

Assessing parental lines and crosses for shoot fly resistance in sorghum [*Sorghum bicolor* (L.) Moench]

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(Received: June 2011; Revised: December 2011; Accepted: January 2012)

Abstract

Shoot fly is a key pest of sorghum in many countries including India. Resistance breeding towards shoot fly is important to address the need of resource poor farmers of the semi-arid tropics. Shoot fly resistance response was studied among 19 parental lines and 78 hybrids in two diverse environments. Cytoplasmic male sterile (CMS) lines, MS 104A and SPSFR 94010A and testers, SFCR 125, SFCR 151, ICSV 705, ICSV 708 and PS 30710 were identified as best for shoot fly resistance traits. The hybrids based on shoot fly resistant CMS and restorer lines had significantly lower proportion of oviposition and dead hearts than the hybrids based on other cross combinations. Leaf glossiness and trichome density revealed high correlation with shoot fly resistance. Thus, while breeding for shoot fly resistance efforts are needed to focus on combining both the traits among the hybrids.

Key words: *Atherigona soccata*- host plant resistance
- glossiness- trichome density-correlations

Introduction

Sorghum [*Sorghum bicolor* (L.) Moench] is an important cereal crop in the semi-arid tropics and is widely cultivated for food, feed, fodder and fuel in Africa and Asia. In India, sorghum is the third important cereal crop after rice and wheat having 7.53 mha area with an annual production of 7.25 mt. Various biotic and abiotic constraints result in relatively low productivity of sorghum (500-800 kg/ha) in India. Sorghum crop is infested by nearly 150 arthropod species from seedling to maturity stages. Insect pest causes considerable losses (~ 32%) of the actual produce of sorghum in India [1]. Sorghum shoot fly (*Atherigona soccata* Rond.) is one of the major pest attacking at seedling stage [2].

Damages leading to as high as 90 per cent dead heart with 75 and 68 per cent losses in grain and fodder yield respectively, have been reported. An increase in 1% of dead hearts due to shoot fly results in loss of 143 kg grain yield per hectare [3].

Chemical methods for the control of shoot fly are not cost effective and ecologically unsafe. Integrated pest management strategies can be effectively followed to control insect damages by utilizing host plant resistance mechanism. Resistant cultivars and hybrids can bring stability to production under low input conditions. Host plant resistance research in sorghum has achieved considerable progress in identifying sources of resistance and transferring resistance to high yielding cultivars [4]. A number of genotypes with resistance to shoot fly have been identified, but the levels of resistance among these genotypes are low to moderate. Most of the earlier studies suggested that the host plant resistance to sorghum shoot fly is of complex nature and is governed by various component traits [5]. For improving the level of resistance in crop plants, identification resistance attributing traits and their individual and combined effects is of paramount importance to select resistant genotypes and to plan effective breeding strategy. The objectives of this work were i) to evaluate parental lines including maintainers and hybrids to discern response of new parental lines to shoot fly, ii) to get insight of the differences amid cytoplasmic male sterile (CMS) lines and their maintainers, and iii) to find out the association amongst shoot fly resistance attributing traits.

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Materials and methods

In the present investigation, six CMS lines and thirteen testers were used. These genotypes were selected on the basis of genetic variation observed for shoot fly response and yield attributing traits. Seeds of the parental lines were obtained from Sorghum Research Station (SRS), Marathwada Agricultural University (MAU), Parbhani, India and International Crop Research Institute for Semi-Arid Tropics (ICRISAT), Patancheru, Hyderabad, India. Among the CMS lines two (MS 104A and SPSFR 94010A) were shoot fly resistant and four (PMS 7A, PMS 8A, PMS 28A and SPSFR 94012A) were susceptible. Six among the testers (SFCR 125, SFCR 151, ICSV 700, ICSV 705, ICSV 708 and PS 30710)

were shoot fly resistant and seven (KR 192, KR 196, KR 199, CS 3541, MR 750, C 43 and ICSV 91011) were susceptible. Details of the lines and testers are presented in Table 1.

