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Assessment of cytoplasmic differences of near-isonuclear male-sterile lines in pearl millet

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Abstract. Four near-isonuclear polycytoplasmic versions of 81A and two of Pb 402A male-sterile lines of pearl millet (*Pennisetum typhoides*) were used in factorial matings with five inbred male testers in different combinations in three sets. The cytoplasmic differences were studied for several agronomic traits using mean values and general combining effects (gca) of male-sterile lines, and specific combining ability effects of hybrids. The fertility/sterility behaviour of different male-sterile lines in crosses with common male parents was also studied. Significant differences among near-isonuclear polycytoplasmic lines were observed in mean values for a few traits such as plant height, leaf length and peduncle length, but the differences for combining ability were more pronounced. The A_3 cytoplasm was a better general combiner than the A_2 cytoplasm for grain yield and both A_2 and A_3 cytoplasms were better general combiners for leaf length and peduncle length. In addition, superiority of A_3 cytoplasm for gca was observed for plant height and ear characters over the A_2 cytoplasm in set II. A differential behaviour of cytoplasms, both in combination with a common pollinator and across pollinators, was observed for several traits. The results provide evidence for the distinctiveness of different cytoplasmic sources in pearl millet and for the influence of cytoplasmic factors on the phenotypic expression of nuclear genes. A diversification of male sterility sources in the breeding of pearl millet hybrids is suggested.

Key words: *Pennisetum typhoides* – Pearl millet – Near-isonuclear polycytoplasmic lines – Male sterility – Cytoplasmic differentiation

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Introduction

Cytoplasmic-genetic male sterility has been extensively exploited in commercial hybrids of pearl millet since the availability of the Tift 23A male-sterile line (Burton 1965). All hybrid pearl millet is produced on the A_1 (Tift 23A) cytoplasmic-genetic sterility system. Two additional sources, A_2 and A_3, were developed (Burton and Athwal 1967) but they have not been commercially exploited. The *violaceum* cytoplasm derived from a wild subspecies, *monodii* (Maire) Brunken, has been reported to be different from the A_1, A_2 and A_3 cytoplasms (Marchais and Pernes 1965). Hanna (1989) designated this system of male sterility as A_4 ($=A_m$). Aken'ova (1985) reported a different cytoplasm in the male-sterile Ex-Bornu (referred to as Gero cytoplasm here) from Africa.

The use of a single cytoplasm to produce commercial hybrids in pearl millet is risky and presents problems of genetic vulnerability owing to a narrow cytoplasm base comparable to that of maize (Hooker 1972) and sorghum (Schertz and Ritchey 1978). Thus diversification of the cytoplasmic base is very essential given that a number of new male-sterile lines are available with different cytoplasmic sources (Gill et al. 1986; Virk and Mangat 1987, 1988, 1989; Virk et al. 1989, 1990a, b, c). However, there still exists a doubt in the mind of pearl millet breeders in respect of the distinctness of different cytoplasms. The cytoplasmic differentiation has so far been assessed by the fertility – restoration behaviour of an inbred line on different male – sterile sources as female lines (Burton and Athwal 1967). This method is not unambiguous in detecting cytoplasmic differences. The cytoplasmic differentiation is, however, facilitated if the same genome is introduced into different cytoplasms.

Such near-isonuclear polycytoplasmic male-sterile lines, developed at the Punjab Agricultural University, Ludhi-

ana (Virk and Mangat 1987, 1988, 1989; Virk et al. 1990 a, c), represent a unique material for studying the influence of nuclear-cytoplasmic interactions on agronomic traits. Therefore, the objective of the present research was to evaluate cytoplasmic effects on the mean performance of agronomic traits and to compare different cytoplasms having the same genomic complement for combining-ability effects in crosses with common testers.

