



## Genetic Diversity of Field Pea Genotypes (*Pisum sativum* L.) in Relation to their Plant Type using Multivariate and Genotype-by-Trait Biplot Analysis

Kedir Yimam Assen\*, Gebeyaw Achenef Haile and Aliyi Robsa Shuro

Ethiopian Institute of Agricultural Research, Kulumsa Agricultural Research Center, Asella (489), Ethiopia

### Corresponding Author

Kedir Yimam Assen

e-mail: [kediryimam81@gmail.com](mailto:kediryimam81@gmail.com)

### Article History

Received on 01<sup>st</sup> August, 2024

Received in revised form on 20<sup>th</sup> October, 2024

Accepted in final form on 07<sup>th</sup> November, 2024

### Abstract

The present study was conducted during June–November, 2019 at Bekoji and Kofele substation of Kulumsa Agricultural Research Center (KARC) with the aim to assess the genetic diversity among field pea genotypes for desired morpho-agronomic traits. A total of 49 Field pea genotypes, representing two different plant types were evaluated for 13 characters. Through cluster analysis, the genotypes were grouped into five categories based on the Euclidean distance matrix using the complete linkage method. Cluster one had the most genotypes (20), while cluster five had the fewest (2). Genetic distances among genotypes estimated by Euclidean distances from 13 traits ranged from 14.76 to 5514.77. Principal component and biplot analyses showed that seed yield, plant height, days to 90% maturity, number of pods per plant and seeds per plant were the main factors contributing to genotype divergence. Additionally, genotypes in the prostrate (leafed) type of field pea had a greater genetic distance (diversity) compared to those in the erect (semi-leafless) type. In general this study showed the presence of considerable diversity for the studied traits in field pea genotypes, with differences between plants types even though the dendrogram and PCA didn't show clear cut (distinct) grouping pattern in field pea genotypes with respect to their plant types and sources. This implies an opportunity for improving desired traits in a field pea breeding program through selection or hybridization of these divergent genotypes. Thus, crossbreeding promising parents, especially selected from advanced prostrate and erect types, can result in a good level of genetic recombination.

**Keywords:** Biplot, cluster, diversity, field pea, genotype, plant type

### 1. Introduction

Field Pea (*Pisum sativum* L.) is a cool-season pulse crop that belongs to the family Leguminosae with a chromosome number of  $2n=14$ . In Ethiopia, it is the fourth most important staple food legume after faba bean, common bean, and chickpea. Field pea is typically grown at elevations between 1800-3000 m.a.s.l, much like faba bean. It covers about 219,927.59 hectares of arable lands, with a total production of 3,762,368.83 quintals and an average yield of  $1.71 \text{ t ha}^{-1}$  (Anonymous, 2020). This accounts for 13% of the total area covered by pulses and 11.76% of the total pulses production in the country during the main growing season.

Ethiopia has a wide range of field pea germplasm, making it the secondary center of genetic diversity (Keneni and Jarso, 2005). This implies that Ethiopia has the potential to improve field peas for desired traits through selection and/or hybridization breeding programs. Field pea plays a significant role in the

livelihoods of agricultural communities in Ethiopia. It is a valuable and cheap source of protein, serving as a source of food and feed. Additionally, it plays a great role in soil fertility restoration as a suitable rotational crop due to its ability to fix atmospheric nitrogen.

Although field pea is a crop of great importance, its production is limited by various factors such as low-yielding local varieties, traditional practices, cultivar instability, lodging, biotic factors (diseases like powdery mildew and ascochyta blight), insect pests (field and storage pests), weed infestation, and abiotic factors (drought, soil salinity, frost, etc.) (Maharjan et al., 2015; Singh and Srivastava, 2015; Tegegn and Teshome, 2017; Teshome and Tegegn, 2017). Therefore, increasing the yield is a major objective of breeding programs for field peas, as in most crop improvement programs (Assen, 2020). To achieve this, breeders evaluate germplasm through collection, introduction, and hybridization to develop high-yielding and stress-tolerant varieties.



The selection or hybridization methods are used to genetically improve the desired traits and increase yield (Keneni and Jarso, 2005; Fikreselassie, 2012; Seboka, 2013). To select superior breeding materials from the population, a high level of genetic variability among the genotypes is necessary (Tiwari and Lavanya, 2012). From crop morphology perspective, there are two major types of field peas categorized as leafy (prostrate) and semi-leafless (erect) (Endres and Kandel, 2021). If only the normal-leafed (prostrate) type field peas are exploited to develop new cultivars, it could limit the genetic diversity and exclude desirable traits that exist only in the semi-leafless genotypes (Endres and Kandel, 2021). It is necessary to exploit a wide range of genetic resources including semi-leafless genotypes to minimize production challenges (Tran et al., 2023). Thus, it is important to assess genetic diversity among genotypes in relation to their plant types to identify unique features and important desirable traits in each plant type.

Several studies have been conducted on the genetic diversity of field pea genotypes in Ethiopia (Keneni and Jarso, 2005; Seboka, 2013; Negisho et al., 2017; Assen, 2020). These studies have explored the magnitude and pattern of genetic diversity among field pea genotypes majorly focused on conventional normal leaf types. However, there is limited information available on diversity studies among genotypes that compare the two plant types (normal vs semi-leaf less type). This lack of information led to the present study, which aimed to assess the genetic diversity among genotypes for desired morpho-agronomic traits. This assessment was done through multivariate and genotype-by-trait (GT) biplot analysis. Furthermore, the study aimed to evaluate the clustering (diversity) pattern of field pea genotypes in relation to their plant types and sources and to identify diverse parents that

can be utilized in crop improvement programs.

## 2. Materials and Methods

### 2.1. Experimental site

The experiment was conducted at two locations of South Eastern Ethiopia namely Bekoji and Kofele substation of Kulumsa Agricultural Research Center during the main cropping season in 2018/2019 under rain fed condition. Bekoji is located at an altitude of 2780 m.a.s.l with a geographic co-ordinate of 07° 32'37"N latitude and 39° 15'21" E longitudes. The area receives mean annual rainfall of 1020 mm. The mean annual maximum and minimum temperature of the site is about 18.6 °C and 7.9 °C, respectively. The geographical location of Kofele is 07° 04'28"N latitude and 38° 47'11" E longitudes with an altitude of 2660 meter above sea level (m.a.s.l). The agro-ecology of the area is characterized by an average annual rain-fall of 1211 mm, with annual mean maximum and minimum temperatures of 18 °C and 7.1 °C respectively.

