Enhanced Utilization of Plant Genetic Resources in Crop Improvement Programmes

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On domestication, the crop species typically inherited a small fraction of the natural genetic variation present in their wild progenitors. On-farm crop diversity diminished further with the advent of modern plant breeding, resulting in the creation of plant varieties that optimized adaptation at the cost of adaptability. Every quantum jump in productivity was also accompanied by a narrowing down of the breeders' crossing block. Once a new thresh hold is reached, plant breeders tend to make crosses within the small and related set of this newly improved germplasm only. This has led us to a predicament of stagnation in genetic gains. Changes in biotic and abiotic stress regimes on account of climate change and natural resource depletion are expected to pose serious challenges in view of the narrow genetic base of breeding programmes. The problem is likely to be accentuated by restricted germplasm flow in deference to propriety concerns. Enhanced utilization of plant genetic resources (PGR) is clearly warranted. Impediments to use of PGR including lack of local adaptation and linkage drag are discussed along with measures to overcome them. 'Genetic incorporation' as a PGR utilization strategy in contrast to the generally followed 'gene introgression' is highlighted. It is a base broadening exercise which gradually improves current adaptation of breeding material and has the potential to cater to unforeseen breeding needs. Crop wild relatives (CWR) are a subset of PGR which show great scope for enhanced utilization as they represent distinct, genetically diverse but underutilized gene pools for crop improvement. Instances of commercial deployment of genes from the wild are listed. Salient pre-breeding work for wheat improvement at PAU is discussed as an illustration for enhancing use of PGR.

Key Words: Crop Improvement, Crop Wild Relatives, Gene Introgression, Germplasm Utilization, Plant Genetic Resources

About 4,00,000 species of flowering plants are estimated to exist today (Govaerts, 2001; Bramwell, 2002). Thousands of these species are or have been used by human beings in one form or the other. Most of these partially domesticated or wild-collected species are found in tropics. Nearly 7,000 species mentioned in the records of Plant Resources Project of South Asia (PROSEA) are used in that region (Jensen *et al.*, 1991) and a similar number of species have been listed as Plant Resources of Tropical Africa (PROTA). Apart from this, several thousand plant species are in use in Mediterranean and temperate regions of the world. The uses of these plant species include food, food additives, feed and fodder, fuel and various uses as household and industrial materials. Two major further uses of plants are as medicinal plants (between 65,000-118,000 species) and ornamentals (about 30,000 species). It was this human-plant contact, happening on an evolutionary time-scale, which led to plant domestication and emergence of agriculture. About 7,000 plant species are, or have been cultivated to some degree, world-wide (Wilson, 1992). A large proportion of these domesticated species catered to the primary human need-food. The number of such species however dwindled sharply with loss of traditional cultures and the shift to high production agriculture for supporting the human population explosion. In the present era only 103 species of plants contribute 90% of the food needs of the world (Prescott-Allen and Prescott-Allen, 1990). Further, just 30 plant species, mainly comprising of staples, supply most of human nutrition and three of these (wheat, maize and rice) provide more than half of the planet's food (Heywood, 2008). Thus, the agricultural revolution that began 10,000 years ago has gradually, but consistently seen a narrowing down of the number of plant species on which humankind depends. At the same time, the loss of natural plant diversity has continued apace and as per 'Gran Canaria Declaration on Climate Change and Plant Conservation' as many as two-thirds of the world plant species are in danger of extinction during the course of 21st century. Shrinking diversity at the species level in both natural and cultivated domains provides the larger context in which the utilization of plant genetic resources may be placed.

