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Study of Genotypic and Phenotypic Variation in F_2 Segregating Generation of Okra [*Abelmoschus esculentus* (L.) Moench]

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ABSTRACT

Background: Hybrid breeding in Okra [*Abelmoschus esculentus* (L.) Moench] is important to improve productivity of this crop. Given that a breeder's primary goal is production, it's critical to understand the connections between the traits that have a direct and indirect impact on yield.

Methods: 45 hybrids generated by crossing 10 diverse parents in structured mating design were evaluated to estimate the magnitude of their genetic variability and heritability in F_{2s}. Mean performance across three replications were calculated for 21 traits and is assessed for their effect on yield. The recorded data were statistically analyzed at 5% level of significance by multivariate analysis using principal component and hierarchical cluster.

Result: For all of the analyzed traits, the analysis of variance revealed a highly significant difference between population. A greater difference between PCV and GCV estimates for days to anthesis, days to 50% flowering and days to first picking indicates a greater degree of environmental control for these traits. Traits such as days to anthesis, days to 50% flowering and days to first picking shows low to moderate heritability. These findings show that there is enough genetic diversity in these parameters to support selection of better accessions. Using cluster analysis and principal component analysis (PCA), crosses were divided into groups based on their performance and the identification of the most discriminating attribute that accounted for the greatest variability. It revealed that yield per plant is highly associated with days to first picking, days to 50% flowering, days to anthesis, plant height and shoot length. Further, principal component shows 1000-seed weight and germination percentage followed by days to 50% flowering as the most discriminating trait. Hence, selection for any trait would favor future hybrids breeding programmes.

Key words: Cluster analysis, Genetic variability, Okra, PCA.

INTRODUCTION

To enhance the productivity of crops, breeding programmes always emphasize on selection of better genotypes and hybrids, especially in the mineral rich crops which are important food in human diet. In 2021, India is the largest producer of Okra with overall contribution of 60% of the total world production, leading producers being the state of Gujarat and Maharashtra (FAOSTAT, 2021). Native to Africa it contains carbohydrates, protein, fiber, minerals and vitamins (Sabitha *et al*., 2012) and is now widely grown in Southern Europe, Middle East Asia. India has a total area of 10,436 with a productivity of 17.96 tons/hectare (Kumar *et al*., 2020). Its short lifecycle, fast growth period and photo-insensitive nature allow geneticists and breeders to grow two or three crops annually and shorten the time it takes to develop a genetic trait.

Conventional breeding and selection program of any crop emphasizes optimization of yield and yield specific quantitative traits for crop improvement. All traits related to yield, earliness and yield enhancement in okra are quantitative in nature. Such characters are controlled by polygenes and are much influenced by environmental fluctuations (Meena *et al*., 2017).

Germplasm serves as a great source of base population and offers capacity for extensive variety and genetic improvement of any crop mostly depends on the amount of genetic variability available in the population (Johnson *et al*., 1955). The development of inbred lines that resemble the parental lines of hybrids is facilitated by the selection of particular features in the F_{1s} and selfing of those over generations. These inbreds with desired traits, such as high yield potential, can be employed as parents for hybrid varieties as well as high-yielding types. The most effective selection of desirable features for creating optimum genotypes is crucial for increasing genetic yield potential.

The estimates of h^2 (Heritability) help the plant breeder in a selection of elite genotypes from diverse genetic populations. When h^2 is investigated in conjunction with genetic advances, heritable variation may be used more

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precisely and efficiently (Johnson *et al*., 1955). PCA is a crucial multivariate method for measuring genotype genetic variation and investigating character associations (Abdi and Williams, 2010).

Based on these facts, the present study was designed to quantify the extent of genetic variability available for total yield per plant and its component traits in F_{2s} of okra, which were developed by selfing of F_{1s} of selected parental genotypes and to estimate the genetic gain that can be made by selection.

