Assessment of genetic diversity in garden pea (*Pisum sativum* L.) and identification of promising lines for hybridization

Abstract

The current study is being conducted to understand the degree of genetic divergence in order to discover more diversified parents for pea genetic advancement. The most important tasks for a pea breeding are development of high yielding varieties with stable productivity, resistance to diseases and unfavorable environmental conditions, different maturing types, high rate of organic matter accumulation during the initial phases of growth, sufficiently high intensity of photosynthesis, increases in protein content, essential amino acids and favorable rations among them. The present investigation was carried out at vegetable research farm and laboratory works in the department of Horticulture (Vegetable and Floriculture), BAU, Sabour, Bhagalpur, Bihar during the Rabi season 2020-21. The genotype Kashi Mukti, Kashi Uday, Kashi Nandini and VM-11 of cluster II exhibited superiority for earliness in first flower, days to 50% flowering and days to first picking and significantly better performance as measured by the lowest cluster mean and a substantial improvement in performance while the genotype Haze-02 of cluster IV exhibited superiority for inter-nodal length , plant height , seeds per pod and total sugars (%) based on maximum cluster mean with significantly better performance. progenies derived from such diverse crosses are expected to show a wide spectrum of genetic variability and a greater scope for isolating transgressive segregants in the advanced generations. Hence these genotypes may be uased in a multiple crossing programme to recovertransgressivesegregants.

Keywords: organic matter, transgressivesegregants, genetic advancement, *Pisum sativum*

Introduction

Garden pea (*Pisum sativum* L sub sp. hortense Asch. and Graebn, 2n=2x=14) is one of the important leguminous vegetable crops around the world, which is grown in India during the *rabi* or cool season. Garden pea is a member of the family *Fabaceae* and originated in Europe and West Asia, while its wild type came from Ethiopia (De Candolle, 1886). It is an autogamous crop with cleistogamous flowers and generally less than 1% naturally out-crossing. Garden pea commonly known as vegetable pea is a highly nutritive vegetable containing high percentage of digestible protein (amino acid, methionine and cystene), carbohydrate, vitamin A, Vitamin C and minerals like calcium, potassium, phosphorus, iron, fiber and low in fat (National Food Administration, 2002). It is crucial for India, where the majority of the population is vegetarian, since it provides vital and affordable sources of protein as a supplement to cereals for the poor who cannot afford to consume proteins from an animal source. Due to its ability to fix nitrogen from the environment with the aid of Rhizobium bacteria that be present in its nodule roots, the pea is included in crop rotation to supply nitrogen to other plants (Rudnicki and Wenda, 2002).

The most important tasks for a pea breeding are development of high yielding varieties with stable productivity, resistance to diseases and unfavorable environmental conditions, different maturing types, high rate of organic matter accumulation during the initial phases of growth, sufficiently high intensity of photosynthesis, increases in protein content, essential amino acids and favorable rations among them (Siddika *et al.*, 2014). The extensive replacement of landraces and conventional pea accessions by present-day cultivars, which is characterised by cultivars with higher tolerance for biotic and abiotic stress, has the effect of reducing genetic variability loss (Handerson *et al.*, 2014). Genetic diversity among germplasm plays a significant role in the genetic development of any crop since it makes it possible to identify the most divergent parents based on the contribution of various quantitative and qualities traits, which can then be used in any hybridization programme. As a result, a breeding program's study of the genetic variety in the available germplasm is a requirement for the efficient selection of the best genotypes. A plant breeder must locate the source of beneficial genes in order to incorporate them into breeding populations and select for a combination of desirable features that might lead to the isolation of productive genotypes and cultivars. As a result, the current study is being conducted to understand the degree of genetic divergence in order to discover more diversified parents for pea genetic advancement.

