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ENHANCING UTILIZATION OF PLANT GENETIC RESOURCES IN CROP IMPROVEMENT

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ABSTRACT

Crop plant genetic resources (PGR) including landraces, old and new cultivars, mutant etc., are vital to crop improvement. These were used in research to develop improved cultivars that has resulted in increase of productivity and production considerably of various crops. The need for collecting and conserving germplasm was realized during 1960s, when there was threat of loss of landraces due to large adoption of improved varieties. Currently over six million-germplasm accessions are held in over 1300 genebanks across the world. This paper discusses assembly and management of genetic resources of sorghum, pearl millet, chickpea, pigeonpea, groundnut and six small millets at the Rajendra S Paroda Genebank at ICRISAT-Patancheru, India and means to further enhance their utilization for sustainable agriculture globally.

Various institutes and organizations worldwide have donated germplasm to the ICRISAT genebank. In addition, two hundred and thirteen germplasm collection missions were organized in 62 countries securing 33,194 germplasm accessions. The entire holding is over 118,800 accessions of the above crops from 130 countries. The germplasm accessions receive high priority for regeneration, characterization, conservation and distribution. The focus of research is on diversity assessment and on developing representative core, mini-core and composite collections to enhance utilization by the breeders. Molecular characterization of diverse germplasm sets is pursued for value addition and to enhance their utilization. Most of the accessions have been characterized. Germplasm seeds are conserved under very precise (cool and dry) conditions. Adequate seed of each accession is conserved to meet the requests of researchers and for posterity.

The ICRISAT genebank has been supplying over 21,000 germplasm samples annually to scientists across the countries. ICRISAT has restored crop germplasm to several countries including India. From the basic germplasm supplied from ICRISAT genebank, 66 varieties were released for cultivation in 44 countries.

Introduction

The wealth of plant genetic resources that includes landraces, old and new cultivars, genetic stocks, mutants etc., has contributed enormously towards achieving the global objectives of food security, poverty alleviation, environment protection and sustainable development. The value of genetic resources in developing superior crop cultivars is well recognized. The utilization of Norin 10 gene in wheat and Dee Geo Woo Gen in rice (sources of reducing plant height) have revolutionized the production of these crops globally. Wheat productivity increased by 137% and of rice by 93% in last 40 years due to the improved cultivars (Table 1), coupled with good agronomic management. Diverse genotypes were used in developing improved cultivars of soybean (resistance to diseases and insect-pests, tolerance to pod shattering, promiscuous nodulation and high yield; cf Dashiell and Fatokun, 1997) and groundnut (broadening genetic base, adding disease resistance and high yield; cf Singh and Nigam, 1997) that resulted in 93.2% productivity increase in soybean and 69.6% in groundnut in the last 40 years. Similarly, diverse germplasm sources having traits of

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short-duration, large seed size and disease resistance were used to develop new and high yielding cultivars of Chickpea (cf Singh *et al*., 1997) and pigeonpea (cf Remanandan and Singh, 1997).

The concern of PGR exploration and ex-situ conservation was not serious until 1960s. The development and spread of high yielding varieties of wheat and other crops by 1960s started replacing the local cultivars very rapidly leading to erosion of plant diversity. This loss of native crop landraces and cultivars prompted the international organizations such as the Food and Agriculture Organization (FAO) and the World Bank to create new institutional structures for the collection and preservation of valuable plant genetic resources in ex-situ genebanks. Since the last four decades, this program has achieved spectacular success. Over six million germplasm accessions have been collected and/or assembled in 1308 genebanks world over (FAO, 1998).

Created in 1971, the Consultative Group on International Agricultural Research (CGIAR) is an association of public and private members supporting a system of 15 Future Harvest Centers that work in more than 100 developing countries to achieve sustainable food security and reduce poverty through scientific research and development activities in the fields of agriculture, forestry, fisheries, policy and environment.

The CGIAR germplasm collections are a unique resource, available to all researchers. Germplasm contributions have helped lay the foundations of recovery by jumpstarting agricultural growth in countries emerging from conflict such as Afghanistan, Angola, Mozambique and Somalia.

The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), one of the 15 CGIAR centers, is responsible for germplasm assembly, characterization, conservation and distribution of germplasm of five mandate crops (sorghum, pearl millet, chickpea, pigeonpea and groundnut) and six small millets (finger millet, foxtail millet, kodo millet, little millet, proso millet and barnyard millet) and their wild relatives.

Germplasm Assembly in the ICRISAT Genebank

When ICRISAT was established in 1972, efforts were begun to assemble the germplasm of the mandate crops that existed with various research institutes in India and other countries. The Rockefeller Foundation had assembled over 16,000 sorghum germplasm accessions from major sorghum areas, and ICRISAT acquired 11,961 accessions of this collection in 1974 that existed in India and USA, besides 2000 pearl millet accessions. ICRISAT also obtained 2000 accessions of pearl millet collected by the Institut Francais de Recherché Scientifique pour le Development en Cooperation (ORSTOM) in francophone West Africa.

