

Use of core and mini core collections in preservation and utilization of genetic resources in crop improvement

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Abstract

Plant genetic resources are the most valuable and essential basic raw material to meet the current and future needs of crop improvement programmes and the demands of increasing populations. The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT, established in 1972) responded to this need by establishing a Genetic Resources Unit (GRU) for assembling, characterizing, evaluating, maintaining, conserving, documenting and distributing germplasm of the mandate crops (sorghum, pearl millet, chickpea, pigeonpea and groundnut) and their wild relatives, and six small millets (finger millet, foxtail millet, barnyard millet, kodo millet, little millet and proso millet). The efforts have yielded the assembly of 113 849 germplasm accessions in the ICRISAT genebank and over 5.5 million accessions globally. Unfortunately, only a small proportion of this large collection has been used in improving crops. Developing core collection (about 10% of the entire collection) has been suggested as a method of enhancing the use of the germplasm. However, even this number could be large and unmanageable if the entire accession is several thousands. A methodology to reduce the size further and select a mini-core that is about 1% of the entire collection, yet represents full diversity of the species has been developed. Core collections of sorghum, pearl millet, chickpea, pigeonpea, groundnut, and finger millet and mini-core collection of groundnut and chickpea have also been developed. The core and mini-core collections of chickpea and groundnut have been evaluated and diverse sources for early maturity, traits related to drought tolerance and large seed size in kabuli chickpea, and early maturity, tolerance to low temperature, and traits related to drought tolerance in groundnut have been identified. Their use in breeding will broaden the genetic base of the cultivars.

Introduction

Plant genetic resources are the most valuable, essential, and basic raw materials for crop improvement programmes to meet the demands of increasing populations. Vavilov (1926) was the first geneticist to realize the essential need for a broader genetic base for crop improvement. He and his colleagues collected germplasm of crops and their wild relatives globally. In the wake of new agricultural development in the early 1970s, the loss of traditional cultivars and landraces seemed to be the most urgent problem, and massive germplasm collecting efforts were made to address it. A network of international centres was executed from early 1980s to enhance the collection, conservation, evaluation, and documentation of the crop genetic resources (Plucknett et al. 1987). ICRISAT responded to this need by establishing a GRU for assembling, characterizing, evaluating, maintaining, conserving, documenting and distributing germplasm of the mandate crops (sorghum, pearl millet, chickpea, pigeonpea and groundnut) and their wild relatives, and six small millets (finger millet, foxtail millet, barnyard millet, kodo millet, little millet and proso millet).

A recent survey conducted revealed that over 5.55 million plant germplasm accessions have been assembled and conserved in 1308 genebanks, globally (FAO 1996). The database indicates that 48% of all accessions in the genebank are cereals, 16% legumes, 10% forages, 8% vegetables and the remaining include fruits, roots and tubers, fibre crops, oil crops and others. Currently this enormous volume of germplasm resources cannot be used effectively to conduct research and also requires funds for managing the genebank. As Frankel and Brown (1984) indicated, germplasm could be used, for a wider range of characters, if a smaller number of well characterized accessions were to be given priority for use in crop improvement research. To pursue the same idea, Frankel (1984) proposed manageable sampling of the collection or 'core

collection'. A core collection contains a subset of accessions from the entire collection that captures most of available diversity of species (Brown 1989a). The core subset thus formed can be evaluated extensively and the information derived could be used to guide more efficient utilization of the entire collection (Brown 1989b). The reduced collection size will also help in reducing expenses required to manage the genebank. The paper enumerates efforts to enhance utilization of germplasm resources in research and how core and mini-core collections have been of greater use.

Assembly of the germplasm

Soon after ICRISAT was established in 1972, efforts were made to assemble and collect the germplasm. The Rockefeller Foundation, working with the Indian Agricultural programme during 1960s, assembled over 16 000 sorghum germplasm accessions from major sorghum areas, and ICRISAT acquired 11 961 accessions of this collection in 1974 that existed in India and USA. Initially, ICRISAT had also acquired over 2000 pearl millet germplasm accessions assembled by the Rockefeller Foundation in India, and another 2000 accessions collected by the Institut Francais de Recherche Scientifique pour le Development et la Cooperation (ORSTOM) in Francophone West Africa.