Seeds of parental lines were staggered sown at 10-15 days interval during 2005-06 post-rainy season to obtain synchronization between male and female parents. All the six male sterile lines were crossed with thirteen testers in line × tester fashion to obtain 78 crosses. The panicle of Each selected female was pollinated at least three to four days continuously for effective and sufficient seed setting. Parental lines were also selfed to obtain pure seeds.

Table 1. List of the lines used in the investigation with their pedigree and response to shoot fly resistance

Genotype	Pedigree	Response to shoot fly [#]
Lines		
PMS 7A	Selection from SPMD 9069	S
PMS 8A	Selection from ICSR 88014 LP	S
PMS 28 A	Tall selection from 296 B (ICSB 94040B × 296B)	S
SPSFR 94012A	[BTx623 × (SC 108-3 × GPR 148)-18-4-1] × (B line bulk)-5-1-2-5) × PS 21194 × SPV 351) -3-1-2-3-3]-13-3-1-4.	S
MS 104 A	Selection from 296B × Swati (SPV 504)	R
SPSFR 94010A	[BTx623 × (SC 108-3 × GPR 148)-18-4-1] ×(B line bulk)-5-1-2-5) × PS 30715-1 × PS 19349 B)-2-4-1	R
Restorers (testers)		
KR 192	Selection from SPV 544 × SPV 462-3	S
KR 196	Early SPS × SPV 1023	S
KR 199	Developed from IS 93094	S
CS 3541	IS 3675 × IS 3541	S
MR 750	ICSR 38= (SC 108-3 × CSV 4)-27-2-1	S
C 43	CS 3541 × IS 235449	S
ICSV 91011	(IS 9608 × [SC 108-3 × Swarna) × IS 9327)-6-2-1)-3-6-2.	S
SFCR 125	(ICSV 705 × YT-3-47)-7-1-1-2	R
SFCR 151	(1011 E 23-2 (PM 12645 × IS 2205)-5-1-2-2.	R
ICSV 700	(IS 1082 × SC 108-3)-1-1-1-1-1	R
ICSV 705	[(125/R × EW 3257-4) × -1-5-6-1-1-1 × (SC 108-3 × CS 3541)-19-1]-3-1-2-3-3.	R
ICSV 708	(IS 5622 × CS 3541)-20-1-1-1-1-1 × (VC 4 V2 × Bulk 7-55)-1-5-1) -5-2-5-1-1	R
PS 30710	[(IS 5622 × CS 3541)-20-1-1-1-1-1-1 × (Uch V2 × Bulk Y-55)-1-5-1]-5-1-6-1	R
Checks		
PVK 801	Selection from GD 34-5-5-3	S
CSH 16	PMS 27A × C 43	S
IS 18551	Originates from Ethiopia, race <i>Durra</i> .	R

[#]S = Susceptible, R = Resistant

All the 78 F₁ hybrids along with parents and three checks, viz., PVK 801 (susceptible variety), CSH 16 (commercial hybrid) and IS 18551 (resistant source) were sown in randomized block design with two replications. Maintainer lines (B lines) were also sown to evaluate their reaction to shoot fly. Each parental line and F₁ hybrids were planted in a single row plot (4 m) with row to row and plant to plant spacing of 45 and 15 cm, respectively. The sowing was carried out on two dates, viz., 22nd July, 2006 for late rainy season screening and 22nd September, 2006 for screening in early post-rainy season. Each replication consisted of 106 entries, which were divided into two tiers to reduce soil heterogeneity and entries within the replication were randomized. After every ten rows of test entries one row each of susceptible (PVK 801) and resistant (IS 18551) checks were sown. Six border rows with susceptible genotype (PVK 801) were sown around experimental plot 20 days before sowing of main experiment. To attain uniform shoot fly pressure under field condition the inter lard fish meal technique [6] was followed. Plant protection measures were avoided up to 30 days after sowing (shoot fly infestation period).

