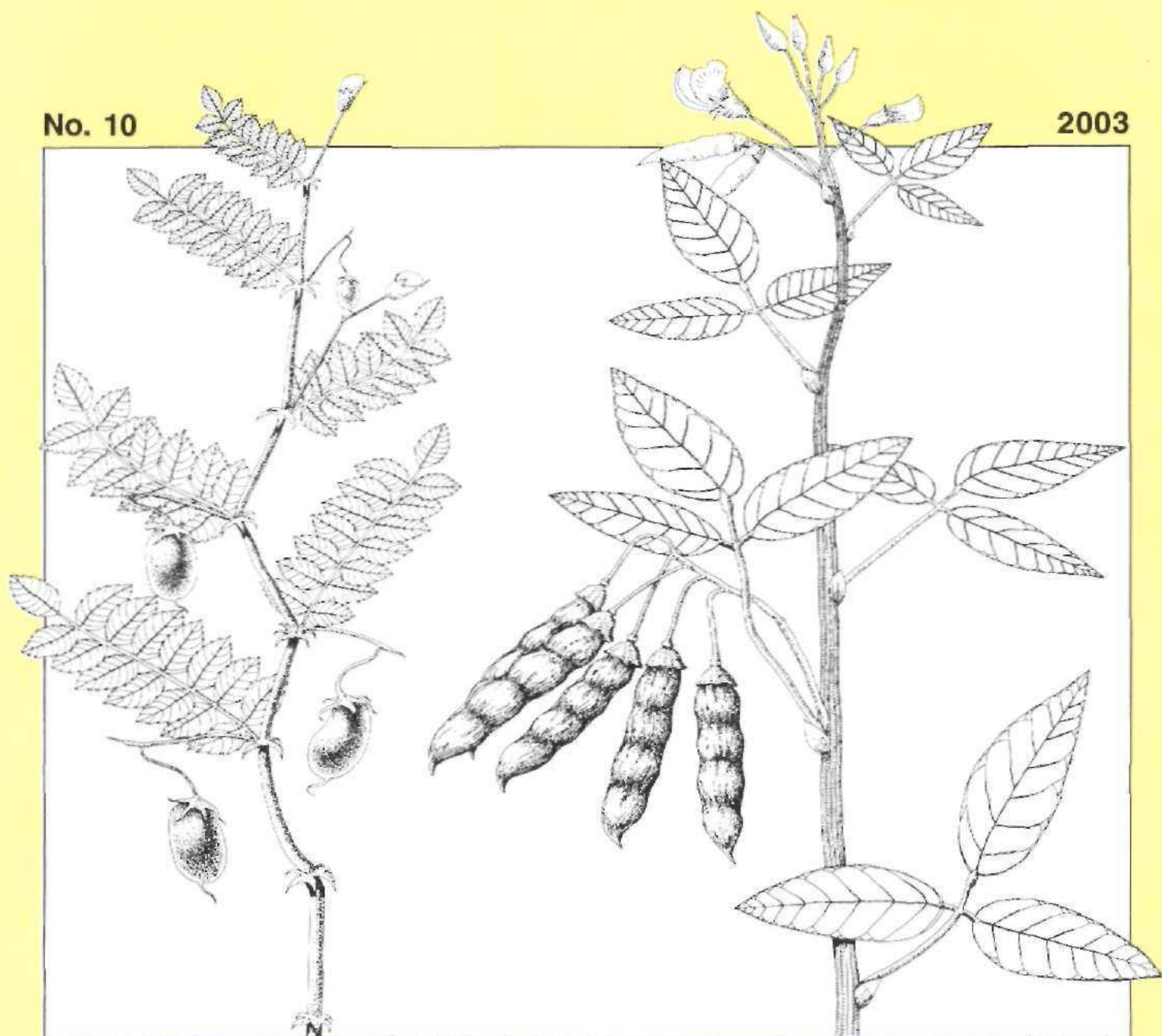




# International Chickpea and Pigeonpea Newsletter

No. 10

2003



# International Chickpea and Pigeonpea Newsletter

## Publishing objectives

The International Chickpea and Pigeonpea Newsletter (ICPN) is published annually by ICRISAT. It is intended as a worldwide communication link for all those who are interested in the research and development of chickpea (*Cicer arietinum* L.), and pigeonpea (*Cajanus cajan* (L.) Millsp.) and their wild relatives. Though the contributions that appear in ICPN are peer-reviewed and edited, it is expected that the work reported will be developed further and formally published later in refereed journals. It is assumed that contributions in ICPN will not be cited unless no alternative reference is available.

ICPN welcomes short contributions (not exceeding 600 words) about matters of interest to its readers.

## What to contribute?

Send us the kind of information you would like to see in ICPN.

- **Contributions should be current, scholarly, and their inclusion well-justified on the grounds of new information.**
- Results of recently concluded experiments, newly released varieties, recent additions to germplasm collections, etc.
- Genome maps and information on probe-availability and sequences, and populations synthesized for specific traits being mapped. Glossy black and white prints of maps should be included, if possible. Partial maps can also be submitted.
- Short reports of workshops, conferences, symposia, field days, meetings, tours, surveys, network activities, and recently launched or concluded projects.
- Details of recent publications, with full bibliographic information and 'mini reviews' whenever possible.
- Personal news (new appointments, awards, promotions, change of address, etc.)

## How to format contributions?

- Keep the items brief - remember, ICPN is a newsletter and not a primary journal. About 600 words is the upper limit (no more than two double-spaced pages). **As the newsletter is devoted to the chickpea and pigeonpea crops, authors should refrain from providing a general introduction to these crops, except if they are being grown in a new area.**
- If necessary, include one or two small tables (and no more). Supply only the essential information; round off the data-values to just one decimal place whenever appropriate; choose suitable units to keep the values small (e.g., use tons instead of kg). Every table should fit within the normal typewritten area of a standard upright page (not a 'landscape' page).
- Black-and-white photographs and drawings (prepared in dense black ink on a white card or a heavy-duty tracing paper) are welcome - photocopies, color photographs, and 35-mm slides are not. Please send disk-files (with all the data) whenever you submit computer-generated illustrations.
- Keep the list of references short - not more than five references, all of which should have been seen in the original by the author. Provide all the details including author/s, year, title of the article, full title of the journal, volume, issue, and page numbers (for journal articles), and place of publication and publishers (for books and conference proceedings) for every reference.
- Express all the quantities only in SI units. Spell out in full every acronym you use.
- Give the correct Latin name of every crop, pest, or pathogen at the first mention.
- Type the entire text in double spacing. Please send a file, which should match the printout, on a double-sided/high density IBM-compatible disk using Microsoft Applications.
- Contact the Editor for detailed guidelines on how to format text and diskettes.
- **Include the full address with telephone, fax, and email numbers of all authors.**

The Editors will carefully consider all submitted contributions and will include in the Newsletter those that are of acceptable scientific standard and conform to requirements. The language of the Newsletter is English, but where possible, articles submitted in other languages will be translated. Authors should closely follow the style of the reports in this issue. Contributions that deviate markedly from this style will be returned for revision, and could miss the publication date. Communications will be edited to preserve a uniform style throughout the Newsletter. This may shorten some contributions, but particular care will be taken to ensure that the editing will not change the meaning and scientific content of the article. Wherever substantial editing is required, a draft copy of the edited version will be sent to the contributor for approval before printing.

**Contributions should be sent before 31 March to:**

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Last year (2002) was a landmark for scientists working on chickpea and pigeonpea. Several scientists received both individual as well as team awards for their achievements. ICRISAT and ICARDA jointly won the King Baudouin Award, the highest accolade conferred by the Consultative Group on International Agricultural Research (CGIAR) for excellence in chickpea research. ICRISAT Chickpea Team also won the Doreen Mashler Award for outstanding contribution to chickpea improvement. Jagdish Kumar, HD Upadhyaya, and P Lava Kumar were also recognized for their achievements described under the section News. Congratulations to one and all! I am sure in the years to come several such laurels will follow. Keep it up!

This issue of the International Chickpea and Pigeonpea Newsletter (ICPN) contains articles on all disciplines of chickpea and pigeonpea. However, most articles still continue to be from Asia and the Indian subcontinent in particular. A substantial research on these crops is being carried out in several other countries of Asia and in Africa, and ICPN can be a good informal vehicle to bring this research to wider readership. A great proportion of results on these crops remain unpublished or are published in the vernacular publications. This deprives a large section of the scientific community to be informed of the outcome of scientific efforts. I urge scientists to share their research results with the readership of ICPN.

Several papers submitted for this issue were not in the ICPN format and had to be sent back to authors for modifications. To reduce time in acceptance of papers for publication in the ICPN, I request authors to follow ICPN guidelines for format and length of submission.

I would like to acknowledge S Chandra, YS Chauhan, R Folkertsma, PM Gaur, JVDK Kumar Rao, V Mahalakshmi, S Pande, A Ramakrishna, GV Ranga Rao, KN Reddy, OP Rupela, KL Sahrawat, DVSSR Sastry, KB Saxena, R Serraj, HC Sharma, KK Sharma, P Singh, and RP Thakur for reviewing ICPN papers, and the Library at ICRISAT for compiling the SATCRIS listing.

I assure you that with cooperation from the contributors and readers, we will try our best to ensure that ICPN continues to maintain high standards in disseminating information efficiently and effectively among chickpea and pigeonpea workers.

**HD Upadhyaya**

## **About Scientists**

**Jagdish Kumar**, Principal Scientist (Chickpea Breeding), who is presently on secondment from ICRISAT and working with Agriculture Environmental Renewal Canada Inc. in Ottawa, Canada was awarded "Millennium ICRISAT Science Award 2002" in recognition of his contribution to chickpea improvement.

**HD Upadhyaya**, Special Project Scientist, Genebank, ICRISAT was awarded "Millennium ICRISAT Science Award 2002" for his Outstanding Scientific Article entitled "A mini core subset for capturing diversity and promoting utilization of chickpea genetic resources in crop improvement" published in Theoretical and Applied Genetics in 2001.

After completing one-year assignment as Visiting Scientist, **PM Gaur** joined as Senior Scientist (Chickpea Breeding) at ICRISAT, Patancheru effective August 2002.

## **ICRISAT and ICARDA Win the King Baudouin Award 2002**

ICRISAT and ICARDA have jointly won the 2002 King Baudouin Award, the highest accolade conferred by the Consultative Group on International Agricultural Research (CGIAR). This Award is given in recognition of the most outstanding scientific work done by the 16 CGIAR centers in partnership with national research and development organizations. The joint submission by ICRISAT and ICARDA was titled "Changing lives in marginal environments - ICRISAT and ICARDA: a winning partnership in chickpea research". The award was given for excellence in chickpea research, particularly for development of new chickpea varieties with higher tolerance to drought and heat, better resistance to pests and diseases that provide stable and economically profitable yields. The benefits of this research are having positive impacts in Bangladesh, Ethiopia, India, Myanmar, Nepal, Syria, and other rainfed agricultural areas. This research partnership involved collaboration between scientists and farmers in more than 30 countries.



(Left to right) Drs William Dar (Director General, ICRISAT), Jagdish Kumar (Principal Scientist, Chickpea Breeding, ICRISAT), Ian Johnson (Chairman, CGIAR), and Adel El-Beltagy (Director General, ICARDA) at the King Baudouin Award ceremony in Manila, Philippines.

ICRISAT has earlier won this award in 1998 for pigeonpea research and in 1996 for pearl millet research. ICRISAT and IITA are the only CG centers that have received this award thrice.

release of 50 high-yielding chickpea varieties. Also, improved crop production and integrated pest management options were developed and disseminated to many of these countries.

## **ICRISAT's Chickpea Team Wins Doreen Mashler Award 2002**

ICRISAT's Chickpea Team had another significant recognition during 2002. The team received the Doreen Mashler Award of ICRISAT for outstanding contribution to chickpea improvement. ICRISAT's Chickpea Team comprised scientists from ICRISAT and national agricultural research systems (NARS) of India, Bangladesh, Nepal, Pakistan, Myanmar, Ethiopia, Kenya, Sudan, Australia, USA, and Canada. In addition, farmers of these countries and some non-governmental organizations (NGOs) are the important team members. The ICRISAT-NARS collaboration in these countries has led to the

## **Chickpea Scientists' Meet at ICRISAT**

A Chickpea Scientists' Meet was organized at ICRISAT, Patancheru, India during 16-17 January 2003. Thirty chickpea scientists from India, Bangladesh, Nepal, Ethiopia, Australia, and Canada participated, along with 14 scientists from ICRISAT. The objectives of the meeting were to: (i) visit the chickpea research activities at ICRISAT, (ii) provide opportunity to scientists to select germplasm and breeding material, (iii) exchange information among scientists from national programs and ICRISAT, and (iv) identify future research thrusts and priorities for chickpea research globally. The technical session was devoted to presentations on future priorities and research



strategies of ICRISAT (PM Gaur), India (Masood Ali), Bangladesh (M Azizur Rahman), Nepal (RK Neupane), Ethiopia (Ketems Daba), Australia (EJ Knights), and Canada (Tom Warkentin) for chickpea improvement. After the technical session, the scientists visited experiments and research facilities of ICRISAT and selected breeding materials and germplasm of their interest.

## Awards for DFID-funded Research on Pigeonpea Sterility Mosaic

Research work on pigeonpea sterility mosaic funded by the Crop Protection Programme (CPP) of United Kingdom Department for International Development (DFID) was recognized for excellent research outputs that helped solve the mystery of sterility mosaic, a serious threat to pigeonpea production in the Indian subcontinent. With funding from CPP-DFID, ICRISAT and the Scottish Crop Research Institute (SCRI) have identified the causal agent of sterility mosaic and methods for its control. For these achievements two team members working in the DFID project bagged the following awards.

- P Lava Kumar, working in the DFID project since October 1996, first as PhD student (October 1996-August 1999) and later as a Special Project Scientist at ICRISAT won three awards for his outstanding research contributions:
  - The Jawaharlal Nehru Award for Outstanding Post Graduate Agriculture Research 2001 for best PhD work in plant pathology presented by the Union Minister of Agriculture, Sri Ajit Singh, on 16 July 2002 at Vigyan Bhavan, New Delhi, India. The award was instituted by the Indian Council of Agricultural Research (ICAR).
  - Millennium ICRISAT Science Award 2002, category 'Promising Young Scientist' presented by Dr Fortunato Battad, President Emeritus, Central Luzon State University, Philippines on the occasion of ICRISAT Loyalty Day on 12 December 2002 at ICRISAT, Patancheru, India.
  - Sri Veerapaneni Narasimham Memorial Gold Medal for the Year 2001 for best research worker in plant pathology presented by the Governor of Andhra Pradesh Sri Surjit Singh Barnala, during the Acharya NG Ranga Agricultural University 35<sup>th</sup> Annual Convocation on 11 March 2003 at Hyderabad, India.

- NK Kulkarni, former PhD student in a DFID-funded project at ICRISAT, won the Prof MJ Narasimhan Academic Merit Award for presentation of a research paper based on his PhD work in the national contest held during the 55<sup>th</sup> Annual Conference of Indian Phytopathological Society on 17 January 2003, at Osmania University, Hyderabad. He is currently working as Research Associate in DFID project at the University of Agricultural Sciences, Bangalore, India.

## New Pigeonpea Varieties Released in Andhra Pradesh

Two new pigeonpea varieties developed by Acharya NG Ranga Agricultural University, Rajendranagar, Hyderabad were released for cultivation in Andhra Pradesh, India during July 2002.

LRG 38 (Ranga Bold) was developed from a cross between C 11 and ICP 7035 by the Regional Agricultural Research Station, Lam, Guntur, Andhra Pradesh. It is a medium-duration variety (170 days) with bold seed (100-seed mass 10.5 g). It matures 10 days earlier than LRG 30 which was released in 1980. It is a robust bushy type with broad leaves. It is suitable for sole and intercropping during kharif (rainy) and rabi (post-rainy) seasons. The yield potential of LRG 38 is 2.0 to 2.2 t ha<sup>-1</sup>.

WRG 27 (Varalu) is a selection from a local landrace and was developed by the Agricultural Research Station, Warangal, Andhra Pradesh. It is a medium-duration variety (170-180 days). It has red flowers and is suitable for sole cropping and intercropping during kharif and rabi seasons. It is tolerant to *Helicoverpa* pod borer. The yield potential of WRG 27 is 2.0 to 2.2 t ha<sup>-1</sup>.

## National Review and Planning Meeting on Chickpea in Pakistan

The national review and planning meeting of Pulses Program was held from 24 to 26 September 2002 at the National Agricultural Research Centre (NARC), Islamabad, Pakistan after a gap of five years. The meeting was coordinated by Dr Muhammad Bashir, National Coordinator (Pulses), NARC and attended by more than 60 participants including scientists from federal and provincial research institutes, personnel from agriculture extension department, representatives of seed companies, Federal Seed Certification and Registration Department, and progressive growers. The overall objective of the meeting

was to share views to develop strategies for the improvement of chickpea in the country based on problem oriented research. The meeting provided a common forum for chickpea scientists to share their previous research results, plan research activities for 2002/03, and make recommendations for researchers, planners, and farmers. The issues related to production constraints of chickpea were thoroughly discussed. It was noted that non-availability of quality seed of improved cultivars, non-adoption of production technology, and lack of credit facilities for chickpea growers are major constraints. The following research priorities were fixed for the improvement of chickpea in the country:

- Improvement for high yield potential and desirable physio-agronomic traits with resistance to biotic and abiotic stresses
  - Drought and cold tolerance
  - Blight and wilt resistance
  - Field and storage insect infestation
- Development of package of production technology for different cropping systems

- Moisture conservation
  - Nutrient management and *Rhizobium* inoculation
  - Seed rate and sowing time
  - Weed control
  - Intercropping
- Production of certified and quality seed

The recommendations and research plan for 2002/03 were approved by the Director General, NARC and Member, Crop Sciences, Pakistan Agricultural Research Council (PARC). The higher authorities of PARC and Ministry of Food and Agriculture were requested to raise funds for chickpea research so that fixed targets may be achieved. The need for short- and long-term training of chickpea scientists was also felt. The house also proposed a seminar on chickpea during March 2003 and a National Conference on Grain Legumes (Pulses) during 2004 at Faisalabad, Pakistan.

**Contributed by:** *Muhammad Bashir*  
*National Coordinator (Pulses), Pulses Program,*  
*National Agricultural Research Centre*  
*Islamabad, Pakistan*

# Research Reports

## Chickpea

### Breeding

#### Effect of Seed Size on Seed Yield and Quality in Chickpea

**SK Varshney** (Department of Seed Technology, Tirhul College of Agriculture, Dholi (Muzaffarpur) 843 121, Bihar, India)

Chickpea (*Cicer arietinum*) is an important rabi (postrainy season) pulse crop of India and occupies a prime position both in area and production in the state of Bihar. Seed size and density affect the seed vigor as they indicate the amount of reserve food supply for seedlings during the period of germination, field emergence, and stress conditions. In seed industry too seed size is considered an important aspect of seed quality. To obtain uniform seed size within a variety, size grading is inevitable. While grading, sizeable portion of oversize and undcrsize seeds are rejected due to their unworthiness in terms of seed quality. Therefore, optimum seed size needs to be determined which may affect both seed yield and quality in chickpea (Vadivelu and Ramakrishnan 1983, Bhor et al. 1988). Keeping in view the above facts, this study was undertaken.

The experiment was conducted with eight treatments including four seed sizes: oversize (OS), graded (G), ungraded (UG), and undersize (US); and two chickpea genotypes P-256 and DHG 82-4. The experiment was laid out in three replications during rabi in 1990/91, 1991/92, and 1992/93 at Tirhut College of Agriculture, Dholi Research Farm, Dholi, Bihar. The plot size for each treatment was 5 x 3 m<sup>2</sup> and recommended agronomic practices were followed to raise the crop. The initial quality of the seed used for experimentation for three years is presented in Table 1. The average 100-seed mass ranged from 13.1 (US) to 35.5 g (OS) in P-256 and from 12.9 (US) to 35.8 g (OS) in DHG 82-4. The 100-seed mass of ungraded and graded seed was at par in both the genotypes. Other seed quality traits like germination percentage, seedling length, and vigor index (seedling length x germination percentage) were superior in OS and G seed as compared to UG and US seed of P-256 but the differences were less in seed of DHG 82-4.

Seed yield and seed quality traits in different seed grades were analyzed (Table 2). The germination percentage and seedling length were assessed as in the procedure laid down by the International Seed Testing Association (ISTA 1985). The vigor index was determined as given by Abdul Baki and Anderson (1973). The results on seed yield indicated that there was no significant difference between different sizes of seed. This clearly indicated that small seeds also have sufficient amount of food reserve for germination and stand establishment in chickpea.

**Table 1. Initial quality of seed of two chickpea genotypes used in the experiment in Dholi, Bihar, India<sup>1</sup>.**

Seed size <sup>2</sup>	100-seed mass (g)		Germination (%)		Seedling length (cm)		Vigorindex	
	Range	Mean	Range	Mean	Range	Mean	Range	Mean
P-256								
OS	32.0-40.6	35.5	92-98	95.0	13.9-21.7	18.6	1278.8-2129.5	1778.9
G	25.0-30.8	27.3	90-100	94.7	12.7-22.0	18.4	1193.8-2068.0	1769.6
UG	27.0-30.0	28.6	90-93	91.7	14.6-20.2	17.7	1314.0-1882.3	1626.6
US	11.0-15.0	13.1	85-90	87.0	13.2-21.1	16.9	1135.2-1793.5	1468.2
DHG 82-4								
OS	33.0-39.4	35.8	95-97	96.0	18.7-21.0	20.2	1791.4-2027.3	1937.9
G	25.0-32.4	28.1	96-97	96.7	17.2-19.6	18.3	1651.2-1896.4	1771.0
UG	27.0-30.0	28.6	93-96	94.3	17.8-20.2	18.9	1655.4-1898.8	1785.4
US	12.0-14.0	12.9	92-97	94.0	19.6-20.5	20.1	1822.8-1989.5	1887.2

1. All mean values represent average over three years. 1990/91, 1991/92, and 1992/93.

2. OS = Oversize; G = Graded; UG = Ungraded; and US = Undersize.

**Table 2. Effect of seed size on seed yield and quality in chickpea in Dholi, Bihar, India<sup>1</sup>.**

Description	Seed yield		100-seed mass		Germination		Seedling length		Vigor index	
	Mean (t ha <sup>-1</sup> )	Increase (%) over UG	Mean (g)	Increase (%) over UG	Mean	Increase (%) over UG	Mean (cm)	Increase (%) over UG	Mean	Increase (%) over UG
<b>Seed size<sup>2</sup></b>										
OS	1.94	9.07	25.17	-5.97	91.0	-	17.2	10.9	1579.0	9.8
G	1.81	2.08	28.73	7.32	92.0	1.09	17.4	12.3	1620.1	12.7
UG	1.77	-	26.77	-	91.0	-	15.5	-	1438.1	-
US	1.55	-12.96	26.30	-1.76	91.0	-	18.0	16.1	1673.9	16.4
<b>Genotype</b>										
P-256	1.69	-	27.03	-	91.3	-	17.0	-	1574.3	-
DHG 82-4	1.85	-	26.47	-	91.0	-	17.1	-	1588.0	-

1. All mean values represent average over three years, 1990/91, 1991/92, and 1992/93.

2. OS = Oversize; G = Graded; UG = Ungraded; and US = Undersize.

Based on the results of three years, we concluded that there should be minimum rejection of small seed during seed processing which may reduce the cost of chickpea production. Similarly, the seed quality traits like 100-seed mass, germination percentage, seedling length, and vigor index were not affected adversely by seed size. These seed quality traits were at par in all the seed produced from plants of seed of different sizes.

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- Bhor SB, Thete RY, Patil RB, and Bharud RW. 1988.** Effect of seed size on growth, yield, yield attributes and seed quality of gram. *Seed Research* 16:143-147.
- ISTA. 1985.** International rules for seed testing. *Seed Science and Technology* 13:299-355.
- Vadivelu KK, and Ramakrishnan V. 1983.** Effect of seed size on quality attributes and yield of seeds in Bengal gram (*Cicer arietinum* L.). *Seed Research* 11:177-181.

## Induced Flower Color Mutations in Chickpea

**BM Atta, M Ahsan ul Haq, TM Shah, M Sadiq, Mahmud ul Hassan, and Hina Syed** (Mutation Breeding Division, Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan)

Three main classes of flower color occur in chickpea (*Cicer arietinum*); pink and white color constitute major classes while blue color constitutes minor class. A survey of world collection of over 12,000 chickpea accessions indicated that 80.67% accessions had pink flowers (includes dark pink, pink, and light pink), 18.87% had white flowers, and 0.46% had blue flowers (Pundir et al. 1985). Only scanty information is available in literature about flower color mutations induced in chickpea.

Dry and healthy seeds with 10% moisture of desi chickpea cultivar Pusa 329 were treated with gamma rays and ethyl methane sulfonate (EMS) for induction of mutations. The doses of gamma rays were 250 and 300 Gray (Gy). Seeds presoaked in water for 2 h were treated with 0.3% and 0.4% aqueous solution of EMS for 1 h. Seeds of individual mutant plants (M<sub>1</sub> generation) in all the treatments (including control) were harvested separately and grown as single plant progenies. Morphological mutations for plant type, growth habit, branching, leaf type, pod size, flower color, and chlorophyll content were scored in M<sub>2</sub> generation.