The chickpea and pigeonpea germplasm initially acquired by ICRISAT consisted of the material originally collected and assembled by the former Regional Pulse Improvement Project (RPIP), a joint project of the Indian Agricultural Research Institute (IARI), the United States Department of Agriculture (USDA), and Karaj Agricultural University in Iran. Sets of this germplasm placed in several agricultural research institutes in India and Iran, and at the USDA were donated to ICRISAT in 1973. ICRISAT also acquired over 1200 chickpea accessions from the Arid Lands Agricultural Development Program (ALAD-Lebanon) Similarly, much of the groundnut germplasm was received from the Indian Groundnut Research Program, now the National Research Center for Groundnut (NRCC-Junagadh), and USDA (Raleigh, USA).

ICRISAT also assumed the responsibility of adding new germplasm of the five mandate crops. Special efforts were made to collect or assemble landraces and wild relatives from areas threatened by genetic erosion. Between 1974 and 1997, ICRISAT launched 212 collection missions in areas of diversity in 61 countries and collected 8957 sorghum, 10 802 pearl millet, 4228 chickpea, 3870 pigeonpea, and 2666 groundnut accessions. Apart from ICRISAT's own collection efforts and the major donations cited above, national programmes of other countries, namely, Ethiopia, Sudan, and India contributed to enrich the germplasm collections at ICRISAT.

The existing collections possess over 80% of the available diversity, yet there is continuing need to rescue the endangered germplasm of mandate crops globally. In recent years, a collection mission was organized and 19 samples of foxtail millet; 15 samples of proso millet, and four samples of pigeonpea in North Vietnam were secured. About 80 samples of pigeonpea with remarkable range of seed colour variability was collected from Bangladesh. The Malian national programme, Institut d'Economie Rural Rurale (IER), was interested in collecting groundnut germplasm in the Mopti region of northern Mali. This region is just below the Sahara Desert and is threatened by desertification. To meet this requirement, a mission that collected 23 germplasm samples from the fields or threshing floors was organized. A similar mission for pigeonpea was launched in Tanzania (2001) and 123 samples were collected. In 2002, 38 pigeonpea germplasm samples from Uganda and 48 samples from Kenya were collected. ICRISAT and Tchad Agricultural Research Institute (ITRAD) signed a memorandum of understanding to collect pearl millet, sorghum, and groundnut germplasm in Tchad. The execution of the work resulted in securing 163 germplasm samples (sorghum: 131, millet: 17, and groundnut: 15). Following the Germplasm Acquisition Agreements, ICRISAT has acquired 48 samples of sorghum, 5 of pearl millet, and one of groundnut collected in 1998-99 in Mauritania. The national programme and ICRISAT conducted these missions jointly and also shared the samples.

Germplasm conservation and distribution

Conservation

Germplasm conservation requires cleaning the seed materials, drying to minimal seed moisture content, storing in cool and dry conditions, and monitoring seed health during storage. In the ICRISAT genebank, the seeds of entire collection are stored in medium-term storage (MTS – 50C; 20% RH) in aluminium cans. Recent seed health monitoring on seeds conserved from 10-25 years (MTS) indicated >75% seed viability for majority of the accessions. Accessions with declining seed viability (<75% seed germination) are regenerated on priority and the old seeds are replenished with fresh stock. Additionally, germplasm seeds are conserved in long-term storage (LTS; -200C) after packing in the vacuumed aluminium foil pouches. Before packing, the seeds are dried to about 5% moisture content with the help of walk-in-drying room (100 m² size; 150C and 15%RH) facility. Conservation of the FAO-designated germplasm in LTS facility to about 70% of the entire collection has been achieved.

Germplasm distribution

The ICRISAT Genebank supplies healthy, viable, and genetically pure seeds to research workers. From the beginning of the research (1973) to 1996, about 593 388 seed samples were supplied to users in 134 countries. During the recent years (1997 to 2003), about 70,826 samples have been distributed. Additionally, 69 363 seed samples were distributed within ICRISAT for research and screening against various stresses.

Safety back-up: ICRISAT's agreement with FAO places the germplasm collections under the auspices of FAO, and requires safety duplication preferably at –18°C in countries outside India. There is a memorandum of understanding with the International Center for Agricultural Research in the Dry Areas (ICARDA) for safety duplication of chickpea germplasm and 2000 accessions have been deposited with the institution. Various options for a cost-effective, secure and long-term strategy for other crops are being explored under the World Bank-funded genebank upgrading project.

