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Genetic Diversity Studies in MAGIC Population of Soybean (*Glycine max* (L.) Merrill) Based on Mahalanobis D² Distance

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Authors' contributions

This work was carried out in collaboration among all authors. Author BRS performed the research, managed the literature searches, performed statistical analysis and wrote the first draft of the manuscript. Author VS designed the study, wrote the protocol and managed the analyses of the study. Authors MRR and SV supervised the experiment, monitored writing the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Soybean MAGIC lines are highly variable breeding material which utilizes both recent and historic genetic recombination events. Present investigation was carried out to assess the genetic diversity present among 95 soybean MAGIC lines along with six checks for 10 different quantitative traits. All the genotypes were grouped into 16 clusters by performing Tocher's clustering method using Mahalanobis D² distance. Cluster I was the largest comprising of 30 genotypes followed by cluster II (23 genotypes), cluster X (15 genotypes) and cluster IX (9 genotypes). The maximum genetic distance (D²) was observed between cluster XI and XV (168.37) followed by cluster III and XV

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(164.3), cluster X and XV (149.64) as well as between cluster XII and cluster XVI (145.99). The cluster mean for most of the traits were high in cluster I and cluster XVI. Oil content contributed maximum (23.86%) towards total genetic diversity followed by number of pods plant⁻¹ (18.97%), seed yield plant⁻¹ (18.63%), 100 seed weight (11.05%) and number of branches plant⁻¹ (10.16%) traits. The soybean MAGIC lines belong to the cluster XI (6-120) and cluster XV (6-30, 6-31, 6-5) were found to be the most divergent hence can be utilised in the recombination breeding programs to exploit maximum heterosis.

Keywords: Soybean; MAGIC lines; Genetic diversity; Mahalanobis D² statistics.

1. INTRODUCTION

Soybean is a diploid species with 2x = 40, belongs to the family Fabaceae and sub-family Favoideae. It is well known as miracle bean or golden bean due to its enormous economic and nutritional values [1]. Globally. India stands fifth in the production of 13.78 MT and having a productivity of 1.2 tonnes per ha which is much lower than the world productivity of 2.8 tonnes per ha [2]. Presence of narrow genetic base, influence of biotic and abiotic stresses, partial exploitation of heterotic potential and poor management practices are few causes for lower yielding ability in Indian soybean cultivars [3]. Hence, the present situation demands for creation and improvement of sufficient genetic diversity in the crop to exploit heterosis and to introgress desirable genes in the present cultivars for achieving higher yield potential. There are many methods to create genetic diversity among which, hybridisation is most commonly used [4]. A multi-way cross hybrid such as MAGIC (Multi-parent Advanced Generation Inter-Cross population) lines are tailor-made source of germplasm which undergoes heavy genetic recombination and also contribute a major variability to the crop gene pool [5]. Multivariate analysis such as Mahalanobis D² statistics is highly used in the genetic divergence study which groups the genotypes such that more diverse genotypes are grouped into most distant clusters and also useful in the assessment of the relative contribution of each trait towards the total divergence [6]. Moreover it provides the insight for selection of appropriate parents for combining new alleles of the desirable character in the crop improvement programme [7]. Hence, the present study was undertaken to assess the magnitude of genetic divergence among selected soybean MAGIC lines in order to utilise them in the future breeding programs to enhance the yielding ability of the crop.

2. MATERIAL AND METHODS

2.1 Experimental Site

The present investigation was carried out at the research farm. ARS (Adilabad). PJTS Agricultural University, Hyderabad under rainfed conditions during south west monsoon season, 2019. The experimental material consisted of 95 soybean MAGIC lines (F $_5$ generation) with six checks viz., EC333901, EC546882, EC572109, NRC86, JS 335 and Basara among which first five checks are founder parents of the studied MAGIC lines and Basara is the local check of Telangana. All the entries were evaluated in Randomised Block Design (RBD) in two replications by keeping inter and intra-row spacing of 45 and 10 cm respectively. 95 MAGIC lines are assigned at random within blocks of adjacent replications, each treatment once per block with checks repeated after every 30 lines.

2.2 Observations Recorded

The observations were recorded on whole plot basis for days to 50 % flowering and days to maturity whereas, data were recorded for plant height (cm), number of branches plant⁻¹, number of pods plant⁻¹, 100 seed weight (g), seed yield plant¹ (g), and harvest index (%)traits based on five randomly selected plants of each entry per replication. The protein content (%) and oil content (%) were estimated as per protocols of AOAC (1990) [8]. The recorded data were subjected to analysis of variance. Genetic divergence analysis was done by using Mahalanobis D2 - statistics [9]. Clustering of genotypes was done according to Tocher's method [10]. Intra and inter-cluster distances generated were used to describe the relationship among the genotypes, formula proposed by [11]. dissimilarity coefficient between the The genotypes was arranged into reasonable hierarchal system and estimated by using D2 value as suggested by [12].

