Proceedings of the Fourth International Food Legumes Research Conference (IFLRC-IV), October 18–22, 2005, New Delhi, India
M. C. Kharkwal (ed.)

Hybrid breeding in grain legumes: A success story of pigeonpea

K. B. SAXENA[1*], R. V. KUMAR[1], V. A. DALVI[1], NALINI MALLIKARJUNA[1], C. L. L. GOWDA[1], B. B. SINGH[2], S. B. S. TIKKA[3], K. B. WANJARI[4], L. B. PANDEY[5], R. M. PARALKAR[6], M. K. PATEL[7], BAO SHIYING[8] AND ZONG XUXIAO[9]

[1]*International Crops Research Institute for Semi Arid Tropics (ICRISAT), Patancheru P.O., 502 324, A.P., India*
[2]*Indian Institute of Pulses Research, (IIPR), Kanpur 208 024, India*
[3]*Gujarat Agricultural University (GAU), SK Nagar 385 506, India*
[4]*Panjabrao Deshmukh Krishi Vidyapeeth (PDKV), Akola 444 104, India*
[5]*MAHYCO, Jalna 431 203, India*
[6]*Ankur Seeds, Nagpur 440 018, India*
[7]*JK Seeds, Hyderabad 500 016, India*
[8]*Yunnan Academy of Agricultural Sciences (YAAS), Kunming, Yunnan 650 205, China*
[9]*Chinese Academy of Agricultural Sciences (CAAS), 12 Zong Guan Cun Nan De Jie, Haidan Distt. Beizing 100 081, China*
**Presenting author: k.saxena@cgiar.org*

Abstract

The nature and action of genes determining yield in cereals and pulses are more or less comparable. But the advances made in improving the yield levels through breeding in cereals are much higher than those of the legumes. Apart from other physiological differences many cereals and legumes, this differential yield advances may be attributed to commercial exploitation of hybrid vigour in the former. Although floral biology of some of the legumes permits limited out-crossing hybrid vigour for yield has not been exploited for commercial use in any other legume with the exception of pigeonpea. Recently, significant progress has been made to develop hybrid pigeonpea technology. This technology is based on cytoplasmic-genic male-sterility systems, developed through crosses between cultivated types with its wild relatives. Some of the hybrids have recorded up to 77% standard heterosis for grain yield. The technology for developing their maintainers and restoration of their fertility, and

hybrid seed production has also been perfected. This paper discusses the scope, challenges, achievements, and prospects for hybrid pigeonpea.

Introduction

Majority of protein needs of people in the developing countries is derived from legumes. Most legumes are grown invariably under unfavourable and risk prone environments resulting in low and unstable productivity. For example in India pulses (food legumes, excluding oil and fodder legume crops) are grown on about 23 m ha but their production (13.0 mt) and mean productivity (600 kg ha^{-1}) are low. To enhance the productivity of pulses, Indian Council of Agricultural Research (ICAR) initiated a multidisciplinary pulses improvement project in 1967 and so far over 200 varieties were released. Although the introduction of these varieties helped in increasing the cropped area but their productivity has remained unchanged around 110 kg ha^{-1} moth bean (*Vigna aconitifolia*) to 1000 kg ha^{-1} field pea (*Pisum sativum*). The non-conventional breeding approaches such as exploitation of hybrid vigour is warranted to break the yield barrier. The quantum jump in yield potential observed in some cereals and vegetable crops in the past was primarily due to exploitation of 'hybrid vigour' or 'heterosis'. In pulses, however, this breeding approach could not be used due to their high self-pollinating nature and inability to produce hybrid seed economically. Faba bean (*Vicia faba*) and pigeonpea [*Cajanus cajan* (L.) Millsp.] are the two pulses where a reasonable level of natural out-crossing occurs and this trait could be used in developing hybrid cultivars. Recently, a breakthrough has been achieved in developing hybrid technology in pigeonpea. The problems and prospects of hybrid breeding are reviewed in this paper. Globally, pigeonpea is cultivated on over 5 m ha and about 85% of area is in India. In India the dehulled split cotyledons are cooked to make a thick soup (*dhal*) for eating with bread (*chapati*) and rice, while in Africa and Central America, whole dry seeds and immature seeds are used as vegetable. The broken seeds, husk, and pod wall are also fed to cattle, while the dry stems are an important household fuel wood.