Insight of the germplasm supplied to users

One of the main areas of research is to assess the patterns of demand for germplasm accessions to guide future strategies for germplasm regeneration and management. The germplasm distribution data of sorghum, pearl millet, chickpea, and groundnut till 1998 were analyzed, and the following patterns of germplasm demand emerged.

Sorghum

Of the 36 727 germplasm accessions held in the genebank, 30 570 (83.2%) accessions were distributed at least once. Of this number, about 83% were landraces, 16% breeding material and 1% wild sorghums. Twelve accessions, including three zera-zera accessions were dispatched more than 99 times.

Pearl millet

Of the 21 392 accessions, 15 366 (71.8%) accessions were distributed at least once during 1973 - 1998. The diversity in the distributed material was almost the same as in the entire collection. A total of 1769 accessions were distributed more than 10 times. IP 4021, an earliest flowering accession from Gujarat, India was distributed 94 times.

Chickpea

A total of 112 818 samples of 16 311 chickpea accessions were supplied to scientists in 81 countries. A maximum of 302 requests were received for ICC 4973 (L 550), a kabuli cultivar from India. Shannon-Weaver diversity index (H') of the accessions distributed was similar to the accessions contained in the global collection, indicating that the diversity available in the entire collection has been distributed.

Groundnut

All assembled accessions except 794 (5.3% of the entire collection), have been distributed. Countries in Africa have received the maximum number of accessions (92.3%) followed by Asia (76.6%), Europe (5.6%), Americas (5.3%), and Oceania (2.2%). A maximum of 292 requests were received for the ICG 799 (Kadiri 3), a hypogaea cultivar from India.

Pigeonpea

The summary of germplasm distributed from 1974 to 2003 revealed that 65 749 germplasm seed samples were supplied to users. This number was out of 10 648 unique accessions indicating that only 78.1% of the accessions held in the genebank have been distributed. Scientists from India were the major recipients of the seeds (68.7% of the total). The pigeonpea accessions requested most frequently were: ICP 7035 (DSLRL-55), a germplasm collection from Madhya Pradesh (distributed 305 times), ICP 26 (T 21, distributed 267 times), and ICP 7182 (BDN 1, distributed 253 times).

Low utilization of germplasm resources

The overall summary of germplasm utilization from the ICRISAT genebank, across the crops, over 10 years (1994-2003) indicated that, annually, 302 seed requests (114 inside ICRISAT and 188 outside ICRISAT) were met by supplying 21 431 (9 534 inside ICRISAT and 11 897 outside ICRISAT) seed samples. These figures are lower than that of Marshall (1989) who opined that the germplasm collection use could be considered adequate when the total number of germplasm samples distributed each year is >20% of the total collections or the number of independent requests a year is more than five a year per 1000 accessions.

Information on use of germplasm in ICRISAT's crop improvement programmes is also scanty. The summary of parental lines used in the ICRISAT chickpea improvement programme, for example, (1978-2001) revealed that 12 184 parents were used in making crosses that included only 83 unique germplasm accessions of chickpea and five of wild *Cicer* species. The two most frequently used cultivars were L 550 (847 times) and K 850 (808 times). Similarly, the summary of parental lines used in the ICRISAT groundnut improvement programme (1986-2002) revealed that 23 547 parents were used in making crosses, but this included only 132 unique germplasm accessions of groundnut and 10 of wild *Arachis* species. The two most often used cultivars were Robut 33-1 (3096 times) and Chico (1180 times). In China, the data on utilization of basic germplasm in groundnut breeding was summarized, revealing that only a few lines were used (Jiang and Duan 1998). In the USA, 41 peanut cultivars of Runner, Spanish, and Virginia market types were studied to determine contributions from the ancestral lines and coefficients. Among the Runner market type peanuts, Dixie Giant was a source in all the pedigrees of Runner market type, and the Small White Spanish-1 also appeared in 90% pedigrees. The two lines contributed nearly 50% of the germplasm of Runner cultivars (Knauff and Corbet 1989).

