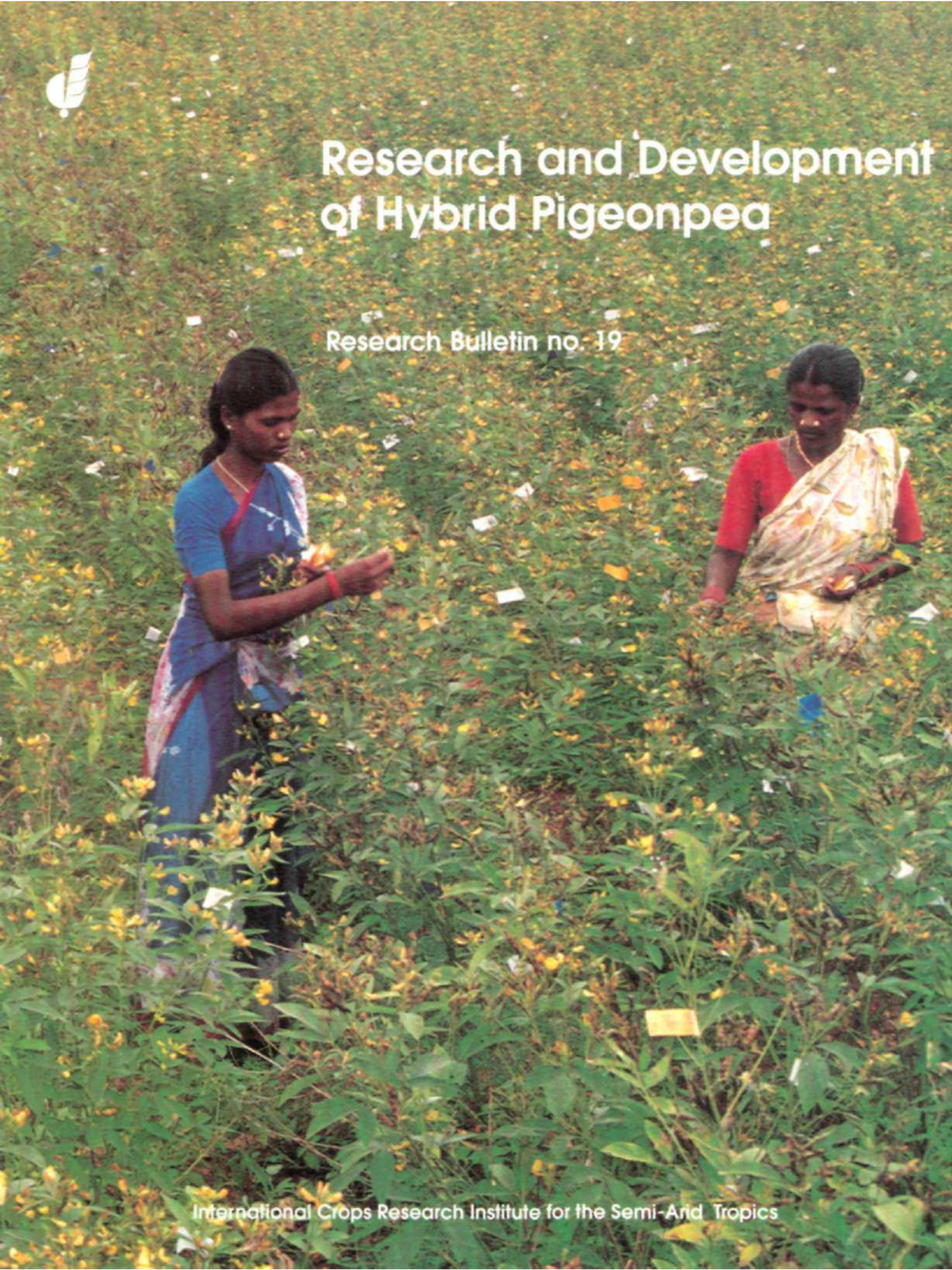
Research and Development of Hybrid Pigeonpea

Research Bulletin no. 19



International Crops Research Institute for the Semi-Arid Tropics

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Abstract

The discovery of stable genetic male-sterility in pigeonpea *(Cajanus cajan* (L.) Millspaugh) and its non- additive genetic variation for yield has paved the way for breeding hybrids. Male-sterile genes have been transferred to promising cultivars to develop male-sterile lines. ICPH 8 the first hybrid pigeonpea was released in 1991. This bulletin describes the advances made in hybrid pigeonpea research and the factors involved in the commercial exploitation of hybrid seed production.

Résumé

La recherche et le développement du pois d'Angole hybride. La découverte de la stérilité mâle génétique stable chez le pois d'Angole (*Cajanus cajan* (L.) Millspaugh), ainsi que la variabilité génétique non-additive de cette culture ont créé des conditions favorables pour la sélection des hybrides. Les gènes mâles-stériles ont été transférés dans les cultivars prometteurs en vue de mettre au point des lignées mâles-stériles. ICPH 8, la première variété hybride du pois d'Angole, était vulgarisée en 1991. Ce bulletin de recherche décrit les développements réalisés en matière de la recherche sur les hybrides du pois d'Angole, ainsi que les divers facteurs régissant l'exploitation commerciale de la production de semences hybrides.

Sumário

Pesquisa e desenvolvimento do grão do bico. A descoberta da estérilidade masculina genética no grão do bico (*Cajanus cajan* (L.) Millspaugh) que é genéticamente estavel, assim como a variação genética não aditiva no rendimento nessa safra tem preparado o tereno para criar híbridos. Os genos da estérilidade masculina foram transferidos para os cultivares prometidores para o desenvolvimento das raças da estérilidade masculina. O primeiro híbrido, ICPH 8, foi libertado em 1991. Esse boletim descreve os avanços feitos nas pesquisas dos híbridos do grão do bico assim como os fatores involvidos na exploração comercial da produção dos híbridos da semente.

Cover photo: *Identifying male-sterile pigeonpea plants in a hybrid seed production plot on the National Seeds Corporation farm at Nandikotkur, Andhra Pradesh, India.*

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ICRISAT

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Contents

Introduction

Pigeonpea *(Cajanus cajan* (L.) Millspaugh) is a short-lived perennial shrub, cultivated as an annual crop in southern and southeastern Asia, eastern Africa, the Caribbean region, and south and central America. It is chiefly grown for its seeds which are eaten either as dry split peas *(dhal)*, or as a green vegetable, and for its stems that provide a good source of fuel wood. Research on the genetic improvement of pigeonpea started with the selection of disease-resistant genotypes from landraces in the early part of this Century. Although some cultivars have recently been developed through hybridization (Singh et al. 1990) most cultivars are landraces or selections from landraces. In experimental plots, these cultivars yield up to 3-4 t ha^{-1}. The average yield in traditional farming systems, however, remains at around 700 kg ha^{-1} due to a number of yield-reducing biotic and abiotic factors. Notable success has been achieved in introducing resistance to such diseases as wilt caused by *Fusarium oxysporum* and sterility mosaic into pigeonpea cultivars (Reddy et al. 1990) but little progress in improving pigeonpea's genetic yield potential is apparent.

