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Performance of A_1, A_2, A_3, $A_{4(M)}$, $A_{4(G)}$ and $A_{4(VZM)}$ cytoplasms based iso-nuclear sorghum hybrids for shoot fly resistance across-rainy and post-rainy seasons

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Abstract

Breeding for resistance to sorghum shoot fly in A_1 CMS system has been only partially successful. To compare the alternate Cytoplasmic Male Sterility (CMS) systems for shoot fly resistance, 72 hybrids produced by crossing 36 A-lines carrying six diverse cytoplasms namely, A_1, A_2, A_3, $A_{4(M)}$, $A_{4(G)}$, $A_{4(VZM)}$, each in six nuclear backgrounds with two common fertility restorers. The hybrids were evaluated during 2006 and 2007 rainy and post rainy seasons in shoot fly screening trials at ICRISAT. ANOVA indicated absence of overall cytoplasmic influence on dead hearts%. The general (GCA) and specific combining ability (SCA) estimates suggested that inheritance for deadhearts was governed by additive-type of gene action. For GCA effects, the A_2 and $A_{4(M)}$ cytoplasms and for SCA effects, the $A_{4(G)}$ and $A_{4(M)}$ cytoplasms were superior over other cytoplasms. Overall, the $A_{4(M)}$ cytoplasm seemed to contribute to shoot fly resistance in hybrid combinations. However, use of all the six alternate cytoplasms should not increase the risk of shoot fly in commercial grain production.

Key words: Sorghum, shoot fly, resistance, hybrid, combining ability, heterosis

Introduction

Shoot fly (*Atherigona soccata* Rond.) is an important biotic constraint to sorghum production causing considerable losses in both the rainy and postrainy seasons. Shoot fly attacks sorghum at the seedling stage (18-30 days after emergence). The larvae damage the growing point of 5-30 days old sorghum seedlings. As a result, the central leaf dries up, resulting in typical dead-heart symptoms (Pont 1972). Infestation rates are higher in late-sown rainy season and early-sown postrainy season sorghum crops. The losses due to this pest have been estimated to reach as high as 85.9 per cent of grain and 44.9 per cent of fodder yield (Sukhani and Jotwani 1980). The levels of infestation may go up to 90-100 per cent (Usman 1972). The annual losses in sorghum production due to shoot fly in India have been estimated at nearly US$200 million (ICRISAT 1992). Adoption of chemical methods for insect control is not economically feasible for resource poor farmers of the semi-arid tropics and the low crop value per acre precludes the use of insecticides for control of pests. Therefore host-plant resistance combined with timely sowing is the most realistic approach for minimizing grain and stover yield losses to insect pests.

In India, while there was 37% reduction in area for sorghum production, yield increased by 80% (USDA 1997) due to concerted effort in the development and expansion of rainy season adapted sorghum hybrids. The commercial hybrids produced so far all over the globe are based on the single cytoplasm designated as *milo* or A_1 (Reddy and Stenhouse 1994; Moran and Rooney 2003). Most of the hybrids grown in India based on *milo* cytoplasm (A_1 cytoplasm) are highly susceptible to shoot fly (Dhillon et al. 2005). Major efforts in breeding A_1 cytoplasmic-nuclear male sterility-based sorghum hybrid seed parents for shoot fly resistance have met with partial success. Cytoplasmic and nuclear genetic diversity of male-sterile (A-) as well as restorer (R-) lines in sorghum is important to avoid the disease outbreak as it happened in 1970 for turcicum leaf blight of corn hybrids possessing a uniform Texas (T) cytoplasm (Tatum

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1971). However, utilization of the non-*milo* CMS systems at commercial level depends on several factors such as influence of cytoplasm on responses to pests and diseases apart from stability of male-sterility, restorer gene frequency in the germplasm and availability of commercially viable heterosis (Reddy et al. 2005). However, a reliable comparison of different cytoplasms in an iso-nuclear hybrid background has not been possible since alloplasmic male-sterile lines with a common genetic background and common fertility restorers were not available. Hence the present study was conducted to determine the influence of different cytoplasms *viz*., A_1, A_2, A_3, $A_{4(M)}$, $A_{4(G)}$ and $A_{4(VZM)}$, on shoot fly resistance using a set of diverse iso-nuclear and allo-cytoplasmic sorghum hybrids.

