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Multi-environment Assessment of Combining Ability, Heterosis of *Bt* Introgressed Lines and Hybrids of Upland Cotton (*Gossypium hirsutum* L.)

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ABSTRACT

A study was conducted at Dharwad, Raichur and Aurangabad in India during the rainy season of 2018 to investigate the environmental impact on combining ability and heterosis estimates in cotton. Combining ability analysis across locations showed significant effects on the performance of genotypes, GCA, SCA, genotype×environment, GCA×environment and SCA×environment interactions for most of the characters. Combining ability variance revealed the predominance of non-additive gene actions for characters under study. The parents and crosses varied with locations for combining ability and better parent heterosis estimates. Out of 22 parents, ten parents recorded high overall *gca* and among them, two BGII, six BGI and two non-*Bt* parents were under study. Similarly, 55 hybrids were exhibited high overall *sca* and better parent heterosis from the study and most of the crosses were derived from lines crossed with high overall *gca* (H) with low overall *gca* (L) or vice versa. This study indicated the requirement of parents with contrasting *gca* effects to realize a higher frequency of heterotic hybrids. The predominance of H×L type of crosses indicated the presence of non-additive gene action suggesting exploitation of good heterotic hybrids from the study. The frequency of hybrids with high overall *sca* and heterobeltiosis was more in BG II hemizygous hybrids.

KEYWORDS: Bt cotton, introgression, combining ability, heterosis

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1. INTRODUCTION

 B^t cotton hybrids were released in India during 2002 BGI (MON531) and BGII (MON15985) in 2008 ensuring protection from American bollworms. Since then most private companies are engaged in developing Bt cotton pure lines and hybrids. Before the introduction of Bt cotton, bollworms were a major cotton pest with 80% of yield damage and required a huge amount of pesticides to control them. (Nagrare et al., 2014). Hybrid cotton cultivation is a major source of Indian production (Anonymous, 2021). Soon after the introduction of Bt cotton, productivity increased from 207 kgs ha-1 (2001-2002) to 456 kg ha⁻¹ in 2012 (Suresh et al., 2014). However, last decade productivity stagnation is observed which is presumed due to the narrow genetic base of germplasm in hybrids. The majority of the studies published on combining ability and heterosis have used non-transgenic cotton lines and hybrids that are popularly called non-Bt (Pushpam et al., 2015, Krithika et al., 2020 and Suryanaik et al., 2020). There are negligible studies published using BGI and BGII lines. Detailed investigation of such Bt lines is important in developing high yielding Bt cotton hybrids. Estimation of combining ability or genetic effects is an important strategy to sort out superior and inferior parents coupled with better crosses. The general combing ability (GCA) is defined as the average performance of a genotype in a series of cross combinations. The specific combining ability (SCA) is the average performance of a specific cross combination expressed as deviation from the population mean. Thus SCA is important for hybrid development whereas GCA is useful in the identification of potential parental genotypes for hybridization. The general combining ability refers to additive gene action and is used to determine the performance of parents in general terms while the specific combining ability effect is determined by additional dominant gene action and is used to indicate the hybrid performance in specific terms. Breeders use these variance components to determine the gene action and to assess the genetic potentials of the parent in hybrid combinations (Sprague and Tatum, 1942). The estimates of gca and sca are usually used for determining the potential of parents and their hybrids for the improvement of crop production. So parent's selection for hybridization can be done because of combining ability

and per se performance. However, the effects of gca and sca are more informative and reliable than per se values as they also provide information about the type of gene action; that is, additive or non-additive (Anjum et al., 2018, Richika et al., 2021 and Gnanasekaran and Thiyagu, 2021). The primary objective of the breeding programme is to improve seed cotton yield that has low heritability which is highly affected by genotype×environment interaction. Identification of stable parental material to develop superior-stable yielding hybrids is important (Chao-zhu et al., 2007, Anandan, 2010, Patil et al., 2017, Bhandari et al., 2021). The cotton yield traits were mainly controlled by genetic and environment interaction effects, whereas ginning out turn and fiber quality traits were mostly determined by main genetic effects (Shahzad et al., 2019). It will enable the breeder to evaluate a larger number of hybrid parents, shorten the testing structure of the breeding program through an initial accurate selection of optimal combinations and possibly reduce the cost of trial evaluation and combination testing. In this study involving multi-location testing, we made effort to identify cotton lines with the combination of characters and develop a high frequency of heterotic hybrids in Bt introgressed lines of cotton to know the overall performance of the hybrid across three locations.

