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Assessment of genetic diversity in exotic collections of fava bean (*Vicia faba* L.)

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Abstract
The present investigation "Assessment of genetic diversity in exotic collections of fava bean (*Vicia faba* L.)" was carried out to assess the extent of genetic variability and divergence of yield contributing characters of seed yield per plant in forty-six genotypes of fava bean at Vegetable Research Farm, Chandra Shekhar Azad University of Agriculture and Technology, Kanpur (U.P.) during *Rabi* 2019-20. The analysis of variance revealed highly significant differences among the forty-six fava bean genotypes for seed yield per plant traits indicating the presence of sufficient variability for all the traits under the study. All the traits indicating the presence of high degree of variability and better scope for further crop improvement. The observations for ten characters were recorded as well as multivariate technique; cluster analysis was applied. The fifty genotypes were grouped into eight distinct clusters on the basis of Euclidean distance. The maximum inter-cluster distances for seed yield were recorded between Cluster V & VII and Cluster V & VIII respectively, indicates the presence of wide diversity between these clusters. Therefore, genotypes from these clusters can be selected for hybridization programme to get desirable recombinants. Number of clusters per plant contributed the maximum per cent towards genetic divergence in forty-six genotypes of fava bean, indicates the presence of wide variability for these traits among the studied genotypes. Therefore, simultaneous selection for above traits is suggested for the improvement of seed yield per plant in fava bean.

Keywords: Fava bean, genetic diversity, cluster analysis

Introduction
Broad bean, horse bean, winter bean, Windsor bean, pigeon bean, and generally known as Bakla and Kala Matar in India are all names for the fava bean. Bionomically, fava pods belong to the fabaceae family, in the genus: *Vicia* scientifically called (*Vicia faba* L.; 2n = 2x = 12, 14) (Singh *et al*., 2013) [33]. It is a tough plant that can withstand cold temperatures. The only bean cultivated as a winter crop is the fava bean. It is widely assumed that it originated in North Africa and the South Caspian Sea (Tanno and Willcox, 2006), and that Arab traders brought it to India. Fava beans play a significant role in global agriculture due to its superior yield performance when compared to other grain legumes. It can also be utilized as a break crop in areas where grain monoculture is prevalent. Because of its biological nitrogen fixation capacity and improved weed and disease control in succeeding crops, it is a highly profitable crop (Preissel *et al*., 2015). The fava bean is a small annual, glabrous herb with erect stem, growing to the height of upto 6 feet plant. The fava bean features big white flowers that grow in clusters on short pedicles, honeybees play a role as pollinators. The pod of the fava bean is a light, green beaded fruit that matures to a blackish-brown colour (Lindemann and Glover, 2003). The green pod of the fava bean is consumed as a vegetable, while the dry seed is utilized as a grain legume. The seeds of the fava bean are high in protein (16%), carbohydrate (6%), fat (1%) and dietary fiber (32%). Fava bean has the highest crude protein content as well as the largest production of protein per hectare among the most widely farmed crops. It contains 18 gm of carbohydrate, 8 gm of protein, 0.7gm of total fat, 0.08 mg of thiamine, 12.0 mg of ascorbic acid, 50 mg of calcium, 25 mg of sodium, 332 mg of potassium, 0.1 gm. of saturated fat and 1.4 mg of iron per 100 gm of edible amount (fdc.nal.usda.gov, 2019) [12]. Fava bean was recognized by the Indian Council of Agriculture Research (ICAR) as a viable grain legume crop and was included in the AICRP programme. The fava bean is classified as the eighth most important grain legume by the Consultative Group on International Agricultural Research (CGIAR) (Sharifi, 2015) [29]. The fava bean is used in a variety of ways to soften stiff limbs, including as an ingredient and as a topical therapy. The seeds of the fava bean are a good source of L-DOPA, which is a precursor to dopamine and is used to treat