Materials and methods

Plant material

Six diverse sources of male-sterility inducing cytoplasms that include A_1, A_2, A_3, $A_{4(M)}$, $A_{4(G)}$ and $A_{4(VZM)}$ in the genetic backgrounds of ICSA 11, ICSA 37, ICSA 38, ICSA 42, ICSA 88001 and ICSA 88004 thus making a total of 36 A-lines were crossed with two varieties (as R-lines), IS 33844-5 and M 35-1-19 that restored fertility in all the six CMS systems to produce 72 hybrids. The six cytoplasms A_1, A_2, A_3, $A_{4(M)}$, $A_{4(G)}$ and $A_{4(VZM)}$ in the genetic backgrounds of ICSA 11, ICSA 38, ICSA 88001 and ICSA 88004 were found to be variable for traits contributing to shoot fly resistance (Dhillon et al. 2005).

Experimental design and layout

A total of 84 entries including 72 hybrids, six B-lines (ICSB 11, ICSB 37, ICSB 38, ICSB 42, ICSB 88001 and ICSB 88004), two R-lines (IS 33844-5 and M 35-1-19) and four checks (296B-high yielding B-line susceptible to shoot fly, RS 29-high yielding R-line, CSH 16-high yielding hybrid and IS 18551-shoot fly resistant line) were evaluated in shoot fly screening blocks at ICRISAT, Patancheru during the 2006 and 2007 rainy and postrainy seasons. The 72 hybrids were planted in a split-split-plot design with three replications considering R-lines as main plots, A-lines as sub-plots and cytoplasms as sub-sub-plots so that the cytoplasms will be assessed with more precision (have more degrees of freedom). The 6 B-lines, 2 R-lines and 4 checks were evaluated in an adjacent block in randomized complete block design with three replications. Each entry was planted in two rows of 2 m length with a spacing of 75 cm between rows and 15 cm between plants in a row.

Nursery management and assessment of resistance against shoot fly

In order to attain the uniform shoot fly pressure under field conditions and also to attract additional shoot flies, the interlard fish meal technique (Nwanze 1997) was followed. Interlards were first planted to build up shoot fly population. Observations on dead-heart incidence (%) were recorded in the two row plots per entry in each replication at 28 DAE (when the dead hearts in susceptible check was 90%) where dead hearts incidence (%) = (number of plants with dead-heart symptoms in each plot)/(total number of plants in the plot observed) x 100.

Data analysis

Data of individual years were subjected to analysis of variance using split-split-plot model treating R-lines as main plots, A-lines as sub-plots and cytoplasms as sub-sub plots, with Genstat 12th edition. Separate analysis of variance (ANOVA) for individual years were done to test the significance of differences among the F_1s. The error variances in the trials conducted in two years were homogeneous, as revealed by Bartlett's test (Bartlett 1937), providing statistical validity to carry out combined ANOVA. The genotypes were considered fixed while the years and replications were considered as random effects. A combined analysis was performed to test the significance of the hybrid ´ year interaction. Line × Tester analysis (Kempthorne 1957) was used to study combining ability estimates using females as lines and males as testers. The main effects of CMS and restorer lines were equivalent to general combining ability (GCA), and the effects of a CMS line with a specific restorer were equivalent to specific combining ability (SCA) (Hallauer and Miranda 1981).

Results and discussion

Significant differences among the years, seasons were recorded and also the interaction effect of year with the season was significant for dead hearts%. Such an environmental sensitivity of shoot fly infestation was also observed by Kumar et al. (2008). Non significant rank correlations for the trait across the seasons and years indicated crossover type of G × E interaction. Hence the individual years and seasons data are reported separately.