Loss of Genetic Diversity in Crop Plants

Plant genetic resources can be broadly considered as all materials that are available for improvement of a cultivated plant species. The entire gamut of plant material, of current as well as potential use in breeding of a crop, thus qualifies as plant genetic resources. In the typical plant breeding sense, however, genetic resources are generally those materials that without selection for adaptation to the target environment do not have an immediate use for the breeders (Hallauer and Miranda, 1981). The relationship of a crop with its plant genetic resources has often been viewed through the concept of primary, secondary and tertiary gene pools (Harlan and de Wet, 1971). Beyond these gene pools, based on ease of sexual genetic transfers, are the rapidly expanding possibilities of transfer of isolated or cloned genes. The gene pools may gradually coalesce into a virtual gene ocean. The entire biodiversity of living species may, thus, come to be viewed as a potential genetic resource (Dhillon and Agrawal, 2004). While the possibilities of horizontal gene transfer offer the prospect of molecular unification of the biosphere, the crop plant species have derived their identity from a process of division and isolation. Domestication created the fundamental demarcation of plant genetic resources into cultivated and wild types. By its very nature of being a rare event, crop domestication represents an acute bottleneck and sampled a small proportion of total diversity of the wild population (the 'founder effect': Ladizinsky, 1985). The narrow genetic base because of bottlenecks at domestication has been highlighted for various crop species including rice, durum and bread wheat, *Phaseolus* beans, tomato, pigeonpea, chickpea, *Citrus*, and possibly *Musa* and yam (Spillane and Gepts, 2001). The bottleneck is perpetuated further by various reproductive isolation factors preventing gene flow. The domesticated plants may be carried by the cultivators to sites far removed from its original habitat. During transfer between latitudes, there may be a further narrowing of the genetic base because the population would not be well adapted to the new day-length conditions, and so only a small number of the genotypes would survive (e.g., potato in temperate areas). Examples of crops with narrow genetic bases arising during migration include soybean in the US, maize in Africa and the US (Tallury and Goodman, 2001), sorghum, millet and lentil in South Asia (Erskine *et al*., 2001). There may be other chance occurrences that narrow the genetic base such as disease epidemics, which may decimate populations. Often domestication is accompanied by an amphiploidization event, as in case of hexaploid wheat, thus erecting barriers to gene flow from diploid progenitors.

Post-domestication, the crops evolved under human selection but continued to possess a breadth of genetic variation in order to overcome challenges from changes in biotic and abiotic milieu. The advent of modern plant breeding resulted in creation of plant varieties that optimized adaptation at the cost of adaptability. On farm diversity was seriously undermined. Every quantum jump in productivity was also accompanied by a narrowing down of the breeders' crossing block. Once a new thresh hold is reached, plant breeders tend to make crosses within the small and related set of this newly improved germplasm only. This places a ceiling on the further progress or gain from a breeding programme on account of insufficient genetic diversity. For instance, once the small initial set of semi-dwarf wheat or rice lines were produced, it became counter productive to involve the much larger set of tall lines in crosses. Tall segregants virtually competed out the dwarf ones and produced a large proportion of undesirable individuals, at least in early segregating generations. Ironically in case of wheat, the two major yield jumps, first on account of semi-dwarf plant type and second resulting from winter wheat x spring wheat hybridization meant that breeders would restrict their subsequent efforts to within the improved sets, though both improvements resulted from use of a divergent gene pool.

The narrowed genetic base of germplasm is often evident from plateau in yield gains as have been observed in several crop plants. Historically, instances of more disastrous consequences have also been observed. Often quoted instances include the blight epidemic caused by *Phytophthora infestans* in potato (*Solanum tuberosum*) in Western Europe in 1845/1846, the havoc caused by *Bipolaris* on T-cytoplasm maize in the USA in 1970 (Campbell and Madden, 1990) and the *Fusarium graminareum* epidemic in wheat and barley in western USA during 1994 to 1996 (FAO, 1996a).

Plant breeders sometimes adopt a deliberate diversity restricting approach to conserve market or industrial processing oriented quality traits. The examples of malt barley have been well studied and breeding with a defined, small set of parents has been argued to be advantageous (Wych and Rasmusson, 1983). Further, narrowing of the genetic base may result from specialization within crops: for example, breeding of winter wheat has been mainly done by using only winter germplasm and breeding of spring wheat by using only spring germplasm (Spillane and Gepts, 2001). The genetic base of hot peppers (*Capsicum annum*) is partitioned by the specialized requirements for distinct uses (e.g. thin pericarp required for drying, whilst fresh use requires small fruit). The genetic base of *Brassica* is similarly partitioned into different morphological types.