Pigeonpea is a partially cross-pollinated crop and traditionally, high-yielding pure lines in different maturity groups have been developed by exploiting, through pedigree selection, its additive genetic variation. A review of the literature on the quantitative genetics of pigeonpea (Saxena and Sharma 1990) shows the presence of a significant level of non-additive genetic variation for yield which could be profitably exploited through heterosis breeding to increase grain yield.

For commercial hybrid seed production there are two pre-requisites, an efficient mass pollen transfer mechanism and a stable male-sterile source. Natural cross-pollination in pigeonpea was observed as early as 1919 (Howard et al. 1919), but could not be utilized in commercial hybrid seed breeding due to the non-availability of a male-sterile source (Singh 1974; Royes 1976). Recently identified sources of genetic male-sterility and the presence of partial natural out-crossing have made it possible to explore a new avenue in pigeonpea breeding. At ICRISAT Asia Center (IAC) and several other research institutions, considerable efforts have been made to identify heterotic cross combinations, and to develop an economic hybrid seed production technology. In 1989 a special project was launched by the Indian Council of Agricultural Research (ICAR) at nine centers to strengthen research on and development of pigeonpea hybrids. The work includes diversification of male-sterile lines, development and evaluation of hybrids, seed production technology, and on-farm research. In 1991 the first commercial hybrid pigeonpea was released. This Research Bulletin summarizes the present status of research in identifying heterotic combinations, the diversification of the male-sterile base, seed production technology, and its transfer to national agricultural research systems (NARS).

Natural cross-pollination

Self-fertilization is not the rule in pigeonpea and a considerable degree of natural cross-pollination occurs (Table 1). This creates serious problems in

Table 1. Natural outcrossing (%) in pigeonpea recorded at various locations worldwide.

	Outcrossing (%)	
Country/place	Mean	Range
India		
Pusa		1.6-12.0
Nagpur	25.0	3.0 - 48.0
Niphad	16.0	11.6-20.8
West Bengal	30.0	
Ranchi		3.8 - 26.7
Coimbatore	13.7	
Varanasi		10.3-41.4
Badnapur		0.0- 8.0
Coimbatore		10.0 - 70.0
Hyderabad	27.9	0.0 - 42.1
Kenya		
Katumani	17.7	
Kibos	12.6	
Makueni	21.0	
Mtwapa	22.0	
Kabete (low pollinators)	23.3	
Kabete (high pollinators)	45.9	
Australia		2.0 - 40.0
Hawaii	15.9	5.9 - 30.0
Puerto Rico		5.5- 6.3
Trinidad	26.4	
Uganda		8.0 - 22.0

Source: Saxena et al. (1990).

Table 2. Flower drop, pods plant^{-1}, and yield plant^{-1} in fertile and sterile sibs of pigeonpea genotypes MS 3A and MS 4A, ICRISAT Asia Center 1976.

	MS 3A		MS 4A		Mean	
Character	Fertile	Sterile	Fertile	Sterile	Fertile	Sterile
Flower drop[1] (%)	42	51**[2]	49	60**	45	56**
Pods plant^{-1} (number)	320	320	247	262	283	291
Yield plant^{-1} (g)	69	66	43	57	55	61

1. Unrelated to floral bud abscission caused by low temperature.
2. ** = significant at 1%.

Source: Saxena et al. (1986).

maintaining the genetic purity of germplasm and released cultivars. There are 24 insect species that affect cross-fertilization (Onim 1981) and the degree of natural cross-pollination at any particular location depends on the presence and activity of these pollinating vectors (Saxena et al. 1990). At IAC *Apis dorsata* and *Megachile* spp. were found to be major pollinators. A study on the feasibility of hybrid seed production in two male-sterile lines revealed that the number of pods plant^{-1} and yield plant^{-1} were similar on fertile and sterile segregants (Table 2). Flower drop, however, was significantly higher in sterile plants as they produced more flowers than fertile plants did to ensure sufficient pod set. The experience at IAC and other locations shows that on average, 20% cross-pollination is sufficient for an effective hybrid seed production program.

Male sterility in pigeonpea

Genetic male-sterility

Prior to the start of ICRISAT's breeding program, male sterility coupled with female fertility had not been reported in pigeonpea, although both male and female steriles were known to exist. In 1974, an extensive search was made at IAC for male sterility among 7216 germplasm lines and 124 F_4 derivatives of crosses between *C. cajan* and its wild relatives. Seventy-two plants with aberrant floral characteristically were grouped broadly into five types (Reddy et al. 1977). The variant type 'translucent male-sterile' was considered most promising as it was totally devoid of pollen grains and its characteristically translucent anthers provided an effective means of identifying male-sterile plants in the field at an early flowering stage. Two male-sterile lines MS 3A (found in ICP 1555) and MS 4A (found in ICP 1596) from the variant type translucent male-sterile were selected for further study and possible utilization in the hybrid pigeonpea research program.

Microsporogenetic studies reveal that in these male-sterile lines, the tetrad fails to separate from the pollen mother cell and gradually degenerates through vacuolation while the tapetum persists. Inheritance studies show that male sterility is controlled by a single recessive gene ms_1 (Reddy et al. 1978). Subsequently, another source of male sterility characterized by brown arrow-head shaped anthers and controlled by a non-allelic single recessive gene ms_2 was identified in Australia (Saxena et al. 1983). Here, male sterility is caused by the degeneration of the tetrad at an early prophase stage (Dundas et al. 1981). Both male-sterile sources are being used in the hybrid pigeonpea breeding program at IAC. Figure 1 shows the two male-sterile anthers in comparison to an anther from a normal fertile plant.

