

Studies on combining ability and heterosis involving diverse cytoplasmic male sterility system in pearl millet

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Abstract

Investigation was undertaken to study the combining ability and to quantify the magnitude of heterosis of alloplasmic isonuclear lines of pearl millet. The results revealed that the lines with A₄ cytoplasm are significantly better general combiner for grain yield per ear, ear weight, ear length and productive tillers per plant than the lines with A₁ and A₅ cytoplasm. Pollinators IP-1497, IP-973, IP-872 and IP-10085 proved their utility for breeding high yielding hybrids. None of the pollinators proved to be good combiners simultaneously for all the traits. Majority of the hybrids carrying A₄ cytoplasm were highly heterotic for the traits viz., days to maturity, plant height, ear head weight, grain yield/ear, grain yield /plant and ear length. On the other hand, majority of the A₅ based hybrids were highly heterotic for productive tillers, ear length, flag leaf area and peduncle length. The mean as well as range of heterosis for days to flowering, days to maturity, plant height, flag leaf area and 1000-grain weight was limited in all the three sources of cytoplasm. The magnitude of heterosis was high for ear weight, grain yield/ear and grain yield/plant and A₄ based hybrids had maximum heterosis for grain yield per plant and other panicle components, followed by A₁ and A₅ indicating a distinct advantage of these cytoplasm.

Key words: General combining ability, cytoplasm, heterotic, heterosis, cytoplasmic male sterility

Introduction

Pearl millet [*Pennisetum glaucum* (L.) R.Br.] is mainly grown for grain production, and is also valued for its fodder (both stover and green forage). It is highly cross-pollinated crop grown over 9.6 mh. In India. Cytoplasmic genetic male sterility system is considered as an efficient genetic tool in pearl millet hybrid breeding. Of the several cytoplasmic male sterility (CMS) sources

available in pearl millet, A₁ source is the most and widely exploited to develop commercial hybrids. However, several alternate CMS systems (A₂, A₃, A₄ and A₅) were identified and developed for use in hybrid breeding programme to diversify the cytoplasm and nuclear genetic base of pearl millet hybrids. Though a large number of A₁ cytoplasm based hybrids were released for commercial cultivation all over India, but the other sources were not being exploited due to non-availability of suitable restorers. The isonuclear lines have also been established in the background of several diverse CMS sources, which provides an opportunity for studying cytoplasmic effects on the expression of different characters. In any hybrid breeding programme based on several CMS sources, the information on combining ability of alloplasmic lines and relative magnitude of heterosis in cytoplasmically diverse hybrids helpful in determining relative chance and quantifying the magnitude and direction of heterosis in hybrids carrying different cytoplasm in pearl millet. Studies have indicated that cytoplasm exhibits pronounced effect on heterosis and combining ability [1-2]. Hence, the investigation was undertaken to study the combining ability of alloplasmic isonuclear lines of pearl millet.

Materials and methods

Three isonuclear lines viz., A₁, A₄ and A₅ cytoplasm in the genetic background of the most widely used commercial seed parent 81B were crossed with eleven diverse pollen parents (IP-9140, 10811, 16197, 12678, 8229, 9149, 873, 804, 577, 7440 and 10085) representing wide genetic variability to study the combining ability effects of alloplasmic lines. Crossing

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programme was taken up in 2004 and the resultant 33 F_1 s and 14 Parents (three corresponding B lines and 11 male parents) formed the experimental material.

The F_1 s and parents were evaluated in randomized block design with three replications during *kharif* 2005 at Regional Agricultural Research Station, Bijapur (UAS, Dharwad), Karnataka. The seed of each entry was sown in two rows of 4-meter length, intra row spacing was 15 cm and inter row spacing was 50 cm. Recommended package of practices were followed to raise the crop. Observations on twelve quantitative traits were recorded on each of the five randomly selected competitive plants tagged in each replication of all the test genotypes. Data were analysed and combining ability variances were worked out by following Line x Tester analysis suggested by Arunachalam [3] and Kempthorne [4]. Heterosis was calculated as percentage increase or decrease of F_1 's over the mid parent (MP) following the method of Turner [5] and Hayes *et al.* [6].