Effects of cytoplasm on hybrid mean performance

When overall mean performance for dead heart% is considered, the effect of all the cytoplasms were at

par with each other during 2006 rainy, 2006 and 2007 postrainy seasons (Tables 1 and 2). However, during 2007 rainy season, the $A_{4(M)}$ and $A_{4(G)}$ cytoplasms were superior over A_2 cytoplasm. In the individual genetic backgrounds, A_3 and $A_{4(M)}$ cytoplasms during 2006 rainy, $A_{4(M)}$ and $A_{4(G)}$ cytoplasms during 2007 rainy (Table 1) and A_2 and A_3 cytoplasms during 2006 postrainy (Table 2) had an edge over other cytoplasms for dead heart%. Thus $A_{4(M)}$ cytoplasm showed comparatively less dead heart% during rainy season. Same set of male-sterile lines in the similar nuclear backgrounds were studied in comparison to maintainer lines for dead hearts % by Dhillon et al. (2005). They also observed that $A_{4(M)}$ cytoplasm to be least susceptible compared to other cytoplasms. Umakanth et al. (2012) also reported lower dead hearts in A_4 cytoplasm across seasons. However, when the mean dead heart% values are observed, there were hardly any cytoplasmic influences on these traits in the hybrids. Similarly, only marginal differences were noted in comparison to maintainer cytoplasm by earlier workers (Dhillon et al. 2005; Umakanth et al. 2012). The male-sterile cytoplasm as such was found to be susceptible to shoot fly compared to maintainer cytoplasm (Reddy et al. 2003; Dhillon et al. 2005). However it is not so in case of other pests wherein A_1 cytoplasm was found to be more resistant to stem borer than the maintainer line cytoplasm (Reddy et al. 2003). Male-sterile lines of the both midge-resistant and midge-susceptible lines were equally susceptible, indicating that resistance to sorghum midge is influenced by factors in the cytoplasm of the B-line (Sharma 2001).

Cytoplasmic effects on GCA, SCA and heterosis

Parents for hybrid breeding can be selected based on either per se performance or general combining ability (GCA) or both. The per se performance, however, is not a reliable index as a line with high yield potential may not necessarily exhibit its superiority in cross combinations (Srivastava et al. 1979). Therefore, potential combiners which can produce combinations superior to the existing ones should be selected. The variance ratio of general to specific combining ability effects was above unity for dead heart% over rainy and postrainy seasons in both the years suggesting the preponderance of additive gene action controlling the trait. In the studies involving other insects of sorghum, GCA was found to be more important in determining tolerance against greenbug [*Schizaphis graminum* (Rondani)] (Dixon et al. 1990), stem borer stem tunneling (Singh and Verma 1988), head bug

Table 1. Mean performance of iso-nuclear allo-plasmic hybrids for dead hearts% in 2006 and 2007 rainy seasons

Hybrid	2006 Rainy season						2007 Rainy season					
	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZ\,M)}$	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZ\,M)}$
ICSA/B 11 x IS 33844-5	94	86	94	93	92	95	74	85	92	87	79	86
ICSA/B 37 xIS 33844-5	88	87	88	89	86	92	86	78	83	71	79	78
ICSA/B 38 x IS 33844-5	85	89	88	91	90	93	66	84	69	83	84	71
ICSA/B 42 x IS 33844-5	90	87	92	80^{cjmn}	92	92	77	76	68	65	82	73
ICSA/B 88001 x IS 33844-5	91	80.0^{afgh}	93	90	96	87	68	74	80	65	53^{k}	60
ICSA/B 88004 x IS 33844-5	82	80	72^{bjk}	85	86	77	80	85	83	68	68	73
ICSA/B 11 x M 35-1-19	100	100	88^{bf}	88^{cg}	89^{dh}	89^{ei}	82	92	75	63^{g}	84	83
ICSA/B 37 x M 35-1-19	89	93	92	96	85^{m}	84^{n}	65	79	84	62	74	78
ICSA/B 38 x M 35-1-19	94	88	90	91	89	91	72	63	56	55	68	59
ICSA/B 42 x M 35-1-19	79	86	74^{fl}	73^{gn}	82	86	68	82	81	79	70	63
ICSA/B 88001 x M 35-1-19	84	89	79^{fjk}	92	89	86	65	74	57	68	63	72
ICSA/B 88004 x M 35-1-19	93	94	95	90	91	91	67	72	69	75	60	72
Mean	89	88	87	88	89	88	73	79	75	70^{g}	72^{h}	72
LSD (between overall mean of hybrids) (P= 0.05)			2.78							6.67		
LSD (between cytoplasms at same levels of A-line and R-line) (P= 0.05)			9.63							23.10		

