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Genetic divergence of pea genotypes (*Pisum sativum* L.) based on multivariate analysis

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Abstract
The present study was conducted to identify the nature and magnitude of genetic divergence among fifty two genotypes based on phenotypical traits using the multivariate analysis. Based on cluster analysis, the genotypes were best fitted into three clusters. The maximum and minimum genotypes grouped in cluster I (50) and minimum were in Cluster II (1) and cluster III (1) respectively. The maximum intra-cluster distance was shown by cluster I (D^2=5.07) indicating maximum difference among the genotypes within and the minimum value was shown by cluster II and cluster III. Maximum value of inter-cluster distance (D^2=16.76) was recorded between cluster I and cluster III revealing that the genotypes of these clusters were highly diverse from others and can be used as divergent parents for hybridization and selection. Thus, for getting high heterosis for recovering transgressive segregants, genotypes from cluster I and III can be used as distant parents in any breeding programme for successful pea improvement. The results of present study could be exploited in the future genetic improvement programme of pea genotypes in Madhya Pradesh region.

Keywords: Genetic diversity, environment, hierarchical clustering, pea

Introduction
Pea (*Pisum sativum* L.) 2n=2x=14 is one of the world's oldest crop which belongs to the family fabaceae (earlier leguminaceae subfamily papilionaceae) grown in all the temperate countries and in the most tropical highlands. It has been grown for several thousand years in India and is a crop native to Syria, Turkey, Israel and Ethiopia, for its versatile uses as pulses and livestock feed (Choudhury *et al*., 2007) [1]. It is grown for its green tender pods, dried seeds, canned, frozen or dehydrated form (Santalla *et al*., 2001) [17]. In India it is cultivated in an area of 311.87 (000, in ha.) which production of 321.87 (000, in MT.) and productivity of 1032 Kg/ha. (Source: www.eands.dacnet.nic.in). Most of the production is produced in Uttar Pradesh, Madhya Pradesh, Bihar, Assam and Orissa, and the total area is about 95 per cent. In Madhya Pradesh the area (000, in ha.), production (000, in MT) and productivity (Kg/ha) are 54.0, 542.2 and 1004, respectively. (Sources: Department of Agriculture, Cooperation and Farmer Welfare). They are rich source of phytonutrients, minerals, vitamins and antioxidants and is known for its superior quality protein like high levels of lysine making it an appropriate dietary complement to the cereals (Dhama *et al*., 2010) [3]. Being a short duration crop it is highly utilized for crop rotation and also have an important role in the modern agricultural systems as it is capable to enhance the soil structure and provides breaks for the disease control (Martin *et al*., 2008) [15]. Substitution of landraces and traditional pea accessions by the modern cultivars is widespread and consequently the genetic variability loss is reduced, in particular replacement with cultivars characterized by superior tolerance for biotic and abiotic stress (Handerson *et al*., 2014) [7]. In the process of genetic improvement of any crop, genetic diversity among germplasm plays a major role, since it opens the way to determine the most divergent parents based on the contribution of different qualitative and quantitative traits, for further utilization in any hybridization programme. Therefore, the exploration of genetic diversity in the available germplasm is a pre-requisite in a breeding programme for effective selection of the superior genotypes (Goyal and Bisen, 2017) [6]. A plant breeder has to identify the source of favorable genes to incorporate them into the breeding populations and select for a combination of desirable traits that might result in the isolation of productive genotypes and cultivars. Thus, present study is undertaken to understand the magnitude of genetic divergence for identifying more diverse parents for pea genetic improvement.

Methods and Materials

Experimental material consists of fifty two genotypes of field pea included two check variety KN-5 and JP-885 were estimated by taking the pooled data of 52 pea genotypes grown in a Randomized Complete Block Design (RCBD) with two replications at three different dates of sowing at an interval of 21 days during both *Rabi* Season i.e. 2018-19 and 2019-20 respectively at Seed Production Farm of BSP (Vegetable) JNKVV, Jabalpur (M.P.). All the agronomic practices were made to raise the healthy crop and development. Each plot consisted of two rows of 3.0 meter length. Observation were recorded on five competitive plants situated under the same field condition for fifteen morphological quantitative traits *viz*., days to flower initiation, first flowering node, day to maturity, number of primary branches/plant, number of secondary branches/plant, number of nodes/plant, pod bearing length (cm), number of pods/plants, pod length (cm), number of seed/pods, number of seeds/plant, hundred seed weight (gm), biological yield/plant (gm), harvest index (%) and seed yield/plant (gm). Averages of the data from the sampled plants in respect of different quantitative characters were used for various statistical analyses. The magnitude of genetic diversity among fifty five pea genotypes was determined by using D^2 Mahalanobis genetic distance statistics (Mahalanobis, 1936)[14]. Hierarchical clustering using Tocher's method, as described by Rao (1952)[16] was followed for the grouping of genotypes into distinct clusters.