The Need for Enhancing PGR Utilization

Multiple cycles of narrowing of genetic diversity in crop plants in recent years have been topped with extensive churning and utilization of the available variation. It is now imperative in most of the situations to explore beyond the elite germplasm or even beyond the species boundary. There is a need to integrate wide hybridization and alien introgression with mainstream plant breeding. The need for enhanced utilization of plant genetic resources is reflected in the Global Plan of Action for the Conservation and Sustainable Use of Plant Genetic Resources for Food and Agriculture (PGRFA) adopted at the International Technical Conference on Plant Genetic Resources held in 1996 (FAO, 1996b). The plan has genetic enhancement and base broadening, besides on-farm diversification as its priority areas.

Presently, the need for enhancing PGR utilization has acquired greater urgency. The need for greater genetic diversity in sources of resistance to biotic stresses had often been emphasized and several successful uses of PGR are known. Recently, the prospects of using genetic approaches for combating abiotic stresses also have improved on account of increased insight into tolerance mechanisms and availability of molecular tags. Relevant variation for pursuing this approach in the cultivated germplasm is however inadequate (Hajjar and Hodgkin, 2007) and use of land races/crop wild relatives has acquired importance. The abiotic stress tolerance agenda has expanded further in response to emerging climate change scenario. Climate change is now a certainty and looms as a threat of unprecedented scale to agricultural systems and food security of the country. It demands an urgent reorientation of breeding programmes in terms of genetic and genomics input as well as screening strategy. Utilization of newer sources of genetic variation is warranted. Another major change in the present situation is the apprehended shrinkage of germplasm exchange. In the emerging IPR regime it has become crucial to impart self-sufficiency to the breeding programmes. Breeding programmes with greater depth of genetic resources are expected to flourish. Continuously developing a wider spectrum of genetic variability thus becomes an essential upstream activity. Regular infusion of diverse genetic variation can help offset negative consequences of shrinking germplasm exchange to some extent. Another aspect of the present situation arises from decades of intensive and exploitative agricultural practices which have taken a toll of valuable natural resources. Resources like water will have to be rationalized in face of scarcity while self imposed restrictions, as on use of nitrogenous fertilizers and pesticides, have become essential for environmental health. This poses an extremely tough challenge to crop improvement as further gains in productivity need to come in spite of reduced inputs. Such variation is again likely to be available in land races and wild relatives rather than improved materials. All these considerations point to an urgent intensification of PGR utilization and reorientation of crop breeding goals and practices.

Impediments to the Use of PGR in Crop Improvement

Lack of environmental adaptation of the PGR to be used as donor is a major impediment to its use in classical plant breeding. Linkage drag is the other major reason restricting the use of PGR in crop improvement. Assuming a target locus in the centre of a 100 cM chromosome, about 53 cM remain around the target gene in the third backcrossing generation (BC_3), and in BC_{10} the average linkage drag is still about 20 cM (Stam and Zeven, 1981; Welz and Geiger, 2000). If this linkage drag contains undesirable alleles from the PGR, the performance of the backcrossing products can be unsatisfactory. Linkage drag accompanying translocated rather than recombining chromosomes can be several times more tenacious and sometimes pose almost insurmountable difficulties in commercial utilization of alien genes. Epistasis or co-adaptation of genes within both breeding population and PGR means that natural or artificial selection has favoured specific combinations of alleles at different gene loci within each type of material. The specific allele combinations are lost after crossing and recombining the two types, leading to so-called ‘recombination losses’ (Haussmann *et al*., 2004). It takes several generations to establish new favourable allele combinations through selection.