The germplasm material of chickpea and pigeonpea originally collected and assembled by the former Regional Pulse Improvement Project (RPIP), a joint project of the Indian Agricultural Research Institute (IARI), the United States Department of Agriculture (USDA) and Karaj Agricultural University in Iran, formed the initial collection. Sets of this germplasm, which were available in several agricultural research institutes in India and Iran, and at the USDA, were donated to ICRISAT in 1973. ICRISAT also acquired over 1,200 chickpea accessions from the Arid Lands Agricultural Development (ALAD) program in Lebanon. Similarly, much of the groundnut germplasm was received from the Indian groundnut research program, [now the National Research Center for Groundnut (NRCG), Junagadh], and USDA. Besides germplasm donations by the All India Coordinated Research Projects on various crops, considerable number of germplasm were received from agricultural universities at

Pantnagar (Uttranchal), Rajendranagar (Andhra Pradesh), Ludhiana (Punjab), Coimbatore (Tamil Nadu), Jabalpur (Madhya Pradesh), Rahuri (Maharashtra) and IARI at New Delhi. Fifteen Indian organizations that donated highest number of germplasm are listed in Table 2. Recently, in 2004-05, we obtained chickpea germplasm samples from Washington State University, Pullman, USA (2083 cultivated, 68 wild) and ICARDA, Syria (682 cultivated, 21 wild). We also received 622 groundnut germplasm samples from the National Institute of Agrobiological Sciences, Japan. Over 400 accessions of sorghum collected in Niger were received from our regional genebank in Niamey.

ICRISAT initiated activities to add new germplasm of its mandate crops from areas that were not adequately represented in the germplasm collection. Between 1975 and 2000, a total of 213 joint missions were launched in 62 countries, from which 33,194 accessions (sorghum 9011; pearl millet 10841; chickpea 4228, pigeonpea 3873, groundnut 2776; and small millets 2465) were collected. A large number of breeding lines or germplasm selections are developed and evaluated at important locations. The promising/improved germplasm lines were also registered in the genebank and conserved for future utilization. The genebank currently holds 118,833 accessions of which 73.8% have been conserved as base collection and 93.0% are designated with FAO (Table 3).

Germplasm Management

Phenotypic characterization and evaluation

Agronomic and morphological characterization is necessary to facilitate the utilization of germplasm. To achieve this, germplasm accessions of all the crops were sown in batches over the years and characterized for morphological and agronomic traits. Germplasm screening against biotic and abiotic stresses were conducted in collaboration with various disciplinary scientists. Grains were tested for nutritional value. Germplasm sets were evaluated over locations jointly with scientists in India, Nepal, Thailand, Indonesia, Ethiopia, Kenya and more intensively with the National Bureau of Plant Genetic Resources (NBPGR), India. The results of joint evaluations have led to a better understanding of the germplasm material.

Regeneration

Regeneration was carried out to meet the seed increase of (1) accessions that had reached a critical low level of seed stock or viability; (2) accessions required for medium-term storage (MTS; 5 ^{o}C, 25-30%RH) or long-term storage (LTS; -20 ^{o}C); and (3) germplasm repatriation, particularly to the NBPGR, India. Some of the germplasm accessions that do not produce seeds under ICRISAT-Patancheru climatic conditions (some wild *Arachis* species) are maintained vegetatively in the greenhouse. Some other accessions (wild *Cicer* species) need long day length and cool weather to grow and produce seeds. These species are also regenerated in greenhouse facilities.

Conservation

Germplasm conservation requires cleaning the seed material, drying to minimal seed moisture content, storing in cool and dry conditions and regular monitoring of seed health during storage. In the ICRISAT genebank, the seeds are stored in medium-term storage (MTS) in aluminium cans. A recent monitoring of the health of seed conserved for 10–25 years (MTS) indicated greater than 75% seed viability for majority of the accessions. Accessions with declining seed viability (less than 75% seed germination) are regenerated on priority and the old stock is replaced with fresh seeds. The germplasm accessions are also conserved in long-term storage (LTS) after packing in vacuum-sealed aluminium foil pouches. Before packing, the seeds are dried to about 5%

moisture content in a walk-in drying room (100 m^3 size; 15 °C and 15% RH) facility. At present, we have about 76% of the FAO designated germplasm in the LTS facility.

Documentation and supply of information

The vast germplasm data gathered on chickpea and pigeonpea germplasm has been summarized and presented to the users in the form of catalogs (Pundir *et al*., 1988; Remanandan *et al*., 1988). During the last 20 years, we had a very purposeful collaboration with NBPGR, India, on germplasm exploration, and evaluation at a number of locations, and results were published as 'Collaboration on Genetic Resources' (ICRISAT 1989). The data on joint germplasm evaluations were analyzed and published two catalogs each on forage sorghum germplasm (Mathur *et al*., 1991, 1992), and pearl millet (Mathur *et al*., 1993b and 1993c), and one on chickpea (Mathur *et al*., 1993a). Core and mini-core collections of ICRISAT mandate crops were established and the information was published for the benefit of fellow research workers. A Manual of Genebank Operations and Procedures was published (Rao and Bramel, 2000) documenting the procedures for germplasm acquisition, maintenance, documentation, conservation, and distribution. Existing procedures were reviewed and revised to maintain the collections according to international standards. A taxonomic key for the identification of wild species of the mandate crops has also been included in the manual.

Global germplasm supply to scientists and institutions

The ICRISAT genebank is holding germplasm that was donated by various institutes, organizations and farm communities and is ever willing to supply the same for research. From the beginning of our work (1973) until 2005, we have supplied 674,108 germplasm samples to scientists in 142 countries (Table 4).

