Revealing the inheritance of qualitative traits and identification of transgressive segregants for productivity traits in early segregating generation of soybean (*Glycine max* (L.)Merril).

Abstract

Breeding effort was carried out to elucidate information on inheritance of qualitative traits and identification of transgressive segregants for productivity traitsin two crosses *viz.*,DSb 23 xMACS 1575 (Cross 1) and DSb 23 x MACS 1460 (Cross 2) in soybean during *kharif* 2020 at Botany Garden, Department of Genetics and Plant Breeding, University of Agricultural Sciences, Dharwad. The observations were recorded on nine traits *viz.*, days to 50% flowering, days to maturity, plant height, pods per plant, 100 seed weight, seed yield per plant, hilum colour, leaf shape and flower colour. Test for goodness of fit revealed that the traits *viz.*, flower colour and leaf shape are controlled by a single dominant gene and hilum colour was controlled by two genes with complementary gene action. In general, percent of transgressive segregants obtained for yield related traits were in a range of 7-17%. The high frequency of favourable segregants were identified for pods per plant (15.33%), 100 seed weight (13.87%) and days to maturity (16.42%) in cross 1 andhigh frequency of favourable segregants in cross 2 were observed for days to 50% flowering (8.95%), plant height (13.75%) and seed yield per plant (14.17%). These segregants can be further utilized in the breeding programme to develop high yielding varieties and morphological traits can be used as markers in molecular breeding.

***Keywords*:**Qualitative traits, Transgressive segregants, Soybean.

1. Introduction

Soybean is regarded as the 'miracle crop' or 'Golden Bean' of the 20th century. Soybean belongs to the *Fabaceae*family and is native to north easternChina. It is a versatile crop with ample opportunities not only to improve economy of farming community but also to benefit industries. Soybean provides high quality protein (40%) and cholesterol free oil (20%). In addition to other essential amino acids, vitamins and minerals, soybean is rich source of lysine (6.4%). The oil is used in the manufacture of antibiotics, paints, adhesives and lubricants etc. Soybean is also known for its benefits to human health, such as the cholesterol reducing effect of protein as recently approved by the United States Food and Drug Administration. Work is underway to assess the potential benefits of soybeans in other areas of human health, such as cancer, osteoporosis and post-menopausal discomfort (Krishna, 2001).

In any crop improvement programme knowledge on morphology of traits, their inheritance pattern, nature and number of genes responsible for that trait are important. This ensures better combination of desirable traits in offspring for any improvement programme. Existence of wide morphological diversity in soybean offers ample opportunity for genetic studies. Qualitative characters are considered as marker characters in the identification of soybean species and varieties, which are less influenced by environmental variations. The genetic constitutions of such marker characters have obvious importance.

A wide spectrum of variability is expected to be shown by progenies derived from a set of diverse crosses, thereby providing a greater scope for isolating high yielding segregants. Transgressive segregants are generated by the accumulation of favourable genes from parents involved in hybridization. Studies on transgressive segregants allow us to figure out their proportions for different characters that contribute to the yield. These characters having high variability and heritability will be of more useful in population development and other breeding programs.

Keeping the aforesaid views, an attempt has been made with the following objectives in the present investigation.

1. To study the inheritance of qualitative traits.
2. To identify transgressive segregants for yield and yield attributing traits.

2. Materials and Methods

The experimental material for the present study comprised of F_2 populations of two crosses involving three diverse parents *viz.,*DSb 23, MACS 1575 and MACS 1460. Among the parents, DSb 23 is a high yielding variety with broad leaves, brown hilum colour and purple flowers. Whereas,MACS 1575 and MACS 1460 are the genotypes from Agharkar Research Institute, Pune, Maharashtra were low yielders compared to DSb 23. MACS 1575 has narrow leaves and black hilum colour but MACS 1460 has white flowers and black hilum collar. To understand the inheritance of qualitative traits and identification of transgressive segregants, DSb 23 was crossed with MACS 1575 and MACS 1460. The seeds of two F_2 populations *viz.,*DSb 23 x MACS 1575 (Cross 1) and DSb 23 x MACS 1460 (Cross 2) were obtained from AICRP on Soybean, Main Agricultural Research Station, University of Agricultural Sciences, Dharwad.List of F_2 populations under study is given in Table 1.