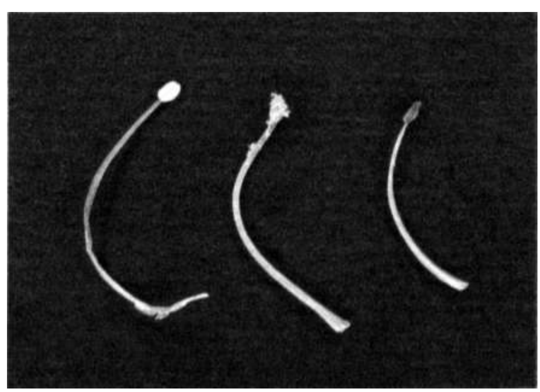


Figure 1. Anthers of **(left to right)** *translucent male-sterile ms_1, normal, and brown arrow-head male-sterile ms_2.*

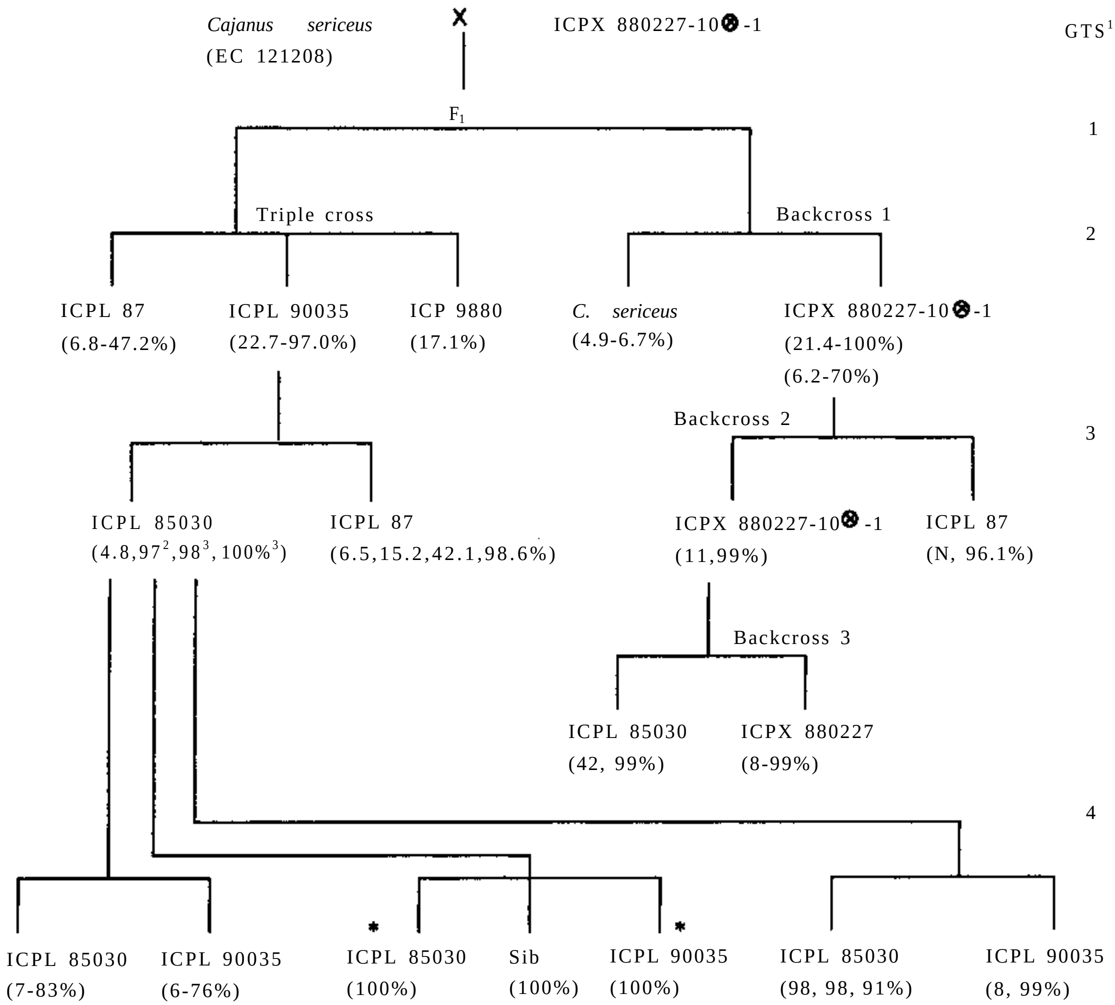
The use of genetic male sterility poses some problems in the commercial production of hybrids, as it is necessary to manually rogue fertile plants from the female parent rows. The identification of marker genes closely linked to the male-sterile gene, as reported in lettuce (Lindquist 1960) and water melon (Watts 1962), would eliminate the need to rogue, however, efforts to identify such genes in pigeonpea at IAC and elsewhere have not been successful as yet. A possible link between the male-sterile gene ms_1 and temperature sensitivity was observed by Singh et al. (1993). Pod set was found to be affected at low temperatures in the male-sterile line ms ICP 3783. When the minimum temperature dropped below 10°C and mean day temperatures were below 18°C, male-sterile plants shed floral buds. Fertile segregants showed greater tolerance of low temperatures. Floral bud abscission at temperatures below 10°C exclusive to male-sterile plants suggests that temperature sensitivity may be linked to the male-sterile gene ms_1. More experiments under varied field and controlled environmental conditions are required to confirm this hypothesis and if it is confirmed, to identify critical day/night temperatures that induce complete floral bud abscission in male-sterile plants. Seed production nurseries can then be raised in suitable locations where fertile plants in female rows need to be rogued once, considerably reducing the labor requirements of such an operation. The possible contamination of male-sterile plants with fertile sibs could also be avoided.

Cytoplasmic male-sterility

Considerable efforts are being made at IAC to develop cytoplasmic male-sterility (CMS) through mutagenesis and wide hybridization. Gamma radiation and mutagenic chemicals specific to cytoplasm organelles such as streptomycin sulfate, mitomycin-C, sodium azide, ethidium bromide, and ethyl methane sulfonate were applied at varying dosages to two ms_1 male-sterile lines, ms Prabhat-determinate (DT) and ms Prabhat-non determinate (NDT); two ms_2 male-sterile lines, QMS 1 and QMS 9; and four cultivars, ICPL 87, Prabhat, ICPL 89021, and ICPL 88039 (Ariyanayagam et al. 1993a). High levels of maternal inheritance for male sterility within the derived lines have been recovered. In one of the M_5 progenies of the sodium azide (0.025%) treated QMS 1, 87% male-sterile plants were recorded. A preliminary electrophoretic assay of esterase indicated that sterile lines developed through mutation were different from the ms_2 male-sterile parent. Together with the segregation pattern, the electrophoretic assay suggests that the mutagenic treatment has an effect on the maternal inheritance of sterility. The maintainer is the fertile sib type ms_2. Fertility restoration occurs with many normal genotypes.

In order to induce CMS through wide hybridization, attempts have been made to place the pigeonpea genome in alien (wild) cytoplasms (Ariyanayagam et al. 1993b). The investigation has indicated the prevalence of differences in the occurrence of male sterility between species, and among accessions within species. For instance, the use of *Cajanus sericeus, C. scarabaeoides,* and *C. acutifolius* as the female parent gave rise to a higher frequency of sterile progenies than C. *albicans.* Similarly, there were differences among accessions within these three promising species. Among the C. *scarabaeoides* accessions PR 4562 was most promising, and among the *C. sericeus* accessions EC 121208 responded well in three-way crosses and backcrosses. A combination of three-way crosses and backcrosses was found to be effective in minimizing such undesirable interactions as morphological abnormalities and female sterility, of the cytoplasmic and genomic parents. In general it has been observed that the expression of female sterility is less prominent in direct backcrossing with pigeonpea lines. Backcrossing, however, was effective in inducing cytoplasmic-genic male-sterility in *C. sericeus* x *C. cajan.* Some derivatives from the crosses involving *C. sericeus* and pigeonpea lines ICPL 90035 and ICPL 85030 look promising. The level of male sterility progressively increases with each backcross generation. Male sterility in different progenies at the genome transfer stage (GTS)-4 ranged between 6-100% (Figure 2). In GTS-6 some progenies exhibit 85% male sterility (Rao et al. 1995).