Efficient utilization of germplasm requires awareness of target traits for which variation in elite germplasm is lacking, followed by identification of suitable donor germplasm. An efficient transfer methodology should be in place and implemented in a timely manner. Further, choice of recipient genotype should ensure commercial viability of the end product. Coordination at all these steps is an essential components for successful PGR utilization as exemplified by incorporation of grassy stunt virus (GSV) resistance from *Oryza nivara* to cultivated rice (Brar and Khush, 1997) and indicated by the following chronology of developments. Dr. SD Sharma had collected wild rice *O. nivara* from Eastern India in 1966 and 6723 accessions were conserved at IRRI, Philippines. At that time GSV was an unimportant disease and no screening work was in progress. Wide adoption of semi-dwarf rice varieties led to emergence of GSV as a major disease. In early 1970s, GSV destroyed 2,87,000 acres of rice in Indonesia, India, Sri Lanka, Vietnam and Philippines. Outbreak of GSV led to screening of this material by Drs. GS Khush and KC Ling at IRRI. Only one accession was found to be highly resistant. Resistance was transferred to IR24 and subsequently several other cultivars including IR36. This resistance from *O. nivara* became widely deployed, covering 74 million acres in Indian subcontinent, China and South East Asia.

PGR utilization involving a pre-breeding step will translate into commercial use in a time frame different from the one applicable to conventional breeding. The long duration of gene transfer process discourages crop breeders to exploit exotic and un-adapted donors. This is well illustrated by another example from rice, the saga of bacterial leaf blight resistance gene *Xa-21*. The broad spectrum resistance conferred by this gene was observed in *Oryza longistaminata* lines from Mali, Africa in 1977. It took almost 20 years of intensive research using molecular as well as conventional tools for commercial deployment of this gene. For donor species with distinct/non-homologous genomes (unlike *O. longistaminata* which has same genome designation as cultivated rice) the gene transfer and commercial utilization is likely to take even longer. Several rust resistance genes (*Yr 40, Lr 57, Lr58* etc.) have been mobilized from non-progenitor wild wheats at Punjab Agricultural University, Ludhiana. Almost two decades of continuous efforts starting with evaluation of donor accessions in the 1980s was required before the genes could be transferred to elite wheat lines (Dhaliwal *et al*., 2003). PGR utilization, particularly when distant sources are involved, requires continuity and sustained efforts over time frames which sometimes may not be harmonious with regular project tenures.

Systematic documentation of information on PGR collections can greatly enhance their utilization for crop improvement. Of particular importance are information on economically important traits, e.g. resistances, quality and specific adaptation traits. Beyond preliminary evaluation, information on genotype x environment interactions and affiliation to heterotic pools (if hybrid breeding is relevant) can facilitate targeted exploitation of PGR. In the Indian context several publications are now available for use by breeders of foodgrain crops (Dhillon *et al.*, 2006b), oilseeds and cash crops (Dhillon *et al.*, 2004b) and horticultural crops (Dhillon *et al.*, 2004c). The breeders also need to look beyond crop specific information and be aware about policy matters and regulations for PGR exchange and utilization (Dhillon *et al.*, 2004a; FAO, 2010).

Approaches to PGR Utilization

It is unlikely that all of the potentially useful combinations have been assembled in any single group of locally adapted stocks (Simmonds, 1962). At the same time it may not be possible for a mainstream breeding programme to exploit new un-adapted germplasm to create new gene combinations that might excel over the best pre-existing commercially deployed materials. While adapted germplasm may be at one type of adaptive peak, the exotic prospective donor will be at a different peak, with a major maladaptive valley preventing gene flow between them (Whitlock *et al*., 1995). Base broadening may be likened to building bridges between different fitness peaks. The conventional breeding methodology will need to be modified to achieve this. While this is a matter of disassembly of co-adapted gene complexes, more serious problems to utilization of PGR are posed when the donor is distant and one or more types of reproductive barriers may be present. In such cases a pre-breeding phase becomes mandatory. Pre-breeding refers to activities designed to identify desirable characteristics/genes from un-adapted plant genetic resource and to transfer them to an intermediate product that breeders can manipulate.

Simmonds (1993), Spoor and Simmonds (2001) have listed two major approaches to utilization of PGR: introgression and incorporation. They have strongly argued in favour of incorporation in contrast to the generally followed single gene transfers or introgression. Salient features of the incorporation approach are as follows:

- Making use of the broadest possible starting materials, consistent with the specific objectives of the programme. Evaluation of such material is, in this context, often irrelevant, since most of the starting material is expected to be un-adapted to the target environments anyway.
- A need for extensive recombination (If natural outcrossing rates are sufficiently high, recombination is easy; otherwise controlled crossing or the use of male sterility genes may be necessary).

