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Genetic diversity and trait association analysis in field pea (*Pisum sativum* L.) genotypes

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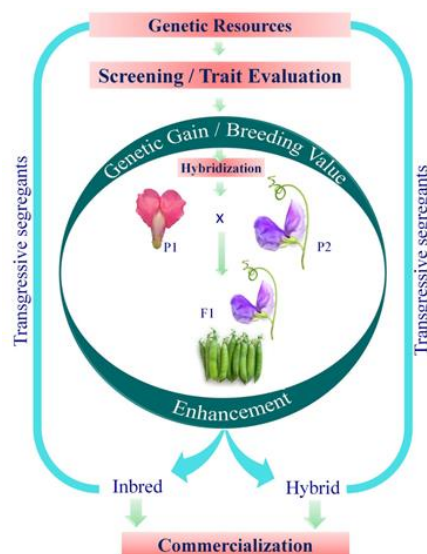
Abstract

Aim: The objective of this study was to scrutinize the genetic diversity using twenty-eight genotypes (Parents + Hybrids) with the help of 12 agro-morphological traits and 18 useful SSR markers.

Methodology: The study carried out with 28 genetically diverse pea genotypes (7 parents and 21 F₁ crosses, developed in half diallel fashion) with recommended agro-practices and evaluation were done under Randomized Block Design (RBD). The data observed were analyzed by SPAR.2.0 software.

Results: It was found that parentage and crosses have genetic variability (among parentage and F₁s) for all the agro-morphological traits. The extent of lowest and highest PCV and GCV was recorded for days to maturity and seed yield per plant respectively. Broad-sense heritability magnitude ranged from 28.17% for number of seeds/pod to 76.31% for Plant height. Along with high genetic advance (>20%) highest heritability (>60%) coupled and was assessed for number seeds/pod (76.31% to 26.35%). The No. of pod per plant ($r = 0.685$ and 0.670 , respectively at $P \leq 0.01$) has substantial genotypic and phenotypic correlation with seed yield/plant followed by pod length (0.639), number of nodes to first flowering (0.576) etc. Using molecular data measured genetic coefficients revealed varying degree of genetic relatedness among the 7 parental lines of field pea and 0.12 to 0.89 range Jaccard's similarity coefficient has been reported. On the basis of 18 SSR markers study 7 parental lines of field pea can be differentiated into two major groups, where with highest Jaccard's similarity coefficient (0.89) Prakash consisted in group 1.

Interpretation: Genetic variability plays essential role in selection of genotypes with desired good trait of interest. Presence of high heritability helps in enhancing the selection efficiency and genetic gain in field pea crop.



Keywords: Genetic diversity, correlation coefficient, agro-morphological traits, *Pisum sativum* L.

Introduction

Pulses having relatively high protein content have given the status of 'wonder crop', which makes the diet more balanced in its nutritive value. Pea (*Pisum sativum* L.) is known to be one of the oldest cultures along with cereals and lens in the world (Zohary *et al.*, 2012) [33]. The main use of Field pea is for human consumption and livestock feeding. Pea is rich source of

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Proteins (21 to 25%) hence, served as potential alternative to soybean in western countries (Barac *et al.*, 2010) [4]. Being second most important pulse after common bean it is widely cultivated worldwide (Esposito *et al.*, 2007) [11] in an area of 68,68,131.0 ha with global production of 1,13,32,772 tons (FAOSTAT, 2014) [12]. Canada is the leading field pea producer country (approximately 3 million metric tons in 2012) followed by France, Russian federation, China mainland and Ukraine (Jansen *et al.*, 2014).

In India, where majority of the population of is vegetarian pulses are the major source of protein. Besides this with the property of good source of carbohydrates and total digestible nutrients (86 to 87%), considered as excellent livestock feed (Enderes *et al.*, 2016). During 2015-16, pulses were grown an area of 24.91 million ha with a production of 16.35 million tone and average productivity was 656 kg/ha. Despite its potential attributes, the per capita availability of pulses in India has declined sharply from 61 g per day during 1951 to 43 g per day during 2016 (Annual Report DPD, 2016-17) In India, during 2015-16, pea was grown over an area of 0.90 million ha with a production of about 0.74 million tone and average productivity was 821 kg/ha (Annual Report DPD, 2016-17), which is highest among Rabi pulses grown in India. However, the current status of productivity of pea is far below than other countries of the globe.