### 2.2. Experimental materials and design

Forty-nine field pea genotypes, including twenty eight prostrate (leafy) and twenty one erect (semi leafless) plant types were used for the study. List of field pea genotypes, code, source, status and plant types are given in Table 1. The experiment was carried out using 7x7 simple lattice designs; each replication containing seven incomplete blocks and each incomplete block containing seven genotypes. Each plot had two rows of 4 m length, with spacing of 20 cm between rows and 5 cm between plants. Each genotype was planted in a plot size of 1.6 m<sup>2</sup>.

Table1. List of experimental materials used for the study

No	Genotype	Code	Source	Plant type	Leaf type	Status
1.	GPHA-05	G-1	HARC	Prostrate	leafy	Advanced line
2.	GPHA-013	G-2	HARC	Prostrate	leafy	Advanced line
3.	GPHA-03	G-3	HARC	Prostrate	leafy	Advanced line
4.	GPHA-019	G-4	HARC	Prostrate	leafy	Advanced line
5.	GPHA-02	G-5	HARC	Prostrate	leafy	Advanced line
6.	GPHA-010	G-6	HARC	Prostrate	leafy	Advanced line
7.	GPHA-07	G-7	HARC	Prostrate	leafy	Advanced line
8.	GPHA-08	G-8	HARC	Prostrate	leafy	Advanced line
9.	GPHA-06	G-9	HARC	Prostrate	leafy	Advanced line
10.	GPHA-012	G-10	HARC	Prostrate	leafy	Advanced line
11.	GPHA-04	G-11	HARC	Prostrate	leafy	Advanced line
12.	GPHA-016	G-12	HARC	Prostrate	leafy	Advanced line
13.	GPHA-09	G-13	HARC	Prostrate	leafy	Advanced line
14.	GPHA-01	G-14	HARC	Prostrate	leafy	Advanced line
15.	GPHA-018	G-15	HARC	Prostrate	leafy	Advanced line



No	Genotype	Code	Source	Plant type	Leaf type	Status
16.	GPHA-017	G-16	HARC	Prostrate	leafy	Advanced line
17.	GPHA-014	G-17	HARC	Prostrate	leafy	Advanced line
18.	GPHA-011	G-18	HARC	Prostrate	leafy	Advanced line
19.	GPHA-015	G-19	HARC	Prostrate	leafy	Advanced line
20.	P-313-010	G-20	ICARDA	Erect	Semi-leafless	Advanced line
21.	P-313-045	G-21	ICARDA	Erect	Semi-leafless	Advanced line
22.	P-313-086	G-22	ICARDA	Erect	Semi-leafless	Advanced line
23.	P-313-082	G-23	ICARDA	Erect	Semi-leafless	Advanced line
24.	P-313-042	G-24	ICARDA	Erect	Semi-leafless	Advanced line
25.	P-313-071	G-25	ICARDA	Erect	Semi-leafless	Advanced line
26.	PDFPTBEK	G-26	ICARDA	Erect	Semi-leafless	Advanced line
27.	G227 63-2C	G-27	HARC	Prostrate	leafy	Released variety
28.	P-313-053	G-28	ICARDA	Erect	Semi-leafless	Advanced line
29.	P-313-070	G-29	ICARDA	Erect	Semi-leafless	Advanced line
30.	P-313-027	G-30	ICARDA	Erect	Semi-leafless	Advanced line
31.	P-313-065	G-31	ICARDA	Erect	Semi-leafless	Advanced line
32.	P-313-026	G-32	ICARDA	Erect	Semi-leafless	Advanced line
33.	P-313-090	G-33	ICARDA	Erect	Semi-leafless	Advanced line
34.	P-313-046	G-34	ICARDA	Erect	Semi-leafless	Advanced line
35.	MILKEY	G-35	HARC	Prostrate	leafy	Released variety
36.	P-313-098	G-36	ICARDA	Erect	Semi-leafless	Advanced line
37.	HASABE	G-37	HARC	Prostrate	leafy	Released variety
38.	HOLETA	G-38	HARC	Prostrate	leafy	Released variety
39.	WALMERA	G-39	HARC	Prostrate	leafy	Released variety
40.	P-313-059	G-40	ICARDA	Erect	Semi-leafless	Advanced line
41.	P-313-061	G-41	ICARDA	Erect	Semi-leafless	Advanced line
42.	P-313-068	G-42	ICARDA	Erect	Semi-leafless	Advanced line
43.	P-313-089	G-43	ICARDA	Erect	Semi-leafless	Advanced line
44.	P-313-067	G-44	ICARDA	Erect	Semi-leafless	Advanced line
45.	P-313-003	G-45	ICARDA	Erect	Semi-leafless	Advanced line
46.	ADI	G-46	HARC	Prostrate	leafy	Released variety
47.	BURKITU	G-47	HARC	Prostrate	leafy	Released variety
48.	BILALO	G-48	KARC	Prostrate	leafy	Released variety
49.	BURSA	G-49	KARC	Prostrate	leafy	Released variety

HARC: Holeta Agricultural Research Center; ICARDA: International Center of Agricultural Research in Dry Areas; KARC: Kulumsa Agricultural Research Center

### 2.3. Collected data

Data were collected on single plant and plot bases. On a plant basis, data were collected from ten randomly selected plants from each genotype in each replication, namely, plant height (PH) (cm), number of pods per plant (NPPP) (number), number of seeds per pod (NSPP) (number) and total number of seeds

plant<sup>-1</sup> (NSPPL) (number).

While the data on plot basis were collected include days to 50% flowering (DF), days to 90% maturity (DM), lodging score (LS), stand count at harvest (SCH), Ascocayta blight (AB), powdery mildew (PM), Frost score, thousand seed weight (TSW) (gram) and seed yield (SYPH) (kg ha<sup>-1</sup>). Assessment of



lodging score was made at physiological maturity using a 1-9 scale (Wang et al., 2006); where, 1=main stems strictly upright, 2=main stems incline slightly, 3=main stems at 60° angle, 4=main stems at 45° angle, 5=main stems at 30° angle, 6=1/2 of the main stems flat, 7=2/3 of the main stems flat, 8=4/5 of the main stems flat and 9=all main stems flat. Ascocayta blight and Powdery mildew disease was recorded using 1–9 scale (Bernier et al., 1993).

