms were compatible with most botanicals, including neem and *Parthenium*. Compatibility of these with stinging nettle is reported here. Commercially available neem oil (Sunny Neem Extracts, Pvt. Ltd., Azadirachtin 0.03%) was used as reference. Neonates of *H. armigera* and *Spodoptera litura* were used as test insects. Use of neonates was preferred over third -instars (widely used while screening efficacy of synthetic pesticides) because focus of the low-cost biopesticides is on their prophylactic use. Active ingredients in the botanicals were extracted by adding 5% of dry mass in hot water, and the filtrate was evaluated.

Efficacy of neem foliage on *H. armigera* was similar (about 58%) to that noted in the past. *Parthenium* foliage affected only on *S. litura* (Table 2). *Anona* fruit rind, *Pongamia* cake and *Calotropis* foliage did not show any effect on *Helicoverpa*. Unlike in previous studies, neem fruit kernel (NFK) showed low activity (52% on *H. armigera*, and 38% on *S. litura*). Age of fruit and/or length of period between powder preparation and use (shelf-life) are suspected to be responsible for low activity, and will be studied in future. *Jatropha* cake showed maximum activity (84%) on *S. litura*. Stinging nettle was active against both the insects (60 and 66% mortality, respectively). Appreciable reduction in insect development was noted with stinging nettle and *Parthenium* foliage in case of *S. litura.* Problems in repeatability of bioefficacy of botanicals have been noted, and seem to be associated with shelf-life, and needs to be studied in future.

Table 2. Screening different botanicals for ability to manage insect-pests (ICRISAT, Patancheru, 2006)

Treatment[a]	**Mortality (%)**		**Mass change (%)**	
	HA	**SL**	**HA**	SL
Control	**30**	**16**	**NA**	**NA**
3% neem oil	**86[b]**	**88[b]**	**-53**	**-80[b]**
***Anona* fruit rind**	**26**	**58**	**39**	**-1**
Calotropis	**28**	**32**	**5**	**-41**
Neem	**58[b]**	**20**	**29**	**-16**
Neem fruit kernel	**52**	**38**	**-7**	**-18**
Parthenium	**32**	**60[b]**	**11**	**-58**
***Jatropha* cake**	**54**	**84[b]**	**-1**	**-5**
***Pongamia* cake**	**28**	**24**	**-15**	**-24**

Treatment[a]	Mortality (%)		Mass change (%)	
	HA	**SL**	**HA**	SL
Stinging nettle (SN)	**66[b]**	**60**	**2.2**	**-61**
BCB 19 + SN	**76[b]**	**NS**	**5.2**	**NA**
BCB 19	**66[b]**	**NS**	**-4**	**NA**
MA + SN	**74[b]**	**NS**	**-31**	**NA**
MA	**62[b]**	**NS**	**13**	**NA**
SE ±	**9.4**	**11.1**	**22**	**23.3**

HA = *Helicoverpa armigera*; SL = *Spodoptera litura.* a = Unless stated otherwise, all botanicals were extracts of foliage (leaves and twigs) dried in shade and powdered. b = Values are statistically significantly different from control at *P* 0.05.
OP Rupela

Milestone: Mass production techniques and stable formulations developed (GVRR/OPR) 2009

Activity 6C.5.3: Develop mechanisms to cope with pest outbreaks and management

Milestone: Chickpea pod borer, red hairy caterpillar, groundnut leaf miner, and Spodoptera prediction models validated (GVRR) 2007

The populations of the aforementioned insect species are being monitored at ICRISAT center using light and pheromone traps, and field scouting. The efforts to develop population prediction models are in progress, which will be validated in the near future.

GV Ranga Rao

Milestone: Tools for the pest identification and nature of damage for different crops developed (HCS/GVRR) 2008

Insect pests damaging the ICRISAT mandate crops are being diagnosed regularly at the ICRISAT research farms, and on the farmers fields to recommend appropriate control measures. The database is being updated regularly.

HC Sharma and GV Ranga Rao

Milestone: Potential kairomones for attracting Helicoverpa adults identified (GVRR/HCS) 2009

The relative attraction of *H. armigera* females to different host plants and their volatile compounds is being studied to identify chemical components that play a major role in host plant selection by the insects. The females of *H. armigera* prefer to lay eggs on pigeonpea than on cotton, indicating the possibility of using the later as a trap crop for pest management in cotton.