Limitations of large collection

Owing to the large number, accessions could not be characterized effectively and the documentation was not achieved up to desired levels. This resulted in under utilization of germplasm resources as already quantified in the previous section. It is also costly to manage accessions in the genebank. An accession of self-pollinating species (chickpea), for example, costs US\$ 145.42 whereas cross-pollinating (pigeonpea) costs US\$ 214.88 for conservation in perpetuity (IPGRI 2002). This cost is enormous when the ICRISAT genebank is holding 113 849 accessions of the different crops. Financial support for genebanks is not increasing proportionately with the increase in volume of work; rather, it is scanty and uncertain. It is a necessity, therefore, to derive maximum benefits from the germplasm resources and make germplasm management and use in the genebanks cost-effective. This can be achieved by efficient characterization of germplasm, developing core and mini core subsets and improving the documentation to enhance access to germplasm data.

Means to enhance use of germplasm

Germplasm characterization and evaluation

Agronomic and botanical characterization is necessary to facilitate the use of germplasm. Evaluation of germplasm accessions for traits of agronomic importance enhances its utility for greater use by the research workers. To achieve the same, germplasm accessions of all the crops were sown in batches over the years and characterized for morphological and agronomic traits. Germplasm screening against stresses were conducted in collaboration with various disciplinary scientists. Grains were tested for nutritional value, such as starch, protein, oil content, cooking time, and so forth. Germplasm sets were evaluated over locations jointly with national agricultural research systems (NARS) in India, Nepal, Thailand, Indonesia, Ethiopia, Kenya, West Africa, and more intensively with the NBPGR (India). Following is the progress in brief on crop germplasm characterization at ICRISAT:

Sorghum

Germplasm characterization work started in 1973 by sowing the germplasm accessions in batches over the years. During 1997, the status of characterization work was reviewed and the gaps were identified. To fill these gaps, a set of 781 new germplasm accessions was sown for characterization. In addition, data were recorded on 2000 accessions for grain characteristics. About 498 germplasm accessions received earlier from South Africa and 348 accessions of converted zera-zera sorghums were characterized and classified. Protein content of 364 accessions analyzed during 2002 ranged between 8.3 to 17.7%. Out of 122 accessions of wild relatives of sorghum screened for shoot fly resistance, 11 accessions were identified as resistant in addition to the two accessions of sorghum (IS 2146 and IS 18551).

Pearl millet

During 1973-96, most of the germplasm accessions of pearl millet had been characterized. Work done was reviewed in 1997 and as result, 197 germplasm accessions were planted in fields in 1997 to record data on some traits that were lacking from earlier years. In subsequent seasons, 1815 and 1394 accessions were sown to record the data on days to flowering, plant height, spike length, and spike thickness.

Chickpea

Work on characterizing chickpea germplasm was almost fully achieved by 1996. Only a set of germplasm (66 accessions) awaited characterization and this was accomplished during the 1997/98 crop season. However, data on some traits on part of the germplasm were missing. To achieve the same, 1419 accessions were planted to record data on plant colour, 1995 for flowering duration, and six for growth habit, plant width, seeds per pod, and pods per plant. Forty-nine accessions of wild Cicer were characterized in the field, and another 100 accessions received from ICARDA were sown under extended day length conditions in the glasshouse and characterized for 22 traits. Preliminary screenings of the chickpea collection of Asian origin indicated the presence of good levels of genetic resistance, particularly to collar rot and *Botrytis* grey mould disease.

Pigeonpea

In continuing the work of germplasm characterization of pigeonpea, 388 accessions received from USA were sown during 1997 crop season for characterization and seed increase. Seed protein content was estimated on 779 accessions. The values ranged between 14.9 and 24.6%. Information on photoperiod reaction on 11 000 pigeonpea accessions was added to the database. A diverse set of 28 accessions of pigeonpea and 12 related wild species were tested for tolerance to damages by pod fly (*Melanagromyza obtusa* Malloch) and pod wasp (*Tanaostigmodes cajaninae* La Salle). The accessions ICPW 141, 278, 280 (*Cajanus scarabaeoides*), ICPW 14 (*C. albicans*), ICPW 214 (*Rhynchosia bracteata*), and ICPW 202 (*Flemingia stricta*) showed resistance to pod fly and pod wasp damage (Sharma et al. 2003).