Pea in India grown and utilized since ancient times but due to introduction of exotic collection and adoption to improved varieties the great heritage is eradicated and disappeared. Genetic diversity in crop is principle tool in formulation of breeding strategies for further invigoration in genetic pool. The magnitude of genetic variability and heritable desirable characters are prerequisite for Crop improvement and successful breeding programme (Kaur *et al.*, 2018) [31]. Similarly, genetic variability, heritability and genetic advance are also play important role for improvement of any crop for selection of superior genotypes and improvement of any traits. Yield is an intricate parameter influenced by several genetic factors interacting with surroundings. Therefore, success of any breeding programme for its improvement depends on the existing genetic variability in the base population and on the efficiency of selection (Kumari *et al.*, 2008) [19]. The knowledge of association of characters is of crucial importance in developing an efficient breeding programme. The proportion of phenotypic variance that is due to genotypes which is heritable defines Heritability. The trait of high heritability serves as useful guide for effective trait selection with genetic advance. It will be possible to decide various breeding programmes for improvement of different characters based on the study of heritability and genetic advance (Kumari *et al.*, 2012) [20].

The objective of this study was to determine genetic diversity and trait association among pea genotypes (Parents + Hybrids) using molecular and agro-morphological markers.

Materials and Methods

Experimental materials and design

The experiment has been carried out at the department of Plant Breeding and Genetics, CAU, Imphal, Manipur using 7 genetically diverse pea varieties (Table 1). The parentages were sown in crossing blocks and F₁s were generated in diallel fashion (single diallel) during Rabi 2013-14. The evaluation was done using Randomized Block design (RBD) with F₁s along with parents (7 parents + 21 F₁ crosses) following three replications during Rabi 2014-15. Seeding was done with a spacing of 30cm x 10cm, each treatment had

one line with 4 m length with all recommended agro-practices. The data were recorded for 12 agro-morphological characters like days to first flowering (DFsF), number of nodes to first flowering (NNFF), days to 50% flowering (DFF), days to maturity (DM), plant height (Pht), number of pods per plant (NPP), pod length (PL), number of seeds per pod (NSP), seed yield per plant (SYP), biological yield per plant (BYP), 100-seeds weight (SW) and harvest index (HI).

Table 1: Genotypes detail and their sources

S. No.	Genotypes/crosses	Developing institute
1	Makyatmubi	CAU, Imphal
2	Makuchabi	CAU, Imphal
3	KPMR851	CSAUAT, Kanpur
4	Prakash	IIPR, Kanpur
5	Pant P 217	GBPUAT, Pantnagar
6	Rachana	CSAUAT, Kanpur
7	VL 58	VPKAS, Almorah
8	21 F ₁ s developed in half diallel fashion at CAU, Imphal	

Data collected and analysis

The data for 12 agro-morphological characters were analyzed by SPAR.2.0 software. Standard statistical techniques were used and the data were subjected to analysis of variance (ANOVA) using the Fisher's least significant difference (LSD) method to test the significance difference between means. To compare the differences among the genotype means the significant data were further analyzed statistically using Least Significant Difference (LSD) test at 5% and 1% probability level. Clustering algorithm of Ward's method was adopted for the cluster analysis. (Ward, 1963) [32] using Statistica for Windows 7.1.

DNA isolation and molecular marker assay

The genetic diversity among parents and hybrid lines was done by utilizing microsatellite markers (SSR) selected from Smykal *et al.* 2007, 2008 [26]. SSR scores were converted into binary data by presence (1) or absence (0) of the selected fragment for genetic similarity, cluster and structural analysis. Jaccard index of similarity (Reif *et al.* 2005) [23] were used to calculate Genetic similarity coefficients using SPSS 12 software (SPSS 2003). Calculation of Polymorphic information content (PIC) for each marker were performed using the following formula: $PIC_i = 1 - \sum P_{ij}^2$, where P_{ij} is the frequency of the j^{th} allele. Based on similarity matrix of Jaccard coefficients, genetic data in factorial space, multidimensional scaling (MDS), visualization was adopted (Kruskal 1964) [21]. Morphological descriptors were analyzed using principal component analysis (PCA).

Results and Discussion

Existing of genetic variability/diversity among germplasm/lines is essential for enhancing the genetic gain/breeding value in crop plants. The experimental material taken under study possess great genetic variability (among parentage and F₁s) for all the traits studied (Table 2) which are also found in concordance with the works of Gixhari *et al.* (2014) and Khan *et al.* (2013) [18]. The substantial heterotic value of resultant hybrid over parental value indicates good combing ability among parentage involved, which is more relevant to be utilized for maximizing genetic gain in field pea hybrids. The existing genetic diversity among parents and crosses might be useful for making strategies for further invigoration of field pea breeding programs (Cupic *et al.*, 2009) [9] as facilitates wide scope for selecting good

combining parents for breeding programmes (Gatti *et al.*, 2011) [13]. Substantial genetic differences among treatments (seven parents + twenty-one hybrids) indicates high breeding value available in the studied materials which is corroborated with the findings of Bisht and Singh (2011) [7] and Esposito *et al.*, (2013) [13].