Pre-requisites for Commercial Hybrid Breeding

Favourable Gene Action

The phenomenon of heterosis has been exploited in cross-pollinated crops, especially for developing open-pollinated synthetics, composites, and/or hybrid varieties. Since dominant genes in the population have evolutionary advantage, the heterosis was initially considered a discernible phenomenon of the cross-pollinated crops but later the commercial exploitation of hybrid vigour in cereal and vegetable crops established its utility in the self-pollinating crops also. It was found that since over-dominance and dominance gene actions are not common for yield in both self as well as cross-pollinated crops, the additive gene action and the additive x additive interallelic and intergenomic interactions play an important role in the expression of hybrid vigour [Sharma and Dwivedi, 1995]. The pollination system of a crop, therefore, poses no restriction in the manifestation of heterosis. In pigeonpea, both additive and non-additive gene actions for yield and yield components have been reported [Saxena and Sharma, 1990] and it offers scope for hybrid breeding.

Presence of Heterosis

The first report of hybrid vigour in pigeonpea was published in 1957 by Solomon *et al.* For grain yield they reported 24.5% heterosis over the better parent. In the heterotic crosses the hybrid vigour for yield was closely associated with hybrid vigour for number of pods per plant, number of primary branches, and plant height. Subsequently, a number of reports have been published on hybrid vigour for yield and yield components (Saxena and Sharma, 1990) in pigeonpea.

Effective Cross-Pollination System

For large-scale economical seed production of hybrids and their parents, it is essential to have an effective and inexpensive mass pollen transfer mechanism. The floral biology of pigeonpea permits partial cross-pollination caused by a number of insect species. Of these, *Magachile* spp. and *Apis mellifera* are the main pollinators. A combination of factors such as number of pollinating insects, number of flowers, location of field, temperature, humidity, and wind velocity determine the extent of cross-pollination at a particular location. Howard *et al.* were the first to report natural out-crossing in pigeonpea in 1919 and subsequently, a number of reports have appeared in literature (Saxena *et al.*, 1990).

Availability of Stable Male-Sterility Systems

High level of male-sterility in the female parent in conjunction with natural out-crossing will make the hybrid seed production easy and affordable. Since no stable male-sterility system was available in pigeonpea, a deliberate search for a male-sterility was made in the germplasm, leading to the identification of male-sterile plants in ICP 1596. In this accession the male-sterility was associated with translucent anthers and it was controlled by a single recessive gene, ms_1 (Reddy *et al.*, 1978). Later, another source of male-sterility, characterized by brown anthers and controlled by non-allelic single recessive gene ms_2, was also reported [Saxena *et al.*, 1983]. Both these male-steriles have prominent anther morphology which helps in their easy identification in field before anthesis. In hybrid breeding, the cytoplasmic-genic male-sterility (CGMS) is known to be more efficient than genetic male-sterility (GMS) system. So far four wild relatives of pigeonpea have been successfully used in breeding CGMS lines. These are (i) A_1 cytoplasm, derived from *Cajanus sericeus* (Saxena *et al.*, 1997); (ii) A_2 cytoplasm, derived from *C. scarabaeoides* (Tikka *et al.*, 1997, Saxena and Kumar, 2003); (iii) A_3 cytoplasm, derived from *C. volubilis* (Wanjari *et al.*, 2001); and (iv) A_4 cytoplasm, derived from *C. cajanifolius* (Saxena *et al.*, 2005a). The instability in the expression of male-sterility and their fertility restoration has, however, limited the use of A_1, A_2 and A_3 cytoplasms in large-scale practical hybrid breeding programmes (Saxena *et al.*, 2005b). However, the CGMS developed with A_4 cytoplasm (shown in Table 1) is considered the best source for commercial exploitation.

Fertility Restoration of Male-Steriles

Fertility restoration is a vital component of CGMS-based hybrid technology. At ICRISAT, during the past three years over 300 germplasm of diverse origin have been crossed to study the fertility restoration of different CGMS systems. The observations on F_1 hybrids indicated

Table 1. Male-sterility in F_1 of cross ICPW 29 (*C. cajanifolius*) x ICP 11501 (*C. cajan*) and backcross generations

Generation	Total plants	Male-sterile plants
F_1	12	12*
BC_1F_1	8	8
BC_2F_1	5	4+1*
BC_3F_1	165	165
BC_4F_1	7	7
BC_5F_1	67	67
BC_6F_1	1133	1133
BC_7F_1	17186	17186
BC_8F_1	19856	19856