## 2.4. Data analyses

### 2.4.1. Cluster analysis

The process of clustering genotypes into different groups, based on multiple traits, was carried out using the complete linkage agglomeration method. This was done by combining mean data using the hclust function of the stats package in R software. To determine the appropriate number of clusters, local peaks of the pseudo-F statistic were examined, along with small values of the pseudo  $t^2$  statistic followed by a larger pseudo  $t^2$  for the next cluster fusion. This was accomplished using SAS version 9.0 (Anonymous, 2002). The dendrogram was built based on the genetic distance using Euclidean distance as a measure of dissimilarity, with a complete linkage method. This was done using the fvizdend function of the factoextra package in R software (Anonymous, 2019).

### 2.4.2. Distance analysis

Genetic distance was determined using the Euclidean distance measure (Green et al., 1974)

$$ED_{jk} = \sqrt{\sum_{i=1}^n (x_{ij} - x_{ik})^2}$$

$ED_{jk}$  = distance between genotypes  $j$  and  $k$ ;  $x_{ij}$  and  $x_{ik}$ =value of phenotypic trait of the  $i^{th}$  character for genotypes  $j$  and  $k$ , respectively; and  $n$ =number of phenotypic traits used to calculate the distance

The average intra and inter cluster distances were calculated using the function cls.scatt.data of clv package in R software.

Square of intra-cluster distance =  $\sum D_i^2 / n$

Square of inter-cluster distance =  $\sum D_i^2 / n_{ij}$

Where;  $\sum D_i^2$ =Sum of distance between all possible combinations,  $n_i$  = number of genotypes in cluster  $i$  and  $n_j$ = number of genotypes in cluster  $j$

### 2.4.3. Principal component analysis (PCA)

The stats package in R software (Anonymous, 2019) was used to perform Principal Component (PC) analysis. To identify the traits that have contributed the most to the total variation among the genotypes, the correlation matrix among traits was taken into consideration as a covariate.

### 2.4.4. Genotype by trait (GT) biplot

The Genotype by Trait (GT) Bi-plot is a novel approach used for visually representing the differences among genotypes, the impact of traits, the characteristics of genotypes for the traits present in each quadrant, and the correlation among traits based on multiple traits. To create this representation, the princomp function of the stats package in R software has been utilized.

## 3. Results and Discussion

### 3.1. Clustering of genotypes

Tables 2a and 2b present the number, proportion, and names of genotypes in each cluster, along with their plant type, status, and sources. The 49 field pea genotypes were categorized into five distinct clusters based on a Euclidean matrix over location, which included two to twenty genotypes (Table 2a) and similarly the dendrogram showed genotypes clustered in different groups based on the 13 measured traits (Figure 1). This indicates that there is considerable diversity among the tested genotypes. Many authors have reported the presence of diversity among field pea genotypes, with different numbers of distinct clusters. For instance, Singh et al. (2021) studied the genetic diversity of 55 pea genotypes based on eleven traits and grouped them into six different clusters. Bhuvaneswari et al (2016) also evaluated 51 field pea genotypes and grouped them into seven distinct clusters. The difference in the number of clusters with the present study was due to the variation in clustering method (dissimilarity matrix), tested genotypes, and the number of variables/traits considered for evaluation. Faiza et al. (2021) also grouped 57 field pea genotypes into five distinct clusters, similar to the present result. Cluster I contained the maximum number of genotypes (20), which represents 40.82% of the total genotypes evaluated. Cluster

Table 2a: Distribution of 49 field pea genotypes in to different cluster groups based on 13 traits

	Clusters				
	I	II	III	IV	V
No. of genotypes	20	4	15	8	2
Proportion (%)	40.82	8.16	30.61	16.33	4.08
Name of Genotypes	G-1, G-2, G-7, G-8, G-10, G-13, G-14, G-15, G-16, G-17, G-21, G-24, G-25, G-27, G-30, G-31, G-37, G-40, G-43, G-45	G-3, G-12, G-18, G-36	G-4, G-5, G-6, G-9, G-11, G-20, G-22, G-29, G-32, G-33, G-34, G-38, G-39, G-41, G-42	G-19, G-23, G-28, G-47, G-49, G-35, G-44, G-46,	G-26, G-48



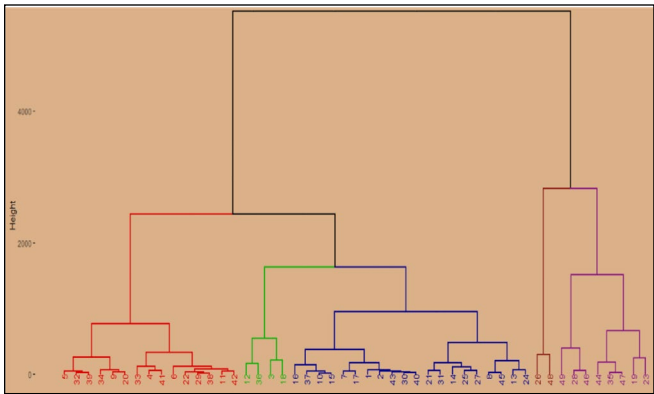


Figure 1: Dendrogram showing 49 field pea genotypes clustering on the basis of 13 traits based on complete linkage method following Euclidian distance

III and IV comprised fifteen (30.61%) and eight (16.33%) genotypes, respectively. Cluster II and V were the smallest and consisted of four (8.16%) and two (4.08%) genotypes, respectively (Table 2a).

Genotypes from different plant types and sources of materials were distributed/grouped under each cluster (Table 2b). The advanced line and released varieties were also grouped under each cluster except cluster II for released variety (Table 2b). This suggest analyses of diversity pattern, among genotypes from different plant type, status and source for quantitative traits revealed existence of phenotypic diversity within plant type, status and source. This may indicates the presence of genetic differences among genotypes from the same plant

Table 2b: Distribution of 49 field pea genotypes over five clusters by two groups of plant types, status and sources based on 13 traits

	clusters					No. of genotypes
	I	II	III	IV	V	
<b>Plant type</b>						
Prostrate	12	3	7	5	1	28
Erect/semi-leafless/	8	1	8	3	1	21
Total	20	4	15	8	2	49
<b>Status</b>						
Advanced line	18	4	13	4	1	40
Released variety	2	0	2	4	1	9
Total	20	4	15	8	2	49
<b>Sources</b>						
Ethiopian (HARC, KARC)	12	3	7	5	1	28
Introduced (ICAR-DA)	8	1	8	3	1	21
Total	20	4	15	8	2	49

type, status and sources. Genotypes from different sources of origin were grouped under the same cluster indicated the germplasm exchange between the national (HARC/KARC) and ICARDA field pea breeding programs, suggesting that these genotypes may share common parents.

