

LinexTester Analysis and Estimating Combining Abilities for Yield and its Components in Soybean

ABSTRACT Combining ability is mostly used by breeders to select appropriate parental cultivars to produce the larger progeny of new combinations through their hybridization. The objectives of this research were to estimate the general combining ability of parents and specific combining abilities of F1 hybrids, to identify suitable parents and hybrids for yield and its contributing traits. In this study, materials comprising 8 F1s derived by crossing 2 lines with 4 testers following the Line x Tester mating design. The F1s and the parental genotypes were grown at the 'Research Farm' of the Tagore University's, Department of Genetics & Plant Breeding during the autumn 2021 using Randomized Block Design (RBD) with three replications. The popular soybean variety JS 335 was found as the best overall performer (good general combiner) for most traits, while the cross-combination JS 93-05 X RVS 2001-4 showed particular promise for specific traits. The cross RVS-24 X JS 95-60 showed desirable traits like high plant height, pod number, and yield, making it a potential candidate for breeding higher-yielding and biomass-rich varieties. Additionally, crosses RVS-24 X JS 95-60, RVS-24 X JS 335, JS 93-05 X RVS 2001-4, and RVS-24 X JS 97-52 exhibited high heterotic responses for seed yield, making them valuable for developing high-yielding lines. The cross JS 93-05 X RVS 2001-4 showed the best potential for genetic improvement in the pod number/plant, seed number per pod, yield/plant, and the harvest index, making it a valuable resource for isolating high-yielding genotypes.

Keywords: -General combining ability and specific combining ability.

INTRODUCTION

In self-pollinated crop soybean (*Glycine max* (L.) Merrill), recombination breeding has been extensively used to develop the variability reservoir for exploitation in a breeding program. In a systematic breeding program, it is essential to identify the elite parents for hybridization and superior crosses to expand the variability reservoirs for the selection of superior genotypes. [17-20] Combining ability studies help in such endeavor. In the present investigation, line × tester design with well-adapted and widely grown varieties of soybean (tester) was used to obtain information on combining the ability of elite lines for five characters of economic importance in soybean. (Tadesse and Tizzazu, 2017). For the genetic improvement of soybeans, the plant breeders have an important role to play in meticulously unravelling the genetic mysteries that govern the inheritance of desired traits. This vital information is especially critical when selecting parents for breeding programs. Here's where the concepts of the combining ability and the gene action come into play. Combining ability essentially tells us how well a parent performs when crossed with different partners. This helps breeders identify parents that consistently produce high-performing offspring, regardless of their mate. On the other hand, gene action delves deeper, revealing the underlying mechanisms by which genes influence traits like yield and its contributing factors. Understanding these mechanisms is essential for choosing the most effective breeding strategy to isolate and amplify desirable traits. Therefore, evaluating breeding materials for both general and specific combining ability, along with investigating the degree of hybrid vigor (heterosis) for yield and related traits, becomes the foundation for any successful

breeding program. It's by unlocking these genetic secrets that breeders can truly unlock the full potential of their crops. Seed yield is a complex trait. It is the result of the interaction of several traits in association with the environment. Reliable information on it facilitates the breeder to decide an appropriate breeding procedure for crop improvement. Similarly, the relative magnitude of additive and non-additive gene action forms the guidelines for handling the segregating material from crosses having diverse parents.

MATERIAL AND METHODS

The experiment was conducted during autumn season of 2021. The field experiment was undertaken at the Institute farm of Rabindranath Tagore University, Raisen, MP.

The materials for the experiments comprising 2 soybean lines (L) and 4 testers (T) were hybridized in line (L) x tester (T) crossing scheme. The crosses resulted in eight F₁ hybrids. These F₁ hybrids, and their parental lines were grown in Randomized Block Design (RBD) with three replications. Here, each experimental entry was planted in a single 3.0m row with a plant-to-plant and row to row spacing of 23 cm and 40 cm, respectively.

The combining ability was done by adapting Kempthorne (1957) procedure. The total variance among F₁ hybrids was further partitioned into variance due to lines, testers and their interaction component, which was used to estimate the additive and non-additive components of variance. Also, the contribution of lines, testers and their interaction towards total variability for each character was computed for assessing their relative importance according to model as suggested by Kempthorne (1957) using the statistical software package of Windostat version 9.1.