Vast variability for plant height which is having positive association with other yield traits was reported in parents (132.7 cm in Pant P 217 to 58.73cm in Prakash) and crosses (139.00 cm in Prakash x Pant P 217 to 109.0 cm in Rachna x VL-58) signify the scope for further yield enhancement in field pea via transgressive as well as heterosis breeding (Table 3). Similar result obtained by Researchers with lengths varying between 65.67 and 132.0 cm (Ceyhan and Avci, 2015) [8], 51.20 and 111.30 cm (Georgieva *et al.*, 2016) [14], 65.67 and 126 cm (Khan *et al.*, 2013) [18]. On the other hand, the average length 63.64 cm is reported by Habtamu and Million (2013) [16] which is lower than that obtained in the present work (90.05 cm). The difference in plant height might

be due to genotype of the lines and environment adaptability (Khan *et al.*, 2013, Solberg *et al.*, 2015) [18, 27].

Extent of variability was determined by analyzing genetic coefficient of variability (GCV) and phenotypic coefficient of variability (PCV) (Table 4). It was reported lowest for days to maturity (GCV=1.08 and PCV=1.97) and highest for seed yield per plant (22.76 and 33.12, respectively). Presence of great phenotypic coefficient of variability (PCV) for (>20%) for seed yield per plant (33.12%), biological yield per plant (28.81%) and number of pod per plant (24.40%) indicates substantial effect of environment on the expression of characters which is strongly supported by findings of Bashir *et al.* (2017) and Meena *et al.* (2017) [5, 22]. While Higher genotypic coefficient of variation (GCV) for seed yield per plant (22.76%), biological yield per plant (18.74%) and number of pods per plant (16.92%) reported to have minor environmental interference, thus, having greater scope to be improved (Bagati *et al.* 2016) [3].

Table 2: ANOVA for different characters of field pea

Source of variation	d.f.	Mean sum of squares											
		DFsF	NNFF	DFE	DM	Pht (cm)	NPP	PL (cm)	NSP	SYP (g)	BYP (g)	100 SW (g)	HI (%)
Replication	2	21.893	0.940	19.393	6.655	59.465	0.155	0.040	0.464	9.27	58.333	9.79*	74.33**
Genotypes	27	25.344**	3.302**	23.275**	7.458**	709.920**	9.815**	0.520**	0.876**	24.10**	96.64**	21.33**	47.08**
Error	54	9.485	1.015	7.936	3.260	66.566	2.599	0.124	0.403	4.057	19.56	2.27	10.55

*, ** Significant at 0.05 and 0.01 levels, respectively

Table 3: Mean value of different characters for morphological trait of parent and crosses in field pea (Half di allele)

Parent/Cross	DFsF	NNFF	DFE	DM	Pht (cm)	NPP	PL (cm)	NSP	SYP (g)	BYP (g)	100 SW (g)	HI (%)
C1	64.67	16.33	68.33	109.33	118.07	10.00	7.04	6.33	13.80	30.52	25.08	45.12
C2	60.67	16.00	67.00	109.00	120.67	13.00	7.13	5.33	15.49	37.02	22.61	41.52
C3	67.00	16.33	71.00	111.00	128.80	9.00	7.31	5.67	12.02	32.29	25.90	37.45
C4	62.67	15.67	66.67	109.00	134.47	10.00	7.27	6.33	13.77	31.73	22.08	43.31
C5	68.67	15.00	73.00	112.00	112.67	8.00	6.33	5.33	12.20	31.53	21.21	38.92
C6	61.33	14.67	67.33	109.67	111.53	9.00	6.56	5.67	11.79	28.80	24.83	46.87
C7	60.00	16.33	65.67	109.00	116.47	10.67	6.75	6.00	13.42	27.88	19.72	47.81
C8	62.67	16.00	67.00	109.00	120.40	10.33	7.11	6.33	13.91	29.02	23.15	47.92
C9	63.00	16.00	67.67	110.00	115.97	8.33	6.92	6.33	12.60	27.06	20.41	46.87
C10	66.33	15.67	71.33	111.00	118.40	9.33	6.96	6.33	10.62	25.23	19.86	42.18
C11	62.00	13.67	67.00	108.00	118.83	13.00	7.19	6.67	15.36	34.13	20.71	45.17
C12	58.33	15.00	65.00	108.33	132.13	10.00	6.43	5.00	11.73	25.43	21.07	46.19
C13	58.00	14.33	65.00	108.00	130.40	12.67	6.17	5.67	11.43	25.80	18.42	44.47
C14	62.33	13.33	64.67	107.67	114.33	11.67	6.49	5.67	8.49	22.00	18.01	38.93
C15	61.33	13.33	63.67	108.33	116.80	8.33	6.33	5.33	9.76	22.87	19.15	43.27
C16	60.00	16.00	64.00	108.33	139.00	8.00	6.57	6.33	11.40	25.26	19.99	45.11
C17	63.33	15.67	67.33	109.00	130.47	8.67	6.43	5.33	9.10	21.07	20.03	43.03
C18	63.67	15.00	68.00	109.33	136.07	10.00	5.76	5.00	11.99	27.08	19.04	44.22
C19	62.00	14.33	65.67	109.33	125.73	8.67	6.35	6.67	11.81	26.98	17.81	43.88
C20	59.33	13.33	64.33	107.67	134.57	8.00	6.10	6.33	10.41	24.70	15.75	45.18
C21	63.00	14.00	66.33	109.33	109.27	8.67	6.51	5.67	7.53	19.80	17.22	38.24
P1	63.00	15.33	69.00	110.33	111.27	6.67	7.27	5.00	8.70	21.47	23.51	40.43
P2	63.67	15.67	69.33	110.67	105.53	7.33	6.82	6.33	6.60	16.46	18.43	40.11
P3	62.33	15.33	67.00	110.00	109.27	8.67	6.37	5.67	7.73	20.96	18.46	36.76
P4	70.33	15.33	74.67	114.00	58.73	6.00	6.37	5.00	6.77	15.78	20.93	43.17
P5	61.67	14.33	68.67	110.00	132.07	7.67	6.61	6.33	7.70	18.14	17.42	42.42
P6	67.00	13.00	71.67	112.67	111.53	7.67	5.99	5.33	5.29	15.72	15.76	33.44
P7	61.67	13.67	70.67	112.33	104.33	7.33	6.49	6.00	6.21	18.48	18.25	33.50