Isolation of CMS in pigeonpea is also in progress at various ICAR centers and at the Bhabha Atomic Research Centre (BARC), Trombay. At BARC, male-sterile plants have been isolated from the cross *C. sericeus* x TT 5. Male sterility in the derived lines was restored by some fertile segregants of the same cross (S E Pawar, BARC, personal communication).



1. GTS - genome transfer stage
2. Progeny displayed 97% male sterility, was temperature-sensitive, partially abnormal for leaf morphology, and reverted to partial fertility at low temperature.
3. Progeny displayed 98% male sterility, was morphologically normal, and retained male sterility throughout.
4. Progeny displayed 100% male sterility had one or two branches with abnormal leaf morphology and maintained male sterility throughout

N = Normal * Cytoplasmic-genic male-sterile progeny

Figure 2. A scheme for interspecific hybridization between* Cajanus sericeus *and* Cajanus cajan *to develop cytoplasmic-genic male-sterile lines. Source: Ariyanayagam et al (1993b).

Heterosis

Considerable non-additive genetic variation has been reported for yield and important yield components of pigeonpea (Saxena and Sharma 1990) and is being exploited in heterosis breeding programs. Solomon et al. (1957) were the first to report 25% hybrid vigor in yield over the better parent. Subsequently, a number of studies confirming the presence of heterosis for yield have been published and are reviewed in Saxena and Sharma (1990).

Nine genetic male-sterile based hybrids were tested at ICRISAT in 1977 and of these two hybrids were found to perform well with 31.6% and 23.6% heterosis over the best varietal control (1.1 t ha^{-1}). Since then over 1200 experimental hybrids have been developed at IAC and several exhibit significant standard heterosis for grain yield. Experimental hybrids that show high heterosis are subsequently tested in multilocational trials for stability and adaptability to different environments. Table 3 presents the yields of some experimental hybrids tested in different trials at IAC. Those that were stable and showed hybrid vigor for grain yield were evaluated again in on-farm trials. IPH 732 has subsequently been released for cultivation in Tamil Nadu.

Broadening the genetic base of male-sterile lines

Traditionally pigeonpea is grown in many cropping systems requiring cultivars that differ in plant type and maturity. These differences are so great that it is impossible to breed cultivars specific to each cropping system. Crop duration, plant type, and resistance to wilt and sterility mosaic diseases are essential components of all pigeonpea breeding programs (Saxena et al. 1986). A search for spontaneous male-sterile mutants carrying genes other than ms_1 and ms_2 now plays an essential role.

The ms_1 gene has been transferred through backcrossing into promising new genotypes. It has also been transferred to a photo-insensitive line (Saxena et al. 1980). To hasten the transfer process, backcrosses were individually made on BC_1F_1s and in succeeding generations backcrosses were made with heterozygous fertile segregants which were identified by a selfed-progeny test. By adopting such a procedure of backcrossing, 5 backcrosses were made in 8 seasons as compared with the 13 seasons required to do this using conventional conversion procedures. To avoid any potentially deleterious effects of having a single source of male sterility, the recurrent parent was used as the female parent in the final backcross. The male-sterile gene ms_2 has also been recovered in a number of genotypes. Male-sterile lines available at IAC and some of their important traits are listed in Table 4.

These male-sterile lines have been utilized to produce experimental hybrids using a wide range of pollinators. Estimates of combining ability have been made to assist in the choice of appropriate parents for desired traits in the hybrids. The general combining ability for important traits, of some of the male-sterile lines listed in Table 4, are presented in Table 5. QMS 7, ms ICPL 288, and ms T 21 have been found to be good combiners for yield. QMS 2, QMS 9, and ms Prabhat (DT) combined well for early flowering and maturity, while good combining ability for plant height and seed size was found in ms ICPL 288. Good combiners that have high heterosis for yield have been subsequently tested in multilocational trials.

Seed production technology

Maintenance of male-sterile lines

For heterosis to be well and uniformly expressed in hybrids it is essential that male-sterile stocks be genetically pure. Since genetic male-sterility in both ms_1 and ms_2 sources is controlled by a single recessive gene it must be maintained as a heterozygote by harvesting seeds from male-sterile plants pollinated by fertile heterozygotes. To multiply male-sterile lines, seeds harvested from male-sterile plants are grown in isolation (Figure 3). At flowering a young bud from each plant is manually opened, and its anthers checked for the presence or absence of pollen grains. Fertile and sterile plants are tagged separately. At maturity seeds are only harvested from male-sterile plants. Immature pods are removed from fertile plants, if necessary, to extend the period of pollen availability.

Table 3. Characteristics of some promising short-duration experimental pigeonpea hybrids, ICRISAT Asia Center, 1988/89.

Trial/Genotype	Time (d) to 50% flowering	Time (d) to maturity	Plant height (cm)	Yield (t ha^{-1})	Superiority over best control (%)
1988 Early Pigeonpea Promising Hybrids Test (Determinate) 1					
IPH 550	66	110	133	5.08	190
ICPL 4 (control)	67	106	108	1.75	
SE	±0.7	±1.0	±5.2	±0.59	
CV(%)	1	1	6	30	
1988 Early Pigeonpea Promising Hybrids Test (Determinate) 2					
IPH 575	68	113	143	4.81	126
ICPL 4 (control)	66	107	125	2.13	
SE	±0.9	±1.7	±4.6	±0.53	
CV(%)	2	2	5	25	
1988 Early Pigeonpea Promising Hybrids Test (Indeterminate) 3					
IPH 526	73	116	213	5.28	66
ICPL 161 (control)	90	133	163	3.18	
SE	±0.9	±1.1	±10.9	±0.52	
CV(%)	2	1	9	22	
1998 Early Pigeonpea Promising Hybrids Test (Indeterminate) 4					
IPH 583	89	130	289	4.26	94
Manak (control)	73	112	260	2.20	
SE	±0.9	±1.2	±7.3	±0.37	
CV(%)	2	1	4	18	
1988 Early Pigeonpea Promising Hybrids Test (Indeterminate) 5					
IPH 732	86	129	193	5.58	160
IPH 700	81	124	163	4.49	109
IPH 719	75	117	198	4.93	129
T21 (control)	82	124	178	2.15	
SE	±0.9	±1.2	±10.3	±0.68	
CV(%)	2	1	8	26	
1989 Early Pigeonpea Promising Hybrids Test (Determinate) 1					
IPH 752	71	120	89	4.53	86
ICPL 151 (control)	73	111	90	2.44	
SE	±1.3	±2.4	±2.6	±0.25	
CV(%)	3	3	5	15	
1989 Early Pigeonpea Promising Hybrids Test (Determinate) 5					
IPH 786	72	119	94	4.64	55
IPH 784	71	118	80	4.10	37
ICPL 87 (control)	78	120	85	2.99	
SE	±0.4	±0.7	±2.0	±0.16	
CV (%)	1	1	3	8	

Plot size 2.28 m^2

Table 4. Important characteristics of pigeonpea male-sterile lines available at ICRISAT Asia Center.