Table 1: List of lines (L), testers (T) and LxT hybrids used in this experiment

S. No.	Parental genotypes		LxT hybrids
	Name of the Testers	Name of the Lines	
1	JS 97-52	JS 93-05	JS 93-05 X JS 97-52
2	JS 95-60	RVS 24	JS 93-05 X JS 95-60
3	RVS 2001-4		JS 93-05 X RVS 2001-4
4	JS 335		JS 93-05 X JS 335
5			RVS 24 X JS 97-52
6			RVS 24 X JS 95-60
7			RVS 24 X RVS 2001-4
8			RVS 24 X JS 335

RESULT

Analysis of variance for combining ability

The data recorded for eight hybrids, two lines and four testers were used for analyzing the combining abilities using the line x tester design (L x T) as given by Kempthorne (1957).

The extent of variance was assessed to find the significant differences among various genotypes, including parents, crosses, lines, and testers. The F-test was used to compare the significant differences (Table 2).

The ANOVA clearly exhibited the existence of significant differences among the genotypes for all characters except pod length. The variance due to treatments was divided into crosses, parents, and parents vs. crosses, and it was found that the variance due to parental lines was significant for all characters except pod length. Crosses too exhibited a significant variance for all the characters except pod length. The variance of the lines was also highly significant for all the traits studied except for pod length and 100-seed weight. Significant MS for the testers were recorded for number of secondary branches/plant, days-to-50% flowering, days-to-maturity, pod length, number of pods/plant, grain yield/plant, biological yield/per plant, and HI, while they were non-significant for 100-seed weight and the number of primary branches/plant. For line x tester interaction, the variances for all the characters were highly significant except for the pod length. The significant Mean squares (MS) for lines and testers contributed to the GCA variance components for the traits. Similarly, the significant MS for the line x tester interaction indicated the contribution of the crosses to the SCA variance component and a significant amount of variation among the hybrids.

Table 2 ANOVA for line x tester for yield and yield attributing traits in soybean

Source	d. f.	Traits										
		Days-to-50% flowering	Days-to-maturity	Plant height (cm)	Primary branches/plant	Secondary branches/plant	Pods/plant	Pod Length (cm)	100-seed weight (g)	Seeds Yield/plant (g)	Biological yield/plant (g)	Harvest index (%)
Replications	2	1.7917	2.1667*	2.3504*	0.2413	0.2304	18.761**	0.0812	1.172*	0.5712	0.2618	19.8004**
Parents	5	82.756**	67.256**	562.00**	3.122*	3.728*	145.820**	0.474	2.524**	9.041**	5.845**	127.823**
Crosses	7	93.232**	31.185**	21.095**	2.562*	8.595**	110.816**	1.000	1.425*	7.157**	11.942*	119.125**
Parents vs crosses	1	60.764**	40.573**	3,02740**	7.261*	18.134*	264.470**	0.056	0.014	9.392**	22.086*	7.549*
Lines	1	301.041**	70.041**	11.9004**	3.72*	5.0417*	71.070**	0.629	0.1067	6.8267**	9.2504*	16.0067**
Testers	3	108.375**	43.597**	19.7715*	0.5726	7.5056*	22.190**	1.6049**	0.9228	2.1744*	14.9126**	151.070**
L X T	3	8.8194**	5.8194**	25.484**	4.151*	10.8694**	212.688**	0.5171	2.3656**	12.250**	9.8682*	121.554**
Error	14	4.5536	8.4524	15.919	0.9693	0.720	19.792	0.312	0.824	0.7455	0.946	19.642

*, ** differ significantly at 5% and 1% level of probability, respectively.

General and Specific combining ability (GCA and SCA) variance:

The variations due to GCA and SCA for different traits under investigation are presented in Tables 3. The GCA variance was highest for the pods/plant (25.95) followed by seeds

yield/plant (9.27), days-to-50% flowering (4.92), days-to-maturity (1.47), biological yield/plant (1.21), 100-grain weight, pod length, number of primary branches/ plant, number of secondary branches/plant, plant height and harvest index.