Results and discussion

Mean squares due to hybrids and parents were highly significant for all the traits indicating sufficient genetic variation in parental lines and hybrids for all characters. Analysis of variance for combining ability revealed that there were significant differences among the restorer lines for all the traits, indicating that selection for the hybrid parents for the study was appropriate. However, due to nuclear similarities of the isogenic lines among the testers, differences was observed only for days to maturity, plant height, flag leaf area and grain yield per plant. The significant mean squares due to the line x tester interaction indicated that hybrids differed significantly in their sca effects for all the traits. Highly significant mean squares due to parents vs. hybrids

showed presence of heterosis in hybrids for all the traits (Table 1).

The general combining ability effects for grain yield /ear head was significant and positive in A_4 cytoplasm and non-significant for A_1 and significant and negative for A_5 . The lines with A_4 cytoplasm also expressed significant gca effect for ear head weight, ear length and productive tillers/ plant. The A_1 cytoplasmic based lines showed positive and significant gca effect for two traits and negative and significant gca effect for three traits. On the other hand A_5 cytoplasmic based lines exhibited no significant gca effect for any of the traits (Table 2). These results revealed that combining ability is strongly influenced by the type of cytoplasm. Of the three CMS sources used in this study, A_4 appeared to have positive effect on many of the productive traits followed by A_1 . Yadav [7] also reported that lines with A_3 and A_4 cytoplasm are significantly better combiners for grain yield than A_1 cytoplasm in pearl millet. Virk and Brar [8] also reported significant effect of diverse cytoplasm on the combining ability in pear millet and Young and Virmani [1] reported similar results in rice.

Among the eleven pollinators, the line IP-16197 exhibited significant and positive *gca* effects for six traits and the line IP-873 for four traits including grain yield/ plant. Additionally the line IP-873 exhibited significant negative *gca* effects for time to flower indicates its utility for producing early maturity hybrids. The other two lines, which showed positive and significant *gca* effect for three traits including grain yield, were IP-577 and IP-10085. As was observed for *gca* effects significant cytoplasmic and nuclear interactions were detected with respect to all traits.

Table 1. Analysis of variance for combining ability in respect of twelve different quantitative characters in pearl millet

Sources of variation	df	Mean sum of squares											
		Days to flowering	Days to maturity	Plant height (cm)	Productive tillers/plant	Ear length (cm)	Ear girth (cm)	Flag leaf area (cm ²)	Peduncle length (cm)	Ear wt./ ear (g)	Grain yield/ ear (g)	Grain yield/ plant (g)	1000 grain wt. (g)
Hybrids	32	6.96**	7.68**	138.77**	0.30**	41.24**	2.36**	612.66**	34.18**	214.78**	126.69**	410.56**	0.98**
Replication	2	6.41	33.39	55.57	0.36	10.94	2.22	12.95	26.05	5.03	3.24	65.7	5.01
Lines	10	10.98**	4.87*	85.52**	0.09**	48.79**	1.27**	696.5*	34.30**	107.62**	94.68**	49.56**	11.16**
Testers	2	3.03	17.46*	24.92**	1.76	49.69	8.01	975.91**	78.09	342.77	113.2	113.16**	1.49
L x T	20	5.34**	8.11**	176.78**	0.25**	36.63**	2.34**	534.41**	29.73**	255.55**	144.04**	399.08**	4.38**
Error	92	1.55	1.34	9.21	0.05	2.59	0.29	197.37	2.32	6.7	1.93	4.55	0.25

* and ** - indicates significance at 5% and 1 % level respectively

Table 2. Estimates of general combining ability effects (*gca*) for twelve different quantitative characters in pearl millet