Groundnut

Almost all the germplasm accessions had been characterized by 1996 for most of the traits. However, data for one or more traits was lacking for a large number of accessions. Such accessions were planted in fields starting from 1997 and the missing data were recorded. About 158 accessions of wild *Arachis* species were characterized using 70 descriptors. Rosette and early leaf spot (ELS) are the most destructive diseases of groundnut in West and Central Africa (WCA) and southern and eastern Africa (SEA). Out of 4420 groundnut germplasm accessions and 80 accessions of wild species evaluated, 20 genotypes in 1997/98 and 28 genotypes in 1998/99 showed low (<20%) disease incidence. Thirty-five additional sources of resistance to ELS were identified. Of these, 30 are from South America (mostly from Peru and Bolivia) and are Valencia types. Eleven accessions of wild *Arachis* species (ICGs 8131, 13211, 13222, 14855, 14856, 14888, 14875, 14907, 14924, 14939, and 14946) were highly resistant and 15 others were resistant to ELS. Eight wild species accessions (ICG 8131, 8193, 8904, 11560, 13212, 13261 and 15875) with low *Aspergillus flavus* colonization and aflatoxin contamination were identified. In West Africa, 500 accessions were screened over two years for tolerance to *Aspergillus* invasion and aflatoxin contamination. Thirty-one lines were consistently tolerant to seed invasion by *Aspergillus* and aflatoxin production. Against late leaf spot disease, 24 promising sources of resistance were identified from several wild species accessions. Out of 48 accessions of wild *Arachis* species screened against peanut bud necrosis disease (PBND), ICG 8131, ICG 8144, ICG 8944 were identified as disease free and ICG 11551 had <5% disease incidence. Insect-pests reduce yields in groundnuts. With concerted efforts at ICRISAT, several germplasm resistant sources that included six accessions for jassids (*Empoasca kerri* Pruthi) 11 for thrips (*Thrips palmi* Karny), one for aphids (*Aphis craccivora* Koch.), 10 for termites (*Odontotermes* spp.), and two for leaf miner (*Aproaerema modicella* Deventer) (Ranga Rao and Wightman 1999) were identified.

Geographical pattern of diversity

Chickpea

Data on 16,820 accessions of chickpea germplasm for seven morphological and 13 agronomic traits, and reaction to fusarium wilt was analyzed to determine phenotypic variation in the accessions from different geographical regions. The means for different agronomic traits differed significantly between regions. The variances for all the traits among regions were heterogeneous. South Asia contained maximum range variation for all the traits. The traits, seed colour and days to 50% flowering showed the highest pooled diversity index. Principal component analysis using 13 agronomic traits and clustering of the first three principal component scores delineated two regional clusters comprising Africa, South Asia, and Southeast Asia in the first cluster and Americas, Europe, West Asia, Mediterranean region, and East Asia in the second cluster (Upadhyaya 2003).

Groundnut

Data on groundnut germplasm (13 342 accessions) held in the ICRISAT Genebank for 16 morphological and 10 agronomic traits in two seasons were analyzed to study geographical patterns of variation. The phenotypic variation was found for most traits in all the regions. The means for different agronomic traits differed significantly among regions. The variances for all the traits among regions were heterogeneous. Principal component analysis using 36 traits and clustering on the first seven principal component scores delineated three regional clusters, consisting of North America, Middle East, and East Asia in the first cluster; South America in the second cluster; and West Africa, Europe, Central Africa, South Asia, Oceania, South Africa, East Africa, Southeast Asia, Central Asia and the Caribbean Islands in the third cluster (Upadhyaya et al. 2002).

Development of core and mini-core collections of mandate crops for enhanced utilization

Core collection

Improvement in yield potential and resistance to biotic and abiotic stresses is an important objective in crop improvement. Plant breeders have successfully improved yield potential of most crops, resulting in large production increases in the past five decades. However, yields have reached a plateau in several crops, and the lack of further significant progress is a cause for concern. One reason for this plateau is that breeders tend to confine themselves to their working collection, consisting largely of highly adapted materials, and rarely use more diverse germplasm sources. In India's chickpea programme, (the world's largest for this crop), for instance, 184 breeding lines evaluated in 2001 included only 13 germplasm lines (mostly for stress resistance) in their pedigrees compared to 284 cultivars/breeding lines. Thus, only a small fraction of available germplasm diversity (ICRISAT's genebank alone contains over 17,000 chickpea accessions) has been used. Studies by Frey (1981) indicate that introduction of new alien germplasm in advanced breeding programmes often increase yield potential. However, it is almost impossible to predict which germplasm accessions(s) will be suitable for use in breeding programmes because of high genotype x environment interactions.