- Weak and progressively decentralized selection. Whenever possible, selection should be based on multiple large populations and carried out over several generations in target environments.
- The maintenance of the above process as a programme distinct from conventional breeding programmes, until usable material is produced.

Examples of application of the incorporation approach include the development of barley (*Hordeum vulgare*) composite-cross at Davis, California (Cooper *et al*., 2001), dynamic gene pool management in wheat (Goldringer *et al*., 2001); pearl millet (*Pennisetum glaucum*) composite populations developed in Africa (Niangado, 2001). Another, long-term base-broadening project in maize is the Hierarchical Open-ended Population Enrichment (HOPE) project in Canada (Kannenberg and Falk, 1995; Kannenberg, 2001). Other projects in maize include the Genetic Enhancement of Maize (GEM) programme in which material was selected on the basis of the evaluation data of the previous Latin American Maize Programme (LAMP) (Pollock and Salhuana 2001). In the PAU maize breeding programme, it was seen that intra-population improvement became more rewarding when accompanied by introgression of new germplasm (Dhillon *et al*., 2006a). Similarly for inter-population three broad based heterotic pools were developed to serve as source of populations for derivation of inbred lines (Dhillon *et al*., 1997).

In addition to introgression and incorporation, Cooper *et al.* (2001) listed pre-breeding as the third PGR utilization approach. The first two approaches are based on freely recombining donors presumably from the primary gene pool and leaves out the major category of crop wild relatives (CWR) which is covered under the third approach. The utilization approach to be followed for CWR would actually depend on its genomic constitution. A full-fledged pre-breeding phase would be typically necessary where the donor species genome is distinct/non-homologous to the recipient crop species. Often a chromosome engineering step would be involved for translocating the relevant donor chromosome segment to the recipient genome. This would entail use of specific genetic stocks (as in case of *Ph1* locus mediated homoeologous recombination in wheat) or ionizing radiation facilitated translocations. A large number of chromosomal translocations from alien species for rust resistance have been obtained in wheat using these two methods. On the other hand, an incorporation based approach can be followed for wild progenitors whose genomes show good homology with the cultivated species. In case of homology with the donor, the AB-QTL method provides an excellent opportunity for transfer of ‘hidden genes’ (e.g., for productivity) following a simultaneous molecular marker analysis and gene transfer approach (Tanksley and Nelson, 1996). This method has helped crop breeders to view un-adapted germplasm and crop wild relatives as potential donor of traits which are not evident in the donor phenotype. In contrast to the AB-QTL method, Eshed and Zamir (1994) suggested the approach of establishing a population of NILs such that the donor chromosome segments are evenly introgressed over the whole recipient genome. Ideally, the total genome of the exotic donor is represented in the established set of NILs. This NIL population, named introgression library (IL), consists of a set of lines, each carrying a single marker-defined donor chromosome segment introgressed from an agriculturally un-adapted source into the background of an elite variety (Zamir, 2001). A major drawback of the AB-QTL and IL approach is that exploration of even a single donor line involves huge amount of breeding as well as molecular marker work. It is hard to extend this approach to large number of donor lines. Moreover, there are no specific guidelines for narrowing down to a smaller set of donors as we are looking mainly for hidden and interactive variation in context of a particular recipient line.