MATERIALS AND METHODS

Field trials were carried out at the Chaudhary Charan Singh University Research Farm, Department of Genetics and Plant Breeding, Chaudhary Charan Singh University, Meerut, during the spring seasons of 2020 to 2023. Ten parental genotypes of *A. esculentus* with various morphological and productive traits were procured from NBPGR, India (Table 1). 45 hybrids of F_1 generation were created in rabi season of 2021-22 by crossing the selected ten genotypes in all conceivable ways. 45 F_2 progenies were grown during *rabi* 2022-23 at the experimental plots to maintain a healthy crop stand, the typical cultural and agronomic guidelines were followed and need-based plant protection measures were employed. The observations were recorded on 21 qualitative and quantitative traits *viz*., days to germination, days to anthesis, days to 50% flowering, number of nodes/plants, distance of nodes/plants, number of primary branch, number of second branch, fruit length, fruit width, days to first picking, number of pod/plant, length of tapering, weight of fruit (edible stage), number of seed/pod, shoot length, root length, root and shoot ratio, height of plants, 1000-seed weight, germination (%) and fruit yield/plant were recorded. The quality parameters were analyzed as per the methods of AOAC, (1984). Mean performance across for parents or F_2 were calculated for each trait and is assessed. The recorded data were statistically analyzed at 5% level of significance following the standard process as stated by Panse and Sukhatme (1967). Then, it was subjected to the following statistical analyses: Analysis of variance (ANOVA) for the design of experiment (Panse and Sukhatme, 1967), Coefficient of variation, Heritability and genetic advance (Burton and deVane, 1953) and Principal component and hierarchical analysis (Searle, 1961) by using Scilab (Campbell *et al*., 2010) to show the highest discriminating trait and level of relationship, respectively, among progenies. Burton and deVane (1953) suggested that the genetic coefficient of variation together with

Table 1: Ten parental genotypes, 45 F_{1s} and 45 F_{2s} used in this study.

S.no	Accession/Hybrid
Genotypes	
1	EC169420
2	EC169430
3	EC169435
4	EC169453
5	EC169462
6	EC169464
7	EC169468
8	EC169470
9	EC169474
10	EC169477
F1s and F2s	
C1	EC 169420 × EC169430
C2	EC 169420 × EC169435
C3	EC 169420 × EC169453
C4	EC 169420 × EC169462
C5	EC 169420 × EC169464
C6	EC 169420 × EC169468
C7	EC 169420 × EC169470
C8	EC 169420 × EC169474
C9	EC 169420 × EC169477
C10	EC 169430 × EC169435
C11	EC 169430 × EC169453
C12	EC 169430 × EC169462
C13	EC 169430 × EC169464
C14	EC 169430 × EC169468
C15	EC 169430 × EC169470
C16	EC 169430 × EC169474
C18	EC 169435 × EC169453
C19	EC 169435 × EC169462
C20	EC 169435 × EC169464
C21	EC 169435 × EC169468
C22	EC 169435 × EC169470
C23	EC 169435 × EC169474
C24	EC 169435 × EC169477
C25	EC 169453 × EC169462
C26	EC 169453 × EC169464
C27	EC 169453 × EC169468
C28	EC 169453 × EC169470
C29	EC 169453 × EC169474
C30	EC 169453 × EC169477
C31	EC 169462 × EC169464
C32	EC 169462 × EC169468
C33	EC 169462 × EC169470
C34	EC 169462 × EC169474
C35	EC 169462 × EC169477
C36	EC 169464 × EC169468
C37	EC 169464 × EC169470
C38	EC 169464 × EC169474
C39	EC 169464 × EC169477
C40	EC 169468 × EC169470
C41	EC 169468 × EC169474
C42	EC 169468 × EC169477
C43	EC169470 × EC169474
C44	EC169470 × EC169477
C45	EC 169474 × EC169477

heritability estimates gave a better picture of the extent of heritable variation. Heritability (h^2_{bs}) and genetic advance (GAM) estimates were interpreted as low, medium and high as per the classification of Johnson *et al.* (1955).

RESULTS AND DISCUSSION

Genotypic variability

The ANOVA showed a highly significant variation at a 5% level for all the traits (Table 2), indicating that the parents included in this investigation exhibited sufficient variability for all the characters studied. The estimates and magnitude of coefficient of genetic variability are presented in Table 2. In crop yield improvement programmes, mainly the genotype component of variation is important since only this component is transmitted to the next generation.

The proportional degree of variability between crop plant attributes is compared using the coefficient of variation (CV%) (Sharma, 1988). The highest coefficient of variation was recorded for the fruit width followed by number of pods/ plant and fruit length (Table 2). These results imply that the fruit width, number of pods/plant and fruit length, in that order, had higher amounts of exploitable genetic variability among the studied Okra F_{2s} attributes. It also suggests that choosing certain traits over others has a higher chance of leading to favorable advancement. Days to 50% flowering showed the lowest %CV, which indicates minimal exploitable genetic variability and as a result, offers less potential for advantageous advance in selecting when compared to other traits.