*Partial male-fertile

that (i) although the fertility restorers were available but their frequency was low, (ii) many genotypes were heterogeneous for fertility restoration gene(s), (iii) some genotypes produced hybrids with partial fertility restoration, and (iv) the restoration was influenced by environment in some crosses. At ICRISAT the fertility restoration was found to be the best in hybrid combinations using A_4 cytoplasm and the F_1 plants compared well with that of control cultivars for pollen fertility. The special feature of the fertility restoration of this group is that the quantity of pollen produced in almost all the F_1 plants matched well with that of control. In contrast, the hybrids derived from the A_1 and A_2 cytoplasms were shy bearers and produced relatively less pollen.

Heterosis for Yield

GMS-Based Hybrids

Initially in the absence of CGMS, the GMS sources were used for developing hybrids. The world's first pigeonpea hybrid ICPH 8 was released for cultivation in 1991. The release of ICPH 8 is considered a milestone in the history since no commercial hybrid is available in pulses. Subsequently, concerted efforts were made to identify new hybrids. Over 10,000 hybrid combinations were evaluated. Of these, 182 F_1 hybrids exhibited more than 20% standard heterosis (Table 2) and some more hybrids were released.

Table 2. Number of hybrids showing significant heterotic advantage in multilocation trials

Year	Standard heterosis (%)					
	20-40	41-60	61-80	81-100	>100	Total
1990-91	18	21	09	07	11	66
1991-92	07	06	02	02	01	18
1992-93	08	04	02	05	01	20
1993-94	18	05	04	05	03	35
1994-95	16	06	-	-	-	22
1995-96	07	03	02	-	-	12
1996-97	03	01	02	03	-	09
Total	**77**	**46**	**21**	**22**	**16**	**182**

In 1993, Punjab Agricultural University (PAU), Ludhiana, identified hybrid PPH 4. In the multi-location trials this hybrid out-yielded the control variety by a margin of 47.4%. Later, another hybrid CoH 1 was released by Tamil Nadu Agricultural University (TNAU), Coimbatore. This hybrid recorded 32% higher yield over control. In 1997, TNAU released another hybrid CoH 2, which out-yielded the control by 35%. Two pigeonpea hybrids, AKPH 4104 and AKPH 2022 were released by Punjabrao Deshmukh Krishi Vidyapeeth (PDKV), Akola. These hybrids also recorded 35-64% superiority over the controls.

CGMS-Based Hybrids

In the early maturing group, all the hybrids evaluated at Patancheru in 2004 produced greater seed yield than control (Table 3). The best hybrid ICPH 2470 matured in 125 days and produced 3205 kg ha^{-1} seed yield with 77.5% advantage over the control. The estimates of productivity per unit of time indicated that ICPH 2470 produced 25.6 kg ha^{-1} day^{-1} grain as compared to 14.1 kg ha^{-1} day^{-1} by the control. In the multilocation medium (Table 4 and 5) and long-duration (Table 6) trials also the hybrids recorded significant superiority over the controls.

Table 3. Performance of promising early-maturing hybrids at Patancheru, 2004

Entry	Days to mature	Seeds pod^{-1}	100-seed mass (g)	Grain yield (kg ha^{-1})	Heterosis over control (%)
ICPH 2470	125	3.8	8.9	3205	77.5
ICPH 2438	122	3.8	9.0	2404	33.1
ICPH 2429	128	3.8	9.4	2351	30.2
ICPH 2431	122	3.4	8.3	2195	21.5
UPAS 120 (control)	128	3.4	9.1	1806	-
SEm	± 3.3	± 0.14	± 0.23	± 171.5	-
CV (%)	3.6	5.63	3.48	10.8	-

Table 4. Yield of some medium maturing hybrids in the coordinated trials of ICAR (mean of four locations)

Entry	Yield (kg ha^{-1})	Superiority (%)
SKNPCH 10	3440	56
SKNPCH 1	3380	54
SKNPCH 5	3206	46
NDPH8	3333	67
NDPCH 33	2986	49

Source: IIPR Report

Other Advantages of Hybrids

Besides high yield, the hybrids have been found to have over 40% greater shoot and root mass and perhaps this ability provides greater stability in the performance of hybrids even under diverse stress-prone environments (Saxena *et al.*, 2005b).