Six M<sub>2</sub> progenies showing mutants for flower color were identified (Table 1). Mutation frequency of blue and white flower mutants on progeny basis was 0.15% and 0.07%

**Table 1. Induced flower color mutations in chickpea genotype Pusa 329.**

M <sub>2</sub> progeny	Treatment <sup>1</sup>	Total number of plants	Ratio of normal:mutant
1762	300 Gy gamma rays	16	15 normal : 1 blue (CM 1762/99)
1965	0.3% EMS	12	11 normal : 1 blue (CM 1965/99)
3268	0.4% EMS	16	15 normal : 1 blue (CM 3268/99)
3358	0.4% EMS	18	17 normal : 1 blue (CM 3358/99)
3339	0.4% EMS	14	13 normal : 1 white (CM 3339/99)
3513	0.4% EMS	15	14 normal : 1 white (CM 3513/99)

1. EMS = Ethyl methane sulfonate.

**Table 2. Distinguishing features of flower color mutants and parent chickpea genotype Pusa 329<sup>1</sup>.**

Character	Pusa 329	CM 1762/99	CM 1965/99	CM 3268/99	CM 3339/99	CM 3358/99	CM 3513/99	SE±
Days to flowering	89	91	93	96	92	96	96	0.94
Plant height (cm)	53.3	50.6	48.4	54.0	56.4	52.0	49.8	1.61
No. of primary branches plant <sup>1</sup>	6.0	6.1	4.4	4.5	5.4	4.3	5.1	0.49
No. of secondary branches plant <sup>1</sup>	9.2	13.2	11.2	12.1	11.8	9.3	9.0	1.46
Total number of pods plant <sup>1</sup>	121.3	180.8	128.8	117.1	141.4	108.2	114.9	8.32
100-seed mass (g)	15.23	8.95	10.20	8.23	16.75	9.15	16.83	0.31
Seed yield (g plant <sup>1</sup> )	30.42	25.80	18.22	15.82	37.16	12.94	32.26	1.85
Flower color	Pink	Blue	Blue	Blue	White	Blue	White	
Seed size	Medium	Small	Small	Small	Medium	Small	Medium	
Seed color <sup>2</sup>	LB	DB	DB	DB	LB	DB	B	
Growth habit <sup>1</sup>	SE	SE	SE	SS	SS	SS	SS	
Wilt (%) <sup>4</sup>	30	10	12	5	4	4	6	

1. Data are averages of three replications with five plants per replication.

2. LB = Light brown; DB = Dark brown; and B = Brown.

3. SE = Semi-erect; and SS = Semi-spreading.

4. Data are averages of three replications with forty plants per replication.

respectively. All the induced flower color mutants bred true in M<sub>3</sub> generation. Morphological data of M<sub>4</sub> is presented in Table 2.

Only two white flower mutants (CM 3339/99 and CM 3513/99) showed increase in seed yield as compared to control (Table 2). The increase in yield may be attributed to increase in 100-seed mass and wilt tolerance in both the mutants and more number of pods plant<sup>-1</sup> in CM 3339/99. Seed size was medium in control and white flower mutants, whereas it was small in all the blue flower mutants with markedly reduced yield per plant. The seed size was categorized according to 100-seed mass as small (<15 g), medium (15-18 g), medium-bold (19-22 g), and bold (>22 g). Phenotypically, white flower color seems to be associated with medium seed size and blue flower color with small seed size. The linkage of blue flowered plants with small seeds was also reported by Kumar et al.

(1982). Genetic studies are planned to investigate their mode of inheritance and associations. Proper utilization of these mutants for better yield can be made through intercrossing with high-yielding desi varieties followed by selection. The induced flower color mutants have been added in gene pool for use as genetic markers in different breeding experiments.

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## Response of Chickpea Genotypes to Different Dates of Sowing in Alfisols of Chittoor District, Andhra Pradesh, India

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In Chittoor district of Andhra Pradesh, India the length of crop growing season is more due to characteristic bimodal distribution of rainfall. Chickpea (*Cicer arietinum*) is a new crop to this region and hence it can be tried as a sequential crop in double cropping system, after groundnut (*Arachis hypogaea*). Among different agronomic practices, selection of suitable variety and optimum time of sowing are important non-monetary inputs for obtaining higher yields. Information regarding these aspects is lacking for the southern agroclimatic zone and hence this study was conducted at the SV Agricultural College Farm, Tirupati, Andhra Pradesh.

A field experiment was conducted during rabi (postrainy season) 2001/02 on sandy loam soils in a factorial randomized block design (RBD), replicated thrice with two factors, viz., three genotypes (ICCV 10, ICCV 2, Annigeri 1) and four dates of sowing (October 15, November 1, November 15, and December 1). A uniform fertilizer dose of 20 kg nitrogen ha<sup>-1</sup> and 50 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> was applied as basal dose to all experimental plots. One seed was hand dibbled per hill in furrows adopting a

spacing of 30 cm x 10 cm. Five plants were randomly selected from net plot area in each experimental plot and tagged for recording observations on growth characters and yield attributes.

ICCV 10 had the tallest plants and highest number of primary and secondary branches plant<sup>-1</sup>, whereas ICCV 2 had the lowest plant height. However, there was no significant difference in number of primary and secondary branches plant<sup>-1</sup> between ICCV 2 and Annigeri 1. ICCV 10 recorded highest number of pods plant<sup>-1</sup> and seeds pod<sup>-1</sup>, while these attributes were lowest in ICCV 2. The variety ICCV 2 took least days to mature (80 days) while ICCV 10 took highest time to mature (102 days) (Table 1).

When sown on November 1, the genotypes had good growth and high yield. Plant height and number of primary and secondary branches plant<sup>-1</sup> were low when the genotypes were sown on December 1. The lowest pod production and seeds pod<sup>-1</sup> were recorded when the genotypes were sown on November 15. The maturity duration was more in genotypes when sown on October 15 and was less when sown on December 1. Hastening of maturity with the delay in sowing was also reported by Aziz and Rahman (1994).

Seed yield was affected significantly by genotypes and time of sowing. Seed yield of ICCV 10 was similar to Annigeri 1 but significantly higher than ICCV 2 (Table 1). High yield was related to higher number of pods plant<sup>-1</sup> and seeds pod<sup>-1</sup> as reported by Reddy and Ahlawat (1998). Chickpea crop sown on November 1 recorded highest seed yield. This might be due to favorable temperature

**Table 1. Effects of different sowing dates and genotypes on agronomic characteristics and yield of chickpea at Tirupati, Andhra Pradesh, India during 2001/02<sup>1</sup>.**

Treatment	Days to maturity	Plant height (cm)	No. of primary branches plant <sup>-1</sup>	No. of secondary branches plant <sup>-1</sup>	No. of pods plant <sup>-1</sup>	No. of seeds pod <sup>-1</sup>	Seed yield (t ha <sup>-1</sup> )
<b>Genotypes</b>							
ICCV 10	102	35.0	3.8	7.9	18.0	1.30	0.48
ICCV 2	80	28.0	2.7	6.6	12.1	0.99	0.35
Annigeri 1	98	30.7	2.7	6.4	16.3	1.27	0.44
SEm ±	0.28	0.23	0.77	0.1	0.39	0.007	0.018
CD (P = 0.05)	1	0.7	0.2	0.3	1.2	0.02	0.053
<b>Sowing dates</b>							
October 15	100	30.7	3.1	7.8	16.0	1.20	0.42
November 1	96	32.9	3.4	8.2	18.0	1.24	0.58
November 15	91	31.0	3.0	6.0	13.3	1.14	0.35
December 1	87	30.3	2.8	5.7	14.5	1.15	0.34
SEm ±	0.33	0.26	0.09	0.12	0.46	0.008	0.021
CD (P = 0.05)	1	0.8	0.3	0.4	1.3	0.03	0.062

1. Data of all characteristics except days to maturity were recorded at harvest.

during crop growth period resulting in increased number of pods plant<sup>-1</sup> and seeds pod<sup>-1</sup>. Chickpea crop when sown on December 1 recorded lowest seed yield. Yield reduction in chickpea when sown earlier or later than the optimal date of sowing was also observed by Paikaray and Misra (1992) and Saini and Faroda (1997). The results indicated that ICCV 10 performed better than Annigeri 1 and ICCV 2, indicating the suitability of this variety to this tract. Sowing of chickpea on November 1 was found to be the best in Chittoor district.

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## Evaluation of Chickpea Genotypes for Cold Tolerance

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In North India, minimum temperature falls below 10°C for two and half months (December, January, and early February) during the crop season of chickpea (*Cicer arietinum*). Due to lack of cold tolerance the recommended chickpea cultivars of the region are unable to set productive pods (with fully developed seeds) at this low temperature. However, cultivars continuously flower and develop few pods with shriveled seeds. The cultivars set productive pods when temperature starts rising from mid-February.

Hence, the maturity period may be prolonged to 150-160 days. Consequently per day productivity of chickpea is low when compared to the most competitive crop of the region, i.e., wheat (*Triticum aestivum*). There is urgent need to develop cold tolerant chickpea cultivars. The cold tolerant genotypes will mature early and also escape from the damage of insect pests. After early harvest of chickpea crop, farmers may have an additional summer crop. We evaluated 57 desi chickpea genotypes for cold tolerance at the Punjab Agricultural University, Ludhiana (30°54' N, 75°48' E, 247 m altitude), Punjab, India.

The test genotypes of chickpea were selected from the International Chickpea Cold Nursery/Winter 1995-96 supplied by ICRISAT, Patancheru, Andhra Pradesh, India. The genotypes were sown on 1 October 2000, 25 days in advance than the recommended time to ensure that flowering occurs during the cold spell. Each genotype was sown in a single row plot of 3 m length with 30 cm interrow spacing. Two plants from each genotype were tagged at flower initiation stage. All except six genotypes flowered by mid-December 2000. Pollen viability, and total pods and productive pods formed per plant were recorded at low (minimum) temperatures of 5°C and 10°C separately to evaluate the genotypes for cold tolerance. Pollen viability of each genotype was studied in the flowers exposed at least three days to minimum temperatures selected for the study (Srinivasan et al. 1999). Total pods plant<sup>-1</sup> and productive pods plant<sup>-1</sup> on the tagged plants were counted for both the minimum temperatures, separately. Minimum temperature remained around 5°C from 21 December 2000 to 29 January 2001 except for seven days. Old flowers and pods were removed from the tagged plants on 21 December 2000. Pods appeared till 29 January 2001: tagged plants were tied with small white thread and pods were allowed to develop further. On 15 February 2001, total pods and number of productive pods were recorded. Empty pods turned pale yellow and productive pods were green. These tagged plants were further allowed to (lower till 28 February 2001 at minimum temperature 10°C, and all the fresh pods that appeared were tied with red color thread on these tagged plants. Thereafter no flower was allowed to develop into pod till 15 March 2001. Final count of pods per plant were taken on 15 March 2001.

Analysis of variance revealed that genotypes differed significantly for pollen viability and pods formed at both minimum temperatures selected for the study (Table 1). The genotypes differed in reaction to low temperatures. Thus the genotypes could be isolated for cold tolerance. Genetic variation in pod set at low temperatures was also noticed earlier under field conditions (ICRISAT 1988) and confirmed both in field and controlled environments

**Table 1. Pollen viability, total and productive pods formed at two minimum temperatures in chickpea genotypes at Ludhiana, Punjab, India.**

Genotype	Pollen viability <sup>1</sup> (%)		Pods formed plant <sup>-1</sup>			Productive pods plant <sup>-1</sup>		
	5°C	10°C	5°C	10°C	Total	5°C	10°C	Total
ICC 3197	7.35	88.38	5.5	74.5	80.0	0	28	28
ICC 3422	2.62	60.72	0.0	157.5	157.5	0	7	7
ICC 3423	1.50	93.42	25.5	0.5	26.0	0	0	0
ICC 3426	1.24	71.75	0.0	33.0	33.0	0	3	3
ICC 3427	0.00	75.62	0.0	64.0	64.0	0	3	3
ICC 3428	1.61	91.84	0.0	0.0	0.0	0	0	0
ICC 3437	2.56	90.60	0.0	199.0	199.0	0	0	0
ICC 3488	1.62	70.62	0.0	0.0	0.0	0	0	0
ICC 3489	0.00	74.39	0.0	3.5	3.5	0	3	3
ICC 3500	2.34	98.92	18.0	6.5	24.5	0	3	3
ICC 3501	0.25	72.42	4.0	0.0	4.0	0	0	0
ICC 3502	2.46	91.92	6.5	0.0	6.5	1	6	7
ICC 3503	0.24	90.91	14.0	7.5	21.5	0	1	1
ICC 3504	0.27	97.68	25.0	40.5	65.5	3	6	9
ICC 3505	0.35	55.07	11.0	3.0	14.0	0	0	0
ICC 3507	0.49	71.80	19.5	4.0	23.5	2	4	6
ICC 3590	0.27	84.76	2.5	26.0	28.5	0	1	1
ICC 4479	0.00	90.92	0.0	4.0	4.0	0	1	1
ICC 4492	0.32	95.95	3.5	19.0	22.5	0	2	2
ICC 7150	0.00	88.76	0.0	0.0	0.0	0	0	0
ICC 7178	0.00	90.87	0.0	21.0	21.0	0	4	4
ICC 7179	0.00	94.88	0.0	12.0	12.0	0	8	8
ICC 11406	0.34	81.27	0.0	42.0	42.0	0	5	5
ICC 11407	0.37	95.10	0.0	3.5	3.5	0	3	3
ICC 11408	0.00	72.60	0.0	75.0	75.0	0	4	4
ICC 11411	0.00	86.86	0.0	17.0	17.0	0	4	4
ICC 11412	0.34	86.27	0.0	17.5	17.5	0	9	9
ICC 11414	0.68	89.12	0.0	22.5	22.5	0	12	12
ICC 11416	0.00	97.24	0.0	34.5	34.5	0	11	11
ICC 11417	1.96	97.24	49.5	40.0	89.5	0	0	0
ICC 11418	0.00	96.94	0.0	17.0	17.0	0	6	6
ICC 11421	0.00	89.12	0.0	1.5	1.5	0	1	1
ICC 12382	2.65	92.32	5.5	0.5	6.0	0	0	0
ICC 12385	0.00	89.90	0.0	1.0	1.0	0	0	0
ICC 12386	1.51	88.95	0.0	11.0	11.0	0	6	6
ICC 12387	0.31	90.94	7.0	12.0	19.0	0	4	4
ICC 12388	0.00	92.84	0.0	26.0	26.0	0	12	12
ICC 12389	0.00	92.93	0.0	0.0	0.0	0	0	0
ICC 12390	0.21	95.12	2.5	3.5	6.0	0	1	1
ICC 12398	0.00	90.17	0.0	2.0	2.0	0	2	2
ICC 12400	2.91	90.47	0.5	5.0	5.5	0	2	2
ICC 12406	1.87	95.86	0.0	13.0	13.0	0	4	4
ICC 12407	0.27	90.32	4.0	0.5	4.5	0	0	0
ICC 12408	1.19	92.70	0.0	16.5	16.5	0	2	2
ICC 12410	0.00	95.11	0.0	19.5	19.5	0	2	2
ICC 12412	1.13	98.40	0.0	23.0	23.0	0	13	13
ICC 12413	0.38	94.75	0.0	38.0	38.0	0	3	3

*continued*



**Table 1.** Continued

Genotype	Pollen viability <sup>1</sup> (%)		Pods formed plant <sup>1</sup>			Productive pods plant <sup>1</sup>		
	5°C	10°C	5°C	10°C	Total	5°C	10°C	Total
ICC 12414	0.69	97.77	0.0	9.5	9.5	0	5	5
ICC 12415	0.53	99.57	4.0	10.0	14.0	2	3	5
ICC 12416	1.02	82.04	5.0	0.0	5.0	1	0	1
ICC 12418	1.11	99.61	2.0	9.0	11.0	0	4	4
ICC 12419	7.07	100.00	7.0	61.0	68.0	3	57	60
ICCV 88501	2.57	96.34	69.5	20.0	89.5	36	3	39
ICCV 88502	1.72	97.20	23.5	11.5	35.0	16	3	19
ICCV 88503	1.95	92.93	32.0	5.0	37.0	6	3	9
ICCV 88505	0.10	94.70	15	38.5	40.0	0	10	10
ICCV 88506	1.74	98.07	7.5	7.5	15.0	1	3	4
CD (5%)	0.53	20.36	0.74	4.78	-	-	-	-

1. Mean of nine observations recorded in one flower of the first plant and two flowers of the second plant.

by Singh et al. (1993) and Srinivasan et al. (1998). Variation in pollen viability among the genotypes was also noticed by Srinivasan et al. (1999).

Pollen viability ranged from 0 to 7.35% at 5°C and 55.07 to 100% at 10°C (Table 1). In general, pollen viability was very low (<3%) in all the genotypes except two genotypes, ICC 3197 (7.35%) and ICC 12419 (7.07%) at 5°C. These results clearly indicated that pollen viability was severely affected at minimum temperature 5°C.

The total number of pods formed plant<sup>-1</sup> ranged from 0 to 69.5 at 5°C and 0 to 199 at 10°C. Of the 57 genotypes, 26 genotypes produced pods and the remaining 31 genotypes did not produce any pod at 5°C. The genotype ICCV 88501 produced highest number of pods (69.5 pods plant<sup>-1</sup>) followed by ICC 11417 with 49.5 pods plant<sup>-1</sup> at minimum temperature of 5°C. At minimum temperature of 10°C, the genotype ICC 3437 developed high number of pods (199 pods plant<sup>-1</sup>), followed by ICC 3422 with 157.5 pods plant<sup>-1</sup>. It appeared that most of the genotypes were sensitive to both the minimum temperatures selected for the study. Only 16 genotypes developed more than 30 pods plant<sup>-1</sup> during the entire cold spell. Correlation studies indicated significant and positive association of total pods plant<sup>-1</sup> with pollen viability and pods formed at 5°C ( $r = 0.403$  and  $0.283$ ) and very strong association with pods formed at 10°C ( $r = 0.938$ ).

The number of productive pods plant<sup>-1</sup> with fully developed seeds varied from 0 to 36 at 5°C and 0 to 57 at 10°C. Of the 57 test genotypes, only 10 genotypes developed productive pods at 5°C. Two genotypes, ICCV 88501 (36 pods plant<sup>-1</sup>) and ICCV 88502 (16 pods plant<sup>-1</sup>), had the ability to develop good number of productive

pods at low temperature. Srinivasan et al. (1998) also reported cold tolerance in these two genotypes at low temperature under field conditions during December and January, but observed that the third genotype ICCV 88503 was better tolerant to low temperature. However, in our study ICCV 88503 did not develop good number of productive pods (6 pods plant<sup>-1</sup>). At minimum temperature of 10°C, 44 genotypes produced productive pods and 13 genotypes did not produce any productive pods. Of the 44 genotypes, only 7 genotypes had 10 or more productive pods plant<sup>-1</sup>. ICC 12419 had highest number of 57 productive pods plant<sup>-1</sup>. Few genotypes, ICCVs 88501, 88502, and 88503, had more number of productive pods plant<sup>-1</sup> at 5°C than at 10°C. These genotypes flowered early (38 days to flowering) as compared to other genotypes. When the plants were tagged for this study these were in full bloom. Pods developed at the end of cold spell of 5°C attained physiological maturity and later a small number of flowers appeared during the second cold spell of 10°C. Therefore, only few pods appeared at minimum temperature 10°C. It is interesting to note that ICC 3437 produced 199 pods plant<sup>-1</sup> at 10°C but none were productive. This confirmed that development of pod may take place at low temperatures but development of seed required high temperature.

On the basis of this study, four genotypes namely ICC 3197, ICC 12419, ICCV 88501, and ICCV 88502 appeared promising for cold tolerance and this trail needs to be further confirmed under phytotron conditions. The seeds of these lines are available for distribution.

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rots, ascochyta blight, stunt, and pod borer. Wilt caused by *Fusarium oxysporum* f. sp. *ciceris* causes severe yield loss. Genetic resistance is the most economical way to control this disease. The Pulses Research Station of Himachal Pradesh Krishi Vishwavidyalaya in Berthin, Himachal Pradesh has identified a small-seeded, wilt resistant, high-yielding line ICCV 90201, designated as Himachal Chana 2. The line was developed from the cross GL769 x P919 at ICRISAT, Patancheru, India. The yield performance of Himachal Chana 2 from 1991/92 to 1999/2000 in various trials conducted in low hills of the state is given in Table 1. The new line gave an average seed yield of 1879 kg ha<sup>-1</sup> at Berthin (Table 1). The improvement in yield over C 235, HPG 17, and Himachal Ghana 1 at Berthin was 92.9%, 18.3%, and 6.8% while at Dhaulakuan it was 20.2%, 59.1%, and 65.3%, respectively. The plants of Himachal Ghana 2 are compact, medium tall, and erect having average 100-seed mass of 16.5 g. In on-farm trials conducted on farmers' fields in low hills and mid-hills of

## Himachal Ghana 2: A New Desi Chickpea Line for Himachal Pradesh, India

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Chickpea (*Cicer arietinum*) is an important pulse crop in the low hills and mid-hills of Himachal Pradesh, India. The major biotic factors for low production are wilt, root

**Table 2. Performance of chickpea line Himachal Chana 2 in farmers' fields in low and mid-hills of Himachal Pradesh, India, 1999/2000.**

Conducting agency	Test set	Seed yield (kg ha <sup>-1</sup> )		
		Himachal Chana 2	Himachal Chana 1	C 235
KVK, Una	1	1087	1050	980
	2	1070	1037	972
RSS, Berthin	1	1215	1192	890
	2	1172	1072	905
Average		1133	1087	936

**Table 1. Performance of chickpea genotypes in low hills and mid-hills of Himachal Pradesh, India.**

Location/Genotype	Seed yield (kg ha <sup>-1</sup> )									
	1991/92	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/2000	Average
Berthin										
Himachal Chana 2	1607	2822	1756	1367	-	2283	1127	2180	1890	1879
Himachal Chana 1	-	-	2178	1517	-	1721	1170	2375	1588	1758
HPG 17	-	2579	1663	1347	-	1347	1152	1422	1571	1587
C 235	1033	1591	703	605	-	1006	689	1131	1033	974
CD	509	513	365	389	-	601	268	632	274	-
Dhaulakuan										
Himachal Chana 2	-	-	2031	-	765	1333	1003	1410	-	1308
Himachal Chana 1	-	-	1545	-	397	410	802	800	-	791
HPG 17	-	-	1382	-	380	927	429	990	-	822
C 235	-	-	1913	-	750	1285	585	910	-	1088
CD	-	-	NS <sup>1</sup>	-	208	356	308	192	-	-

1. NS = Not significant.

the state, Himachal Chana 2 gave an average yield of 1133 kg ha<sup>-1</sup> as against 1087 kg ha<sup>-1</sup> in Himachal Ghana 1 and 936 kg ha<sup>-1</sup> in C 235 (Table 2). Himachal Chana 2 exhibited stable resistance to wilt (an average disease score of 1.6 during 1990/91 to 1993/94) when screened in wilt sick plots using the technique developed by Nene et al. (1981). It showed resistance to wilt also at ICRISAT in Patancheru, Jabalpur (Gupta 1995), and Hisar. In 1994, the All India Coordinated Research Project recommended Himachal Chana 2 as a donor for wilt resistance (Anonymous 1994).

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## Gujarat Gram 1: A High-yielding Wilt Resistant Desi Chickpea Variety for Central Zone of India

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The chickpea breeding program at Pulses Research Station, Gujarat Agricultural University, Junagadh, Gujarat, India aims to develop early-maturing, high-yielding, fusarium wilt resistant desi chickpea (*Cicer arietinum*) varieties with better seed quality for rainfed and irrigated conditions. The genotype Gujarat Gram 1 (GCP 101) was developed from the cross GCP 2 x ICCV 2 by pedigree method of selection and evaluated in various state and coordinated trials during 1994/95 to 1996/97 (Table 1). This variety (also called GG 1) was released and notified in 1997 by the Central Variety Release Committee.

Gujarat Gram 1 has high yield potential, wilt resistance, pod borer (*Helicoverpa annigera*) tolerance, good milling and cooking qualities, and better storage ability. It is an early-maturing variety (107 days) with medium seed size

**Table 1. Mean yield performance of chickpea variety Gujarat Gram 1 in coordinated and state varietal trials in central zone of India.**

Genotype	Seed yield (t ha <sup>-1</sup> )				Yield increase (%) over check cultivars
	1994/95	1995/96	1996/97	Mean	
Coordinated trials					
Gujarat Gram 1	2.39 (5) <sup>1</sup>	1.62 (7)	1.81 (10)	1.94 (22)	—
C 235 (check)	1.86(4)	0.96 (6)	1.43 (10)	1.41 (20)	37.58
Vijay (check)	2.30 (4)	1.35 (7)	1.66(10)	1.77 (21)	9.60
State trials (Irrigated)					
Gujarat Gram 1	2.42 (3)	1.76(3)	2.63(5)	2.27 (11)	—
Dahod Yellow (check)	1.91 (3)	1.24(3)	2.09 (5)	1.75 (11)	29.?:
ICCC 4 (check)	1.59(3)	1.22(3)	2.48 (5)	1.76(11)	28.98
State trials (Rainfed)					
Gujarat Gram 1	1.47(3)	1.24(5)	1.19(5)	1.24 (13)	—
Chaffa (check)	1.00(3)	0.85 (5)	1.08(5)	0.97 (13)	27.83

1. Figures in parentheses indicate number of locations.

**Table 2. Fusarium wilt incidence (%) in chickpea variety Gujarat Gram 1 in wilt sick plots at different locations in India.**

Location	1995/96		1996/97	
	Gujarat Gram 1	JG 62	Gujarat Gram 1	JG 62
Bharari	8.5	-	-	-
Hisar	18.6	-	8.5	100.0
Dholi	8.6	-	24.8	100.0
ICRISAT (Patancheru)	34.6	100.0	-	-
Sehore	82.1	100.0	15.3	96.0
Rahuri	20.2	-	10.1	-
Ludhiana	7.4	100.0	2.8	-
Berhampore	38.5	94.4	29.9	-
Junagadh	29.0	100.0	28.6	100.0
Dharwad	0.0	-	-	-
Kanpur	3.2	100.0	51.9	-
Delhi	-	-	25.8	97.9
Badanapur	-	-	18.0	100.0
Faridkot	-	-	10.4	100.0
Gulbarga	-	-	3.3	-

(100-seed mass 18.2 g) and is suitable for rainfed as well as irrigated conditions.

The yield of Gujarat Gram 1 in coordinated and state varietal trials is presented in Table 1. In 22 coordinated varietal trials (1994/95 to 1996/97) conducted at different locations in the central zone of India, Gujarat Gram 1 recorded a mean seed yield of 1.94 t ha<sup>-1</sup> as against 1.77 t ha<sup>-1</sup> in the control cultivar Vijay (an increase of 9.60%) and 1.41 t ha<sup>-1</sup> in the control cultivar C 235 (an increase of 37.58%). In 11 trials conducted under irrigated condition in Gujarat during the same period, the mean seed yield of this variety was 2.27 t ha<sup>-1</sup> as against 1.76 t ha<sup>-1</sup> in the control cultivar ICC 4 (an increase of 28.98%) and 1.75 t ha<sup>-1</sup> in the control cultivar Dahod Yellow (an increase of 29.71%). It also performed well in trials conducted under rainfed condition in the state during the same period, producing 1.24 t ha<sup>-1</sup> seed yield as against 0.97 t ha<sup>-1</sup> of local variety Chaffa (an increase of 27.83%). This variety was also evaluated in 27 front line demonstrations in the state during 1997/98, 1998/99, and 2001/02 under irrigated and rainfed conditions. It gave 1.80 t ha<sup>-1</sup> mean seed yield as against 1.59 t ha<sup>-1</sup> in Dahod Yellow (an increase of 13.21%) under irrigated condition. Under rainfed condition, this variety gave 1.47 t ha<sup>-1</sup> mean seed yield with an increase of 16.67% than local cultivar Kankaria (1.26 t ha<sup>-1</sup>).