Materials and methods

The material consisted of near-isonuclear polycytoplasmic lines of 81 A_1 and Pb 402A_3 male-sterile lines whereby the same genome was incorporated into variable cytoplasms through successive back crossing. The male-sterile line 81A_1 (=ICMA 1) was developed at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, and has been extensively exploited in hybrid breeding programmes in India because of its good combining ability. The 81B (maintainer of 81A) genome was substituted into the A_2 and A_3 cytoplasms at Punjab Agricultural University by backcrossing of the 81B line onto the Pb 305A_2, Pb307 A_2 and Pb 405 A_3 male-sterile lines in A_2 and A_3 cytoplasms (Burton and Athwal 1967) which resulted in Pb 310A_2 (=81A_2), Pb3111A_2 (=81A_2) and Pb 406A_3 (=81A_3) near-isonuclear male-steriles, respectively. The lines Pb 311 A_2 and Pb 406A_3 had completed seven and Pb310A_2 six, back crosses by the rainy season of 1990. Thus four near-isonuclear polycytoplasmic male-sterile lines (81A_1, Pb310A_2, Pb311A_2 and Pb 406A_3) in A_1, A_2 and A_3 cytoplasms, together with the 81B maintainer line in normal cytoplasm, were finally available (Virk and Mangat 1987, 1988, 1989; Virk et al. 1990 a, c). Similarly, the genome of the Pb402A_3 male-sterile line with A_3 cytoplasm (L67A of Burton and Athwal 1967) was substituted into A_1 cytoplasm (Tift 23A cytoplasm) by backcrossing Pb 402B with the 81A_1 male-sterile line. Thus Pb 402A_1, Pb 402A_3 male-sterile and Pb 402B male-fertile (maintainer) versions were also available.

The near-isonuclear polycytoplasmic lines, along with some other available male-sterile lines were used in factorial matings for attempting F_1 crosses during the summer season of 1990 for combining ability studies. Set I involved seven male-sterile lines (Pb208A_1, Pb211A_1, Pb311A_2, Pb406A_3, Pb402A_1, Pb402A_3 and Pb405A_3) and three male testers (13082-2-5-2, PIB 1045 and Pb307B). Set II involved three male-sterile lines (Pb 310A_2, Pb 311A_2 and Pb 406A_3) and three male testers (81B, Pb 307B and SM 697 Togo 30). Set III represented five male-sterile lines (81A_1, Pb111A_1, Pb 311A_2, Pb 402A_3 and Pb 406A_3) and three male inbred testers (81B, 13082-2-5-2 and SM 697 Togo 30). The parents and F_1 crosses of all the sets were evaluated during the rainy season of 1990 (July to October) in a randomized complete block design with two blocks having 3-m long single row plots spaced at 60 cm. Thinning was done to maintain a plant-to-plant distance of about 15 cm within rows. Data were recorded on a plot basis for days to 50% earing. Observations on five random plants in a plot were recorded for height at maturity (cm), ear girth (cm), leaf length (cm), number of tillers per plant, ear weight (g) per plant and grain yield (g) per plant.

Analyses of variance for the design and combining ability were computed following Kempthorne (1957). The combining ability effects of the parents and crosses were calcualted. The *Isd* at $P=0.05$ for comparing mean values, the general combining ability of female lines, and the specific combining ability of near-isonuclear polycytoplasmic female lines when pollinated with a common male parent, were computed.

Results and discussion

Analyses of variance (data not given) were computed for all set-character combinations. The items due to females within parents and hybrids were sub-divided into orthogonal comparisons that corresponded to differences between the near-isonuclear polycytoplasmic lines for mean performance and general combining ability, respectively. Similarly, the item females × males within hybrids was sub-divided into the corresponding orthogonal comparisons reflecting the specific combining-ability differences of hybrids involving near-isonuclear male-sterile lines. Further analysis was restricted to characters that exhibited significant variation. Since our interest centres around near-isonuclear polycytoplasmic male-sterile lines only, the relevant data in respect of mean values and estimates of general combining ability (gca) effects of the near-isonuclear polycytoplasmic male-sterile lines for different traits are presented in Table 1. The specific combining ability (sca) estimates of hybrids based on near-isogenomic lines have been presented in Tables 2, 3 and 4 for sets I, II and III, respectively.