In general clustering of genotypes based on the morpho-agronomic traits revealed no distinct plant type and material source grouping patterns in which genotypes from the same plant type, status and source appeared in different clusters or the cluster did not necessarily included all genotypes from the same plant type, status and source. In agreement with the present finding, Keneni et al. (2005) and Negisho et al. (2017) reported that there was no clear diversity pattern between accessions and geographic location (source/origin of collection) even though the high level of intra and inter genetic diversity among field pea genotypes. However, Tran et al. (2023) were observed clear cut grouping pattern of field pea genotypes through cluster dendrogram in to normal leafed and semi-leafless with a few exception in opposite to the present finding.

3.2. Cluster mean analysis

Table 3 lists the mean values of 13 traits for 49 field pea genotypes belonging to five different clusters. Cluster I comprised twenty genotypes, including twelve prostrate and eight erect plant-type field peas. These genotypes had certain characteristics such as late days to 50% flowering, short plant height, susceptibility to lodging and Ascochyta blight disease, and low yields, which were inferior to those of Cluster II. In agreement to this result, similar finding were reported by Yimam et al. (2024). Cluster II contained four genotypes and was characterized by early days to 50% flowering and 90% maturity, short plant height, low number of stands at harvest, seeds per pod and seeds per plant, lower seed size and yield. They were relatively resistant and susceptible to Ascochyta blight and lodging, respectively.

Fifteen genotypes, including seven prostrate and eight erect-type field peas, made up Cluster III, which was characterized by intermediate plant height and pods per plant. These genotypes were also relatively susceptible to frost. Cluster IV had intermediate plant height, a high number of pods and seeds per plant, and relative resistance to lodging and powdery mildew disease. Yirga and Tsegay (2013) were reported similar with the current results. They had higher seed yield next to Cluster V. Cluster V was characterized by late days to 90% maturity, taller plant height, high number of stands at harvest and seeds per pod, higher in seed yield and seed size. They were relatively susceptible and resistant to powdery mildew and frost, respectively. Based on the special merit of each cluster, superior genotypes such as G-15, G-3, G-33, G-44, and G-48 could be selected from Clusters I, II, III, IV, and V, respectively (Table 4). This finding is similar with the finding of Assen, 2020 reported that, field pea genotypes were showed different reaction to powdery mildew disease.



Table 3: Mean value of five clusters for 13 traits in 49 field pea genotypes

Cluster	DF	DM	PH	SCH	LS	PPP	SPP	SPPL	TSW	SYPH	AB	PM	Frost
I	<b>78.67</b>	142.28	106.92	84.38	<b>5.05</b>	8.13	5.05	40.98	190.35	3035.29	<b>4.36</b>	2.70	2.64
II	<b>75.38</b>	<b>141.36</b>	<b>102.91</b>	<b>80.41</b>	4.84	<b>7.71</b>	<b>4.64</b>	<b>35.62</b>	<b>175.46</b>	<b>2237.57</b>	<b>3.89</b>	2.75	2.68
III	77.30	142.99	114.52	82.71	4.73	8.34	4.94	41.36	184.06	3950.54	4.28	2.69	<b>2.79</b>
IV	77.37	142.90	115.75	84.29	<b>4.60</b>	<b>9.10</b>	5.08	<b>46.27</b>	189.98	5348.59	4.34	<b>2.67</b>	2.72
V	77.24	<b>144.57</b>	<b>118.47</b>	<b>90.14</b>	4.70	8.46	<b>5.40</b>	45.43	<b>228.23</b>	<b>7318.57</b>	4.17	<b>2.89</b>	<b>2.06</b>

DF: days to 50% flowering; DM: days to 90% maturity; PH: plant height; SCH: stand count at harvest; LS: lodging score; PPP: pods per plant; SPP: seeds per pod; SPPL: seeds per plant; TSW: thousand seed weight; SYPH: seed yield per hectare; AB: ascocayta blight; PM: powdery mildew

Table 4: Mean of superior genotype selected in each cluster based on the special merit of each

Cluster	Cluster												
	DF	DM	PH	SCH	LS	PPP	SPP	SPPL	TSW	SYPH	AB	PM	Frost
I (G-15)	78.50	143.82	124.18	91.21	7.07	8.36	5.21	43.21	190.82	2996.71	4.14	2.29	2.71
II (G-3)	76.25	143.54	111.75	75.61	4.79	8.07	4.82	38.43	155.82	2164.18	2.89	2.68	2.71
III (G-33)	77.71	141.79	105.18	77.54	3.14	9.25	4.43	40.71	179.18	3957.21	5.04	2.46	2.82
IV (G-44)	77.50	141.86	91.89	84.18	1.93	6.54	4.14	25.50	184.46	5299.29	3.96	2.86	2.93
V (G-48)	77.86	146.18	113.54	93.14	5.32	8.46	5.29	44.61	235.50	7167.18	4.79	3.21	1.93

DF: Days to 50% flowering; DM: Days to 90% maturity; PH: Plant height; SCH: Stand count at harvest; LS: Lodging score; PPP: Pods plant<sup>-1</sup>; SPP: Seeds pod<sup>-1</sup>; SPPL: Seeds plant<sup>-1</sup>; TSW: Thousand seed weight; SYPH: Seed yield ha<sup>-1</sup>; AB: Ascocayta blight; PM: Powdery mildew

In the present study genotypes from different plant types, statuses, and sources of materials were distributed among each cluster, indicating phenotypic diversity. The study has also shown that grouping of genotypes from different plant types and sources of origin under the same cluster indicates that differences in these factors do not necessarily imply differences in genetic makeup. This finding suggests that field pea improvement programs should be focused not only on genetic diversity between different plant types and material sources but also within them. This could lead to more comprehensive and effective strategies for improving field pea genotypes in the future.