The phenotypic variance of the traits under study was divided into heritable (genotypic variance) and non-heritable (environmental variance) components (Table 2). The magnitude of genotypic variances was higher than their corresponding environmental variances for all the traits which indicates that the genotypic component of variation was the major contributor to total variation in the studied traits, except for days to anthesis, days to 50% flowering and days to first picking where environment variation was higher, indication of the environmental influence in these traits. The highest phenotypic and genotypic coefficient of variation (PCV and GCV) was obtained for the root length followed by root to shoot ratio shoot length and number of pods/plant, while, the least was recorded for days to anthesis, days to 50% flowering and days to first picking. Thus, a greater potential is expected in selecting root length, root to shoot ratio shoot length and number of pods/plant among the studied progenies. Similar type of variations in genotypic and phenotypic coefficient was reported by several other researchers (Ashraf *et al*., 2020; Shwetha *et al*., 2020; Ranga *et al*., 2021). Using the coefficient of variation (CV%), crop plant attribute proportional degrees of variability are compared. Other workers who made similar observations also reported them (Oyetunde and Ariyo, 2014; Makdoomi *et al*., 2018).

Low environmental effect is indicated by high heritability in the observed variation. High h^2_{bs} was recorded in all the traits, except for days to anthesis, days to 50% flowering and days to first picking which shows low to moderate heritability (Table 2). These findings show that there is enough genetic diversity in these traits to support selection of better accessions. High heritability combined with high genetic gain was observed for root length, shoot length and root to shoot ratio. This suggests that these traits are more additively expressed by genes than by the environment and that their inheritance is more common (Panse and Sukhatme, 1957). High heritability with moderate genetic gain was recorded for plant height, germination percentage and number of pods/plant. This indicated that the traits were governed by additional gene interaction. High heritability coupled with low genetic gain was recorded for days to anthesis, days to 50% flowering and days to first picking indicating non-additive gene action. Similar results were reported in recent studies (Walling *et al*., 2020; Temam *et al*., 2020 and Sandeep *et al*., 2022).

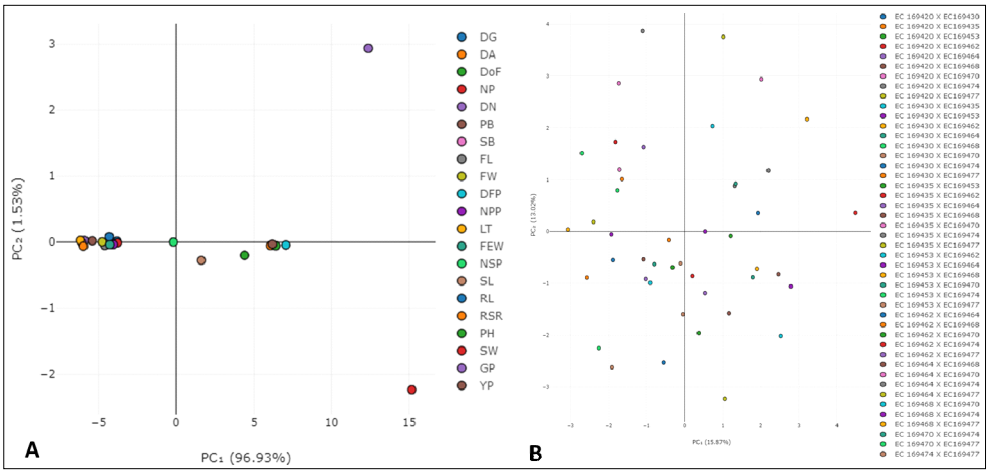

Fig 1: Loading of variables into PC_1 and PC_2 components of okra F_{2s} (A) Association an differences of 21 traits; (B) Effect of traits discrimination on progenies.

Table 2: Mean square and genetic parameters for some quantitative traits in okra F_{2s} combinedwith parental genotypes.