**Table 3. Reaction of chickpea variety Gujarat Gram 1 to *Helicoverpa* pod borer in India.**

Genotype	Pod damage (%)		
	1995/96	1996/97	Mean
Gujarat Gram 1	12.65(9) <sup>1</sup>	12.66(9)	12.66(18)
C 235 (check)	41.20(5)	6.63 (5)	23.92 (10)
Vijay (check)	50.96 (5)	8.57 (3)	29.77 (8)

1. Figures in parentheses indicate number of locations.

Gujarat Gram 1 was found resistant or moderately resistant to fusarium wilt under wilt sick plot conditions at most of the locations during 1995/96 and 1996/97 (Table 2). This variety was also observed to be less damaged (12.66%) by *Helicoverpa* pod borer as compared to the check cultivars C 235 (23.92%) and Vijay (29.77%) (Table 3).

Gujarat Gram 1 has semi-spreading habit and small, light green leaflets; the seeds are smooth, round, reddish brown, and medium in size (100-seed mass 18.2 g). This new desi chickpea variety offers a good opportunity to the farmers of central zone to augment their economic growth and also increase the total pulse production of the region.

## Gujarat Gram 4: A New Desi Chickpea Variety for Northeastern India

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North East Plain Zone (NEPZ) of India needs chickpea (*Cicer arietinum*) varieties which have high yield potential coupled with early maturity and resistance to ascochyta blight, fusarium wilt, and botrytis gray mold. These varieties should also be suitable for late planting. The variety Gujarat Gram 4 (GCP 105) released in 2000 by the Central Variety Release Committee fulfills most of the above-mentioned requirements. This variety (also called GG 4) was developed from the segregating population ( $F_2$  generation) of the cross ICCL 84224 x Annigeri supplied by ICRISAT, Patancheru, India. The line was developed through pedigree selection and evaluated in various coordinated trials during 1996/97 to 1998/99 in NEPZ.

Performance of Gujarat Gram 4 under rainfed and irrigated conditions is given in Table 1. It produced 1.72 t ha<sup>-1</sup> seed yield, 10.97% more than C 235 (1.55 t ha<sup>-1</sup>)

under rainfed condition. Under irrigated condition, it gave 1.51 t ha<sup>-1</sup> seed yield, 12.69% more than the control BG 256 (1.34 t ha<sup>-1</sup>). It matured in 131 days compared to 132 days for C 235 and 134 days for BG 256. The yield performance of Gujarat Gram 4 from 1996/97 to 1998/99 in various coordinated varietal trials conducted in NEPZ is given in Table 2. This variety gave an average yield of 1.95 t ha<sup>-1</sup>, 18.18% increase over control cultivar C 235 (1.65 t ha<sup>-1</sup>).

The reaction of Gujarat Gram 4 and control cultivars C 235 and BG 256 to fusarium wilt, ascochyta blight, and botrytis gray mold is given in Table 3. Over two years,

**Table 1. Performance of chickpea variety Gujarat Gram 4 under different sowing conditions in North East Plain Zone of India during 1998/99.**

Genotype	Mean seed yield (t ha <sup>-1</sup> )		Yield increase (%) over control	
	Rainfed	Irrigated	Rainfed	Irrigated
Gujarat Gram 4	1.72(4) <sup>1</sup>	1.51 (4)	-	-
C 235 (control)	1.55(4)	1.25(4)	10.97	20.80
BG 256 (control)	1.37(4)	1.34(4)	25.55	12.69

1. Figures in parentheses indicate number of locations.

**Table 2. Seed yield of chickpea variety Gujarat Gram 4 in coordinated varietal trials in North East Plain Zone of India.**

Genotype	Mean seed yield (t ha <sup>-1</sup> )				Yield increase (%) over control
	1996/97	1997/98	1998/99	Mean	
Gujarat Gram 4	1.78(2) <sup>1</sup>	1.96(3)	2.11 (4)	1.95 (9)	
C 235 (control)	1.62(2)	1.81 (4)	1.51 (4)	1.65 (10)	18.18
BG 256 (control)	1.34(2)	1.80(3)	1.64(4)	1.59(9)	22.64

1. Figures in parentheses indicate number of locations.

**Table 3. Reaction of chickpea variety Gujarat Gram 4 to different diseases in coordinated pathological nurseries under artificially inoculated conditions in India.**

Genotype	Fusarium wilt (%)			Disease reaction <sup>1</sup>	
	1996/97	1998/99	Mean	Ascochyta blight	Botrytis gray mold
Gujarat Gram 4	16.9 (12) <sup>2</sup>	30.0(14)	23.6 (26)	8.2 (3)	7.0 (2)
C 235 (control)		55.0(13)	55.0(13)	7.9 (4)	8.0 (2)
BG 256 (control)	35.8(12)	45.4 (14)	40.6 (26)	8.6 (4)	9.0(1)

1. Reaction during 1998/99 on 1-9 scale, where 1-3 = resistant and 7-9 = susceptible.

2. Figures in parentheses indicate number of locations.

the average incidence of wilt was 23.6% in Gujarat Gram 4 as compared to 55.0% and 40.6% in C 235 and BG 256, respectively. The incidence of aschochyta blight in Gujarat Gram 4 was slightly higher than C 235. But the incidence of botrytis gray mold was lower than both the control cultivars. The culinary and nutritional quality of Gujarat Gram 4 is also good. The 100-seed mass of this variety is 17.10 g. Seeds of Gujarat Gram 4 contain 23.18% protein and 66.48% carbohydrate. The new variety offers a better opportunity to the farmers of the NEPZ of India.

## Annigeri in the High Barind Tract of Bangladesh - Performance of a Chickpea Variety Out of its Zone of Adaptation

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Annigeri is a long-standing chickpea (*Cicer arietinum*) variety selected for the tropical zone of peninsular India (ICRISAT 1992, Krishnamurthy et al. 1996, Kumar et al. 1996). It is still widely used as a local check in varietal evaluations in peninsular India because of its consistently high yields over locations and seasons. The High Barind Tract (HBT) is situated in the northwest of Bangladesh, in the subtropics. Here, the chickpea growing period is considered to be longer because of the cooler and more prolonged winter period of the subtropics. Thus medium- or long-duration chickpea genotypes should be better adapted rather than short-duration types evolved in tropical regions with warm, short winters. However, in the particular conditions of the HBT, the surface soil dries quickly, and temperatures rise rapidly (to maximum >30°C) from the end of February, forcing the crop to terminal drought stress. Therefore, shorter duration varieties than those normally grown in the subtropics may have an advantage in this particular environment.

During the late 1990s, a study was conducted on comparative rooting behavior of chickpea genotypes, for their ability to acquire water and nutrients, across a range of environments and soil conditions (Ali 2000). The locations used were ICRISAT Center, in peninsular India, and the HBT. Annigeri was included as a common control variety in all experiments because it remained a dominating

variety in peninsular India and it was also thought as possibly suitable for the shorter duration subtropical environment of the HBT. Indeed, Annigeri outperformed other test genotypes in the first year of the study in the HBT, 1998/99. It was therefore tested against other genotypes known to perform well in the HBT in subsequent seasons and at several locations. This paper reports these comparisons.

Test chickpea genotypes were grown rainfed in farmers' fields, with a minimum of three replications. Plot size was 4 x 5 m<sup>2</sup>, but at Chabbishnagar the plot size was 1 *bigha* (1.333 m<sup>2</sup>) from 1999/2000 onwards. When Annigeri was sown prior to mid-November (Table 1), it matured 3-7 days earlier than the Barichola varieties. However, when it was sown later, date of maturity was about the same as for the other genotypes due to forced maturity of all plants by heat and soil moisture stress. In each season and at each location Annigeri out-yielded the genotypes against which it was tested (Table 1). Yield variation between tests was primarily due to date of sowing, affecting moisture status of the seedbed, and rainfall received during the growing period. There was no rain during 1998/99 and 2000/01 seasons; there was 71 mm of rain during 1999/2000 and 16 mm during 2001/02. No major diseases were observed on Annigeri, apart from minor incidence of collar rot (*Sclerotium rolfsii*) and chickpea stunt virus, but to no greater extent than for other genotypes. Pod borer (*Helicoverpa armigera*) attack was minimal in the low rainfall years but substantial in the higher winter rainfall year of 1999/2000. No genotypic differences in susceptibility to pod borer attack were observed.

Seed mass of Annigeri was greater than that of the Barichola lines and local varieties (Table 1). However, it was less than that of ICC 4958, another line introduced from ICRISAT to be used as a parent line for breeding for drought resistance particularly in the HBT because it has prolific rooting characteristics. Both of these lines have proven attractive to consumers in the HBT, particularly for confectionery purposes because of their large seed size and attractive color. Annigeri seed is bright yellow and reportedly tastes better as whole fry than other available varieties. Further, protein concentration in dhal of Annigeri, at around 25%, is greater than that of other comparable varieties (ICRISAT 1989).

The shorter duration, consistently good yield performance, no greater susceptibility to major pests and diseases of the HBT, and attractive consumer characteristics of Annigeri when compared to other local varieties have justified its proposal for release as variety for the HBT. Annigeri has been proposed for release by the Bangabandhu

**Table 1. Seed yield and seed mass of Annigeri in relation to other chickpea genotypes adapted to rainfed conditions at three locations in the High Barind Tract of Bangladesh.**

Genotype	Chabbishnagar				Nachole		Niamatpur
	1998/99 (22 Nov) <sup>1</sup>	1999/2000 (1 Nov)	2000/01 (28 Oct)	2001/02 (20 Nov)	2000/01 (15 Nov)	2001/02 (7 Nov)	2000/01 (8 Nov)
<b>Duration (days)</b>							
Annigeri	110	130	133	115	118	120	127
<b>Seed yield (t ha<sup>-1</sup>)</b>							
Annigeri	1.12	2.04	2.70	2.01	2.50	1.81	2.80
ICC 4958	1.09	1.80			1.63		
Barichola 2	0.85	1.89	2.20	1.80	2.23	1.49	2.29
Barichola 3			2.00	1.75	1.95	1.30	
Barichola 5		1.90	2.39	1.85	2.31	1.59	2.53
Local		1.80	1.83	1.81	1.88	1.10	1.79
SE±	0.075	0.189	0.224	0.141	0.211	0.128	0.246
<b>100-seed mass (g)</b>							
Annigeri	19.96	19.53	19.91	19.45	19.61	19.64	19.59
ICC 4958	29.91	28.10				28.80	
Barichola 2	14.32	13.30	13.10	13.65	13.49	13.30	13.10
Barichola 3			15.50	15.53	15.61	15.30	
Barichola 5		13.63	12.80	13.70	13.51	13.31	13.20
Local		12.01	12.02	12.80	12.60	12.48	12.71
SE±	0.365	0.477	0.358	0.531	0.462	0.441	0.511

1. Date of sowing.

Sheikh Mujibur Rahman Agricultural University, Salna, Gazipur, Bangladesh and the proposal is currently being evaluated. This experience shows that although a variety may have evolved in quite a different agro-ecological zone than the one under test, specific traits of the variety may cause it to "click" in an alien environment.

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## Priming of Chickpea Seeds with Water and Mannitol Overcomes the Effect of Salt Stress on Seedling Growth

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Poor crop establishment due to drought, lack of irrigation facilities, and salinity is a common problem in developing countries. Fast emergence of seedlings usually leads to healthier crops. In an earlier study, we had reported that osmo-priming (with mannitol) and hydro-priming of chickpea (*Cicer arietinum*) seeds for 24 h improved seedling growth under water deficit stress in comparison with non-primed control (Kaur et al. 2002b). The beneficial role of priming was attributed to the alterations in enzyme activities of carbohydrate metabolism in the seedlings. Kaur et al. (2002a) reported that activities of amylase, invertase (acid and alkaline), sucrose synthase and sucrose phosphate synthase in shoots, sucrose synthase and invertase in roots, and sucrose phosphate synthase in cotyledons increased in primed stressed seedlings as compared to non-primed stressed seedlings. Chickpea seedlings raised in 4% mannitol and water primed chickpea seeds showed better performance in terms of yield as compared to non-primed seeds (Kaur et al. 2002b). Harris et al. (1999) have also reported that overnight priming of seeds with water promoted seedling vigor, yield, and crop establishment of chickpea, maize

(*Zea mays*), and rice (*Oryza sativa*) in India. Similar results have been reported with chickpea crop raised from overnight water primed seeds in Bangladesh (Musa et al. 1999). This study was planned to see if priming of seeds with mannitol, water, sodium chloride (NaCl), gibberellic acid (GA<sub>3</sub>), and potassium nitrate (KNO<sub>3</sub>) could overcome the negative effect of salt stress on seedling growth in chickpea. GA<sub>3</sub> was primarily selected because of its role in increased seedling growth under NaCl imposed stress (Kaur et al. 1998).

Chickpea (PBG 1) seeds were washed with water, surface sterilized with 0.1% mercuric chloride for 5 min and again washed with water. The priming of seeds was done with 50, 100, 150, 200, 500, and 1000 mM NaCl; 50, 100, 150, and 200 mM KNO<sub>3</sub>; 4% mannitol; 3 and 6 μM GA<sub>3</sub>; 3 μM GA<sub>3</sub> + 4% mannitol, and 6 μM GA<sub>3</sub> + 4% mannitol; and water. For priming, the washed chickpea seeds were fully immersed in these solutions under aseptic conditions for 24 h at 25°C. The seeds were then washed with water and dried on a filter paper at 25°C. The primed and non-primed seeds were germinated in conical flasks at 25°C in dark on Murashige and Skoog (1962) medium without sucrose. The salt stress was created by including 75 mM NaCl in the medium. The length and biomass of roots and shoots of each seedling were recorded at 7 days after sowing.

In general it was observed that priming with water and mannitol causes early emergence of germination under salt stressed conditions. Priming with 4% mannitol and water increased the length and biomass of roots and shoots of salt stressed chickpea seedlings as compared to non-primed controls (Table 1). Priming with 3 and 6 μM GA<sub>3</sub> alone and in combination with 4% mannitol did not show any additional beneficial effect on seedling growth

**Table 1. Effect of priming of chickpea seeds with 4% mannitol, water, and gibberellic acid (GA<sub>3</sub>) on growth of seedling under salt stress at 7 days after sowing<sup>1</sup>.**

Priming treatment	Root growth seedling <sup>-1</sup>			Shoot growth seedling <sup>-1</sup>		
	Length (cm)	Fresh biomass (mg)	Dry biomass (mg)	Length (cm)	Fresh biomass (mg)	Dry biomass (mg)
Control <sup>2</sup>	3.6 ± 0.36	44.5 ± 1.5	4.3 ± 0.23	1.9 ± 0.18	35.7 ± 0.9	3.6 ± 0.10
4% mannitol	8.5 ± 0.02	53.7 ± 4.3	5.1 ± 0.35	3.8 ± 0.59	77.6 ± 7.6	7.8 ± 0.61
4% mannitol + 3 μM GA <sub>3</sub>	8.6 ± 0.69	58.3 ± 4.3	5.0 ± 0.29	3.3 ± 0.28	73.3 ± 4.8	7.4 ± 0.14
4% mannitol + 6 μM GA <sub>3</sub>	8.7 ± 0.79	69.7 ± 0.9	5.9 ± 0.30	3.5 ± 0.15	77.9 ± 2.7	7.3 ± 0.62
Water	7.1 ± 0.34	55.3 ± 3.3	5.4 ± 0.15	3.8 ± 0.26	88.1 ± 4.3	7.9 ± 0.45
3 μM GA <sub>3</sub>	7.5 ± 0.41	48.0 ± 2.0	4.8 ± 0.15	3.6 ± 0.31	73.0 ± 4.4	7.3 ± 0.75
6 μM GA <sub>3</sub>	7.1 ± 0.33	49.7 ± 3.9	4.8 ± 0.30	3.7 ± 0.08	73.1 ± 3.6	7.1 ± 0.25

1. Data represent mean ± SD of 3 replications with 10 seedlings in each replication.

2. Non-primed seedlings grown under salt (75 mM sodium chloride) stress.



**Table 2. Effect of priming of chickpea seeds with different concentrations of potassium nitrate (KNO<sub>3</sub>) and sodium chloride (NaCl) on growth of seedling under salt stress at 7 days after sowing<sup>1</sup>.**

Priming treatment	Root growth seedling <sup>-1</sup>			Shoot growth seedling <sup>-1</sup>		
	Length (cm)	Fresh biomass (mg)	Dry biomass (mg)	Length (cm)	Fresh biomass (mg)	Dry biomass (mg)
Control <sup>2</sup>	3.6 ± 0.36	44.5 ± 1.5	4.3 ± 0.23	1.9 ± 0.18	35.7 ± 0.9	3.6 ± 0.10
50 mM KNO <sub>3</sub>	3.7 ± 0.05	51.5 ± 0.5	5.0 ± 0.03	2.6 ± 0.02	68.8 ± 6.3	6.7 ± 0.45
100 mM KNO <sub>3</sub>	4.2 ± 0.94	64.7 ± 4.5	6.4 ± 0.38	2.4 ± 0.07	48.5 ± 3.0	4.7 ± 0.15
150 mM KNO <sub>3</sub>	4.4 ± 0.95	53.3 ± 5.1	5.3 ± 0.46	2.6 ± 0.41	54.4 ± 6.4	5.5 ± 0.60
200 mM KNO <sub>3</sub>	4.2 ± 0.24	50.4 ± 5.8	5.0 ± 0.46	2.1 ± 0.28	53.4 ± 7.9	5.1 ± 0.69
50 mM NaCl	3.6 ± 0.22	32.5 ± 1.9	3.1 ± 0.25	1.8 ± 0.33	39.0 ± 0.4	3.7 ± 0.09
75 mM NaCl	3.1 ± 0.01	31.0 ± 2.0	3.2 ± 0.20	1.6 ± 0.39	32.2 ± 2.2	3.1 ± 0.22
100 mM NaCl	2.8 ± 0.03	29.5 ± 1.5	2.8 ± 0.15	1.5 ± 0.04	32.0 ± 2.0	3.0 ± 0.25
150 mM NaCl	2.4 ± 0.32	28.7 ± 0.9	2.8 ± 0.10	1.7 ± 0.11	34.7 ± 1.5	3.0 ± 0.24
200 mM NaCl	1.9 ± 0.23	28.5 ± 0.5	2.7 ± 0.10	1.1 ± 0.10	25.5 ± 2.5	2.5 ± 0.20

1. Data represent mean ± SD of 3 replications with 10 seedlings in each replication.

2. Non-primed seedlings grown under salt (75 mM NaCl) stress.

though the addition of exogenous GA<sub>3</sub> to the medium of chickpea seedlings growing under saline conditions has been reported to increase seedling growth (Kaur et al. 1998). Osmo-conditioning of cucumber (*Cucumis sativus*) seeds with mannitol had also been reported to alleviate the adverse effects of salt stress on germination and growth of seedlings (Passam and Kakouriotis 1994).

Priming of chickpea seeds with NaCl had an adverse effect on seedling growth. Increasing the concentration of NaCl from 50 to 200 mM reduced the growth of primed seedlings (Table 2). The seeds primed with higher concentrations of NaCl (500 and 1000 mM) failed to germinate. However, priming of tomato (*Lycopersicon lycopersicum*) seeds with NaCl had been reported to improve seedling growth under salt stress (Cayuela et al. 1996). Although priming with different concentrations of KNO<sub>3</sub> though increased the biomass of roots and shoots, the increase was less as compared to water and mannitol primed seedlings (Tables 1 and 2). In tomato seeds, priming with KNO<sub>3</sub> has been reported to increase seedling growth under water and salt stressed conditions (Kang et al. 1996).

Priming of chickpea seeds with mannitol and water improved seedling growth under salt stressed conditions. This information can be employed by chickpea growers for improving the performance of crop in the field under adverse abiotic conditions. Harris et al. (1999) and Musa et al. (1999) have reported that seed priming increases yield of chickpea under rainfed conditions.

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## Response of Chickpea to Sources and Levels of Sulfur

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Chickpea (*Cicer arietinum*) is the most important pulse crop in Madhya Pradesh, India. It is cultivated on nearly 2.74 million ha, accounting for 6.1% of the total cropped area under pulses in the state. Madhya Pradesh contributed about 46% of the total chickpea production in the country. In general, the yield of chickpea in the state is almost stagnant (900-1000 kg ha<sup>-1</sup>) even with adoption of improved production technologies. Sulfur (S) fertilization improves both growth and seed yield of chickpea (Singh 1998). Most of the soils in Madhya Pradesh are generally becoming deficient in S due to high

cropping intensity, neglect of organic manures, and restricted use of S fertilizers. This investigation was therefore undertaken to study the effect of sources and levels of S on the yield of chickpea.

A field experiment was conducted during post-rainy season of 1993/94, 1994/95, and 1995/96 at the Main Pulse Research Station, RAK College of Agriculture, Sehore, Madhya Pradesh. The experimental soil was clay loam having pH 7.5, organic carbon 0.35%, available nitrogen (N) 200 kg ha<sup>-1</sup>, phosphorus (P) 9.2 kg ha<sup>-1</sup>, potassium (K) 310 kg ha<sup>-1</sup>, and S 9 mg kg<sup>-1</sup>. The experiment was laid out in a randomized block design with 4 replications. Different levels of S (0, 20, and 40 kg ha<sup>-1</sup>) were applied through elemental S (85% S), gypsum (18% S), single super phosphate (12% S), ammonium sulfate (24% S), and pyrite (22% S) at the time of sowing. Fertilizer dose of N, P, and K at 35, 53.5, and 20 kg ha<sup>-1</sup> respectively were applied as basal dressing.

The required quantity of N and P was applied through urea and triple super phosphate as per treatment. Chickpea cultivar JG 74 was sown in rows at 30 cm apart in the first week of November and harvested in the second week of March.

Sulfur level at 20 kg ha<sup>-1</sup> enhanced grain yield of chickpea significantly in all 3 years (Table 1). The yield was enhanced by 29% over control. Although similar increase in yield was observed with 40 kg S ha<sup>-1</sup>, the net benefit was not high. Plant height, branches plant<sup>-1</sup>, pods plant<sup>-1</sup>, and 100-seed mass increased significantly with application of 20 kg S ha<sup>-1</sup>. However, the differences in these characters in treatments with 20 kg S ha<sup>-1</sup> and 40 kg S ha<sup>-1</sup> were statistically not significant. The beneficial effect of S fertilization on growth and yield attributes may be due to better availability of S and its translocation which in turn increased the yield of chickpea. Maximum additional net return of Rs 2074 ha<sup>-1</sup> and incremental benefit-cost ratio of 3.84 were obtained with 20 kg S ha<sup>-1</sup>. Similar results were obtained by Singh (1998). Among the S sources, single super phosphate and gypsum proved superior to other sources with respect to growth, yield components, seed yield, and additional net return. The incremental benefit-cost ratio (4.60) with single super phosphate was also highest. Higher response to gypsum in respect of seed yield might be due to readily available S in gypsum compared to other S sources. Similar results were reported by Ram and Dwivedi (1992). Chickpea crop fertilized with 20 kg S ha<sup>-1</sup> through gypsum or single super phosphate may prove to be more productive and profitable.