Experimental site

The present experiment was carried out during *kharif* 2020-21 at Botany Garden, Department of Genetics and Plant Breeding, University of Agricultural Sciences, Dharwad.

Methods

The experiment comprising of F_2 populations of the two crosses *viz.,*DSb 23 x MACS 1575 (Cross 1) and DSb 23 x MACS 1460 (Cross 2) along with the parents DSb 23, MACS 1575, MACS 1460 were sown in a row length of 5 m with spacing 30 cm between rows and 10 cm between plants. The experiment was laid out in an un-replicated trial during *kharif* 2020-21 under *rainfed* condition. The experimental material was sown on 12th July 2020. The recommended package of practices was followed to raise a good crop. Observations on individual plants were recorded in two F_2 populations.In each F_2 population individual plants were tagged for recording observations on 10 different quantitative and qualitative traits.

Observations were recorded on qualitative traits like leaf shape, hilum colour, flower colour and qualitative traits like yield and yield attributing traits.

Table 1: List of F_2 populations under study

Sl. No	Cross	Parents	Number of F_2 seeds	Plant stand (at harvesting)	Germination (%)
1	Cross 1	DSb 23 x MACS 1575	357	274	76.75
2	Cross 2	DSb 23 x MACS 1460	589	480	81.49

Identification of transgressive segregants

In both the segregating populations, the number of plants which performed better than mean + one standard deviation for group of traits *viz.,* plant height, pods per plant, 100 seed weight and seed yield per plant were noted as transgressive segregants and expressed in percentage. Whereas for days to maturity and days to 50% flowering selection criteria for identifying transgressive segregants was mean - one standard deviation and are also expressed in percentage.

Test for goodness of fit

The chi squaretest based on the enumerative data (qualitative) is a valuable tool for the breeder to decide the observed data is according to an expected ratio or it agrees well with expected or theoretical frequency distribution. It is done as per method suggested by Panse and Sukhatme(1967). χ^2 is expressed as follows.

$$\chi^2 = \sum_{i=1}^{n} \frac{(O-E)^2}{E}$$

Where,

O = Observed frequency of the class

E = Expected frequency of the respective class

The table χ^2 value was taken at (n-1) degrees of freedom and at 0.05 probability level. The null hypothesis H_0 is rejected, if observed χ^2 is found to be greater than the theoretical χ^2 and there by this difference is said to be significant. Otherwise, the null hypothesis is accepted and the difference between observed and theoretical frequency distribution are not significant.

3. Results

3.1. Inheritance studies for leaf shape and hilum colour in cross 1 (DSb 23 x MACS 1575)

χ^2 value and probabilities for segregation of leaf shape and hilum colour in cross 1 are presented in Table 2 and Table 3 respectively.

3.1.1. Leaf shape

The parental lines DSb 23 and MACS 1575 were differed for leaf shape. DSb 23 was a genotype with broad leaves and MACS 1575 was having narrow leaves. In F_2 population 274 individual plants were observed for leaf shape. 210 plants were having broad leaf shape whereas 64 plants were bearing narrow leaves. The calculated chi square value indicates that the observed value for leaf shape fitted well with an expected ratio 3 broad: 1 narrow leaf shape which indicate that this character is controlled by a single dominant gene.

3.1.2. Hilum colour

In cross between DSb 23 xMACS 1575, observations were recorded for hilum colour in order to study its mode of inheritance. In DSb 23 the colour of hilum is brown whereas in MACS 1575 it is black. In F_2 population, individual plants were observed for their hilum colour, 167 plants out of 274 plants showed black hilum colour whereas 107 plants were having brown hilum. The calculated chi square value indicates that the observed value for hilum colour fitted well with an expected ratio of 9 black: 7 brown. This denotes the character is controlled by two genes with complimentary gene action.