HC Sharma and GV Ranga Rao

Milestone: Tools for detection and quantification of NPV samples developed (GVRR/PLK) 2008

Serological and nucleic acid-based diagnostics tests for *Ha*NPV, *Sli*NPV, and *Ambl*NPV:
Nucleopolyhedroviruses (NPVs) are commonly used as biological pesticides to control insect pests over a wide array of agricultural crops on commercial basis under integrated pest management (IPM) programs. Many viral products are failing to meet acceptable standards because of poor inability to accurately assess the product quality. Therefore, it is necessary to have an efficient strategy for virus production and diagnostic tools for ensuring quality control of the NPVs: *Helicoverpa armigera nucleopolyhedrovirus* (*Ha*NPV), *Spodoptera litura nucleopolyhedrovirus* (*Sli*NPV), and *Amsacta albistriga nucleopolyhedrovirus* (*Ambl*NPV).

To compliment our ongoing NPV production activities, we produced polyclonal antibodies against polyhedra protein of *Ha*NPV, *Sli*NPV, and *Ambl*NPV infecting *H. armigera, S. litura,* and *A. albistriga*, respectively, to develop an enzyme-linked immunosorbent assay (ELISA) for diagnosis and quality control of the NPVs. Poly occlusion bodies (POBs) were purified from NPV infected insect cadavers and used for the purification of

polyhedrin protein by alkali treatment, followed by differential centrifugation and isoelectric focusing at 5.6 pH. The purified polyhedrin protein was used as antigen. Three New Zealand White inbred rabbits were used for immunization with 0.5 mg ml^{-1} purified polyhedrin protein of *Ha*NPV, *Sli*NPV, and *Ambl*NPV emulsified in Freund's complete adjuvant through intramuscular route. Four injections were given at weekly intervals. A week after 4th injection, the rabbit was bled for polyclonal antiserum at weekly intervals for 4 weeks, and then the animals were boosted with polyhedrin protein in Freund's incomplete adjuvant. The purity and titer of the antiserum was assessed by direct-antigen coating ELISA and western immunoblotting. The polyclonal antibodies did not react with the insect protein extracts reveling high specificity of the antibodies, and the antibody titer in various bleeds ranged between 1: 2,000 to 1: 40,000 for detecting respective polyhedrin proteins. In addition, the antibodies of each virus were cross-reacted with polyhedrins of other viruses in the study, indicating high conservation in polyhedrin protein among the three species of NPVs. The detection limits of antiserum in indirect ELISA was 20 ng of polyhedrin in case of *Ha*NPV and *Sli*NPV, whereas 8 ng in case of *Ambl*NPV. Further work is continuing towards quantification of polyhedrin for the early diagnosis of infection progress in the field, as well as quality control of NPV to accurately quantify the virus titer in the formulations.

In addition, three degenerate oligonucleotide primers (PgC, PgN, and RedPgC) were developed for the amplification of polyhedrin gene from by polymerase chain reaction (PCR). Using these primers, *Ha*NPV polyhedrin gene (~750 bp) was amplified by PCR and cloned into pUC18 vector. This sequence is being characterized, and the sequence information will be used to assess the diversity of *Ha*NPV isolates occurring in India. Attempts are also being made to develop oligomers for the amplification of hyper variable regions in *Ha*NPV genome for studying virus diversity.

Lava Kumar, GV Ranga Rao and Farid Waliyar

Milestone: Tools for the integrated pest management in different crops developed (HCS/GVRR) 2010

Development of insect pest diagnostic and management tools: Crop losses caused by biotic stresses are estimated to be in tens of billions of dollars across the world in spite of the huge investments in plant protection. Pest epidemics continue to occur causing severe hardships to poor farmers. We developed computer based insect pest diagnostic and management tools for all ICRISAT mandate crops to minimize crop losses and increase crop productivity without jeopardizing environmental safety. In this process, the E-learning systems developed in collaboration with Knowledge Management and Sharing unit on groundnut and chickpea pest diagnosis and management have been shared with NARS and updated. This is an effective extension tool for NARS extension staff and scientists.