MATERIALS AND METHODS

The present investigation was carried out at vegetable research farm and laboratory works in the department of Horticulture (Vegetable and Floriculture), BAU, Sabour, Bhagalpur, Bihar (87°2’ 42” East and 25° 15’ 40” North; 45.57 m above mean sea level) during the Rabi season 2020-21. The experimental material comprised of 26 genotypes of pea (Pea Vasundhra, Nikhar-10, Punjab- 89, P-10, Goldie, NS-1100, Azad Pea-3, UBL-10, Kashi Shakti, Kashi Ageti, EC-507771, IC-109696, Nirali, Kashi Samridhi, Kashi Mukti, Kashi Uday, Kashi Nandini, P-3771, VM-12, P-3824, VM-10, EC-269571, Muze-01, VM-11, EC-598572, Haze-02)collected from Department of Vegetable Science & Floriculture, BAU, Sabour were grown in a Randomized Block Design (RBD) with three replications. In each replication the seeds were sown in a plot of 2 m × 1.5 m in which row to row and plant to plant spacing was 30 cm and 10 cm respectively. All the recommended agro-practices were followed to ensure a healthy crop growth and development. Observation were recorded on five competitive plants situated under the same field condition for seventeen morphological quantitative qualitative traits *viz*., days to first flower, days to 50% flowering, days to first picking, plant height (cm), number of primary branches per plant, nodes per plant, inter-nodal length, pod length (mm), pod diameter (mm), seeds per pod, shelling (%), number of pods per plant, pod yield per plant (g), number of pickings, total soluble solids (˚brix), ascorbic acid (mg/100g), total sugar (%), protein (%). The data collected were subjected to multivariate analysis utilizing Mahalanobis D^2 statistics as suggested by Mahalanobis (1936) and Rao (1952). Genotypes were grouped into various clusters following Tocher’s method as suggested by Rao (1952).

Result and Discussion

The analysis of variance revealed highly significant among the genotypes foryield its components and quality traits (Table-1), which indicated that considerable amount of genetic variability present in the genotypes. Hence, there is ample scope for inclusion of promising genotypes in breeding program for yield and its component characters.The degree of genetic diversity plays an essential role in a crop's varietal development programme. D^2 statistics is a useful technique for assessing genetic diversity among various genotypes and identifying parents for hybridization in order to generate suitable recombinants. The computation from co-variance matrix gave non-hierarchical clustering based on Mahalanobis D^2 values among 26 genotypes and grouped them into five clusters (Table-2 &Figure-1).

Amongst different five clusters, cluster I was found to be the largest one. Out of the 5 clusters of 26 genotypes, cluster I comprised of maximum 12 genotypes (Goldie, P-3824, EC-269571, P-10, NS-1100, Kashi Shakti, Nikhar-10, Muze-01,Kashi Ageti, EC-598572, UBL-10, P-3771) followed by cluster III with 6 genotypes (VM-12, VM-10, Punjab-89, IC-109696, Pea Vasundhra, NS-1100) and cluster II with 4 genotypes (Kashi Mukti, Kashi Uday, Kashi Nandini, VM-11) and cluster V with 3 genotypes (EC-507771, Nirali, Kashi Samridhi) and only one cluster IV exhibited monogenotypic i.e., containing one genotype which is showed in Table 2. Saddik *et al.* (2014) also reported 5 clustering in field pea. Kumar and Kumar (2016) reported 4 cluster and studied on 54 different genotypes of garden pea, Singh and Singh (2003) studied genetic divergence for 10 traits and had 11 clusters and Vikas and Singh (1999) had 9 clusters for 45 pea genotypes. The highest intra-cluster distance was exhibited by V (125.55) followed by cluster III (119.96), cluster II (91.3) and cluster I (81.69) which indicated that hybridization involving genotypes within the same clusters may result in cross combination (Table-3). The highest inter-cluster distance was observed between cluster II and V (588.09) subsequently cluster II and clusters III (342.33), cluster II and IV (283.84), clusters II and I (235.59), clusters I and V (233.67).A wide range of inter-cluster genetic distance among the different clusters of pea genotypes have also been reported by Tiwari *et al.* (2004), Kumar *et al.* (2007), Singh *et al.* (2007), Sharma *et al.* (2013), Georgieva *et al.* (2016) and Khan *et al.* (2017). These lines may be utilized in further breeding programme for the exploitation of hybrid vigour and suggesting wide diversity between them and genotypes in these clusters could be used as parents in hybridization programme to develop desirable type because crosses between genetically divergent lines will generate heterotic segregants. Therefore, crosses between the genotypes of clusters separated by inter-cluster distances are likely seemed to be beneficial for further improvement.