Mean performance

The mean values for set I (Table 1) showed non-significant differences between Pb 311A_2 and Pb 406A_3 male-sterile lines except that the plant height of Pb 311A_2 was significantly shorter than that of Pb 406A_3. The only significant difference observed in set II was for leaf length for which Pb 310A_2 (28.60 cm), Pb 311A_2 (36.00 cm), and Pb 406A_3 (30.70 cm) differed significantly. It may be noted that the leaves of Pb 310A_2 were significantly smaller than those of Pb 311A_2 inspite of having been derived from the same A_2 source. In set III also the leaf length of Pb 311A_2 (36.00 cm) was significantly greater than that of Pb 406A_3. The lines 81A_1 and Pb 406A_3 did not differ significantly for leaf length. The peduncle length of 81A_1 (25.20 cm) was significantly smaller than Pb 311A_2 (29.10 cm) and Pb 406A_3 (29.20 cm).

The male-sterile line Pb 402A_3 exhibited significantly taller plants (145.50 cm) than its A_1 version, Pb 402A_1 (117.50 cm), but they did not differ for the mean expression of any other character (Table 1).

General combining ability effects

The general combining ability effects of Pb 406A_3 were significantly smaller for ear girth but higher for ear weight and grain yield than Pb 311A_2 thereby revealing the better combining ability of A_3 cytoplasm for yield than of A_2 cytoplasm. In set II (Table 1), the A_3 cytoplasm male-sterile line Pb 406A_3 had significantly higher and positive gca effects for height at maturity, leaf length, ear weight and grain yield compared to the A_2 cytoplasm line Pb 310A_2. The gca effects of Pb 406A_3 were signifi-

Table 1. Mean performance (x̄i) and general combining ability (gi) effects for characters where significant differences were observed among near-isonuclear male-sterile lines of pearl millet

Female line	DE	HT		PL		EL	EG	EW	LL		GY
	gi	x̄i	gi	x̄i	gi	gi	gi	gi	x̄i	gi	gi
Set-I											
Pb $311A_2$	0.19	90.50[a]	0.36			0.61	0.21[a]	1.38			1.62[a]
Pb $406A_3$	1.02	112.50	−4.64			−1.38	−0.53**	5.38**			5.45**
Pb $402A_1$	−0.48[a]	117.50[a]	13.02**			−3.46**[a]	−0.24[a]	−5.12**			−4.71**
Pb $402A_3$	−3.31**	145.50	0.86			1.26	−0.90**	−3.62**			−2.05
lsd 0.05	2.27	21.60	12.46			2.31	0.67	4.37			3.61
SE gi	0.73		4.00			0.74	0.22	1.40			1.16
SE (gi−gi')	1.11		6.10			1.13	0.33	2.14			1.77
Set-II											
Pb $310A_2$			−5.22[a]			−0.89	−0.02	−4.11*[a]	28.60[a]	−1.71*[a]	−3.72*[a]
Pb $311A_2$			−7.89[a]			−0.49	−0.44**[a]	−0.11[a]	36.00[a]	0.16	−0.39[a]
Pb $406A_3$			13.11**[a]			1.38*	0.42*[a]	4.22**[a]	30.70[a]	1.56*[a]	4.11*[a]
lsd 0.05			15.92			2.32	0.56	3.47	4.59	2.66	3.86
SE gi			4.28			0.62	0.15	0.93		0.71	1.04
SE (gi−gi')			7.42			1.08	0.26	1.62		1.24	1.80
Set-III											
$81A_1$			−12.20*	25.20[a]	−2.57**[a]				31.00	−3.61**[a]	
Pb $311A_2$			−8.20	29.10	0.46				36.00[a]	1.01	
Pb $406A_3$			−1.53	29.20	0.68				30.70	0.19	
lsd 0.05			17.77	3.98	2.30				5.33	3.09	
SE gi			5.42		0.70					0.94	
SE (gi−gi')			8.57		1.11					1.49	