Data were recorded in each row excluding border plants on leaf glossiness, seedling vigour, oviposition percentage, dead heart percentage and trichome density on adaxial (upper) and abaxial (lower) surface of lamina. Evaluation for leaf glossiness was performed according to the scale given by Sharma *et al.* [7]. Seedling vigour (a combination of height, leaf growth, and robustness) was evaluated on a 1 to 5 scale at 9 Days after emergence (DAE). Oviposition percentage was calculated at 14 and 21 DAE by multiplying with 100 the ratio of number of plants with eggs to total number of plants. Similarly, the dead heart percentage was calculated by calculating the ratio of number of plants with dead heart to total number of plants and multiplying with 100 at 21 and 28 DAE. The observations on oviposition and dead hearts were angular transformed for statistical analysis. Trichome density was recorded at 12 DAE on central portion of the fifth leaf (from the base) of three random seedlings from each genotype in each replication [5]. Number of trichomes per microscopic field was recorded in adaxial (upper) and abaxial (lower) surfaces of the leaves.

The data were subjected to analyses of variance. The parents were classified as resistant and susceptible on the basis of their reaction to shoot fly. Correlation between shoot fly resistance as defined by oviposition and dead heart percentage with shoot fly resistance

attributing traits like leaf glossiness, seedling vigour, trichome density on adaxial and abaxial surface of lamina were estimated using SPSS ver. 13.0. The contribution of each trait to dead heart (%) alone and in combination was also studied. The parents and hybrids were grouped based on different trait combinations. Mean dead heart (%) of each group was calculated and their significance was tested using t-test.

Results and discussion

Non-preference for oviposition has been considered as one of the predominant mechanisms responsible for shoot fly resistance [6]. However, seedling resistance due to inability of the larvae to penetrate through the leaf sheaths and survival of the new tillers formed when the growing shoot is destroyed (recovery resistance) has also been suggested as another mechanism of resistance [7]. In this study we have measured the degree of resistance of sorghum genotypes to shoot fly in terms of numbers of plants with eggs and percentage of dead hearts. The other traits (leaf glossiness, seedling vigour and trichome density) associated with non-preference were also assessed in all the genotypes in two diverse screening environments.

Response of parental lines and their hybrids to shoot fly

Shoot fly resistant CMS, maintainer, and restorer lines were glossy in nature and recorded glossiness score of 1.69 to 2.10, while the shoot fly susceptible CMS, maintainer, and restorer lines were non-glossy (Score 3.37 to 3.66) (Table 2). Parental performance and hybrid mean performance for leaf glossiness was consistent across both the screening environments with few exceptions, indicating consistent and reliable evaluation of the trait. Jayanthi *et al.* [8] also reported stable expression of glossiness across seasons. For seedling vigour, lack of stability in the expression across test environments revealed influence of environmental factors on this trait. Even though means of crosses suggested consistent performance in both the environments, there were very few hybrids exhibiting consistent vigour score in these screening environments (data not shown). This implies that screening environments [9] had a definite role. For seedling vigour scores all shoot fly resistant (CMS, maintainers and restorer) lines exhibited better scores (2.62 to 2.83) than susceptible parents (3.17 to 4.00). The hybrids derived from the resistant (R × R) parents revealed good vigour (2.99) as against other cross combinations. The lines with only few trichomes on upper surface are termed

trichomeless. Lines with typical trichomes on both the surfaces of the leaf are recognized as trichomed lines. Trichome density was greater in early post-rainy screening environment than in late rainy season indicating that the character was under genetic control with some environmental influence in expression in different seasons [10]. The shoot fly susceptible parents had very less number of trichomes at abaxial surface (1.91 to 14.91) whereas, resistant parents recorded high trichome density (37.45 to 52.94). Hybrids between resistant CMS lines (SFRA) and testers (SFRR) had higher trichome count on both the surfaces of leaf lamina (90.10 and 38.60) as compared to SFSA × SFRR (59.36 and 23.39) and SFSA × SFRR (22.79 and 8.72) combinations. Across the environments, trichome density on adaxial surface was greater than that on abaxial surface. Among the checks, IS 18551 a shoot fly resistant land race exhibited superior performance for all the resistance governing traits across the environments. None of the parents and hybrids showed superiority over IS 18551. However, R × R hybrids

revealed enhanced resistance as compared to the commercial hybrid CSH 16. This supported the earlier observations [5].

