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Abstract—Increasing the grain yield and its stability in dry lands of the semi-arid tropics of Asia and Africa, recognized as the home to a vast impoverished humanity, continues to be a major challenge for agricultural research and development agencies. Development of genetically enhanced cultivars with high yield potential, appropriate maturity, and resistance to biotic and abiotic stress factors is an effective approach to increasing and stabilizing the production in these regions. Advances possible through this approach are illustrated with the impacts made in pearl millet (*Pennisetum glaucum*), a warm-season cereal; and chickpea (*Cicer arietinum*), a cool-season legume, which are amongst the five mandate crops of the International Crops Research Institute for the Semi-arid Tropics (ICRISAT). The research presented in this paper is largely the work done by ICRISAT in partnership with the national agricultural research systems and advanced research institutes. The conventional approach of genetic enhancement both in pearl millet and chickpea has been highly successful. Progress has also been made in the areas of molecular marker-assisted selection in both crops, and in wide hybridization and transgenic technology in chickpea to improve those traits that are less amenable to genetic manipulation through conventional methods of genetic enhancement.

Key words: dry land, semi-arid tropics, pearl millet, chickpea, genetic enhancement

1. Introduction

_Semi-arid_tropical_regions_of_the_world_are_home_to_about_850_million_people, largely_dependent_on_ agriculture as the major source of their livelihood. The crop production environments of these regions are characterized by low soil fertility, low and erratic rainfall, high temperatures, and negligible use of external inputs--all leading to low crop productivity. Improved management of water, soil and nutrients would play a major role in achieving increased and stable production in these regions. Improved crop cultivars can also play a significant role in this direction (Serraj et al., 2003). For enhanced and stable crop production in the dry areas of the semi-arid tropics, the three key elements of seed-based solutions are the choice of crops adapted to these environments, development of improved cultivars of these crops with higher grain yield and improved resistance to biotic and abiotic stresses, and efficient seed production and distribution mechanisms. The International Crops Research Institute for the Semi-arid Tropics (ICRISAT) has a mandate for genetic improvement of some of the most drought tolerant crops that include sorghum [Sorghum bicolor (L.) Moench] and pearl millet [Pennisetum glaucum (L.) R. Br.] as cereals; pigeonpea [Cajanus cajan.) Millsp.] and chickpea [Cicer arietinum L.] as pulses; and groundnut [Arachis hypogaea L.] as an oilseed crop. Significant progress has been made in developing improved cultivars of these crops that perform better than the traditional ones due to a combination of factors that includes improved yield potential, early maturity, and improved resistance to biotic and abiotic stresses. This paper deals with past achievements and future prospects by illustrating the work on two contrasting crops---pearl millet (a warm season cereal) and chickpea (a cool season legume) using both the conventional and biotechnological research approaches that have been followed at ICRISAT for genetic enhancement and cultivar development.

2. Conventional Breeding Approaches

2.1 Selection for Grain Yield and Adaptation: Genetic advances from direct selection for grain yield in drought-prone environments result from genetic changes in yield potential, drought tolerance, and appropriate maturity that helps escape drought. Resistance to target pests and diseases is considered an integral part of this breeding strategy. This breeding strategy has proved successful both in pearl millet and chickpea. For instance, ICTP 8203, an open-pollinated variety (OPV) of pearl millet was developed at ICRISAT-Patancheru from progenies derived from an *iniari* (early-maturing) landrace from northern Togo that had been selected for high grain yield, acceptable agronomic traits (shorter plant height, early maturity, compact panicles, good exsertion, and no tip sterility) and high levels of resistance to the most dreaded pearl millet disease-downy mildew (DM) caused by *Sclerospora graminicola* (Sacc.) Schroet. (Rai et al.,1990). This variety, released in 1988 specifically for cultivation in Maharshtra state of India, was rapidly adopted, covering about 0.8 million ha (about 50%) of the