** Significant at the 1% probability level
[a] Significant difference between near-isonuclear lines
DE = days to 50% earing; HT = height at maturity; PL = peduncle length
EL = ear length; EG = ear girth; EW = ear weight
LL = leaf length; GY = grain yield

cant and greater than those of Pb $311A_2$ for plant height, ear girth, ear weight and grain yield. The male-sterile lines Pb $310A_2$ and Pb $311A_2$, in general, showed similar gca effects in both direction and magnitude except for ear weight where the gca of Pb $310A_2$ was negative and significant, and significantly smaller than that of the Pb $311A_2$ line carrying the same cytoplasm (Table 1). In set III, the A_2 (Pb $311A_2$) and A_3 (Pb $406A_3$) lines showed significantly superior and positive gca effects for peduncle length compared with the A_1 ($81A_1$) cytoplasm line but agreed in the mean expression of this trait. Also the gca effects of A_2 (Pb $311A_2$) and A_3 (Pb $406A_3$) male-sterile lines were positive and significantly higher than the A_1 (81 A_1) cytoplasm line for leaf length (Table 1). The pair of near-isonuclear lines Pb $402A_1$ and Pb $402A_3$ in set I revealed (Table 1) that the A_3 cytoplasm was significantly better in combining for earliness, and longer and thinner ears.

Mean performance and specific combining ability of crosses

Comparisons between cytoplasms were made for those crosses where near-isonuclear male-sterile lines were pollinated with a common male parent. In set I, Pb $406A_3$ produced a significantly later flowering and higher-yielding hybrid with the 13082-2-5-2 male parent than with the Pb $311A_2$ female line. The estimate of sca for the hybrid involving Pb $406A_3$ was significantly higher than that of the hybrid involving Pb $311A_2$ for grain yield (Table 2). In combination with the PIB 1045 male parent the hybrid of Pb $311A_2$ had a significantly higher sca for ear weight and grain yield than the Pb $406A_3$ line. With the Pb 307B male parent the hybrid of Pb $311A_2$ had a shorter height and a lower grain yield performance than that of the Pb $406A_3$ line. The hybrid Pb $406A_3 \times$ Pb 307B had a significantly better sca for earliness, with taller plants and more grain yield, than the hybrid Pb $311A_2 \times$ Pb 307B.

In set II, when 81B was used as male parent (Table 3), involving maintenance backcrosses of near-isonuclear polycytoplasmic lines, the A_3 version (Pb $406A_3$) was significantly late in earing with taller plants having longer peduncles, longer leaves, higher ear weight and higher grain yield than the A_2 version, Pb $310A_2$. A similar trend was observed for the sca effects of these lines. The differences between Pb $406A_3$ and the second

Table 2. Mean performance (x̄i) and specific combining ability (sca) effects of hybrids of near-isonuclear male-sterile lines of pearl millet – set I

Female	Male	Days to 50% earing		Height at maturity		Ear length		Ear weight		Grain yield		Fertility (F)/ sterility (S)
		x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	
Pb 311A$_2$	13082-2-5-2	57.00	0.55	183.50	16.07**	26.70	1.38	23.50	−3.81	13.00[a]	−6.05[a]	F
Pb 406A$_3$	13082-2-5-2	61.00[a]	2.62*	163.00	0.57	25.60	−0.49	31.00	0.31	22.50	−0.38	F
Pb 311A$_2$	PIB 1045	57.50	0.17	176.00	0.07	25.50	0.73	39.00	8.12**[a]	29.50	8.10**[a]	S
Pb 406A$_3$	PIB 1045	59.00	0.83	159.00	−11.93**	25.90	0.37	36.00	1.12	24.50	−0.74	F
Pb 311A$_2$	Pb 307B	59.00	0.38[a]	143.00[a]	−16.14**[a]	22.60	−2.11*	22.00	−4.31*	16.00[a]	−2.05	S
Pb 406A$_3$	Pb 307B	56.00	−3.45**	165.50	11.36*	25.60	0.12	29.50	−0.81	23.00	1.12	S
Pb 402A$_1$	13082-2-5-2	56.00	−0.88	174.00	−6.10	22.90	1.64	27.00	6.19*	19.00	6.29*	F
Pb 402A$_3$	13082-2-5-2	53.00	−1.05	160.50	−7.43	22.60	−0.86	31.00	8.69**	20.50	5.12**	F
Pb 402A$_1$	PIB 1045	58.00	1.33	206.00[a]	17.40**	24.60	3.90**[a]	15.50	−8.88**	9.50	−5.57**	F
Pb 402A$_3$	PIB 1045	55.00	1.17	182.50	6.07	22.40	−0.50	19.50	−6.38**	14.00	−3.74	F
Pb 402A$_1$	Pb 307B	57.50	−0.45	160.50	−11.31	15.10[a]	−5.54**[a]	22.50	2.69	11.00	−0.71	S
Pb 402A$_3$	Pb 307B	55.00	−0.12	161.00	1.36	24.20	1.36	19.00	−2.31	13.00	−1.38	S
lsd 0.05		3.92	3.21	21.60	17.64	4.00	3.27	7.58	6.19	6.25	5.11	
SE sij			1.03		5.66		1.05		1.99		1.64	
SE(sij−skj)			1.57		8.64		1.60		3.03		2.50	