### 3.3. Distance analysis

#### 3.3.1. Inter and intra cluster distance

The inter-cluster distance ranged from 797.90 to 5081.32,

Table 5. Average intra (Bold diagonal) and inter (off bold) Euclidian Cluster distance

Cluster	I	II	III	IV	V
I	<b>242.79</b>	797.90	915.31	2313.33	4283.48
II		<b>179.03</b>	1713.04	3111.10	5081.32
III			<b>245.99</b>	1398.07	3368.33
IV				<b>414.83</b>	1970.36
V					<b>151.69</b>

with the largest distance observed between cluster II and V (5081.32), followed by cluster I and V (4283.48) Table 5. This suggests that crosses with parents belonging to the most divergent clusters (Fikreselassie, 2012) will result in the maximum amount of heterosis, making genotypes extracted out of clusters II and V and clusters I and V good choices as parents for hybridization. This is because they are expected to generate desirable segregates with a broad genetic base, which could be further improved through selection in segregating generations. The intra-cluster distances ranged from 151.69 to 414.83, with the maximum distance found in cluster-IV (414.83) followed by cluster-III (245.99), indicating that genotypes grouped in these clusters were more divergent than genotypes in other clusters. Conversely, the minimum intra-cluster distance was observed in cluster V (151.69) followed by cluster II (179.03), indicating that the genotypes in these clusters were genetically closer than any other groups.

#### 3.3.2. Genotypic distance among evaluated genotypes

The genetic distance between field pea genotypes was analyzed in this study using the Euclidian distance method. The range of genetic distance was from 14.76 to 5514.77 with a mean of 1311.50 (Table 6). The largest genetic distance was found between G-18 and G-26 (5514.77) followed by G-3 and G-26 (5306.2), G-18 and G-48 (5212.05), G-12 and G-26 (5136.26), G-3 and G-48 (5003.67), and G-15 and G-48 (4170.73) (Table 7). The smallest genetic distance was between



G-29 and G-38 (14.76), followed by G-10 and G-15 (18.51), G-35 and G-47 (20.72), G-32 and G-39 (23.49), G-8 and G-48 (25.16), and G-22 and G-38 (26.16). Out of the 49 field pea genotypes, 14 (28.57%), including 9 prostrate and 5 erect types, or 9 advanced lines and 5 released varieties, had a mean genetic distance higher than the overall mean of 1311.50, while 35 genotypes (71.43%) had a mean genetic distance below 1311.50 (Table 6). The study indicated variation among genotypes that could be used for field pea breeding programs. It calculated the average genetic distance of each genotype compared to others. The most distant genotypes were G-26, G-48, G-49, G-28, and G-46. While G-42, G-24, G-29, G-38 and

G-13 had lowest mean Euclidian Distance (ED) in ascending order (Table 6). In regard to plant types, the highest Euclidian genetic distances were observed between prostrate and erect type field pea, while the lowest genetic distances were estimated among erect type field pea genotypes. According to Table 7, the genetic distances among erect-type field pea genotypes were lower compared to the prostrate-type genotypes. In other words, there was a wider diversity among prostrate types.

The genetic distance was also observed to be greater in the normal-leafed (prostrate) field pea genotypes than the semi-leafless (erect) ones. This suggested that there was ample

Table 6: Minimum, maximum and mean euclidian distance of each field pea genotypes compared to other genotypes (in each pair)

Genotype	Min.	Max.	Mean	Genotype	Min.	Max.	Mean
G-1	52.0	4680.4	1146.9	G-25	55.0	4313.3	974.3
G-2	28.1	4717.7	1169.2	G-26	303.4	5514.8	3743.8
G-3	170.9	5306.2	1682.5	G-27	55.0	4354.1	988.6
G-4	60.7	3587.1	954.7	G-28	51.5	3831.9	2169.3
G-5	33.9	3336.1	1039.8	G-29	14.8	3804.5	908.2
G-6	47.2	3732.0	919.0	G-30	28.0	4707.2	1162.2
G-7	49.7	4797.8	1230.0	G-31	51.6	4223.8	952.2
G-8	25.2	4078.3	923.7	G-32	23.5	3299.3	1054.6
G-9	33.4	3119.4	1142.0	G-33	83.3	3513.1	976.7
G-10	18.5	4484.1	1038.5	G-34	45.7	3078.2	1164.8
G-11	42.4	3820.2	908.2	G-35	20.7	3179.8	1644.9
G-12	164.0	5136.3	1523.0	G-36	132.8	4973.5	1377.4
G-13	69.6	3951.8	910.9	G-37	40.1	4498.6	1045.8
G-14	97.7	4365.6	998.0	G-38	14.8	3799.3	908.4
G-15	18.5	4473.4	1033.7	G-39	23.5	3317.8	1046.8
G-16	117.0	4596.8	1102.9	G-40	28.0	4700.9	1158.6
G-17	49.7	4842.1	1265.5	G-41	60.7	3626.3	943.6
G-18	213.5	5514.8	1886.5	G-42	42.4	3848.8	905.9
G-19	233.0	2935.4	1476.1	G-43	37.4	4672.8	1140.0
G-20	33.4	3140.2	1131.4	G-44	170.8	3344.0	1771.6
G-21	51.6	4255.4	957.8	G-45	25.2	4094.5	925.7
G-22	26.6	3773.1	912.0	G-46	51.5	3803.5	2145.0
G-23	251.1	2828.4	1316.1	G-47	20.7	3168.2	1636.4
G-24	50.3	3890.0	906.7	G-48	303.4	5212.1	3447.5
				G-49	368.0	4197.8	2496.5
Over all mean of euclidian distance							
Mean of minimum 81.4							
Mean of maximum 4090.6							
Mean of mean 1311.5							



Table 7: Minimum and maximum euclidian distance between genotypes selected from all genotype combinations

Genotype combination	Minimum euclidian distance	Genotype combination	Maximum euclidian distance
B/n G-8 & G-48	25.16	B/n G-3 & G-26	5306.2
B/n G-10 & G-15	18.51	B/n G-3 & G-48	5003.67
B/n G-22 & G-38	26.58	B/n G-12 & G-26	5136.26
B/n G-29 & G-38	14.76	B/n G-15 & G-48	4170.73
B/n G-32 & G-39	23.49	B/n G-18 & G-26	5514.77
B/n G-35 & G-47	20.72	B/n G-18 & G-48	5212.05
B/n= between			

opportunity to improve seed yield and other traits through selection or hybridization of distant field pea genotypes. Furthermore, the Euclidean distance values were higher among advanced lines than released varieties, indicating that there was greater potential for improvement with regard to the status of materials.