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pearl millet area in this state at the peak of its popularity in 1995 (Fig.1). Even in the face of stiff competition with hybrids, ICTP 8203 was estimated to be cultivated on 0.4-0.5 million ha in Maharashtra in 1999, and it still occupies about 0.3 million ha, mostly in very dry areas with low soil fertility. ICRISAT-Patancheru developed ICMV 88908, another OPV of *iniari* background that was morphologically similar to ICTP 8203 but had about 10% grain yield advantage over ICTP 8203. This variety was released as Okashana 1 in Namibia where it rapidly spread, increasing from about 1% of the total pearl millet area in 1990 to 49% in 1996 (Rohrbach et al., 1999). Okashana 1 outyields locals by 49% in fertilized field conditions (20 kg N and 15 kg P ha⁻¹) under low and late rainfall, and by 87% under the similar fertilized fields when rainfall is moderate or timely.

The development of hybrid HHB 67 that matures in <65 days remains the greatest landmark in breeding early-maturing pearl millet cultivars. Researchers from CCS Haryana Agricultural University (CCSHAU) observed that this hybrid, selected for high grain yield in a short growing season, gives 12-15% less grain yield than the fullseason hybrids maturing in 75-80 days (Table 1). However, when sowing is delayed due to late arrival of monsoon rains, HHB 67 gives 45-72%

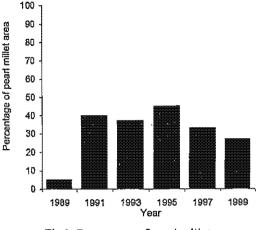


Fig.1: Percentage of pearl millet area

Table 1. Effect of sowing time on grain yield

Pearl	Grain yield		
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Hybrid	Normal sowing	Late sowing	maturity
HHB 50	3.09	0.93	79
HHB 60	3.21	1.10	75
<u>HHB 67</u>	2.72	1.60	61

more grain yield than the full-season hybrids and permits farmers to clear their fields in time for the subsequent winter season crops such as chickpea and mustard [*Brassica juncea* L.]. In the drier pearl millet zones, early maturity of HHB 67 allows it to escape terminal drought stress that will reduce yields of the full-season cultivars. HHB 67 has gained immense popularity with farmers and is currently estimated to be cultivated on more than 0.4 million ha in some of the driest areas of Haryana and Rajasthan.

Breeding for high grain yield combined with earliness and disease resistance has been equally successful in chickpea. For instance, cultivation of kabuli (large-seeded) chickpeas has normally been confined to cooler areas with longer growing season. ICRISAT developed an extra-early-maturing kabuli variety, ICCV 2, that flowers in 25 days and matures in 85 days at Patancheru (18°N), India. It combines higher yield potential with earliness and wilt resistance, thus making it adaptable to tropical environments. In the All India Coordinated trials conducted across 13 year × location environments in the southern zone, ICCV 2 gave 25% more seed yield than the control variety L 550, matured 18 days earlier, had 25% larger seed size and 12% wilt incidence against Patancheru isolate compared to 43% in L 550 (Table 2). ICCV 2, first released in India in 1989 as Swetha, has wide adaptation as evidenced by its good performance compared to local varieties and its consequent release as Wad Hamid in Sudan in 1998 and as Yezin 3 in Myanmar in 2000. A survey in 2003 showed that ICCV 2 had replaced 50% of the chickpea area in Myanmar within 3 years of its release. The success of ICCV 2 has led to the development of several early-maturing and large-seeded kabuli varieties such as ICCV 3, KAK 2, JGK 1, and Vihar in India. A large number of early-maturing desi (small-seeded) chickpea varieties have also been released in India. These early-maturing desi and kabuli chickpea varieties have stimulated the expansion of chickpea area in southern India, with the total chickpea area in the two southern states (Andhra Pradesh and Karnataka) increasing from 189 000 ha to 532 000 ha during the past two decades. Early-maturing chickpea varieties have also made significant impacts in Maharashtra, Madhya Pradesh and Gujarat states of India, and also led to significant spillover impacts in Australia and Canada.