**Table 1. Growth, yield components, and incremental benefit-cost ratio of chickpea with different sources and levels of sulfur at Sehore, Madhya Pradesh, India.**

Treatment	Growth and yield attributes'				Seed yield (kg ha <sup>-1</sup> )				Additional net return (Rs ha <sup>-1</sup> )	Incremental benefit-cost ratio
	Plant height (cm)	No. of branches plant <sup>-1</sup>	No. of pods plant <sup>-1</sup>	100-seed mass (g)	1993/94	1994/95	1995/96	Mean		
<b>Sulfur level (kg ha<sup>-1</sup>)</b>										
0	29.30	4.50	31.70	15.90	1218	1088	759	1021	-	-
20	31.92	5.48	40.20	16.12	1543	1391	998	1314	2074	3.84
40	32.00	5.92	42.26	16.24	1539	1384	1035	1319	2035	2.64
SE±	0.89	0.18	1.12	0.03	26.78	35.20	21.55	35.3	-	-
CD (5%)	1.27	0.63	3.16	0.11	87	101	79	99.0	-	-
<b>Sulfur source</b>										
Elemental sulfur	31.80	5.20	38.95	16.15	1513	1368	900	1260	1588	2.65
Gypsum	33.50	6.45	43.70	16.40	1518	1467	1120	1368	2624	2.75
Single super phosphate	31.65	5.85	43.50	16.20	1635	1452	1077	1388	3233	4.60
Ammonium sulfate	30.95	5.45	39.15	16.00	1500	1351	984	1278	1822	3.20
Pyrite	31.85	5.55	40.85	16.15	-	1336	1003	1169	1005	3.00
SE±	0.21	0.92	0.65	0.82	29.66	31.02	32.20	32.2	-	-
CD (5%)	0.61	NS'	1.96	NS	98	90	88	96.0	-	-

1. Data is mean of three years.

2. NS = Not significant.

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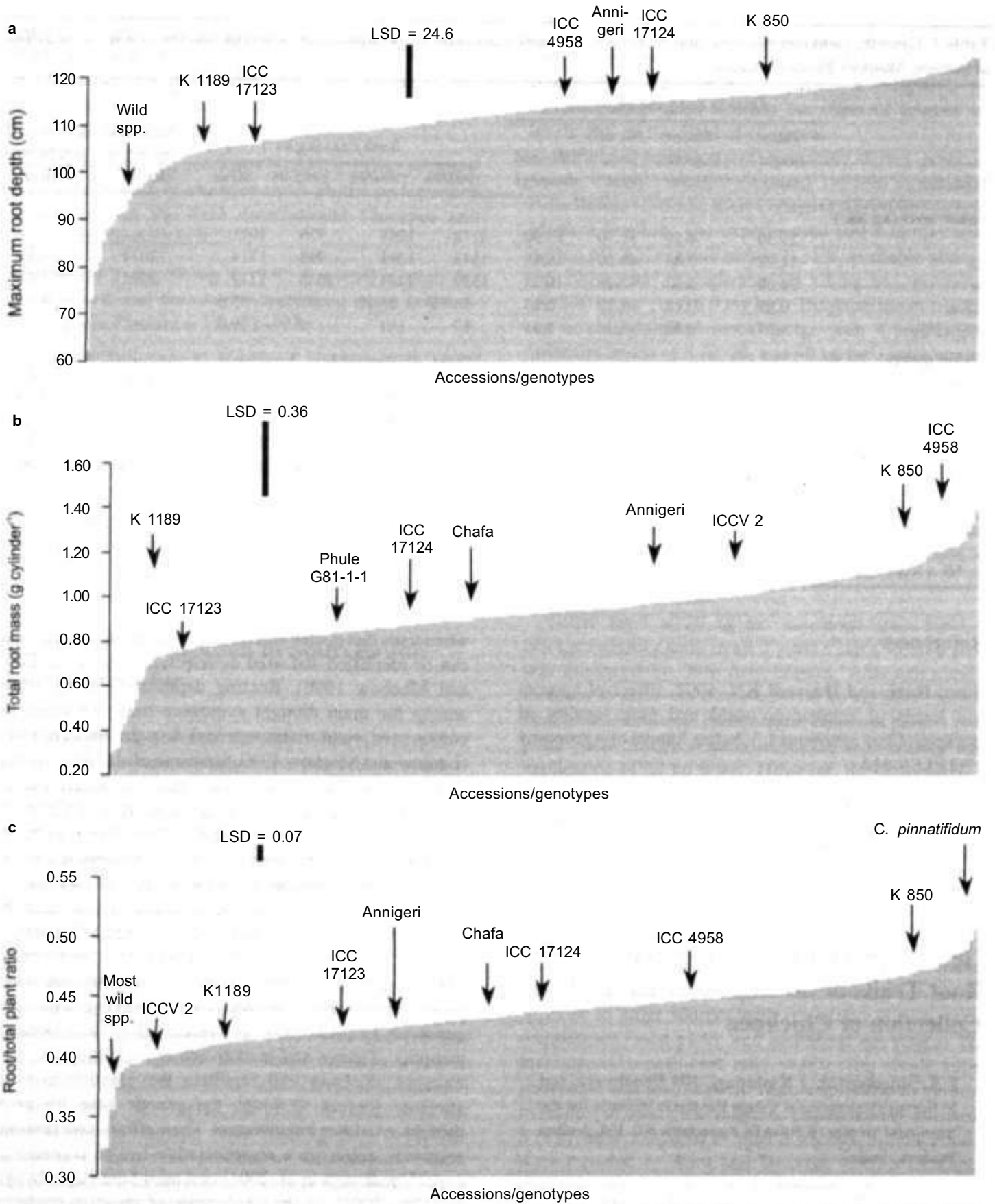
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## Genetic Diversity of Drought-avoidance Root Traits in the Mini-core Germplasm Collection of Chickpea

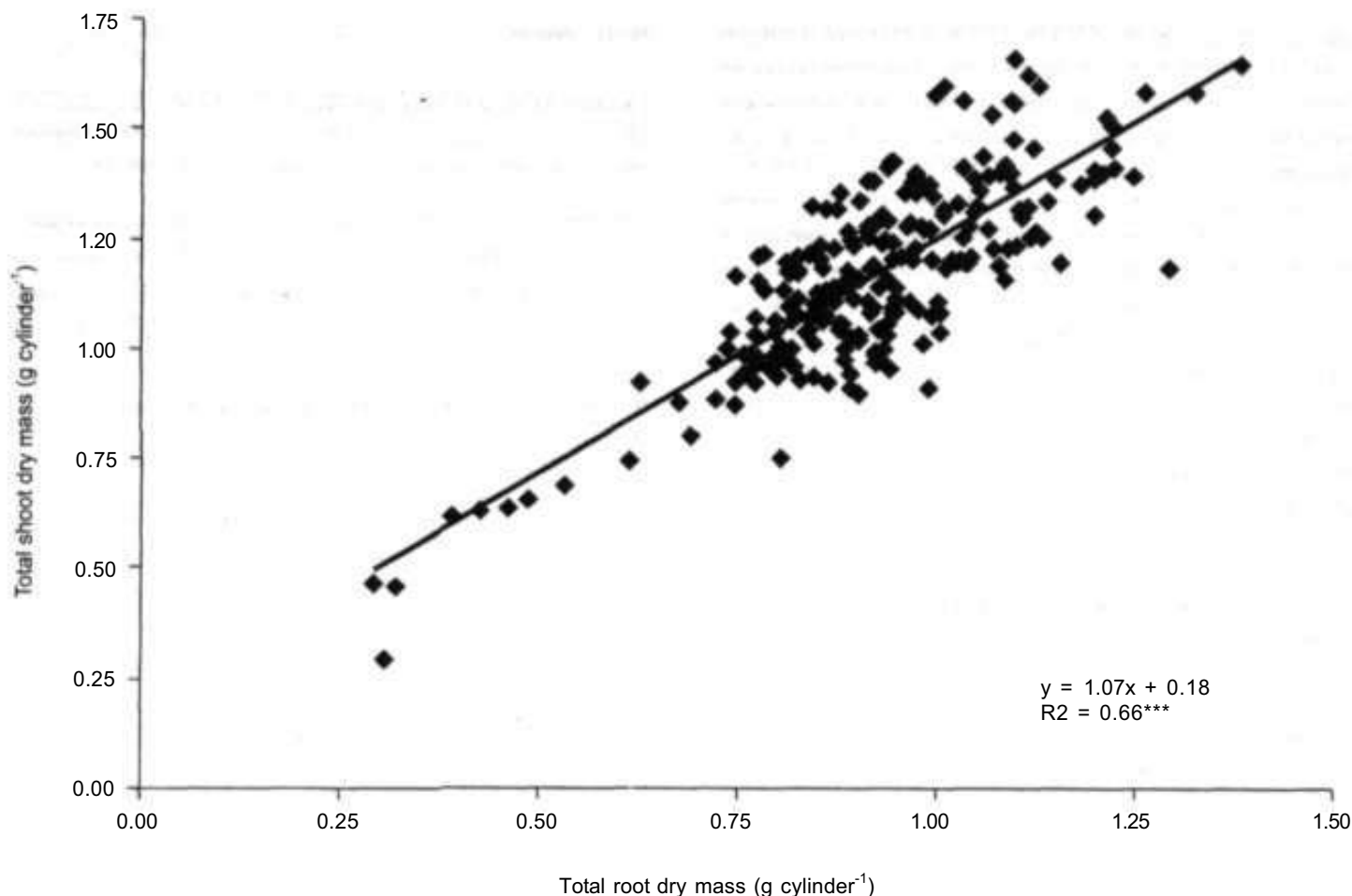
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Drought stress is a major cause for yield losses in chickpea (*Cicer arietinum*). A large portion of the losses can be prevented through crop improvement. Better drought-adapted genotypes could more effectively be bred

when traits that confer yield under water-limited conditions can be identified and used as selection criteria (Ludlow and Muchow 1990). Rooting depth and density were among the main drought avoidance traits identified to confer seed yield under terminal drought environments (Ludlow and Muchow 1990, Subbarao et al. 1995, Turner et al. 2001). Though they were rated as highly useful traits, these were also categorized as very difficult to screen. Efforts made at ICRISAT, Patancheru, India to identify sources for deep and large root system, led to the identification of the chickpea variety ICC 4958 (Saxena et al. 1993) and later to the development of drought tolerant varieties by incorporating the deep and large root system of chickpea into a well-adapted genetic background (Saxena 2002). However, most of these studies were based on a narrow genetic base involving only one genotype, i.e., ICC 4958. The recent efforts in molecular mapping of genes and marker-assisted selection for root traits in chickpea will facilitate the identification of alternate sources to widen the genetic base for crop drought-avoidance improvement. These efforts have become relatively easier as a representative list of accessions, core (Upadhyaya et al. 2001) and mini-core (Upadhyaya and Ortiz 2001), for the whole range of variation has been made available. The main objectives of this study were to assess the extent of genetic variation available for the root system traits (Figs. 1 and 2) in the mini-core germplasm of



**Figure 1. Rooting depth and density of the mini core chickpea germplasm accessions (n=211), 12 cultivated genotypes and 10 accessions of wild species at 35 days after sowing: (a) Maximum root depth attained; (h) Total root dry mass; and (c) Root/total plant ratio. (Note: The values are means of two replications.)**



**Figure 2. Relationship between total root dry mass and total shoot dry mass of the mini-core chickpea germplasm accessions, some wild species and chickpea cultivars.**

chickpea, to identify accessions with contrasting root growth in the early stages of development, and to compare them with familiar cultivars and wild relative species.

The whole mini-core germplasm collection of *C. arietinum* (211 accessions) along with 12 cultivars (Annigeri, ICC 4958, JG 62, JG 74, ICC 42, Phule G81-1-1, Chafa, K 850, K 1189, ICCV 2, KAK 2, and ICC 898) as references and 10 accessions of wild annual species (ICC 17116 of *C. yamashitae*, ICC 17123 and ICC 17124 of *C. reticulatum*, ICC 17156 of *C. bijugam*, ICC 17200 and ICC 17210 of *C. pinnatifidum*, ICC 17241 of *C. chorassanicum*, ICC 17148 and ICC 17180 of *C. judaicum*, and ICC 17162 of *C. cuneatum*) were evaluated by growing three plants in PVC cylinders (18 cm diameter, 120 cm long). The cylinders were filled with an equi-mixture (w/w) of Vertisol and sand, with initial soil water content equivalent to 70% field capacity. The plants were allowed to grow under receding soil moisture conditions thereafter, to mimic field terminal

drought. The cylinders were placed in pits to avoid heating due to direct solar radiation. The experiment was conducted in an Alpha design (6 x 40) with two replications. The sampling was done at 35 days after sowing, a time when early duration genotypes (well adapted to the lower latitudes) are known to exhibit maximum differences in root growth (Saxena et al. 1993). The data was analyzed using REML (residual maximum likelihood) analysis treating accessions as the random components.

The differences of entries were significant at <0.001 level for both root and shoot traits presented (Fig. 1). The root and shoot growth of the wild species was relatively poor compared to *C. arietinum* lines. However, the growth of *C. reticulatum* (ICC 17123 and ICC 17124) was relatively good and close to *C. arietinum* accessions (Fig. 1a). The maximum root depth of ICC 17241 (*C. chorassanicum*) was the least (62 cm). The range (73-91cm) of maximum root depth of the rest of the wild species, except *C. reticulatum*, was not significant. The

maximum root depth of ICCV 2, ICC 4958, and Annigeri was 115, 114, and 114 cm, respectively. The maximum root depth differences among cultivars were not statistically significant. Some of the accessions with a deep root system are ICCs 1431, 8350, 15697, 3512, and 11498.

Total root dry mass of the accessions of wild species except *C. reticulatum* was about one third of the maximum value (Fig. 1b). The linear growth phase of the root occurs later in most accessions of the wild species compared to the cultivated species as the growth duration of these are longer. As a result, maximum root depth and the root dry mass were poor in these accessions. The root dry mass of ICC 4958 and K 850 was significantly higher than that of K 1189 and Phule G81 -1 -1 (Fig. 1b). The top germplasm accessions for this trait were ICCs 5337, 7255, 13077, 15294, and 8261 with a root dry mass of more than 12 g cylinder<sup>1</sup>.

Ratio of root to total plant biomass also showed a vast range of variation (Fig. 1c). Most wild species showed very low ratio of root to total plant biomass (<0.39). Most of the cultivated genotypes and *C. reticulatum* exhibited a moderate value. Some of the accessions exhibiting significantly higher values of about 0.48 were ICC 17200 from *C. pinnatifidum* and ICCs 16207, 1397, 13077, 11627, and 12307 from *C. arietinum*.

Total root dry mass of the test entries showed a close linear relationship with the total shoot dry mass (Fig. 2) as well as the total leaf area of the plants. This relationship is very valuable for further root trait screening as it permits a less cumbersome preliminary selection of genotypes for large root mass on the basis of above ground shoot biomass or visual scores on shoot biomass or leaf area.

The germplasm accession ICC 4958 was previously used as the only source for deep and large root system parent or control in most of the drought avoidance related studies. The new genotypes identified, if confirmed, could be utilized as valuable alternative sources for diversification of mapping populations with varying growth duration and to get the required polymorphism for successfully mapping the root traits of chickpea.

This screening of the mini-core germplasm is being repeated during 2002/03 to confirm the results obtained. Any queries related to this study may be directed to Dr R Serraj, Principal Scientist, Crop Physiology, ICRISAT.

**Acknowledgment.** The authors thank the staff of Gene Bank, ICRISAT for supplying the seeds of mini-core chickpea germplasm and the wild species used in this study and the staff of Crop Physiology Lab for their technical help.

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## Root and Shoot Growth Dynamics of Some Chickpea Genotypes Under Two Moisture Levels

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Chickpea (*Cicer arietinum*) is usually grown under progressively receding soil moisture and terminal drought stress conditions. It is often grown on land, less preferred for cultivation of cereals, where soils are generally marginal in their physico-chemical characteristics. The chickpea root system gains importance under such environment as the yield stability depends more on the

root's ability to supply water and nutrients. More than 1500 chickpea germplasm accessions were previously screened for identification of drought-adapted genotypes. Among these, ICC 4958 was found to be the best and the vigorous early root system was found responsible for the relative drought tolerance (Saxena et al. 1993). Another drought tolerant genotype, Annigeri, was one of the best adapted for the peninsular Indian conditions and used often as a control cultivar for yield evaluations. The root system of ICC 4958, assessed under normal growing season, was 30% more than that of Annigeri. Mapping population developed for identification of molecular markers for root depth and root prolificacy by crossing ICC 4958 and Annigeri are currently under evaluation.

Before progressing further, it is necessary to evaluate the root system performance of these two parents in varying environments such as late planted condition to confirm their suitability for inclusion in expensive marker studies. Similarly, a mapping population developed by crossing JG 62, a double-podded genotype (better partitioning), and ICCV 2, an extra-early kabuli (drought escape), to identify molecular markers for both high yield under drought and earliness are being studied. Better root system in this population, if present, can be of an additional advantage for yield stability under drought. The root systems of ICC 4958 and Annigeri are already well documented mostly during normal growing season (Krishnamurthy et al. 1996) whereas those of JG 62 and ICCV 2 need to be studied yet. Thus, it became necessary to compare the root system characteristics of these genotypes in the off-season before conclusions can be drawn on the genetic value of these parents' root system

across environments and seasons, and to continue the use of the already available recombinant inbred line (RIL) populations for mapping studies.

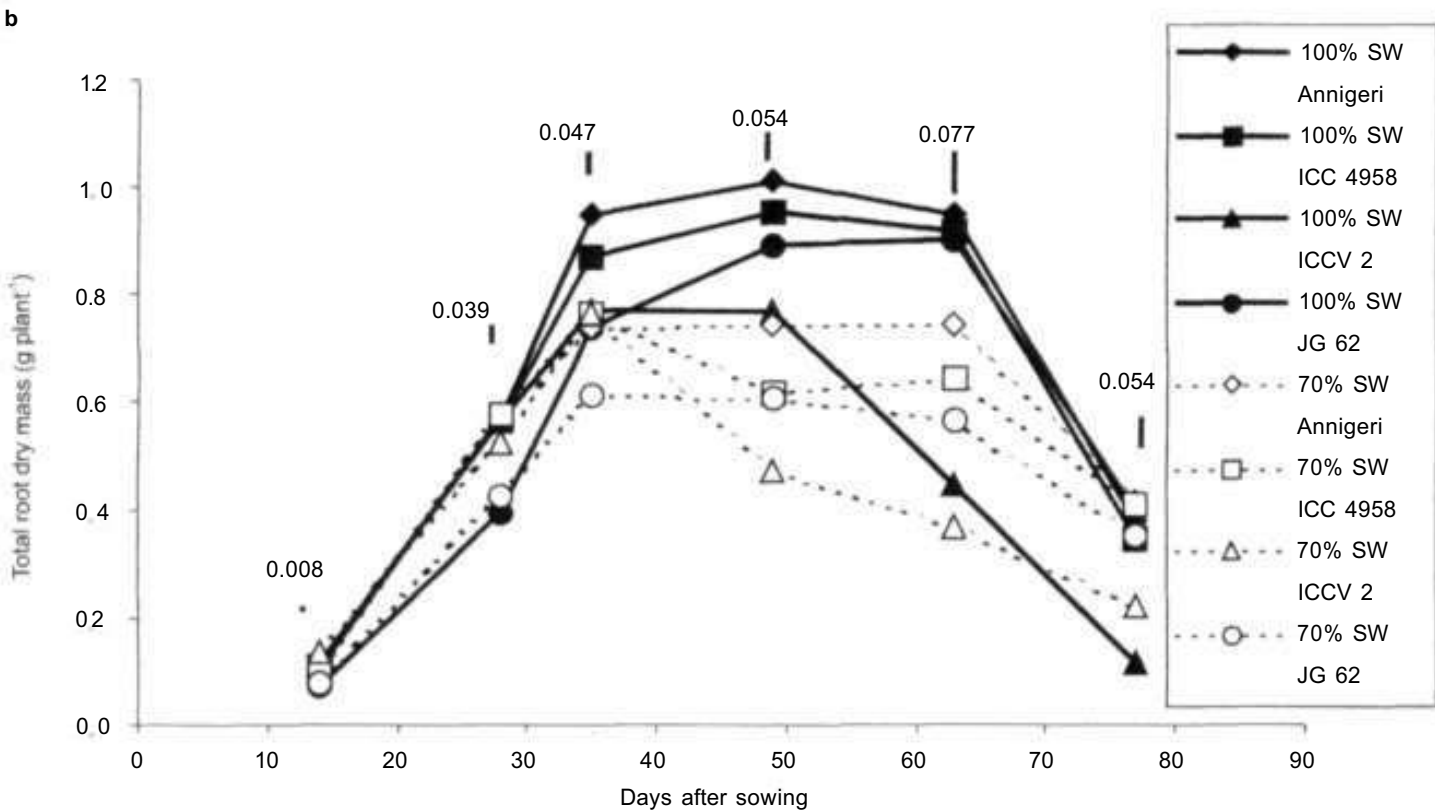
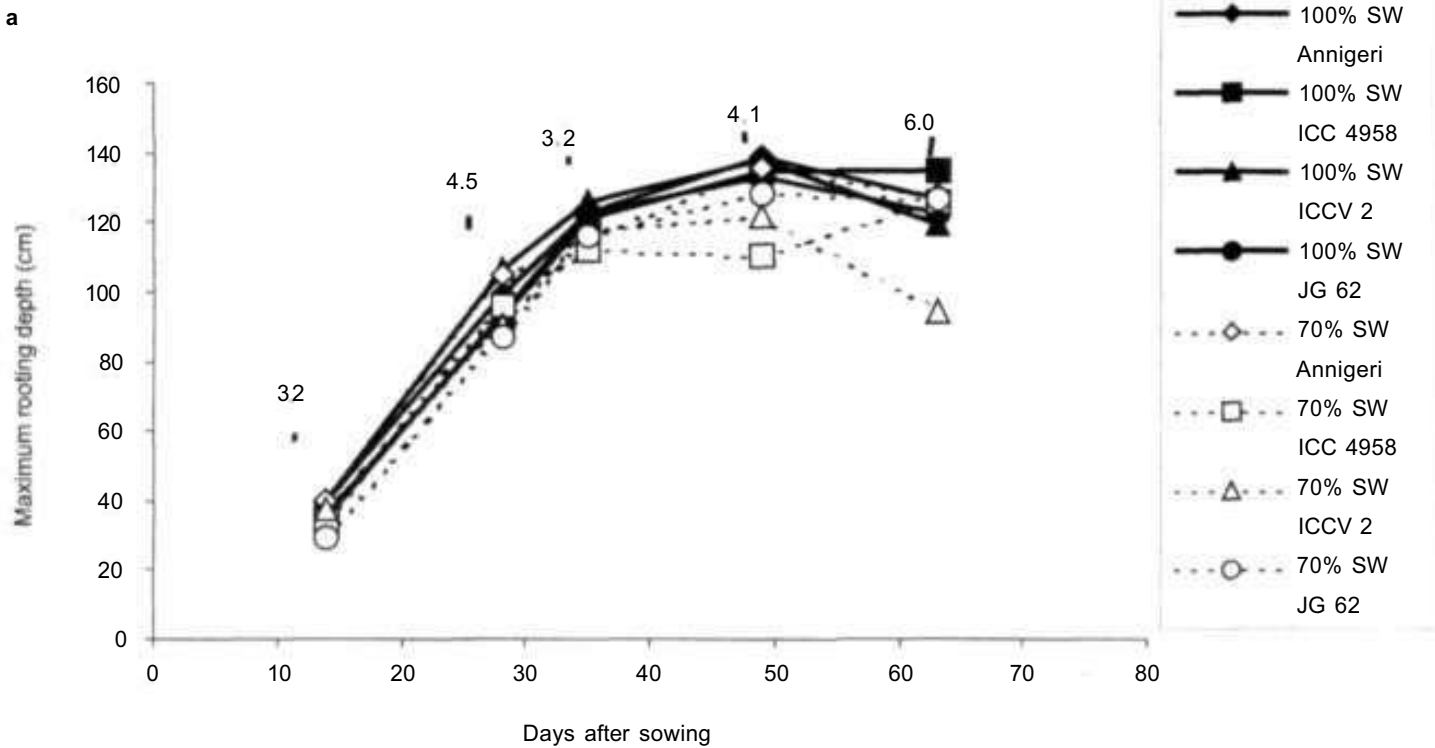
Root growth of four chickpea genotypes, viz., Annigeri, ICC 4958, JG 62, and ICCV 2 was evaluated in 2002 by growing plants in PVC cylinders (18 cm diameter, 120 cm long) under two moisture environments, in a randomized block design with five replications. The cylinders were filled with an equi-mixture of (w/w) Vertisol and sand, mixed with water to a level equivalent to 70% field capacity uniformly: (1) without further irrigation; or (2) irrigated adequately at 28 days after sowing (DAS). Two plants per cylinder were retained after thinning. The cylinders were wrapped with a thick layer of paddy straw to avoid direct solar heating. The root and shoot sampling was done at 14, 28, 35, 49, 63, and 77 DAS. The crop was sown late, on 17 January 2002.

The genotype ICCV 2 was the earliest in maturity and all the plants matured at 63 DAS. ICC 4958 was closer to maturity at this stage with very few pods. But Annigeri and JG 62 did not bear any pod and the plants shed most of the lower leaves at 77 DAS. Maximum rooting depth of the genotypes did not show any significant difference except at 49 DAS (Table 1; Fig. 1a). In the treatment with 70% field capacity soil moisture, ICC 4958 exhibited a plateau in gaining depth and ICCV 2 reached its maximum at this stage. However, at 63 DAS the rooting depth of ICC 4958, Annigeri, and JG 62 did not show any increase as they started showing symptoms of forced maturity by dropping most of the lower leaves. The plants grown irrigated showed relatively a normal pattern of flowering and pod filling until 63 DAS.

**Table 1. Analysis of variance and its significance for water regimes, genotypes, and sampling time and their interactions.**

Source of variation	Mean sum of squares and significance level <sup>1</sup>			
	Maximum rooting depth (cm)	Total root dry mass (g plant <sup>-1</sup> )	Total shoot dry mass (g plant <sup>-1</sup> )	Leaf area (cm <sup>2</sup> plant <sup>-1</sup> )
Water regime	2823***	0.566***	25.28***	60593***
Genotype	467**	0.334***	0.38**	4956***
Sampling time	58264***	2.891***	15.02***	33452***
Water regime x Genotype	474**	0.016 <sup>NS</sup>	0.05 <sup>NS</sup>	1927***
Water regime x Sampling time	158 <sup>NS</sup>	0.188***	4.64***	12494***
Genotype x Sampling time	381***	0.080***	0.19***	2658***
Water regime x Genotype x Sampling time	141 <sup>NS</sup>	0.009 <sup>NS</sup>	0.16**	1017***
Residual	90.0	0.13	0.073	228

1. Significant at \* = <0.05 level, \*\* = <0.01 level, and \*\*\* = <0.001 level; NS = Not significant.



**Figure 1.** Changes in root growth of four chickpea genotypes over the growing period at two soil water (SW) levels in cylinders: (a) maximum rooting depth; and (b) total root dry mass. (Note: Values are means of five replications. The vertical bars and the values are the standard errors for comparison of genotypes in a sampling time.)



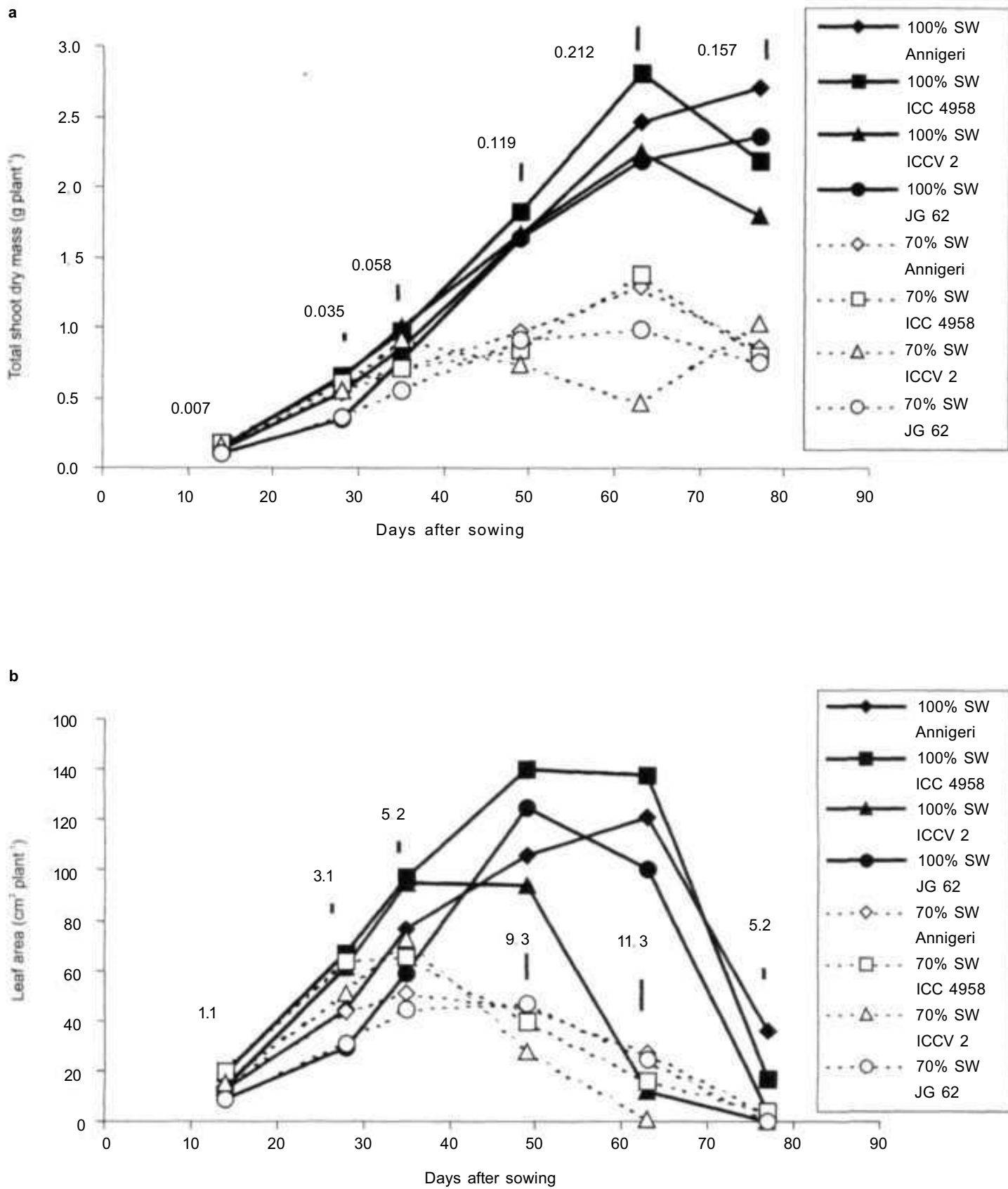


Figure 2. Changes in shoot growth of four chickpea genotypes over the growing period at two soil water (SW) levels in cylinders: (a) total shoot dry mass; and (b) leaf area. (Note: Values are means of five replications. The vertical bars and the values are the standard errors for comparison of genotypes in a sampling time.)