2.3 Statistical Package

Recorded data for eight morphological traits and two qualitative traits were subjected to Analysis of variance by the software Windostat version 9.3 of Indostat services.

3. RESULTS AND DISCUSSION

3.1 Grouping of Accessions into Various Clusters

Based on D^2 values, the grouping of 101 soybean accessions was carried out by Tocher's method. A total of 16 clusters were constructed among which, seven clusters were containing more than one genotypes and the remaining nine were of solitary clusters indicating that the genotypes are highly diverse in nature. The accessions viz., 10-06, 10-53, EC572109, 9-144, 8-53, 7-94, 6-120,7-112 and Basara belongs to clusters III, IV, V, VI, VII, VIII, XI, XIII and cluster XVI respectively indicating their distinct genotypic composition from the remaining genotypes. Cluster I was found to be the largest comprising of 30 genotypes followed by cluster II (23 genotypes), cluster X (15 genotypes), cluster IX (9 genotypes), as well as cluster XII and XIV

with six genotypes each and cluster XV comprising of three genotypes are presented in Table 1.

The Indian cultivars such as NRC 86, JS 335 was grouped with the exotic cultivar EC546882 in the cluster I, which indicates that the genetic diversity has no relation with the geographical diversity, and the genetic diversity may have resulted from selection and genetic drift rather than merely geographic distribution factor. The result is in agreement with the result obtained by [13] and [14].

3.2 Average Inter and Intra-cluster Distances

The intra-cluster D^2 values were observed ranging from zero to 57.59. Among 16 clusters, the maximum intra-cluster distance was recorded in cluster XIV (57.59) followed by cluster X (37.31), cluster IX (33.59), cluster XII (33.27), cluster XV (23.8), cluster II (21.98) and cluster I (20.77), indicating that some amount of genetic divergence exist in the genotypes belonging to these clusters (Table 2). Carefully selection of the genotypes from these clusters may be used in further recombination breeding program.

Table 1. Grouping of all the soybean accessions into different clusters based on D² values

Clusters	Accessions	Total
Cluster I	7-92, 10-34, 8-68, 9-97, 8-107, 8-41, 10-51, 8-67, 10-10, 8-29, 6-100, 7- 144, 10-32, 9-141, 9-132, 7-4, 9-149, NRC 86, 8-134, 8-6, 10-33, 9-24, 6-43, 8-99, 9-115, 9-14, 9-94, 9-116, EC546882, JS 335	30
Cluster II	7-85, 7-141, 7-124, 9-145, 10-48, 8-95, 7-78, 7-53, EC333901, 10-41, 9- 120, 7-122, 10-1, 10-2, 7-82, 9-131, 8-92, 10-52, 9-125, 9-91, 10-24, 9- 122, 7-126	23
Cluster III	10-06	1
Cluster IV	10-53	1
Cluster V	EC572109	1
Cluster VI	9-144	1
Cluster VII	8-53	1
Cluster VIII	7-94	1
Cluster IX	10-35,10-54, 9-12, 10-50, 10-3, 8-125, 6-32, 8-98, 8-106	9
Cluster X	8-11, 8-13, 9-143, 9-135, 10-42, 8-69, 7-51, 7-80, 8-42, 7-3, 8-86, 8-120, 10-36, 6-48, 9-114	15
Cluster XI	6-120	1
Cluster XII	6-97, 7-36, 6-23, 6-124, 6-46, 10-31	6
Cluster XIII	7-112	1
Cluster XIV	6-62, 6-68, 8-118, 10-38, 8-137, 8-39	6
Cluster XV	6-30, 6-31, 6-5	3
Cluster XVI	Basara	1

The intra-cluster distance of remaining ten clusters was zero, as these clusters comprised of single genotypes each.

The inter-cluster distance was ranging from 6.36 to 168.37. The highest inter-cluster distance was observed between cluster XI and XV (168.37) followed by cluster III and XV (164.3), cluster X and XV (149.64) and between cluster XII and cluster XVI (145.99). Similarly, the lowest inter-cluster distance was observed between cluster IV and VII (6.36) followed by cluster V and VIII(6.65), cluster VIII and XIII (13.76), cluster III and V (17.64) as well as cluster V and XIII with value of 19.41. More the distance (D²) between the clusters, more the genetic divergence between the accessions belonaina that clusters. to Crossing between the genotypes of diverse clusters results into more variability in the gene pool. Similar results were also reported in soybean by [15]. The soybean MAGIC lines belong to the cluster XI (6-120) and cluster XV (6-30, 6-31, 6-5) were found to be the most divergent therefore may be used in the recombination breeding programs to exploit the maximum heterosis (Table 2).