Line	Origin	ms gene	Growth habit[1]	Plant spread[2]	Time to 50% flower-ing (d)	Plant height (cm)	Seeds pod^{-1}	100-seed mass (g)	Seed color[3]	Disease reaction[4]	
										Wilt	SM
IMS 1	Australia	ms_1	DT	C	56	75	3.5	7.4	B	S	S
IMS 1 (Imp)	ICRISAT	ms_1	DT	C	58	90	6.0	10.0	B	S	S
QMS 9	Australia	ms_2	DT	C	61	89	3.9	7.3	B	S	S
ms Prabhat (DT)	ICRISAT	ms_1	DT	C	68	111	3.7	7.5	B	S	S
QMS 2	Australia	ms_2	DT	C	69	105	3.6	10.4	B	S	S
QMS 7	Australia	ms_2	NDT	SS	70	117	3.3	7.7	B	S	S
ms ICPL 87091	ICRISAT	ms_1	DT	C	72	115	6.5	13.4	Cr	S	S
ms ICPL 288	ICRISAT	ms_1	NDT	C	78	174	3.2	9.3	Cr	R	R
ms Prabhat (NDT)	ICRISAT	ms_1	NDT	SS	80	158	4.5	6.9	B	S	S
ms T 21	ICRISAT	ms_1	NDT	SS	99	225	3.5	8.9	B	S	S
ms ICP 3783	ICRISAT	ms_1	NDT	SS	118	180	4.0	9.2	Cr	R	R

1. DT = Determinate; NDT = Indeterminate
2. C = Compact; SS = Semi-spreading
3. B = Brown; Cr = Cream
4. S = Susceptible; R = Resistant

Table 5. General combining ability effects of six pigeonpea male-sterile lines, ICRISAT Asia Center, rainy season 1992.

Line	Time to 50% flowering (d)	Time to maturity (d)	Plant height (cm)	100-seed mass (g)	Seeds pod^{-1}	Pods $plant^{-1}$	Seed yield $plant^{-1}$(g)	Plot yield (t ha^{-1})
QMS 2	-3.06**[1]	-2.85**	-16.97**	0.64**	0.02	-5.58	-1.17	-333.41*[2]
QMS 7	9.51**	9.82**	17.94**	-0.20	-0.03	11.95*	2.91	565.06**
QMS 9	-8.87**	-6.13**	-21.97**	-0.98**	-0.05	-11.04	-4.71*	-834.17**
ms Prabhat (DT)	-4.92**	-5.85**	-18.63**	-0.94**	0.08	-0.62	-2.44	-268.94*
ms ICPL 288	1.17	-1.04	17.79**	1.26**	-0.04	-2.64	2.68	401.68*
ms T 21	6.17**	6.06**	21.84**	0.22	0.03	7.93	2.73	829.59**
SE (gi)[3]	±0.530	±0.439	±2.456	±0.104	±0.065	±4.131	±1.314	±90.408
SE (gi-gj)[4]	±0.750	±0.622	±3.473	±0.172	±0.092	±5.842	±1.859	±127.856

1. ** Significant at 1%
2. * Significant at 5%
3. (gi) Estimated GCA effect of 'i'th treatment
4. (gi-gj) Estimated difference of GCA effect between 'i'th and 'j'th treatment

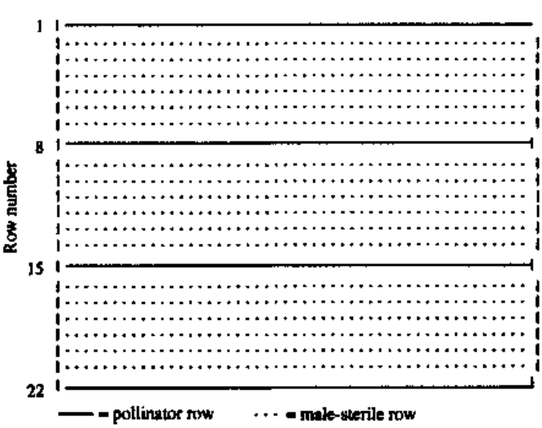


Figure 3. Layout plan for male-sterile maintenance block (in isolation).

A problem often encountered in the maintenance of male-sterile stocks is the incorrect identification of fertile and sterile plants. This is largely due to the bushy nature of the plants and the consequent intermingling of branches of neighboring plants. This problem is overcome by widely spacing plants and maintaining 6 separate rows of male-sterile to each row of normal plants.

Pollinator lines

The genetic purity of pollinators is also essential for uniform expression of hybrid vigor. To prevent genetic contamination the pollinator must be grown in isolation and off-types, if any, should be rogued before flowering starts. Small quantities of pollinator seeds can be produced under insect-proof cages.

Hybrids

Identification of heterotic crosses generally requires testing a large number of combinations. Seeds of these experimental hybrids are best produced by hand-pollinating male-sterile plants. A trained person can hand-pollinate about 400 flowers in a day. A pod set of 30-40% can be expected and the resultant seed is sufficient for the hybrid to be included in a small replicated trial.

To produce large quantities of hybrid seed, seeds from male-sterile plants in the maintenance block are sown in isolation with the required pollen-parent. Tests at IAC indicate that full pod set is obtained if one pollinator row is sown after every six male-sterile rows (Figure 4). Seed production using genetic male-sterility requires fertile segregants within female rows to be rogued. The first bud that appears on each plant has to be examined. Male-sterile plants are tagged, while fertile segregants have to be rogued before their flowers open and insects transfer their pollen to sterile plants. In pollinator rows flowering terminates when the potential pod load is realized. In the absence of sufficient pod load in sterile plants, due to lack of insect vectors and/or non-synchrony of flowering in parents,

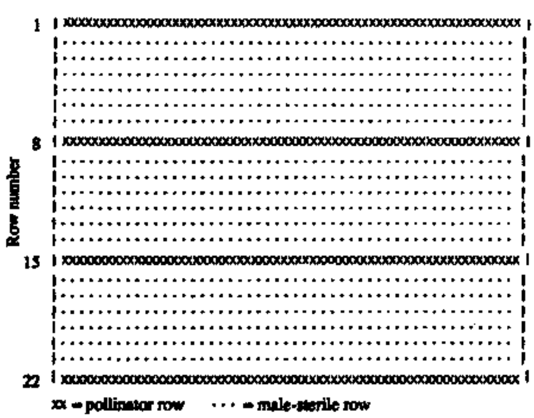


Figure 4. Layout plan for pigeonpea hybrid seed production in isolation.

flowering continues until the potential number of pods are set. To ensure an adequate hybrid yield, flowering in the pollinators can be extended by periodically removing young developing pods and frequently irrigating the plants. The ratio of male-sterile and pollinator rows may have to be changed if the recommended 6:1 ratio is not optimal because there are insufficient pollinating insect vectors, and/or plant growth is variable.