The root dry mass reached near maximum at 35 DAS in all the genotypes (Table 1; Fig. 1b). Root dry mass of field grown chickpea is known to continue until about 10 days to physiological maturity when grown under normal season (Krishnamurthy et al. 1996). The early cessation of root and shoot growth was likely due to the increasing temperature after mid-February. Root dry mass of ICCV 2 in 70% field capacity started declining after this stage and under irrigation at 49 DAS. After imposing the soil moisture treatments the positive irrigation response in root mass appeared in all genotypes. JG 62 produced significantly the least root biomass at 14 and 28 DAS whereas due to a rapid growth at later stages the difference was minimized and not significant. There was no difference in root dry mass among Annigeri, ICC 4958, and ICCV 2 in the early stages and between Annigeri and ICC 4958 in the later stages. ICC 4958 was previously shown to produce large root mass in the early stages of crop growth both at ICRISAT, Patancheru, India as well as in the spring sown conditions at the International Center for Agricultural Research in Dry Areas (ICARDA), Aleppo, Syria (Krishnamurthy et al. 1996, Saxena 2002). A comparison of the two genotypes ICCV 2 and JG 62 exhibits a contrasting temporal interaction for root mass. This provides a classic example on the significance of growth stage while defining the superiority of any genotype. ICCV 2 produced large root mass at early stages but JG 62 produced at later stages of crop growth.

The biomass production of shoot was similar to that of the root (Table 1; Fig. 2a). All the genotypes except ICCV 2 did not have enough time to pod and mature normally. Though there were flowers, the flowers did not set pods; the partitioning was very poor and consequently the plants remained green but lost the leaf area (Fig. 2b). ICCV 2 produced significantly high shoot biomass in the early stages and JG 62 the lowest, as the linear phase of the growth varied between these two genotypes. There was no difference between Annigeri and ICC 4958 in any of the stages. Under field conditions, the early growth vigor of ICC 4958, at least up to flowering over Annigeri

is apparent visually as well as by dry matter production. The absence of such a difference in shoot growth indicates genotype x environment interaction in this experiment.

In conclusion, genotypic variation was observed for root and shoot growth among the four cultivated genotypes. The linear growth phase of the genotypes was different leading to a crop growth stage x genotype interaction. Such interactions would create difficulties in identifying the best rooting progenies, as this superiority needs to be seen in a temporal context. The absence of a significant difference in root or shoot growth between extremely late planted Annigeri and ICC 4958, emphasizes the need for further comprehensive investigation of the whole germplasm collection to choose the best parental lines to identify quantitative trait loci (QTL) for the root traits across different growth conditions. The root growth of ICCV 2 at the early stages was good and therefore some of the existing RILs of JG 62 x ICCV 2, though were not consciously bred for, can also be expected to possess a better root system.

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# Pathology

## Status of Chickpea Diseases in Himachal Pradesh, India

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Chickpea (*Cicer arietinum*) is an important pulse crop grown in submontane, low hill subtropical zone of Himachal Pradesh, India. It is cultivated between 30°4' to 31°35' N and 71°5' to 76°55' E. The area under chickpea cultivation in Himachal Pradesh is 2500 ha (Anonymous 1998). It is mostly cultivated in rainfed situation on small farms. The average yield of chickpea is 700 kg ha<sup>-1</sup> and is below potential yields. Diseases are the most important factor limiting production. There is limited information available on diseases affecting this crop (Kapoor et al. 1991). Therefore, there is a need to conduct systematic surveys to determine the incidence of diseases affecting chickpea in Himachal Pradesh.

Surveys from 1992 to 1998 were conducted in the major chickpea-growing areas of Himachal Pradesh. Farmers' fields, demonstration plots in farmers' fields laid out by the extension division, research stations, and seed farms were surveyed in 7 districts at 161 locations. At each location, disease observations were recorded in 3 to 8 individual fields. In each field, number of total plants and plants infected by different diseases in one m<sup>2</sup> area at 10 randomly selected spots were counted. From these observations the average disease incidence in each district was calculated.

Fungal isolates were made from all plant parts of diseased plants and cultures were maintained in potato dextrose agar (PDA) medium. The pathogenicity of isolates of *Fusarium* spp and *Rhizoctonia solani* was tested on susceptible cultivar JG 62 by using pot culture inoculation methods developed by Nene and Haware (1980). For pot culture, inoculum was grown in a chickpea flour-sand mixture. This inoculum mixture was thoroughly mixed with autoclaved soil in a pot (1:20) and sowing was done 4 days later. Control plants were grown in a comparable mixture of non-infected sand and chickpea flour and autoclaved soil.

Inoculum of leaf pathogen was produced on PDA and chickpea seed meal dextrose agar. Aerial parts of the plants were sprayed uniformly with a spore suspension of pathogens. The plants were then covered with transparent

polyethylene bags for 4 days and incubated at 23±2°C with 12 h photoperiod. Disease assessment for leaf pathogen was made at 20 and 40 days after inoculation. Experiments with each of the pathogens were replicated 5 times, a 15-cm pot with eight seedlings forming a replication.

Eight fungal and four viral diseases were identified. The incidence of diseases varied in the districts (Table 1). Most of these diseases have been reported previously (Kapoor et al. 1991); however, cucumber mosaic virus (CMV), bean yellow mosaic virus, and a gemini virus were recorded for the first time from Himachal Pradesh. The diseases that were encountered during the survey period are described below.

**Collar rot.** Collar rot (*Sclerotium rolfsii*) was observed in farmers' fields at all the locations. In infested soil, germinating seeds are attacked in the pre-emergence phase. Seedlings and young plants get infected at the collar region and the affected plants dry up. The incidence decreases with the age of the crop. The disease is favored by good soil moisture and high soil temperature. Optimum temperature for disease development is 30°C (Mathur and Sinha 1968).

**Wilt.** Wilt was first reported from India (Butler 1918). *Fusarium solani*, *F. oxysporum* f. sp *ciceris*, *R. solani*, and *R. bataticola* were isolated from wilted plants. The incidence of *F. solani* was high although *F. oxysporum* was mainly associated with chickpea wilt. The disease occurred at all the stages of plant growth. High incidence was noticed at Una and Sirmour where temperature is normally high (>24°C).

**Black root rot.** Black root rot is caused by the fungus *F. solani*. The disease was observed at all stages of plant growth. Excessive moisture and moderately high temperatures (25 to 30°C) encourage disease development (Nene and Reddy 1987). High incidence of disease was observed at Bilaspur.

**Stem rot.** Stem rot is caused by *Sclerotinia sclerotiorum*. The disease can affect the crop at any stage. However, maximum incidence was noticed in February and March when the crop canopy covered the ground below the crop. Excessive vegetative growth, high soil moisture, and cool weather (20°C) favor disease development.

**Dry root rot.** Dry root rot caused by *R. bataticola* was a serious disease in non-irrigated chickpea-growing areas. The incidence of disease was more in Una and Sirmour districts.

**Table 1. Incidence (%) of fungal and viral diseases in chickpea in submontane, low hill subtropical zone of Himachal Pradesh, India, 1992-98.**

District	No. of locations surveyed	Wilt	Black root rot	Collar rot	Stem rot	Dry root rot	Wet root rot	Blight	Gray mold	Viruses <sup>1</sup>		
										a	b	c
Kangra	9	0.00	0.65	12.17	0.72	0.00	0.63	0.00	0.00	0.00	0.03	0.00
Hamirpur	17	0.51	1.96	13.33	2.50	0.72	0.01	0.00	0.00	0.01	0.00	0.00
Una	20	0.72	2.58	6.70	3.30	4.86	0.00	0.70	0.00	0.01	0.01	0.00
Sirmour	30	0.63	1.72	20.37	5.00	3.04	0.06	0.65	0.06	0.63	0.02	2.00
Solan	25	0.31	2.32	15.50	3.50	2.32	0.02	0.63	0.06	0.50	0.00	0.00
Bilaspur	45	0.52	4.86	20.56	4.25	2.58	0.65	0.55	0.02	0.31	0.02	5.00
Mandi	15	0.00	1.50	7.80	2.25	0.00	0.01	0.00	0.00	0.00	0.00	0.00

1. Symptoms: a = stunting; b = mild mosaic; c = reduced terminal buds.

**Wet root rot.** Wet root rot caused by *R. solani* was observed mainly in Kangra and Bilaspur area. Although it was a minor disease the incidence was more in fields having higher moisture content.

**Blight.** Among the leaf pathogens, blight caused by *Ascochyta rabiei* was most important and caused considerable losses. The disease appeared in epiphytotic form in parts of Himachal Pradesh, Punjab, Haryana, and Uttar Pradesh in 1969, due to appearance of a new race of the pathogen (Vir and Grewal 1974). During the survey it was observed mainly in the seed farms and research stations because chickpea is grown normally in the same field every year. However, the disease occurrence was seldom in farmers' fields. The disease incidence was high at flowering period in February and March when mean maximum temperature was 22°C and minimum temperature was 5°C and humidity was high.

**Gray mold.** Gray mold was a minor disease caused by *Botrytis cinerea*. It was observed only in Sirmour, Solan, and Bilaspur, and the incidence was very low (1%).

**Viral diseases.** Plants showing viral disease symptoms such as stunting, mild mosaic, and reduced terminal buds were collected and maintained by periodic inoculation on chickpea cultivar HPG 17. Identification of virus isolates were made on the basis of reaction on diagnostic hosts, transmission, and serological tests. Diseased plants were tested by the direct antigen coating enzyme linked immunosorbent assay (DAC-ELISA) method (Hobbs et al. 1987) with antisera of the luteo virus, pea leaf roll virus (PLRV), and CMV. Of the 86 samples assayed, four reacted positively with the CMV antiserum and 14 reacted

with PLRV antiserum. Samples that showed stunting but did not react with the antiserum of PLRV were sent to ICRISAT, Patancheru, Andhra Pradesh, India for further identification. The pathogen was identified as chickpea chlorotic dwarf virus (CCDV), a leaf hopper transmitted gemini virus.

The presence of CMV was also confirmed by the symptoms produced on tobacco cultivar Xanthi (systemic infection) and on *Chenopodium amaranticolor* (local lesions) after mechanical sap inoculation. The symptoms incited by CMV on chickpea were similar to those described by Dhingra et al. (1979) and Singh et al. (1994). The symptoms of chickpea stunt caused by PLRV and CCDV were same, both causing stunting of plants due to shortening of internodes and phloem browning and were similar to those described by Horn et al. (1996). Both the viruses were not sap or seed transmissible.

The incidence of CMV in commercially cultivated chickpea cultivar HPG 17 (bold seeded) and C 235 (small seeded) remained low (<1%) but was higher in those plots where nearby plots were grown with cucurbitaceous crops. Chickpea stunt was observed in all chickpea-growing areas of Himachal Pradesh. The incidence ranged from 2 to 5% in farmers' fields. At the Regional Research Station in Dhaulakuan and Pulses Research Station in Berthin, it reached up to 20% in few entries.

**Acknowledgment.** The authors are thankful to Dr DVR Reddy, Principal Scientist, ICRISAT, Patancheru for identification of CCDV and Dr YS Paul, Senior Scientist, Plant Pathology, Himachal Pradesh Krishi Vishwa-vidyalaya, Palampur for his help in identification of fungal diseases.

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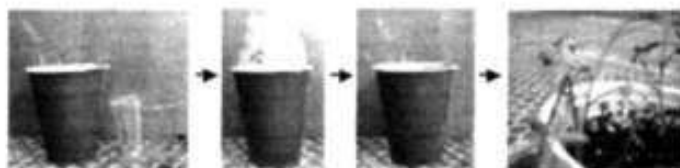
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## An Improved Technique for Virulence Assay of *Ascochyta rabiei* on Chickpea

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A reliable quantitative bioassay is required to study host-pathogen interactions. A number of screening techniques have been reported for ascochyta blight of chickpea (*Cicer arietinum*) caused by *Ascochyta rabiei* (Reddy et al. 1984, Haware et al. 1995). However, it has been a problem to get desired results using those techniques under our growth chamber and greenhouse conditions. This study was initiated to develop a reproducible technique for virulence assay of *A. rabiei* on chickpea.

Bioassays were carried out in a Conviron growth chamber or in a greenhouse set at 20°C day and 16°C night temperature regimes (Trapero-Casas and Kaiser 1992). Two-week old seedlings of chickpea were sprayed with conidia ( $1 \times 10^5$  spores  $\text{ml}^{-1}$ ) to incipient runoff. Inoculated seedlings were immediately covered with a transparent plastic cup to form a mini-dome. The purpose of the mini-dome is to provide a uniform high level of relative humidity for infection to occur. Hence, this improved screening method is called the mini-dome technique (Fig. 1). The mini-domes were removed after 24 h. Disease severity was rated 14 days after inoculation. Two methods were used to rate disease severity. The first



**Figure 1. Illustration of the mini-dome technique: (from left) two-week old chickpea seedlings sprayed with conidia ( $10^5$  spores  $\text{ml}^{-1}$ ) of *Ascochyta rabiei*; inoculated seedlings covered with a plastic cup to form a mini-dome; mini-dome removed after 24 h; and disease severity evaluated 14 days after inoculation.**

method was visual rating using the 1-9 rating scale of Reddy and Singh (1984). In the second method, the number of leaves showing symptoms as well as the total number of leaves on each plant were counted. The percentage of infected leaves was then calculated.

Initially the mini-dome technique was tested for various time periods (0, 6, 12, 24, and 48 h) for which

plants were covered by the mini-dome. It was shown that covering for 24 h was sufficient for infection to occur. Subsequently 24 h cover under the mini-dome was employed in all experiments. Then seven inoculum concentrations ( $0, 10^2, 10^3, 10^4, 10^5, 10^6,$  and  $10^7$  spores  $ml^{-1}$ ) were tested on four host germplasm lines Dwelley, FLIP 84-92C, PI 359075, and Spanish White. On the

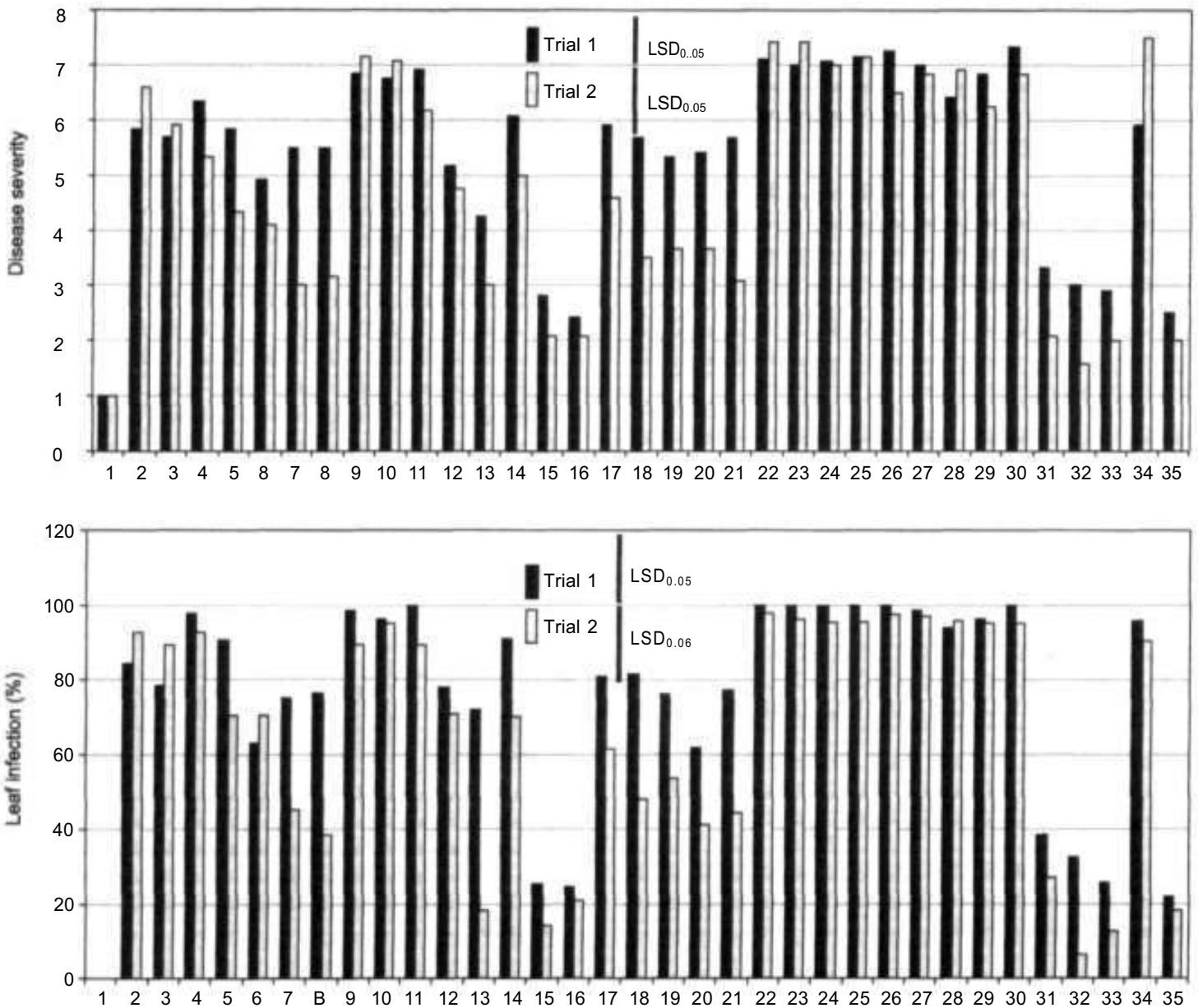


Figure 2. Comparison of *Ascochyta rabiei* isolates in two virulence trials on cultivar Dwelley using the mini-dome technique, and correlation of the two disease rating methods (visual rating of 1 to 9 scale, and leaf count expressed as percentage of infected leaves). (Note: Each bar represents a mean of six replications. Isolate code 1: non-inoculated control; Codes 2 to 12: isolates from Pullman, Washington, USA; Codes 13 and 14: isolates from Genesee, Idaho, USA; Codes 15 and 16: isolates from Walla Walla, Washington; Codes 17 to 21: isolates from Fresno, California, USA; Codes 22 to 30: isolates from Sutter County, California; Codes 31 to 35: isolates from Walt Kaiser's archival collection, four from USA and one from Syria.)

susceptible lines PI 359075 and Spanish White, inoculum concentration of  $10^4$  spores  $\text{ml}^{-1}$  caused significant disease. On the resistant lines Dwelley and FLIP 84-92C, inoculum concentration of  $10^5$  spores  $\text{ml}^{-1}$  caused appreciable disease. A spore concentration of  $10^5$  spores  $\text{ml}^{-1}$  was, therefore, chosen as a standard concentration for all experiments.

A set of 34 isolates from various chickpea-growing areas in USA (one isolate from Syria) was tested twice on cultivar Dwelley using the mini-dome technique. Considerable pathogenic variation was detected among the 34 isolates (Fig. 2), but very little variation was observed among replications of a given isolate. The disease severity based on visual rating highly correlated with the severity rating based on percentage of infected leaves ( $r = 0.88$ ). The isolates that were highly virulent in the first experiment remained the most pathogenic in the second experiment (isolate codes 9 to 11, 22 to 30, and 34) (Fig. 2) as well, and also the isolates that were less virulent in the first experiment remained only slightly pathogenic in the second experiment (isolate codes 15, 16, 31 to 33, and 35) (Fig. 2). Results also showed pathogenic variation related to geographic locations. For example, the nine isolates, 22 to 30, from Sutter County, California were consistently more virulent than the five isolates (17 to 21) from Fresno, California (Fig. 2).

The mini-dome technique does not require any expensive equipment and is easy to carry out. This technique gives reproducible results and much reduced level of variation among replications of treatments, which will enhance sensitivity of the bioassay in detecting pathogenic variations among isolates. The two disease rating methods (visual rating vs leaf counting) gave similar results and were highly correlated. The visual rating method is simpler than the leaf counting, but it requires experience and can be subjective. The leaf counting method is time-consuming, but is more objective than the visual rating method. This mini-dome technique, being simple and reproducible, could enable to study the genetics of pathogenicity of *A. rabiei*. It can also be implemented in screening progenies in resistance breeding.

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## Entomology

### Effect of *Helicoverpa* Nuclear Polyhedrosis Virus on Pod Borer Larvae in Chickpea Crops in Bangladesh

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A major constraint to chickpea (*Cicer arietinum*) in the High Barind Tract (HBT) of Bangladesh, and wherever the crop is grown in South Asia, is the gram (chickpea) pod borer (*Helicoverpa armigera*), which can damage all the pods under severe infestation. The larvae can be killed by a range of chemical insecticides, if applied before they grow too large. But reliance on protection with such synthetic chemicals is not recommended because the insect can develop resistance to these chemicals. Chemical insecticides have toxic effects on natural enemies of pod borer and other beneficial organisms and there are toxicity hazards to humans. Therefore, integrated pest management (IPM) techniques offer an ecologically safe approach to management of pod borer. The use of *Helicoverpa* nuclear polyhedrosis virus (HNPV), which is specific to *H. armigera* and harmless to other organisms, is a potential biological pesticide for managing this pest (Ranga Rao and Rameshwar Rao 2001). This could be combined with other components of IPM for *Helicoverpa* currently being used, or under test, for chickpea in the HBT. These include: early recognition of pod borer infestation so that effective action can be taken; intercropping of chickpea with linseed (*Linum usitatissimum*), coriander (*Coriandrum*

**Table 1. Effect of spraying HNPV on number of *Helicoverpa armigera* larvae on chickpea in farmers' fields at different locations in the High Barind Tract of Bangladesh during 2001/02 season.**

Spray period (2002)	Number of comparisons	Sample size per plot	Unit of larval density	Number of larvae		Statistical significance <sup>1</sup>
				In plots sprayed with HNPV	In unsprayed plots	
5 & 23 Jan	4 <sup>2</sup>	5 m x 1 m	Larvae m <sup>2</sup>	0.8	27.1	P <0.001
24 Jan-10 Mar	34 <sup>2</sup>	5 m x 1 m	Larvae m <sup>2</sup>	1.1	5.0	P <0.05
17-30 Mar	17 <sup>3</sup>	5 x 10 plants	Larvae on 10 plants <sup>4</sup>	15.2	53.2	P <0.001

1. According to paired "t" test.

2. Two sprays were given at 1-2 week intervals.

3. One spray was given.

4. There were, on average, about 10 plants m<sup>-2</sup> and thus the values given approximate larvae m<sup>2</sup>.

*sativum*), and/or barley (*Hordeum vulgare*) to encourage natural enemies and discourage oviposition of *Helicoverpa* moths; placement of bird perches to encourage birds to feed on larvae; and picking of larvae by children if larvae grow too large for either HNPV or chemicals to be effective.

The key to this IPM strategy, however, is the effectiveness of HNPV and having enough of it to meet demand. The People's Resource Oriented Voluntary Association (PROVA), a non-governmental organization promoting rainfed agriculture in the HBT of Bangladesh, has embarked on a program to test the efficacy of HNPV application in managing pod borer on chickpea in the HBT and its commercial production and distribution. The protocol for HNPV production and use described by Ranga Rao and Rameshwar Rao (2001) was followed. To multiply the virus inoculum, larvae of *H. armigera* were collected from fields of chickpea, pigeonpea (*Cajanus cajan*), and tomato (*Lycopersicon lycopersicum*), and infected with the HNPV inoculum originally derived from ICRISAT, Patancheru, India. Dead larvae, in which the virus had multiplied, were blended and the virus concentrate extracted after centrifugation. The HNPV was applied by knapsack sprayer to chickpea fields at a rate of 12 drops of HNPV extract per 12 liters of water. Fields were sprayed at dusk, and 12 ml of "Robin Blue" was added to the solution, to prevent damage to HNPV by ultraviolet radiation. Farmers' fields, of usually around 1 *bigha* area (7.5 *bigha* - 1 ha), were equally divided and one half was sprayed with HNPV while the other half was not sprayed. A total of 19 *bigha* was sprayed at 7 locations in Godagari, Nawabganj Sadar, and Nachole Upazilas of the HBT. Data on larval numbers were recorded at 5-6 days after spray application. Spray applications were made during three periods during January, February, and March 2002.

Spraying of HNPV effectively reduced the number of *H. armigera* larvae on chickpea in farmers' fields (Table 1). It was noted that larval density in unsprayed plots was initially high, with small larvae, decreased during February, and again increased during March. These results show promise for use of HNPV in pod borer management but further testing in the 2002/03 season is necessary, including measurement of effects of HNPV application on actual grain yield. The production system for HNPV also needs to be scaled up and its commercial viability, in comparison with reliance on chemical pesticides, evaluated.