Kedir et al. (2024) were reported that normal-leaved (prostrate) field pea genotypes are relatively more variable in thousand seed weight and genotypes in semi-leafless type were more diverse in plant height, ascocayta blight and lodging score in parallel to this finding. Based on the present findings, the most optimal crosses for obtaining the highest amount of heterosis are those between parents chosen from G-18 ×G-26, followed by G-3 ×G-26, G-18 ×G-48, G-12 ×G-26, G-3 ×G-48, and G-15 ×G-48.

Table 8: Range (Minimum and maximum) euclidian genotypic distance regarding to plant type and status of field pea genotypes

prostrate type	Erect type	Prostrate & Erect	Released	Advanced	Advanced and Released
18.51( b/n G-10 & G-15) - 5212.05 (b/n G-18 & G-48)	27.95 (b/n G-30 & G-40) - 4973.45 (b/n G-26 & G-36)	14.76 (b/n G-29 & G-38) - 5514.77 (b/n G-18& G-26)	20.72(b/n G-35 & G-47) - 4195.98 (b/n G-37 & G-48)	18.51(b/n G-10 & G-15) - 5514.77 (b/n G-18 & G-26)	14.76(b/n G-29 & G-38) - 5212.05 (b/n G-18 & G-48)
b/n= between					

### 3.4. Principal component analysis (PCA)

The results of the principal component analysis for 13 traits among 49 field pea genotypes are summarized in Table 8. The first five principal components with eigenvalues greater than one (3.28, 2.25, 1.62, 1.12, and 1.05) accounted for 71.67% of the total phenotypic variation among the genotypes. This is because factors with eigenvalues less than one were ignored following Gutten's lower bound principle. The first two

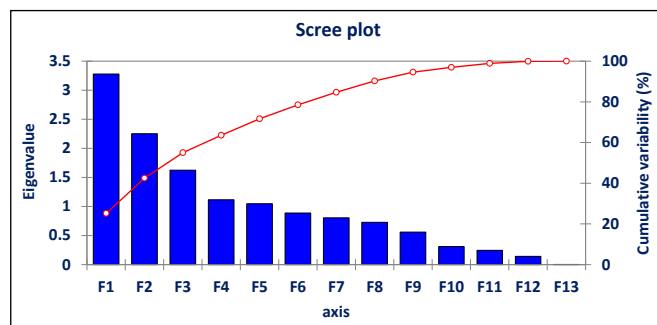


Figure 2: Scree plot showing Eigen value and cumulative variability of each principal component

principal components, PC1 and PC2, were the most significant and accounted for 42.53% of the total variation. A previous report such as Hadis and Dergie (2013) also found that the first five PCs explained 78.9% of the total variation, with PC1 and PC2 explaining the most variability (Figure 2 and Table 9).

Traits such as number of seeds per plant, number of pods per plant, plant height, days to 90% maturity, and number of seeds

Table 9: Eigenvectors, eigenvalues, proportion and cumulative percentage of variation explained by five principal components (PCs) for 13 traits in 49 field pea genotypes over location

Traits	PC1	PC2	PC3	PC4	PC5
DF	0.13	0.27	0.29	<b>0.32</b>	<b>0.36</b>
DM	<b>0.34</b>	<b>0.29</b>	0.11	<b>-0.38</b>	0.17
PH	<b>0.40</b>	-0.25	0.14	-0.22	-0.20
SCH	0.21	<b>-0.45</b>	0.13	-0.19	0.07
LS	0.25	<b>-0.39</b>	<b>0.38</b>	0.00	-0.22
PPP	<b>0.41</b>	0.10	-0.10	0.08	-0.28
SPP	<b>0.31</b>	0.25	-0.06	<b>0.35</b>	0.07
SPPL	<b>0.47</b>	0.19	-0.11	0.26	-0.17
TSW	0.08	<b>-0.43</b>	<b>-0.32</b>	0.17	<b>0.35</b>
SYPH	<b>0.29</b>	0.02	<b>-0.41</b>	-0.21	0.28
AB	0.09	<b>-0.31</b>	0.07	<b>0.40</b>	<b>0.45</b>
PM	0.06	0.17	<b>0.39</b>	<b>-0.39</b>	<b>0.47</b>
Frost	-0.05	0.01	<b>0.51</b>	<b>0.30</b>	-0.10
Eigenvalue	3.28	2.25	1.62	1.12	1.05
Proportion (%)	25.22	17.31	12.49	8.59	8.07
Cumulative %	25.22	42.53	55.02	63.61	71.67

PC: Principal component, Bold value in the table under each PC represent high vector loading



per pod and seed yield  $\text{ha}^{-1}$  had a relatively high cumulative contribution effect to the first PC, which explained about 25.22% of the total variation. This implies that these traits were responsible for the differentiation of genotypes into different clusters and had a greater contribution to the total diversity. Similarly, it was observed that the variance explained by PC1 was mainly due to traits like plant height and pods per plant, and days to 90% maturity and grain yield per hectare.

In PC2, the observed 17.31% of the variation was mainly contributed by stand count at harvest, thousand seed weight, lodging score, Ascocayta blight, and days to 90% maturity. The third component accounted for about 12.49% of the total variation. Frost score, seed yield per hectare, powdery mildew, lodging score, and thousand seed weight contributed more to the variation in PC3 than other traits. In PC4, Ascocayta blight, powdery mildew, days to 90% maturity, number of seeds per pod, days to 50% flowering, and frost score had relatively more contribution.

In conclusion, more than three traits with small contributions accounted for each principal component, and the total contribution of the PC to the total divergence observed among genotypes. Traits with the largest values closer to unity within the first principal component influence the clustering more than those with lower absolute values closer to zero. Thus, the differentiation of the genotypes into different clusters and greater contribution to the total variation was due to the cumulative effect of several traits rather than the smaller contribution of all traits or the large contribution of a few traits.

### 3.5. Genotype by Trait (GT) Bi-plot

The GT data is represented on a GT bi-plot using PC1 and PC2 (Figure 3). This plot can help visualize the relationships between different traits, the contribution of individual traits to the overall variation, the nature of genotypes in each quadrant, the trait profiles of different genotypes, and the divergence of genotypes. The bi-plot has several interpretations:

- The cosine of the angle between two trait vectors approximates the Pearson correlation between them. An angle smaller than  $90^\circ$  indicates a positive correlation, an angle greater than  $90^\circ$  indicates a negative correlation, and an angle of  $90^\circ$  indicates zero correlation.
- The angle between a genotype and a trait indicates the relative level of the genotype for that trait. An acute angle indicates that the genotype is above average for the trait, an obtuse angle indicates that the genotype is below average for the trait, and a right angle indicates that the genotype is average for the trait.
- The vector length (i.e., the distance from the bi-plot origin) of a trait indicates how well the trait is represented in the bi-plot. A short vector indicates that the variation of the trait across genotypes is either small or not well presented in the bi-plot, which is due to its weak or lack of correlation with other traits.