Repatriation of germplasm to national programs

The global collections held at ICRISAT serve the purpose of restoration germplasm to the source countries when national collections are lost due to natural calamities, civil strife, etc. We supplied 362 sorghum accessions to Botswana; 1827 sorghum and 922 pearl millet to Cameroon; 1723 sorghum and 931 chickpea to Ethiopia; 838 sorghum and 332 pigeonpea to Kenya; 1436 and 445 sorghum accessions respectively to Nigeria and Somalia; and 71 pigeonpea accessions to Sri Lanka. The germplasm collection maintained in the ICRISAT genebank includes 44,822 accessions received from or jointly collected with the Indian National Programs. The National Bureau of Plant Genetic Resources (NBPGR), India requested ICRISAT for restoration of this germplasm. As part of ICAR/ICRISAT Partnership Projects, the genebank has repatriated almost full set of this germplasm by July 2004 (Table 5). Thus the NARS of several countries have regained their precious heritage which could have been lost if this was not conserved in the ICRISAT genebank.

Impact of germplasm supplied to NARS worldwide

Besides the utilization of germplasm in ongoing research at other institutes, 66 germplasm accessions (sorghum 30, pigeonpea 7, chickpea 19, groundnut 6, finger millet 2, and 1 each of pearl millet and barnyard millet) supplied from the ICRISAT genebank have been directly released as cultivars in 44 countries (Figure 1). Pigeonpea germplasm accession ICP 8863 collected from farmer's field in India was found very promising against fusarium wilt and was purified for the trait. The purified line was found high yielding and it was released for cultivation in 1986 as Maruthi in Karnataka state, India. This variety is also

grown on large hectarage in adjacent states, namely, Maharashtra and Andhra Pradesh (Bantilan and Joshi 1996).

A sorghum variety, Parbhani Moti was released in Maharashtra, India, in 2002. This variety is an excellent Maldandi-type [predominant postrainy (Rabi) sorghum landrace in Maharashtra and Karnataka states of India) with large lustrous grains and high yield. This was selected from a germplasm collection from Ghane Gaon, Sholapur, Maharashtra, made by ICRISAT genebank staff during 1989.

Another example is the release of barnyard variety (PRJ 1) in Uttranchal state during 2003. This variety yielded 45.4% higher grain yield compared to the check variety VL 29. It provides substantial fodder yield as well. This variety is a selection from ICRISAT germplasm collection IEC 542 that originated in Japan.

Present scenario of PGR utilization

Much progress has been in developing stable and high-yielding cultivars using diverse germplasm resources. This has resulted in area increase under some crops. During the last 40 years, area under soybean increased by 250.9%; pigeonpea: 60.7%; groundnut: 47.9% and rice: 22.4%. For other crops such as wheat and chickpea, area remained nearly unchanged. Productivity has improved considerably in most of the crops (Table 1). However, in future, there is much to be done to further improve productivity of the crops to meet the food requirement of ever increasing population.

A glance of ICRISAT genebank service to researchers revealed that on an average, 21,065 germplasm samples are supplied annually to users outside the ICRISAT (mean from 1974 to 2005). According to Marshall (1989), this figure indicates satisfactory germplasm distribution service of the genebank. However, the use of basic germplasm in breeding programs is scanty. For example, the summary of parental lines used in the ICRISAT groundnut-breeding program at ICRISAT (1986-2002) revealed that 986 unique parents were used in developing 8279 breeding lines, but this included only 132 unique germplasm accessions of groundnut and 10 of wild *Arachis* species. The two most often used cultivars were Robut 33-1 (3096 times) and Chico (1180 times). In the ICRISAT chickpea-breeding program (1978-2004), 12,887 parents (586 unique parents) were used in developing 3548 breeding lines, which included only 91 unique germplasm accessions of chickpea and five of wild *Cicer* species (Upadhyaya *et al*., 2006). The two most frequently used cultivars were L 550 (903 times) and K 850 (851 times). The data analysis from the Indian chickpea research program revealed that during 1967 - 2003, a total of 86 varieties was developed through hybridization that traced back to 95 unique parents. The top 10 parents contributed more than 35% to the genetic base of the released varieties. Most frequently used parents were Pb 7, IP 58, F 8, Rabat and S 26. About 41% varieties developed have Pb 7 as one of the parents in their pedigree (Kumar *et al*., 2004). There are similar reports from China (Jiang and Duan, 1998), and the USA (Knauft and Gorbet, 1989) in groundnut.

Strategies to enhance germplasm utilization

Assessment of diversity in the germplasm collection

The germplasm characterization and assessment of diversity is important to plant breeders for crop improvement and to genebank curators for efficient and effective management of their collection.

The chickpea germplasm collection (16,820 accessions) was characterized for seven morphological and 13 agronomic traits and reaction to fusarium wilt to determine phenotypic variation in different geographical regions. The means for different agronomic traits differed significantly between regions.

The variances for all the traits among regions were heterogeneous. South Asia region contained the largest range of variation for all the traits. The Shannon-Weaver (Shannon and Weaver, 1949) diversity index (H') was variable in different regions for different traits. Analysis revealed the need to secure more germplasm collections from Mediterranean countries and Ethiopia. Cluster analysis delineated two regional clusters consisting Africa and South and Southeast Asia in the first cluster; and the Americas, Europe, West Asia, Mediterranean and East Asia in the second cluster (Upadhyaya, 2003) (Figure 2). An earlier study of chickpea germplasm data at ICRISAT (Pundir *et al*., 1988) revealed that in general, Indian accessions were highest yielding and the accessions from Chile had higher plant height and greater seed mass. The accessions from Spain and Syria had longer flowering duration and the accessions from Greece and Russia had erect growth habit. Resistance to fusarium wilt was more common in accessions from Bangladesh than from other countries.