Characters	Days to flowering	Days to maturity	Plant height (cm)	Productive tiller/plant	Ear length (cm)	Ear girth (cm)	Flag leaf area (cm ²)	Peduncle length (cm)	Ear weight/ear (g)	Grain yield/ear (g)	Grain yield/plant (g)	1000 grain weight (g)
Testers												
A ₁	0.00	0.37	0.78	-0.21*	-1.37	0.52*	6.15*	-1.52*	-3.55**	0.05	0.26	0.21
A ₄	-0.30	0.47	-0.93	0.25*	0.38	-0.47	-1.98	1.56*	2.75*	1.82**	1.71	0.00
A ₅	0.30	-0.84	0.15	-0.04	1.00	-0.05	-4.17	-0.04	0.80	-1.88**	-1.97*	-0.21
S.E (gi)	0.21	0.19	0.52	0.03	0.27	0.09	1.10	0.26	0.44	0.23	0.33	0.08
S.E.(gi-gi)	0.30	0.28	0.73	0.05	0.38	13.00	1.56	0.37	0.63	0.33	0.47	0.12
Lines												
IP-9140	-0.56	1.37*	0.84	0.08	0.10	0.30	-9.83**	-1.99**	-5.63**	0.29	0.23	1.86**
IP-10811	1.00	-0.18	-0.40	0.02	-2.25**	0.44	-2.67**	1.47*	-0.71	-0.93	0.56	2.08**
IP-16197	2.00**	-0.63	4.62**	-0.05	-0.93	0.09	-5.44	1.80**	3.46**	5.71**	11.95**	0.86**
IP-12678	0.56	0.93	-3.91**	-0.01	4.19*	0.53*	-7.68*	2.99**	-2.44*	-3.28**	-11.28**	-0.58*
IP-8229	0.89	0.37	-2.14	-0.16	0.36	-0.14	4.95	-2.12**	-2.74*	-3.12**	-8.84**	-0.76**
IP-9149	-0.89	0.15	-6.23**	-0.09	-0.90	0.34	8.22**	-1.25	-2.91*	-2.05**	-2.48*	-1.07**
IP-873	-1.78**	-0.96	0.49	-0.04	-3.33**	-0.25	14.21**	0.31	1.50	2.74**	7.10**	-0.49*
IP-804	-1.11	0.04	2.94*	0.14	0.83	-0.32	2.81	1.68*	-1.34	-3.84**	-2.95**	-0.46
IP-577	0.22	-0.96	0.75	0.19	-2.67**	-0.01	8.69**	0.29	5.71**	4.95**	9.49**	-0.29
IP-7440	-0.67	-0.40	1.75	0.00	1.94*	-0.32	3.12	0.09	1.13	-0.02	-6.22**	0.01
IP-10085	0.33	0.26	1.29	-0.08	2.67**	-0.66	-6.40*	-3.28**	3.96**	-0.45	2.44*	-1.16**
S.E.(gi)	0.35	0.33	0.86	0.06	0.45	0.15	1.83	0.43	0.73	0.39	0.56	0.14
S.E(gi-gi)	0.26	0.24	0.63	0.04	0.33	0.11	1.35	0.32	0.54	0.29	0.41	0.1
C.D @ 5%	1.166	1.08	2.84	0.2	1.5	0.5	6.02	1.42	2.42	1.29	1.99	0.47
C.D @ 1%	1.54	1.43	3.76	0.27	2.00	0.67	7.98	1.85	3.21	1.72	2.69	0.62

* and ** : indicate significance at 5% and 1 % level respectively

Table 3. Average heterosis (%) of pearl millet hybrids carrying A₁, A₄ and A₅ cytoplasm

Characters	Range		Average heterosis (%) (A ₁ , A ₄ & A ₅)
	Maximum	Minimum	
Days to 50% flowering	4.30	-7.90	-2.35
Days to maturity	3.39	-4.91	-1.03
Plant height (cm)	9.42	-6.83	2.03
Productive tillers/plant	41.67	-18.52	7.57
Ear length (cm)	45.54	-8.75	21.08
Ear girth (cm)	23.59	-10.43	7.68
Peduncle length (cm)	43.90	-18.30	10.40
Flag leaf area (cm ²)	41.92	-45.06	3.98
Ear weight (g)	48.61	-11.67	39.70
Grain yield/Ear (g)	86.12	-18.20	21.80
Grain yield/plant (g)	68.39	-17.69	18.80
1000-seed weight (g)	33.12	-1784	5.70

Table 3 shows that wide range of heterosis was observed among the crosses for all the characters under study. High average heterosis was noticed for ear length, ear weight and grain yield/ear. With a view to separate out the cytoplasmic effects on heterosis for different traits the hybrids were grouped by their cytoplasmic sources (A₁, A₂ and A₃) and mean values (F₁s and MP's) were calculated in each of the group for all the twelve traits (Table 3). The mean heterosis in hybrids grouped by cytoplasmic type appeared to be of similar magnitude for days to maturity, days to 50 % flowering, flag leaf area and 1000-grain weight. But for other traits the cytoplasmic effect was conspicuous. The A₅ cytoplasmic source exhibited pronounced heterotic effect on productive tillers/plant and peduncle length compared to other sources (A₁ and A₄). Similarly the A₄ cytoplasmic source had greater heterotic effect on ear length, ear