*, ** Significant at the 5% and 1% probability levels, respectively
[a] Significant difference between near-isonuclear lines

Table 3. Mean performance (x̄i) and specific combining ability (sca) effects of hybrids of near-isonuclear male-sterile lines of pearl millet – set II

Female	Male	Days to 50% earing		Height at maturity		Peduncle length		Leaf length		Ear weight		Grain yield		F/S
		x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	
Pb 310A$_2$	81B	60.00[a]	−1.67[a]	91.00[a]	−11.16[a]	27.55[a]	−2.68*	28.60[a]	−2.42*[a]	13.50[a]	−3.56*[a]	7.50[a]	−2.94[a]	S
Pb 311A$_2$	81B	62.00	−0.83	90.50[a]	−9.44[a]	29.10	−0.98[a]	36.00[a]	3.11**[a]	16.00[a]	−5.06**[a]	9.50[a]	−4.28*[a]	S
Pb 406A$_3$	81B	66.00[a]	2.50*[a]	142.00[a]	21.06**[a]	33.40[a]	3.67**[a]	33.60[a]	−0.69[a]	34.00[a]	8.61**[a]	25.50[a]	7.22**[a]	S
Pb 310A$_2$	Pb 307B	59.00	2.00[a]	161.00	9.72	31.70	0.65	34.20	0.94	28.50[a]	5.94**[a]	20.00[a]	5.39**[a]	S
Pb 311A$_2$	Pb 307B	59.00	0.83	143.00	−5.61	31.60	0.70	34.30	−0.82	22.00[a]	−4.56**[a]	13.00[a]	−4.94**[a]	S
Pb 406A$_3$	Pb 307B	56.00	−2.83*[a]	165.50	−4.11	29.20	2.03*	36.40	1.48	29.50[a]	−1.39[a]	22.00[a]	−0.44[a]	S
Pb 310A$_2$	SM 697	53.00	−0.33	156.00	1.89	34.00[a]	−1.35	34.10	−0.12	22.00[a]	−2.39[a]	14.00[a]	−2.44[a]	F
Pb 311A$_2$	SM 697	54.50	0.00	166.50	15.06*[a]	32.10	0.28	32.20	−2.29*	38.00[a]	9.61**[a]	29.00[a]	9.22**[a]	F
Pb 406A$_3$	SM 697	55.50	0.33	155.50	−16.94*[a]	29.15[a]	−2.32*	36.70	0.81	25.50[a]	−7.22**[a]	17.50[a]	−6.78**[a]	F
lsd 0.05		4.95	4.05	27.56	22.50	4.55	3.71	4.59	3.75	6.01	4.91	6.67	5.45	
SE sij			1.09		6.06		1.00		1.01		1.32		1.47	
SE (sij−skj)			1.89		10.49		1.73		1.75		2.29		2.54	

*, ** Significant at the 5% and 1% probability levels, respectively
[a] Significant difference between isonuclear lines
SM 697 = SM 697 Togo 30; F = fertility; S = sterility

Table 4. Mean performance (x̄i) and specific combining ability (sca) effects of hybrids of near-isonuclear male-sterile lines of pearl millet – set III