The groundnut germplasm collection (13,342 accessions) was characterized for 16 morphological and 10 agronomic traits in two seasons to determine the phenotypic variation in different geographical regions. The means for different agronomic traits differed significantly among regions. The variances for all the traits among regions were heterogeneous. South America, which showed 100% range variation for 12 of the 16 morphological traits, also revealed highest range variation. From South America among regions, primary seed color among morphological traits and leaflet length among agronomic traits showed highest pooled H'. Three of the six botanical varieties, aequatoriana, hirsuta, and peruviana were poorly represented indicating the need to be collected. PCA using 38 traits and clustering on first seven PC scores delineated three regional clusters; consisting North America, Middle East, and East Asia in the first Cluster, South America in the second cluster, and West Africa, Europe, Central Africa, South Asia, Oceania, Southern Africa, Eastern Africa in second cluster and Southeast and Central Asia and the Caribbean in the third cluster (Upadhyaya *et al*., 2002b) (Figure 3).

The pigeonpea germplasm collection (11,402 accessions from 54 countries grouped into 11 regions) was analyzed for patterns of variation for 14 qualitative and 12 quantitative traits. Semi-spreading growth habit, green stem color, indeterminate flowering pattern, and yellow flower color were predominant among qualitative traits. Primary seed color had maximum variability and orange color, followed by cream were the two most frequent seed colors in the collection. Variances for all the traits were heterogeneous among regions. The germplasm accessions from Oceania were conspicuous by short growth duration, short height, fewer branches, pods with fewer seeds, smaller seed size, and lower seed yields. The accessions from Africa were of longer duration, taller, with multiseeded pods, and larger seeds. The germplasm diversity, indicated by H' pooled over all traits, was highest for Africa and lowest for Oceania. The cluster analysis delineated three clusters: cluster 1 includes accessions from Oceania; cluster 2 from India and adjacent countries, and cluster 3 from Indonesia, Thailand, The Philippines, Europe, Africa, America and the Caribbean countries. Pigeonpea-rich countries such as Myanmar, Uganda, and others like Bahamas, Burundi, Comoros, Haiti, and Panama are not adequately represented in the collection, and need priority attention for germplasm exploration (Upadhyaya *et al*., 2005c).

Developing core collections

One of the reasons that plant breeders are using less basic germplasm in research is the lack of information on traits of economic importance, which often shows high genotype x environment interactions and requires

replicated multilocational evaluations. Evaluation is very costly and resource-demanding task owing to the large size of the germplasm collections. To overcome this, our research now focuses on studying the diversity of germplasm collection and developing "core collections," which are about 10% of the entire collection, but represent almost full diversity of the species. From the germplasm collection in the ICRISAT genebank, we have already developed core collection of sorghum (2,247 accessions, Grenier *et al*., 2001); pearl millet (1,600 accessions, Bhattacharjee, 2000); chickpea (1,956 accessions, Upadhyaya *et al*., 2001a); groundnut (1,704 accessions, Upadhyaya *et al*., 2003); groundnut Asia core (504 accessions, Upadhyaya *et al*. 2001c); pigeonpea (1,290 accessions, Reddy *et al*., 2005); finger millet (622 accessions, Upadhyaya *et al*., 2005a) and foxtail millet (155 accessions, Upadhyaya – unpublished data) (Table 6).

Developing mini-core collection

When the size of the entire collection is very large, even a core collection size becomes unwieldy for evaluation by breeders. To overcome this, ICRISAT scientists developed a seminal two-stage strategy to develop a mini-core collection, which consists of 10% accessions in the core collection (and hence only 1% of the entire collection) (Upadhyaya and Ortiz, 2001). This mini-core collection still represents the diversity of the entire core collection. The first stage involves developing a representative core collection (about 10%) from the entire collection using all the available information on origin, geographical distribution, and characterization and evaluation data of accessions. The second stage involves evaluation of the core collection for various morphological, agronomic, and quality traits, and selecting a further subset of about 10% accessions from the core collection. At both stages standard clustering procedures should be used to form groups (clusters) of similar accessions and then select desired number of accessions from each cluster. At ICRISAT, we have already developed mini-core collections of chickpea consisting of 211 accessions (Upadhyaya and Ortiz, 2001), groundnut (184 accessions) (Upadhyaya *et al*., 2002a), pigeonpea (146 accessions), and finger millet (65 accessions) (Upadhyaya – unpublished data) (Table 6).

Developing composite collection

The revolution in molecular biology, bioinformatics, and information technology has provided the scientific community with tremendous opportunities for solving some of the world's most serious agricultural and food security issues, and has led to the formation of Generation Challenge Program (GCP) entitled "Unlocking Genetic Diversity in Crops for the Resource-Poor (www.generationcp.org)". The GCP is designed to utilize molecular tools and comparative biology to explore and exploit the valuable genetic diversity existing in germplasm collections held at the CGIAR and NARS genebanks, with particular focus on drought tolerance. In recent years, several studies conducted on plants have detected DNA markers associated with ecology, geography, disease resistance, and quantitative traits (Thornsberry *et al*., 2001; Turpeinen *et al*., 2001; Ivandic *et al*., 2002, 2003; Russel *et al*., 2003; Sun *et al*., 2001, 2003; Gebhardt *et al*., 2004; Sabharwal *et al*., 2004; and Amirul Islam *et al*. 2004) demonstrating that it is a viable alternative to classical QTL analyses, which were time taking and costly measurements.