GV Ranga Rao

Synergism of host plant resistance to *Helicoverpa* with insecticides its implications for ETLs in pigeonpea: We evaluated the effect of different protection regimes on *H. armigera*-resistant (ICPL 332) and susceptible (ICPL 87119) genotypes of pigeonpea to quantify the contribution of host plant resistance in *Helicoverpa* management. The plots were sprayed at the 10% flowering, 75% flowering, 50% podding, and dough stages in different combinations. There were three replications in a RCBD for each variety. Untreated plots served as a control. Percentage pod damage was 13.2 and 47.4% in the completely protected and unprotected plots of ICPL 87119, respectively. The protected plots of ICPL 332 had a pod damage of 7.0% compared to 37.6% in unprotected plots. The grain yield was 2491 kg ha^{-1} in completely protected plots of ICPL 87119 compared to 1950kg in the unprotected plots. The protected plots of ICPL 332 yielded 2539 kg ha^{-1} compared to 1811 kg ha^{-1} in the unprotected plots. The results clearly suggest the usefulness of combining insect-resistant varieties with insecticides for management of *H. armigera.*

Synergism of host plant resistance to *Helicoverpa* with insecticides its implications for ETLs in chickpea: We evaluated the effect of different protection regimes on *H. armigera*-resistant (ICC 506 and ICCV 10) and susceptible (ICC 3137 and ICCC 37) genotypes of chickpea to quantify the contribution of host plant resistance in management of *H. armigera*. The plots were sprayed at vegetative, flowering, and podding stages with methomyl. There were three replications in a factorial design. In the untreated control plots, there were 37.7, 86.7, 11.7, 32.3, and 26.0 larvae per 5 plants in Annigeri, ICC 3137, ICC 506, ICCC 37, and ICCV 10, and the pod damage was 15.3, 29.6, 5.2, 15.8, and 13.55%, respectively. Grain yield was 1431 to 1704 kg ha^{-1} in Annigeri, ICC 506, ICCV 10, and ICCC 37 compared to 926.1 kg in ICC 3137. In the plots that were protected at the vegetative and flowering stages, the pod damage was 0.6% in ICC 506, 8.3% in Annigeri, and 11.2% in ICCV 10 as compared to 60.5% in ICC 3137, suggesting that host plant resistance in combination insecticides is quite effective in minimizing the pod borer

damage. The plots that were protected at the vegetative, flowering, and podding stages had 1.7, 4.7, 1.3, 2.3, and 1.3 larvae per 5 plants, and suffered 9.5, 5.7, 0.1, 2.4, and 4.8% pod damage in Annigeri, ICC 3137, ICC 506, ICCC 37, and ICCV 10, respectively, indicating that three applications of insecticides at the critical stages provides a good protection against *H. armigera*. The grain yield was 1879kg ha^{-1} in ICCV 10, followed by 1924 kg in Annigeri, and 1663 kg in ICC 506 as compared to 1376 kg in case of ICC 3137. Further analysis is in progress to compute the economic thresholds for cultivars with different levels of resistance/susceptibility to *H. armigera.*

HC Sharma

Milestone: Tri-trophic interactions involving insects, host plants, and natural enemies for effective pest management studied (HCS) 2009

Identification of potential natural enemies of *Helicoverpa* in chickpea and pigeonpea eco-systems: Natural enemies of *H. armigera* have been collected from different eco-systems. Cultures of the parasitoids, *Campoletis chlorideae*, and *Cotesia* sp.; and the predators, *Cheilomenes sexmaculatus*, and *Chrysoperla carnea* have been established in the laboratory for further studies on bio-efficacy, and develop protocols for mass multiplication of these natural enemies for use in studies on effects of transgenics on non-target organisms, and biological control of *H. armigera*.

HC Sharma

Relative efficiency of *Campoletis chlorideae* to parasitize different insect hosts and *Helicoverpa armigera* larvae on different host plants: *Helicoverpa armigera* is a serious pest of cotton, grain legumes, and cereals. Complex intercropping systems and large scale deployment of *Bt*-transgenic crops with resistance to *H. armigera* have potential consequences for the development and survival of *C. chlorideae*. Therefore, we studied the tritrophic interactions of *C. chlorideae* involving eight insect host species and six host crops under laboratory conditions. The recovery of *H. armigera* larvae following release was greater on pigeonpea and chickpea as compared to cotton, groundnut, and pearl millet. The parasitism by *C. chlorideae* females was least with reduction in cocoon formation and adult emergence on *H. armigera* larvae released on chickpea. Host insects also had significant effect on the development and survival of *C. chlorideae*. The larval period of *C. chlorideae* was prolonged by 2 to 3 days on *Spodoptera exigua*, *Mythimna separata*, and *Achaea janata* as compared to *H. armigera*, *H. assulta*, and *S. litura*. Maximum cocoon formation and adult emergence were recorded on *H. armigera* (82.4 and 70.5%, respectively) than on other insect hosts. This information can be used to devise appropriate cropping systems to encourage the activity of natural enemies for biological control of insect pests.