**Acknowledgments.** We thank Dr GV Ranga Rao, ICRISAT, for providing training to PROVA staff, along with staff of other organizations in Rajshahi, Bangladesh in the production and use of HNPV, in December 2000. He also provided the mother culture of HNPV from which subsequent batches of HNPV solution were produced for use in these studies. We are grateful to Babul Aktar and Omar Faroukh for competent technical assistance in the conduct of these studies, and the farmers on whose fields the trials were conducted. This work is the output from Plant Sciences Research Programme Project R7540 funded by the UK Department for International Development (DFID) and administered by the Centre for Arid Zone Studies, University of Wales, UK for the benefit of developing countries. The views expressed are not necessarily those of DFID.

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### Functional Genome Analysis Using DDRT for Ascochyta Blight Resistance in Chickpea

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Chickpea (*Cicer arietinum*) (2n=2x=16) is the third most important pulse crop worldwide and first in India (FAO 1998). Despite its importance, conventional breeding has increased yields only 0.6% annually in recent years. Growth in productivity has been low mainly due to widespread susceptibility to ascochyta blight caused by *A. rabiei* that has the potential to cause 100% yield loss in chickpea (Nene 1984). To analyze chickpea-*Ascochyta rabiei* interaction, we studied the genes that are up- and/or down-regulated during infection by the pathogen. To determine the gene expression profile during infection by pathogen. Differential Display Reverse Transcription (DDRT) approach was deployed where many RNAs can be simultaneously analyzed (Liang and Pardee 1992). A comparative analysis was performed of expression patterns of resistant and susceptible cultivars upon infection by the pathogen.

Ten-day-old seedlings of FLIP 84-92C (ascochyta blight resistant cultivar of *C. arietinum*) and PI 489777 (ascochyta blight susceptible accession of *C. reticulatum*) were inoculated with  $1 \times 10^6$  conidia ml<sup>-1</sup> suspension of a virulent strain of *A. rabiei* (A20) in the mist chamber. The mist chamber, measuring 66 cm in height, 121.5 cm wide, and 95 cm deep, was constructed and covered with 6 mil clear polyethylene. The mist control system was known as "Automatic Misting System" (Phytotronics, USA) and had tork brand timers. Control plants were sprayed with water outside the mist chamber. Leaf samples were collected from control and infected seedlings on 1, 2, 3, 7, and 8 days after inoculation. The plants started showing disease symptoms after 7 days. Total RNAs were extracted from all the samples separately using RNeasy Kit from Qiagen, Valencia, California, USA. RNAimage, involving components for reverse transcription as well as polymerase chain reaction (PCR) amplification, was obtained from GenHunter, Nashville, Tennessee, USA for DDRT analysis.

The DDRT products were analyzed on 6% polyacrylamide gels and silver stained (Fig. 1). The differentially expressed bands were extracted from the gel, reamplified using the same primers and run on 1% agarose gels. Fragments eluted from the agarose gels were cloned into the pGEM-T easy plasmid vector (Promega, USA). Sequencing of the cloned cDNA fragments was performed on an AB1 Prism 377 DNA sequencer (Applied Biosystems, USA) using the dideoxy sequencing method with T7 universal primer. The homology search was carried out using [www.ncbi.nlm.nih.gov/BLAST](http://www.ncbi.nlm.nih.gov/BLAST).

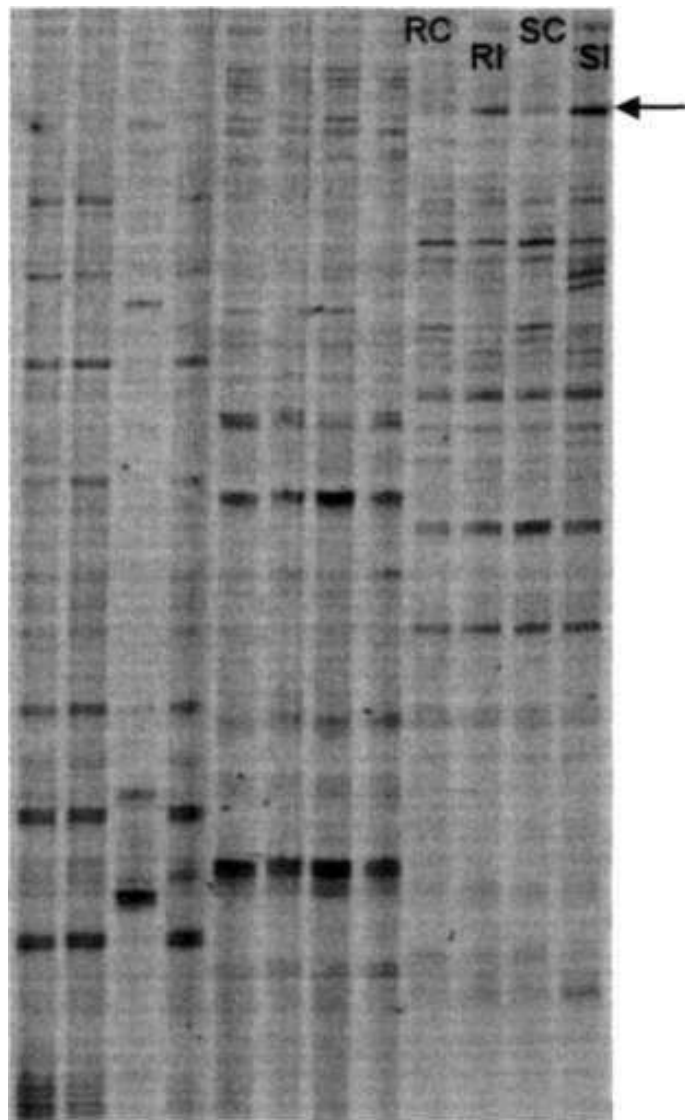


Figure 1. A representative silver stained gel picture of DDRT products using H-AP7 and H-T<sub>(11)</sub>G primer. (Note: Arrow indicates upregulated partial cDNAs; RC = Resistant control; RI = Resistant infected; SC = Susceptible control; SI = Susceptible infected.)



Pooled mRNA samples of resistant control, resistant infected, susceptible control, and susceptible infected were used for DDRT analysis. This strategy permitted the mRNAs which are differentially expressed at low level at a given time to amplify. Important modifications in our DDRT study are the exclusions of radioactive dNTP in the PCR and visualization by autoradiography. Cold PCR reactions were run and the products were visualized by employing silver staining on 6% polyacrylamide gels (Fig. 1). Some DDRT reactions were performed with Resistant Gene Analog (RGA) primers in place of arbitrary primers. We amplified two partial cDNA clones using primer pairs H-T<sub>(11)</sub>A and H-AP26, and H-T<sub>(11)</sub>A and Pto kin 1.

Tentative identities of these clones were established when they showed 87% homology with serine hydroxy methyl transferase and 88% homology with aldolase of pea (*Pisum sativum*) (Figs. 2 and 3). Both sequences have been submitted to Genbank and their numbers are AF416481 and AF416480, respectively.

Aldolase is a prerequisite for the glycolytic/gluconeogenic pathway as well as the pentose phosphate cycle and Calvin cycle in plants. Serine hydroxy methyl transferase (SHMT) is a key enzyme in photorespiration. This is the first report on SHMT sequences in chickpea although complete cDNA sequences of the aldolase gene from chickpea are available (AJ005041). The role of these *Ascochyta* responsive genes in blight resistance needs to be established.

As large sequence databases become available for plants, the number of genes to be monitored becomes too large for traditional analyses such as northern blots. DDRT is a cost effective and an efficient technique that covers 96% of expressed genes at a given time. Further extensive analysis using advanced methods such as microarrays at the expression level will reveal the responses of various known genes to infection by *A. rabiei*. Knowledge on the behavior of different genes during chickpea-*A. rabiei* interaction will ultimately facilitate isolation of blight resistance genes. To our knowledge, this is the first report of efforts to study the blight resistance genes using DDRT technique.

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## Enzymatic Degradation of Oligosaccharide Content of Chickpea

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Chickpea (*Cicer arietinum*) is a unique legume because it is used to prepare a variety of food products in different parts of the world. A number of nutritional, biochemical, and biotechnological investigations on chickpea have been reported (Singh 1985). Flatulence is caused by oligosaccharides such as raffinose, stachyose, and verbascose. In the human digestive tract these oligosaccharides are not digested, since the intestinal juice lacks  $\alpha$ -galactosidase. Microorganisms in the large intestine ferment these sugars that lead to flatus (Price et al. 1988). The enzyme  $\alpha$ -galactosidase hydrolyzes raffinose, stachyose, and verbascose. Commercial production of chickpea flour free from oligosaccharides using  $\alpha$ -galactosidase would add value and could expand the use of chickpea as an excellent source of cholesterol free vegetable protein. Crude preparation of  $\alpha$ -galactosidase from microbial sources have been used to hydrolyze the oligosaccharides in soymilk (Mulimani and Ramalingam 1995). However, the crude preparation of  $\alpha$ -galactosidase from a microbial source increases the cost of production of legume flour free from flatulence-causing sugars. We report here a commercial application of crude  $\alpha$ -galactosidase from guar (*Cyamopsis tetragonoloba*) seeds that can be used to remove oligosaccharides from chickpea flour.

The chickpea cultivar Annigeri was collected from the Agricultural Research Station, Gulbarga, Karnataka, India. Oligosaccharide concentration was determined in 100 g of powdered chickpea Hour according to the method of Tanaka et al. (1975).

Whole raw chickpea seed (100 g) was soaked in distilled water (1L) at 4, 8, 12, 16, and 20 h. The soaked water was decanted and replaced with fresh water. Whole chickpea seed (100 g) was cooked in distilled water (1 L)

at 1:10 ratio for 20, 30, 40, 50, and 60 min. Five grams of chickpea flour (fraction which passes through 600  $\mu\text{m}$  sieve) was treated with 50 ml of crude  $\alpha$ -galactosidase of germinating guar (0.45 units  $\text{ml}^{-1}$ ). The above mixture was placed in a waterbath maintained at 45°C for 2 h with occasional stirring. For the control, 50 ml of phosphate buffer (0.1 M, pH 6.8) was added instead of the enzyme solution to 5 g of chickpea flour.

Soaking for 16 h resulted in the mean decrease of 76.3% for verbascose plus stachyose and 75% for raffinose (Fig. 1). The removal of verbascose plus stachyose is known to be stronger in the increase of flatulence than raffinose (Price et al. 1988). Cooking of chickpea for 60 min showed mean reduction of 29.6% for verbascose plus stachyose and 52.3% for raffinose (Fig. 1). Iyengar and Kulkarni (1977) observed 59.4% reduction in raffinose family sugars in chickpeas after cooking. The treatment of chickpea flour with crude  $\alpha$ -galactosidase from guar seeds resulted in average reduction of 89.6% for verbascose plus stachyose and 88.5% for raffinose over control experiments (Fig. 1). Shivanna et al. (1989) have reported the reduction of raffinose family sugars present in soymilk with partially purified  $\alpha$ -galactosidase from germinating guar and observed 80% and complete hydrolysis of stachyose and raffinose respectively by 30 min incubation. This is the first report on the use of  $\alpha$ -galactosidase from guar for the hydrolysis of oligosaccharides present in chickpea flour. Crude enzyme treatment was efficient in elimination of galacto-oligosaccharides from chickpea than cooking and soaking techniques. The guar seed was chosen as a source of enzyme because of its easy and

abundant availability. It is also a rich source of enzyme. Although the crude enzyme treatment reduced the levels of oligosaccharides, the acceptability of final product, cost, safety, and palatability of enzyme treated flour need to be determined before commercial application of this process. Also suitability of using the enzyme treated flour in preparation of traditional dishes should be established before scale-up process.

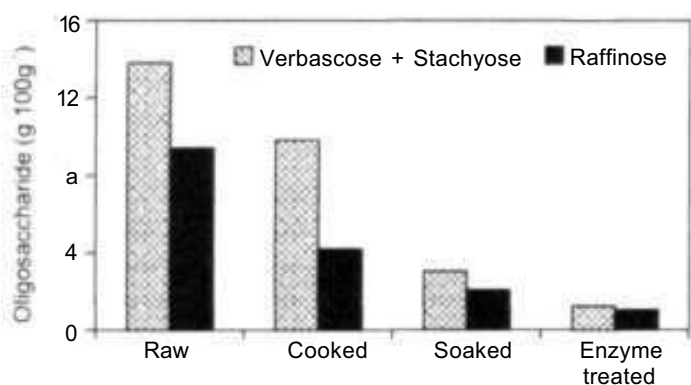
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## Diversity in Advanced Breeding Lines of Chickpea

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The diversity in germplasm can be assessed at morphological (Dasgupta et al. 1987, Kumar et al. 1998) as well as at protein or DNA levels. Morphological characters are generally environment sensitive whereas seed storage protein is more stable. In this investigation, morphological diversity in advanced breeding lines of chickpea (*Cicer arietinum*) was assessed based on Tocher's method using



**Figure 1. Mean level of verbascose plus stachyose and raffinose in chickpea seed with different treatments: raw (whole seed), cooked (60 min), soaked (16 h), and flour with enzyme ( $\alpha$ -galactosidase).**

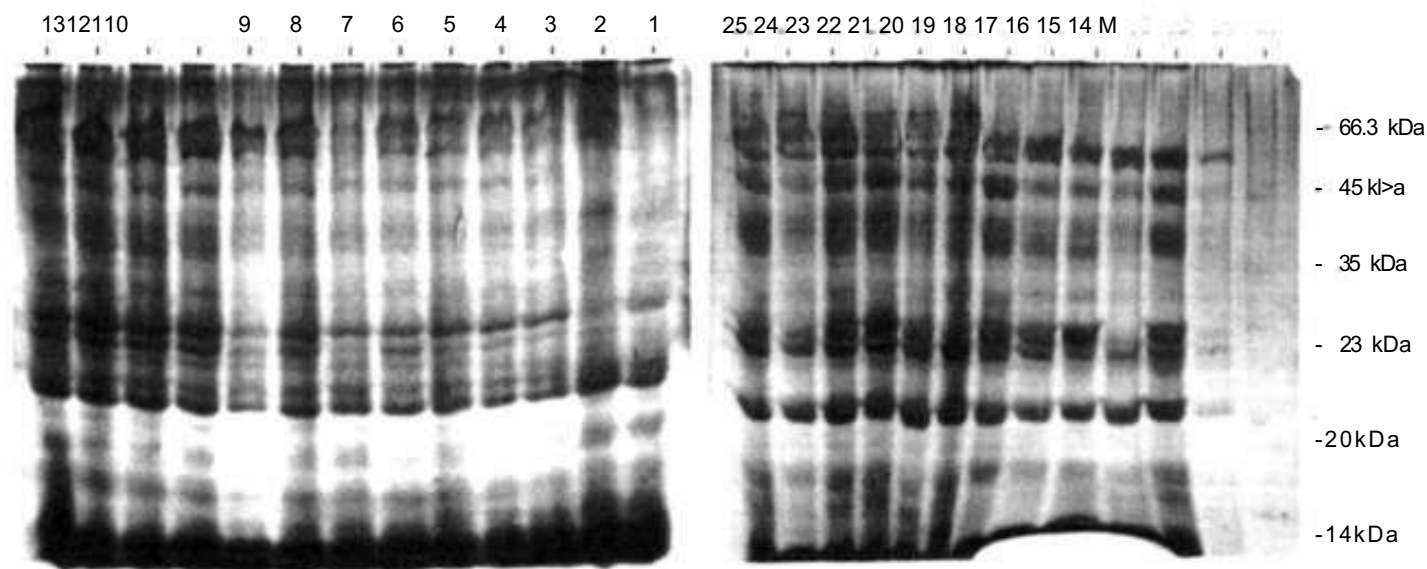
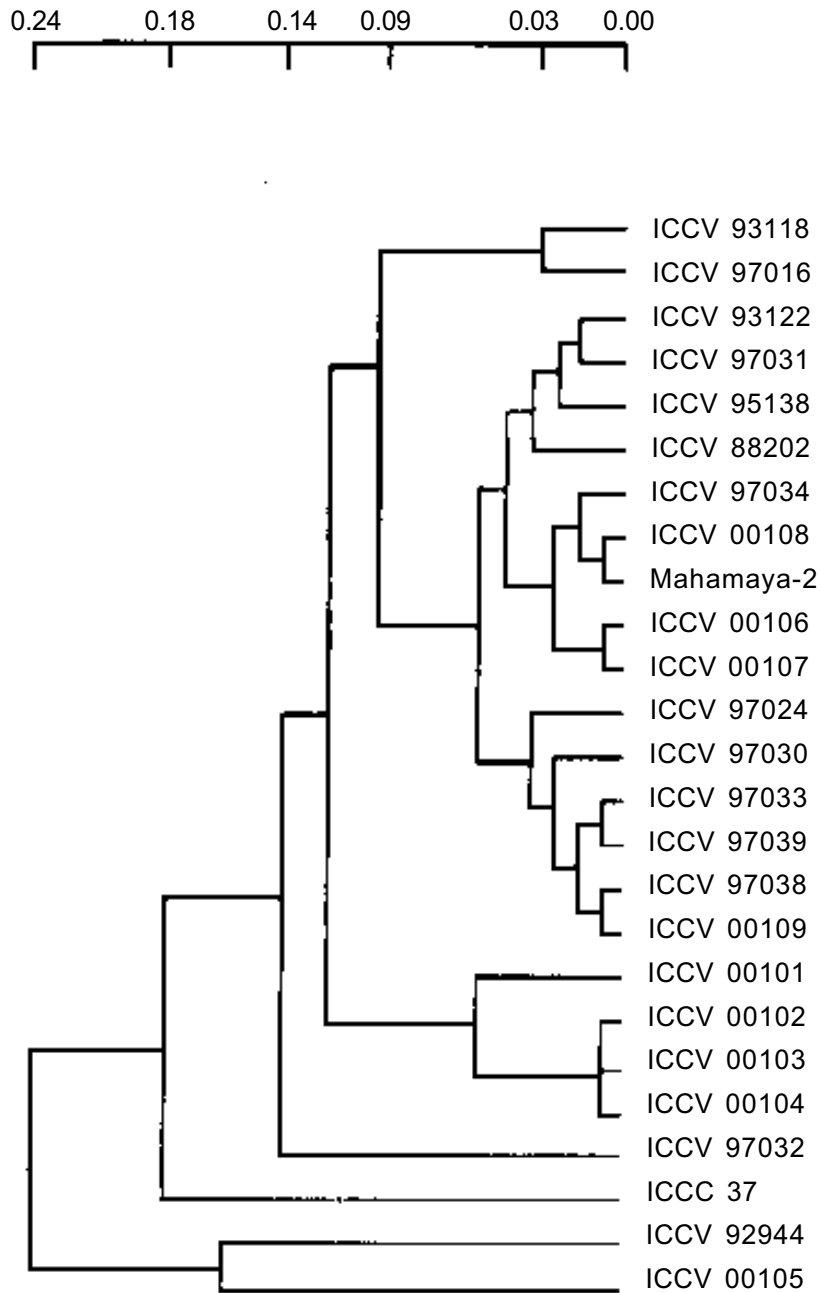


Figure 1. SDS-PAGE analysis of total protein of 25 chickpea genotypes along with marker protein (M). (Note: Names of entry numbers 1 to 25 are given in Table 1.)

Table 1. Chickpea genotypes exhibiting parentage and clustering pattern at morphological and molecular levels.

Entry no.	Entry name	Parentage	Clustering pattern	
			At morphological level	At protein level
1	ICCV 93118	R476M x ICCL 85216	I	I
2	1CCV 93122	(Annigeri x ICC 506-EB) x (Annigeri x ICC 12237)	II	II
3	ICCV 95138	(ICCC 42 x ICC 1069) x CT Line 2112	VIII	II
4	ICCV 97016	Dhanush x BG 276	IV	I
5	ICCV 97034	(AKG 33 x ICC 4958) x (ICCC 42 x ICCV 10)	III	II
6	ICCV 97024	ICCL 82108 x Annigeri	MI	III
7	ICCV 97030	(BBN 9-3 x Avrodhi) x (GF 16 x ICCL 82108)	IV	III
8	ICCV 97031	(JG 62 x ICC 12237) x ICC 12237	II	II
9	ICCV 97032	ICCC 42 x ICCV 10	I	V
10	ICCV 97033	(ICCV 10 x K 850) x (ICCV 89230 x JG 74)	11	III
11	ICCV 97038	(ICCV 10 x ICC 10448)	III	III
12	ICCV 97039	(Annigeri x GW 5/7) x (ICC 12237)	VI	III
13	ICCV 88202	PRR 1 x ICCC 1	II	II
14	ICCV 92944	(GW 5/7 x P 326) x ICCL 83149	V	VII
15	ICCV 00101	IG 9216 x ICCV 10	V	IV
16	ICCV 00102	IG 9215 x ICCV 10	VII	IV
17	ICCV 00103	JG 74 x ICCL 83105	II	IV
18	ICCV 00104	JG 74 x ICCL 83105	III	IV
19	ICCV 00105	Kalburgi x ICCV 2	III	VII
20	ICCV 00106	Kalburgi x ICCV 10	III	II
21	ICCV 00107	IG 9216 x ICCV 10	I	II
22	ICCV 00108	IG 9216 x ICCV 10	II	II
23	ICCV 00109	IG 9216 x ICCV 10	IV	III
24	ICCC 37 (common check)	NA <sup>1</sup>	III	VI
25	Mahamaya-2 (local check)	NA	I	II

1. NA = Information not available.



**Figure 2. Dendrogram of 25 chickpea genotypes based on similarity index in SDS-PAGE analysis.**

Mahalanobis  $D^2$  distance statistics (Rao 1952). Genetic diversity was also measured from seed storage protein profile banding through SDS-PAGE method. It is useful to assess the correlation between both the methods and the diversity of lines generated through the hybridization program.

The experimental materials comprised 23 advanced breeding lines of short-duration desi chickpea and 2 control cultivars (Table 1). The seeds were sown on 11 December 2000 in randomized complete block design with 3 replications. Each plot had an area 4 m x 1.5 m.

Observations were recorded on ten randomly selected plants in each replication. Data were recorded on plant height (cm), days to 50% flowering, days to maturity, number of branches per plant, number of pods per plant, pod length (cm), number of seeds per pod, 100-seed mass (g), harvest index (%), and seed yield per plant (g). To study seed protein polymorphism, one dimensional SDS-PAGE (15% separating gel and 4% stacking gel) was carried out following Laemmli (1970) in a mini-vertical gel system.

The genotypes were significantly different for all ten characters as indicated by ANOVA. The inter-varietal  $D^2$  values were computed for all possible 300 pairs of comparison and ranged from 94.89 (between ICCV 93118 to ICCV 95138) to 76065.95 (between ICCV 93118 and ICCV 00107).  $D^2$  values between varieties were quite high in majority of comparisons indicating high variability among the lines. Using  $D^2$  values, the lines could be grouped into 8 clusters (Table 1). Cluster II consisted of 6 lines, while clusters VI, VII, and VIII each comprised of single line. Inter-cluster distance ( $D^2 = 70323.51$ ) was maximum between clusters V and VIII, while it was minimum between clusters II and III ( $D^2 = 2105$ ). SDS-PAGE indicated that the band numbers 5, 6, 7, 8, 13, 17, and 20 having relative mobility (Rmf) values 0.175, 0.213, 0.288, 0.363, 0.563, 0.70, and 0.85 respectively were present in all lines. Protein bands 2, 3, 8, 9, 11, and 16 having molecular weight less than 45 kDa were found to be more promising in distinguishing chickpea lines as these bands were present in a few lines (Fig 1). The genetic similarity between lines i and j were calculated as  $S_{ij} = 2a/(2a+b+c)$  (Nei and Li 1979) where 'a' is the number of bands present in both samples i and j, 'b' is the number of bands present in i and absent in j, and 'c' is the number of bands present in j and absent in i. The resulting similarity matrix was used for construction of a dendrogram by UPGMA method (Sneath and Sokol 1973) and the lines were grouped into 7 clusters (Fig. 2). Cluster II consisted of 9 lines while cluster V and VI consisted of single line. It is interesting to note that in cluster II some breeding lines developed from a single common parent. The lines ICCV 95138 and ICCV 97034 were developed from the parent ICCV 42. Similarly, ICCV 00108 and ICCV 00107 were developed from the parent IG 9216. It was also found that the lines ICCV 00103 and ICCV 00104 were developed from the parent JG 74 and both lines were present in cluster IV. Thus, it appeared that in some cases parentage of lines influenced the composition of cluster. The composition of clusters in  $D^2$  and SDS-PAGE method in general differed. However, there was similarity in some cases. The lines ICCV 93122, ICCV 97031,

ICCV 88208, and ICCV 00108 were present in cluster II consistently in both methods of grouping. Similarly, ICCV 97024 and ICCV 97038 were consistently present in cluster II or in cluster III in both  $D^2$  method and similarity index banding. This indicates consistency of grouping of these lines. Combining these two methods of clustering could more reliably help in assessing the diversity of lines or varieties.

**Acknowledgment.** The authors are grateful to Dr Jagdish Kumar, Senior Scientist (Chickpea Breeding), ICRISAT, Patancheru, India for providing chickpea advanced breeding lines and also to the information center of Bose Institute, Kolkata, India for statistical analysis.

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# Pigeonpea

## Breeding

### BRG 1: A High-yielding and Bold-seeded Pigeonpea Variety for Dhal and Vegetable Purpose

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Pigeonpea (*Cajanus cajan*) seeds can be used as dhal (dry split decorticated seed) or as a green vegetable. For green vegetable, pods are harvested when the seeds attain physiological maturity, i.e., when the seed accumulates most of its dry matter. Vegetable pigeonpea variety should have large pods and seeds for easy shelling. The

seeds should be sweet and tender to meet the requirement of the consumers (Faris and Singh 1990).

In recent years, farmers around towns and cities of southern Karnataka, India have started growing vegetable pigeonpea for local markets, where the demand is high. Hy 3C, a bold-seeded variety released in 1976 is being grown in these areas to meet the requirement of the consumers. However, this variety is susceptible to *Helicoverpa* pod borer infestation and phyllody disease.

BRG 1, a new high-yielding and bold-seeded vegetable type, has been developed recently at Gandhi Krishi Vignana Kendra (GKVK) Campus, University of Agricultural Sciences, Bangalore, Karnataka (Fig. 1). This variety was developed by pedigree method from the segregating material of a cross between Hy 3C and local vegetable type, collected from the farmers' fields at Chemachanahalli in Devanahalli taluka, Karnataka. BRG 1 was tested for its performance in state multilocation trials from 1997/98 to 2001/02 (AICPIP 1998, 1999, 2000, 2002). BRG 1 produced a mean dry seed yield of 1423 kg ha<sup>-1</sup>, 12.2% more than the control cultivar Hy 3C (1268 kg ha<sup>-1</sup>) (Table 1). During 2001/02, it produced a mean green pod yield of 4238 kg ha<sup>-1</sup>, 40.5% more than Hy 3C (Table 2).