The SCA variance was highest for the pods/plant (64.30) followed by seeds yield/plant (38.35), harvest index (33.97), No. of secondary branches/plant (3.38), plant height (3.18), biological yield/plant, days-to-50% flowering, number of primary branches/plant, 100 seed weight, pod length and days to maturity.

The result in the tables indicate that the SCA variance was greater than the GCA variance and the variance-ratio was <1 for all the traits except days-to-50% flowering, which showed higher GCA variance than SCA variance. It suggests that the SCA is playing a more important role than the GCA in the control and expression of the traits.

Table 3 Estimates of genetic components of variances for yield and yield-attributes in soybean

Source	Traits										
	Days-to-50% flowering	Days-to-maturity	Plant height (cm)	Primary branches per plant	Secondary branches per plant	Pods per plant	Pod length (cm)	100-seed weight (g)	Seeds yield per plant (g)	Biological yield per plant (g)	Harvest index (%)
σ^2_{gca}	4.92	1.47	-0.25	-0.09	-0.13	25.95	0.02	-0.05	9.27	1.21	-0.14
σ^2_{sca}	1.42	-0.87	3.18	1.06	3.38	64.30	0.03	0.51	38.35	2.97	33.97
$\sigma^2_{gca}/\sigma^2_{sca}$	3.46	-1.68	-0.08	-0.08	-0.03	0.40	0.66	-0.10	0.24	0.40	0.004

The estimate of GCA and SCA effects and Gene action –

The values of GCA and SCA effects for all the traits under study are presented in the Tables 4 and 5, respectively. Trait-wise, the results are as follows:

Days-to-50% flowering

Estimated GCA effects for days-to-50% flowering were negative for genotypes JS 93-05 (-3.54), JS 95-60 (-3.79), and RVS 2001-4 (-2.29), while parent RVS-24 (3.54) and JS-335 (5.88) exhibited a positive and significant GCA effects. However, the GCA effects for the crosses were non-significant.

Days-to-maturity

Significant negative GCA effects were observed for days to maturity in JS 95-60 (-3.29), while the JS 97-52 (3.04) showed a significant and positive GCA effect. Here, no one of the crosses exhibited either significant positive or significant negative SCA effects.

Plant height (cm)

A non-significant GCA and SCA effects were recorded for plant height in the parents and the crosses.

Number of primary branches per plant

Like plant height, for primary branches per plant also, no one of the parents showed either positive or negative significant GCA effects. However, only one cross i.e., JS 93-05 X JS 97-52 (0.23) showed a significant positive SCA effect for primary branches/plant. The SCA variance for the number of primary branches/plant appeared to be greater than the GCA variance signifying the prevalence of non-additive gene actions for it.

Number of secondary branches/plant

The parental line JS 335 (1.22) exhibited a positive and significant GCA effect, while RVS 2001-4 (-1.32) showed a negative significant GCA effect for the No. of secondary branches/plant. The SCA effects of the crosses viz., RVS-24 X JS 95-60 (1.49), and JS 93-05 X JS 97-52 (1.27) showed significant positive effects while, other two crosses i.e. RVS-24 X JS 97-52 (-1.27) and JS 93-05 X JS 95-60 (-1.49) exhibited a significant negative SCA effect for the secondary branches/plant. The greater SCA variance for secondary branches/plants than the GCA indicated a predominance of the non-additive gene action in the expression of the trait.

Number of pods/plant

For the No. of pods/plant, a significant GCA effect was recorded in the parental genotypes viz., RVS-24 (1.72), JS 93-05 (-1.72) and JS 335 (0.76) while, parent JS 93-05 (-1.72) exhibited a significant and negative GCA effect. The cross JS 93-05 X RVS 2001-4 (6.39) showed significant positive SCA effects while, cross RVS-24 X RVS 2001-4 (-6.39) exhibited a significant negative SCA effect for the trait. The variance due to SCA was higher than the GCA variance indicating involvement of the non-additive gene action for the trait.