Table 4. Mean and range of F₁, MP heterosis (%) in pearl millet hybrids carrying different cytoplasm

Characters	Mean & range	Diverse cytoplasmic sources					
		A ₁		A ₄		A ₅	
		F ₁	HMP (%)	F ₁	HMP (%)	F ₁	HMP (%)
Days to 50% flowering	Mean	46.33	-1.77	46.64	-2.52	46.06	-2.77
	Max.	48.67	4.30	50.00	4.20	47.67	1.06
	Min.	44.67	-6.61	44.33	-7.90	44.33	-7.65
Days to maturity	Mean	83.33	0.07	82.12	-2.55	83.42	-0.61
	Max.	86.33	3.39	84.33	-0.20	85.67	2.19
	Min.	80.67	-3.20	80.67	-4.91	81.00	-3.76
Plant height (cm)	Mean	177.74	1.90	177.11	3.29	176.02	0.89
	Max.	188.00	1.19	188.43	9.13	190.10	9.42
	Min.	168.40	-3.04	171.07	-0.64	162.40	-6.83
Productive tillers/plant	Mean	2.59	-1.78	2.77	5.99	3.05	18.50
	Max.	2.80	14.86	3.10	22.50	3.40	41.67
	Min.	2.33	-18.53	2.07	-22.01	2.80	6.33
Ear length (cm)	Mean	24.76	14.06	27.13	25.79	26.51	14.92
	Max.	30.20	44.96	31.53	43.77	30.30	45.54
	Min.	20.53	-8.75	23.83	-6.34	20.17	-4.87
Ear girth (cm)	Mean	10.11	12.83	9.54	8.07	9.12	2.12
	Max.	11.00	20.90	10.73	22.13	10.57	23.59
	Min.	9.27	2.02	8.30	-10.43	8.53	-6.93
Flag leaf area (cm ²)	Mean	86.05	5.72	75.76	6.07	77.95	0.15
	Max.	98.17	24.47	90.23	26.89	111.90	41.92
	Min.	71.80	-16.64	55.80	-10.93	40.80	-45.06
Peduncle length (cm)	Mean	21.55	2.16	23.03	10.35	24.62	18.73
	Max.	25.70	22.77	28.73	42.43	30.27	43.90
	Min.	17.27	-18.30	20.33	-4.01	19.23	-9.70
Ear weight/ ear (g)	Mean	46.21	30.15	52.51	48.61	50.56	40.55
	Max.	59.83	66.05	62.93	81.68	61.07	76.82
	Min.	31.17	-11.67	40.63	13.40	36.73	4.60
Grain yield /ear (g)	Mean	26.32	21.54	28.09	29.53	24.38	14.37
	Max.	38.03	71.58	40.23	86.12	30.47	50.79
	Min.	19.70	-17.63	18.13	-18.20	18.00	16.21
Grain yield /plant (g)	Mean	61.57	14.13	63.02	30.70	59.34	11.81
	Max.	73.10	45.20	82.97	68.39	81.17	47.58
	Min.	44.17	-23.41	43.37	-17.69	45.37	-17.10
1000 grain weight (g)	Mean	11.35	6.76	11.77	7.06	11.55	3.43
	Max.	13.93	33.12	14.47	32.72	13.97	26.20
	Min.	9.07	-16.82	9.37	-17.84	10.13	-14.65

head weight, grain yield /ear, grain yield /plant compared to other two sources. On the other hand, A₁ source had highest heterotic effect on ear girth. These results indicated the differential effect of diverse cytoplasm on the expression of different quantitative characters. The considerable variation in expression of heterosis in hybrids based on different cytoplasm particularly for productivity traits suggest that there exist ample opportunity to exploit these diverse sources in pearl millet hybrid breeding to raise the productivity to a greater height.