The cluster V exhibited highest mean values for yield and yield contributing characters (Table- 5) namely pod diameter, number of pods per plant, number of primary branches per plant, total soluble solid, ascorbic acid, protein percentage (%), shelling (%) and pod yield per plant. The genotypes of cluster II revealed lowest mean values for days to first flower along with days to 50% flowering, number of nodes per plant and days to first picking. To develop high yielding varieties along with early maturing type these groups can be used in hybridization programme. Cluster I exhibited highest mean values for pod length and plant height while cluster IV exhibited highest mean for number of seed per pod and total sugar and desirable minimum inter-nodal length and on the basis of mean values which could be utilized for hybridization programme for the development of high yielding pea genotypes. Variable cluster means for different plant growth and fruit yield characters have also been reported by Surejaand Sharma (2001), Tiwari *et al.* (2004), Kumar *et al.* (2007), Kumari *et al.* (2013), Georgieva *et al.* (2016), Gupta *et al.* (2017) and Bijalwan*et al.* (2018) in garden pea.

The per cent contribution of seventeen traits towards total genetic divergence is listed in (Table-5). The selection and choice of parents mainly depends upon contribution of characters towards divergence. In the present investigation the highest contribution in manifestation of genetic divergence was exhibited by total sugar (28%) followed by days to 50% flowering (24.92%), days to first picking (12%), pod diameter (8.62%), ascorbic acid (6.77%), plant height (5.54%), days to first flowering (4.31%), inter-nodal length (4%), number of nodes per plant (2.15%), number of seed per pod (1.54%), pod yield per plant (0.92%), total soluble solid (0.62%), protein percentage, shelling percentage contributed (0.31%).Therefore, these characters may be given importance during hybridization programme.

Selection of genotypes for future hybridization program

The selection of superior genotypes and specific characters for breeding improvement programme presented in Table 6. Genotypically distant parents usually able to produce higher heterosis (Falconer 1960; Moll *et al.* 1962; Ramanujam *et al.*, 1974; Ghaderi *et al.*, 1984; Siddika *et al.* 2014). The genotypes belonging to the distance clusters could be used in hybridization programme for obtaining a wide spectrum of variation among segregates. The genotypes EC-507771, Nirali and Kashi Samridhi group in cluster V showed superiority for pod yield per plant, primary branches per plant, pod diameter, shelling (%), pods per plant, T.S.S., ascorbic acid and protein (%) and take maximum duration for first picking, based on their D^2 value of cluster mean superiority and *per se* performance. The genotype Kashi Mukti, Kashi Uday, Kashi Nandini and VM-11 of cluster II exhibited superiority for earliness in first flower, days to 50% flowering and days to first picking and significantly better performance as measured by the lowest cluster mean and a substantial improvement in performance while the genotype Haze-02 of cluster IV exhibited superiority for inter-nodal length , plant height , seeds per pod and total sugars (%) based on maximum cluster mean with significantly better performance.

The hybridization programme involving genotypes EC-507771, Nirali and Kashi Samridhi into Kashi Mukti, Kashi Uday, Kashi Nandini and VM-11 could be undertaken to isolate high yielding sergeants with earliness, since these genotypes have pod yield per plant, primary branches per plant, pod diameter, shelling (%), pods per plant, T.S.S. (˚Brix), ascorbic acid and protein (%). These parents could be selected for hybridization on the basis of their large inter-cluster distance for isolating useful recombinants in the segregating generations. Siddika *et al.* 2014 reported that selection of parents from distantly placed clusters exhibited significant high heterotic segregants. Therefore, progenies derived from such diverse crosses are expected to show a wide spectrum of genetic variability and a greater scope for isolating transgressive segregants in the advanced generations. Hence these genotypes may be uased in a multiple crossing programmeto recovertransgressivesegregants.