Table 5. Yield of ICPH 2671 at different locations during 2005 and 2006 rainy seasons

Year	Location	Fertility restoration(%)	ICPH 2671	Maruti (control)	SEm	CV (%)	% heterosis
2005	Medchal	100	2996	1041	+331.6	21	188
	Coimbatore	98	4262	2538	252.7	17	71
	Patancheru	100	2671	1677	207.7	14	59
	Jalna	100	3416	2541	NR	7	34
2006	Parbhani	98	4779	2648	173.4	6	80
	Jalna	100	1948	1092	91.8	10	78
	Coimbatore	100	1823	1100	324.7	28	66
	Phaltan	-	3208	2243	147.3	20	43
	Patancheru	100	2660	1919	140.7	8	39
	Bangalore	-	1603	1462	289.9	20	10
	Mean	-	**2937**	**1826**	-	-	**61**

Table 6. Performance of some late maturing hybrids developed and evaluated by MAHYCO, 2004

Hybrid no.	Maturity (days)	Yield kg ha^{-1}	Superiority (%)	Yield kg ha^{-1} day^{-1}	Superiority (%)
MRT-9801	245	3777	49	15.4	44
MRT-9802	243	3465	37	14.3	33
BAHAR (control)	237	2535	-	10.7	-
LSD @ 5%	2.5	327	-	-	-
CV (%)	6.0	16.7	-	-	-

Seed Production Technology

An efficient seed production system that could provide quality seeds at economically viable costs is the lifeline of any hybrid-breeding programme. Since out-crossing in pigeonpea is affected by insects, a safe isolation distance is essential to produce quality seed of the parents and hybrids. So far, in pigeonpea no isolation distance study has been conducted using male-steriles but it is assumed that the information generated on isolation specifications of inbred cultivars could be utilized in producing seeds of pigeonpea hybrids and their parents. At ICRISAT isolation distance of over 300 m was found to be safe to produce breeders' seed.

Tests at ICRISAT also indicated that full pod set on the male-sterile plants is obtained if one pollinator row is sown after every six male-sterile rows. This row ratio may have to be changed if the recommended 6:1 ratio is not optimal due to insufficient insect pollination. The hybrid seed production technology and crop management practices are critical in determining the seed production costs. A large variation in the production costs of male-sterile and hybrid seeds was observed at PAU. In 1990, 275 kg ha^{-1} seed of a male-sterile line was produced at a cost of Rs. 39.4 kg^{-1}. Experiments at ICRISAT demonstrated that adoption of multiple harvest system can reduce the hybrid production costs significantly, particularly in early maturing materials.

Some Issues in Realizing the Reality of Pigeonpea Hybrids

The released GMS-based hybrids have exhibited a considerable heterosis for yield. This was perhaps due to the fact that both the parents invariably were agronomically superior. In CGMS based hybrids, however, most of the R-lines are derived from the germplasm and the F_1s exhibit variability and it might limit the breeding value. A successful CGMS hybrid breeding programme in the long run should involve research in the following areas:

- *Development of Diverse A-lines*: This is somewhat easy, because well-adapted agronomically superior genotypes with high general combining ability can be crossed; and if the F_1 hybrids are male-sterile then the genotype of the pollen parent is reconstituted by successive backcrossing.
- *Identification of Stable Fertility Restorers*: We need to cross a large number of genotypes with the CGMS lines, test F_1s in diverse environments and identify stable fertility restorers.
- *Breeding of Agronomically Superior R-Lines*: This can be done either by backcrossing agronomically superior genotypes to the source R-line or by inter-mating R-lines and recovering new superior R-lines through pure line breeding.
- *Synthesis and Evaluation of Hybrids*: To select high performing specific crosses for wide adaptation a number of test crosses should be made and evaluated for their performance in multilocation trials.
- *Grow-Out Test for Hybridity*: This is an important component of commercial hybrid breeding programmes and genetic and/or molecular approaches need to be persued.

It may also be realized that the success of any hybrid breeding programme will depend a great deal on the strength of conventional pure line breeding programme. It is likely that at the first instance one may find reasonable good combinations in a limited number of crosses but over the period of time the frequency of such crosses will decline. The development of CGMS in pigeonpea is a path breaking achievement but a lot more efforts are needed to make pigeonpea hybrid varieties a commercial reality. In recent years, pigeonpea production in India has recorded a significant growth rate and it is primarily due to the development of early maturing and disease resistant varieties. The early varieties have found new niches such as crop rotation with winter crops. Similarly, the newly developed disease resistant varieties have replaced the susceptible landraces and cultivars. But these achievements are insufficient to meet the ever-growing global demand for pigeonpea. Since the scope for horizontal expansion of area is limited, the attention of researchers needs to be focused on increasing the productivity.