Narrowing down to a smaller set of prospective donors is not just an issue for AB-QTL and IL based approach but also essential for taking up detailed evaluation and subsequent utilization. This can be achieved by assembling core collections. Core collection may be defined as a limited set of accessions representing, with a minimum of repetition, the genetic diversity of a crop species and its wild relatives (Frankel, 1984). In context of specific gene bank collection, the core collection represents the genetic spectrum in the whole collection and should include as much of its genetic diversity as possible (Brown, 1995). The practical norm is to limit the entries in a core collection to ~10%, using the sampling theory of selectively neutral alleles, with a ceiling of 3000 per species. This level of sampling is effective in retaining 70% of alleles of entire collection. However, core collections based on basic passport and characterization data for major morphological characters, and developed primarily to make genetic diversity available to researchers have limited value unless this is evaluated extensively for traits of economic importance. This will make the core collection and eventually entire collection more useful to plant breeders and other crop improvement scientists (Upadhyaya *et al*., 2008). In some major crops, size of the entire germplasm collection is very large, even a core collection size becomes difficult for extensive evaluation by breeders or researchers. To overcome this, the concept of 'Mini-core collections', was given. A mini-core collection consists of 10% accessions in the core collection and only 1% of the entire collection (Upadhyaya and Ortiz, 2001) and represents the diversity of the entire core collection. These can be thoroughly evaluated and the information so derived can be utilized for improving the efficiency of breeding programmes. Molecular biology and bioinformatics can facilitate assessment and utilization of genetic diversity e.g., Generation Challenge Program (GCP) on 'Unlocking Genetic Diversity in Crops for the Resource-Poor' (www.generationcp.org) is designed to utilize molecular tools and comparative biology to explore and exploit the valuable genetic diversity existing in germplasm collections held at the CGIAR and national gene banks, with particular focus on drought tolerance.

Utilization of Crop Wild Relatives

Crop wild relatives (CWR) are species closely related to crops, including crop progenitors. CWR have been identified as critical resources that are vital for wealth creation, food security and environmental stability in the future (Meilleur and Hodgkin, 2004; Stolton *et al*., 2006; Maxted *et al*., 2008). Historically, the commercial use of wild relatives started in the late 19th century when wild *Vitis* species were used as rootstocks to protect grapes cultivars from *Phylloxera* aphids and *Meloidogyne* nematodes (Prescott-Allen and Prescott-Allen, 1988). In sugarcane, virus resistance was incorporated from *Saccharum spontaneum* in the first half of the 20th century and later all sugarcane varieties were developed using three to five species (Stalker, 1980). In 1941, first tomato variety having *Fusarium* resistance gene from *Lycopersicon pimpinellifolium* was released and subsequently a large number of wild species have been used to introgress genes into cultivated tomato (Rick and Chetelat, 1995). In the middle of 20th century, the value of CWR was widely recognized and breeding efforts to explore the potential of wild relatives were initiated in many crops which paid rich dividends e.g. late blight resistance from *Solanum demissum* and *S. stoloniferum*, resistance to viruses from these species and from *S. chacaoense* and *S. acaule* in potato (Ross, 1986). Starting with the work of Sears (1956), large set of stocks carrying the wheat-alien translocations conferring resistance to diseases and insect pests were developed and characterized in wheat (Friebe *et al*., 1996). The use of wild relatives increased in 1970s and 1980s (Hodgkin and Hajjar, 2008) and in the mid 1980s, Prescott-Allen and Prescott-Allen (1988) asserted that the achievements were substantial enough to recognize the potential of wild relatives. By this time there are about 31 crops where CWR have been used to the extent that cultivars with wild genes were available (Prescott-Allen and Prescott-Allen, 1988). International Board for Plant Genetic Resources (IBPGR) working with national programmes, initiated a number of collecting missions that primarily focused on CWR (IBPGR, 1991). With the improvement in interspecific hybridization techniques and advent of molecular markers for tagging and mobilization of useful genes, breeders evinced greater interest in use CWR for crop improvement.

The availability of total holdings of wild and weedy relatives of different crops as revealed by SINGER (http://singer.grinfo.net/) range from about 400 accessions (sorghum) to more than 5000 (wheat). The proportion of wild or weedy relatives in gene bank holdings has significantly increased in a span of 20 years starting from 1983 (Plucknett *et al*., 1987) to 2004 (http://singer.grinfo.net/). During this period, CWR representation increased from 0.0 to 4.95% in wheat, 0.5 to 7.08% in common bean, 0.001 to 5.27% in barley, and 0.4 to 4.97% in pigeonpea (Hodgkin and Hajjar, 2008). The increase in proportion of wild relatives in gene banks reflects the expectations of collectors and gene bank managers regarding the usefulness of wild relatives.

Summarizing the use of wild relatives for improvement of major crop species in the last 20 years, Hajjar and Hodgkin (2007) have listed the number of traits for whose improvement CWR was used in a particular crop. They showed that extent of utilization varies from crop to crop. Tomato takes lead with 55 traits followed by rice and potato with 12 traits each. Using CWR, wheat was improved for 9 traits and sunflower for 7 traits. Millet featured on the list with 3 traits and maize and chickpea with 2 traits each.