3.3 Cluster Means of the Characters

A considerable difference was noticed among the cluster means for all the characters (Table 3.). The cluster mean was found highest for number of pods plant⁻¹(78), days to 50 % flowering (47) and days to maturity (114) in cluster XVI while the lowest mean was observed in cluster XV (28.77), cluster XIII (35) and cluster XIV (95.42) for the respective characters. The maximum cluster mean for plant height (68.33), 100 seed weight (16.12) and protein content (40.55) was observed in cluster XII while lowest mean was observed in cluster XV (44.07), cluster XVI (10.45) and cluster VIII (36.15) for the respective characters. Cluster mean for number of branches plant⁻¹ was highest (9.4) in cluster XV and lowest (3) in cluster III. The maximum (19.28) and the minimum [15] cluster mean for seed yield plant⁻¹ was observed in cluster XI and XV respectively. The cluster mean for oil content was highest (20.21) in cluster X and lowest in cluster VIII (16.45). Harvest index exhibited the highest (44.6) and lowest (33.33) cluster mean value in cluster XII and cluster IV respectively. Similar results were also reported in soybean by [16] and [17].

The cluster mean for most of the characters in the cluster I and cluster XVI were noticed to be

higher than the overall cluster mean. Hence the soybean accessions such as Basara, EC546882, NRC 86, JS 335, 7-92, 10-34, 8-68, 9-97, 8-107, 8-41, 10-51, 8-67, 6-100, 7-144 and other MAGIC lines belongs to these clusters may directly be utilized in the breeding program to enhance the production potential of this crop (Table 3).

3.4 The Contribution of Each Traits towards Total Divergence

The contribution of each trait towards total diversity is computed based upon the number of times the character appears in the first rank. The trait oil content appeared a maximum of 1205 times in the first rank indicating maximum contribution towards the total diversity. Whereas, harvest index appeared the lowest of 66 times in the first rank suggesting the less contribution of it towards total diversity. The relative contribution of each trait towards total diversity is mentioned in Table 4.

The results revealed the highest contribution of oil content (23.86%) towards total diversity followed by number of pods plant¹ (18.97%), seed yield plant¹ (18.63%), 100 seed weight (11.05%) and number of branches plant¹ (10.16%) traits. Whereas, the contribution of harvest index (1.31%) was found to be lowest towards total diversity followed by plant height (2.48%), days to 50 % flowering (2.81%), days to maturity (4.36%) and protein content (6.38%).

The result suggests that the soybean accession selected for the present study are mostly divergent for oil content, number of pods plant⁻¹ traits, and seed yield plant⁻¹ and 100 seed weight as these four characters contributed 72.51% to the total diversity. The present findings are in agreement with the results obtained by [18] for oil content, [19] for seed yield plant⁻¹ and number of pods plant⁻¹, [14] for days to 50 % flowering, Dubey et al. [20] for protein content and [13] for plant height, harvest index and 100 seed weight traits.

The accessions belong to the most distant clusters *viz.*, (III and XV), (XI and XV) and (X and XV) and cluster I and II which having higher intra-cluster distance may be utilized directly or as a parent in the soybean hybridization program by emphasizing selection for oil content and number of pods plant⁻¹ to improve yield potential of the crop.

Clusters		11	III	IV	V	VI	VII	VIII	IX	Х	XI	XII	XIII	XIV	XV	XVI
l	20.77	54.78	27.41	27.42	26.27	32.7	32.9	39.29	32.31	43	38.66	61.03	47.48	57.22	112.13	56.95
11		21.98	56.47	48.39	52.51	40.16	45.46	65.34	55.15	92.58	119.65	54.11	96.91	96.39	99.51	113.03
111			0	52.89	17.64	21.6	65.21	30.29	23.58	70.85	40.02	78.76	41.6	66.58	164.3	79.12
IV				0	55.94	49.08	6.36	81.26	48.58	35.01	76.64	36.65	93.28	76.59	96.2	65.05
v					0	21.68	60	6.65	27.04	62.11	33.02	90.44	19.41	43.59	123.36	67.7
VI						0	51.4	29.2	26.79	80.05	70.25	91.28	56.39	79.28	126.52	46.24
VII							0	81.2	57.63	37.98	91.92	42.36	95.23	88.71	89.73	68.65
VIII								0	37.72	84.29	40.78	112.29	13.76	53.82	125.59	73.84
IX									33.59	70.94	53.23	77.74	52.13	62.25	129.01	66
х										37.31	57.82	72.91	87.71	83.7	149.64	86.17
XI											0	113.75	33.62	54.19	168.37	75.7
XII												33.27	123.59	103.71	117.83	145.99
XIII													0	50.12	129.46	102.78
XIV														57.59	113.89	104.6
XV															23.8	140.05
XVI																0