ICRISAT and collaborating institutes have constituted composite collections of chickpea (Upadhyaya *et al*., 2006a) and sorghum (3000 accessions each) and groundnut, pigeonpea, finger millet (1000 accessions each) (Table 7) that contain maximum diversity known in the species, accessions with economic traits and some representation of the related wild species. The composite collections will be genotyped using SSR markers. The data generated will

be used to define the genetic structure of the collection for functional and comparative genomics. The analysis of genetic diversity will help to elucidate population structures that influence the analysis of the associations between molecular markers and the morphological or reaction traits. Using all available information, about 10% accessions will be selected containing maximum diversity and those could be used in the breeding programs.

Identification of new sources for traits of economic importance for use in crop improvement program

Due to the reduced size, the core collection can be evaluated extensively to identify the useful parents for crop improvement. By evaluating core collection of chickpea, we identified new sources of important traits, namely, early maturity (28 accessions), large seeded kabuli (16 accessions) and high-yielding (39 accessions) types. The clustering of 28 early maturing accessions along with four controls revealed three clusters. Cluster-1 was formed of five entries including three controls (ICCVs 2, 96029 and Harigantars). Cluster-2 was formed of 14 entries including control Annigeri. Thirteen entries constituted cluster-3 and no control among them. It can be presumed that these 13 accessions are more distant from controls than other accessions (Figure 4). The phenotypic diversity index was highest between ICC 14648 and ICCV 96029, compared to the other entry pairs. Such information has high value to chickpea breeders.

The evaluation of groundnut core collection resulted in identification of 21 accessions with early maturity (Upadhyaya *et al*., 2005c). The cluster analysis done on these 21 accessions and three controls revealed three clusters. Cluster-1 comprised of four entries including two controls (Gangapuri and Chico). Cluster-2 contained 13 entries including one control (JL-24). Seven test accessions formed cluster-3 and these accessions are more distinct from the three controls used in this study (Figure 5). In the groundnut core, 158 accessions had low temperature tolerance at germination (Upadhyaya *et al*., 2001b). Also found were 15 Valencia, 20 Spanish, and 25 Virginia type germplasm lines in groundnut with high yield, good shelling percentage and 100-seed weight through multilocational evaluation of the 'Asia region core collection' (Upadhyaya *et al*., 2005b). These new sources performed better than or similar to the best control cultivars for particular trait (s), but were diverse from them. Holbrook *et al*. (1997) achieved similarly through examining all accessions in the groundnut core collection (Holbrook *et al*., 1993) for resistance to the groundnut root-knot nematode (*Meloidogyne arenaria* (Neal) race 1) and resistance to pre-harvest aflatoxin contamination (PAC) (Holbrook, 1998) while Franke *et al*. (1999) later did similarly for resistance to Rhizoctonia limb rot (*Rhizoctonia solani* Kuhn AG-4).

The mini-core collections of chickpea and groundnut have been evaluated and diverse sources of useful traits were identified. From the chickpea mini-core, 18 accessions having traits related to drought tolerance (Kashiwagi *et al*., 2005) and 29 accessions tolerant to soil salinity (Serraj *et al*., 2004) have been identified. Similarly, Pande *et al*. (2006) screened the mini-core collection for resistance to various diseases and identified 67 accessions resistant/highly resistant to fusarium wilt, moderate resistance to ascochyta blight in 3 accessions, botrytis grey mold in 55 accessions, and to dry root rot in 6 accessions. Some accessions also with multiple resistances were identified. The evaluation of groundnut mini-core resulted in identification of 18 diverse accessions with high water use efficiency (Upadhyaya, 2005). The evaluation of chickpea mini-core at the Indian Institute of Pulses Research (IIPR), Kanpur, India during 2002 to 2004 seasons revealed 12 very promising accessions. Of these six accessions

were involved in hybridization to develop large seeded kabuli cultivars. The evaluation of groundnut mini-core in Thailand (2004-05) indicated ten accessions high-yielding. The groundnut mini-core evaluation in China during 2005 resulted in identification of 14 accessions highly resistant to bacterial wilt, six with high oil content and four with high Oleic and low Linoleic acid. Three accessions had highest Oleic: Linoleic acid ratio.

Molecular characterization of germplasm

Characterization of germplasm with molecular markers can help improve their utilization. It can form the basis for mining and cloning of genes of agronomically important traits.

Genotyping chickpea accessions

A total of 288 chickpea accessions including 211 mini-core subset accessions consisting of 75% desi type (Upadhyaya and Ortiz, 2001), 57 accessions of kabuli chickpea, and 20 accessions of wild *Cicer* species from ICARDA were genotyped using 40 SSR markers. The results indicated that the chickpea mini-core developed at ICRISAT was allelically more diverse than the germplasm from ICARDA. The accessions from ICARDA consisted of more heterozygous individuals compared with mini-core accessions. The dendogram constructed based on shared allele distance using unweighted pair group mean average (UPGMA) method indicated two main groups: one consisting mainly of accessions from the Indian subcontinent and the other group of accessions from Mediterranean, Middle-East and Ethiopia. The accessions of wild species (*C. reticulatum* and *C. echinospermum)* formed two groups of their own flanking two ends of the chickpea accessions (Upadhyaya *et al*., 2006b).