Ethics approval - Not Applicable

Availability of data and material - Not Applicable

Code availability - Not Applicable

Ethics approval - Not Applicable

Consent to participate - Not Applicable

Consent for publication - Not Applicable

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Fig: 1: Dendrogram showing grouping of 26 pea genotypes generated using D^2 cluster analysis (Tocher's method)

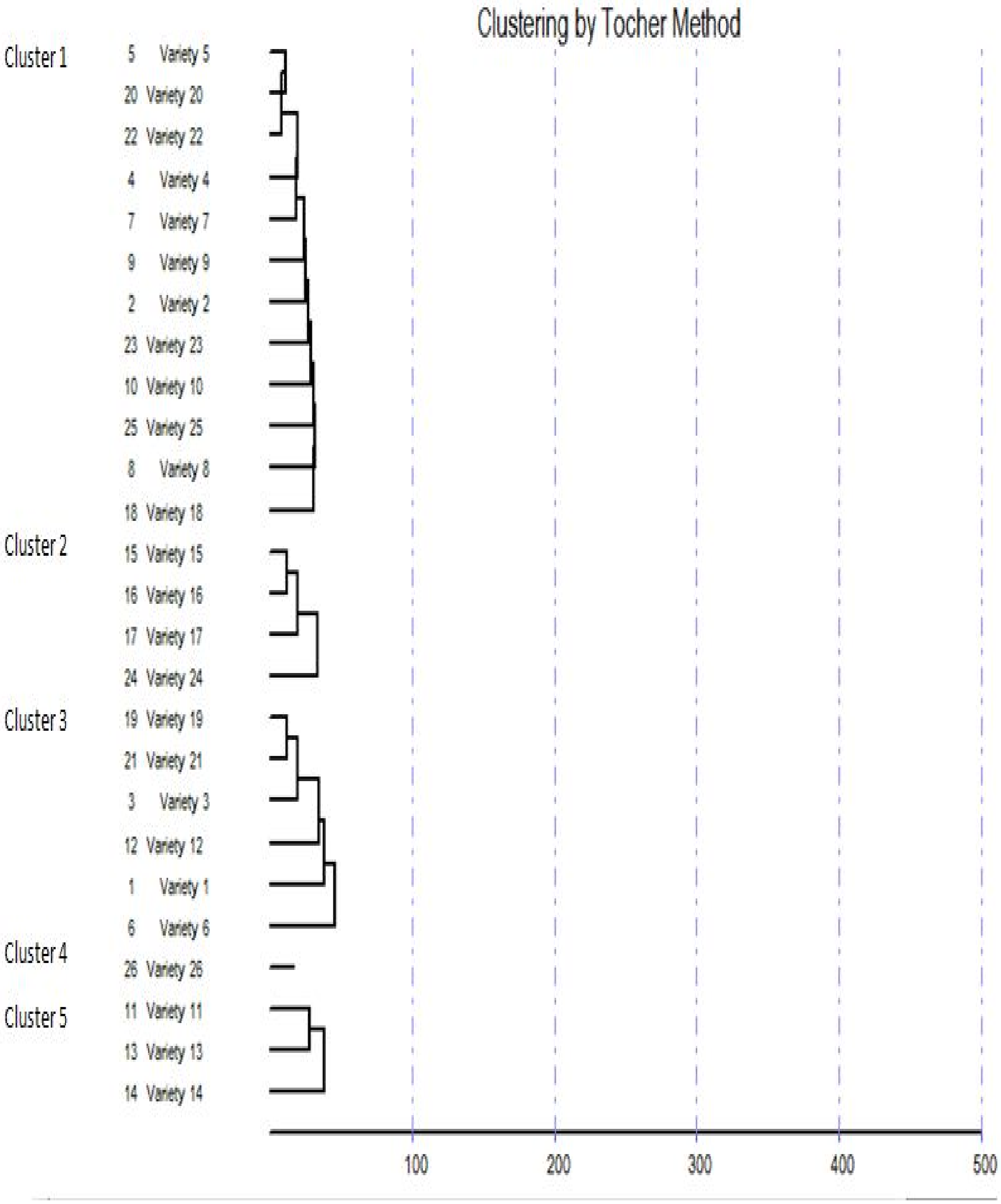

Fig: 2: Cluster distances showing grouping of 26 pea genotypes generated using D^2 cluster analysis (Tocher's method)