Table 2. Average inter and intra-cluster distances (D²) values of 101 soybean accessions for ten characters

^{*} Diagonal bolded values indicate intra-cluster distances *Above diagonal values are inter-cluster distances.

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Clusters	Days to 50 % flowering	Days to maturity	Plant height (cm)	Number of branches plant ⁻¹	Number of pods plant ⁻¹	100 seed weight (g)	Seed yield plant ⁻¹ (g)	Oil content (%)	Protein content (%)	Harvest Index (%)
Cluster I	41.13	106.45	60.30	4.64	54.60	14.10	13.10	18.40	38.89	39.96
Cluster II	41.00	106.46	63.40	4.59	29.90	12.30	8.20	17.70	38.57	36.19
Cluster III	41.00	106.00	60.20	3.00	60.40	14.60	12.80	16.90	37.85	38.52
Cluster IV	41.00	108.00	59.80	5.00	50.40	14.20	9.71	20.00	40.05	33.33
Cluster V	38.00	102.00	60.60	4.60	52.40	12.00	13.70	17.10	36.65	38.72
Cluster VI	41.00	112.00	58.80	4.50	56.00	11.20	11.70	16.70	39.30	37.50
Cluster VII	40.00	109.00	62.20	5.20	44.80	13.20	9.33	20.20	39.25	41.16
Cluster VIII	39.00	106.00	68.00	5.20	51.90	12.30	15.50	16.50	36.15	40.90
Cluster IX	40.78	105.17	65.40	4.47	56.30	13.30	11.70	17.10	39.08	38.75
Cluster X	39.00	102.97	64.50	4.44	51.50	13.40	14.50	20.20	39.66	40.85
Cluster XI	42.00	101.00	56.60	4.40	64.60	15.10	19.30	17.90	38.80	42.48
Cluster XII	41.75	103.92	68.30	4.18	30.20	16.20	9.58	19.40	40.55	36.40
Cluster XIII	35.00	104.00	58.20	5.30	55.00	14.90	16.40	16.60	36.45	44.60
Cluster XIV	37.17	95.42	62.30	5.98	51.10	13.80	12.50	17.50	38.26	37.57
Cluster XV	41.67	105.67	44.10	9.40	28.80	14.80	6.70	18.50	38.93	39.13
Cluster XVI	47.00	114.00	67.20	6.70	78.00	10.50	13.10	18.20	39.95	41.08
Overall mean	40.41	105.50	61.20	5.10	51.00	13.50	12.40	18.00	38.65	39.20

Table 3. Cluster means and contribution towards total diversity for 10 yield and yield contributing traits in 101 soybean accessions

S.No.	Character	Number of times ranked first	Contribution (%)
1	Plant height (cm)	125	2.48
2	Number of branches plant ⁻¹	513	10.16
3	Number of pods plant ⁻¹	958	18.97
4	Days to 50 % flowering	142	2.81
5	Days to maturity	220	4.36
6	100 seed weight (g)	558	11.05
7	Seed yield plant ⁻¹ (g)	941	18.63
8	Oil content (%)	1205	23.86
9	Protein content (%)	322	6.38
10	Harvest Index (%)	66	1.31

. Table 4. Relative contribution of different characters towards total genetic diversity

4. CONCLUSION

From the findings of the present investigation it is evident that the selected soybean MAGIC lines are highly diverse as they have undergone heavy genetic recombinations during development which form a total of 16 clusters. The accessions belong to the most distant clusters viz., (XI and XV), (III and XV) and (X and XV) and cluster XIV and X which were having higher intra-cluster distance may be utilized directly or as parents in the soybean hybridization program by emphasizing selection for oil content and number of pods plant⁻¹ to improve yield potential of the crop.

5. RECOMMENDATIONS

Rational use of both the morphological and molecular clustering pattern may be useful for selecting desirable genotypes which can be utilized in future breeding programs for exploiting heterosis or in the introgression of genes for biotic and abiotic stress tolerance.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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