Pod length (cm)

None of the parents showed positive significant GCA effect for pod length while, parent RVS 2001-4 (-0.70) exhibited a significant negative GCA effect. A non-significant GCA and SCA effect for pod length was noted in the cross combinations.

100-Seed weight (g)

For 100 seed weight, the GCA and SCA effects were non-significant. However, the variance due to SCA was higher than the GCA, suggesting that the non-additive gene action primarily determined the seed weight.

Seed yield/plant

Parental lines viz., RVS-24 (0.62), RVS 2001-4 (0.20) and JS 335 (0.68) showed significant positive GCA effects, while parents JS 93-05 (-0.62) and JS 97-52 (-0.13) exhibited significant negative GCA effects for the trait seed yield/plant. This means that the first three parents tend to pass on genes that increase seed yield, while the latter two tend to pass on genes that decrease seed yield.

The crosses RVS-24 X JS 335 (1.40) and JS 93-05 X RVS 2001-4 (1.78) exhibited positive, significant SCA effects, while crosses RVS-24 X RVS 2001-4 (-1.78) and JS 93-05 X JS 335 (-1.40) expressed a significant negative SCA effects for the trait seed yield/plant.

It was observed that the variance due to SCA for the trait was greater than the variance due to GCA, indicating the predominance of non-additive gene action in controlling the trait.

Biological yield/plant (g)

The GCA effects for biological yield per plant were positive and significant in parental lines RVS-24 (0.62), JS 95-60 (1.00), and JS 335 (1.55). This means these parents tend to pass on genes that increase biological yield to their offspring. Parental lines JS 93-05 (-0.62) and RVS 2001-4 (-1.92) showed significant negative GCA effects, indicating they tend to pass on genes that decrease the biological yield/plant. The cross RVS-24 X JS 95-60 (1.26) exhibited a significant and positive SCA effect, as against the crosses RVS-24 X JS 97-52 (-1.35) and JS 93-05 X JS 95-60 (-1.26) that exhibited a significant and negative SCA effect. This means the specific combination of genes in these crosses leads to an increase or decrease in biological yield. Similarly, the variance due to SCA effect was larger to the variance due to GCA effects, which indicated that the preponderance of non-additive gene action in controlling the biological yield/plant.

Harvest index (HI) (%)

For harvest index, the GCA effect was positive and significant only in the parental line RVS 2001-4 (6.17), while it was negative and significant in JS 95-60 (-5.93). This means RVS 2001-4 tends to pass on genes that increase harvest index, while JS 95-60 tends to pass on genes that decrease it. The cross JS 93-05 X RVS 2001-4 (6.50) found to have a significant and positive SCA effect, while the cross RVS-24 X RVS 2001-4 (-6.50) exhibited significant and negative SCA effects. This means the specific combination of genes in these crosses would lead to an increase or decrease in harvest index, what would be beyond the expected value from the individual parents. Further, the variance due to SCA effect was larger than the same for the GCA, which indicated the prevalence of non-additive gene actions in controlling the harvest index.

Table 4 GCA effects in the parental genotypes for yield and yield-attributing traits in soybean

Crosses	GCA effects of various characters										
	Days-to-50% flowering	Days to maturity	Plant height (cm)	Primary branches per plant	Secondary branches per plant	Pods per plant	Pod length (cm)	100-seed weight (g)	Seeds yield per plant (g)	Biological yield per plant (g)	Harvest Index (%)
LINES											
RVS-24	3.54 **	1.71	0.70	0.40	0.46	1.72 **	-0.16	-0.07	0.53*	0.62 *	0.82
JS 93-05	-3.54 **	-1.71	-0.70	-0.40	-0.46	-1.72	0.16	0.07	-0.53*	-0.62 *	-0.82

						**					
TESTER											
JS 97-52	0.21	3.04 *	-0.51	0.02	-0.47	2.35	-0.07	-0.39	-0.13*	-0.62	0.95
JS 95-60	-3.79 **	-3.29 *	-0.80	-0.30	0.57	-1.85	0.41	0.04	-0.75	1.00 *	-5.93 **
RVS 2001-4	-2.29 *	-0.79	-1.36	-0.15	-1.32 **	-1.25	-0.70*	-0.17	0.20*	-1.92 **	6.17 **
JS 335	5.88 **	1.04	2.67	0.42	1.22 **	0.76 **	0.36	0.52	0.68*	1.55 **	-1.18

*, ** differ significantly at 5% and 1% level of probability, respectively.