(C1= Makyatmubi x Makuchabi, C2= Makyatmubi x KPMR851, C3= Makyatmubi x Prakash, C4= Makyatmubi x Pant P 217, C5= Makyatmubi x Rachna, C6= Makyatmubi x VL 58, C7= Makuchabi x KPMR851, C8= Makuchabi x Prakash, C9= Makuchabi x Pant P 217, C10= Makuchabi x Rachna, C11= Makuchabi x VL 58, C12= KPMR851 x Prakash, C13= KPMR851 x Pant P217, C14= KPMR851 x Rachna, C15= KPMR851 x VL 58, C16= Prakash x Pant P 217, C17= Prakash x Rachna, C18= Prakash x VL 58, C19= Pant P 217 x Rachna, C20= Pant P 217 x VL 58, C21= Rachna x VL-58, P1= Makyatmubi, P2= Makuchabi, P3= KPMR851, P4= Prakash, P5= Pant P 217, P6= Rachna, P7= VL 58)

Table 4: Genetic value for different morphological characters in field pea

S. No	Variables	Vg	Vp	GCV%	PCV%	H ²	GA	GAM
1.	Days to first flowering (DFsF)	5.29	14.77	3.66	6.12	35.76	2.83	4.51
2.	No. of nodes to first flowering (NNFF)	0.76	1.78	5.84	8.92	42.90	1.18	7.88
3.	Days to 50% flowering (DFF)	5.11	13.05	3.34	5.33	39.18	2.92	4.30
4.	Days to maturity (DM)	1.40	4.66	1.08	1.97	30.04	1.34	1.22
5.	Plant height (cm) (Ph)	214.45	281.01	12.35	14.15	76.31	26.35	22.24
6.	No. of pods per plant (NPP)	2.41	5.00	16.92	24.40	48.06	2.22	24.16
7.	Pod length (cm) (PL)	0.13	0.26	5.48	7.63	51.63	0.54	8.11
8.	No of seeds per pod (NSP)	0.16	0.56	6.83	12.86	28.17	0.43	7.46
9.	Seed yield per plant (g) (SYP)	5.86	12.40	22.76	33.12	47.24	3.43	32.23
10.	Biological yield/plant (g) (BYP)	21.86	51.66	18.74	28.81	42.31	6.27	25.11
11.	100 seed weight (g) (SW)	6.21	8.89	12.36	14.78	69.85	4.29	21.27
12.	Harvest Index (%) (HI)	12.18	22.73	8.24	11.26	53.58	5.26	12.43

The magnitude of broad-sense heritability ranged from 28.17% for number of seeds per pod to 76.31% for plant height (Table 4). The highest heritability (>60%) coupled with high genetic advance (>20%) was assessed for number of seeds per pod (76.31% to 26.35%). It is reported that high heritability and genetic advance trait could be due to additive gene action and have high response to selection. During selection breeding importance should be given to these traits, whereas, traits exhibiting low genetic advance accompanied by moderate to high heritability indicate the non-additive gene action, thus selection should be practiced with care in respect of low heritable traits. Regarding this, further explanation of Sardana *et al.* 2007 [25] suggested that traits with high heritability might not necessarily lead to higher genetic advance. Low heritability coupled with low genetic advance was assessed for the most of the traits, indicating ineffectiveness of direct selection.