Validating the chickpea mini core collection:

Discriminant function analysis was used to determine the level of congruence between the genotypic data set and the 28 phenotypic clusters of the chickpea mini-core (Upadhyaya and Ortiz, 2001) based on morphological and agronomic traits. For DFA analysis, genotypic data from 210 accessions screened with 40 SSR markers was used. Overall most individuals were assigned with a high degree of confidence to the original (phenotypic) clusters from which accessions constituting mini core collection were selected. Only 27% of the individuals were re-assigned into new clusters according to genotypic data, which were mainly identified within clusters 4, 6, and 7 of the mini-core (ICRISAT, 2004). This confirmed that the chickpea mini core was well selected.

Genotyping chickpea accessions of varying maturity duration

Sixty-two chickpea germplasm accessions (50 early-, 6 medium- and 6 late-maturing) were analyzed with 37 SSR markers. A total of 673 alleles were found. The number of alleles per marker varied from 4 to 28 with an average of 18. The polymorphic information content (PIC) values ranged from 0.53 to 0.94 with an average of 0.85. Mean heterozygosity was low (0.0276). The principal component analysis (PCA) plot of Rogers's distance indicated three distinct clusters (ICRISAT, 2004).

Genotyping groundnut accessions

In groundnut, 26-accessions were analyzed with random amplified polymorphic DNA (RAPD) assays. The genetic similarity (S_{ij}) ranged from 59.0 to 98.8% with an average of 86.2%. Both multidimensional scaling and unweighted pair-group method with arithmetic averages (UPGMA) dendograms revealed the existence of five distinct clusters. Some accessions with diverse DNA profiles (ICGs 1448, 7101, and 1471, and ICGVs 99006 and 99014) were identified for mapping and genetic enhancement in groundnut (Dwivedi *et al*., 2001). Molecular marker based diversity estimates are useful to select diverse lines for developing populations that may be used for

mapping studies to identify DNA markers linked with resistance to rosette disease in groundnut. Nine amplified fragment length polymorphism (AFLP) using primer pairs were performed on nine rosette resistant and one susceptible accessions. Across the 10 accessions, the nine primer pairs identified 94 unique markers, with an average of 10.4 markers per primer pair. The genetic dissimilarity (D_{ij}) values ranged from 3.92 to 50.53% with an average of 19.56%. Groundnut accessions, namely, ICG 11044 with ICGs 3436, 9558 and 11968 showed greater genetic diversity (36.59 to 50.53%) amongst the nine rosette resistant accessions used. These accessions possess high levels of resistance to rosette, average d"2% compared to e"90% infection in susceptible control ICG 7827 across four seasons' evaluation at Lilongwe, Malawi. These accessions therefore could be inter-crossed among themselves to produce diversified rosette resistant breeding populations (Dwivedi *et al*., 2003).

Conclusion

Crop genetic resources have contributed enormously towards sustainability of agriculture and alleviation of poverty. These are being assembled and conserved at several genebanks for future use. Using raw germplasm resources, a large number of crop varieties and hybrids have been developed and released for cultivation. New strategies on core and mini-core collections were developed to enhance the precision of germplasm characterization and reducing cost on germplasm regeneration and conservation. Composite sets of ICRISAT mandate crops are being developed under the Generation Challenge Program. Phenotypic and genotypic characterization of these sets will provide vast scope of identifying useful and unique germplasm resources for utilization in crop improvement. Molecular characterization of the germplasm of agronomic importance has been pursued for value addition and to enhance their utilization.

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Table 1. Area under cultivation and productivity of the selected crops during last four decades[1]

Crop	1963-65	1983-85	2003-05
Area: m ha			
Wheat	213.2	230.4	213.1
Rice (Paddy)	123.5	143.8	151.2
Soybean	25.3	51.7	88.8
Sorghum	47.3	47.7	43.8
Chickpea	11.7	9.8	10.6
Groundnut in shell	16.9	18.4	25.0
Pigeonpea	2.8	3.5	4.5
Grain yield kg ha^{-1}			
Wheat	1196	2173	2841
Rice (Paddy)	2062	3201	3976
Soybean	1172	1747	2265
Sorghum	970	1466	1328
Chickpea	577	682	780
Groundnut in shell	853	1089	1447
Pigeonpea	632	750	708

Table 2. Institutions in India that donated a large number of germplasm to ICRISAT, 1973–2003.

Institution	Sorghum	Pearl millet	Chick pea	Pigeonpea	Groundnut	Small millets	Total
AICSIP, Hyderabad	175	-	-	-	-	-	**175**
AICRPO, Hyderabad	-	-	-	-	529	-	**529**
ANGRAU, Hyderabad	115	-	-	3,035	1,366	285	**4,801**
ARS, Niphad, Maharashtra	-	-	345	-	-	-	**345**
GAU, Junagadh	-	66	-	-	1,167	-	**1,233**
GBPUAT, Pantnagar	-	155	96	-	-	-	**251**
HAU, Hisar	-	-	211	-	-	-	**211**
IARI, New Delhi	33	-	3,022	174	-	-	**3,229**
JNKVV, Jabalpur	-	164	127	479	-	-	**770**
MPKV, Rahuri	-	234	173	191	267	-	**865**
NBPGR, New Delhi	90	170	149	-	161	469	**1,039**
PAU, Ludhiana	-	106	1,029	-	496	-	**1,631**
RAU, Samastipur, Bihar	-	-	-	-	197	-	**197**
TNAU, Coimbatore	13	45	63	40	590	531	**1,282**
Rockefeller Foundation (India)	11,370	2,022	-	-	-	1,246	**14,638**
Total	11,796	2,962	5,215	3,919	4,773	2,531	**31,196**

Table 3. Germplasm holdings in the Rajendra S Paroda Genebank, ICRISAT, Patancheru, December 2004.

