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Research Article:

Performance of sorghum-maize interspecific derivatives for forage attributes

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SUMMARY : Over decades of crop improvement sorghum has led to yield plateau with persisting problems of biotic (shoot fly, stemborer, grain mold) and abiotic (drought, heat) causing yield losses in marginal conditions where the crop is grown. An attempt was made to cross the superior genotypes from sorghum and maize genera and study the derivatives after stabilizing through advancing and selection for several years. The derivatives resembled sorghum though anthocyanin pigmentation was observed in early generations. The derivatives with good forage traits were tested across two locations, Pantnagar and Hisar in India. Two genotypes were found promising for high biomass and quality traits (T2-2251-3-12) and high biomass, high tillering and low HCN (T3-2252-7-1). These can be utilized as new sources of variability in forage sorghum improvement programs.

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<u>KEY WORDS:</u> Intergeneric hybrids, Maize, Sorghum, Forage, HCN

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BACKGROUND AND OBJECTIVES

Sorghum occupies a prominent place among the food grains securing the food and nutritional security of the poor people residing in the semi-arid tropics and contributing a vital share of energy, proteins, vitamins and minerals (Sanjana Reddy 2017). While, it a staple food crop in the semi-arid areas of Asia and Africa, it is used as a poultry and animal feed in Americas, Australia and China. In India, the rainy season grown crop is utilized for poultry and animal use while the post rainy season grown crop is used for human consumption. The cultivars grown in both the seasons vary widely (Rana *et al.* 1997). The crop residue (stover) left after harvest is a valuable fodder resource in the areas of its cultivation. With the advent of hybrid technology and introduction of temperate germplasm, significant advances were made in rainy season grown sorghum. Significant advances in crop improvement of sorghum have been achieved in realizing the grain yield through utilizing variability within the gene pool of cultivated sorghum, S. bicolor. However, much progress could not be made in achieving tolerance to shoot fly, stemborer and grain mold among biotic constraints while abiotic constraints pose challenge. A plateau has been reached in realizing yield gains and further improvement required introduction of variation through other different means like wide hybridization. Wide hybridization through crossing between individuals belonging to two different genera has been attempted to create, establish variation and develop pre- breeding material for ready use in plant breeding programs. Intergeneric hybridization can widen the available genetic pools so that novel genetic variation can be utilized by plant breeders. For traits that are quantitatively inherited, introgression provides the most logical and effective approach to gene transfer, assuming that interspecific or intergeneric hybridization can be achieved (Hodnett et al., 2010). Intergeneric hybrids were attempted between maize - sorghum, sugarcane - sorghum and rice - sorghum (Jun and Qingguo 1994). Differential expression profiling of sucrose-related genes in intergeneric hybrids involving sugarcane and sorghum was reported by Ramalashmi et al. (2014). With an objective of realizing new variability, the promising sorghum lines were crossed with maize lines at IIMR and the resultant segregates showing forage attributes were advanced with selection. The segregants were studied with an objective to determine their performance for forage traits vis a vis standard checks as well as to determine the association of the traits with forage yield.

RESOURCES AND METHODS

The plant material used in the study included four inbred lines of maize (CM118, CM130, CM208, and CM211) and nine cytoplasmic male sterile (CMS) lines of sorghum. Maize lines were obtained from Maize Research Station, Professor Jayashankar Telangana State Agricultural University (PJTSAU), Hyderabad, India. Nine male sterile lines of sorghum, that included four lines in A1 CMS background (27A, 126A, 296A, 356A), three lines in A_2 CMS background (C43 A_2 , MR750A₂, 7A₂), one line each in A₃ and A₄ CMS background (8A₃ and 9A₄), were obtained from Germplam Unit, Indian Institute of Millets Research (former Directorate of Sorghum Research), Hyderabad, India. During the 2008 post rainy season, all the sorghum CMS lines were sown at 15d intervals in pots for synchronization of flowering with maize inbred lines. After emerging from the boot leaf and 1-2 days before opening of the florets, the sorghum panicles were covered with paper bags to avoid any cross pollination. Each experiment was carried out on flower heads which were protected from foreign pollen by bagging. Initially, pollen