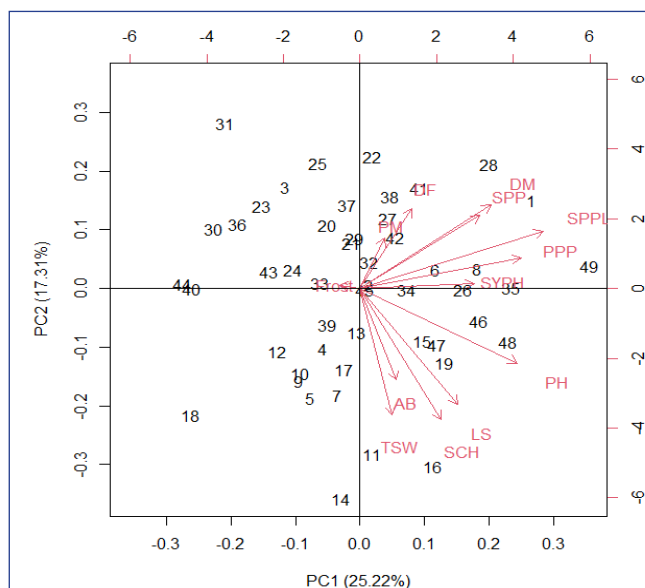


Figure 3: Genotype by trait (GT) biplot based on the original genotype by trait data showing the contribution and association of 13 traits (red line arrows) and distribution of field pea genotypes (light-black 1-49) under PC1 and PC2

• The distance of genotypes from the bi-plot origin and the way of genotype positioning (overlapping, near/far from each other). Based on these principles, the following observations can be made from Figure 3. Seed yield was positively correlated with a number of other traits, including the number of seeds per plant, pods per plant, and seeds per pod, plant height, and days to 90% maturity. The number of seeds per plant was also positively correlated with days to 90% maturity, plant height, pods per plant, and seeds per pod. Plant height was positively correlated with days to 90% maturity, pod per plant, and seeds per pod, stand count at harvest, lodging score, and thousand seed weight.

This indicates that genotypes taking longer to mature and with a high number of stands have taller plant height. Taller genotypes also tend to have less lodging resistance. Frost's score was not strongly correlated with any traits, as suggested by its short vector. These statements can be verified from the Pearson correlation table for the association of most of the traits (Table 10). The first and the second PC bi-plots explained 42.53 % of the total variability among the genotypes, displaying that number of seeds per plant, number of pods per plant, plant height, days to 90% maturity, number of seeds per pod and seed yield per hectare were considered the most discriminating traits. The genotypes positioned on the right top quadrant were characterized by late maturity, high performance for seeds per plant, pods per plant, and seeds per pod and seed yield per hectare. The genotypes depicted in the right bottom quadrant had tallest plant height, high stand count at harvest, larger lodging score (larger lodging score indicates more lodging and less lodging resistance) and large seed size.

Table: 10 Pearson correlation between Traits in 49 Field pea genotypes over Location

Variables	DF	DM	PH	SCH	LS	PPP	SPP	SPPL	TSW	SYPH	AB	PM	Frost
DF	<b>1</b>	<b>0.396</b>	-0.021	-0.111	-0.002	0.133	0.243	0.214	-0.099	-0.079	0.037	0.139	0.165
DM		<b>1</b>	<b>0.375</b>	0.030	0.077	<b>0.361</b>	<b>0.306</b>	<b>0.418</b>	-0.183	<b>0.356</b>	-0.185	<b>0.306</b>	-0.097
PH			<b>1</b>	<b>0.444</b>	<b>0.709</b>	<b>0.386</b>	0.200	<b>0.397</b>	0.167	<b>0.301</b>	0.142	0.013	-0.044
SCH				<b>1</b>	<b>0.505</b>	0.165	-0.095	0.084	<b>0.403</b>	0.132	0.230	0.057	0.037
LS					<b>1</b>	0.169	0.040	0.156	0.207	-0.120	0.262	0.025	0.179
PPP						<b>1</b>	0.218	<b>0.848</b>	-0.081	<b>0.339</b>	0.083	-0.022	-0.108
SPP							<b>1</b>	<b>0.695</b>	-0.001	0.254	-0.023	0.075	-0.002
SPPL								<b>1</b>	-0.035	<b>0.388</b>	0.065	0.019	-0.073
TSW									<b>1</b>	<b>0.301</b>	<b>0.304</b>	<b>-0.291</b>	-0.203
SYPH										<b>1</b>	0.044	0.002	-0.200
AB											<b>1</b>	0.062	0.020
PM												<b>1</b>	0.115
Frost													<b>1</b>

The bi-plot of the first two principal components revealed that the studied genotypes were scattered in all the quadrants (Figure 3) even though clear-cut grouping of these genotypes not occurred, which showed the high level of genetic diversity in the evaluated genotypes.

The genotypes distributed around the origin and overlap to each other had similar genetic characteristics, while the genotypes that were found far from the origin are considered unrelated (genetically distinct) (Figure 3).

In relation to plant type, it was observed that, prostrate type field pea genotypes were found far from the biplot origin and sparsely distributed. It implies that prostrate type field pea genotypes have considerable diversity than erect type field pea. Advanced field pea genotypes were also positioned apart from the origin and scattered. Therefore, the divergent genotypes from prostrate type (G-49,G-18,G-48,G-14,G-16 and G-11), erect type (G-44,G-40,G-31,G-28,G-22 and G-25), advanced line (G-14,G-16,G-18,G-31,G-44 and G-40) and released variety (G-49 and G-48) could be used as potential parents for successful hybridization to develop heterotic groups in the field pea breeding program.

In overall, field pea genotypes show considerable diversity in studied traits, with differences between plant types. Prostrate type and advanced line genotypes have wider diversity compared to erect types and release varieties, respectively. This variation can be utilized in breeding programs to develop high-yielding varieties with desirable traits. Crossbreeding promising parents, especially selected from prostrate and erect types, can result in a good level of genetic recombination. Advanced lines such as G-18 and G-26, G-3, and G-26, and prostrate types such as G-18 and G-48 are good examples of promising parents for crossbreeding. This could generate desirable segregates with a broad genetic base.