The direct contributor of yield in field pea i.e the size of pod has reported great variability, ranged from 5.99 cm in Rachna and 7.27 cm Makyatmubi; and among crosses and recorded longest in cross Makyatmubi x Prakash (7.31 cm) and shortest in the cross Prakash x VL-58 (5.76 cm). Another dominant trait in field pea, i.e Pod size is mainly depending on plant vigor (Khan *et al.*, 2013) [18] which is proved in current study as cross Makyatmubi x Prakash recorded longest pod involved parent having longest pods. Besides these wide range of variation was noticed for peduncle length (3.10 - 9.75 cm). Substantial genetic variability for yield traits among parents and crosses was also recorded which shows the cross combination Pant P-217 x Rachna and Makuchabi x VL-58 differed significantly from the other genotypes with an average of 6.67 grain per pod (Table 3) represented the high value. However, crosses KPMR-851 x Prakash (5.00) and Prakash x VL-58 (5.00) recorded minimum grain per pod. The experimental results obtained in the studied experimental are comparatively higher than those (2.87 and 5.73 grain per pod) reported by Ceyhan and Avci (2015) [8]. Hence, it was found that parentage involved in the study may be very useful in breeding field pea for grain number per pod. The combination of Makyatmubi x Prakash (25.90 g) which is recorded highest test weight (100-seed weight) might be useful in development of transgressive segregants as well as exploitation of heterosis in field pea. The crosses recorded highest seed yield and biological yield per plant (Makyatmubi x KPMR-851) 15.49g and 37.02g respectively might be useful in development of breeding strategies and varieties with maximum genetic gain. Recombination breeding through multiple crosses involving these hybrids would be desirable to breed genotypes combining these characters. The

experimental results found to be concordance with Singh *et al.* (2005) [28], and Brar *et al.* (2012) [6]. Makuchabi x Prakash (47.92%) recorded maximum harvest index followed by Makuchabi x KPMR-851 (47.81%) while, the cross combination Makyatmubi x Prakash (37.45%) and parent rachna (33.44%) recorded the minimum harvest index.

Overall on the basis of results of mean performance, *sca* effects and standard heterosis, the Makyatmubi x KPMR-851, Makuchabi x VL-58 and Makuchabi x Prakash were identified as the most promising cross combinations to give transgressive segregants.

The trait association analysis revealed that that number of pods per plant ($r = 0.685$ and 0.670 , respectively at $P \leq 0.01$) has substantial genotypic and phenotypic correlation with seed yield per plant followed by pod length (0.639), number of nodes to first flowering (0.576) etc. (table 5). However, days to 50 percent flowering and days to maturity has substantial negative correlation with seed yield per plant. Hence, the traits having positive correlation might be very useful for selection of high yielding genotypes.

Biological yield however has shown substantial positive correlation with seed yield per plant ($r = 0.953$ genotypic and 0.950 phenotypic, respectively at $P \leq 0.01$) revealed good coordination between source and sink in field pea. Among component traits like number of nodes to first flowering, pod length, biological yield per plant (g) has substantial positive correlation with 100-seed weight which is also positively correlated with seed yield per plant, hence, in field crop selection for above component traits will have substantial effects on overall yield. Number of seeds per plant was correlated positively and significantly with pod length ($r = 0.403$ phenotypic and $r =$ genotypic respectively at $P \leq 0.05$) showed close proximity with earlier works (Tofiq *et al.*, 2015) [30].

The PCA results revealed the first PC components accounted for 75.0% of the variation (41.0, 25.0 and 9.0 for PC1, PC2 and PC3 respectively (table 6, Fig. 1 and 2). The first component was showed positive relation for almost all 9 traits except days to first flowering, days to fifty percent flowering and days to maturity which showed negative correlation. However, PC2 explained 25% of total variation where all traits except plant height, number of pods per plant and number of seed per pod, rest are shown positive relation to the major yield contributing traits. Besides, number of seeds per pod and pod length recorded positive relation with yield contributing traits in PC3 shown positive relation with yield contributing traits in field pea. These results showed close association with the findings of Gixhari *et al.* (2014).