Crop	Active collection[1]	Base collection[2]	Accessions held in-trust[3]
Sorghum	37,257	31,669	35,836
Pearl millet	21,594	15,150	21,329
Chickpea	20,116	15,984	16,970
Pigeonpea	13,632	10,266	12,712
Groundnut	16,041	6,820	14,419
Finger millet	5,949	4,620	4,979
Foxtail millet	1,535	1,054	1,535
Proso millet	842	576	835
Little millet	466	384	462
Kodo millet	658	630	656
Barnyard millet	743	487	743
Total	118, 883	87,640	110,476

1. Active collection: germplasm seeds stored in medium-term storage facility and available for current utilization.

2. Base collection: germplasm seeds stored in long-term storage facility for utilization in posterity.

3. Accessions held in-trust: FAO designated germplasm freely available for use to the researchers.

Table 4. Global distribution of germplasm samples to scientists, 1974 - 2005

Crop	1974-83	1984-1993	1994-2005	Total
Sorghum	58,627	158,762	31,382	248,771
Pearl millet	15,302	62,769	11,536	89,607
Chickpea	52,015	45,413	24,893	122,321
Pigeonpea	19,546	30,593	16,278	66,417
Groundnut	20,908	44,034	29,182	94,124
Small millets	20,067	17,352	15,449	52,868
Total	186,465	358,923	128,720	674,108

Table 5. Restoration of basic germplasm from ICRISAT genebank to different countries

	Number of accessions						
Country	**Sorghum**	**Pearl millet**	**Chickpea**	**Pigeonpea**	**Groundnut**	**Small millets**	**Total**
Botswana	362						362
Cameroon	1,827	922					2,749
Ethiopia	1,723		931				2,654
Kenya	838			332			1,170
Nigeria	1,436						1,436
Somalia	445						445
Sri Lanka				71			71
India	14,637	7,189	7,488	5,988	6,060	3,460	44,822

Table 6. Core and mini -core collections of ICRISAT mandate crops.

Crop	Number of accessions used	Number of traits involved	Number of accessions
Core			
Sorghum	22,473	20	2,247
Pearl millet	16,063	11	1,600
Chickpea	16,991	13	1,956
Pigeonpea	12,153	14	1,290
Groundnut	14,310	14	1,704
Finger millet	5,940	14	622
Foxtail millet	1,474	13	155
Asian core			
Groundnut	4,738	15	504
Mini-core			
Groundnut	1,704	31	184
Chickpea	1,956	22	211
Pigeonpea	1,290	16	146
Finger millet	622	14	65
Foxtail millet	155	13	-

Table 7. Composite collections of seected crops

Crop	Size of the composite collection (accessions)	Genetic markers used	Institutes collaborating with ICRISAT
Chickpea	3000	50 SSR markers	ICARDA, Syria
Sorghum	3000	50 SSR markers	CIRAD, FranceCAAS, China
Groundnut	1000	20 SSR markers	EMBRAPA, Brazil
Pigeonpea	1000	20 SSR markers	Only ICRISAT
Finger millet	1000	20 SSR markers	AICSMIP, India

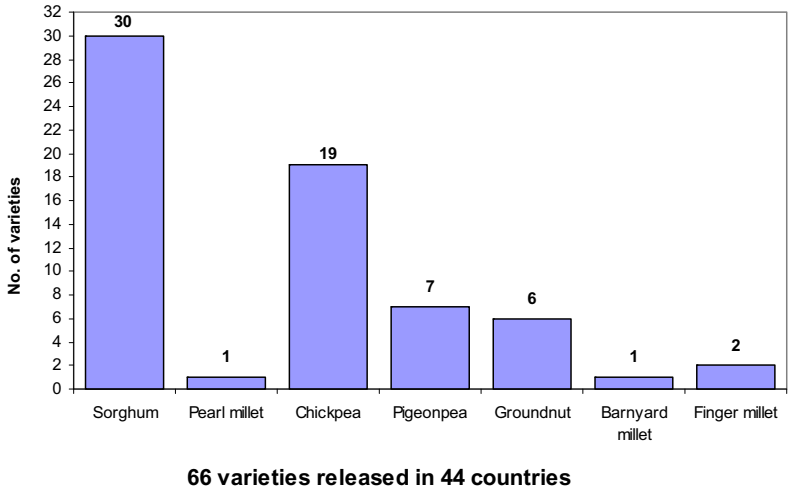

Fig 1. Number of cultivars released worldwide from the basic germplasm supplied from ICRISAT genebank 1976-2003