Results and Discussion

The data obtained from the observations recorded on fifteen morphological quantitative traits were subjected to the statistical scrutiny. It was evident from the analysis of variance that mean sum of squares due to 52 genotypes were highly significant for all the traits (Table 1), giving the clear picture of presence of wide spectrum of variability among the genotypes. These results were in agreement with the findings of Lal *et al*. (2011) [13]; Supe *et al*. (2013) [22]; Georgieva *et al*. (2016) [5] and Kumar and Bisen (2016) [9]. Although the analysis of variance revealed sufficient variability among the genotypes, but the extent of genetic diversity present among the genotypes could not be explained, therefore, cluster analysis was performed to quantify the genetic divergence between any two genotypes or group of genotypes. Based on the relative magnitude of their Mahalanobis D^2 values using Torcher's method, all the 52 genotypes of pea under study were grouped into three clusters. The clustering patterns of pea genotypes into three clusters are presented in Table 2. Maximum number of genotypes (50) was grouped in cluster I namely: Rachna, FP-9-539, RP-3, FP-7-562, FP-7-596, DDR-55, DDR-52, P-3, VL-3, ARKA SAMPURNA, DDR-27, ARKEL, VRP-5, PUSA PRAGATI, PSM-3, DDR-54, JAYANTI, B-22, JM-6, GS-10, HFP-94-13, NDVP-4, PP-155, FP-14-56, IFP-99-25, AMAN, HVP-2, FP-16-86, FP-14-46, FP-13-30, FP-94-12, PP-14-17, PP-14-82, FP-14-13, FP-14-8, FP-14-21, FP-14-56, FP-14-27, FP-14-33, FP-18-30, JP-180, KMPR-30, KPMR-302, KPMR-402, KPMR-502, KPMR-585, MATAR RANGPUR, GOL BATRA TEDUHA, SAFEED BATRA GUDDA, JP-885 (Check 1). Whereas, cluster II and cluster III both contained one genotypes each where, cluster I comprises of genotypes namely: KN-5 (Check 2); and cluster V consisted of genotypes namely: KPMR-327. No parallelism was shown by the grouping pattern of the genotypes between the genetic diversity and geographical origin of genotypes. Similar confirmations were also reported by the findings of Singh *et al*. (2007) [21]; Dhama *et al*. (2009) [4]; Katiyar and Dixit (2009) [8]; Yadav *et al*. (2009) [23]; Devi *et al*. (2010) [2]; Shrivastava *et al*. (2012) [19]; Supe *et al*. (2013) [22] and Kumar and Kumar (2016) [10]. The average intra and inter-cluster D^2 values with their corresponding intra and inter-cluster distance are presented in Table 3. The inter-cluster distances were greater than intra-cluster distances, which indicated the presence of considerable amount of genetic diversity among the genotypes studied. The greater the magnitude of intra and inter cluster distance the higher the variability among the cluster and within the cluster and *vice versa*. The results are in concurrence with the findings of Kumar *et al*. (2006) [11]; Singh *et al*. (2007) [21]; Singh and Mishra (2008) [20]; Katiyar and Dixit (2009) [8]; Sen and De (2017) [18]. The least value of intra cluster distance was found in cluster II and cluster III (D^2=0.0) indicating the presence of less heterogeneous genotypes grouped in this cluster. Whereas, maximum value of intra-cluster distance was observed in cluster I (D^2= 5.07) revealing the existence of maximum differences among the genotypes falling in this cluster. Hence, selection within these clusters may be exercised based on the highest area of desirable traits. In any breeding programme where the nature of crosses is to be evaluated, choice of diverse parents is of paramount importance as they produce superior off-springs in the segregating generation than the closely related ones. The inter-cluster distance (D^2) being the main criterion for selection of genotypes was also worked-out as crossing of genotypes within the same cluster would not produce superior off-springs. A range of 11.68 to 24.63 was observed when inter-cluster D^2 values were used to study the diversity among the clusters. The minimum value of inter-cluster distance (D^2 =11.68) was found between cluster I and II indicating close relationship and similarity for most traits among the genotypes included in these clusters. Whereas, cluster II and III showed maximum value of inter-cluster distance (D^2 =24.63), followed by cluster I and III (D^2 =16.76) indicating that the genotypes included in these clusters are not so closely related showing good amount of diversity. Hence, these genetically diverse genotypes can be used as promising parents for hybridization.

These results are corroborated with the findings of Kumar *et al*. (2007) [12]; Singh *et al*. (2007) [21]; Devi *et al*. (2010) [2] and Shrivastava *et al*. (2012) [19] as they also gave similar conclusion. Diversity among the genotypes was also estimated based on the considerable amount of variation in cluster means for different character. Different clusters exhibited distinct mean values for almost all the fifteen characters which reflect the genetic differences between the clusters (Table 4). It is evident from the cluster mean table that the genotypes in cluster I had highest mean values for days to maturity, number of nodes/plants and harvest index (%). Whereas, the genotypes of cluster II showed the maximum mean for pod length (cm), number of seeds/pod and seed/yield/plant (gm). The genotypes of cluster III showed the maximum mean for days to first flowering, first flowering node, and number of primary branches/plant, number to secondary branches/plant, pod bearing length, number of pods/plants, number of seeds/plants, hundred seed weight (gm) and biological yield/plant (gm). Comparative assessment of cluster means showed that for improving specific characters, the genotypes should be selected from the cluster having high mean value for that particular character.