2. MATERIALS AND METHODS

2.1. Plant material

The present investigation was carried out in three research locations of Indo-American Hybrid Seeds (I) Pvt. Ltd., Dharwad and Raichur (Karnataka) and Aurangabad (Maharastra) India during the rainy season 2018. The experimental material comprises 15 lines and seven testers (Table 1) and that was crossed in line×tester fashion to develop 105 hybrids. All parental lines were proprietary genetic material belonging to Indo-American Hybrid Seeds (I) Pvt. Ltd (IAHS). A total of 105 hybrids were synthesized that comprised, 69 BGII, 34 BGI and two non-Bt hybrids. These hybrids were planted replicated trial along with parents and checks. The plot size was maintained by two rows of ten dibbles spacing of 90×90 cm² with a single plant per dibble. Observations on days to flowering (DOF), days to 50% flowering (DFF) were taken along with seed cotton yield plot⁻¹ (later converted in kg ha⁻¹; SCY), average boll

Table 1: Description of parents and hybrids used for the study									
	Lines		Testers						
BGII	BGI	Non-Bt	BGII	BGI	Non-Bt				
IAC7-1, IAC7-4, IAC7-5, IAC7-7, IAC7-8, IAC7-9, IAC7-10, IAC7-13, IAC7-14	IAC7-2, IAC7-3, IAC7-11, IAC7-12,	IAC7-6 and IAC7-15	IAC7-17	IAC7-16, IAC7-19, IAC7-21, IAC7-20, IAC7-22	IAC7-18				

weight of 20 randomly picked bolls (g; BW), seed index (100 seed weight in g; SI) and lint percentage (LP). Further plant growth characters like plant height (cm; PH), the number of sympodia (SYM), monopodia (MONO) and the number of bolls per plant (NOB) were recorded in five randomly selected plants in each genotype in all replications.

2.2. Statistical analysis

The pooled data was recorded from three locations subjected to Analysis of Variance (ANOVA) and Line×Tester analysis using Analysis of Genetic Designs with R for Windows (AGD-R) software (Rodriguez et al., 2015). Line ×Tester Analysis was used to compute the *gca* and *sca* effects following the procedure of Kempthorne (1957). Data of F_1 hybrids were subjected to combining ability analysis following line×tester linear model (Kempthorne, 1957).

As quantitative traits are correlated either positively or negatively, it is usual to find, for a particular parent and a hybrid, gca, sca effects and better parent heterosis (BPH) respectively in the desirable direction for some characters and the undesirable direction for others. Hence, the overall status of parents concerning their gca effects and the hybrids for their sca effects and BPH across ten characters were determined (Arunachalam and Bandopadhyay, 1979; Anilkumar and Lohithaswa, 2018). As per the procedure suggested by Arunachalam and Bandopadhyay (1979), the determination of the overall status of parents for their gca effects and the hybrids for their sca effects and BPH across all characters should be based on only significant gca, sca and heterotic effects. The consideration of only significant gca, sca and heterotic effects results in loss of information on several parents and crosses. To overcome such shortcoming, we considered the estimates of gca, sca and heterotic effects irrespective of their statistical significance. The estimates of gca effects of parents, sca effects and BPH of hybrids were ranked by assigning the lowest rank for the parent or the cross which manifested the highest gca or sca effects and BPH, respectively in the desirable direction. The highest rank was assigned for the parent or the cross which manifested the lowest gca or sca effects and BPH, respectively in the desirable direction. The rank obtained by parents or hybrids were summed up across all the characters to arrive at a total score for each of the parents or crosses. Further, the mean of the total scores of all the parents or crosses across the traits was computed which was used as the final norm to ascertain the status of a parent or a hybrid for their gca or sca effects and BPH. The parent or hybrid whose total rank exceeds the final norm were given low (L) overall gca or sca or BPH status, respectively. On the other hand, the parent or a hybrid, whose total rank was less than the final norm were given high (H) overall gca or sca or BPH, respectively. Based on the overall gca status of the parents,

crosses were classified into HH (both the parents in a cross with high overall *gca* status), HL (one parent with high and the other with low overall *gca* status) and LL (both the parents with low overall *gca* status) categories (Anilkumar and Lohithaswa, 2018).