Table 5: Genotypic and phenotypic correlation coefficient in field pea

Traits	Genotypic correlation/ Phenotypic correlation	NNFF	DFF	DM	Pht (cm)	NPP	PL (cm)	NSP	SYP	BYP (g)	100 SW(g)	HI (%)
DFsF	G	0.192	0.877**	0.871**	-0.793*	-0.487*	0.084	0.486*	-0.256	-0.083	0.346	-0.499**
	P	0.134	0.862**	0.739**	-0.349	-0.392*	0.041	-0.040	-0.265	-0.181	0.059	-0.316
NNFF	G		0.164	0.047	0.062	0.062	0.792**	0.121	0.576**	0.546**	0.833**	0.4807**
	P		0.165	0.106	0.038	0.017	0.326	0.038	0.313	0.227	0.455*	0.2233
DFF	G			1.051**	-0.802**	-0.474*	-0.087	-0.518**	-0.292	-0.121	0.287	-0.565**
	P			0.853**	-0.385	-0.439*	0.079	-0.035	-0.351	-0.269	0.098	-0.365
DM	G				-1.018**	-0.748**	-0.035	-0.573**	0.554**	-0.414*	0.069	-0.672**
	P				-0.399	-0.545**	-0.072	-0.081	-0.469*	-0.384*	0.027	-0.443*
Pht (cm)	G					0.464*	0.037	0.440*	0.532**	0.518**	-0.046	0.352
	P					0.305	-0.020	0.130	0.353	0.346	-0.008	0.183
NPP	G						0.344	0.317	0.685**	0.677**	0.207	0.348
	P						0.082	0.005	0.670**	0.655**	0.116	0.288
PL (cm)	G							0.319	0.639**	0.711**	0.825**	0.185
	P							0.403*	0.260	0.268	0.492**	0.109
NSP	G								0.442*	0.369*	-0.244	0.349
	P								0.160	0.097	0.013	0.209
SYP (g)	G									0.953**	0.681**	0.762**
	P									0.950**	0.418*	0.548**
Biological yield per plant (g)	G										0.747**	0.535**
	P										0.404*	0.278
100 seed weight (g)	G											0.299
	P											0.280

*(1st row of each trait) for genotypic value, (2nd row of each traits) for phenotypic value

Table 6: Matrix of eigenvalues and eigenvectors of principal components for studied characters in field pea.

Eigen vectors												
	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11	PC12
Eigen values variances	4.93	3.05	1.08	0.89	0.65	0.60	0.33	0.19	0.15	0.11	0.02	0.00
% contribution	0.41	0.25	0.09	0.07	0.05	0.05	0.03	0.02	0.01	0.01	0.00	0.00
% cumulative	0.41	0.66	0.75	0.82	0.87	0.92	0.95	0.97	0.98	0.99	0.99	0.99
DFsF	-0.29	0.35	-0.05	0.18	0.26	0.23	-0.11	0.76	-0.11	0.02	0.20	0.00
NNFF	0.13	0.41	0.11	-0.41	-0.20	0.25	0.69	0.05	-0.19	0.08	-0.07	0.00
DFF	-0.32	0.35	-0.02	0.19	0.17	0.18	0.05	-0.20	0.50	-0.11	-0.61	-0.07
DM	-0.38	0.27	0.03	0.07	0.11	0.13	0.07	-0.51	0.09	-0.05	0.68	0.12
Pht (cm)	0.31	-0.18	0.02	0.11	-0.37	0.72	-0.13	0.08	0.39	-0.05	0.17	0.02
NPP	0.33	-0.02	-0.32	0.43	0.15	-0.29	0.50	0.12	0.39	0.22	0.19	0.05
PL (cm)	0.17	0.39	0.35	0.15	-0.41	-0.41	-0.10	0.13	0.17	-0.53	0.11	0.02
NSP	0.17	-0.02	0.82	0.28	0.26	0.05	0.01	-0.06	-0.04	0.38	-0.03	0.00
SYP(g)	0.35	0.26	-0.21	0.32	0.12	0.18	-0.12	-0.19	-0.41	-0.15	-0.18	0.58
BYP (g)	0.15	0.47	-0.17	-0.18	-0.24	-0.14	-0.43	-0.06	0.14	-0.64	0.02	0.00
100 SW (g)	0.31	0.04	0.08	-0.57	0.55	-0.03	-0.13	0.08	0.36	-0.20	0.09	0.26
HI (%)	0.39	0.22	-0.13	0.09	0.09	0.12	-0.11	-0.18	-0.19	-0.19	0.09	-0.74

18 informative micro-satellite markers are utilized to revealed substantial degree of genetic diversity among the parentage and hybrids involved in this study. Measuring genetic coefficients using molecular data revealed varying degree of genetic relatedness among the 7 parental lines of field pea. Jaccard's similarity coefficient ranged from 0.12 to 0.89 which is due to genetic disparity in the morphology and pedigree among the genotypes. Clustering results showed a clear distinction into major and minor groups in 7 parental genotypes by using 18 microsatellite markers. All these 18 markers could able to distinguish 7 parental lines of field pea into two major groups. The group-I, found as smallest group and contained 1 genotype; Prakash (with 0.76 Jaccard's similarity coefficient). While, Group II was largest, contained 6 parent genotypes including Makyatmubi, Makuchabi, KPMR851, Pant P 217, Rachna and VL 58.