Table 5: SCA effects of L x T hybrids for yield and its component traits in soybean

Crosses	SCA effects of various traits										
	Days-to-50% flowering	Days to maturity	Plant height (cm)	Primary branches per plant	Secondary branches per plant	Pods Per plant	Pod length (cm)	100-seed weight (g)	Seeds yield per plant (g)	Biological yield per plant (g)	Harvest index (%)
RVS-24 X JS 97-52	1.12	0.96	-1.39	-0.23	-1.27 *	-3.75	0.38	0.47	-0.48	-1.35 *	1.42
RVS-24 X JS 95-60	0.46	0.63	1.63	0.99	1.49 **	5.18	-0.34	-0.33	0.87	1.26 *	1.20
RVS-24 X RVS 2001-4	-1.71	-1.21	-2.14	-1.00	-0.99	-6.39 *	-0.02	-0.72	-1.78 **	-0.82	-6.50 *
RVS-24 X JS 335	0.12	-0.38	1.90	0.24	0.77	4.96	-0.02	0.58	1.40 *	0.91	3.88
JS 93-05 X JS 97-52	-1.12	-0.96	1.39	0.23	1.27 *	3.75	-0.38	-0.47	0.48	1.35 *	-1.42
JS 93-05 X JS 95-60	-0.46	-0.62	-1.63	-0.99	-1.49 **	-5.18	0.34	0.33	-0.87	-1.26 *	-1.20
JS 93-	1.71	1.21	2.14	1.00	0.99	6.39	0.02	0.72	1.78	0.82	6.50 *

05 X RVS 2001-4						*			**		
JS 93- 05 X JS 335	-0.12	0.38	-1.90	-0.24	-0.77	- 4.96	0.02	-0.58	-1.40 *	-0.91	-3.88

*, ** differ significantly at 5% and 1% level of probability, respectively.

DISCUSSION

Genetic makeup and the differences between the parents used in a hybridization program influence how genes are expressed and how well the hybrid offspring performs. So, it's important to assess the genetic potential of the parent lines by considering their specific and general combining abilities, which reflect the effects of the non-additive and additive gene actions, respectively. The L x T analysis method (Kempthorne, 1957) is well-suited for this purpose, and it's widely used to estimate these combining ability effects and their variations.

GCA and SCA variance

The GCA variance of parents and SCA variance of hybrids are important basic parameters in any breeding program. The GCA variance, which reflects the average performance of parents, was high for all the traits under consideration. Similarly, the SCA variance, which reflects the specific performance of hybrids, was the highest in pods/plant, followed by seeds yield/plant, harvest index, No. of secondary branches/plant, plant height, biological yield/plant, days-to-50% flowering, No. of primary branches/plant, 100 seed weight, pod length, and days-to-maturity. However, the SCA variance was greater than the general combining ability variance for all traits except days to 50% flowering, resulting in a variance ratio less than one. This indicates the predominance of dominance variance over additive variance for most of the characters studied, and the greater SCA variance than GCA also suggests the preponderance of non-additive gene action. Therefore, a hybrid breeding program would be highly effective for the improvement of these characteristics. Similar findings were reported for days-to-maturity (Raut *et al.*, 2000; Singh *et al.*, 2005), No. of primary and secondary branches/plant (Singh *et al.*, 2019; Patel, 2012), seed yield/plant (Saul *et al.*, 2017), biological yield/plant (Gill *et al.*, 2010), 100-seed weight and number of pods/plant (Saul *et al.*, 2017).

GCA & SCA effects and Gene action

Information on General Combining Ability and Specific Combining Ability are crucial parameters in breeding programs for various crops, including soybeans. GCA reflects a parent's ability to transmit its genetic potential reliably to offspring, while SCA reveals the unique performance of specific hybrid combinations.