3. RESULTS AND DISCUSSION

A nalysis of Variance (ANOVA) showed significant differences among the cotton genotypes for seed cotton yield (SCY) and its contributing characters (Table 2) across the locations. Results indicated that estimated mean squares of lines, testers and crosses for ten agronomic characters were highly significant. Higher magnitude of mean squares of testers noticed in seed cotton yield and its contributing characters like the number of monopodia and sympodia, bolls, boll weight and lint percentage compare to lines and indicated greater diversity among the testers and these testers can be pursued for developing heterotic groups with high combining ability (Andayani et al., 2018). Whereas, line × tester variance was significant for all characters under study except the number of monopodia and sympodia.

Furthermore, interactions mean squares between crosses×location, line×location, tester×location were significant for all the characters. Likewise, lint×tester×location interaction exhibited a significant mean sum of squares for all the characters except for the number of monopodia, seed index and lint percentage. This indicated that parents and crosses performed differently across locations. Furthermore, Patil et al. (2017) suggested that line and/ or tester interactions with multi-environment affect the agronomic performance of parents which also results in a change in cross performance.

The results of pooled combining ability estimates were presented in Table 3. GCA variance is larger than the SCA variance for all the characters except days to first and fifty per cent flowering in the pooled analysis. The closer the ratio GCA: SCA is to unity, the greater the predictability of progeny performance based on GCA alone and better the transmission of trait to progenies. Less than one predictability ratio was observed in all characters revealing that these traits were purely under non-additive gene action (Patil et al., 2017, Khokar et al., 2018 and Richika et al., 2021).

Both lines and testers differed widely in their combining abilities for all the traits. The differences in *gca* effects are attributable to differences in frequencies of genes with the additive effects (Falconer and Mackay, 1996). The differences in gene frequencies among the lines and testers suggested their significant genotypic differences, thus justifying their selection for the present study. As expected, different lines and testers were desirable general combiners

the recutions											
Source of variation	Df	DOF	DFF	PH	MONO	SYM	NOB	BW	SCY	SI	Lint %
Locations	2	5.24	33.95*	70162.76**	13.21**	15.55**	15644.86**	57.04**	68409025.74**	84.11**	169.36**
Replications	3	9.98	7.07	18.96	0.03	2.01	23.34	0.37	371168.4	5.93	6.52
Crosses	104	8.26**	11.26*	959.06**	0.52^{*}	9.16*	431.07**	1.68**	1298138.42**	3.47*	21.00**
Line	14	18.01**	27.65**	4481.72**	2.09**	29.15**	1696.65**	5.64**	4168265.01**	17.84**	56.53**
Tester	6	10.36**	15.38*	4465.15**	2.27**	64.14**	2099.94**	12.99**	8203480.88**	8.81**	185.54**
Line×tester	84	6.49**	8.24*	121.52**	0.14	1.9	100.94**	0.22**	326545.24**	0.70^{*}	3.32*
Location× Crosses	208	9.92**	13.20**	139.00**	0.22*	3.45**	110.23**	0.27**	323142.51**	0.76*	3.03*
Location× line	28	27.06**	37.48**	378.83**	0.52**	8.76*	326.60**	0.78**	999313.39**	2.52**	8.66**
Location× tester	12	14.27*	26.33**	393.52**	0.40**	7.63*	187.34**	1.06**	748222.62**	2.00*	6.76**
Location× line×tester	168	6.76*	8.22*	80.85**	0.16	2.26**	68.66**	0.13*	180084.49**	0.38	1.82
Residuals	312	3.62	5.16	12.59	0.13	1.88	16.55	0.06	41954.7	0.36	1.81

Table 2: Analysis of variance (ANOVA) for the combining ability estimates for seed cotton yield and its components across the locations

*, **: Significant at (*p*=0.05 and *p*=0.01 probability levels respectively. DOF: Days to flowering, DFF: Days to 50% flowering PH: Plant height; MONO: Number of monopodia, SYM: no of sympodia, NOB: Number of bolls; BW: Boll weight in g; SCY: Seed cotton yield in kg ha⁻¹; SI: Seed Index, Lint %