MK Dhillon and HC Sharma

Output target 6C.6: New technologies evaluated, disseminated and their impact documented

Activity 6C.6.1: Exchange improved technologies and new knowledge with ARIs, NARs, NGOs, private sector and farmers' groups

Milestone: Pest management packages developed and disseminated through mass media, literature and e-learning (GVRR/OPR/HCS/CLLG/SNN/SP) Annual

E-learning system for chickpea and groundnut insect pest diagnosis and management strategies updated and shared with Indian NARS and ICARDA.

ICRISAT Participated in Doordarshan (TV) programs on legumes IPM with emphasis on bio-pesticide usage in plant protection.

ICRISAT Participated in an All India Radio (AIR) farmers' phone-in program covering ICRISAT's involvement in Agriculture Research for the betterment of Sat farmer's livelihoods.

ICRISAT Presented a topic on innovative use of bio-pesticides on the National Geographic channel.

G V Ranga Rao and OP Rupela

Exchange of knowledge and supply of trait-specific advanced breeding lines for evaluation for local adaptation (SNN/RA) Annual

Capacity building and knowledge exchange: Eight researchers from different countries (China - 2, India - 1, Nepal - 1, Philippines - 2, and Vietnam - 1) were trained in groundnut breeding and seed production technologies. Queries related to different aspects of groundnut cultivation from farmers and students were attended to on various occasions. One CD on ICGV 91114 in Anantapur district was prepared and information shared.

International trials and advanced breeding lines: We supplied trait-specific 20 sets of international trials and 144 advanced breeding lines and segregating populations to collaborators in Afghanistan, China, Eritrea, India, Nepal, Philippines, Uzbekistan, Vietnam, and Zambia.

Variety releases/ likely releases by the NARS

China: Groundnut variety Huayn 23 was released for cultivation in the Shandong province in China. It is derived from ICGS 37. Huayn 23 produced 13.5% more pod yield over the local control Luhua 12 at 22 locations in the province.

India: The Chief Minister of Andhra Pradesh, India, Dr. Y. S Rajashekhar Reddy, in a special function at ICRISAT, Patancheru, dedicated the drought-tolerant, early-maturing, high-yielding variety ICGV 91114 to Anantapur farmers. This variety was tested in farmer-participatory varietal selection trials for four consecutive years in drought prone locations in Anantapur, the largest groundnut growing district in India. Farmers liked the variety and accepted it as a possible replacement of a six-decade old variety TMV 2. This variety was released formally on 2 June 2006 by the State Varietal Release Committee of Andhra Pradesh.

ICGV 89290 - a Spanish variety with tolerance to foliar diseases and insect pests, has been identified by the All India Coordinated Research Program on Groundnut (AICRP-GN) for its release in Zone II, comprising Gujarat and Rajasthan states of India. This variety is already released as SG 99 for spring season cultivation in Punjab state of India.

ICGV 92195 - a short-duration groundnut variety, has been proposed by Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India, was released by the Central Varietal Release Committee as 'Pratap Mungphali–2' for zone II (Rajasthan and Gujarat) in India.

ICGV 93468 - a short-duration variety, is proposed for release as 'Avtar' in Uttar Pradesh, India for spring season cultivation. The variety is already popular with the farmers and was cultivated on 59,000 ha during the 2005 spring season.

AK 303 - a confectionery variety,, selected from an F_4 population ((ICGV 88384 x JL 24) x (ICGV 88438 x ICG 5240)), has been proposed by Mahatma Phule Krishi Viswavidyalaya, Jalgaon, for release in Maharashtra, India.