In the recent past significant achievements in hybrid pigeonpea breeding have been recorded. A comparison of pigeonpea with other crops such as maize, cotton, rice, millet and sorghum where commercial hybrids are in cultivation shows (Table 7) that the magnitude of realized heterosis for yield in pigeonpea is more or less similar to those of other crops.

Table 7. Standard heterosis reported in various field crops in India

Crop	Range of heterosis (%)
Maize	15.0-51.0
Pearl millet	18.9-53.7
Sorghum	60.0-80.0
Cotton	05.3-28.4
Rice	16.2-44.2
Pigeonpea	20.0-47.4

The experience with QMS based hybrid technology has demonstrated that in pigeonpea the exploitation of hybrid vigour is feasible. With the CGMS being available, the commercial hybrids are a near reality now. Considering the suitability of hybrids in the stress-prone environments, we feel that the hybrids will adopt well in the 'very difficult to cultivate' such as the hilly areas of southern China. In summary, the recent developments in hybrid pigeonpea breeding technology are very encouraging and this technology is here to stay and soon the farmers will reap its benefits.

References

Howard, A., Howard G.L.C. and Khan, A. R. 1919. Studying the pollination of Indian crops. I. Memoirs, Dept Agric. (Botanical series) 10: 195-200.

Reddy, B.V.S., Green, J.M. and Bisen, S.S. 1978. Genetic male-sterility in pigeonpea. Crop Sci., 18: 362-364.

Saxena, K. B., Wallis, E. S. and Byth, D. E. 1983. A new gene for male-sterility in pigeonpea [*Cajanus cajan* (L.) Millsp.]. Heredity, 51: 419-421.

Saxena, K. B. and Sharma, D. 1990. Pigeonpea Genetics. p. 137-158 *In* Nene Y. L., Hall S. D. and Sheila V. K. (ed.). The Pigeonpea, CAB International, Wallingford, U.K.

Saxena, K. B., Singh, L. and Gupta, M. D. 1990. Variation for natural out-crossing in pigeonpea. Euphytica 39: 143-148.

Saxena, K.B., Kumar, R. V., Singh, L. and Raina, R. 1997. Development of a cytoplasmic-nuclear male-sterility system in pigeonpea. p. 19. Progress Report Fourth Consultative Group Meeting on Cytoplasmic Male-Sterility in Pigeonpea March 3-4, 1997. Bhaba Atomic Research Centre, Trombay, India.

Saxena, K. B. and Kumar, R. V. 2003. Development of a cytoplasmic-nuclear male-sterility system in pigeonpea using *C. scarabaeoides* (L.) Thours. Indian J. Genet., 63: 225-229.

Saxena, K.B., Kumar, R. V., Srivastava, N. and Shying, B. 2005a. A cytoplasmic-nuclear male-sterility system derived from a cross between *Cajanus cajanifolius* and *C. Cajan.* Euphytica, 145: 291-296.

Saxena, K.B., Srivastava, D.P., Chauhan, Y.S. and Masood Ali. 2005b. Hybrid Pigeonpea. p. 96-133. *In* Masood Ali and Shiv Kumar (ed.), Advances in Pigeonpea Research, Indian Institute of Pulses Research, Kanpur, India.

Sharma, D. and Dwivedi, S. 1995. Heterosis in grain legume crops - Scope and use. p. 960-979 *In* Genetic Research and Education: Current Trends and the Next Fifty Years, Indian Society of Genetics and Plant Breeding, New Delhi, India.

Solomon, S., Argikar, G. P., Solanki, M. S. and Morbad, I. R. 1957. A study of heterosis in *Cajanus cajan* (L.) Millsp. Indian J. Genet., 17: 90-95.

Tikka, S. B. S., Parmar, L. D. and Chauhan, R. M. 1997. First record of cytoplasmic-genic male-sterility system in pigeonpea [*Cajanus cajan* (L.) Millsp.] through wide hybridization. GAU Res. J., 22: 160-162.

Wanjari, K. B., Patil, A. N., Manapure, P., Manjayya, J. G. and Manish, P. 2001. Cytoplasmic male-sterility in pigeonpea with cytoplasm from *Cajanus volubilis.* Ann. Plant Phyol., 13: 170-174.