Table 1. Seed yield of pigeonpea genotypes BRG 1 and Hy 3C at Bangalore, India.

Year <sup>1</sup>	Seed yield		
	BRG 1 (kg ha <sup>-1</sup> )	Hy 3C (kg ha <sup>-1</sup> )	Increase (%) over Hy 3C
1997/98	916	900	1.8
1998/99	1654	1419	16.6
1999/00	1076	928	15.9
2001/02	2048	1825	12.2
Mean	1423	1268	12.2

1. Data not available for 2000/01.

Table 2. Green pod yield of pigeonpea genotypes BRG 1 and Hy 3C at different pickings at Bangalore, India.

Harvest	Pod yield		
	BRG 1 (kg ha <sup>-1</sup> )	Hy 3C (kg ha <sup>-1</sup> )	Increase (%) over Hy 3C
1 <sup>st</sup> picking	1182	1381	-16.8
2 <sup>nd</sup> picking	1778	1238	43.6
3 <sup>rd</sup> picking	1278	397	221.9
Total	4238	3016	40.5



Figure 1. New vegetable pigeonpea variety BRG 1.



**Table 3. Distinguishable features of pigeonpea variety BRG 1 over Hy 3C.**

Characteristics	BRG 1	Hy 3C
<b>Morphological</b>		
Stem color	Green	Purple
Flower arrangement	Sparse	Clusters
Flower color	Pink	Red
Pod color	Green with few black streaks	Light green with black streaks
Seed color (dry)	Dull white and mottled	White and plain
Seed color (fresh)	Light green and mottled	Light green and plain
Plant height (cm)	150-170	140-160
Days to 50% flowering	90-95	85-90
Days to maturity	175-190	170-185
100-seed mass (dry) (g)	19.1	16.0
100-seed mass (fresh) (g)	41	38
100-pod mass (fresh) (g)	352	229
No. of pods plant <sup>-1</sup>	70-90	50-60
No. of seeds pod <sup>-1</sup>	5-6	3-5
<b>Cooking quality</b>		
Cooking time (min)	29	34
Water absorption (%)	39.1	41.6
Solids in the aqueous extract (%)	1.97	1.58
<b>Incidence of pests</b>		
<i>Helicoverpa</i> pod borer (%)	4.5	19.6
<i>Maruca</i> (%)	2.3	4.4

BRG 1 is a medium-duration variety with indeterminate growth habit and many distinguishable features over Hy 3C (Table 3). The flowers of this variety are pink and pods are green with few black streaks. Time to 50% flowering ranges from 90 to 95 days and maturity from 175 to 190 days. BRG 1 has bold, white, mottled seeds with 100-seed mass of 18.5 to 19.4 g (average 19.1 g).

Since the variety has been identified for vegetable purpose, it was evaluated for its cooking quality parameters. BRG 1 takes 29 min for cooking as compared to 34 min by Hy 3C. Further, solids in the aqueous extract were 1.97% in BRG 1 compared to 1.58% in Hy 3C. BRG 1 also had lower incidence of *Helicoverpa* pod borer (4.5%) and *Maruca* (2.3 %) than the control Hy 3C (19.6 and 4.4%, respectively).

We believe that the new variety BRG 1 will replace Hy 3C and will be widely grown in those areas where pigeonpea pods are harvested for vegetable purpose.

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## Identification of 'Phosphorus' Efficient Pigeonpea Genotypes Based on Phosphate Solubilizing Bacteria in the Rhizosphere

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Among the pulse crops, pigeonpea (*Cajanus cajan*) occupies an important place in Indian agriculture. Yield of the crop, however, has remained low. Phosphorus (P) appears to be one of the most important nutrients limiting pigeonpea production in Indian soils. A major portion of soil phosphorus (95%) is unavailable to plants being in the form of insoluble inorganic phosphate and organic phosphorus complexes, thereby limiting plant growth. Moreover, a large portion of inorganic phosphate applied

to soil as fertilizer is rapidly immobilized after application and becomes unavailable to plants. Thus, release of insoluble and fixed forms of P is an important aspect of increasing soil P availability. Some phosphate solubilizing microorganisms play an important role in phosphorus nutrition of plants by solubilizing insoluble phosphates and making these available to the plants. Soil and seed inoculation with phosphate solubilizing bacteria (PSB) improves solubilization of fixed soil phosphorus and applied phosphates resulting in higher crop yields. To overcome these problems through the activity of PSB, a study was conducted during kharif (rainy season) 2001 at the GB Pant University of Agriculture and Technology, Pantnagar, India to identify P efficient pigeonpea genotypes.

The genotypic variability was recorded among 20 short-duration pigeonpea genotypes, with respect to colonization of PSB in the rhizospheric zone. The lines that had more colonization of PSB were considered P efficient as PSB was reported to solubilize the unavailable or bound P by secreting organic acids and acid phosphatases in the rhizosphere (Gyaneshwar et al. 1998), thus making it available to the plants. The population of naturally occurring PSB in the rhizospheric samples

**Table 1. Population of phosphate solubilizing bacteria (PSB) in rhizospheric soil of pigeonpea at different crop growth stages.**

Genotype	PSB population ( $\times 10^4$ cfu $g^{-1}$ soil)			Seed yield (g plant <sup>1</sup> )
	Vegetative stage	Flowering stage	Maturity stage	
UPAS 120	1.23	1.56	2.03	36.40
H 82-1	1.03	1.53	1.83	32.17
Manak	0.86	2.00	1.33	17.26
AL 1430	1.33	1.86	1.43	40.50
T 21	1.46	1.53	1.10	44.53
Pusa 33	1.20	1.76	1.26	20.36
Pusa 208	1.26	1.30	2.76	28.15
Pusa 2001	0.93	1.03	1.63	22.30
BDN 1	1.13	1.56	1.50	27.74
BWR 10	1.50	1.56	1.53	34.72
ICPL 87	1.80	1.76	1.46	26.10
1CPL 84023	1.03	1.13	1.60	12.50
ICPL 85010	1.76	1.03	1.53	9.81
ICPL 88039	1.43	1.30	2.03	25.89
ICPL 98010	1.66	1.60	1.53	22.10
PA 106	1.30	2.10	1.80	14.20
PA 128	1.66	1.13	1.23	35.83
PA 134	1.60	1.33	2.33	25.46
PA 234	1.70	1.46	1.43	47.46
PA 243	1.33	1.43	1.36	33.20
Mean	1.33	1.48	1.63	27.83
SEm $\pm$	1.331	1.546	1.281	3.606

were determined by counting the number of colonies with clear transparent zone on Pikovskaya's agar medium and the colony forming units (cfu) g<sup>-1</sup> of rhizospheric soil were estimated (Sundara Rao and Sinha 1963). There was neither an external application of P nor PSB inoculation in the experiment. PSB population in the rhizosphere was determined at the vegetative, flowering, and maturity stages.

Analysis of variance for PSB population showed significant differences among the pigeonpea genotypes (Table 1). Plant species differ in the efficiency with which they acquire and utilize nutrients. The results of absolute PSB count and yield were consistently high in the determinate genotype ICPL 87 and indeterminate genotype BWR 10 and PA 243. High yield as well as increasing trend of PSB population from vegetative to maturity stage were observed in UPAS 120 (1.23 x 10<sup>4</sup> to 2.03 x 10<sup>4</sup> cfu g<sup>-1</sup> soil), H 82-1 (1.03 x 10<sup>4</sup> to 1.83 x 10<sup>4</sup> cfu g<sup>-1</sup> soil), and Pusa 208 (1.26 x 10<sup>4</sup> to 2.76 x 10<sup>4</sup> cfu g<sup>-1</sup> soil). High yield and near stable trend of PSB count in all the growth stages were noted in PA 234 and AL 1430. Pigeonpea cultivars were studied for native PSB isolate for the first time and no conclusion regarding high PSB population at all the growth stages and yield could be drawn in the investigation as its relative role changed with the genotype.

At vegetative stage, PSB count showed significant correlation with 100-seed mass (0.85, 0.55) and plant height (0.67, 0.45) at genotypic and phenotypic levels, respectively. Seed yield (0.51) had significant relationship at genotypic level only. At flowering stage, only pod length (-0.53) showed significant correlation at genotypic level. None of the yield traits showed significant correlation with PSB at maturity stage. The low magnitude of association between PSB and yield traits suggests that probably simple correlation does not account for such complex interrelationship.

In conclusion, results of this study revealed that the interactions between PSB and crop plants are complex and this may be affected by genotype, crop growth stage, and environment. The study is based on limited genotypes tested at a single location for one year; therefore, further investigation is required.

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## Pathology

### Evaluation of Pigeonpea Genotypes to Root-knot Nematode *Meloidogyne incognita*

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Pigeonpea (*Cajanus cajan*), also known as red gram or arhar, is a good source of protein and is one of the most important pulse crops in India. The root-knot nematode *Meloidogyne incognita* causes damage to pigeonpea crop and reduces the yield. Therefore, we evaluated 14 pigeonpea genotypes for resistance to *M. incognita*.

The genotypes which were used in this study were obtained from the Directorate of Pulses Research, Kalyanpur, Uttar Pradesh, India. About 4-5 seeds of each genotype were sown in 15 cm diameter clay pots containing 1 kg autoclaved soil-manure mixture in the ratio of 3:1. Prior to sowing, seeds were treated with *Rhizobium* using a 5% sucrose solution. When the seedlings were 20 days old, they were inoculated with 3000 freshly hatched second stage larvae or juveniles (J2) of *M. incognita*. For obtaining J2, eggmasses of nematodes were picked from the roots of tomato (*Lycopersicon lycopersicum*) plants grown in concrete culture beds. These eggmasses were then placed in 7.5 cm diameter sieves of 1 mm pore size and lined with double layer of tissue paper and placed in 10 cm diameter petri dishes containing water. The petri dishes were left at room temperature (25 ± 1 °C) for three days and thereafter water containing J2 that meanwhile hatched out was collected. The number of J2 per unit volume of the suspension was determined by counting them with the help of counting dish under a stereoscopic microscope. Appropriate amounts of J2 suspension were added to the test plants by making holes in the soil around the root system, so that each plant was inoculated with 3000 J2. There were five replications for each inoculated and uninoculated genotypes. Uninoculated plants served as control.

The plants were uprooted at 90 days after inoculation and were assessed for root gall, plant mass, plant length, and root nodulation. Roots and shoots were separated by cutting and the total length and mass were determined.

**Table 1. Reaction of pigeonpea genotypes to the root-knot nematode *Meloidogyne incognita*<sup>1</sup>.**

Genotype	Treatment <sup>2</sup>	Shoot length (cm)	Root length (cm)	Total plant length (cm)	Fresh plant mass (g)	Dry plant mass (g)	Root-gall index <sup>1</sup>	Root-nodule index <sup>4</sup>	Disease reaction <sup>5</sup>
AF-239	I	45.0	13.0	58.0	5.9	2.8	1.0	3.5	MR
	C	46.0	14.4	60.4	7.0	4.0	-	4.0	
C AUP 9004	I	32.0	10.0	42.0	3.2	1.6	5.0	1.0	HS
	C	34.0	10.5	44.5	4.5	2.0	-	2.6	
KE 22	I	36.0	9.5	45.5	4.5	2.4	4.0	1.5	S
	C	42.2	10.0	52.2	5.4	3.0	-	2.0	
KM 33	I	39.5	8.5	48.0	4.6	2.3	3.0	2.0	S
	c	43.0	9.0	52.0	5.3	2.6	-	3.0	
P 609	I	42.0	12.0	54.0	5.4	2.5	2.0	2.5	MS
	c	42.5	13.5	56.0	6.6	3.4	-	3.0	
H 9013	I	38.0	10.0	48.0	4.8	2.4	3.5	2.0	S
	c	40.0	10.5	50.5	5.5	2.8	-	3.0	
H 9014	I	49.0	11.0	60.0	6.4	3.0	1.0	4.0	MR
	c	50.0	12.5	62.5	7.8	4.0	-	4.5	
H 9125	I	40.2	10.0	50.4	4.7	2.2	3.0	2.0	S
	c	41.0	11.5	52.5	5.6	2.5	-	2.5	
Pusa Pigeonpea	I	50.0	14.5	64.5	7.4	3.5	0.0	4.5	HR
	c	52.0	16.5	68.5	8.4	4.5	-	5.0	
Pusa 17	I	38.5	10.0	58.5	5.0	2.6	4.0	1.5	S
	c	40.0	12.8	52.8	5.6	2.8	-	3.0	
Pusa 25	I	40.4	10.0	52.4	5.2	2.5	3.0	2.5	S
	c	42.0	11.0	53.0	6.0	3.2	-	3.0	
Pusa 26	I	44.0	12.0	56.0	6.0	3.0	1.0	4.0	MR
	c	45.0	12.4	57.4	7.5	4.4	-	4.5	
Pusa 28	I	32.0	8.0	40.0	3.0	1.5	5.0	1.0	HS
	c	33.5	10.0	43.5	4.4	2.4	-	2.0	
KM 34	I	42.0	11.0	53.0	6.2	2.5	2.0	3.0	MS
	c	43.0	12.5	55.5	6.5	3.4	-	4.0	
CD ( <i>P</i> = 0.05)				3.35	0.96	0.72		0.84	

1. Data are means of five replications.

2. I = Inoculated; C = Control.

3. Scored on 0-5 rating scale, where 0 = no galls, and 5 = >100 galls per root system.

4. Scored on 0-5 rating scale, where 0 = no nodulation, and 5 = very high nodulation.

5. HR = Highly resistant; MR = Moderately resistant; MS = Moderately susceptible; S = Susceptible; HS = Highly susceptible.

For determining dry mass, the roots and shoots were dried in an oven at 60°C and the total dry mass was determined. Root-gall index was assessed on 0-5 rating scale (Sasser et al. 1984), where 0 = no galls, and 5 = >100 galls per root system. Root-nodule index was also assessed on 0-5 rating scale where 0 = no nodulation, and 5 = very high nodulation. Data was analyzed for critical difference (Panse and Sukhatme 1978).

Among 14 genotypes tested, Pusa Pigeonpea was resistant and AF-239, H 9014, and Pusa 26 were moderately resistant to *M. incognita* infection (Table 1).

Other genotypes showed varying levels of resistance. The resistant genotypes showed low number of root galls and more root nodules whereas the susceptible genotypes showed higher number of root galls and less number of root nodules. However, as compared with uninoculated control all the resistant and susceptible genotypes have less fresh and dry mass and root nodulation (Table 1). Our results are in accordance with Sasser and Hartman (1985) and Anver and Alam (1994), who have also reported resistance in some other cultivars of pigeonpea on the basis of root-gall index.

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## Entomology

### Reaction of Eight Short-duration Pigeonpea Genotypes Against Pod Borer Complex in Tamil Nadu, India

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Pigeonpea (*Cajanus cajan*) is an important grain legume crop of the semi-arid tropics and is consumed as green peas as well as dry seeds (Tabo et al. 1995). India is the largest producer of pigeonpea in the world (Nene and Sheila 1990). The pod borers *Helicoverpa armigera* and *Maruca vitrata* are the major insect pests that constrain pigeonpea production (Lateef and Reed 1990). Annual losses due to *H. armigera* and *M. vitrata* have been estimated at US\$ 317 million and US\$ 30 million worldwide respectively (ICRISAT 1992). To increase pigeonpea production, the major focus has been on short-duration pigeonpea cultivars. To identify suitable short-duration cultivars for Tamil Nadu, India the promising genotypes developed by ICRISAT, Patancheru, India were evaluated against the pod borer complex at the

National Pulses Research Centre, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu during 1995/96 and 1996/97 cropping seasons.

An experiment was conducted in a randomized complete block design with three replications involving eight short-duration pigeonpea entries [four determinate types: ICPL 151, ICPL 4, ICPL 86012, and ICPL 87 (check); and four indeterminate types: ICPH 8, ICPL 88034, JCPL 2, and UPAS 120 (check)]. Each entry was planted in 4.8 m<sup>2</sup> plot in four rows, with 30 cm interrow spacing and 10 cm plant spacing. Normal agronomic practices were followed for raising the crop. The entries were grown under unprotected condition. Damage due to pod borer complex was assessed on pods collected from five plants at random. Podfly (*Melanagromyza obtusa*) seed damage was assessed on seed obtained from 5 plants selected at random from the middle two rows leaving the border rows. At harvest the seed yield per plot was also recorded. The percentage pod damage was converted to Arcsine transformed values and data for pod borer and podfly damage, and yield were statistically analyzed.

During 1995/96 season, pod borer damage ranged from 42.7% in ICPL 4 to 91.8% in ICPL 87, whereas in 1996/97, the damage ranged from 40.5% in ICPL 4 to 74.0% in ICPL 87. The mean damage over two years ranged from 41.6% to 82.9% (Table 1). ICPL 4 was least susceptible with 49.8% less damage than the susceptible check ICPL 87. ICPL 151, ICPL 86012, and ICPL 88034 suffered 34.4, 28.5, and 15.6% less damage, respectively over the check entry ICPL 87. The performance of ICPL 2 was on par with UPAS 120. The mean podfly damage ranged from 6.3% in ICPL 4 to 13.7% in UPAS 120 (Table 1). The decrease in seed damage was highest (53.3%) in ICPL 4, followed by ICPL 151 (38.5%). Podfly damage was similar in UPAS 120 (13.7%) and ICPL 87 (13.5%). Maximum seed yield of 328.5 kg ha<sup>-1</sup> was recorded in ICPL 4, followed by ICPL 88034 (308.5 kg ha<sup>-1</sup>) and ICPL 86012 (251.4 kg ha<sup>-1</sup>) (Table 1). The lowest seed yield of 161.2 kg ha<sup>-1</sup> was recorded in ICPL 87. More than 100% yield increase was recorded in ICPL 4 over ICPL 87, followed by ICPL 88034 (69.8%) and ICPL 86012 (56.0%).

The genotypes ICPLs 4, 151, 88034, and 86012 showed more than 15% reduction in pod damage as compared to ICPL 87 and UPAS 120. Similarly, more than 35% podfly seed damage reduction was recorded in ICPL 4, ICPL 151, and ICPH 8 as compared to ICPL 87 and UPAS 120. In Madhya Pradesh, India the short-duration genotypes ICPL 151 and ICPL 86012 have been reported to suffer low pod borer damage than the check entry ICPL 87 (Anonymous 1997-98). In our study we

**Table 1. Pod borer and podfly damage and seed yield in short-duration pigeonpea genotypes in Tamil Nadu, India.**

Entry	Lepidopteran pod borer damage <sup>1</sup> (%)			Podfly seed damage (%)			Seed yield (kg ha <sup>-1</sup> )		
	1995/96	1996/97	Mean	1995/96	1996/97	Mean	1995/96	1996/97	Mean
ICPL 151	52.8 (46.6)	55.9 (48.4)	54.4	9.2	7.3	8.3	206.8	201.3	204.1
ICPL 4	42.7 (40.8)	40.5 (39.5)	41.6	7.2	5.4	6.3	393.7	263.3	328.5
ICPL 86012	65.6 (54.1)	52.9 (46.7)	59.3	13.4	12.5	13.0	331.4	171.3	251.4
ICPH 8	72.9 (58.7)	53.9 (47.3)	63.4	10.4	7.0	8.7	168.2	136.0	152.1
ICPL 88034	51.5 (45.8)	61.7 (51.8)	56.6	8.4	12.8	10.6	345.9	271.0	308.5
ICPL 2	67.0 (55.0)	68.0 (55.6)	67.5	9.9	15.6	12.8	197.1	164.0	180.6
UPAS 120 (check)	71.6 (57.8)	62.2 (52.1)	67.1	11.8	15.5	13.7	196.4	167.0	181.7
ICPL 87 (check)	91.8 (73.5)	74.0 (59.4)	82.9	11.7	15.2	13.5	204.6	117.7	161.2
CD ( $P = 0.05$ )	(2.92)	(2.87)		2.60	4.82		14.61	12.84	
SE±	1.36	1.34		1.21	2.25		6.81	5.99	
Mean	64.5	58.6		10.3	11.4		255.5	186.5	

1. Data represent pod damage. Figures in parentheses are Arcsine transformed values

observed similar results. In multilocal trials conducted at ICRISAT (Andhra Pradesh), Akola (Maharashtra), SK Nagar (Gujarat), and Vamban (Tamil Nadu), ICPL 4 suffered low pod borer damage, followed by ICPL 151 and ICPL 86012. The seed yield was also higher in these entries than ICPL 87. Short-duration types that performed well under Indian conditions have also been reported in Africa (Singh et al. 1994).

We conclude that ICPLs 151,4, 86012, and 88034 are tolerant to both pod borer and podfly, and may have factors contributing to resistance.

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## New Methodology for Assessment of Damage by Pigeonpea Pod Wasp *Tanaostigmodes cajaninae*

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Pigeonpea (*Cajanus cajan*) is one of the important pulse crops of India, and is widely grown throughout the country. Under field condition, the yields are low due to damage by insect pests and diseases. More than 200 insect species damage pigeonpea, of which few species are known to occur in India (Lateef and Reed 1990). Among these pests the pod borers are the main cause for reduction in seed yield. Of late, pigeonpea pods are damaged by the pod wasp *Tanaostigmodes cajaninae* LaSalle (Hymenoptera: Tanaostigmatidae). Although it was considered earlier as a minor pest, in recent years, the damage is on the increase. This pest was first reported from India by Lateef (1977) and Lateef et al. (1985).

It inflicts more damage on the research stations where there are more chances for the host availability for a longer period rather than in farmers' fields (Reed et al. 1989). The pods infested by this pest often fail to develop and even if they develop, the pod size remains small (Ranga Rao and Shanower 1999). If the pest incidence occurs during later stages of pod development, one or two seeds may develop at the distal portion of the pods.

Field surveys conducted in Tamil Nadu, India during 1993 revealed that there was about 25% to 75% pod damage by this pest in farmers' fields (Durairaj and Ganapathy 1996). A survey in some of the pigeonpea-growing areas in Tamil Nadu revealed that there was about 25% pod damage in farmers' fields under rainfed conditions. At the Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu the pod damage varied from 25% to 50% in different varieties of pigeonpea (Durairaj et al. 2001). In general, the pod damage due to different pod borers is assessed by collecting 300 to 500 pods at random, and counting the healthy and affected pods at harvest. The same methodology is also being adopted for pod wasp damage assessment (Lateef et al. 1985). This method of observation may be useful for the lepidopteran borers as the damage is caused in the fully developed pods and the symptoms are prominent. But the pod wasp damage starts immediately after fertilization of the pods. The infested pods fail to develop (atrophied), and even if they develop, these are very small and escape the attention of the field staff. Usually, the pod samples are collected at the time of harvest for assessing the pod borer damage, but by this time the pods damaged by the pod wasp are dried and shed, and may not be represented in the pod samples collected. Hence, to overcome this problem and to assess the real damage caused by the pod wasp, the following damage assessment methodology was developed at the Tamil Nadu Agricultural University, Coimbatore.

The pod wasp damage should be assessed when 50 to 70% of the pods have matured. At this stage even the pods which were infested by the pod wasp in the early stages of pod development are on the plant. From each plot, 5 to 10 plants should be selected at random depending upon the plant population and nature of trials. In each plant, 1 to 2 branches may be selected at random. The total number of pods including the underdeveloped pods due to pod wasp damage present at 0.5 m from the tip of the branch should be counted. If there are more number of accessions to be screened, one or two branches at 0.5 m length may be collected as described earlier and tied with labels, and the damage can be assessed in the laboratory by counting the total number of pods and the infested pods. By following this method all the pod wasp infested pods will be represented in the pod sample. This will give a better estimate of the damage caused by pod wasp, which is not taken care of in the normal method of pod borer complex damage assessment. Thus, pod damage values were higher in the new method of assessment than in the existing method (Table 1).

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**Table 1. Assessment of pigeonpea pod damage by the pod wasp by two methods at the Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India.**

Genotype	Period of observation	Pod wasp damage (%)	
		Existing method	New method
Vamban 1	February 2001	9.0	15.0
ICPL 86012	February 2001	22.0	37.2
ICPL 87	February 2001	17.0	21.4
APK 1	February 2002	15.0	23.5
CO 5	February 2002	23.0	31.3

# Effect of Insecticides on Loss in Seed Mass and Yield of Pigeonpea by Pod Borer

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Pigeonpea (*Cajanus cajan*) is attacked by more than 200 species of insect pests. Among the various species of insect pests of pigeonpea, the pod borers cause serious damage in North India. At Varanasi, Uttar Pradesh, podfly (*Melanagromyza obtusa*) and pod borer (*Helicoverpa armigera*) are the major pests of pigeonpea and result in

55.94%, 32.47%, and 19.19% loss in pods, seeds, and seed mass, respectively (Kumar and Nath 2002). Insecticide application is one of the most effective methods for controlling these pests. Therefore, we evaluated several insecticides for their relative efficacy to minimize the losses due to pod borers in pigeonpea.