Female	Male	Height at maturity		Peduncle length		Ear girth		Ear weight		Grain yield		F/S
		x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	x̄i	sca	
$81A_1$	81B	130.00[a]	4.90[a]	29.40[a]	0.35[a]	6.75	0.16	25.00[a]	−2.17[a]	18.00[a]	−2.13	S
Pb $311A_2$	81B	90.50[a]	−38.60**[a]	29.10[a]	−1.98[a]	6.60	−0.08	21.00[a]	−4.17[a]	15.50[a]	−4.47**[a]	S
Pb $406A_3$	81B	142.00[a]	6.23[a]	33.40[a]	2.10*[a]	6.80	−0.39	34.00[a]	6.17**[a]	25.50[a]	4.37**[a]	S
$81A_1$	13082-2-5-2	145.50[a]	−7.20[a]	29.10[a]	−2.17*[a]	7.10	0.47	26.00	0.37	19.00	1.17	F
Pb $311A_2$	13082-2-5-2	183.50[a]	26.80**[a]	36.90[a]	2.60*[a]	7.20	0.48	23.50[a]	−0.87	17.50	0.17	F
Pb $406A_3$	13082-2-5-2	163.00	−0.37[a]	36.20[a]	1.68[a]	7.20	−0.03	31.00[a]	3.97**	22.50	3.67*	F
$81A_1$	SM 697 Togo 30	153.00	2.30	30.50	0.82[a]	6.60[a]	−0.62*[a]	37.50[a]	2.53[a]	25.50[a]	0.97[a]	F
Pb $311A_2$	SM 697 Togo 30	166.50	11.80	32.10	−6.13**[a]	6.90[a]	−0.41[a]	38.00[a]	5.03**[a]	29.00[a]	4.63**	F
Pb $406A_3$	SM 697 Togo 30	155.50	−5.87	29.15	−3.78**[a]	8.25[a]	0.43[a]	25.50[a]	−10.13**[a]	17.50[a]	−8.03**[a]	F
lsd 0.05		30.80	25.14	3.98	3.26	0.97	0.81	7.05	5.77	6.12	5.00	
SE sij			7.67		0.99		0.24		1.76		1.52	
SE (sij−skj)			12.12		1.57		0.39		2.78		2.41	

*, ** Significant at the 5 and 1 percent probability level, respectively
[a] Significant difference between near-isonuclear lines
F = fertility and S = sterility

version of the A_2 cytoplasm, Pb $311A_2$, presented a similar trend for mean values and sca effects, except for days-to-earing where the differences were non-significant. Also the mean values of these two lines, unlike their sca effects, were not significantly different for peduncle length and leaf length. The two A_2 cytoplasm versions, Pb $310A_2$ and Pb $311A_2$, did not differ for most of the traits in respect of mean values and sca effects; the only exception was for leaf length where the mean value and sca effects of Pb $310A_2$ were significantly smaller than those of Pb $311A_2$. When the male parent Pb 307B was used, Pb $406A_3$ showed a significantly better sca for earliness but a lower sca for ear weight and grain yield than the Pb $310A_2$ line, though without corresponding changes in their mean values. The trend for mean values of ear weight and grain yield was similar when the hybrids of Pb $406A_3$ and Pb $311A_2$ were compared. The mean values and sca effects of Pb $311A_2 \times$ Pb 307B were significantly lower than Pb $310A_2 \times$ Pb 307B for ear weight. With the male parent SM 697 Togo 30, the hybrid with Pb $406A_3$ had a significantly smaller mean peduncle length than the hybrid with Pb $310A_2$. Both mean values and sca effects for ear weight and grain yield for the hybrid involving the Pb $310A_2$ line were significantly lower than the hybrid with Pb $311A_2$

Set III allows a comparison of A_1, A_2 and A_3 cytoplasms (Table 4) since 81B was used as male parent with all of these lines. The A_3 cytoplasm had a significantly higher expression for the mean value of peduncle length and higher mean values and sca effects for ear weight and grain yield as compared to the A_1 cytoplasm. Compared with the A_2 cytoplasm, that of exhibited higher values for both mean expression and sca effects for plant height, peduncle length, ear weight and grain yield. The A_2 cytoplasm line (Pb $311A_2$) had smaller mean and sca values for plant height than the A_1 cytoplasm line ($81A_1$). With 13082-2-5-2 as male parent, the combination with Pb $406A_3$ had a significantly higher mean expression and sca for peduncle length than the combination with $81A_1$. This combination also had a significantly lower sca value for plant height than the combination with $81A_1$. This combination also had a significantly lower sca value for plant height than the combinations with line Pb $311A_2$. The A_2 cytoplasm (Pb $311A_2$) hybrid had a significantly greater plant height and peduncle length along with higher sca estimates than the hybrid involving the A_1 system ($81A_1$). When the SM 697 Togo 30 male parent was used the hybrid with Pb $406A_3$ had a significantly higher ear girth, and a lower ear weight and grain yield, along with a significantly lower sca for peduncle length, ear girth, ear weight and grain yield, than the hybrid with $81A_1$. The hybrid of this male parent with Pb $311A_2$ had smaller ear girth, greater ear weight and a higher grain yield, along with a significantly higher sca for ear weight and grain yield, than the hybrid with Pb $406A_3$. The