To overcome this problem, Frankel (1984) proposed manageable sampling of the collection or forming a 'core collection'. The core subset can be evaluated extensively and the information derived used to guide more efficient use of the entire collection (Brown 1989b). ICRISAT has developed core collections of pearl millet (Bhattacharjee 2000), chickpea (Upadhyaya et al. 2001a), sorghum (Prasada Rao and Ramanatha Rao 1995; Grenier et al. 2001), groundnut (Upadhyaya et al. 2003), and pigeonpea (Reddy et al. 2004) for research globally.

Mini-core collection

In several crops having thousands of accessions in the *ex situ* collections in the genebanks, even a core collection could be unmanageably large and unwieldy. The International Rice Research Institute (IRRI) rice collection, for example, contains over 80 000 accessions, so even a core subset would be expensive and time-consuming to evaluate. The challenge is to further reduce the size of the core subset without losing the spectrum of diversity. A strategy for sampling the entire and core collections was developed to build up a mini-core subset involving two stages (Upadhyaya and Ortiz 2001). First, a representative core collection (10%) is developed from the entire collection, using information on origin, geographical distribution, and characterization and evaluation data. The core collection is then evaluated for various morphological, agronomic, and quality traits selected, and ultimately, a subset of 10% accessions from the core subset (that is, 1% of the entire collection) that captures most of the useful variation in the crop. At both stages, standard clustering procedures are used to separate groups of similar accessions, and various statistical tests are used to evaluate representatives of the core and mini core collections. We have developed mini-core collections of chickpea (Upadhyaya and Ortiz 2001), groundnut (Upadhyaya et al. 2002).

Evaluation of core and mini-core subsets to identify useful germplasm

Groundnut

Like elsewhere, the use of genetic resources has been limited in Asian groundnut breeding programmes, resulting in a narrow genetic base of cultivars. Utilization of exotic germplasm in breeding programmes is needed to enhance the diversity of cultivars. The Asian groundnut core collection (Upadhyaya et al. 2001b) consisting of 504 accessions (29 accessions of subsp. *fastigiata* var. *fastigiata*, 245 of subsp. *fastigiata* var. *vulgaris*, and 230 of subsp. *hypogaea* var. *hypogaea*), along with four control cultivars, was evaluated in multi-environments for 22 agronomic traits to select diverse superior germplasm accessions for use as parents in improvement programmes. Analysis of data, using the residual maximum likelihood (REML) approach, indicated that variance components due to genotypes were significant for all the 22 traits, and genotypes x environment interaction were significant for eight traits. Estimates of

broad sense heritability ranged from 35.45% for pod yield per plant to 97.96% for days to cessation of flowering, indicating relative reliability of selection for different traits. On the basis of performance compared to control cultivars in different environments, 15 *fastigiata*, 20 *vulgaris*, and 25 *hypogaea* accessions from 14 countries were selected. The selected accessions and control cultivars were grouped using scores of the first 15 principal components (PCs) in *fastigiata*, 20 PCs in *vulgaris*, and 21 PCs in *hypogaea*. The clustering by Ward's method indicated that the selected accessions were diverse from the control cultivars. These 60 diverse parents will provide the germplasm that can be used in improving programmes to broaden the genetic base of groundnut cultivars.

From the core of groundnut consisting of 1704 accessions (Upadhyaya et al. 2003), 21 early maturing landraces were selected and evaluated at 12400Cd (equivalent to 75 days after sowing (DAS) and 14700 Cd (equivalent to 90 DAS) in rainy season at Patancheru along with early maturing cv. Chico. ICGs 4558, 4729 and 9930 (1.97-2.15 t/ha) were as early as Chico, but produced 25-36% more pod yields than the early maturing cv. JL 24 at 12400 Cd in the 2001/02 post rainy season (ICRISAT 2003).