The study revealed that A₄ based hybrids had maximum heterosis for grain yield per plant and other panicle components, followed by A₁ and A₅ indicating a distinct advantage of these cytoplasmic sources. However, other equally important consideration in the use of any CMS source at commercial scale depends upon its stability, availability of strong and agronomically superior restorers. Elaborate studies conducted across wide range of environments have established that A₄ and A₅ sources are more stable than A₁, but the utility of these sources has been constrained by the lack of availability

of restorers [9]. However, by intensifying the restorer-breeding programme on these sources, the cytoplasmic base of the hybrids in pearl millet can be diversified successfully.

A comparison is made (Table 5) between the most productive and the most heterotic crosses of three different sources of male sterile lines. It is observed that most productive crosses figures as the most heterotic crosses and vice versa. In A_4 based hybrids, the mean grain yield was highest but the group as such showed the lowest mean heterotic value. It indicates that per cent heterosis as an indication of combining ability can be misleading, since a poor hybrid can show a high per cent of heterosis. With these observations it can be inferred that the low yielding (very poor) parental lines

may give highly heterotic cross, but these crosses may not be the most productive ones. It is also indicated (Table 5) that most productive as well as most heterotic crosses belonged to the hybrids based on A_4 cytoplasmic male sterile line. Hence it is clear that potentiality of A_1 and A_5 cytoplasmic sources are comparatively limited for exploitation of heterosis on commercial scale. The present study thus indicated that, the A_4 based hybrids had maximum heterosis for grain yield and other panicle components followed by A_1 and A_5 based hybrids, indicating distinct advantage of A_4 cytoplasm. But the utility of this source has been constrained by lack of availability of restorers. However, by intensifying the restorer breeding on A_4 source, the cytoplasmic base of the hybrids in pearl millet can be diversified.

Table 5. (A) Most Productive and (B) Most heterotic crosses of the study

Crosses	(A) Most productive		Crosses	(B) Most heterotic	
	<i>Per se</i> yield	HMP (%)		<i>Per se</i> yield	HMP (%)
A_1 Based			A_1 based		
1. $81A_1 \times IP-10085$	73.10	24.64	1. $81A_1 \times IP-10811$	71.53	45.20
2. $81A_1 \times IP-10811$	71.53	45.20	2. $81A_1 \times IP-873$	70.63	44.84
A_4 Based			A_4 Based		
1. $81A_4 \times IP-9140$	82.97	60.12	1. $81A_4 \times IP-873$	73.63	70.58
2. $81A_4 \times IP-16197$	81.50	68.39	2. $81A_4 \times IP-16197$	81.50	68.39
A_5 Based			A_5 Based		
1. $81A_5 \times IP-10085$	81.17	47.58	1. $81A_5 \times IP-10085$	81.17	47.58
2. $81A_5 \times IP-577$	71.53	24.01	2. $81A_5 \times IP-873$	60.97	44.84

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References

1. **Young J. B. and Virmani S. S.** 1990. Heterosis in rice over environment. *Euphytica*, **51**: 87-93.
2. **Yadav O. P.** 1994. Genetic divergence in pearl millet accessions of Indian and exotic origin. *Indian J. Genet.*, **54**: 89-93.
3. **Arunachalam V.** 1989. Genetic bases of Plant Breeding. In: *Plant Breeding Theory and Practices* (ed.) V.L. Chopra, Oxford & IBH Publishing Co. Ltd. New Delhi.
4. **Kemphorne O.** 1957. An introduction to genetic statistics. New York, John Wiley and Sons, 1 Edn. pp. 456-471.
5. **Turner J. H.** 1953. A study of heterosis in upland cotton. II. Combining ability and inbreeding effects. *Agron. J.*, **43**: 487-490.
6. **Hayes H. K., Immer F. R. and Smith D. C.** 1955. *Methods of Plant Breeding*. Mc Graw Hill Book 10. Inc. New York.
7. **Yadav O. P.** 1999. Heterosis and combining ability in relation to cytoplasmic diversity in pearl millet. *Indian J. Genet.*, **59**: 445-450.
8. **Virk D. S. and Brar J. S.** 1993. Assessment of cytoplasmic difference of near isonuclear male sterile lines in pearl millet. *Theor. Appl. Genet.*, **87**: 106-112.
9. **Gowda C. L. L., Rai K. N., Reddy B. V. S. and Saxena K. B.** 2006. Hybrid Parent Research at ICRISAT. Publishers Int. Crops Res. Institute for semi Arid Tropics, Patancheru, Andhra Pradesh (India). pp. 24-46.