Table 3: Variance components for gene action across the location										
Source of variation	Df	DOF	DFF	PH	MONO					
Days to flowering	0.48	0.16	0.65	1.91	0.41					
Days to 50 percent flowering	0.51	0.28	1.12	2.05	0.52					
Plant height	18.15	77.22	308.88	72.62	0.89					
Number of monopodia	0.00	0.04	0.14	0.00	0.99					
No of sympodia	0.00	0.67	2.68	0.02	0.99					
Number of bolls	14.07	30.44	121.75	56.26	0.81					
Boll weight	0.03	0.14	0.54	0.10	0.91					
Seed cotton yield	45890.07	109292.68	437170.70	183560.29	0.79					
Seed Index	0.06	0.26	1.02	0.22	0.90					
Lint percentage (%)	0.25	1.63	6.52	1.01	0.93					

in both direction and magnitude for different traits. Thus, no single line or tester was a desirable combiner for all the traits. As it is true to lines and testers for *gca* effects, the hybrids differed significantly for their *sca* and better-parent heterotic effects. These results indicated that while the performance of a few hybrids is attributable only to their parental genes with additive effects, which of other hybrids is attributable to non-additive effects of their parental genes in addition to their additive effects (Arunachalam, 1976). It should, however, be noted that the estimates of *gca* and *sca* effects are relative to and are dependent on a particular

set of parents included in the experiment.

Similar to lines and testers for their *gca* effects, the different hybrids displayed desirable *sca* and heterotic effects for different traits. Among BGII parents, IAC7-7 was a good combiner for seed cotton yield and IAC7-1 for boll weight and seed index. Among the hybrids crosses involved in both BGII parent, BGII×BGI, BG II×NBt, BGI×BGI, BG I×NBt and NBt×NBt resulted in BGII homozygous, BGII hemizygous, BGI homozygous, BGI hemizygous of hybrids respectively. Among the five groups of hybrids, BGII hemizygous (60 hybrids) comprise more

number of hybrids compared to others and the least with Non-Bt hybrids (2). All the hybrids behaved variedly over environments. IAC7-15NBt×IAC7-17BGII showed significant *sca* effects for seed cotton yield in all locations. The crosses interacted more markedly with environments, suggesting the hybrids did not have the same relative performance across locations (Rojas and Sprague, 1952 and Shahzad et al., 2019). The most peculiar outcome is all the characters were having significant *sca* in BGII hemizygous group to other. Different parents and their crosses lead to different results in *gca* and *sca*, heterosis effects made to analyse the overall status of *gca*, *sca* effects and heterosis for parents and crosses under study.

Out of 22 parents, seven lines and three testers showed high overall *gca* status across the environment and the remaining exhibited low *gca* status (Table 4). Among them, three BG II (IAC7-1, IAC7-7 and IAC7-10), two non-Bt (IAC7-6 and IAC7-15) and three BGI lines (IAC7-3, IAC7-11 and IAC7-12) lines showed high gca status. Likewise, three BGI testers (IAC7-16, IAC7-20 and IAC7-21) displayed high gca. This indicates around 66% of BGI (6 out of 9 BGI) lines showed high overall gca effects compared to 30% of BGII lines (3 out of 10). Although these are a matter of genetic background of the material, however, set of isogenic lines will enlighten the influence of BGII and BGI on overall combining ability. The lines and testers with high overall gca effects could be used to develop hybrids from which we can derive the superior lines with multiple characters. Among the 105 crosses, IAC7-15NBt×IAC7-17BGII recorded a high overall sca effect over the environment followed by IAC7-5BGII× IAC7-18NBt and IAC7-3BGI×IAC7-20BGI, in which either of the parents scored H for overall gca effects (Table 5).