Estimated cost of seed production

Seed cost plays an important role in the wide-scale adoption of hybrid pigeonpea and management practices are clearly a critical factor in determining production costs. Preliminary studies jointly conducted by ICRISAT and the Maharashtra Hybrid Seed Company (MAHYCO) in the rainy season of 1979/80 showed that when the cost of land was not taken into consideration, a kg of medium-duration hybrid pigeonpea seed could be produced for less than Rs 2 (1 US$ = Rs 8) (Saxena et al. 1986). In the rainy season of 1988 another study was undertaken by the Institute and Tamil Nadu Agricultural University (TNAU) to determine the cost of producing hybrid pigeonpea seed. Parents of hybrid ICPH 8 were sown according to the recommendations in an isolated block measuring 0.16 ha. A single harvest yielded 813 kg ha^{-1} of hybrid seed resulting in a 1:32 seed-to-seed ratio. The estimated cost was Rs 6.25 kg^{-1} (1 US$ = Rs 13) and roguing cost 45.12% of the total production cost (Table 6). Two hundred labor days, equivalent to employing 15 workers for a fortnight, were required to rogue a seed production area of 1 ha (Murugarajendran et al. 1990).

Studies conducted by the Punjab Agricultural University (PAU), Ludhiana showed a large variation in the production costs of male-sterile and hybrid seeds. Two hundred and seventy five kg seed ha^{-1} of the male-sterile line ms Prabhat (DT) were harvested in 1990 at a cost of Rs 39.4 kg^{-1} (1 US$ = Rs 17). In 1992 rainy season, 1040 kg ha^{-1} seed were harvested, at a significantly reduced production cost of Rs 3.7 kg^{-1} (1 US$ = Rs 27) (Table 7). The estimated production cost of hybrid PPH 4 seed was Rs 13.8 kg^{-1} (Srivastava and Asthana 1993). These studies demonstrate that with good management genetic male-sterility can be exploited as a

Table 6. Cost of hybrid pigeonpea seed production at Tamil Nadu Agricultural University, India, rainy season, 1988.

Gross expenditure (Rs ha^{-1})		
Field preparation	=	1142
Inputs (Fertilizer, irrigation, insecticides, etc.)	=	1220
Labor (sowing, weeding, spraying, harvesting, seed cleaning)	=	1524
Roguing fertiles	=	3200
Total		7086
Returns per hectare		
Hybrid seed yield	=	813 kg
Pollen parent		
Seed yield	=	304 kg
Value[1]	=	Rs 1824
Value of pigeonpea stubbles	=	Rs 200
Cost of hybrid seed		
Gross expenditure	=	Rs 7086
Cost of producing	=	Rs (7086)-(1824+200)
813 kg of hybrid seed	=	5062
Cost of one kilogram of hybrid seed	=	Rs 6.25

1. For these calculations, it was assumed that the cost of parent seeds was Rs 6.00 kg^{-1} (1 US$ = Rs.13).

Source: Murugarajendran et al. (1990).

Table 7. Cost (Rs) of producing hybrid (PPH 4) and male-sterile (ms Prabhat (DT)) seeds at Punjab Agricultural University, Ludhiana, India.

	ms Prabhat (DT)			PPH 4
Item	1990	1991	1992	1992
Gross expenditure	13194	13194	13194	13194
Seed yield (kg ha^{-1})	275	630	1040	800
Fertile plants yield (kg ha^{-1})	315	720	1275	257
Value of commercial grains (Rs)	2205	5040	8924	1799
Value of byproduct (Rs)	375	375	375	375
Value of total seed (Rs)	10614	7779	3894	11020
Cost of one kg seed (Rs)	39.4	12.3	3.7	13.8

1. Estimated cost does not include such fixed costs as land rent, land revenue, depreciation, and interest on fixed cost.
2. Net seed yield after 15% losses in cleaning and grading.

Source: Srivastava and Asthana (1993).

means of producing hybrid seed at a cost acceptable to both farmers and seed growers.

Production costs can be reduced by adopting a multiple harvest system. In tropical environments with warm winters, pigeonpea produces several flushes of pods within a year (Chauhan et al. 1987) and the perennial nature of this crop can be exploited to produce quality hybrid seed at low cost (Saxena et al. 1992). Multiple harvests help to substantially reduce the cost of hybrid seed production as there is no need to rogue after the first harvest and the same seed production nursery can be used in subsequent years. Plants in such a production system need to be ratooned to a manageable height as they otherwise grow tall, making insect control and harvesting difficult.

Isolation specifications

Isolation specifications for pigeonpea differ considerably on account of the varying degrees of natural cross-pollination. Ariyanayagam (1976) cites the Food Agricultural Organization (FAO) as recommending a minimum isolation distance of 180 m and a maximum of 360 m, while Agrawal (1980) recommends distances of 400 m for the production of foundation and 200 m for certified seeds. Faris (1985) suggests that for quality varietal seed production, two varieties must be separated by at least 100 m, whilst a distance of 200 m between varieties is essential if the seed is to be used by breeders. Experience at IAC suggests that although uniform isolation standards are difficult to specify, a distance of 200-300 m is suitable, both, for the production of hybrid pigeonpea seeds and the maintenance of male-sterile and pollinator lines.

Commercial pigeonpea hybrids

ICPH 8

ICPH 8 is the first hybrid of a pulse crop to be released for commercial cultivation (ICRISAT 1993). ICPH 8 was bred by hybridizing a male-sterile line ms Prabhat (DT) and a male-fertile inbred line ICPL 161, both developed at the Institute. The ICPH 8 plants are vigorous, semispreading, and indeterminate in growth habit with profuse branching, flow-

Table 8. Zonal weighted mean seed yield of pigeonpea hybrid ICPH 8 and controls UPAS 120 and Manak in different zones in India, 1981-89.

Zone	Years	Number of trials	Yield (t ha^{-1})			Increase (%) over	
			ICPH 8	UPAS 120	Manak	UPAS 120	Manak
Northwest plains	6	36	2.85	2.10	2.34	35.0	31.0
Central	4	30	1.56	1.16	0.93	32.9	52.5
Southern	4	30	1.42	1.22	1.26	23.6	27.3
Northwestern hills	1	2	1.56	1.50	1.19	4.3	31.0
Northeastern hills	1	1	1.68	1.15	-	45.6	-
Western	1	1	2.06	1.41	1.59	45.6	29.5
Mean			1.99	1.53	1.35	30.5	34.2

Source: Saxena et al (1992).

ering, and podding characteristics. ICPH 8 flowers 80-85 days after sowing, and matures in about 140 days. Though naturally susceptible to major pigeonpea diseases, the hybrid escapes both fusarium wilt and sterility mosaic diseases because of its short duration.