Parkinson's disease. It has anti-oxidant properties as well. For chronic disease prevention and health enhancement, the fava bean is a good dietary source of natural antioxidants (Oomah *et al*., 2006). Fresh pods and immature seeds of the Fava bean contain anti-nutritional elements such as polyphenols, which add a beany flavour (Bjerg *et al*., 1988) and are known to produce astringency. The vicine and co-vicine found in fava bean seeds cause haemolytic anaemia by oxidising erythrocytes. Heat treatment in boiling water, as well as pre-soaking, can diminish the action of anti-nutritional agents (Batra *et al*., 1990). The fava bean is a self-pollinating crop that is somewhat allogamous (5-20 percent). The presence of sufficient variety in a crop's fundamental genetic material is a need for starting a systemic breeding programme for agricultural improvement. The study of genetic diversity is crucial in breeding for broad adaption. It aids in the identification of appropriate parents for a successful hybridization scheme. Divergent groupings emerge as a result of hybridization, which is the primary goal of plant breeding programmes. D^2 approaches (based on multivariate analysis) developed by Mahalanobis are a useful tool for accessing genetic divergence and a good index of genetic variety. For many quantitative aspects, there is a lot of variability in fava bean genotypes, indicating that there is room for improvement.

Materials and Methods

The materials for present investigation comprised of forty-five fava bean genotypes and one varietal check i.e., Vikrant were grown in a randomized complete block design (Federer, 1956) during *Rabi* 2019-20 at Vegetable Research Farm, of Chandra Shekhar Azad University of Agriculture and Technology, Kanpur (U.P.), with recommended agronomical package of practices. Each genotype was sown in three row of 3 m length by adopting a spacing of 30 cm between rows and 10 cm between plants in the replication. The technique of random sampling was adopted for the observation of 10 quantitative characters namely Days to 50 per cent flowering days to maturity, plant height (cm), number of branches per plant, number of clusters per plant, pod length (cm), number of pods per plant, number of seeds per pod, 100-seed weight (g) and seed yield per plant (g).

The experimental data thus recorded on these characters were subjected to statistical and biometrical analysis for analysis of variance and variability (Fisher, 1925) and Genetic divergence and non-hierarchical Euclidean cluster analysis (Mahalanobis and Rao, 1952) [28].

Results and Discussion

Analysis of Variance

The analysis of variance (ANOVA) for ten characters is presented in Table 1. It is revealed that there were highly significant differences among the treatments for all the characters under study, showing wide range of variation in 46 genotypes of fava bean.

Table 1: Analysis of variance (ANOVA) for ten characters in forty-six fava bean genotypes.

Source of variation	d.f.	Days to 50% flowering	Days to maturity	Plant height (cm)	Number of branches per plant	Number of clusters per plant	Pod length (cm)	Number of pods per plant	Number of seeds per pod	100 seed weight (g)	Seed yield per plant (g)
Replication	2	10.19	46.98 **	11.10	0.18	1.09	0.57	3.75	0.24	5.76	3.28
Treatment	45	50.39**	104.59**	107.10 **	1.72 **	55.16 **	1.78 **	18.88 **	0.87 **	183.93 **	23.31 **
Error	90	3.83	0.84	19.97	0.51	0.53	0.46	1.91	0.10	2.69	1.53

*Significant at 5%, ** Significant at 1%

Genetic divergence analysis

The amount of diversity available in the crop decides the success of any crop improvement programme with manifold objectives. Assemblage and assessment of divergence in the germplasm is essential to know the spectrum of diversity. Different research worker has successfully utilized the procedure of selection of parents based on the extent of genetic diversity in different crop species Moll *et al.* (1962) and Murthy (1966). In this regard the concept of genetic distance has been of vital utility in differentiating well defined population Arunachalam (1967). Improvement in grain yield is normally attained through involvement of the genetically diverse parents in breeding programmes. For identifying such diverse parents for crossing, Mahalanobis D^2 statistics has been used in several crops. It is a powerful tool used to quantify the genetic divergence between the genotypes and to relate clustering pattern with the geographical origin.