3.2. Inheritance studies for flower colour and hilum colour in cross 2 (DSb 23 x MACS 1460)

χ^2 value and probabilities for segregation of flower colour and hilum colour in cross 2 (DSb 23 x MACS 1460) are presented in Table 4 and Table 5 respectively.

3.2.1. Flower colour

The parental lines exhibited variability for flower colour. DSb 23 has purple flowers, whereas the other parent, MACS 1460 is having white flowers. In the cross DSb 23 x MACS 1460, observations were recorded for flower colour in order to study mode of inheritance of flower colour. In F_2 population, 480 individual plants were observed for flower colour. 388 plants were having purple flower color whereas 92 plants were bearing white colored flowers. The calculated chi square value indicates that the observed value for flower colour fitted well with an expected ratio 3 purple: 1 white which indicates that this character is controlled by a single dominant gene.

Table 2: χ^2 value and probabilities for segregation of leaf shape in cross 1 (DSb 23 x MACS 1575)

	Observed	**Expected**	**$(O-E)^2/E$**	**χ^2**	**P**
Broad	210	205.5	0.10	0.39	0.53 (0.6-0.5)
Narrow	64	68.5	0.30		

Total	274				

Table 3: χ^2 value and probabilities for segregation of hilum colour in cross 1 (DSb 23 x MACS 1575)

	Observed	**Expected**	**$(O-E)^2/E$**	**χ^2**	**P**
Black	167	154.12	1.08	2.46	0.12 (0.2-0.1)
Brown	107	119.88	1.38		
Total	274				

Table 4: χ^2 value and probabilities for segregation of flower colour in cross 2 (DSb 23 x MACS 1460)

	Observed	**Expected**	**$(O-E)^2/E$**	**χ^2**	**P**
Purple	388	360	12.47	49.88	0.003 (0.004-0.003)
White	92	120	37.41		
Total	480				

Table 5: χ^2 value and probabilities for segregation of hilum colour in cross 2 (DSb 23 x MACS 1460)

	Observed	**Expected**	**$(O-E)^2/E$**	**χ^2**	**P**
Black	289	270	1.34	3.06	0.08 (0.09-0.08)
Brown	191	210	1.72		
Total	480				

3.2.2 Hilum colour

In the cross between DSb 23 and MACS 1460, observations were recorded for hilum colour in order to study its mode of inheritance. In DSb 23, the colour of hilum is brown whereas in MACS 1460 it is black. In F_2 population, individual plants were observed for their hilum colour, 289 plants out of 480 plants showed black colour whereas 191 plants were having brown hilum. The calculated chi square value indicates that the observed value for hilum colour fitted well with an expected ratio of 9 black: 7 brown. This denotes the character is controlled by two genes with complementary gene action. Similar result was also observed in the cross DSb 23 xMACS 1575 for hilum colour.

3.3. Identification of transgressive segregants

Per cent transgressive segregants in F_2 population of both the crosses for days to 50% flowering, days to maturity, plant height, pods per plant, 100 seed weight and seed yield per plant are given in Table 6. The criterion followed for selection of transgressive segregants was mean+one standard deviation. However, for days to 50% flowering and days to maturity, mean–one standard deviation was considered for identifying the superior segregants.

3.3.1. Frequency of transgressive segregants for six traits in Cross 1 (DSb 23 xMACS 1575)

3.3.1.1 Days to 50% flowering

Amongthe two parents, MACS 1575 was early flowering (38 days) and DSb 23 was late flowering (45 days). In F_2 segregating generation the average days taken for 50% flowering was 41 days. Twenty-onetransgressive segregants with early flowering were obtained which accounted for 7.66%. These transgressive segregants recorded a range of 38-40 days for days 50% flowering.

3.3.1.2 Days to maturity

Amongthe two parents, MACS 1575 was early maturing with 84 days and DSb 23 was late which recorded 95 days to mature. In F_2 segregating generation the average days to maturity was 95 days with standard deviation of 17.22 days. The number of transgressive segregants obtained was 45 (16.42%). The mean of superior segregants ranged from 79-90 days for maturity.

3.3.1.3 Plant height

The mean values of parents DSb 23 and MACS 1575 for plant height were 50.4 cm and 41 cm, respectively. In F_2 segregating generation the average plant height was 43.85 cm.