ICPH 8 was released for cultivation in the central zone of India in 1991 and its performance has been outstanding in diverse environments (Table 8). Evaluations from 100 trials showed ICPH 8 yields 30.5% more than the control cultivar UPAS 120, and 34.2% more than the control cultivar Manak. In the northwest plains, central and southern zones where ICPH 8 was tested extensively it clearly established its superiority over the control cultivars. ICPH 8 was tested in minikit trials at 12 locations in the central zone during 1989. It gave 25.6% higher seed yield than variety ICPL 87 (Table 9).

PPH 4

Short-duration pigeonpeas are cultivated extensively in Punjab in a pigeonpea-wheat rotation. As ICPH 8 was found to be unsuitable for cultivation in Punjab because of its growth duration Punjab Agricultural University in Ludhiana developed and released PPH 4 in 1994. PPH 4, a short-duration, indeterminate and semispreading hybrid, was developed by crossing ms Prabhat (DT) with AL 688 NDT. With an average plant height of 260 cm and 100-seed mass of 7.5 g, PPH 4 has been reported to have an acceptable level of resistance to various biotic stresses and yields up to 20% more seeds and 25% more fuel wood than such popular cultivars as T 21, AL 15 and AL 210. On-farm trials conducted in 1994 (Verma et al. 1994) showed that PPH 4 yielded 20% more seeds than the control AL 201 (Table 9).

Table 9. Commercial pigeonpea hybrids released in India.

Name	Year	Zone	Pedigree	Origin	Maturity	Growth habit	No. of trials	Mean yield (t ha^{-1})[1]		Superiority over control (%)
								Hybrid	Control	
ICPH 8	1991	Central	ms Prabhat (DT) x ICPL 161	ICRISAT	Short-duration	NDT	12	1.80	1.44	25.6
CoH 1	1994	South	ms T 21 x ICPL 87109	TNAU/ ICRISAT	Short-duration	NDT	17	1.03	0.78	32.0
PPH 4	1994	North	ms Prabhat (DT) x AL 688	PAU	Short-duration	NDT	20	1.68	1.40	20.0

1. On-farm trials.

CoH 1

In 1994 an indeterminate short-duration hybrid IPH 732, developed by crossing ms T 21 as the female parent and ICPL 87109 as a pollen parent, was released in Tamil Nadu by TNAU as CoH 1 (Rathnaswamy et al. 1994). CoH 1 has an average plant height of 106 cm and a growth duration of 115 to 120 days. It has yellow flowers and its pods are green with purple streaks when young. CoH 1 plants have, on average, 154 pods plant^{-1} compared with 116 in ms T 21 and 48 in ICPL 87109, but its 100-seed mass (10 g) is less than that of ICPL 87109 (12 g). In 17 on-farm trials conducted in 1992 (Murugarajendran et al. 1995), CoH 1 recorded a 32% higher yield than the control VBN 1 (Table 9).

Figure 5. Comparison of seedling vigor in a hybrid (ICPH 73) and its parents (ICPL 87 and ICPL 84023), 3 weeks after sowing.

Physiological studies

Growth rate

Pigeonpea, a slow-growing crop in its early stages, is usually intercropped with crops that have a higher growth rate. Pigeonpea hybrids despite their small seeds have more seedling vigor than their parents (Figure 5). The early growth vigor of such hybrids as ICPH 8 becomes pronounced with time (Table 10) and makes them suitable for sole cropping as they establish quickly and utilize light and water resources efficiently.

Population responses

Hybrids such as ICPH 8 and ICPH 9 have shown good plasticity at plant densities ranging from 16 to

Table 10. Shoot and root mass (g plant^{-1}) of short-duration pigeonpea genotypes and hybrids (grown in pots of Alfisol) measured on different days after sowing (DAS), rainy season 1986.

Genotype	19 DAS	30 DAS	40 DAS	50 DAS	Mean
			Shoot mass (g plant^{-1})		
1CPL 87	0.36	1.11	2.93	6.21	2.65
ICPL 151	0.16	1.33	3.37	5.91	2.69
ICPH 8	0.26	0.60	4.02	7.69	3.39
UPAS 120	0.17	1.20	3.10	5.99	2.61
121	0.17	1.08	3.27	5.48	2.50
SE for comparing genotypes		±0.119**[1]			
SE for comparing genotypes x DAS		±0.311			
			Root mass (g plant^{-1})		
ICPL 87	0.09	0.29	0.75	0.16	0.70
ICPL 151	0.06	0.31	0.95	1.56	0.72
ICPH 8	0.11	0.41	1.15	2.06	0.93
UPAS 120	0.08	0.32	0.92	1.55	0.72
T 21	0.07	0.26	0.85	1.52	0.67
SE for comparing genotypes		±0.034**			
SE for comparing genotypes x DAS		±0.085			

1 ** = Significant at 1%.

66 plants m^{-2} without adversely affecting seed yield (Saxena et al. 1992). Cultivars of comparable duration require an optimum plant population of 33 plants m^{-2} necessitating a seed rate in excess of 30 kg ha^{-1}. The higher degree of plasticity in hybrids reduces the seed rate to 10-15 kg ha^{-1}.

Physiological basis of heterosis

In pigeonpea, variation in yield is primarily accounted for by differences in crop growth rates (Chauhan et al. 1995). Hybrid ICPH 8 yields more than varieties because of its uniformly higher crop growth rate. Although increasing plant population results in a higher biomass in most cultivars, high plant densities do not always increase seed yield as partitioning is adversely affected by plant competition (Chauhan et al. 1995). Pigeonpea hybrids retain their partitioning ability at least to the same level as that of traditional varieties, even at high plant populations. They also have a higher pod density and produce more seeds pod^{-1} than commonly grown varieties (Saxena et al. 1992).

Biomass production

High biomass production in hybrids is in part due to their high crop growth rate. Total biomass production in excess of 20 t ha^{-1} has been recorded in hybrid ICPH 8 in sub-tropical environments (Chauhan et al. 1995). A significant proportion (18-20%) of this harvestable biomass is 'lost' in leaf-fall but could be considered to add to the organic matter pool of the soil. Harvested stems provide useful fuel wood. Hybrid vigor in biomass production could be exploited in agroforestry systems where pigeonpea is grown as a perennial. However, when left to grow over two or three seasons, plants grow tall and have dense canopies introducing some difficulties with pesticide applications.