Table 4: Overa	ll general combin	ing ability s	tatus of pare	ents in cot	ton				
Line ^a	Status	Overa	ıll rank	Over	all rank	Overall rank		Overall rank	
		E1	Status	E2	Status	E3	Status	Ep	Status
IAC7-1	BG II	68	Η	80	Η	79	Η	80	Η
IAC7-2	BG I	91	L	79	Η	90	L	92	L
IAC7-3	BG I	87	L	65	Н	71	Н	65	Н
IAC7-4	BG II	120	L	106	L	93	L	119	L
IAC7-5	BG II	93	L	86	L	103	L	90	L
IAC7-6	NBt	56	Н	53	Н	53	Н	57	Н
IAC7-7	BG II	76	Н	51	Н	88	L	74	Н
IAC7-8	BG II	98	L	83	L	55	Н	86	L
IAC7-9	BG II	92	L	94	L	76	Н	88	L
IAC7-10	BG II	60	Н	86	L	90	L	72	Н
IAC7-11	BG I	66	Н	98	L	78	Н	72	Н
IAC7-12	BG I	45	Н	31	Н	86	L	49	Н
IAC7-13	BG II	95	L	102	L	127	L	107	L
IAC7-14	BG II	90	L	122	L	64	Н	90	L
IAC7-15	NBt	63	Н	64	Н	47	Н	58	Н
Tester ^b									
IAC7-16	BG I	29	Н	29	Н	26	Н	28	Н
IAC7-17	BG II	55	L	64	L	47	L	55	L
IAC7-18	NBt	43	L	46	L	53	L	47	L
IAC7-19	BG I	32	Н	30	Н	47	L	41	L
IAC7-20	BG I	33	Н	40	Н	27	Н	31	Н
IAC7-21	BG I	40	Н	35	Н	31	Н	35	Н
IAC7-22	BG I	48	L	36	Н	49	L	43	L

a Final norm: 80 b final norm 40; H: High overall *gca* status; L: Low overall *gca* status; E1: Aurangabad; E2: Dharwad; E3: Raichur; Ep: Across the locations

Similarly, 50% of hybrids (55) were exhibited overall high *sca* status and such heterotic status have also been reported in maize (Anilkumar and Lohithaswa, 2018). Environmental

influences on better parent heterosis could reflect changes in hybrid performance, changes in inbred performance or a combination of both (Labroo et al., 2021). Based on *sca*

	1		0											
Lines	Testers													
	IAC	27-16	IAC	27-17	IAC	27-18	IAC	27-19	IAC	27-20	IAC	27-21	IAC	27-22
	(]	H)	(L)	(]	L)	(]	[)	(]	H)	(H)	(.	L)
	Total	Status	Total	Status	Total	Status	Total	Status	Total	Status	Total	Status	Total	Status
	score		score		score		score	-	score		score		score	
IAC7-1 (H)	699	L	780	L	658	L	385	Η	308	Η	501	Η	472	Η
IAC7-2 (L)	451	Η	376	Η	547	L	717	L	712	L	410	Η	443	Η
IAC7-3 (H)	452	Η	495	Η	422	Η	763	L	292	Η	644	L	659	L
IAC7-4 (L)	304	Н	657	L	687	L	702	L	426	Н	411	Н	482	Н
IAC7-5 (L)	466	Н	603	L	257	Н	623	L	625	L	501	Н	549	L
IAC7-6 (H)	864	L	318	Н	517	Н	453	Η	470	Н	451	Н	428	Н
IAC7-7 (H)	611	L	704	L	475	Н	501	Η	420	Н	651	L	391	Н
IAC7-8 (L)	568	L	692	L	355	Н	357	Η	699	L	564	L	566	L
IAC7-9 (L)	533	L	594	L	678	L	639	L	537	L	397	Н	338	Н
IAC7-10(H)	336	Н	511	Н	419	Н	431	Η	841	L	506	Н	621	L
IAC7-11 (H)	623	L	483	Η	394	Н	495	Η	590	L	641	L	399	Н
IAC7-12 (H)	588	L	309	Η	597	L	357	Η	603	L	651	L	688	L
IAC7-13 (L)	303	Н	707	L	670	L	699	L	235	Н	614	Н	515	Н
IAC7-14 (L)	301	Н	834	L	787	L	327	Η	380	Н	405	Н	613	L
IAC7-15 (H)	801	L	161	Η	492	Н	525	Н	663	L	598	L	687	L

Table 5: Overall specific combining ability status of the cross in cotton from across the location

Final norm 530; H: High overall gca status; L: Low overall gca status

effects, the crosses were classified into H×H, H×L or L×H and L×L (Table 6). The number of hybrids with high (H) overall status was more in H×L type of crosses. Thus, the present study indicated the requirement of parents with contrasting gca effects to realize a higher frequency of heterotic hybrids. The predominance of H×L type of crosses indicated the presence of non-additive gene action suggesting exploitation of good heterotic hybrids from the study. In many cases, it was observed that at least one good general combining parent (H) was involved in heterotic hybrid having desirable sca effects (Ahuja and Dayal, 2007). The superiority of H×L crosses in producing a high magnitude of heterosis over the number of characters, is of practical utility to a breeder. It is worthwhile to initiate H×L type of crosses for realizing hybrids with high heterosis to optimize resources. The support for the utility of combining ability as one of the criteria for choosing the parents comes from the theoretical results which have indicated higher heterosis in the hybrids derived from parents differing in the frequencies of the genes (Cress, 1966). The parental differences in combining abilities are

attributed to differences in gene frequency (Falconer and Mackay, 1996).