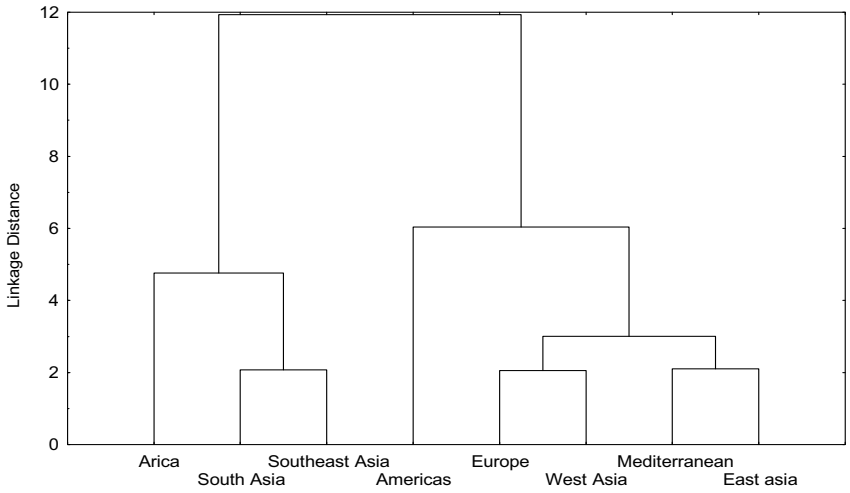

Fig 2. Dendogram of eight regions for the entire chickpea germplasm based on first three principal components.

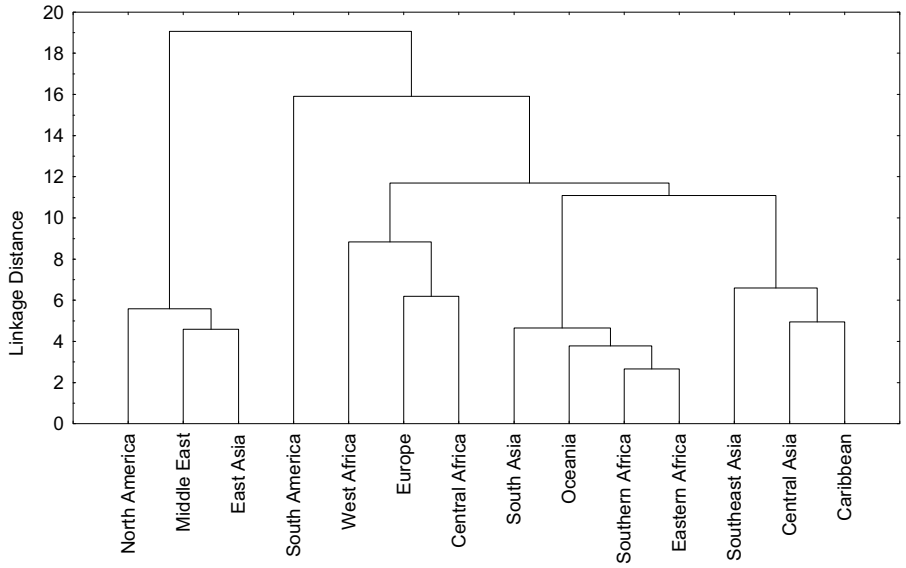

Fig 3. Dendogram of 14 regions in entire groundnut germplasm based on sores of first seven principal components.

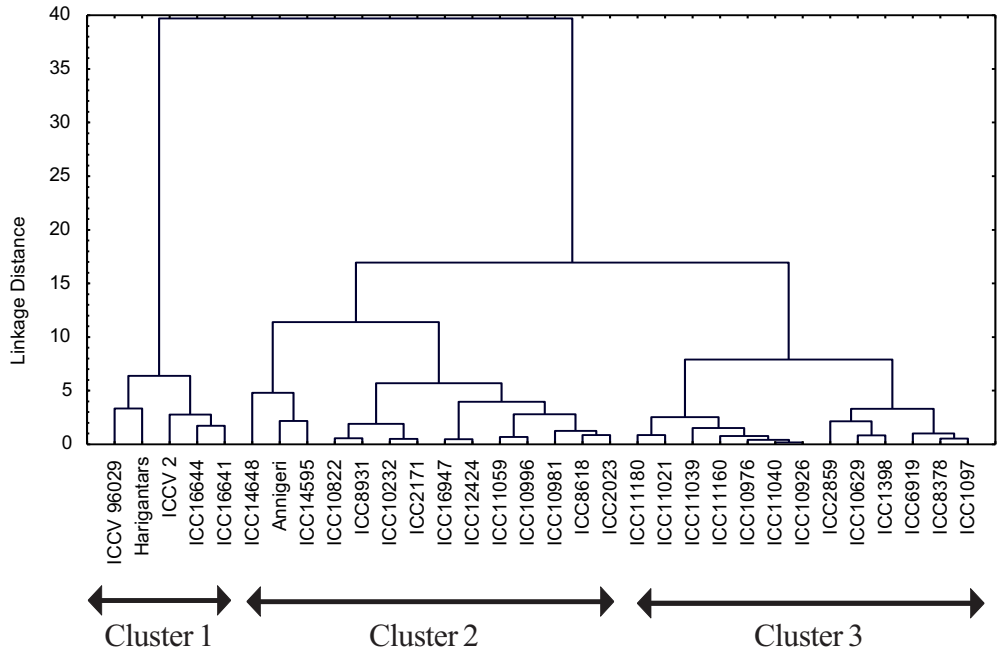

Fig 4. Dendogram based on first three principal components of 16 quantitative traits of 28 early-maturing germplasm lines and four control cultivars capturing (74.3%) variation.

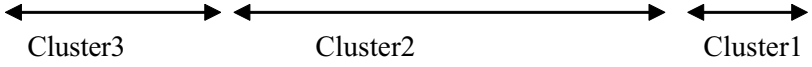

Fig 5. Dendogram of 21 early maturing groundnut landraces with three control varieties based on the first 10 principal components