Drought resistance

As pigeonpea is generally grown under rainfed conditions it is subject to intermittent drought stress. Drought resistance is an important requirement in the crop. Figure 6 shows the yield response

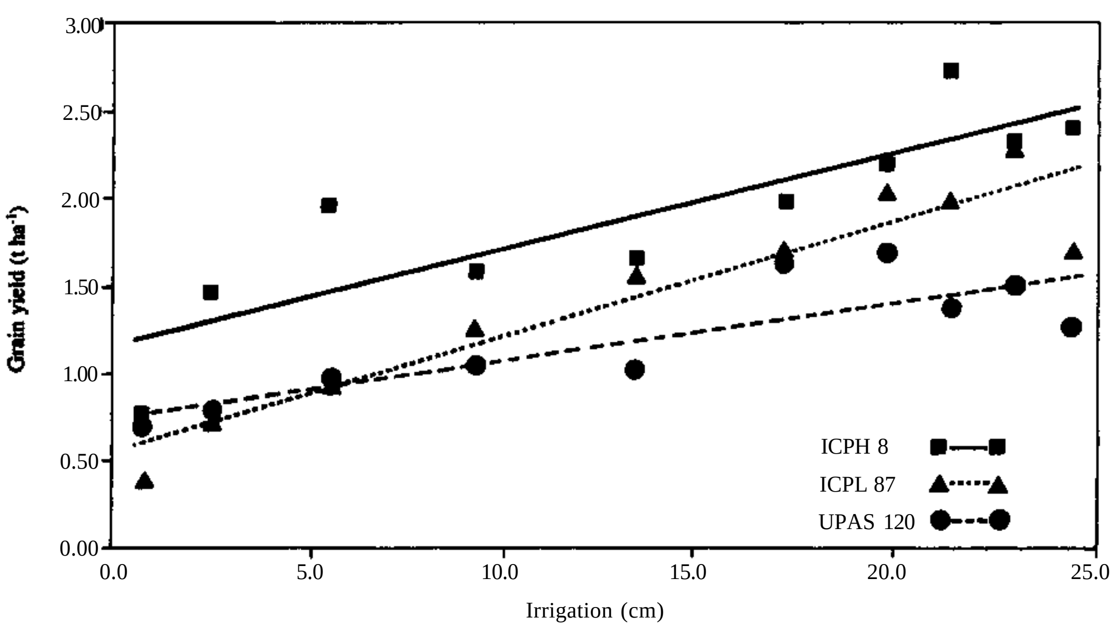


Figure* 6. *Grain yield response of ICPH 8, ICPL 87, and UPAS 120 to different levels of irrigation (cm) applied at reproductive stage, ICRISAT Asia Center, rainy season 1986/87.

of ICPH 8, ICPL 87, and UPAS 120 to differing amounts of irrigation provided at the reproductive stage of crop growth. ICPL 87 and UPAS 120 are two popular short-duration cultivars that have been released in recent years. The results in Figure 6 are consistent with reports of the good performance of ICPH 8 at locations with varying moisture regimes. Hybrids have been shown to have a more vigorous root system than varieties (Table 10) and this may account for their superior performance under drought conditions.

Technology transfer to NARS

Scientists at 1AC and ICAR have collaborated closely in the development of a technology to produce hybrid pigeonpea and the first hybrid, ICPH 8, was released for commercial cultivation in 1991. Encouraged by the Institute's initial success and the promise hybrid pigeonpea holds for increasing yields in the dry regions of India, in 1989 ICAR launched a pigeonpea research and development program at nine centers situated in different agroecological zones. The Indian Institute of Pulses Research at Kanpur coordinates national research and development activities on hybrid pigeonpea. Scientists at IAC are actively involved with these endeavors and have made the technology and parental seeds available to several private and public seed companies for large-scale seed production. ICRISAT offers training and consultation, and organizes group meetings and field visits for scientists and farmers. To effectively transfer technology, a number of training programs have been undertaken to enhance the technical skills of scientists in the public and private sector.

Conclusions

Pigeonpea is a unique pulse crop because its reproductive biology permits the utilization of both additive and non-additive genetic variance in breeding programs. However, as it is predominantly a self-pollinated crop, breeding methods in the past were tailored to exploit its additive genetic variance to develop high-yielding homozygous varieties. Until recently pigeonpea's non-additive genetic variance and hybrid vigor could not used to enhance its genetic yield potential because stable male-sterility was not available. A successful search for easily identifiable and stable male-sterility at ICRISAT in 1974 paved the way for the commercial exploitation of hybrid vigor in pigeonpea, and the first hybrid, ICPH 8, was released in 1991. Physiological studies have shown that pigeonpea hybrids are superior to homozygous cultivars with respect to seedling vigor, crop growth rate, biomass production, drought resistance, seed rate, and yield.

Large-scale breeding of hybrid pigeonpea based on genetic male-sterility suffers from an inherent difficulty. The need to manually rogue fertile segregants within female rows greatly increases the labor component of production costs. Studies on estimating production costs show that with good management and improved agronomic practices, using genetic male-sterility hybrid seeds can be produced at a cost acceptable to farmers and seed growers. However, despite its promise, hybrid pigeonpea has not been widely adopted. Discussions with public and private seed companies have revealed that there is a demand for hybrid seeds and farmers have willingly purchased hybrid pigeonpea seeds at a price as high as Rs 100 kg^{-1}. The nonavailability of quality seeds is a constraint to the widespread use of hybrid pigeonpea. Although fertile segregants are easy to distinguish from sterile ones, farmer-growers are reluctant to invest in the labor-intensive activity of roguing, and seeds supplied to seed companies are not of the required quality. Seed companies often defer payment to farmer-growers until they are assured of seed quality. This delay in payment perhaps further disinclines farmers from growing hybrid seed. Some of the factors that inhibit the large-scale production of hybrid pigeonpea seed include:

- limited seed availability of the male-sterile parent
- poor insect management in seed-production plots, resulting in low yields
- high labor costs incurred in the removal of fertile segregants
- inadequate knowledge of multiple and perennial seed production systems
- competition from more remunerative crops.

The availability of CMS holds the key to a successful commercial exploitation of hybrid vigor in pigeonpea. Preliminary work on the use of chemical mutagens and wild relatives of pigeonpea to induce CMS has shown considerable promise. ICRISAT is working on the development of a CMS system for pigeonpea, and on ways of using molecular markers to classify CMS lines. At the request of the Indian Institute of Pulses Research, the Institute has also established a working group on CMS systems, involving Indian NARS and seed companies. In 1994, two advanced-generation male-sterile progenies based on a cross between cultivated pigeonpea and *Cajanus sericeus* were found promising in greenhouse tests at IAC. Seeds from their progenies were sown in experimental plots to see whether CMS would be effective under field conditions. Pod and seed set were good in the experimental plots, and female fertility was adequately maintained. About 60-70% of plants were male-sterile, compared to the 100% in stable CMS. Further experiments are in progress; and as the percentage of male-sterile plants improves, so will the prospects of commercial production of cheap hybrid pigeonpea seeds.

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