The CMS lines were preferred for oviposition and recorded 63.45 per cent plants with eggs and 60.06 per cent dead hearts at 28 DAE than the maintainer lines (61.48 per cent plants with eggs and 55.53 per cent dead hearts), while the difference among the shoot fly resistant CMS and maintainer lines was not perceptible (Table 2). Interestingly the hybrids in general recorded high oviposition and dead hearts as compared to their parental lines. The most direct measure of shoot fly damage is that recorded in terms of dead heart incidence (%). Therefore, the variability observed for this trait in breeding material is of prime importance for selecting resistant and/or tolerant genotypes. The shoot fly damage in sorghum occurs up to four weeks after seedling emergence [4]. Therefore, the data on relative resistance/susceptibility of CMS, maintainer and restorer lines were recorded at 21 and 28 DAE on dead heart

Table 2. Across seasons summary of reaction of shoot fly resistant and susceptible parental lines and their hybrids towards shoot fly and their morphological traits associated with resistance

Parents and hybrid combinations	Morphological traits				Response to shoot fly			
	Leaf glossiness*	Seedling vigour#	Trichome density		Oviposition (%)		Dead hearts (%)	
			(Adaxial)	(Abaxial)	(14DAE)	(21DAE)	(21 DAE)	(28 DAE)
A lines								
Susceptible	3.58	4.00	21.08	8.12	40.38	63.45	43.86	60.06
Resistant	1.75	2.62	84.29	37.45	16.50	35.20	16.60	31.26
B lines								
Susceptible	3.37	3.59	30.92	14.91	38.75	61.48	40.34	55.53
Resistant	1.69	2.63	81.54	44.66	15.94	32.75	15.25	28.93
Restorers								
Susceptible	3.66	3.17	5.63	1.91	42.63	63.74	42.21	59.34
Resistant	2.10	2.83	134.34	52.94	17.36	33.22	18.16	32.06
Hybrids								
SFSA × SFRR [®]	3.98	3.32	22.79	8.72	43.92	65.28	45.35	64.28
SFSA × SFRR [®]	3.21	3.14	59.36	23.39	37.05	56.93	37.17	52.74
SFRA × SFRR [®]	3.61	3.20	39.04	11.78	38.87	60.37	39.17	55.59
SFRA × SFRR [®]	2.55	2.99	90.10	38.60	30.52	49.67	30.58	43.91
Checks								
PVK 801	4.19	3.25	0.00	0.00	57.46	67.92	53.26	64.53
CSH 16	3.63	3.19	13.75	6.67	44.29	62.95	46.61	55.81
IS 18551	1.07	2.32	157.91	65.67	8.87	27.73	10.85	21.05
LSD (P=0.05)	0.26	0.34	4.16	3.29	2.58	3.44	3.25	3.39

*Leaf glossiness score (1= high glossiness, and 5= non-glossy), #Seedling vigour score (1= high vigour, and 5= poor), [®] Hybrid combinations (SFSA = Shoot fly susceptible A-lines, SFRA = Shoot fly resistant A-lines, SFRR = Shoot fly susceptible restorer, SFRR = Shoot fly resistant restorer), DAE= Days after emergence.

percentage. SFRA × SFRR crosses in general gave lower percentage of oviposition (49.67) and dead hearts (43.91) as compared to other crosses. Dead heart formation due to shoot fly damage was high during the early post-rainy season as compared to the late rainy season (data not shown).

The CMS and maintainer pairs of SPSFR 94010A and MS 104A showed good resistant reaction to shoot fly, while such pairs of PMS 7A, PMS 8A and PMS 28A were susceptible to shoot fly across observations intervals and environments. This was mainly because of the fact that resistance to sorghum shoot fly is largely based on oviposition non-preference [4]. The restorer lines SFCR 125, SFCR 151, ICSV 700, ICSV 705, ICSV 708 and PS 30710 showed resistant reaction to sorghum shoot fly, while KR 192, KR196, KR 199, CS 3541, MR750, C 43 and ICSV 91011 were susceptible across observation intervals and environments. Similar differences in the reaction of resistant and susceptible lines for dead heart formation have been reported in earlier studies [11].