germination of maize lines was standardized using four different pollen germination media (PGM1-PGM4). Percentage of pollen grains germinated and length of the pollen tube were examined under microscope to optimize the best pollen germination medium. Pollen of maize was collected in a petriplate through dusting of florets or through gentle squash of mature anthers. It was immediately carried to recipient sorghum plant and 1-2 drops of PGM-2 was added to the pollen grains in the petriplate and was gently mixed with a soft brush. Paper bag on the recipient sorghum plant was removed and immediately the pre-germinated pollen was placed on the stigma of sorghum florets with a soft paint brush. Sorghum panicle was covered immediately with the same paper bag. The above process was initiated one or two days before the sorghum florets were open and the stigma emerged and repeated for 5-6 days on every plant with the pollen from the same maize line. Each male sterile sorghum line was repeatedly pollinated with pregerminated pollen from each of the four inbred lines of maize, CM118, CM130, CM208, and CM211. Seeds were allowed mature on the sorghum plant and F₁ seeds were harvested. In some cases the seeds were shriveled and embedded deep in the glumes. These were carefully taken out with hand. The F₁ seeds were germinated in paper cups filled with soil and vermiculate mixed in the ratio of 3:1. They were grown in the laboratory until three leaf stage and were transferred to pots. For initial 5-6 days, these plants were grown under shade and later transferred to direct sunlight in pots. The F₁ seeds from 36 crosses, designated as SM1-SM36 were raised. Panicles in F₁ plants were covered with paper bags to allow self-pollination and F₂ seeds were harvested at maturity. The F₂ plants were grown in field and promising progenies for diverse morphological traits were selected and advanced. Similarly, 227 lines were selected in F₃ generation. The 227 F_4 progenies were evaluated during two crop seasons, the rainy and the post-rainy seasons, in augmented design along with elite genotypes as checks. The sweet sorghum variety, SSV84 was used as check for comparison of sweet sorghum traits; the multicut forage sorghum variety SSG 59-3 was used as check for evaluation of forage traits, the early flowering genotype, 2219B was used to evaluate and select for earliness and the popular post rainy sorghum variety, M35-1 was used as check to compare the morphological grain quality traits. The parental lines (27A₁, 126A₁, C43A₂ and MR750A₂), the parents of 227 selected lines were

also planted for comparison. Observations were recorded on the traits such as days to flower, plant height, tiller number, panicle length, panicle width, brix content in stalk juice.

Among the 227 lines tested, 25 lines showed forage traits such as multiple tillers, narrow leaves and loose panicles. Number of tillers ranged from 3-16 per plant. Sixteen of them were found to be promising. They included 14 lines from SM12 ($126 \times CM208$), one line each from SM27 (MR750 $\times CM208$) and SM6 ($27A \times CM211$). The 16 forage varieties along with two checks (SSG 59-3 and CSV 21F) were evaluated in a three replicated RBD design across two locations, Pantnagar and Hisar in India.

Data were recorded for the traits green fodder yield for 2 cuts (GFY 2 cuts), dry fodder yield for 2 cuts (DFY 2 cuts), percentage total soluble sugars (TSS%), ppm HCN, percentage of protein, percentage invitro dry matter digestibility (IVDMD%), early vigor (on a scale of 1 to 4, where 4= good and 1=poor), plant height in cm, number of leaves per plant, number of tillers per plant, leaf length in cm, leaf breadth in cm, leaf to stem ratio and stem girth in cm. Analysis of variance for randomized complete blocks design was carried out using Indostat and trait associations were generated using Microsoft Excel.