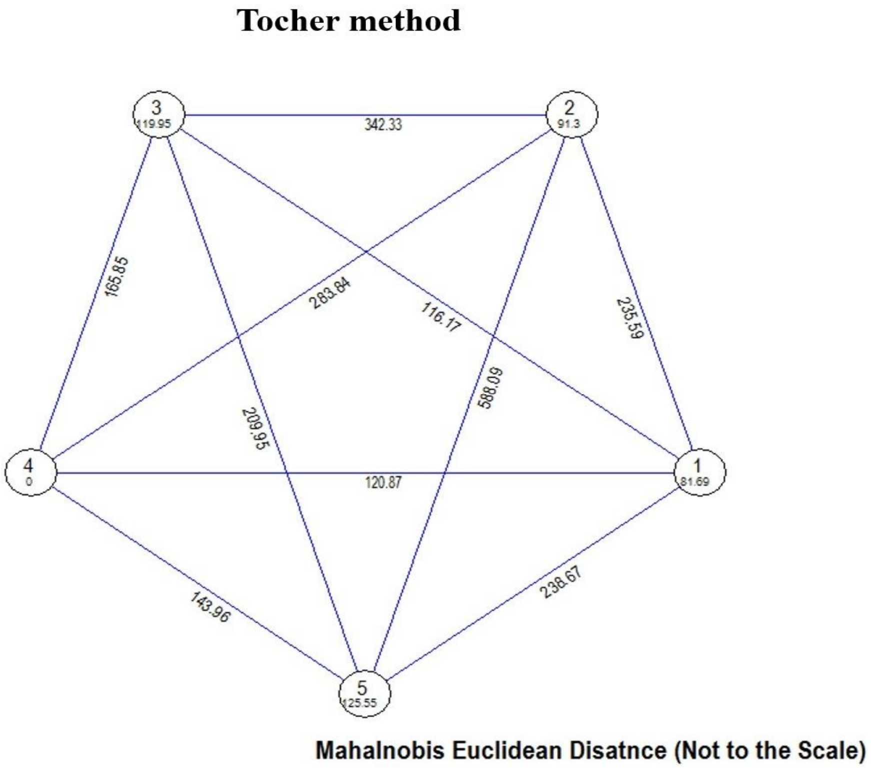

Table-1: Analysis of variance for seventeen characters in garden pea

Characters	Source d.f.	Mean sum of Square		
		Replications (2)	**Treatments (25)**	**Error (50)**
Growth parameters				
Days to first flower		3.50	226.19**	2.35
Days to 50%flowering		4.50	229.97**	1.83
Days to first picking		6.78	246.39**	4.59
Number of primary branches		0.08	0.77**	0.09
Inter-nodal length(cm)		0.16	1.79**	0.05
Nodes per plant		0.56	19.07**	0.66
Plant height (cm)		15.63	347.97**	9.45

Yield contributing traits				
Pod length (mm)		0.01	6.14**	0.17
Pod diameter (mm)		0.01	6.14**	0.17
Seeds per pod		0.08	1.87**	0.15
Shelling (%)		3.13	12.55**	6.34
Pods per plant		0.02	17.98**	1.49
Yield per plant (g)		42.29	903.01**	56.37
Quality characters				
T.S.S. (˚Brix)		0.52	3.57**	0.27
Total sugar (%)		0.01	3.57**	0.04
Ascorbic acid (mg/100g)		0.69	20.01**	0.64
Protein (%)		0.18	5.58**	0.66

** Significant at 1% probability level.

Table: 2: Distribution of twenty-six pea genotypes in various clusters

Cluster	**No. of genotypes**	**Name of genotypes**
I	12	Goldie, P-3824, EC-269571, P-10, NS-1100, Kashi Shakti, Nikhar-10, Muze-01, Kashi Ageti, EC-598572, UBL-10, P-3771
II	4	Kashi Mukti, Kashi Uday, Kashi Nandini, VM-11
III	6	VM-12, VM-10, Punjab-89, IC-109696, Pea Vasundhra, NS-1100
IV	1	Haze-02
V	3	EC-507771, Nirali, Kashi Samridhi