The superiority of parents like SPSFR 94010A, MS104A, SFCR 125, SFCR151, ICSV 705, ICSV 708 and PS30710 for the dead heart percentage and other characters was associated with shoot fly resistance. Similarly, the hybrids like SPSFR 94010A × SFCR 125, MS 104 × PS 30710, PMS 8A × SFCR 125, SPSFR 94012A × ICSV 91011, MS 104 × ICSV 705, SPSFR 94010A × ICSV 705 and MS 104A × ICSV 705 performed better for the resistance attributing traits. These parents in the hybrid combinations can generate useful population and shoot fly resistant progenies.

In this study, the CMS lines were more susceptible to shoot fly than the maintainer lines indicating that the resistance/susceptibility to shoot fly was influenced by the factor associated with cytoplasmic male sterility, but there were few exceptions. The resistance/susceptibility

is influenced not only by the cytoplasm, but nuclear genes are also involved in the expression of this reaction. Discrimination of such interaction from cytoplasmic effects is very complicated. Higher susceptibility of CMS lines than their corresponding maintainer lines to sorghum shoot fly have also been reported earlier [5]. This also suggested that maintainer lines possess the factors that influence expression of resistance to this pest [12]. Similar results on the influence of CMS on genotypic susceptibility to pests have been reported for bacterial leaf blight (*Xanthomonas oryzae* pv. *oryzae*) resistance in rice hybrids [13]. Hence diversification of the CMS systems is required to combat the shoot fly incidence. Among the various cytoplasm backgrounds in sorghum the A₄M cytoplasm is less susceptible to shoot fly and also possesses the better recovery resistance resulting in production of few synchronous tillers on the main shoot which grow rapidly and most of which survive to produce harvestable earheads. This source can be exploited for developing resistant hybrids. Another avenue for resistance to this pest is the utilization of wild relatives of sorghum. Wild relatives belonging to sections *Parasorghum*, *Stiposorghum* and *Heterosorghum* can be used as sources of alternate genes to increase the resistance levels and diversify the basis of resistance to shoot fly [14].

Correlation among resistance attributing traits and their contribution to shoot fly resistance

Shoot fly resistance is reflected by the oviposition behavior and dead heart formation. We studied the correlation among different shoot fly resistance attributing traits. Resistance to shoot fly showed highly significant correlation with the shoot fly resistance attributing traits with $P = 0.01$ (Table 3). Correlations between oviposition and leaf glossiness ($r = 0.77$ to 0.78) and seedling vigour ($r = 0.39$ to 0.43) were positive, while these with trichome density at adaxial ($r = -0.74$

Table 3. Correlation between shoot fly resistance traits, oviposition and dead heart percentage

	Oviposition (14 DAE)	Oviposition (21 DAE)	Dead hearts (21 DAE)	Dead hearts (28 DAE)
Leaf glossiness	0.78**	0.77**	0.79**	0.79**
Seedling vigour	0.43**	0.39**	0.47**	0.49**
Trichome density (Adaxial)	-0.74**	-0.75**	-0.68**	-0.74**
Trichome density (Abaxial)	-0.75**	-0.71**	-0.67**	-0.73**

** Significant at $P=0.01$

to -0.75) and abaxial surface of leaf lamina ($r = -0.71$ to -0.75) were negative. Dead hearts also showed high correlation with leaf glossiness ($r = 0.78$ to 0.79), while negative correlation with trichome density (adaxial and abaxial leaf surface) ($r = -0.67$ to -0.74). Though seedling vigour exhibited positive correlation with both oviposition ($r = 0.39$ to 0.43) and dead heart ($r = 0.47$ to 0.49) but the magnitude was low as compared to other attributing traits. Genotypic correlation confirmed that the number of trichomes on both surfaces of lamina and leaf glossiness contributed resistance for shoot fly. Weaker association of seedling vigour with oviposition and dead heart percentage has been also reported earlier [15]. Therefore, magnitude of resistance by two or more resistance characters together is higher than the magnitude of resistance by the single trait alone.