OBSERVATIONS AND ANALYSIS

The results obtained from the present study as well as discussions have been summarized under following heads :

Mean performance :

The analysis of variance showed significant genetic variability for green fodder yield, dry fodder yield, HCN content and early vigor. The green fodder yield per plant ranged from 218.7q/ha (T13-2276-14-1) to 692.2q/ha (T2-2251-3-12). Except two genotypes, all of them were on par with the best check T17-CSV22F (576q/ha) while seven genotypes were numerically superior. The dry fodder yield per plant ranged from 43.6 q/ha (T13-2276-14-1) to 166.5q/ha (T18-2222-8-8). Except two genotypes, all of them were on par with the best check T17-CSV22F (139.7q/ha) while four genotypes were numerically superior. The HCN content ranged from

Table 1 : Forage yield, quality and phenological traits of sorghum-maize intergeneric derivatives													
Sr. No.	Entry	GFY 2 cuts q/ha)	Rank	DFY 2 cuts (q/ha)	Rank	T SS (%)	Rank	HCN (ppm)	Rank	Protein (%)	Rank	IVDMD (%)	Rank
1.	T1-2251-3-8	275.5	17	54.2	17	3.75	7	68.2	7	8.03	18	55.6	8
2.	T2-2251-3-12	692.2	1	160	2	5.75	1	64.3	8	8.8	2	56.9	3
3.	T3-2252-7-1	617.3	4	127.2	8	5.25	2	54.6	17	8.37	10	53	15
4.	T4-2252-7-2	570.8	10	120.9	12	4.75	4	79.5	2	8.3	13	57.2	1
5.	T5-2252-7-8	671.5	2	141.3	4	5	3	56.6	15	8.8	1	55.8	7
6.	T6-2253-12-2	603.4	5	125.7	9	5	3	55.7	16	8.4	9	56.3	6
7.	T7-2253-12-6	494.7	15	112.4	14	4.75	4	58.4	12	8.57	5	56.4	5
8.	T8-2253-12-16	585.9	7	131.6	6	4.5	5	63.6	9	8.25	15	52.8	16
9.	T9-2254-8	527	14	111.7	15	4.5	5	54.1	18	8.26	14	57	2
10.	T10-2254-10-3	555.2	11	124	10	4	6	57.2	14	8.49	7	54.8	9
11.	T11-2254-10-8	453.6	16	104.5	16	4.5	5	73.2	5	8.24	16	53	13
12.	T12-2254-10-12	554.6	12	119.3	13	5.75	1	74.3	4	8.77	3	54.6	10
13.	T13-2276-14-1	218.7	18	43.6	18	4.75	4	86.7	1	8.05	17	56.7	4
14.	T14-2316-3-3	602	6	149.4	3	5	3	75.2	3	8.51	6	53.7	12
15.	T15-2316-3-7	530.1	13	121.4	11	4.75	4	57.7	13	8.34	11	54	11
16.	T16-SSG59-3	571.3	9	128.3	7	5	3	71.3	6	8.67	4	53	14
17.	T17-CSV22F	576	8	139.7	5	4.5	5	60.6	11	8.33	12	52.3	17
18.	T18-2222-8-8	661.9	3	166.5	1	4	6	63.3	10	8.45	8	52.2	18
	LOC. Mean	542.3		121.2		4.75		65.3		8.42		54.7	
	C.D. (P=0.05)	185.6		53.4		2.21		8.2		0.71		5.4	
	C.D. (P=0.01)	255		73.3		3.04		11.3		0.97		7.4	



86.7ppm (T13-2276-14-1) to 54.1ppm ((T9-2254-8). Compared to the best check for low HCN content (CSV 22F: 60.6ppm), eleven genotypes were on par while seven genotypes had numerically low HCN content. The second check SSG 59-3 had significantly higher HCN (%) (71.3ppm) compared to CSV 22F. The vigor score ranged from 1 to 4. Seven genotypes had significantly superior vigor score as compared to the best check SSG 59-3