hybrid $81A_1 \times$ SM 697 Togo 30 had a significantly higher sca for peduncle length than the hybrid Pb $311A_2 \times$ SM 697 Togo 3 (Table 4).

The differences between the hybrids of Pb $402A_1$ and Pb $402A_3$ with PIB 1045 and Pb 307B male parents were significant for fewer traits (Table 2). With the PIB 1045 male parent the hybrid involving Pb $402A_1$ had a greater mean plant height (206.0 cm) and a higher sca value for ear length than its hybrid with Pb $402A_3$. When the Pb 307B male was used the A_1 cytoplasmic combination had a significantly smaller mean expression and sca for ear length.

The prevalent criterion for the classification of cytoplasmic sources, on the basis of the fertility/sterility behaviour of male-sterile lines in crosses with a common pollinator, was also used and the results are given in Tables 2, 3 and 4. In general, different near-isonuclear polycytoplasmic lines did not show differential fertility restoration with a common pollinator. The only exception was the fertile hybrid of Pb $406A_3$ as compared to the sterile hybrid of Pb $311A_2$ with a PIB 1045 male parent. Apparently, this criterion is not foolproof for studying cytoplasmic differentiation and can result in wrong conclusions concerning the similarity of cytoplasm, as in the present case. The use of mean, gca and sca criteria reveals that they are complementary and not-exclusive ways of discerning cytoplasmic differences. The simultaneous application of multiple criteria will always be advantageous in such a study.

The gca and sca effects are more revealing than the mean performance of a line in crosses with a number of male lines, and hence is closer to the genotypic mean of the line. The sca will reflect the performance of near-isonuclear lines in crosses with a common tester. Both gca and sca involve test crosses and hence are apt to reflect true differences. It may, however, be noted that the line $\times$ tester analysis used in the present study is on a fixed-effects model and some variation of results between different sets cannot be ruled out. There were differences in the outcome of these criteria in their ability to differentiate cytoplasms and these differences were more clear when different versions of 81A were pollinated with its maintainer 81B in sets II and III.

Significant differences between near-isonuclear poly-cytoplasmic male-sterile lines carrying the same nucleus can arise either due to a differential influence of the cytoplasms or to cytoplasmic-nuclear genic interactions. This may result in favourable cytoplasmic-genomic combinations e.g. the height of plants with A_3 cytoplasm was greater than that of plants with A_2 cytoplasm. This can be explained on the basis of genetic activity in mitochondria and chloroplasts (Rao and Fleming 1980). On the other hand, unfavourable combinations between the cytoplasm and the nucleus, e.g., the smaller peduncle length of A_1 than A_2 and A_3 cytoplasms, can arise due to the plasmon-sensitivity hypothesis (Renner and Kupper 1921) in which the maternal cytoplasm provides an unsuitable substrate for the paternal genes.