Seedling vigour did not show any significant contribution towards dead heart (%) when considered along with glossiness and trichome density (data not shown). However, when the parental lines and hybrids were grouped in to combinations on the basis of the presence or absence of both the glossiness and trichomes significant difference between the groups were recorded (Fig. 1). The results revealed that the presence of glossy and the trichome traits have independent and apparently additive effects in reducing the dead hearts under shoot fly pressure. In case of parental lines and hybrids as well, glossiness alone exhibited enhanced resistance to shoot fly as compared

to trichome density alone. However, combination of these two traits exhibited significant reduction in dead hearts (Fig.1).

It can be concluded that through hybrid breeding it is possible to develop shoot fly resistant hybrids. Higher susceptibility of CMS lines to shoot fly as compared to the maintainers suggested the influence of male sterile cytoplasm towards susceptibility to shoot fly. Season specificity was observed in the expression of traits associated with resistance, particularly seedling vigour and trichome density. Glossy trait revealed strong association with shoot fly resistance and this is of immense importance for identifying shoot fly resistant genotypes in preliminary screening of germplasm and breeding populations. However, incorporation of glossiness and trichome traits into sorghum breeding lines can further improve field resistance to shoot fly. Thus, it may be concluded that breeding shoot fly resistance hybrids is feasible by ensuring resistance in both the parents. Alternate sources like non *milo* cytoplasm and wild relatives can be utilized for improving the levels and basis of resistance for shoot fly.

Acknowledgements

The authors wish to thank Dr. H.C. Sharma, Principal Scientist, International Crop Research Institute for Semi-arid Tropics, Patancheru, India for providing parental line seeds and valuable suggestions for the study.

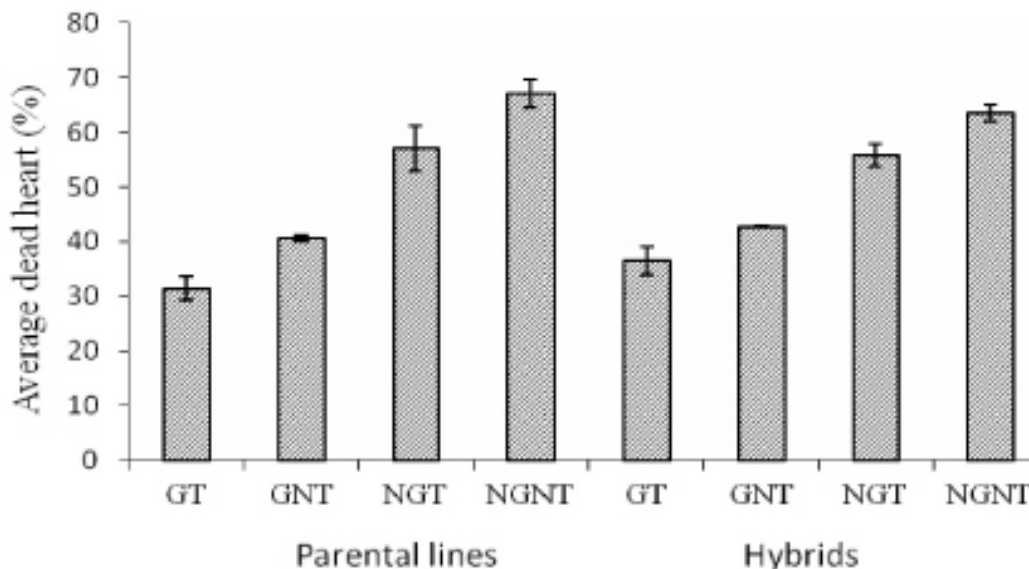


Fig. 1. Response of significant trait combinations to shoot fly among parental lines and hybrids. (GT: Glossy-Trichomed; GNT: Glossy-Trichomeless; NGT: Non glossy- Trichomed; NGNT: Non glossy- Trichomeless)

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