Table 3: Average intra and inter-cluster distance values among five clusters for twenty-six genotypes of pea

Cluster	**I**	**II**	**III**	**IV**	**V**
I	81.69	235.59	116.17	120.87	233.67
II		91.3	342.33	283.84	588.09
III			119.96	165.85	209.96

IV				0	143.96
V					125.55

Table-4: Contribution of various characters towards genetic divergence in pea

Sl. No.	**Character**	**Times rank first**	**Contribution %**
1	Days to first flower	14	4.31%
2	Days to 50% flowering	81	24.92%
3	Days to first picking	39	12%
4	No. of primary branches	0	0%
5	Inter-nodal length (cm)	13	4%
6	Nodes per plant	7	2.15%
7	Plant height (cm)	18	5.54%
8	Pod length (mm)	0	0%
9	Pod diameter (mm)	28	8.62%
10	Seeds per pod	5	1.54%
11	Shelling (%)	1	0.31%
12	Pods per plant	0	0%
13	Pod yield per plant (g)	3	0.92%
14	T.S.S. (˚Brix)	2	0.62%
15	Ascorbic acid (mg/100g)	22	6.77%
16	Total sugars (%)	91	28%
17	Protein (%)	1	0.31%

Table 5: Cluster mean values of seventeen characters of 26 genotypes in pea

Sl. No.	Characters	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Mean
1.	Days to first flower	45.86	29.5	51.11	46	55.33	45.56
2.	Days to 50% flowering	49.92	34.17	55.67	50.33	61.67	50.35
3.	Days to first picking	84.36	75.92	87.11	83.00	97.00	85.48
4.	No. of primary branches	2.58	1.75	2.62	2.67	3.00	2.52
5.	Inter-nodal length (cm)	4.61	3.48	3.94	2.91	3.13	3.61
6.	Nodes per plant	17.17	12.58	16.00	15.53	16.17	15.49
7.	Plant height (cm)	62.01	46.84	57.66	71.70	58.91	59.42
8.	Pod length (mm)	89.35	77.85	86.31	85.54	86.66	85.14
9.	Pod diameter (mm)	11.38	12.42	11.90	11.91	13.30	12.18
10.	Seeds per pod	7.24	7.32	7.16	7.42	7.40	7.30
11.	Shelling (%)	48.20	47.13	46.53	47.27	48.75	47.57
12.	Pods per plant	10.67	8.28	9.83	11.97	13.74	10.89
13.	Pod yield per plant (g)	68.68	51.3	60.34	77.10	95.02	70.48
14.	T.S.S. (˚Brix)	14.51	14.35	14.25	15.13	15.36	14.72
15.	Ascorbic acid (mg/100g)	19.09	17.66	18.36	19.66	20.97	19.14
16.	Total sugars (%)	4.29	4.04	3.94	6.57	6.09	4.98
17.	Protein (%)	21.56	21.79	21.12	22.77	22.28	21.90

Table 6: Cluster combinations and possible cross combinations of twenty-six selected genotypes of pea

S. No.	Cluster Combination	D^2 values	Cross Combination	Characters
1.	II × V	588.09	Kashi Mukti, Kashi Uday, Kashi Nandini, VM- 11 × EC- 507771, Nirali, Kashi Samridhi	Days to first flower, Days to 50% flowering, Days to first picking, No. of primary branches, Pod diameter (mm), Shelling (%), Pods per plant, Pod yield per plant (g), T.S.S. (˚Brix), Ascorbic acid (mg/100g) and Protein (%)
2.	II × III	342.33	Kashi Mukti, Kashi Uday, Kashi Nandini, VM- 11 × VM-12, VM-10, Punjab- 89, IC-109696, Pea Vasundhra, NS- 1100	Days to first flower, Days to 50% flowering, Days to first picking
3.	II × IV	283.84	Kashi Mukti, Kashi Uday, Kashi Nandini, VM- 11 × Haze- 02	Days to first flower, Days to 50% flowering, Days to first picking, Inter-nodal length (cm), Plant height (cm), Seeds per pod, Total sugars (%)