Heterosis will be the greatest, as pointed out by Falconer (1996) when one allele is in a homozygous state in one parent and the other allele in the other parent. Since the heterotic response of F_1 is by and large associated with the diversity of the inbred parents, the common approach of selecting the parents only based on per se performance and adaptation does not necessarily lead to much gainful result (Allard 1960). Some lines produce outstanding progenies on the crossing, while certain others, apparently equally desirable, turn out to be poor parents. In our study, cross which is having high overall gca scored low heterobeltiosis from pooled location (Table 7). Among the crosses, IAC7-12BGI ×IAC7-16BGI (H×H) was scored high better parent heterosis for seed cotton yield and its contributing character followed by IAC7-3BGI×IAC7-20BGI and IAC7-10BGII×IAC7-16BGI. All these crosses belonged to both parents having high overall gca (H×H) for yield and its contributing characters. This revealed both additive and non-additive genetic components to be responsible for

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Table 0. Overa	in neter	oue (DI	II) stat	us of the		II COLLOII	monn av	loss the	locatio	11				
Lines							Tes	ters						
	IAC	27-16	IAC	27-17	7 IAC7-18			7-19	IAC7-20 (H)		IAC7-21		IAC	27-22
	(]	H)	(L)	(]	L)	(]	_)			(]	H)	(]	Ĺ)
	Total	Status	Total	Status	Total	Status	Total	Status	Total	Status	Total	Status	Total	Status
	score		score		score		score		score		score		score	
IAC7-1 (H)	569	L	831	L	609	L	496	Η	338	Н	494	Η	533	L
IAC7-2 (L)	504	Η	670	L	658	L	617	L	580	L	417	Η	498	Н
IAC7-3 (H)	478	Η	595	L	436	Η	584	L	264	Н	445	Η	470	Η
IAC7-4 (L)	555	L	826	L	781	L	846	L	534	L	604	L	724	L
IAC7-5 (L)	703	L	806	L	650	L	724	L	673	L	577	L	728	L
IAC7-6 (H)	695	L	594	L	587	L	547	L	456	Н	447	Н	481	Н
IAC7-7 (H)	528	Η	617	L	471	Н	449	Н	331	Н	506	Н	447	Н
IAC7-8 (L)	502	Η	662	L	466	Η	448	Н	480	Н	537	L	466	Η
IAC7-9 (L)	528	Η	725	L	700	L	615	L	478	Η	514	Η	458	Η
IAC7-10(H)	274	Η	542	L	470	Н	483	Н	596	L	501	Н	484	Н
IAC7-11 (H)	522	Η	406	Н	464	Н	430	Н	442	Н	567	L	436	Н
IAC7-12 (H)	256	Η	302	Н	378	Н	293	Н	343	Н	405	Н	318	Н
IAC7-13 (L)	389	Η	343	Н	637	L	753	L	450	Н	684	L	533	L
IAC7-14 (L)	312	Η	670	L	620	L	378	Н	322	Н	487	Н	499	Н
IAC7-15 (H)	665	L	534	L	553	L	577	L	550	L	535	L	695	L

Table 6: Overall heterotic (BPH) status of the cross in cotton from across the location

Final norm 530; H: High overall gca status; L: Low overall gca status

Table 7: Distribution of crosses with high overall sca and heterotic status concerning overall parental gca status in cotton									
Parental GCA	No. of	Mean SCY	Range of seed cotton yield	sca effects	status in F ₁	BPH (%)			
combination	hybrids	(kg ha ⁻¹)	(kg ha ⁻¹)	High	Low	High	Low		
H×H	24	3096	2366 - 3717	9	15	17	8		
$H \times L$ or $L \times H$	53	2869	2003 - 4095	37	16	30	23		
L×L	28	2435	1646 - 3288	9	19	8	20		
Total	105			55	50	55	51		

the high heterosis (Singh and Gupta, 2019). Furthermore, the superiority of the nine crosses having both the parents with low *gca* showed their specific gene combinations resulted in high *sca* effects (Table 6). Therefore, results suggested that both *gca* and *sca* effects are important for the expression of heterosis and should be precisely tested with the appropriate testers in any hybrid breeding program.