Traits	Mean	Range	CV%	PV	GV	EV	PCV%	GCV%	ECV%	h^2_{bs}(%)	GAM (%)	MSg
DG	9.5558	7.8-11.9	3.0047	1.0505	0.968	0.0824	10.7257	10.2963	3.004	92.1523	20.361	83.54**
DA	38.298	35-41.1	3.1463	3.307	1.4519	1.855	4.7483	3.5563	3.146	56.0942	5.4868	21.78**
DoF	39.297	36-42.1	2.9661	3.2449	1.3587	1.8862	4.5839	3.4948	2.966	58.1285	5.4889	23.69*
NP	9.6361	6.6-12.6	3.5941	2.096	1.9761	0.1199	15.0243	14.5881	3.593	94.2775	29.179	109.85**
DN	3.4691	2.6-4.3	3.6028	0.1441	0.1284	0.0156	10.9409	10.3307	3.600	89.1562	20.0943	23.91**
PB	4.9027	3.8-5.8	3.8931	0.2163	0.1799	0.0364	9.4864	8.6507	3.891	83.1582	16.2507	83.29**
SB	3.0204	2.5-3.7	3.8915	0.0854	0.0716	0.0138	9.6741	8.8569	3.889	83.819	16.7041	10.69*
FL	7.3136	6.1-8.1	4.0609	0.3615	0.2733	0.0882	8.2206	7.1476	4.061	75.5975	12.8021	4.27**
FW	6.7827	5.4-8.4	4.1063	0.836	0.7584	0.0776	13.4802	12.8395	4.107	90.7206	25.1923	57.97**
DFP	41.173	38.5-43.8	3.3954	2.8546	0.9002	1.9544	4.1035	2.3044	3.395	31.535	2.6657	386.19**
NPP	8.8493	5.6-12.2	4.0807	2.2968	2.1664	0.1304	17.1261	16.6329	4.081	94.3227	33.2769	243.44**
LT	2.7302	2.12-3.28	3.7863	0.0963	0.0856	0.0107	11.3683	10.7192	3.789	88.9073	20.8209	0.01
FEW	8.2224	6.8-10.4	3.9934	0.7542	0.6464	0.1078	10.5621	9.7781	3.993	85.7049	18.6476	15.65**
NSP	20.129	15.6-24.2	3.6947	5.0993	4.5462	0.5531	11.2183	10.5924	3.695	89.1529	20.603	13.74**
SL	25.445	15.3-40.3	3.6488	32.0697	31.2077	0.8621	22.2551	21.9539	3.649	97.3119	44.6131	1184.11**
RL	8.1292	3.94-13.44	3.9891	5.2159	5.1108	0.1052	28.0944	27.8097	3.990	97.9839	56.7076	330.39**
RSR	3.2917	1.9-4.5	3.6425	0.7286	0.7142	0.0144	25.9314	25.6743	3.646	98.0269	52.3647	26.26**
PH	33.584	21.7-49.3	3.5118	47.8983	46.5073	1.391	20.6076	20.3062	3.512	97.0959	41.2189	23.96**
SW	65.051	48.1-79.8	3.1068	80.9363	76.8517	4.0846	13.8298	13.4763	3.107	94.9533	27.0516	2147.33**
GP	57.036	39.5-81.1	3.1729	113.880	110.605	3.2749	18.7101	18.4391	3.173	97.1243	37.4343	476.56**
YP	38.682	36.1-41.9	3.1938	3.4087	1.8824	1.5263	4.7729	3.5468	3.194	55.2236	5.4297	83.54**

(DG= Days to germination; DA= Days to anthesis; DoF= Days to 50% flowering; NP= Number of nodes/plant; DN= Distance between nodes; PB= Number ofprimary branch; SB= Number of secondary branch; FL= Fruit length; FW= Fruit width; DFP= Days to first picking; NPP= Number of pods/plant; LT= Length of tapering; FEW= Edible fruit weight; NSP= Number of seeds/pod; SL+ shoot length; RL= Root length; RSR= Root shoot ratio; PH= Plant height; SW= 1000-seed weight; YP= Fruit yield per plant in kg; MSg: Mean square of genotypes).

Table 3: Eigen vectors and total percentage variation for the principal component axes of okra F_{2s}.

Traits*	PC_1	PC_2
DG	-3.8191	0.0155
DA	6.0713	-0.05374
DoF	6.4154	-0.05617
NP	-3.7882	-0.0108
DN	-5.9101	0.02473
PB	-5.4166	0.01964
SB	-6.0648	-0.00711
FL	-4.5915	-0.05275
FW	-4.7629	0.003646
DFP	7.0628	-0.04454
NPP	-4.0558	-0.03651
LT	-6.1653	0.02614
FEW	-4.2785	-0.03767
NSP	-0.1781	0.00061
SL	1.6187	-0.2767
RL	-4.3136	0.08118
RSR	-5.9748	-0.06234
PH	4.4134	-0.2004
SW	15.173	-2.236
GP	12.3652	2.9347
YP	6.1996	-0.03146
% of Variance	96.9345	1.5284
Cumulative (%)	96.9345	98.4629

(DG= Days to germination; DA= Days to anthesis; DoF= Days to 50% flowering; NP= Number of nodes/plant; DN= Distance between nodes; PB= Number of primary branch; SB= Number of secondary branch; FL= Fruit length; FW= Fruit width; DFP= Days to first picking; NPP= Number of pods/plant; LT= Length of tapering; FEW= Edible fruit weight; NSP= Number of seeds/pod; SL+ shoot length; RL= Root length; RSR= Root shoot ratio; PH= Plant height; SW= 1000-seed weight; YP= Fruit yield per plant in kg; MSg: Mean square of genotypes).