The mini-core set of accessions (Upadhyaya et al. 2002) has been useful in identifying better sources for drought resistance. The set was planted in the 2001-02 post rainy season at Patancheru and data were recorded on specific leaf area (SLA) and soil plant analysis development (SPAD) chlorophyll meter reading (SCMR). These two traits can be used as surrogate traits for selecting for water use efficiency (WUE). SCMR values were more strongly correlated with pod yield and other economic traits like 100 seed weight and harvest index than SLA. On the basis of higher heritability and lower proportion of genotype x season interaction variance to phenotypic variance, SCMR appeared to be more stable than SLA. On the basis of SLA and SCMR values compared to control cultivars, six *vulgaris* and 13 *hypogaea* accessions were selected. These accessions and control cultivars were grouped using scores of the first 15 principal components. The clustering by UPGMA method indicated that the selected accessions were diverse and can be used in groundnut improvement programmes to develop WUE cultivars with broad genetic base (our unpublished data).

Promising accessions for low temperature tolerance were selected using core collection of groundnut (Upadhyaya et al. 2003). The set of 1704 core accessions was screened for low temperature tolerance at germination under laboratory conditions. Of the tolerant accessions identified, landraces were selected as they have the original variation and were used to examine other traits of economic importance and genetic diversity. Five accessions that showed tolerance to low temperature, high pod yield and phenotypic diversity were: ICG 4331, 8797, 9515, 11 130 and 13 539. These accessions can be used in breeding programmes to develop high yielding, low temperature tolerant cultivars and to broaden the genetic base (Upadhyaya et al. 2001c).

Holbrook et al. (1997) examined all accessions in the groundnut core collection (Holbrook et al. 1993) for reaction to the groundnut root-knot nematode (*Meloidogyne arenaria* (Neal) Chitwood race 1). Thirty-six core accessions showed a reduction in root galling, egg-mass rating, egg count per root system, and egg count per gram of root compared with Florunner. Twenty-one accessions showed a 70% reduction in egg count per root system and per gram of root, and two accessions showed a 90% reduction of these variables compared with Florunner. The striking geographical patterns observed were the relatively large numbers of resistant accessions from China and Japan.

Use of groundnut core collection has also resulted in the first report of resistance to preharvest aflatoxin contamination (PAC) in the US groundnut germplasm collection (Holbrook 1998). Fourteen core accessions were observed to have, on average, a 70% reduction in PAC in multiple years of testing. Six of these accessions exhibited a 90% reduction in PAC in multiple years of testing.

The most agronomically acceptable portion of the core collection has also been evaluated for resistance to *Rhizoctonia* limb rot (*Rhizoctonia solani* Kuhn AG-4) (Franke et al. 1999). This subset of the core collection consisted of 66 accessions having a spreading or spreading bunch growth habit. Six core accessions had a high level of resistance to *Rhizoctonia* limb rot.

Chickpea

From the chickpea mini-core collection consisting of 211 accessions (Upadhyaya and Ortiz 2001), 16 germplasm lines having large seed size were identified (up to 63 g 100⁻¹) and evaluated in replicated trials during the 2001/2002 post rainy season. All the 16 lines had greater 100-seed weight than the control cultivars (L 550 and ICCV 2) and 10 of them were also better for seed yield. ICCs 8155, 14194, 14199, 14204, 14205, 14926, and 16670 showed a good combination of large seed size (100-seed weight 42-54 g) and seed yield (1.13-1.56 t ha⁻¹ compared to 1.11 t ha⁻¹ for ICCV 2). Diversity analysis indicated that L550 and ICCV 2 were closely related, while the new large-seeded lines from them were diverse. These lines can be used to develop diverse, large-seeded kabuli cultivars.

From the same chickpea mini-core, 28 early maturing accessions were selected and evaluated in a replicated trial along with four controls (ICCV 2, Annigeri, ICCV 96029, and Harigantars) during the 2001/2002 post-rainy season. Seventeen lines had maturity similar to that of Harigantars and greater seed yield. ICC 10629, 10996, 10976, 11021, and 14648 were some of the high-yielding lines that were better than Annigeri (1.44 t ha⁻¹). Diversity analysis indicated that the Harigantars, ICCV 2, and ICCV 96029 were closely related and the new lines from them were diverse. Using these lines in breeding programmes will broaden the genetic base of the resulting cultivars.

The mini-core of chickpea (Upadhyaya and Ortiz 2001) was also screened to identify accessions with drought tolerance traits. The depth of root and density are presumed to be the main drought avoidance traits identified to confer seed yield under terminal drought conditions (Turner et al. 2000). The accessions with deepest roots were: ICC 1431, 8350, 15697, 3512 and 11498. These accessions had deeper roots than the already known drought tolerant cultivars, ICCV 2, ICC 4958, and Annigeri (Krishnamurthy et al. 2003).