Mahalanobis D^2 statistics was used for quantitative assessment of genetic divergence for yield and contributing characters among forty-six fava bean genotypes were presented in Table 2 to 6. The D^2 values were computed in each possible combination of fava bean (*Vicia faba* L.) genotypes. The genotypes were grouped into eight different clusters according to closeness of genotypes in respect to their D^2 values in Table 2. Average intra and inter cluster distance were calculated and presented in Table 4. Intra cluster distance showed divergence among genotypes within a cluster while inter cluster distance expressed relation divergence between the clusters. The detailed description of different cluster are given here as under:

Table 2: Grouping of forty-six genotypes of fava bean into eight clusters on the basis of D^2 analysis.

Cluster	Genotypes	Number of genotypes per clusters
Ⅰ	L2014-096, IC FB235, AREC16071, L2014-107, L2014-106, L2014-108, IC FB246, L2014-105, AREC16072, L2014-099, AREC16079	11
Ⅱ	L2014-010, L2014-094, IC FB239, AREC16065, L2014-104	5
Ⅲ	L2014-130, L2014-017, L2014-095, IC FB232, Vikrant, IC FB242	6
Ⅳ	IC FB238, BASABEER, IC FB241, IC FB237, AREC16076	5
Ⅴ	IC FB245, IC FB243	2
Ⅵ	L2014-129, AREC16066, L2014-005,L2014-101, IC FB233, IC FB240, L2014-121, IC FB236, AREC16068, L2014-120, AREC16078, L2014-119	12
Ⅶ	L2014-131, L2014-100	2
Ⅷ	IC FB234, AREC16077, IC FB247	3

Table 3: Cluster mean among eight clusters for forty six genotypes in fava bean.

Cluster	Days to 50% flowering	Days to maturity	Plant height (cm)	Number of branches per plant	Number of clusters per plant
Ⅰ	75.39	125.18	55.27	4.30	7.36
Ⅱ	72.20	121.27	57.00	4.13	13.67
Ⅲ	75.50	124.83	55.39	4.00	10.94
Ⅳ	74.40	124.00	49.73	5.27	15.53
Ⅴ	73.83	138.17	60.83	4.67	17.17
Ⅵ	72.83	122.50	56.72	4.82	5.86
Ⅶ	65.00	107.83	57.33	4.33	4.93
Ⅷ	65.01	113.00	59.67	4.22	14.22
Cluster	**Pod length (cm)**	**Number of pods per plant**	**Number of seeds per pod**	**100 seed weight (g)**	**Seed yield per plant (g)**
Ⅰ	4.52	24.21	4.00	41.94	39.67
Ⅱ	4.00	24.00	3.60	43.27	41.07
Ⅲ	4.83	23.08	3.19	35.28	37.06
Ⅳ	4.13	21.67	3.53	32.67	37.93
Ⅴ	4.50	24.50	2.67	34.00	37.67
Ⅵ	4.44	23.64	3.61	25.72	34.92
Ⅶ	4.67	23.33	3.50	39.17	39.67
Ⅷ	3.78	23.56	3.89	35.22	37.56

Table 4: Intra and inter cluster distance (D^2) among eight clusters for forty six genotypes in fava bean.

Cluster	Ⅰ	Ⅱ	Ⅲ	Ⅳ	Ⅴ	Ⅵ	Ⅶ	Ⅷ
Ⅰ	74.55	164.95	110.83	243.38	506.31	197.82	521.74	397.67
Ⅱ		43.67	120.71	115.75	482.68	323.97	476.67	168.66
Ⅲ			59.68	114.16	343.97	170.41	587.58	306.06
Ⅳ				58.13	318.72	298.38	721.64	248.67
Ⅴ					78.01	659.83	1610.00	912.51
Ⅵ						98.08	494.14	396.05
Ⅶ							168.28	305.37
Ⅷ								71.31

(a) Group constellation

Grouping of genotypes into different clusters were done by the method suggested by the Tocher and described by the Rao, 1952 [28]. The forty-six genotypes were grouped into eight clusters are presented in Table 2. The maximum number of genotypes were found in the Cluster Ⅵ i.e. twelve followed by Cluster Ⅰ with eleven genotypes; Cluster Ⅲ with six genotypes; Cluster Ⅱ and Cluster Ⅳ with five genotypes; Cluster Ⅷ with three genotypes; Cluster Ⅴ and Cluster Ⅶ with two genotypes respectively.