Principal component Analysis (PCA) and cluster analysis for F_2 generation

The first two components accounted for 98.46% of the cumulative variation in the population in F_{2s} (Table 3). PC1 accounted for 96.93% of the total variation and was also positively and highly associated with 1000-seed weight, plant height, days to 50% flowering and germination percentage. The PC2 explained 1.52% of the total variation and was positively related to germination percentage whereas 1000-seed weight was high but negative. The results revealed 1000-seed weight and germination percentage followed by days to 50% flowering as the most discriminating trait explaining greater variability in okra (Fig 1A). As shown in Fig 1A, which shows the score plot of both components to visualize association and differences of studied traits that yield per plant is highly associated with days to first picking, days to 50% flowering, days to anthesis, plant height and shoot length. This partly agrees with results by other authors (Amoatey *et al*., 2015; Ranga *et al*., 2021 and Sandeep *et al.,* 2022) who reported the high contribution of fruit length, test weight, number of seeds per fruit and fruit yield per plant towards total variation by bivariate analysis. Loading of the variables is shown in Fig 1B, which shows C4, C21, C22, C23, C24, C25 and C27 as the most promising progenies for further heterosis programmes.

F_{2s} (coded as C1 to C45; Table 1) were classified into nine clusters on the similarity axis based on PCA (Fig 2). Based on the result of the cluster analysis and a comparison of the means, it was shown that cluster H and cluster I expressed the best agronomic characteristics and yield potentials. However, cluster G, H and I had the lowest value for days to anthesis and days to 50% flowering. This is also an advantage because it encouraged earliness in fruit maturity of the progenies in the cluster.

The clustering pattern of different progenies did not follow their parental geographical distribution and was fairly random. This suggests that progenies of the same parental

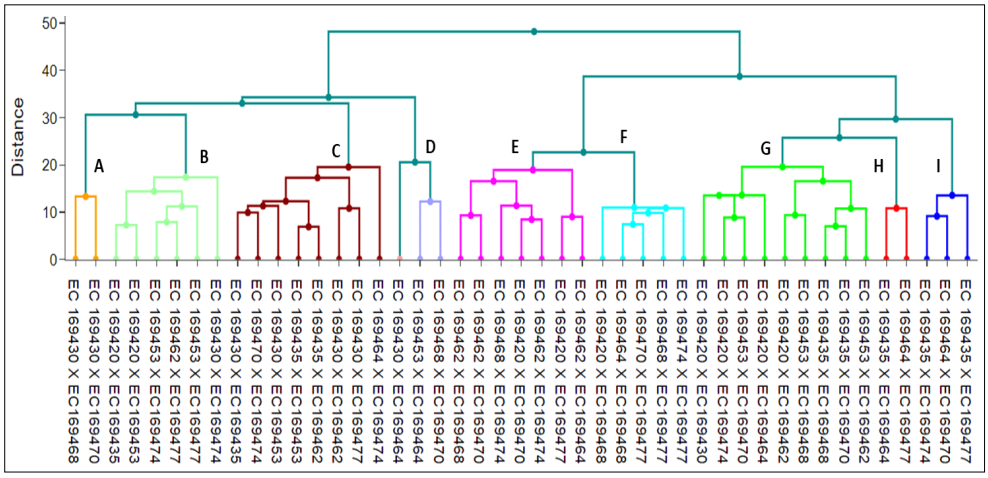

Fig 2: Dendrogram showing the classification of okra F_{2s} into nine major clusters (colored).

origin that are included in different clusters are an indication of the broad genetic base of the hybrids belonging to the origin. Osawaru *et al.* (2013) reported similar results in their genetic variability study among 53 accessions of West African okra (*Abelmoschus caillei*).

CONCLUSION

The genetic diversity studies about okra progenies quantitative traits based on the multivariate analysis using principal component and hierarchical clustering identified most important components in selection for higher yield in okra, explaining greater variability. The grouping of progenies into separate clusters shows that genetic variability in parental genotypes is relatively large. According to the findings, there is enough genetic variation among the progenies under study to justify selection for F_{2s}.

Conflict of interest

All authors declare that they have no conflicts of interest.

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