Table 2. Mean performance of iso-nuclear allo-plasmic hybrids for dead hearts% in 2006 and 2007 postrainy seasons

Hybrid	2006 Rainy season						2007 Rainy season					
	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZM)}$	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZM)}$
ICSA/B 11 x IS 33844-5	90	93	94	85	92	94	41	40	36	41	41	40
ICSA/B 37 x IS 33844-5	84[bde]	92	100	88[j]	94	95	39	33	38	46	39	41
ICSA/B 38 x IS 33844-5	97	86[a]	92	89	88	90	39	39	24	35	31	29
ICSA/B 42 x IS 33844-5	90	91	97	92	87[k]	82[ln]	54	34	42	37	32[d]	33
ICSA/B 88001 x IS 33844-5	91	86	83	92	91	89	41	45	50	39	33	50
ICSA/B 88004 x IS 33844-5	95	92	83[bfjk]	93	93	90	46	33	43	38	51	46
ICSA/B 11 x M 35-1-19	96	96	98	94	92	93	43	29	34	29	37	43
ICSA/B 37 x M 35-1-19	94	96	88	94	89	93	19[c]	22[g]	23[j]	48	40	35
ICSA/B 38 x M 35-1-19	91	93	97	93	91	95	32	36	29	39	22	27
ICSA/B 42 x M 35-1-19	95	95	84[bf]	91	86	92	43	25	32	24	37	29
ICSA/B 88001 x M 35-1-19	92	78[afghi]	93	92	94	97	45	31	41	24	31	31
ICSA/B 88004 x M 35-1-19	90	93	92	93	93	90	28	43	42	42	31	38
Mean	92	91	92	91	91	92	39	34	36	37	35	37
LSD (between overall mean of hybrids) (P= 0.05)			2.69							6.25		
LSD (between cytoplasms at same levels of A-line and R-line) (P= 0.05)			9.31							21.64		

Table 3. Estimates of SCA effects of iso-nuclear allo-plasmic hybrids for dead hearts% in 2006 and 2007 rainy seasons

Hybrid	2006 Rainy season						2007 Rainy season					
	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZM)}$	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZM)}$
ICSA/B 11 x IS 33844-5	3.96	0.29	-1.21	0.29	-0.38	-3.38	-5.61	3.14	-2.90	4.22	6.47	1.07
ICSA/B 37 x IS 33844-5	-2.21	-1.21	-1.88	1.29	5.79	0.12	-8.91*	-6.75	6.80	0.85	2.05	4.94
ICSA/B 38 x IS 33844-5	5.96	1.62	4.46	0.62	-6.04[d]	4.29	-2.88	7.92	-3.10	-2.63	7.50	-2.20
ICSA/B 42 x IS 33844-5	-0.29	-0.21	0.29	-1.88	-1.88	0.29	-4.73	6.20	0.95	-6.71	11.27	-1.46
ICSA/B 88001 x IS 33844-5	3.12	-0.38	-0.54	-3.38	0.29	-2.04	4.80	0.84	4.45	-6.53	1.22	3.52
ICSA/B 88004 x IS 33844-5	-2.38	-6.54	1.46	-0.71	2.46	-0.04	-4.10	-4.83	-3.96	-2.45	-2.71	-5.80
ICSA/B 11 x M 35-1-19	-3.96	-0.29	1.21	-0.29	0.38	3.38	5.61	-3.14	2.90	-4.22	-6.47	-1.07
ICSA/B 37 x M 35-1-19	2.21	1.21	1.88	-1.29	-5.79	-0.12	8.91	6.75	-6.80	-0.85	-2.05	-4.94
ICSA/B 38 x M 35-1-19	-5.96[d]	-1.62	-4.46	-0.62	6.04	-4.29	2.88	-7.92	3.10	2.63	-7.50	2.20
ICSA/B 42 x M 35-1-19	0.29	0.21	-0.29	1.88	1.88	-0.29	4.73	-6.20	-0.95	6.71	-11.27*	1.46
ICSA/B 88001 x M 35-1-19	-3.12	0.38	0.54	3.38	-0.29	2.04	-4.80	-0.84	-4.45	6.53	-1.22	-3.52
ICSA/B 88004 x M 35-1-19	2.38	6.54	-1.46	0.71	-2.46	0.04	4.10	4.83	3.96	2.45	2.71	5.80
SE± (Sij)				4.10						8.58		
SE± (Sij-Skl)				5.79						12.14		