The group-II was further categorized in to two sub groups i.e. Sub-group-IIA and Sub-group-IIB. The Subgroup Sub-group-IIA contains three genotypes i.e. Pant P 217, Rachna and VL 58 (with 0.37 Jaccard's similarity coefficient). Another sub

group (Sub-group-IIB) contains three genotypes; however, within this sub-group Makyatmubi, Makuchabi were more genetically similar (with 0.52 Jaccard's similarity coefficient). Based on the result, genotypes had more genetic distance recorded higher seed yield per plant. Thus, those parental lines having more genetic diversity can be used for more enhancement of genetic gain in field pea (Tar'an *et al.* 2005) [29].

By "Average linkage" method (Bhuvanewari *et al.*, 2017) [24] pattern of genetic diversity was also analyzed using cluster analysis based on similarity index. Total 28 genotypes (7 parents and 21 hybrids) categorised into 2 main clusters (Fig.2). The member within cluster being with more genetic relatedness than the members of another cluster. The number of genotypes among clusters varied from 1 to 27. The maximum numbers of genotypes (Fig. 3) were included in cluster II and there was only one genotype, Prakash in Cluster I. the cluster II is further divided into two sub-cluster Sub-cluster IIa Sub-cluster IIB with 0.95 Euclidean distance for linkage. Cluster IIa Consisting 10 genotypes including

parental genotype Pant P 217 and cluster IIb consisted 17 genotypes where most of the parental lines present. Makyatmubi, Makuchabi, KPMR851, Rachna and VL 58.

The hybridization results showed that distantly related parental combination has substantial yield heterosis in hybrids generated.