(b) Intra and inter-cluster distances

Intra and inter relation of clusters were judged based on the average D^2 values. The average D^2 values of intra and inter clusters distances are presented in Table 4. The intra cluster average D^2 values ranged from 43.67 to 168.28. Among the clusters, Cluster Ⅶ had the maximum intra cluster distance (168.28), while the minimum was recorded in Cluster Ⅱ.

The maximum inter cluster D^2 value was recorded between Cluster Ⅴ and Ⅶ (1610.00), while the minimum D^2 value was found between Cluster Ⅰ and Ⅲ (110.83).

(c) Cluster means analysis for different characters

The cluster mean regarding ten characters across the eight clusters are mentioned in Table 3. Considerable differences between clusters were observed for most of the characters studied. Cluster Ⅰ consist eleven genotypes i.e., L2014-096, IC FB235, AREC16071, L2014-107, L2014-106, L2014-108, IC FB246, L2014-105, AREC16072, L2014-099 and AREC16079. It had maximum distance from Cluster Ⅶ (521.74) and minimum distance from Cluster Ⅲ (110.83). The average intra cluster distance was (d^2=74.55).This cluster have maximum mean value for the character number of seeds per pod (4.00) while it does not have minimum value for any character. Cluster Ⅱ consist five genotypes i.e., L2014-010, L2014-094, IC FB239, AREC16065 and L2014-104. It had maximum distance from Cluster Ⅴ (482.68) and minimum distance from Cluster Ⅳ (115.75).The average intra cluster distance was (d^2=43.67).This cluster have maximum mean value for the 100 seed weight (43.27) and seed yield per plant (41.07). Cluster Ⅲ consist of six genotypes i.e., L2014-130, L2014-017, L2014-095, IC FB232, Vikrant and IC FB242. It had maximum distance from Cluster Ⅶ (587.58) and minimum distance from Cluster Ⅳ (114.16).The average intra cluster distance was (d^2=59.68).This cluster had maximum mean value for days to 50 per cent flowering (75.50) and pod length (4.83) while minimum mean value for the number of branches per plant (4.00). Cluster Ⅳ consist five genotypes i.e., IC FB238, BASABEER, IC FB241, IC FB237and AREC16076. It had maximum distance from Cluster Ⅶ (721.64) and minimum distance from Cluster Ⅷ (248.67). The average intra cluster distance was (d^2=58.13). This cluster had the maximum mean value for number of branches per plant (5.27) and minimum mean value for the characters plant height (49.73) and number of pods per plant (21.67). Cluster Ⅴ consist of two genotypes namely IC FB245and IC FB243. It had maximum distance from the Cluster Ⅶ (1610.00) and minimum distance from the Cluster Ⅵ (659.83). The average intra cluster distance was (d^2=78.01).This cluster had the maximum mean value for the days to maturity (138.17), plant height (60.83), number of clusters per plant (17.17) and number of pods per plant (24.50) and minimum mean value for the number of seeds per pod (2.67). Cluster Ⅵ had maximum twelve genotypes i.e., L2014-129, AREC16066, L2014-005, L2014-101, IC FB233, IC FB240, L2014-121, IC FB236, AREC16068, L2014-120, AREC16078 and L2014-119.It had maximum distance from the Cluster Ⅶ (494.14) and minimum distance from the Cluster Ⅷ (396.05). The average intra cluster distance (d^2=98.08). This cluster had minimum mean value for the 100 seed weight (25.72) and seed yield per plant (34.92). Cluster Ⅶ consist two genotypes i.e., L2014-131and L2014-100.It had maximum distance from Cluster Ⅴ (1610.00) and minimum distance from the Cluster Ⅷ (305.37). The average intra cluster distance was

(d^2=168.28). This cluster had minimum mean value for the days to 50 per cent flowering (65.00), days to maturity (107.83) and number of clusters per plant (4.93). It does not have maximum mean value for any character. Cluster Ⅷ consist three genotypes namely IC FB234, AREC16077, and IC FB247. It had maximum distance from the Cluster Ⅴ (912.51) and minimum distance from the Cluster Ⅱ (168.66). The average intra cluster distance was (d^2=71.31).This cluster had minimum mean value for the character pod length (3.78).