The experiments were conducted at the Agriculture Research Farm, Banaras Hindu University (BHU), Varanasi during kharif (rainy season) 1994 and 1995 to evaluate the relative efficacy of synthetic insecticides in minimizing the losses by pod borer (*H. armigera*) and podfly (*M. obtusa*) on pigeonpea cultivar UPAS 120 and Bahar. The insecticides such as monocrotophos 36 SL (0.04%), fenvalerate 20 EC (0.02%), cypermethrin 25 EC (0.006%), carbaryl 5 D (5.00%), deltamethrin 2.8 EC (0.004%), endosulfan 35 EC (0.07%), and malathion 5 D (5.00%) were applied in two different schedules. In the first schedule, all the insecticides were applied only once at flowering and pod formation stage, while in the second schedule,

**Table 1. Effect of insecticides on loss in pigeonpea seed mass and yield due to pod borer and podfly damage<sup>1</sup>.**

Insecticide	Seed mass loss (%) (Bahar)			Seed yield (kg ha <sup>-1</sup> )					
				Bahar			UPAs120		
	Schedule 1	Schedule 2	Average	Schedule 1	Schedule 2	Average	Schedule 1	Schedule 2	Average
Monocrotophos (0.04%)	3.26 (10.67)	2.52 (9.05)	2.89 (9.86)	2323	2647	2485	1762	1893	1828
Fenvalerate (0.02%)	4.63 (12.37)	3.39 (10.51)	4.01 (11.44)	1938	2240	2090	1603	1655	1630
Cypermethrin (0.006%)	4.25 (11.87)	2.87 (10.52)	3.56 (11.19)	1979	2295	2137	1582	1715	1649
Carbaryl (5.00%)	5.32 (13.26)	4.56 (12.17)	4.94 (12.71)	1799	2096	1948	1380	1529	1455
Deltamethrin (0.004%)	4.93 (12.78)	4.20 (11.75)	4.57 (12.27)	1847	2154	2001	1440	1583	1512
Endosulfan (0.07%)	3.91 (11.35)	2.87 (9.63)	3.39 (10.49)	2120	2436	2292	1675	1830	1753
Malathion (5.00%)	6.00 (14.15)	4.99 (12.84)	5.50 (13.50)	1747	2024	1886	1356	1513	1434
Control	17.55 (24.73)	17.84 (24.97)	17.70 (24.85)	1323	1308	1316	963	996	979
Average	6.23 (13.90)	5.41 (12.68)	5.82 (13.29)	1884	2153	2019	1470	1589	1530
LSD for comparing insecticides ( <i>P</i> = 0.05)			(0.57)			28			32
LSD for comparing schedules ( <i>P</i> = 0.05)			(0.29)			14			16
LSD for comparing insecticides x schedules ( <i>P</i> = 0.05)			(0.82)			39			45

1. Figures in parentheses are angular transformed values.



the insecticides were applied twice (first application was given at pod formation stage and second application was given 25 days later). The pods from five randomly selected plants from three middle rows of each plot were collected to record the damage caused to seed by podfly and pod borer. Pest damage was assessed by recording the mass of healthy and damaged seeds from 50 pod samples from each plot. The yield data was subjected to analysis of variance.

The pigeonpea seed mass loss was assessed by using the following formula:

$$\text{Percentage seed mass loss} = \frac{\text{Mass of damaged seeds}}{\text{Mass of healthy seeds}} \times 100$$

The insecticidal treatments reduced the seed mass loss due to both pest species compared to untreated control. Among the various treatments, the minimum seed mass loss was recorded in plots treated with monocrotophos (2.89%), followed by endosulfan (3.39%), cypermethrin (3.56%), fenvalerate (4.01%), deltamethrin (4.57%), carbaryl (4.94%), and malathion (5.50%). Highest loss in seed mass was recorded in untreated control plots (17.70%) (Table 1).

The maximum seed yield in UPAS 120 was obtained from monocrotophos treated plots (1828 kg ha<sup>-1</sup>), followed by endosulfan (1753 kg ha<sup>-1</sup>), cypermethrin (1649 kg ha<sup>-1</sup>), fenvalerate (1630 kg ha<sup>-1</sup>), deltamethrin (1512 kg ha<sup>-1</sup>), carbaryl (1455 kg ha<sup>-1</sup>), malathion (1434 kg ha<sup>-1</sup>), and untreated control (979 kg ha<sup>-1</sup>). Jakhmola and Bhadauria (1998) had earlier reported that monocrotophos application resulted in highest yields (1575 kg ha<sup>-1</sup>) in UPAS 120.

Bahar yielded 2485 kg ha<sup>-1</sup> in monocrotophos treated plots, followed by endosulfan (2292 kg ha<sup>-1</sup>), cypermethrin (2137 kg ha<sup>-1</sup>), fenvalerate (2090 kg ha<sup>-1</sup>), deltamethrin (2001 kg ha<sup>-1</sup>), carbaryl (1948 kg ha<sup>-1</sup>), malathion (1886 kg ha<sup>-1</sup>), and untreated control (1316 kg ha<sup>-1</sup>). Siddappaji et al. (1985) and Sinha and Srivastava (1989) reported that application of monocrotophos, cypermethrin, fenvalerate, and deltamethrin resulted in high seed yields in pigeonpea. The effectiveness of sprays containing 0.07% endosulfan and the dust formulations of carbaryl and malathion have also been found to increase seed yield significantly (Chaudhury and Rastogi 1980). In our experiments, the plots under second schedule recorded more seed yield compared to the plots treated under first schedule of insecticide application.

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## Natural Enemies of Pigeonpea Insect Pests at Varanasi, Uttar Pradesh, India

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Pigeonpea (*Cajanus cajan*) is damaged by a number of insect pests. Several natural enemies help in restricting the population of potential pests to non-damaging levels. A large number of parasites and predatory insects, several species of spiders, lizards, and birds have been recorded in pigeonpea (Reed et al. 1989). Pigeonpea is attacked by 23 species of insects belonging to 6 orders and 20 families. Among these the pod borers cause losses of 55.94% pod damage, 32.47% seed damage, and 19.19% seed mass loss at Varanasi, Uttar Pradesh, India (Kumar and Nath 2002).

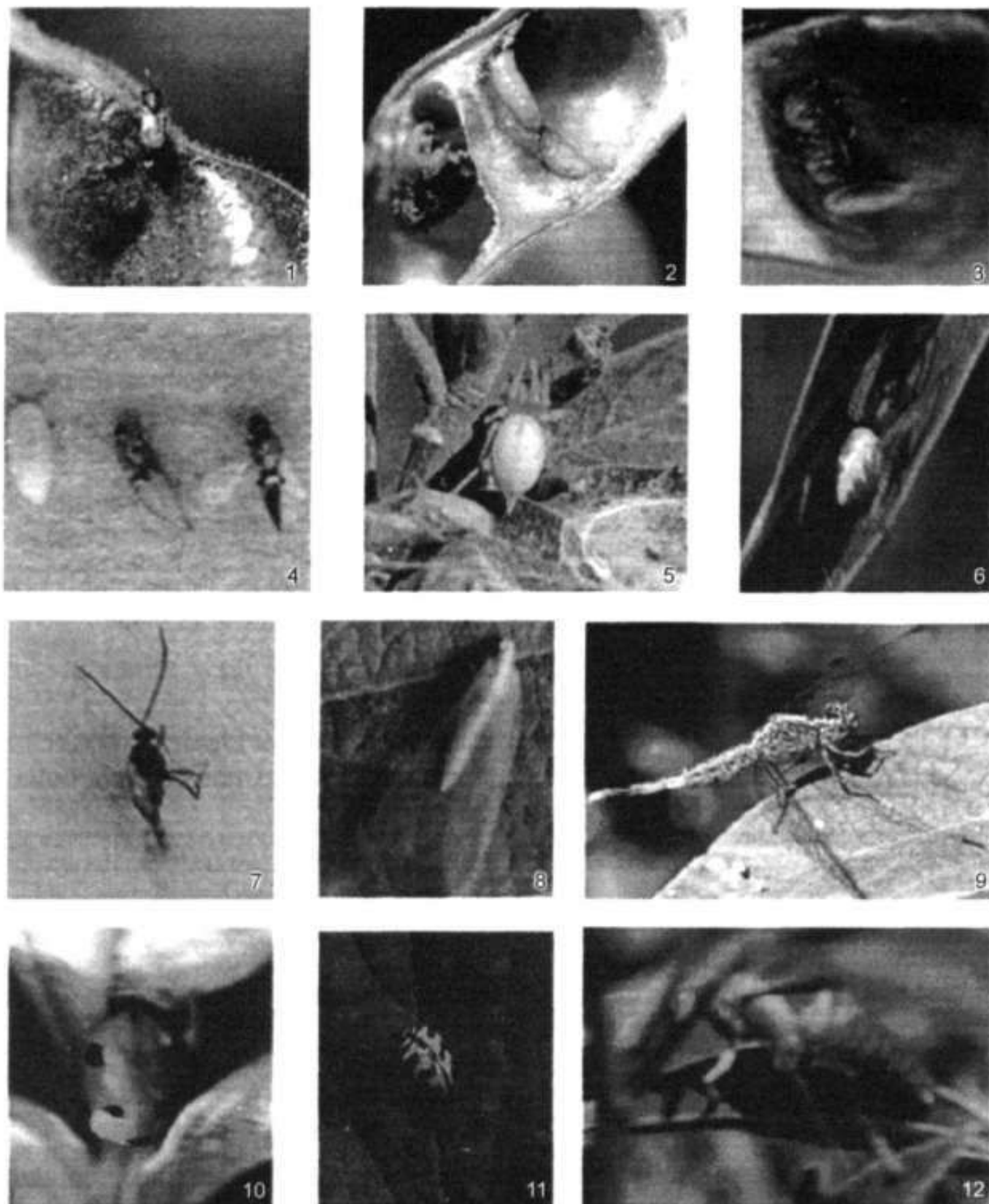


Figure 1. Natural enemies of pigeonpea insect pests: (1) Male and female adults of *Euderus lividus*; (2) Open pod showing parasitized larvae of poddy by *E. lividus*; (3) Larvae of *E. lividus*; (4) Larva, pupa, and adult of *E. lividus*; (5) *Cluhiona* sp; (6) *Araneus* sp; (7) Adult of braconid (*Apanteles* sp); (8) Green lacewing (*Chrysoperla cornea*); (9) Dragonfly (*Crocothemis servilla*); (10) Ladybird beetle (*Coccinella septempunctata*); (11) Ladybird beetle (*Cheilomenes sexmaculatus*); and (12) Common wasp (*Vespa orientalis*).

**Table 1. Natural enemies of insect pests in early (E) and medium-late (ML) cultivars of pigeonpea.**

Common name	Scientific name	Family	Order	Host	Cultivars
Eulophid parasitoid	<i>Euderus lividus</i> Ashm.	Eulophidae	Hymenoptera	Podfly	E. ML
Braconid wasp	<i>Apanteles</i> sp	Braconidae	Hymenoptera	Pod borer and hairy caterpillar	E. ML
Ladybird beetle	<i>Coccinella septempunctata</i> Linn.	Coccinellidae	Coleoptera	Aphid and jassid	E. ML
Ladybird beetle	<i>Cheilomenes sexmaculatus</i> Fab.	Coccinellidae	Coleoptera	Aphid and jassid	ML
Mirid bug	<i>Cyrtorhinus lividipennis</i> Reut.	Miridae	Hemiptera	Thrips	E. ML
Praying mantis	<i>Mantis religiosa</i> Linn.	Mantidac	Dictyoptera	Aphid and grasshopper	E. ML
Dragonfly	<i>Crocothemis servilia</i> Drury	Gomphidae	Odonata	Pod borer larvae	E. ML
Green lacewing	<i>Chrysoperla carnea</i> Stephens.	Chrysopidae	Neuroptera	Aphid, thrips, and jassid	E. ML
Common wasp	<i>Vespa orientalis</i> Linn.	Vespidae	Hymenoptera	Pod borer	ML
Sac spider	<i>Clubiona</i> sp.	Clubionidae	Araneae	Leaf webber and legume pod borer	E. ML
Spider	<i>Araneus</i> sp	Araneidae	Araneae	Leaf webber and legume pod borer	E. ML
Indian mynan	<i>Acridotheris tristis</i> L.	Sturnidae	Passeriformes	Grasshoppers	ML
King crow	<i>Dicrurus macrocercus</i> Vieillot	Dicruridae	Passeriformes	Pigeonpea pod borer	ML

The natural enemies on pigeonpea cultivars were recorded during seedling stage to podding of the crop at the Agriculture Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi during 1994/95 and 1995/96. The natural enemies of insect pests associated with pigeonpea cultivar UPAS 120 belonged to seven insect families and one family of spiders, while the medium-late cultivar Bahar harbored eight families of insects, two families of spiders, and two avian predators. The natural enemies of pigeonpea pests observed in UPAS 120 included braconid wasp (*Apanteles* sp, *Euderus lividus*), ladybird beetle (*Coccinella septempunctata*), mirid bug (*Cyrtorhinus lividipennis*), praying mantis (*Mantis religiosa*), dragonfly (*Crocothemis servilia*), green lacewings (*Chrysoperla carnea*), and spiders (*Araneus* sp, *Clubiona* sp). A total of 13 species of natural enemies were recorded in the medium-late cultivar Bahar. The natural enemies observed in UPAS 120 were also present in Bahar. The common wasp (*Vespa orientalis*), ladybird beetle (*Cheilomenes sexmaculatus*), sac spider (*Clubiona* sp), Indian mynah (*Acridotheris tristis*), and king crow (*Dicrurus macrocercus*) were observed in Bahar (Fig. 1 and Table 1).

The eulophid parasitoid and the spiders were more prevalent than the other natural enemies. Singh and Mavi (1984) reported a spider (*Clubiona abbottii*) as a predator of lycaenids in pigeonpea. Sahoo and Senapati (2000) reported the activities of predators such as spiders,

praying mantis, and wasp between mid-August and mid-December. The eulophid is a potential parasitoid of the podfly *Melanagromyza obtusa*. Singh (1991) reported *E. lividus*, which was reared from immature stage of *M. obtusa* infesting early and late varieties of pigeonpea in Uttar Pradesh. The diversity and prevalence of natural enemies was observed to be more in medium-late varieties than in early-maturing varieties.

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## Egg Parasitoid *Gryon* sp on Pigeonpea Pod Bug *Clavigralla gibbosa* in Tamil Nadu, India

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Pigeonpea (*Cajanus cajan*). is an important pulse crop of India. More than 200 insect pests have been recorded on pigeonpea, of which few species are known to occur in India (Lateef and Reed 1990). Among the pod infesting insects the pod bug *Clavigralla gibbosa* (Heteroptera: Coreidae) is one of the regular and potential pest of pigeonpea (Singh et al. 1989). Though this insect also feeds on lablab (*Lablab purpureus*) and cowpea (*Vigna unguiculata*), it has a high preference for pigeonpea (Rawat et al. 1983). Feeding by nymphs and adults results

in premature shedding of flower buds, flowers, and pods, deformation of pods, and shriveling of grains leading to substantial yield loss (Mishra and Odak 1981). This pest is widely distributed in India and Sri Lanka. In recent years, it has assumed the status of a major pest in Tamil Nadu, India on many pulse crops including pigeonpea, mung bean (*Vigna radiata*), black gram (*Vigna mungo*), cowpea, lablab, and *mochai* (*Lablab purpureus* var. *lignosus*). During field observations, it was found that the eggs of this bug were parasitized by an egg parasitoid *Gryon* sp, which may play an important role in regulating its populations under natural conditions. Hence, a study was conducted at the Tamil Nadu Agricultural University, Coimbatore, India to know the level of egg parasitism by this parasitoid and the seasonal incidence on *C. gibbosa* eggs during 2000-02.

Eggs of *C. gibbosa* bugs are laid on the leaves and pods in groups of 2 to 62 (Shanower et al. 1996). The eggs are round to oval shaped, 2-3 mm long, and dark brown in color. The eggs were collected at monthly intervals and kept in petri dishes on a moist filter paper. These eggs were observed everyday for the hatching of nymphs and emergence of egg parasitoids. The number of unhatched eggs were also recorded. As the egg period of this bug varies from 3 to 7 days, the eggs which did not hatch after this period were treated as unhatched. The percentage of egg parasitism, nymphal emergence, and unhatched eggs were estimated every month. These observations were made from June to March on short-duration pigeonpea varieties when maximum *C. gibbosa* activity was noticed.

**Table 1. Extent of parasitism by *Gryon* sp on pod bug eggs in Tamil Nadu, India during 2000-02.**

Month	Nymphal emergence (%)			Parasitism (%)			Unhatched eggs (%)		
	2000	2001	2002	2000	2001	2002	2000	2001	2002
June	40.8	31.3	- <sup>1</sup>	36.8	41.2	-	22.4	27.3	-
July	24.5	25.6	-	48.6	49.7	-	26.8	24.5	-
August	4.7	25.1	-	72.6	47.9	-	22.6	26.9	-
September	5.3	9.7	-	70.1	56.1	-	24.4	34.1	-
October	4.2	7.1	-	74.3	71.7	-	21.4	21.2	-
November	9.3	1.7	-	64.5	91.6	-	26.1	6.7	-
December	12.1	22.6	-	59.5	60.5	-	28.3	16.7	-
January	-	54.0	59.3	-	37.2	24.4	-	8.2	16.3
February	-	58.3	59.4	-	29.1	15.1	-	12.5	25.4
March	-	64.3	68.2	-	27.1	23.8	-	8.2	7.9
Mean	14.4	30.0	62.3	60.9	51.2	21.1	24.6	18.6	16.5
SE±	4.75	6.63	2.41	4.83	5.96	2.46	0.90	2.87	4.13

1. - = No pigeonpea crop in the field.

The result of this study revealed that the activity of *C. gibbosa* started from June and reached the peak during July to December. The bug population gradually decreased from December onwards and reached a low level during March. The abundance of eggs in the field is directly related to the population in the field. The egg parasitoid identified on *C. gibbosa* was *Gryon* sp (Hymenoptera: Scelionidae). A maximum of 74.3% of the eggs were parasitized during October 2000, and 91.6% in November 2001 (Table 1). The lowest parasitization of 15.1% was recorded in February 2002. The nymphal emergence from the eggs was inversely related to the level of parasitism. Natural mortality of eggs was also observed under laboratory conditions during the course of this investigation. Earlier reports indicated that only a few natural enemies parasitize the eggs of this bug (Shanower et al. 1999). The activity of the egg parasitoid *Gryon clavigrallae* has been reported from Andhra Pradesh, India (Madhuri 1997). Shanower et al. (1996) observed that *G. clavigrallae* parasitized up to 69% of eggs of *Clavigralla* in India. In our studies, 15.1 to 91.6% eggs of *C. gibbosa* were parasitized by *Gryon* sp with a peak activity during August to December. Hence, *Gryon* sp may be considered as a potential biocontrol agent of *C. gibbosa*.

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## Bioassay of *Metarhizium anisopliae* Against Pigeonpea Pod Borer *Helicoverpa armigera*

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*Helicoverpa armigera* is a polyphagous pest on several crops. It is a major constraint in pigeonpea (*Cajanus cajan*) production. Yelshetty and Sidde Gowda (1998) reported 90 to 100% yield loss due to this pest in Karnataka, India. Considering the environmental implications of chemical control, it is important to exploit novel tactics to combat this pest. Microbial control is an attractive method of pest management as it helps to minimize the use of synthetic pesticides. Species of *Helicoverpa* are susceptible to almost all groups of entomopathogens (Deva Prasad et al. 1990). A deuteromycetes fungus, *Metarhizium anisopliae* (Metch.) Sorokin infects a wide range of insect species belonging to Lepidoptera, Hemiptera, Coleoptera, and Orthoptera. Deva Prasad et al. (1990) reported its efficacy against *H. armigera*. Considering its significance in pest management, in vitro studies were carried out during 2001-02 at the College of Agriculture, Nagpur, Maharashtra, India to quantify the conidial concentration to achieve 50% mortality in laboratory reared second instar larvae of *H. armigera*.

*Metarhizium anisopliae* var. *anisopliae* used in the present studies was obtained from the Sugarcane Breeding Institute, Coimbatore, Tamil Nadu, India. The fungus was subcultured on potato dextrose agar (PDA) medium and used for further studies. The conidial suspension for the bioassays was obtained from 15-day-old culture of the fungus, cultured on Sabouraud dextrose agar + yeast (SDA + Y) medium. The fungal mat (20 g) with homogenous fungal growth from the culture medium was suspended thoroughly in 70 ml sterilized distilled water containing 0.1 % Tween-80, by using a rotary mixer for 20 minutes. It was poured through muslin cloth, and then filtered through Whatman No. 1 filter paper. The filtrate was made up to 100 ml by adding sufficient quantity of sterilized distilled water. An improved Neubauer's hemocytometer was used to assess the conidial concentration of the fungal suspension, and through series of dilutions, desired concentrations ranging from  $2.28 \times 10^6$  to  $2.28 \times 10^{10}$  conidia ml<sup>-1</sup> were standardized. Larvae of *H. armigera* collected from

pigeonpea plants in the field were reared individually in transparent plastic vials (3.5 cm x 4 cm). Ten newly moulted second instar larvae were surface sterilized with 1% sodium hypochlorite and rinsed twice with sterilized distilled water. Excess water was removed by blotting paper. The larvae were then placed in a petri dish lined with filter paper, and topically treated with 2 ml conidial suspension ranging from  $2.28 \times 10^6$  to  $2.28 \times 10^{10}$  conidia  $\text{ml}^{-1}$  using a hand atomizer. Control larvae were sprayed with 0.1% Tween-80 in sterilized distilled water. After air drying, the treated larvae were carefully transferred to plastic vials individually and reared at  $25 \pm 2^\circ\text{C}$  temperature and 90% relative humidity. The vials were placed in plastic trays containing moist absorbent cotton and covered with a glass plate. There were six treatments including control replicated four times in a randomized complete block design.

The larval mortality was recorded at 24, 48, 72, 96, 120, 144, 168, and 192 h after treatment. From these, the percentage of larval mortality and period (h) required to kill host larvae were calculated. Cumulative mean larval mortality at 192 h (8<sup>th</sup> day) after treatment was considered for evaluation. Corrected mortality data were subjected to probit analysis and the critical conidial concentration for effecting 50% larval mortality ( $\text{LC}_{50}$ ) and time required to effect 50% larval mortality ( $\text{LT}_{50}$ ) were worked out accordingly.

The bioassay studies against second instar larvae of *H. armigera* revealed that  $\text{LC}_{50}$  was  $1.47 \times 10^5$  conidia  $\text{ml}^{-1}$  of fungal suspension ( $\chi^2 = 0.32$ ,  $y = 3.24 + 0.34X$ , "fiducial limit" =  $4.78 \times 10^3$  to  $4.57 \times 10^6$ ). The fungus at  $2.28 \times 10^{10}$  conidia  $\text{ml}^{-1}$  caused highest larval mortality of 97.5%, followed by 92.5%, 85.0%, 80.0%, and 67.5% in the fungal suspensions containing  $2.28 \times 10^9$ ,  $2.28 \times 10^8$ ,  $2.28 \times 10^7$ , and  $2.28 \times 10^6$  conidia  $\text{ml}^{-1}$  respectively, at 192 h after treatment (Table 1). Kenchareddi and Jayaramaiah (1997) reported  $\text{LC}_{50}$  values of  $6.07 \times 10^4$  and  $6.15 \times 10^5$  conidia  $\text{ml}^{-1}$  against first and third instar larvae of *H. armigera*, respectively while Gopalkrishnan and Narayanan (1989) reported 100% larval mortality in early instars at  $1.8 \times 10^9$  conidia  $\text{ml}^{-1}$ . These results seem to be consistent with the present findings. The  $\text{LT}_{50}$  value for second instar was inversely proportional to the conidial concentration of the inoculum. Similar findings have earlier been reported by Walstad et al. (1970). The  $\text{LT}_{50}$  value was 79.43 h for  $2.28 \times 10^{10}$ , 85.11 h for  $2.28 \times 10^9$ , 97.72 h for  $2.28 \times 10^8$ , 104.71 h for  $2.28 \times 10^7$ , and 123.02 h for  $2.28 \times 10^6$  conidia  $\text{ml}^{-1}$  of fungal suspension (Table 1).

**Table 1. Time mortality response of second instar larvae of *Helicoverpa armigera* to various concentrations of *Metarhizium anisopliae*.**

Concentration (conidia $\text{ml}^{-1}$ )	Cumulative mean larval mortality (%) at 192 h	Time required for 50% larval mortality ( $\text{LT}_{50}$ ) (h)
$2.28 \times 10^{10}$	97.5	79.43
$2.28 \times 10^9$	92.5	85.11
$2.28 \times 10^8$	85.0	97.72
$2.28 \times 10^7$	80.0	104.71
$2.28 \times 10^6$	67.5	123.02
Control	5.0	-

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**Kiran S, Bramel PJ, Reddy LJ, and Vara Prasad KS. 2002.** Traditional pigeonpea cultivation practices in north coastal Andhra Pradesh - A tribal legacy. Patancheru 502 324, Andhra Pradesh, India: ICRISAT. 48 pp.

An effort is made to: (1) Document the different pigeonpea landraces grown in the tribal areas of north coastal districts of Andhra Pradesh, India; (2) Document folk names, crop production aspects, seed production and conservation methods, and storage structures used to store pigeonpea seed; (3) Give a word of appreciation, on record, to the farming communities that have been maintaining the landraces in situ; (4) Emphasize on the growing importance of maintaining landraces in situ; and (5) Create awareness among the younger generations in these areas and motivate them to take up the challenge of maintaining their traditional wealth.

**Piara Singh, Vijaya D, Srinivas K, and Wani SP. 2002.** Potential productivity, yield gap, and water balance of soybean-chickpea sequential system at selected benchmark sites in India. Global Theme 3: Water, Soil, and Agrobiodiversity Management for Ecosystem Health. Report No. 1. Patancheru 502 324, Andhra Pradesh, India: ICRISAT. 52 pp.

Soybean is the predominant crop in the target region of India and has the potential to be followed by chickpea crop on a larger scale in the post-rainy season. Using the CROPGRO models of soybean and chickpea, this study examined the potential yields, yield gap, and water balance of the soybean-chickpea sequential system for the 24 selected benchmark sites within the soybean production zones of India. Considering the variability in soils and climate, this simulation study showed that the average potential productivity of the soybean-chickpea system under rainfed situation ranged from 1390 to 4590 kg ha<sup>-1</sup> across sites. The current level of productivity of the system across sites ranges from 970 to 1780 kg ha<sup>-1</sup>. The yield gap of 200 to 300 kg ha<sup>-1</sup> for the system indicates the potential to increase productivity with improved management under rainfed situation. However, higher increases in yields would be possible in good rainfall years or with supplemental irrigation.

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**Amarteifio JO, Munthali DC, Karikari SK, and Morake TK. 2002.** The composition of pigeon peas (*Cajanus cajan* (L.) Millsp.) grown in Botswana. *Plant Foods for Human Nutrition* 57(2):173-177.

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# Notes

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# Notes

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The semi-arid tropics (SAT) encompass parts of 48 developing countries including most of India, parts of Southeast Asia, a swathe across sub-Saharan Africa, much of southern and eastern Africa, and parts of Latin America. Many of these countries are among the poorest in the world. Approximately one-sixth of the world's population lives in the SAT, which is typified by unpredictable weather, limited and erratic rainfall, and nutrient-poor soils.

ICRISAT's mandate crops are sorghum, pearl millet, chickpea, pigeonpea, and groundnut - five crops vital to life for the ever-increasing populations of the SAT. ICRISAT's mission is to conduct research that can lead to enhanced sustainable production of these crops and to improved management of the limited natural resources of the SAT. ICRISAT communicates information on technologies as they are developed through workshops, networks, training, library services, and publishing.

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