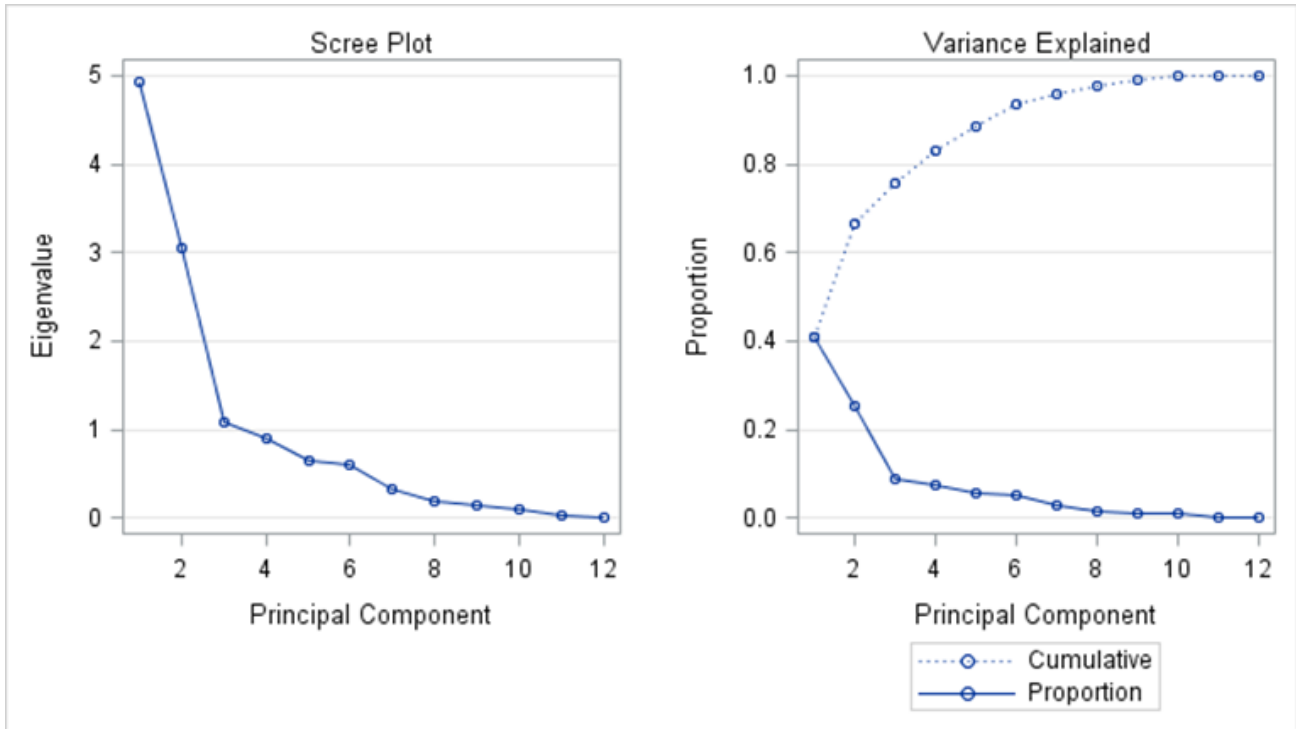


Fig 1: Screen plot of PCA components

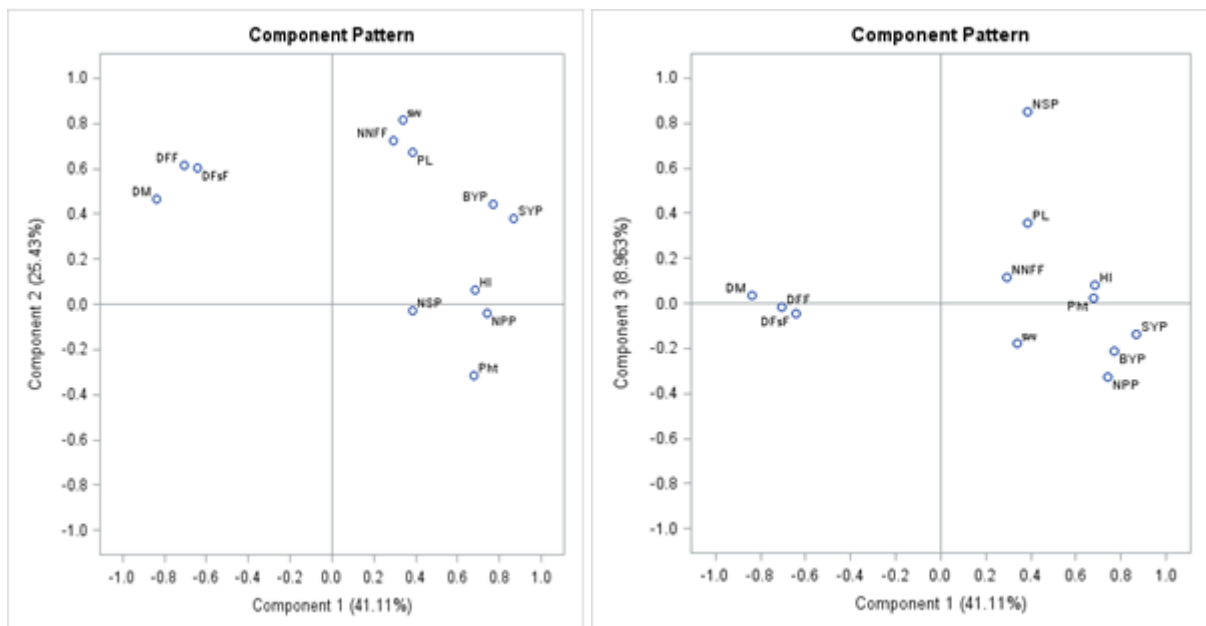


Fig 2: Patterns of PCA components

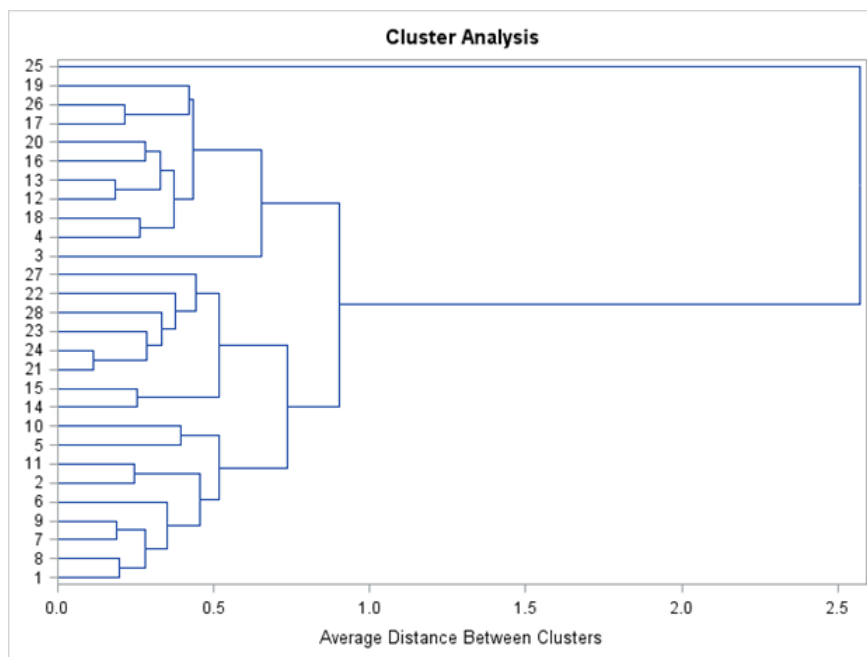


Fig 3: Phenotype based relatedness patterns among parentage and respective hybrids

Conclusion

Based on overall analysis, it is concluded that existence of wide range of genetic variability in yield and its contributing traits are valuable to be utilized in breeding programme for further maximizing genetic gain in field pea. The genotypes like Makyatmubi and Makyatmubi, Makuchabi, KPMR851, Rachna and VL 58 can be utilized for maximizing genetic gain in field pea through heterosis/transgressive breeding.

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Conflict of Interests

The manuscript does not have any personnel/institutional conflict of interest at any level.

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