Significant differences in the mean performance of Pb $310A_2$ and Pb $311A_2$ male-sterile lines derived from the same cytoplasmic source but using different male-sterile lines within it involved only leaf length (Table 1). These minor differences between lines carrying the same cytoplasm derived from different parents could arise due to plasmon differences. Equally, they could be due to sampling error. In general, the direction and magnitude of gca and sca effects were similar in both lines but significant differences were observed in gca for leaf length and ear weight (Table 1), and in sca for leaf length (Table 3). The differences in gca and sca could arise as a result of cytoplasmic-nuclear interactions of the male-sterile line and the pollinator. According to Clement (1975) there is likely to be considerable cytoplasmic variability in divergent lines originating from the same cytoplasmic source. It may be concluded from the differences between near-isonuclear polycytoplasmic male-sterile lines that the cytoplasm can influence agronomic traits. The same nucleus showed differential behavior in different cytoplasms as a result of cytoplasm-nuclear (genic) interaction. Thus the expression of the phenotype is under the influence of nuclear genes, cytoplasmic factors, and the interaction of the two, and certain nuclear-genotypic combinations perform better than others. Cytoplasmic specificity in polyamine levels in anthers was also detected by Dhillion-Grewal et al. (1992) in near-isonuclear polycytoplasmic lines. The results lend clear support to the hypothesis that cytoplasmic variation in pearl millet is a genuine phenomenon which needs to be exploited in the diversification of the female parent in pearl millet hybrids.

References

Aken'ova ME (1985) Confirmation of a new source of cytoplasmic-genic male sterility in bullrush millet [*Pennisetum typhoides* (L.) Leeke]. Euphytica 34:34–38

Burton GW (1965) Pearl millet Tift 23A released. Crop and Soils 17:19

Burton GW, Athwal DS (1967) Two additional sources of cytoplasmic male sterility in pearl millet and their relationship to Tift 23A. Crop Sci 7:209–211

Clement WM Jr (1975) Plasmon mutations in cytoplasmic male-sterile pearl millet (*Pennisetum typhoides*). Genetics 79:583–588

Dhillion-Grewal R, Virk DS, Mangat BK, Basra RK, Basra AS (1992) Polyamine levels in anthers of poly-cytoplasmic isonuclear male-sterile lines of pearl millet. Bot Acad Sinica 33:97–100

Gill KS, Virk DS, Singh NB, Srivastava M (1986) Pb 405A-a new male-sterile line of *bajra* in A_3 source of sterility. Seeds and Farms 12:29–32

Hanna WW (1989) Characteristics and stability of a new cytoplasmic-nuclear male-sterile source in pearl millet. Crop Sci 29:1457–1459

Hooker AL (1972) Southern leaf blight of corn-present status and future prospects. J Envir Qual 1:244–249

Kempthorne O (1957) An introduction to genetical statistics. John Wiley and Sons, New York

Marchais L, Pernes J (1985) Genetic divergence between wild and cultivated pearl millet (*Pennisetum typhoides*). I. Male sterility. Z Planzenzuchtg 95:103–112

Rao AP, Fleming AA (1980) Cytoplasmic-nuclear genic effects in the C121 (Athens) *Zea mays* L. inbred with three cytoplasms. Euphytica 29:227–231

Renner O, Kupper W (1921) Artkreuzungen in der Gattung *Epilobium*. Ber Dtsch Botan Ges 39:201–206

Schertz KF, Ritchey JM (1978) Cytoplasmic-genic male-sterility systems in sorghum. Crop Sci 18:890–893

Virk DS, Mangat BK (1987) Substitution of CMS 81B genome into A_2 cytoplasm. Millet News Lett 6:6

Virk DS, Mangat BK (1988) Substitution of 81B genome into A_3 cytoplasm. Millet News Lett 7:3

Virk DS, Mangat BK (1989) Alloplasmic lines of L 67A in pearl millet. Millets News Lett 8:3

Virk DS, Mangat BK, Gill KS (1989) Pb208A_1 and Pb 305A_2, male-sterile lines of pearl millet. J Res Punjab Agric Univ Ludhiana 26:732

Virk DS, Mangat BK, Gill KS (1990a) Pb 310A_2 and Pb 406A_3 isonuclear male-sterile lines of pearl millet. J Res Punjab Agric Univ Ludhiana 27:379

Virk DS, Mangat BK, Gill KS (1990b) Pb 407A_3 and Pb 408A_3, male-sterile lines of pearl millet. J Res Punjab Agric Univ Ludhiana 27:548

Virk DS, Mangat BK, Gill KS (1990c) Pb 501$A_{(g)}$ and Pb 601$A_{(v)}$ male-sterile lines of pearl millet in diverse cytoplasm. J Res Punjab Agric Univ Ludhiana 27:549