Contribution of character towards genetic divergence

Contribution of each character towards genetic divergence has been estimated from the number of times that each character appeared in the first rank presented in Table 6. The percent contribution of seed yield and its attributing characters in genetic divergence were reported maximum for number of clusters per plant (36.33%) followed by days to maturity (28.41%), 100 seed weight (25.4%), seed yield per plant (3.38%), number of pods per plant (2.71%) and number of seeds per pod (1.74%). The rest of the characters had minute per cent contribution in genetic divergence i.e., plant height (0.97%), pod length (0.6%), days to 50 percent flowering (0.19%) and number of branches per plant (0.19%). The above results imply that, in order to select genetically diverse genotypes for hybridization, the material should be screened for the important traits like, number of clusters per plant, days to maturity, 100 seed weight, seed yield per plant, number of pods per plant.

Table 5: Range of cluster mean value for ten characters in fava bean.

S. No.	Characters	Range			
		Minimum		Maximum	
		Value	Cluster	Value	Cluster
1.	Days to 50% flowering	65.00	Ⅶ	75.5	Ⅲ
2.	Days to maturity	107.83	Ⅶ	138.16	Ⅴ
3.	Plant height (cm)	49.73	Ⅳ	60.83	Ⅴ
4.	Number of branches per plant	4.00	Ⅲ	5.26	Ⅳ
5.	Number of clusters per plant	4.93	Ⅶ	17.16	Ⅴ
6.	Pod Length (cm)	3.77	Ⅷ	4.83	Ⅲ
7.	Number of pods per plant	21.66	Ⅳ	24.21	Ⅰ
8.	Number of seeds per pod	2.66	Ⅴ	4.00	Ⅰ
9.	100 seed weight (g)	25.72	Ⅵ	43.26	Ⅱ
10.	Seed yield per plant (g)	34.91	Ⅵ	41.06	Ⅱ

Table 6: Contribution of each character to genetic divergence in fava bean.

S. No.	Characters	Contribution % towards divergence
1.	Days to 50% flowering	0.19
2.	Days to maturity	28.41
3.	Plant height (cm)	0.97
4.	Number of branches per plant	0.19
5.	Number of clusters per plant	36.33
6.	Pod Length (cm)	0.68
7.	Number of pods per plant	2.71
8.	Number of seeds per pod	1.74
9.	100 seed weight (g)	25.4
10.	Seed yield per plant (g)	3.38

Conclusion

On the basis of maximum cluster mean value the ten characters were falling in distinguish clusters. Genotypes with maximum number of days to 50 per cent flowering and pod length were observed in Cluster III, while, the genotypes with maximum number of days to maturity, plant height and number of clusters per plant were observed in Cluster V. Genotypes with a smaller number of days to 50 per cent flowering and days to maturity, were observed in Cluster Ⅶ Cluster Ⅱ have more 100 seed weight and seed yield per plant. Cluster Ⅰ having a greater number of pods per plant and number of seeds per pod. Cluster Ⅳ having the greater number of branches per plant and dwarf plant height. Hence, it is worthy to note that in calculating cluster means, the superiority of particular genotypes in respect to a given character get diluted by other genotype that are related and grouped in the same cluster but which are inferior or intermediary for that character in question. Hence, apart from selecting genotypes from the clusters which have high inter-cluster distance for hybridization, one can also think of selecting genotypes based on extent of genetic divergence in respect to a particular character of interest. This is to mean that, if breeder's intention is to improve seed yield, he can select genotypes which are highly divergent with respect to these characters.

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