This comparison indicates that clusters I and III had better cluster means for most of the characters, therefore, these clusters might be considered better for selecting genotypes as divergent parents. The similar results are exhibited with the findings of Kumar *et al*. (2006) [11]; Devi *et al*. (2010) [2] and Shrivastava *et al.* (2012) [19].

Table 1: Analysis of variances for yield and yield attributing traits of pea genotypes over environments

Env.	Source of Variation	DF	Mean sums of square														
			DFI	FFN	DM	NPB	NSB	NNP	PBL	NPP	PL	NSPI	NSPII	HSW	BYP	HI	SYP
POE	Replication	1	6.98	0.26	0.00	1.87	0.11	12.69	223.31	10.70	0.06	0.14	12.21	0.26	13.11	266.45	0.01
	Genotypes	51	358.83**	9.63**	514.55**	11.30**	3.06**	72.97**	5425.53**	194.58**	5.65**	5.53**	3026.36**	44.33**	116.62**	1041.86**	31.18**
	Error	51	6.83	0.33	10.18	0.26	0.18	5.60	76.15	6.36	0.27	0.23	123.52	1.46	13.51	136.91	3.52

DFI=Days to first flowering, FFM=First flowering node, DM=Days to maturity, NPB=Number of primary branches/plant, NSB=Number to secondary branches/ plant, NNP=Number of nodes/plants (main branch) PBL=Pod bearing length, NPP = Number of pods/plants, PL= Pod length cm, NSPI= Number of seeds/pod NSPII =Number of seeds/plants, HSW=Hundred seed weight (gm), BYP =Biological yield/plant (gm), HI= Harvest index (%), SYP= Seed/yield/plant (gm), POE= Pooled over environments

Table 2: Clustering pattern of 52 pea genotypes on the basis of their mahalanobis genetic divergence using tocher's methods

Clusters	Number of Genotypes	Name of genotypes included
I	50	Rachna, FP-9-539, RP-3, FP-7-562, FP-7-596, DDR-55, DDR-52, P-3, VL-3, ARKASAMPURNA, DDR-27, ARKEL, VRP-5, PUSA PRAGATI, PSM-3, DDR-54, JAYANTI, B-22, JM-6, GS-10, HFP-94-13, NDVP-4, PP-155, FP-14-56, IFP-99-25, AMAN, HVP-2, FP-16-86, FP-14-46, FP-13-30, FP-94-12, PP-14-17, PP-14-82, FP-14-13, FP-14-8, FP-14-21, FP-14-56, FP-14-27, FP-14-33, FP-18-30, JP-180, KMPR-30, KPMR-302, KPMR-402, KPMR-502, KPMR-585, MATAR RANGPUR, GOL BATRA TEDUHA, SAFEED BATRA GUDDA, JP-885 (Check 1)
II	1	KN-5 (Check 2)
III	1	KPMR-327

Table 3: Average of intra and inter cluster genetic distance

Clusters	I	II	III
I	5.07	11.68	16.76
II	11.68	0.00	24.63
III	16.76	24.63	0.00

Table 4: Cluster wise mean values of 11 morphological traits in pea (*Pisum sativum* L.)

Cluster	Characters														
	DFI	FFN	DM	NPB	NSB	NNPP	PBL	NPPP	PL	NSPP(I)	NSPP(II)	HSW	BYP	HI	SYP
I	44.37	3.67	83.5	2.17	1.24	13.92	47.38	13.15	4.78	4.05	48.76	13.93	14.06	56.15	7.44
II	30.83	2.53	62.62	1.97	1.28	9.43	9.49	10.26	5.52	4.65	46.72	16.08	15.87	45.2	7.56
III	50.77	4.11	77.15	8.15	1.75	13.9	75.26	22.28	4.07	2.98	65.95	16.24	16.26	46.91	7.39

DFI=Days to first flowering, FFM=First flowering node, DM=Days to maturity, NPB=Number of primary branches/plant, NSB=Number to secondary branches/ plant, NNP=Number of nodes/plants (main branch) PBL=Pod bearing length, NPP = Number of pods/plants, PL= Pod length cm, NSPI= Number of seeds/pod NSPII =Number of seeds/plants, HSW=Hundred seed weight (gm), BYP =Biological yield/plant (gm), HI= Harvest index (%), SYP= Seed/yield/plant (gm)

Conclusion

Based on Mahalanobis D^2 analysis it can be concluded that the pea germplasm in the present study can be successfully used for planning future breeding programmes. The inter-crossing of genotypes showing the greater genetic divergence for most of the characters studied should result in superior heterotic crosses and also, generate valuable segregants in the later generations. It is expected that better performing varieties could be generated to increase productivity in field pea. Therefore, from the present study genotypes of cluster I and cluster III based on their high values for inter cluster distance and cluster means can be hybridized as the potential parents to produce superior off-springs in the segregating generations and to improve pea productivity.

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