Number of transgressive segregants obtained was 33 with frequency of 12.04 per cent and recorded a range of 55-76 cm for plant height.

3.3.1.4 Pods per plant

It is evident from Table 8 that the parental lines DSb 23 and MACS 1575 recorded mean of 74.6 and 38 pods per plant respectively. In F_2 segregating generation the average number of pods per plant was 66.23 with standard deviation of 30.19 pods per plant. Pods per plant in cross 1 recorded highest frequency of transgressive segregants (42 plants out of 274 plants) compared to other studied characters, which accounted for 15.33%. Transgressive segregants for pods per plant ranged from 97-160 pods.

3.3.1.5 100 Seed weight

In F_2 segregating generation the average of 100 seed weight was 11.43 g with standard deviation 2.47 g. Data presented in Table 8, showed that the mean value of 100 seed weight in parental lines DSb 23 and MACS 1575 were 12.8 g and 13.7 g, respectively. The number of transgressive segregants in F_2 for 100 seed weight was 38 with the frequency of 13.87 percent with a range of 14-15.5 g.

Table 6: Per cent transgressive segregants for different characters in F_2 populations of the two crosses of soybean

Characters	**Cross**			**Transgressive segregants**	
		Mean	**SD**	**Number of plants**	**%**
Days to 50% flowering	DSb 23 x MACS 1575	41	2.89	21	7.66
	DSb 23 x MACS 1460	40	5.56	43	8.95
Days to maturity	DSb 23 x MACS 1575	95	17.22	45	16.42
	DSb 23 x MACS 1460	95	18.23	63	13.12
Plant height (cm)	DSb 23 x MACS 1575	43.85	10.54	33	12.04
	DSb 23 x MACS 1460	41.28	7.54	66	13.75
Pods per plant	DSb 23 x MACS 1575	66.23	30.19	42	15.33
	DSb 23 x MACS 1460	73.87	29.92	65	13.54

100 seed weight (g)	DSb 23 x MACS 1575	11.43	2.74	38	13.87
	DSb 23 x MACS 1460	10.34	2.47	57	11.87
Seed yield per plant (g)	DSb 23 x MACS 1575	16.50	9.26	29	10.58
	DSb 23 x MACS 1460	18.71	10.91	68	14.17

3.3.1.6 Seed yield per plant

From table 8, it is clear that the mean value for seed yield per plant in parent DSb 23 and MACS 1575 were 28.5 g and 14.7 g, respectively. In F_2 segregating generation the average of seed yield per plant was 16.50 g with standard deviation of 9.26 g. The number of transgressive segregants in F_2 was 29 with the frequency of 10.58 percent and these transgressive segregants ranged from 26.5 g to 52.5 g for seed yield per plant.

3.3.2 Frequency of transgressive segregants for six traits in Cross 2 (DSb 23 xMACS 1460)

3.3.2.1 Days to 50% flowering

Amongthe two parents, MACS 1460 was early flowering (39 days) and DSb 23 was late flowering (45 days). In F_2 segregating generation the average days taken for 50% flowering was 40 days with standard deviation of 5.56 days. Forty-three transgressive segregants with early flowering were obtained which accounted for 8.95%. These transgressive segregants recorded a range of 35-39 days.

3.3.2.2 Days to maturity

The mean values of days to maturity, in parents DSb 23 and MACS 1460 were 95 and 86 days, respectively. In F_2 segregating generation the average days to maturity was 95 days. The number of transgressive segregants obtained was 63 (13.12%). Transgressive segregants for days to maturity ranged from 78-88 days for maturity.

3.3.2.3 Plant height

The mean values of parents DSb 23 and MACS 1460 for plant height were 50.4 cm and 43.2 cm, respectively. In F_2 segregating generation the average plant height was 41.28

cm. Number of transgressive segregants obtained was 66 with frequency of 13.75 per cent and recorded a range of 49-60cm for plant height.