Table :																
Sr. No.	Early vigour	Rank	Plant height (cm)	Rank	No. of leaves/ plant	Rank	No. of tillers/ plant	Rank	Leaf length (cm)	Rank	Leaf breadth	Rank	Leaf: stem ratio	Rank	Stem girth (cm)	Rank
1.	3.25	4	163	17	16.7	16	2.45	14	74.9	17	6	1	0.43	2	4.95	13
2.	4	1	220	1	18.2	14	3	12	77.2	16	5.7	3	0.34	14	6.83	1
3.	4	1	202	10	24.9	1	5.08	1	80.4	10	3.85	18	0.4	8	6.25	4
4.	3.75	2	193	15	15.8	18	4.33	3	89.4	2	4.25	15	0.41	4	5.55	9
5.	4	1	204	9	23	2	4.33	3	79.8	13	4.95	9	0.38	10	5.13	12
6.	4	1	215	3	19.4	10	3.18	10	89.4	1	4.3	14	0.42	3	6.25	4
7.	3.5	3	215	4	19	12	3.6	7	79.7	14	4.4	13	0.44	1	5.13	12
8.	4	1	201	11	20.7	5	3.5	8	79.5	15	4.55	12	0.36	11	5.75	7
9.	4	1	201	12	21.9	4	3.65	6	81.5	9	5.15	6	0.38	9	5.33	11
10.	3.75	2	192	16	19.9	6	3.75	5	80.3	11	4.2	16	0.38	10	5.5	10
11.	3.25	4	195	13	16.6	17	2.5	13	84.5	5	4.88	10	0.33	15	6.23	5
12.	3.75	2	211	7	19.7	7	3.18	10	88	3	5.45	4	0.32	16	6.3	3
13.	2.25	5	130	18	19.2	11	3.15	11	64.5	18	5.08	7	0.38	9	5.63	8
14.	3.75	2	212	5	22.1	3	4.43	2	82.1	8	5	8	0.41	5	6.23	5
15.	3.75	2	212	6	19.4	9	3.33	9	83.9	6	4.7	11	0.36	12	4.95	14
16.	3.75	2	194	14	17.5	15	3.83	4	80	12	4.15	17	0.4	7	4.53	15
17.	3.25	4	210	8	18.2	13	2.25	15	85.9	4	5.85	2	0.35	13	6.65	2
18.	4	1	218	2	19.5	8	3.6	7	83.8	7	5.2	5	0.41	6	6.08	6
LOC. Mean	3.67		199		19.5		3.51		81.4		4.87		0.38		5.73	
C.D.(P=0.05)	0.73		48		7		1.57		11.8		1.32		0.11		1.85	
C.D.(P=0.01)	1.01		65		9.7		2.16		16.3		1.81		0.16		2.54	
C.V. (%)	9.48		11.3		17.1		21.25		6.9		12.83		14.29		15.31	
F (Probability)	0.01		0.1		0.51		0.09		0.06		0.08	-	0.63	-	0.43	

Table 2 : Trait associations among the sorghum-maize intergeneric derivatives for forage yield and quality attributes													
	GFY 2 cuts (q/ha)	DFY 2 cut s (q/ha)	T SS (%)	HCN (ppm)	Protein	IVDMD (%)	Early vigour	Plant height (cm)	No. of leaves/plant	No. of tillers/plant	Leaf length (cm)	Leaf breadth	Leaf: stem ratio
DFY 2 cuts (q/ha)	0.98**	1.00											
T SS (%)	-0.26	-0.21	1.00										
HCN (ppm)	0.69**	0.62**	-0.21	1.00									
Protein (%)	0.80**	0.74**	-0.01	0.91**	1.00								
IVDMD(%)	0.81**	0.73**	-0.22	0.94**	0.97**	1.00							
Early vigour	0.17	0.19	0.88**	0.14	0.41	0.20	1.00						
Plant height (cm)	0.94**	0.89**	-0.26	0.84**	0.92**	0.94**	0.18	1.00					
No. of leaves/plant	0.72**	0.69**	0.23	0.70**	0.87**	0.80**	0.58**	0.78**	1.00				
No. of tillers/plant	-0.28	-0.23	0.98**	-0.26	-0.05	-0.27	0.87**	-0.31	0.22	1.00			
Leaf length (cm)	0.87**	0.81**	-0.23	0.91**	0.96**	0.98**	0.21	0.98**	0.79**	-0.27	1.00		
Leaf breadth	0.00	0.04	0.89**	0.16	0.35	0.16	0.93**	0.08	0.50*	0.84**	0.13	1.00	
Leaf : stem ratio	-0.48*	-0.42	0.96**	-0.43	-0.25	-0.45	0.76**	-0.49*	-0.02	0.96**	-0.46	0.78**	1.00
Stem girth (cm)	0.03	0.08	0.92**	0.11	0.32	0.11	0.95**	0.07	0.50*	0.88**	0.11	0.96**	0.80**