2.2 Trait-based Selection for Drought Tolerance: Morpho-physiological traits have also been used to select lines adapted to droughty environments, although the progress has been slow compared to selection for earliness (permits drought escape) and yield *per se*. A pearl millet study evaluated the effectiveness of panicle harvest index (PNHI = grain mass/panicle mass) as a selection criterion in a bi-directional selection program for combining ability for high and low PNHI in restorer and maintainer lines (Bidinger et al., 2000). In both selection experiments, the difference between the high and low PNHI selections for combining ability for PNHI and grain yield was generally small (1% for PNHI and 2% for grain yield) and statistically non-significant when tested in the irrigated (non-stress) environment, but generally significant and of greater magnitude (5-8% for PNHI and 9-13% for grain yield) when tested under the managed stress environments. These results showed that

Table 2. Mean performance of kabuli chickpea varieties in southern zone of India.							
Variety	Yield (t ha'')	Days to maturity	100-seed mass (g)	Wilt resistance (% mortality)			
ICCV 2	1.45	88	25.0	12			
KAK 2	1.46	91	37.6	10			
L 550	1.15	106	20.8	43			

selection for combining ability for PNHI under terminal stress had little effect on the combining ability of elite parental lines under non-stress conditions, but it resulted in a significant difference in their combining ability for both PNHI itself and for grain yield under terminal stress. Application of this selection criterion led to the development of an open-pollinated variety ICMV 88904 (released as ICMV 221 in India) that outyielded the popular open-pollinated variety ICTP 8203 by 15% across 21 trials in nine locations (Witcombe et al., 1997). ICMV 221 has wider adaptation. Based on its superior performance for grain yield, especially under drought conditions, and downy mildew resistance, this variety was later released as KAT/PM 3 in Kenya and as *Kona* in Eritrea, both in eastern Africa.

In chickpea, large root mass was found to be involved in drought avoidance in ICC 4958, while smaller leaf area was the most important drought tolerance trait in ICC 10448 and ICC 5680. Smaller leaf area in ICC 10448 is due to narrow pinnules, and that in ICC 5680 it is due to fewer pinnules (Saxena, 2003). The larger root system of ICC 4958 was found to be effective in rapid and greater amount of soil water extraction during the early growth period of the crop coinciding with flowering and early seed filling stages of the crop (Krishnamurthy et al., 1996). The smaller leaf surface area resulting from fewer pinnules in ICC 5680 effectively reduced transpirational water loss by 30% compared to ICC 4958 (Saxena, 2003). Efforts to combine large root traits with smaller leaf area have produced several lines (ICCV 98901 to 98907) with large roots and fewer pinnules derived from cross ICC 4958 × ICC 5680 that show greater degree of drought avoidance (Saxena, 2003).

3. Biotechnological Approaches

3.1 Molecular Marker-assisted Selection: Molecular marker-assisted selection is particularly useful for traits that are more intractable and have low heritability (e.g., drought tolerance in pearl millet and chickpea), or for which heritability may be moderate to high but for which relevant selection environments may often be difficult to create and reliably reproduce (DM of pearl millet). This technology is best suited to identify and locate different genes related to a trait and guide their pyramiding to enhance the level of character expression. Molecular markers for more than 60 quantitative trait loci (QTL) affecting DM resistance in pearl millet have been identified. Some of these have been used in applied breeding programs. For instance, realizing the vulnerability of single-cross hybrid HHB 67 to DM, a marker-assisted backcross breeding succeeded in developing resistant versions of this hybrid and its parental lines that have grain yield and flowering comparable to HHB 67, but greatly enhanced DM resistance for the Jodhpur pathotype (Table 3). This breeding tool is now being successfully used at ICRISAT to pyramid DM resistance genes into parental lines of some of the other widely grown commercial hybrids. Molecular markers for terminal drought tolerance have been identified and marker-assisted breeding for drought tolerance in pearl molecular markers for terminal drought tolerance have been identified and marker-assisted breeding for drought tolerance in pearl millet has just begun.