3.3.2.4 Pods per plant

It is evident from Table 8 that mean value of the parental lines DSb 23 and MACS 1460 was 74.6 and 44.2 pods per plant respectively. In F_2 segregating generation the average number of pods per plant was 73.87 with standard deviation of 29.92 pods per plant. The number of transgressive segregants obtained was 65 (13.54%). Transgressive segregants for pods per plant ranged from 104-175 pods.

3.3.2.5 100 Seed weight

In F_2 segregating generation the average of 100 seed weight was 10.34 g. Data presented in Table 8, showed that the mean value of 100 seed weight in parent DSb 23 and MACS 1460 were 12.8 and 13.3 g, respectively. The number of transgressive segregants in F_2 for 100 seed weight was 57 with the frequency of 11.87 percent and ranged between 16-20.5 g.

3.3.2.6 Seed yield per plant

From Table 8, it is clear that the mean value of parent DSb 23 and MACS 1460 recorded 28.5 g and 15.2 g seed yield per plant, respectively. In F_2 segregating generation the average of seed yield per plant was 18.71 g with standard deviation of 10.91 g. The number of transgressive segregants in F_2 was 68 with the frequency of 14.17 percent and mean of these superior segregates ranged between 30-55 g.

From the above information, it can be concluded that high frequency of favourable segregants were identified in F_2 generation of cross 1 for pods per plant (15.33%) followed by 100 seed weight (13.87%) and days to maturity (16.42%) and in cross 2, high frequency of favourable segregants in F_2 generation were observed for days to 50% flowering (8.95%) followed by plant height (13.75%) and seed yield per plant (14.17%).

4. Discussion

4.1. Inheritance of qualitative traits in segregating generation

The present study was aimed at understanding the segregation behaviour for qualitative characters in F_2 generation to understand the population structure with different crosses emerging from different genetic back ground used as parents. Among both the crosses, the parents used were having contrasting characters for hilum colour, in addition, parents in cross 1 (DSb 23 x MACS 1575) were having contrasting characteristics for leaf shape i.e., DSb 23 was bearing broad leaves and MACS 1575 was bearing narrow leaves and in cross 2 (DSb 23 x MACS 1460) parents differed with contrasting characteristics for flower colour i.e., DSb 23 was having purple coloured flowers whereas MACS 1460 was having white flowers which made it easy for identification and recording the data in F_2 generation. From Table 2and Table 4 it is clear that the calculated χ 2 value indicated that the observed value for leaf shape and flower colour fitted well with an expected ratio of 3 broad: 1 narrow for leaf shape and 3 purple: 1 white for flower colour, respectively, which indicated that these characters are controlled by a single dominant gene. Similar results were observed by Takahashi (1934) and Sawada (1988) for leaf shape in soybean. Nguyen (2012), Vu (2013) and Vu et al. (2017) reported that single gene with complete dominant effect is responsible for flower colour in mung bean. Calculated χ 2 value for hilum colour fitted well with an expected ratio of 9 black: 7 brown colour which indicated that this character is controlled by two genes with complementary gene action. Pipolo et al. (2015) reported that hilum colour is controlled by two genes segregating independently in soybean. Table 3 and Table 5 represent the χ^2 value and probabilities for segregation of hilum colour in cross 1 and cross 2 respectively. The above information will help the plant breeder to understand the inheritance of various qualitative characters of economic importance and their use as marker characters in the varietal identification as also in formulating a suitable breeding programme to evolve new superior varieties.