3.1. Transgenic trait status and combining ability estimates

The frequency of hybrids with overall high *sca* and heterobeltiosis was more in BGII hemizygous hybrids compared to BGII homozygous condition with H×L type (Table 8). The same results were comparable with BGI hemizygous hybrids also. This indicates that to get high overall *sca* and heterobeltiosis one of the parents should have

BGII or BGI and either of the parents have high overall *gca* (H) for all characters may give high chances of successful heterotic hybrid (Anilkumar and Lohithaswa, 2018).

3.1. Association among per se performance, combining ability estimates and heterosis

A strong relationship between *gca* effects and line *per se* performance for all characters were observed except the number of bolls per plant, days to flowering and fifty per cent flowering (Table 9). The strong association between *per se* performance and *gca* effects of parents gives direction to the selection of parents (Anandan, 2010). The correlation on combining ability and *per se* performance are strongly related to most of the characters under study and it takes breeder to select the parents based on high *per se*

Groups	Parental <i>gca</i> combination	No.of hybrids	No. of hybrids having high					
		,	sca	BPH (%)				
BG II	H×H	9	5	7				
Hemizygous	$H \times L$ or $L \times H$	32	24	20				
	L×L	19	8	6				
BG II	$H \times L$ or $L \times H$	2	1	0				
Homozygous	L×L	7	0	1				
BG I	H×H	6	2	2				
Hemizygous	$H{\times}L~~or~L{\times}H$	7	5	4				
	L×L	1	0	0				
BG I	H×H	9	2	8				
Homozygous	$H{\times}L~~\text{or}~L{\times}H$	9	5	7				
	L×L	2	1	1				
Non-Bt	H×L	2	2	2				

Table 8: Distribution of crosses with high overall *sca* and heterotic status concerning overall parental *gca* status among the transgenic group of cotton

Table 9: Correlation coefficients between r (gca, LP) of parents, r (F_1 , BPH) and r (sca, BPH) from across the location in cotton

Characters	r (gca, LP)	r (F ₁ , sca)	r (<i>sca</i> , BPH)	r (F ₁ , BPH)
Days to flowering	0.29*	0.80**	0.66**	0.58**
Days to 50% flowering	0.34*	0.77**	0.74**	0.62**
Plant height (cm)	0.85**	0.32*	0.16	0.39*
No. of monopodia plant ⁻¹	0.73**	0.46**	0.62**	0.52**
No. of sympodia plant ⁻¹	0.77**	0.41**	0.60**	0.38*
No. of bolls plant ⁻¹	0.21	0.43**	0.45**	0.44**
Boll weight (g)	0.78**	0.32*	0.67**	0.50**
Seed cotton yield (kg ha ⁻¹)	0.53**	0.45**	0.59**	0.39*
Seed index (g)	0.80**	0.40**	0.26	0.44**
Lint percentage (%)	0.76**	0.36*	0.21	0.27^{*}

*, **: Significant at (*p*=0.05) and (*p*=0.01) probability levels respectively

performance by ignoring combining ability. However, as we noticed a high frequency of overall high *sca* effects and better parent heterosis was observed in the H×L group only. This shows the importance of combining ability study in heterosis breeding. Likewise, a strong association between *per se* performance of F_1 and *sca* effects were found for all characters except lint percentage (r=0.36) followed by plant height and boll weight (r=0.32). Furthermore, *sca* effects were showed a strong positive association with better parent heterosis for flowering traits, number of monopodia and sympodia per plant, number of bolls, boll weight and seed cotton yield (Table 9). Same way better parent heterosis has positively linked all characters indicating that as the performance of cross increases heterosis also increases.

4. CONCLUSION

Significant variations were observed for gca., per se performance and sca across locations for all the traits. The gca for seed cotton yield and its contributing characters were found to be positively correlated with per se performance indicating that high general combiners are more likely to have high seed cotton yield. Hemizygous hybrids with one Bt parent (BGI or BGII) were comparatively more heterotic. Parents with stable gca and higher per se performance were observed for further breeding.

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