Documentation and information exchange

Documentation

The vast germplasm data gathered on chickpea and pigeonpea germplasm has been summarized and presented to the users in form of catalogues (Pundir et al. 1988; Remanandan et al. 1988). Multilocational evaluation data were summarized separately and two catalogues on forage sorghum germplasm (Mathur et al. 1991, 1992), one on chickpea (Mathur et al. 1993a), and two on pearl millet (Mathur et al. 1993b and 1993c) were produced. A Manual of Genebank Operations and Procedures was published (Rao and Bramel 2000) documenting the history of the collections, procedures for germplasm acquisition, maintenance, documentation, conservation, and distribution. Existing procedures were reviewed and revised to maintain the collections according to international standards. A taxonomic key for the identification of wild species of the mandate crops has also been included in the manual. Besides these catalogues, several research articles on germplasm research have been published.

Genebank Information System

The genebank operations span from collection and conservation of germplasm to its distribution and use. The need to adhere to international standards of germplasm conservation and ensure transparency requires that these operations are computerized. This measure will help greatly in automating the routine operations of the genebank in a workflow system and in an efficient and timely dissemination of the information. The online genebank management system, developed in Visual Basic[®] 6.0 with SQL Server[®] 7.0 as the backend, is structured to query the available information.

System-wide Information Network for Genetic Resources (SINGER)

The passport information on ICRISAT germplasm collections was updated for the geographic coordinates and location using Microsoft Interactive World Atlas 2000[™]. The available evaluation data have been computerized for nearly 30 000 accessions. The prototype of the new SINGER front-end developed and germplasm databases were web-enabled using SGRP Toolkit. The passport data of sorghum and pearl millet were updated with available geographic information. In sorghum, information on latitude and longitude of the collection sites has been

updated for 15 391 accessions and in pearl millet for 15 745 accessions, including 661 accessions of wild species. The small millets passport and characterization data were harmonized, validated, and replicated to SINGER database. The West and Central Africa groundnut germplasm data were made available on the web. A common but stand alone seed dispatch system for Africa with MS ACCESS' as backend and Visual Basic' as front end was completed. An online core selector programme was developed based on Hintum (1999).

Gaps in the germplasm collections

In the initial years, assembling germplasm that existed with other institutes in India and abroad were given priority. Subsequently, germplasm collecting missions were introduced in areas that were poorly represented. With passport and characterization data and their summarization, specific germplasm types that are not adequately represented in the genebank (genetic gaps) and also less represented regions were arrived at.

Way forward

Some gaps, such as low number of groundnut botanical variety *hirsuta* and *aequatoriana*, and sources for yellow endosperm and white seeds in pearl millet are apparent in the germplasm collection. There is need to secure more germplasm of these types. Germplasm should also be secured from priority regions.

Most germplasm accessions have already been characterized for a minimum set of descriptors. However, some accessions still need to be characterized and this should be given priority.

One of the significant features of the work on core subset has been the impetus that it is giving to exploring the ways in which PGR data can be collated and used more efficiently. Thus there is considerable interest in combining data from evaluation work with that obtained from biochemical or molecular studies in carrying out multivariate analyses of large data sets involving many hundred of accessions.

Germplasm conservation is a dynamic process. Knowledge of a gene pool is never complete and must be improved continually. This raises the question of the way in which changes to the constitution of the core subset should be managed. Accessions shown to have significant new variants that are absent from the core should undoubtedly be added to it, but on what basis and in what way?

If core collections are to be effective as part of a process of improving the conservation and use of plant germplasm, they should be properly documented. Core subsets can act as communication devices and, for this to be effective, the data needs to be widely accessible and used. This requires database management procedures that allow optimal and full exchange and use of the information.

A basic component of the development of core subsets is the collaboration between genebank managers, plant breeders, and other agricultural research scientists. This collaboration will increase qualitatively the effectiveness of the conservations efforts as well as the effectiveness of the breeding and related research necessary for crop improvement. It will also draw on resources that may not otherwise be available to those concerned with conservation.

The core and mini-core subsets developed should be characterized and evaluated extensively to draw benefits for the users. Molecular characterization of the mini-core subsets will help in elucidating the genetic diversity of the collections.

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