4.2. Identification of transgressive segregants

Recombination breeding is one of the important breeding methods to incorporate and to get the favourable quantitative and qualitative traits for both the parents in its progenies. 75 Transgressive segregation refers to the phenomenon through which we get variation in F_2 or later generation outside the range of both the parents. Selection in F_2 generation for transgressive segregants is likely to help the breeder to pick out the most favourable genotypes. In the present investigation, 754 progeny lines from crosses viz., DSb 23 x MACS 1575 and DSb 23 x MACS 1460 have generated greater variability and useful segregants. Transgressive segregants were recorded in each of the two crosses for yield and yield contributing traits based on mean ± 1 standard deviation. Per cent transgressive segregants in F_2 population of both the crosses for days to 50% flowering, days to maturity, plant height, pods per plant, 100 seed weight and seed yield per plant are given in Table 6. In general, high frequency of favourable segregants were identified in F_2 generation of cross 1 for pods per plant (15.33%), 100 seed weight (13.87%) and days to maturity (16.42%) and in cross 2, high frequency of favourable segregants in F_2 generation were observed for days to 50% flowering (8.95%), plant height (13.75%) and seed yield per plant (14.17%). This could be due to fact that parents used in these two crosses were diverse for the characters studied. Karkute and Girase (2002) observed highest transgressive segregants for plant height (27%) followed by pods per plant and seed yield per plant in chickpea. Massey et al. (2008) reported transgressive segregation in F_2 generations of soybean for days to maturity. Rajavindran*et al*. (2000) reported high frequency of transgressive segregants for plant height (16.88%) and seed yield per plant (13.92%) in sesame. Aditya et al. (2013) reported transgressive segregants in F_2 and F_3 generations for plant height, number of pods per plant 100 seed weight and grain yield per plant in soybean. For yield and its component traits viz., days to 50% flowering, days to maturity, plant height, pods per plant and 100 seed weight considered simultaneously, superior segregants were identified in each of the two crosses. Transgressive segregants for three or more yield component traits were identified as superior transgressive segregants. The possible explanation of simultaneous transgression for two or more yield components is that yield is dependent on yield attributing characters. The details of which are given in Table 7.

Table 7: Superior transgressive segregants identified with rust reaction

Sl. No.	Cross	Plant no.	Days to 50% flowering	Days to maturity	Pods per plant	100 seed weight (g)	Seed yield per plant (g)
1	DSb 23 x MACS 1575	9	38	85	65	13.5	16.5
		26	38	81	96	11.5	28.5
		35	38	80	144	14	35
		38	40	84	108	12.5	29.5
		74	40	86	122	14.5	34
		88	38	80	70	11	16
		92	40	83	99	13.5	30.5
		134	38	82	55	10.5	15
		267	39	84	110	12	33.5

	Mean±SD		41±2.89	95±17.22	66.23± 30.19	11.43± 2.74	16.50±9.26
2	DSb 23 x MACS 1460	21	38	82	111	13	33.5
		23	38	84	123	12.5	37.5
		24	38	85	141	13.5	41.5
		60	35	81	93	11.5	28.5
		85	38	79	114	11	34.5
		201	38	85	100	12.5	31.5
		205	38	79	104	12.5	32
		265	38	81	110	13.5	36
		274	38	83	104	10.5	29.5
	Mean±SD		40±5.56	95±18.23	73.87± 29.92	10.34± 2.47	18.71± 10.91
3	DSb 23 (Parent)	-	41.2	95.6	74.6	12.8	28.5
4	MACS 1575 (Parent)	-	38.4	84.8	38	11.7	12.7
5	MACS 1460 (Parent)	-	39.8	81.8	44.2	13.3	14.2

5. Conclusion

The F_2 populations of the crosses viz., DSb 23 x MACS1575 (cross 1) and DSb 23 x MACS 1460 (cross 2) were grown and observations were recorded on days to 50% flowering, days to maturity, plant height, number of pods per plant, hundred seed weight and seed yield per plant on individual plants from each cross. In addition to this, observations on some qualitative traits like flower colour, leaf shape and hilum colour were recorded.

From the inheritance study, it was observed that flower colour and leaf shape were under monogenic gene action. Purple flower colour was dominant over white, broad leaf shape was dominant over narrow and hilum colour was controlled by two genes with complementary gene action.

High frequency of favourable segregants were identified in F_2 generation of cross 1 for pods per plant (15.33%), 100 seed weight (13.87%) and days to maturity (16.42%) and in cross 2, high frequency of favourable segregants in F_2 generation were observed for days to 50% flowering (8.95%), plant height (13.75%) and seed yield per plant (14.17%).

Therefore, these qualitative traits can be used as morphological markers in molecular breeding and the superior segregants identified for yield and its component traits need to be further tested for their superiority in replicated trails.

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