Impressive instances of commercial deployment genes from the wild species are now available in several crops. Gene introgression from synthetic wheats developed at CIMMYT using *Ae. tauschii* and *T. turgidum*, have resulted in cultivars having improved water-logging tolerance (Villareal *et al*., 2001) and disease resistance (Mujeeb-Kazi *et al*., 2001). Several wheat-alien translocations conferring rust resistance have been commercialized.

The classical example of successful commercial deployment of grassy stunt virus resistance from *Oryza nivara* in rice varieties grown on a global scale has already been discussed (Brar and Khush, 1997). In potato, resistance to late blight from *Solanum demissum* and *S. stoloniferum*, resistance to viruses from these species and from *S. chacoense* and *S. acaule*, and resistance to potato cyst nematode from *S. vernei* and *S. spegazinii* has been introgressed in several lines, whereas blight resistance in cultivar 'Biogold' was transferred from *S. bulbocastanum* (Bradshaw and Ramsey, 2005). Resistance to herbicides (imodazolinone and sulfonylurea) has been transferred from *Helianthus annuus* in sunflower hybrid cultivar 'Clearfield' (Seiler and Gulya, 2004). A chickpea variety 'BG 1103' having drought and high temperature tolerance derived from *C. reticulatum*. In barley, six cultivars having drought tolerance derived from *H. sponataneum* have been released by ICARDA (Hodgkin and Hajjar, 2008).

As far as heterosis breeding is concerned, CWR played a significant role by contributing sterile cytoplasm in different crops. In rice, CMS source derived from wild rice *O. sativa f. spontanea*, were used to produce hybrid cultivars in 1976 and presently about 45% area of rice acreage is under hybrid varieties in China. Similarly in other crops CMS sources have been derived from various wild relatives e.g. in sunflower from *H. annuus* and *H. petiolaris*, in wheat from *T. timopheevi* and in pigeonpea from *Cajanus cajanifolius* and *C. scarabaeoides*.

CWR are now well acknowledged as donors of enhanced yield potential and promising materials have been generated in several crops, particularly rice (Cheema *et al*., 2008). Earlier, yield enhancing QTLs affecting tillers and other traits were introgressed from *O. rufipogon* in rice (Xiao *et al*., 1998). Similarly in other crops yield QTLs derived from wild relatives have been reported, e.g. in tomato (Tanksley *et al*., 1996) and chickpea (Singh and Ocampo, 1997; Singh *et al*., 2005).

With respect to quality traits, a few instances of CWR utilization are reported. Improved protein quality (HMW) in durum wheat from related species, *T. dicoccum* and *T. dicoccoides*; double protein contents in cassava from *Manihot oligantha*; increased fruit size and soluble solids in tomato from wild species; increased amount of anti-cancer compounds in broccoli from *Brassica villosa*. The *T. dicoccoides* gene *GpcB1* conferring high grain protein content has found its way into several wheat cultivars (Brevis and Dubcovsky, 2010).

Some estimates of economic impact of genetic transfers from CWR are available (Frison and Attah-Krah, 2008). For example, traits incorporated from wild relatives into sunflower are worth USD 267-384 million annually to the sunflower industry in USA. A wild tomato accession has contributed 2.4% increase in TSS worth USD 250 million. Wild groundnut has contributed resistance to root knot nematodes that cost groundnut growers around the world approximately USD 1 billion annually. The utilization of CWR so far is indicative of the great potential that this category of PGR holds. The resource remains grossly underutilized and the above achievements represent no more than the proverbial tip of the iceberg. Systematic pre-breeding efforts would however be needed to harness the variation for crop improvement.