Significant progress has been made in identifying molecular markers for *Fusarium* wilt resistance, and research is underway to identify such markers for root mass and pod borer (*Helicoverpa armigera* Hubner) resistance in chickpea. Molecular marker studies have helped develop a better understanding of genes for *Fusarium* wilt resistance. For instance, it has been found that one of the three genes for resistance to race 1, one of the two genes for resistance to race 4, and the gene for resistance to race 5 are located in the same linkage group, while the gene for resistance to race 0 is not linked to these genes (Ratnaparkhe et al., 1998; Tekeoglu et al., 2000). ICRISAT has developed 257 recombinant inbred lines (RILs) from a cross involving ICC 4958, the drought avoidant line with high root volume and Annigeri, a highly adapted variety with low root volume. These RILs have been extensively phenotyped for root traits and are now being genotyped with molecular markers to map QTL for root traits. RILs from one cross have been phenotyped for *Helicoverpa* resistance under unsprayed field conditions, where the leaf damage ratings have varied from 2.0 to 7.5 (Sharma et al., 2003).

Table 3. Performance of DM resistant versions of hvbrid HHB 67 and their parental lines

Hybrid	Pedigree $(P_1 \times P_2)_{-}$	Hybrid p	DM incidence (%)			
	_	Grain yield (t ha ⁻¹)	50% flower (days)	Hybrid	P ₁	Р
ICMH 01107	01027A × ICMR 0100	1 2.6	39	4	24	54
ICMH 01129	01027A × ICMR 01004	4 2.3	40	5	24	32
HHB 67	843A × H 77/833-2	1.9	41	79	93	98
LSD (P = 0.05)	0.76	2.5	-		20-20

3.2 Tissue Culture and Wide Hybridization: Wild species serve as excellent sources of germplasm for resistance to biotic and abiotic stresses. Amongst the problems most serious in dry areas where the use of wild germplasm can have a major impact on production is *Helicoverpa* resistance in chickpea as the resistance levels in landraces, breeding lines and varieties appear to be low compared to those in the wild species. Results of a preliminary screening under natural infestation at Manali in Kullu district of Himachal Pradesh, India, showed that perennial wild species like *C. microphyllum* and *C. canariense* had pod borer damage rating as low as 1, while *C. judaicum*, reported earlier as a source of resistance, had a damage rating of 4, and cultivated chickpea genotypes had high leaf and pod damage ratings of 8.5 and 9 (>70% leaf and/or pod damage). Thus, these two perennial species of *Cicer* have been developed and successfully used (Mallikarjuna, 1999). The major challenge now is to develop hybridization and tissue culture protocols to successfully transfer the resistance genes from these perennial species to cultivated chickpeas.

3.3 Transgenic Technology: The major thrust of this technology at ICRISAT is on legumes, including chickpea, for which efficient tissue culture regeneration and genetic transformation protocols have been developed (Jayanand et al., 2003). Successes have been achieved in developing chickpea plants with resistance to *Helicoverpa* by using genes derived from the bacterium *Bacillus thuringiensis* (Bt cry1Ab) and soybean trypsin inhibitor (SBTI). While the molecular characterization and insect bioassays are ongoing, the effectiveness of alternative sources of insecticidal genes, including those derived from B. *thuringiensis* (Bt), is currently being evaluated at ICRISAT in collaboration with research partners from CIRAD, France. The first field trial of putative *Helicoverpa* resistant transgenic chickpea plants is likely to be in 2005. ICRISAT is also involved in developing transgenic chickpeas for tolerance to abiotic stresses such as drought and low temperatures. This is being attempted by using the *P5CSF* gene from a *Vigna* species that results in overproduction of proline and reduction of free radicals. In another approach, the regulatory sequences from the Drought Responsive Elements (DRE) of *Arabidopsis thaliana* are being introduced into chickpea that are presumed to induce native gene expression related to ABA-independent stress response. To make this approach effective, the *DREBIA* cDNA is being driven by a stress-responsive promoter *rd29A*.

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