Agric. Update, **12** (TECHSEAR-9) 2017 :2346-2351 Hind Agricultural Research and Training Institute (score:2). Overall non-significant differences were observed for other traits implying that the intergeneric derivatives were on par with the checks for the remaining traits. The total soluble sugars ranged from 3.75 to 5.75%. As compared to the best check, SSG 59-3 (5%), two genotypes were numerically superior with 5.75% (T2-2251-3-12, T12-2254-10-12). The protein content ranged from 8.03 to 8.8%. As compared to SSG 59-3 (8.67%), the genotypes T2-2251-3-12 (8.8%), T5-2252-7-8 (8.8%) and T12-2254-10-12 (8.77%) were numerically superior. The invitro dry matter digestibility ranged from 52.2 to 57.2%. Compared to SSG 59-3 (53%), 11 genotypes were numerically superior. The plant height ranged from 130 to 220cm. Compared to the best check CSV22F (210 cm), seven genotypes were numerically superior. The number of leaves per plant ranged from 16 to 25. Compared to the checks with 18 leaves per plant, 12 genotypes were numerically superior. The number of tillers ranged from 2 to 5. Compared to the best high tillering check, SSG 59-3, 4 lines had numerically greater number of tillers. Leaf length ranged from 64-5 cm to 89.4 cm. Compared to the best check, three lines had greater leaf length. Leaf breadth ranged from 3.85 to 6 cm. The line T1-2251-3-8 had more leaf width compared to best check. Leaf to stem ratio ranged from 0.32 to 0.44. Six genotypes were numerically superior (0.41 to 0.44) compared to best check. The stem girth ranged from 4.53 to 6.83 cm. The genotype T2-2251-3-12 (6.83cm) was numerically superior to the best check CSV 22F (6.65 cm). Among all the evaluated genotypes, T2-2251-3-12 was the best performing with high biomass, total soluble sugars, protein content, IVDMD, early vigour and plant height as compared to the best checks. The genotype T3-2252-7-1 was the best performing for biomass, low HCN content, number of leaves per plant and number of tillers per plant. These two genotypes can be further exploited in plant breeding programs.

The superior performance of the intergeneric crosseshas been recorded in earlier studies. The rice and sorghum cross possessed better canopy spatial architecture, higher photosynthetic rate and better tolerance to adverse conditions and higher field yields even under stress conditions (Tang *et al.* 2002). Interspecific hybridization was observed between *S. halepense* \times *S. bicolor* and *S. sudanense* \times *S. bicolor*. In these crosses, there were significant phenotypic variations on vegetative traits

with F_1 progenies having profuse tillering and branching than the parental populations (Magomere, 2014).

Trait associations :

Dry fodder yield increased with increase with green fodder yield and both were significantly and positively associated with protein content, IVDMD, HCN, plant height, number of leaves per plant, leaf length. Green fodder yield was significantly and negatively associated with leaf to stem ratio. Hence, with an increase in plant height, number of leaves and leaf length, biomass can be increased. This in turn will lead to an increase in protein content and IVDMD. However, HCN content also increased significantly (Table 2). Hence, while intro gressing these genotypes for incorporation of new genetic variability, disruptive selection should be made for low HCN content.

Conclusions :

Sorghum-maize intergeneric derivatives were tested for the first timein sorghum history for evaluating and identifying new variability for forage sorghum improvement. Two genotypes were found promising for high biomass and quality traits (T2-2251-3-12) and high biomass, high tillering and low HCN (T3-2252-7-1). High biomass was positively related to forage quality traits and can be targeted for simultaneous selection while the linkage with high HCN needs to be broken with disruptive selection.

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