During the Annual Rabi/Summer Season Groundnut Workshop 2005/2006 of AICRP-GN, 21 new varieties were proposed for inclusion in the coordinated trials. Of these, 10 varieties had either ICRISAT supplied germplasm or breeding lines in their parentage or were direct introduction (ICGV 91114, ICGV 98281, ICGV 98223, ICGV 96110, ICGV 97045, ICGV 98396, and ICGV 98412). In addition to these, 47 other lines have been included in different state-level multi-location trials.

Nepal: Nepal Agricultural Research Council (NARC) released two high-yielding varieties ICGV 86300 (as Rajarshi) and ICGV 90173 (Baidehi) for general cultivation in Nepal.

Timor Leste: After one more year of on-farm trials, the Ministry of Agriculture intends to propose two groundnut varieties, ICGV 88438 and ICGV 95278, for release in the country.

Uzbekistan: ICGV 86155 - a short-duration variety, as 'Salomat;' and ICGV 94088 - a medium-duration variety, as 'Mumtoz,' have been released for cultivation in Uzbekistan. Salomat is recommended for planting as main crop and

also as double crop in Kaskkadarya and Surkhandarya provinces of Southern Uzbekistan. Mumtoz is recommended as a main crop throughout the country.

Philippines: The Ilagan Agricultural Research Station, Ilocos, has introduced ICGV 86564 - a large-seeded variety in the region. This variety has outperformed the local as well as improved varieties in the region, both, in terms of yield and seed size. It has been named as ASHA in India, which means 'Hope'. It has been introduced in the farmers' fields in the Northern province of the country, Ilagan. Farmers are impressed with its performance. It has been included in the state trials so that it gets released officially.

Ghana: The groundnut research in Asia has spill over benefits for Africa also. Recently, the Savanna Agricultural Research Institute, Ghana, released two groundnut varieties, which were developed at ICRISAT Center, Patancheru, India. ICGV 92099 - released as Gusie-Balin, is an early-maturing high-yielding variety with resistance to leaf spots. ICGV 90084 - released as Kpaniely, is a medium-duration high yielding variety with high oil content and resistance to leaf spots.

SN Nigam and R Aruna

Milestone: Breeder seed of improved varieties made available to NARS, NGOs, private sector, and farmer groups (SNN/RA) Annual

Breeder seed (25.5 t) of five varieties (ICGS 44, ICGS 76, ICGV 91114, ICGS 11, and ICGV 86564) was produced in the 2005/2006 post-rainy and the 2006 rainy seasons, and 21.6 t was distributed to different public and private sector seed producing agencies and farmers for further seed multiplication.

Milestone: Technical information and public awareness literature/documents developed and disseminated (Annual)

The 2006 issue of International *Arachis* Newsletter with 16 articles from 5 countries, and news and views items from different parts of the world was published.

SN Nigam

Activity 6C.6.2: Strengthen the NARS and farmers capacity in application of diagnostic tools and integrated aflatoxin management technologies

Milestone: Integrated aflatoxin management technologies disseminated thru farmer participatory trials and village level training courses (FW/SNN) Annual

On-farm evaluation of components of aflatoxin management in groundnut: Integrated aflatoxin management trials using compost, gypsum, and *Trichoderma viride* was conducted during the 2005 rainy season in 10 farmers' fields at threevillages in Pileru area of Chittoor district, Andhra Pradesh, India. In a 100^2m experimental plot, compost (5 t ha^{-1}) was incorporated in the soil after field preparation, *Trichoderma* (sand coated 100 kg ha^{-1}) was applied in the soil at sowing and gypsum (500 kg ha^{-1}) was applied at the flowering time. The plantings were carried out during the second fortnight of July using local variety TMV 2, which is highly susceptible to aflatoxin contamination. In Anantapur district, Andhra Pradesh, India, only *T. viride* was tested at Rekulakunta village on ten farmers' fields with a plot size 100^2m. The *Trichoderma* was applied adjacent to the rows one week after germination.