Significant differences between [a]A_1 and A_2, [b] A_1 and A_3, [c]A_1 and $A_{4(M)}$, [d]A_1 and $A_{4(G)}$, [e]A_1 and $A_{4(VZM)}$, [f]A_2 and A_3, [g]A_2 and $A_{4(M)}$, [h]A_2 and $A_{4(G)}$, [i]A_2 and $A_{4(VZM)}$, [j]A_3 and $A_{4(M)}$, [k]A_3 and $A_{4(G)}$, [l]A_3 and $A_{4(VZM)}$, [m]$A_{4(M)}$ and $A_{4(G)}$, [n]$A_{4(M)}$ and $A_{4(VZM)}$, [o]$A_{4(G)}$ and $A_{4(VZM)}$ cytoplasms

(*Eurystylus oldi*) and midge (*Stenodiplosis sorghicola*) (Ratnadass et al. 2002). However, in the current study, there was inconsistency in the influence of cytoplasm on GCA effects for dead hearts%. During rainy season, in the genetic background of ICSA 38, the gca effects of $A_{4(M)}$ and $A_{4(G)}$ were significant and superior over

Table 4. Estimates of SCA effects of iso-nuclear allo-plasmic hybrids for dead hearts% in 2006 and 2007 postrainy seasons

Hybrid	2006 Rainy season						2007 Rainy season					
	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZM)}$	A_1	A_2	A_3	$A_{4(M)}$	$A_{4(G)}$	$A_{4(VZM)}$
ICSA/B 11 x IS 33844-5	-1.64	4.51	-1.28	-4.19[m]	6.09	-1.03	0.08	-2.81	-0.12	2.78	-4.44	-1.22
ICSA/B 37 x IS 33844-5	5.16	1.48	-0.64	-0.58	2.54	2.53	-0.22	-5.82	0.58	9.53	1.96	-1.04
ICSA/B 38 x IS 33844-5	2.46	-4.61	0.66	1.29	-5.39	1.29	-2.49	5.09	-9.11	6.01	1.96	1.68
ICSA/B 42 x IS 33844-5	-0.04	1.99	-0.63	-1.19	-2.78	-2.19	6.66	2.04	-3.71	-1.94	-12.67	1.83
ICSA/B 88001 x IS 33844-5	-1.21	1.79	-2.29	-0.48	-3.63	-2.93	-2.62	-5.27	-2.74	6.73	3.56	3.56
ICSA/B 88004 x IS 33844-5	6.98	0.58	-1.36	-1.61	-0.83	1.14	6.49	8.06	-0.67	-7.76	-0.19	-3.72
ICSA/B 11 x M 35-1-19	1.64	-4.51	1.28	4.19	-6.09[m]	1.03	-0.08	2.81	0.12	-2.78	4.44	1.22
ICSA/B 37 x M 35-1-19	-5.16	-1.48	0.64	0.58	-2.54	-2.53	0.22	5.82	-0.58	-9.53	-1.96	1.04
ICSA/B 38 x M 35-1-19	-2.46	4.61	-0.66	-1.29	5.39	-1.29	2.49	-5.09	9.11	-6.01	-1.96	-1.68
ICSA/B 42 x M 35-1-19	0.04	-1.99	0.63	1.19	2.78	2.19	-6.66	-2.04	3.71	1.94	12.67	-1.83
ICSA/B 88001 x M 35-1-19	1.21	-1.79	2.29	0.48	3.63	2.93	2.62	5.27	2.74	-6.73	-3.56	-3.56
ICSA/B 88004 x M 35-1-19	-6.98*	-0.58	1.36	1.61	0.83	-1.14	-6.49	-8.06	0.67	7.76	0.19	3.72
SE± (Sij)				3.35						7.60		
SE± (Sij-Skl)				4.74						10.74		