Illustrating Utilization of Plant Genetic Resources: PAU Wheat Improvement Programme as an Example

A broad genetic base, handled through a precise and fast breeding technology would be the hallmark of the new era breeding programmes. Wheat and rice improvement research at PAU has already made significant progress in this direction. New germplasm streams are being created accompanied by molecular marker interventions allowing their rapid channelization for commercial use. The diversifying input has come from well characterized sets of wild/related species of wheat (1500 acc.) and rice (2000 acc.). Using these donors several thousand introgression lines have been generated which are at various stages of utilization in the breeding programme. Salient research activities aimed at utilization of plant genetic resources, particularly the wild relatives in case of wheat are listed:

- A new major gene (*Yr40/Lr57*) for resistance to stripe as well as leaf rust was introgressed from *Ae. ovata* into bread wheat and stably translocated using *Ph* locus manipulation (Kuraparthy *et al*., 2007a). It is highly effective against prevalent races of both rusts and has been mobilized into high yielding backgrounds using molecular markers.
- A novel major gene for leaf rust resistance (*Lr58*) was introgressed from *Ae. triuncialis* (Kuraparthy *et al*., 2007b) and is being transferred to elite wheat lines using cytogenetic and molecular techniques.
- Potentially new gene (s) for resistance to both stripe and leaf rust has been incorporated from *Aegilops umbellulata* into elite wheat lines including PBW 343. It is in the process of being mapped and designated.

- QTLs for stripe rust resistance from diploid A genome species (*Q yrtm.pau.2A* from *Triticum monococcum*, and *Q yrtb.pau.5A* from *T. boeoticum*) have been tagged with molecular markers and transferred to relevant wheat cultivars. These adult plant resistance genes offer the prospect of durable resistance (Singh *et al*., 2007).
- *Aegilops kotschyi* and *Ae. tauschii* lines possessing high iron and zinc content in the grain were identified and are being used as donors (Chhuneja *et al*., 2006; Rawat *et al*., 2009).
- QTLs for higher iron and zinc content in the grain (*Q GFe.pau-2A*, *Q GFe.pau-7A* and *Q GZn.pau-7A*) have also been identified and transferred from *T. boeoticum*.
- QTLs for cereal cyst nematode resistance (*Q cre.pau-1A*, *Q cre.pau-2A*) have been transferred from *T. monococcum* (Singh *et al*., 2010).
- To improve processing quality in wheat, novel HMW glutenin subunits have been transferred from *T. urartu* and *T. diccocoides* to wheat variety PBW 343, resulting in improved sedimentation value.
- A major gene (*GpcB1*) for high protein content and enhanced micronutrient content, originally derived from *T. dicoccoides* has been transferred to a wide range of wheat genotypes using marker assisted selection (Pal, 2010).
- About 100 accessions of *Aegilops tauschii* have been characterized for cellular thermotolerance traits such as membrane thermostability and TTC cell viability and heat tolerant accessions identified (Gupta *et al*., 2010).
- *Aegilops speltoides* is being used for transfer of stay green habit to both tetraploid and hexaploid wheat.
- CMS lines based on different alien cytoplasms including *T. timopheevi* have been developed in wheat (Adugna *et al*., 2003). Restorer gene pool has been developed by using diversifying genetic input from synthetic hexaploid wheats.
- PAU collection of *Aegilops tauschii*, the D genome donor of wheat, was subjected to diversity analysis based on SSR markers and agromorphological traits (Chhuneja *et al*., 2010).
- An efficient hybridization and trait transfer protocol has been designed for introgression from *Aegilops tauschii* to bread wheat (Sehgal *et al*., 2010). The method is based on direct hybridization (bridging species not involved) and is being used for transfer of components of heat tolerance.

The precision and speed imparted by marker assisted gene tagging and transfer is being used to offset some of the difficulties associated with use of wild/weedy and un-adapted donor germplasm. Complementary research efforts in the Department of Plant Breeding and Genetics and School of Agricultural Biotechnology and the excellent context for wheat improvement provided by the dynamic and responsive farming community of Punjab state has helped orient this programme to real needs.

In the face of challenges ahead, it is thus imperative to strengthen PGR utilization efforts. Future needs of crop improvement in terms of biotic and abiotic stresses, and sometimes even for consumer preference, cannot be pre-judged completely. A broad genetic base is our safeguard against all such exigencies. The diversity also ensures continued genetic gains. It is the key to sustainable crop improvement.

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