From each plot, about one kg pod sample was drawn and pods were sorted based on size into small and large pods. The *A. flavus* infection and aflatoxin levels ranged between 1 - 62% and 0 -5233 µg kg^{-1}, respectively. In bulk seed, 68% reduction in *A. flavus* seed infection was observed in plots treated with compost and gypsum, over the control, which had 11% seed infection. Similarly, in large seed lots, about 53% reduction in *A. flavus* was observed with compost, *Trichoderma* and compost + *Trichoderma* + gypsum treated plots. Corresponding aflatoxin in large seed lots showed that 97% reduction in toxin level with compost + *Trichoderma* + gypsum treatment, followed by compost (23%) and *Trichoderma* (14%) over 125 µg ha^{-1} in control. However, in small seed lots, reduction in *A. flavus* infection was low (21%) with a combination compost + *Trichoderma* + gypsum application. In small seed samples, highest reduction (72%) in toxin level was observed with *Trichoderma*, followed by compost + *Trichoderma* + gypsum (41%), and gypsum (33%) as against 133 µg ha^{-1} in control. Kernels from damaged pods showed very high levels (>510 µg ha^{-1}) of aflatoxin. *Trichoderm*a, gypsum, and compost + *Trichoderma* + gypsum

application showed 28 - 58% reduction in aflatoxin contamination over the control (1207 µg ha^{-1}). This trial was repeated in the 2006 rainy season on 10 farmers' fields (plot size 100^2m) in Cherlopalli village in Anantapur. The trial was harvested in Nov. 2006. Post-harvest sampling is being done for *A. flavus* infection and aflatoxin analysis, and the results are awaited.

Farid Waliyar

Low-cost agro-practices for management of groundnut aflatoxin via integrated management: To develop low cost options for the management of aflatoxins contamination in groundnut, a field trial was laid-out at ICRISAT-Patancheru during the 2006 rainy season. The trial comprised of 4 treatments (application of compost, gypsum and their combination, and untreated control), with susceptible genotype JL 24, and planted in 6 replications using randomized complete block design. Highly toxigenic strain (AF 11-4) of *A. flavus* multiplied on maize/sorghum seed was broadcasted in the field before sowing, followed by row application of inoculum at fortnightly intervals - starting from 25 days after sowing. Terminal drought was imposed 30 days before harvest to facilitate the seed infection and aflatoxin contamination. Harvesting at 110 days after sowing was done by up-rooting the plants and the produce dried under sunlight for 5 - 7 days before the pods were stripped. Post-harvest sampling for aflatoxin analysis is in progress and the results are awaited.

Farid Waliyar

Milestone: Training courses in mycotoxin detection technologies conducted for NARS (FW/PLK) Biennual

A training course in mycotoxin detection technologies for NARS will be conducted in 2007.

Farid Waliyar and P Lava Kumar

About ICRISAT®

The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) is a non-profit, non-political organization that does innovative agricultural research and capacity building for sustainable development with a wide array of partners across the globe. ICRISAT's mission is to help empower 600 million poor people to overcome hunger, poverty and a degraded environment in the dry tropics through better agriculture. ICRISAT belongs to the Alliance of Centers of the Consultative Group on International Agricultural Research (CGIAR).

Contact Information

ICRISAT-Patancheru (Headquarters)
Patancheru 502 324
Andhra Pradesh, India
Tel +91 40 30713071
Fax +91 40 30713074
icrisat@cgiar.org

Liaison Office
CG Centers Block
NASC Complex
Dev Prakash Shastri Marg
New Delhi 110 012, India
Tel +91 11 32472306 to 08
Fax +91 11 25841294

ICRISAT-Nairobi (Regional hub ESA)
PO Box 39063, Nairobi, Kenya
Tel +254 20 7224550
Fax +254 20 7224001
icrisat-nairobi@cgiar.org

ICRISAT-Niamey (Regional hub WCA)
BP 12404
Niamey, Niger (Via Paris)
Tel +227 20722529, 20722725
Fax +227 20734329
icrisatwc@cgiar.org

ICRISAT-Bamako
BP 320
Bamako, Mali
Tel +223 2223375
Fax +223 2228683
icrisat-w-mali@cgiar.org

ICRISAT-Bulawayo
Matopos Research Station
PO Box 776,
Bulawayo, Zimbabwe
Tel +263 83 8311 to 15
Fax +263 83 8253/8307
icrisatzw@cgiar.org

ICRISAT-Lilongwe
Chitedze Agricultural Research Station
PO Box 1096
Lilongwe, Malawi
Tel +265 1 707297/071/067/057
Fax +265 1 707298
icrisat-malawi@cgiar.org

ICRISAT-Maputo
c/o IIAM, Av. das FPLM No 2698
Caixa Postal 1906
Maputo, Mozambique
Tel +258 21 461657
Fax +258 21 461581
icrisatmoz@panintra.com

Visit us at www.icrisat.org

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