Significant differences between [a]A_1 and A_2, [b] A_1 and A_3, [c]A_1 and $A_{4(M)}$, [d]A_1 and $A_{4(G)}$, [e]A_1 and $A_{4(VZM)}$, [f]A_2 and A_3, [g]A_2 and $A_{4(M)}$, [h]A_2 and $A_{4(G)}$), [i]A_2 and $A_{4(VZM)}$, [j]A_3 and $A_{4(M)}$, [k]A_3 and $A_{4(G)}$, [l]A_3 and $A_{4(VZM)}$, [m]$A_{4(M)}$ and $A_{4(G)}$, [n]$A_{4(M)}$ and $A_{4(VZM)}$, [o]$A_{4(G)}$ and $A_{4(VZM)}$ cytoplasms

A_2 and A_3 during 2006 while it was vice-versa during 2007. In the genetic background of ICSA 88001, $A_{4(G)}$ were significant and superior over all other cytoplasms during 2006 while A_2 was superior over $A_{4(M)}$ and $A_{4(G)}$ during 2007. During postrainy season of 2006, the gca effects of A_1 cytoplasm was signicant and superior over A_2, A_3, $A_{4(G)}$ and $A_{4(VZM)}$ in ICSA 88004 genetic background while during 2007 postrainy season, the gca effects of $A_{(M)}$ cytoplasm was significant and superior to A_1, A_3 and $A_{4(VZM)}$ cytoplasms in ICSA 42 genetic background. Thus, the $A_{4(G)}$ cytoplasm during 2006 rainy season, A_2 cytoplasm during 2007 rainy season, A_1 during 2006 postrainy season and $A_{4(M)}$ cytoplasm during 2007 postrainy season had an edge over other cytoplasms for combining for low dead hearts%. Overall, A_2 and $A_{4(M)}$ cytoplasms seemed to have marginal superiority over others for good GCA for shoot fly resistance.

SCA effects reflect differential interaction of cytoplasms with nuclear genes of A-lines as well as R-lines and it is this interaction in higher magnitude and desired direction that results in superior hybrid performance. Similar to the GCA effects, there was inconsistency in the influence of cytoplasm on SCA effects for dead hearts%. The A_1 and $A_{4(G)}$ cytoplasms during 2006 rainy season (Table 3), $A_{4(M)}$ and $A_{4(G)}$ cytoplasms during 2006 postrainy season (Table 4) had an edge over other cytoplasms for combining for low dead hearts%. Overall $A_{4(G)}$ and $A_{4(M)}$ cytoplasms seemed to have marginal superiority over others for good SCA for shoot fly resistance. Earlier studies have also indicated that heterosis breeding would not be rewarding in breeding for resistance to shoot fly (Dhillon et al. 2005). Considering the mean performance and combining ability, the $A_{4(M)}$ cytoplasm holds promise for the development of shoot fly resistant hybrids during both rainy and postrainy seasons. Similar findings were reported by Dhillon et al. (2005) on comparison of male-sterile with maintainer lines. For potential use of $A_{4(M)}$ cytoplasm, it has to be screened in diverse nuclear backgrounds as interactions between cytoplasmic and nuclear genes possibly control the expression of traits associated with resistance to sorghum shoot fly in the F_1 hybrids as has also been observed by Sharma et al. (2006).

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