Among the parent lines, JS 93-05, JS 95-60, and RVS 2001-4 displayed a tendency to pass on genes for earlier flowering (negative GCA), while RVS-24 and JS 335 tended towards later flowering (positive GCA). Interestingly, none of the hybrid combinations showed

significantly earlier or later flowering compared to the expected average, indicating no significant SCA effects. This aligns with findings by Gill *et al.* (2010), suggesting that additive gene effects play a more dominant role in determining days to flowering in soybeans.

Similar to days to flowering, the parent JS 95-60 demonstrated a tendency towards early maturity (negative GCA), while JS 97-52 leaned towards later maturity (positive GCA). However, none of the hybrid combinations exhibited significantly earlier or later maturity compared to the expected average, suggesting no significant SCA effects. This finding is consistent with previous research, indicating that additive gene effects likely influence maturity in soybeans.

Neither parents nor hybrid combinations displayed significant and positive or negative GCA or SCA effects for the plant height, 100-seed weight, pod length, or primary branches/plant. Only one cross, JS 93-05 X JS 97-52, showed a significant and positive SCA effect towards the number of primary branches, aligning with observations by Cho Youngkoo *et al.* (2000), Rahangdale *et al.* (2002), and Painkra (2014).

Parent JS 335 exhibited a positive GCA effect for the number of secondary branches, while RVS 2001-4 showed a negative effect. Two crosses, RVS-24 X JS 95-60 and JS 93-05 X JS 97-52, had significant-positive SCA effects; however, two others, RVS-24 X JS 97-52 and JS 93-05 X JS 335, displayed significant-negative SCA effects.

Several parents and crosses exhibited significant GCA and SCA effects for the number of pods per plant. Parents RVS-24, JS 93-05, and JS 335 possessed positive GCA effects, while JS 93-05 also had a negative effect. Crosses JS 93-05 X RVS 2001-4 and RVS-24 X RVS 2001-4 displayed significant positive and negative SCA effects, respectively, aligning with findings by Kunta *et al.* (1997) and Saul *et al.* (2017).

Both RVS 2001-4 and JS 335 displayed positive GCA effects for biological yield, indicating their potential as good general combiners. Similarly, crosses RVS-24 X JS 335 and JS 93-05 X RVS 2001-4 exhibited a significant positive SCA effects, on the other hand crosses RVS-24 X RVS 2001-4 and JS 93-05 X JS 335 had significant negative SCA effects. These findings resonate with observations by Gill *et al.* (2010) in soybeans.

Parents RVS-24, JS 95-60, and JS 335 possessed a positive GCA effect for the seed yield, while JS 93-05 and RVS 2001-4 exhibited negative effects. Cross RVS-24 X JS 95-60 displayed a significant positive SCA effect, while crosses RVS-24 X JS 97-52 and JS 93-05 X JS 95-60 had significant negative SCA effects. These observations are consistent with previous research by Ponnosamy *et al.* (1999), Cho Youngkoo *et al.* (2000), and Saul *et al.* (2017).

For harvest index, the parent RVS 2001-4 showed positive significant GCA effect, while parents JS 95-60 showed significant negative GCA effect. The cross JS 93-05 X RVS 2001-4 showed significant positive SCA effect, while cross RVS-24 X RVS 2001-4 showed the negative significant SCA effects for harvest index. This results are in the line with those reported by Kunta *et al.*, (1997).

Summary and Conclusion

The cross RVS-24 X JS 95-60 showed desirable traits like high plant height, pod number, and yield, making it a potential candidate for breeding higher yielding and biomass-rich varieties. Additionally, crosses RVS-24 X JS 95-60, RVS-24 X JS 335, JS 93-05 X RVS 2001-4, and RVS-24 X JS 97-52 exhibited high heterotic responses for seed yield, making them valuable for developing high-yielding lines. The popular soybean variety JS 335 was found as the best overall performer (good general combiner) for most traits, while the cross-combination JS 93-05 X RVS 2001-4 showed particular promise for specific traits. The cross **JS 93-05 X RVS 2001-4** showed the best potential for genetic improvement in the pod number/plant, seed number per pod, yield/plant, and the harvest index, making it a valuable resource for isolating high-yielding genotypes.

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