#### 4. Conclusion

The present study showed the Presence of considerable diversity for the studied traits in field pea genotypes, with differences between plants types. The study has also shown that grouping of genotypes from different plant types and sources of origin under the same cluster indicates that differences in these factors do not necessarily imply differences in genetic makeup. This finding suggests that field pea improvement programs should focus not only on genetic diversity between different plant types but also within them

#### 5. References

- Anonymous, 2020. Agricultural sample survey, Report on, area and production for major crops (private peasant holdings, meher season). Volume I. Addis Ababa, Ethiopia, 13. Available from: HA37.11 .A742 2012/2013-2013/2014.
- Anonymous, 2019. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL <https://www.R-Project.Org/>. Available from: <https://www.gnu.org/copyleft/gpl.htm>.
- Anonymous, 2002. SAS Institute Inc. Statistical Analysis Software Version 9.0. Cary, NC: SAS Institute Inc., USA.
- Assen, K.Y., 2020. Diversity analysis and identification of promising powdery mildew resistance genotypes in field pea (*Pisum sativum* L). American Journal of Biological and Environmental Statistics 6(1), 7–16. <https://doi.org/10.11648/j.ajbes.20200601.12>.
- Bernier, C.C., Hanounik, S.B., Hussein, M.M., Mohamed, H.A., 1993. Field manual of common Faba bean diseases in the Nile Valley. In: Inf. Bull. No. 3. International Centre for Agricultural Research in the Dry Areas (ICARDA).



- Bhuvaneswari, S., Sharma, S.K., Punitha, P., Shashidhar, K.S., Naveenkumar, K.L., Prakash, N., 2016. Evaluation of morphological diversity of field pea [*Pisum sativum* subsp. *arvense* (L.)] germplasm under sub-tropical climate of Manipur. Legume Research – An International Journal 40(2), 215–223. <https://doi.org/10.18805/Ir.v01of.10756>.
- Endres, G., Kandel, H., 2021. Field pea production. NDSU Carrington Research Extension Center, P.3–4
- Faiza, A., Neelam, A., Syed, M.A.S., 2021. Genetic diversity among pea (*Pisum sativum* L.) genotypes for maturity and yield traits. Sarhad Journal of Agriculture 37(2), 386–397. DOI | <https://dx.doi.org/10.17582/journal.sja/2021/37.2.386.397>.
- Fikreselassie, M., 2012. Variability, heritability and association of some morpho-agronomic traits in field pea (*Pisum sativum* L.) genotypes. Pakistan Journal of Biological Science 15(8), 358–366.
- Green, R.F., Sneath, P.H.A., Sokal, R.R., 1974. Numerical taxonomy. Biometrics 30(2), 372–373. <https://doi.org/10.2307/2529664>.
- Kedir, Y, Deresa, T., Gizachew, Y., Temesgen, A., 2024. Genetic variability and characters association for lodging, yield and related traits of field pea (*Pisum sativum* L.) genotypes in contrasting plant types. Elsevier (Ecological genetics andgenomics) 33. <https://doi.org/10.1016/j.egg.2024.100289>.
- Keneni, G., Jarso, M., Watira, T.W., Adem, G.D., 2005. Extent and pattern of genetic diversity for morpho-agronomic traits in Ethiopian highland pulse landraces: I. Field Pea (*Pisum sativum* L.). Genetic Resources and Crop Evolution 52(5), 539–549. DOI 10.1007/s10722-003-6016-6.
- Maharjan, A., Bhatta, B., Acharya, R.P., Sagar, G.C., Shrestha, S., 2015. Efficacy assessment of treatment methods against powdery mildew disease of pea (*Pisum sativum* L.) caused by *Erysiphe pisi* var. *pisi*. World Journal of Agricultural Research 3(6), 185–191.
- Negisho, K., Teshome, A., Keneni, G., 2017. Genetic diversity in Ethiopian field pea (*Pisum sativum* L.) germplasm collections as revealed by SSR markers. Ethiopia Journal Agricultural Science 27(3) 33–47.
- Seboka, H., Erena, M.F., 2013. Multivariate analysis of some Ethiopian field pea (*Pisum sativum* L.) genotypes. International Journal of Genetics and Molecular Biology 5(6), 78–87. DOI: 10.5897/IJGMB2013.0080.
- Singh, A.K., Srivastava, C.P., 2015. Effect of plant types on grain yield and lodging resistance in Pea (*Pisum sativum* L.). Indian Journal of Genetics 75(1), 69–74.
- Singh, S., Sharma, V.R., Nannuru, V.K.R., Singh, B., Kumar, M., 2021. Phenotypic diversity of pea genotypes (*Pisum sativum* L.) based on multivariate analysis. Legume Research– An International Journal 44(8), 875–881.
- Tegegn, A., Teshome, E., 2017. Grain yield and yield components of field pea (*Pisum sativum* L.): as influenced by ascochyta blight (*Mycosphaerella pinodes*) disease in the highlands of Bale, Oromia. American Scientific Research Journal for Engineering, Technology and Sciences (ASRJETS) 35(1), 15–24.
- Teshome, E., Tegegn, A., 2017. Comparative study of powdery mildew (*Erysiphe polygoni*) disease severity and its effect on yield and yield components of field pea (*Pisum sativum* L.) in the southeastern Oromia, Ethiopia. Journal of Plant Pathology and Microbiology 8, 410. doi: 10.4172/2157-7471.1000410.
- Tiwari, G., Lavanya, G.R., 2012. Genetic variability. In: character association and component analysis in F4 generation of field pea (*Pisum sativum* var. *arvense* L.). Karnataka Journal of Agricultural Science 25(2), 173–175.
- Tran, C.T., Besieger, T.M., Becker, H., Horneburg, B., 2023. Genetic diversity of pea (*Pisum sativum* L.) genotypes differing in leaf type using SNP markers. Genet Resource and Crop Evolution 70(1), 1085–1095. <https://doi.org/10.1007/s10722-022-01487-3>.
- Wang, T.F., Gossen, B.D., Slinkard, A.E., 2006. Lodging increases severity and impact of *mycosphaerella* blight on field pea. Canadian Journal of Plant Science 86, 855–863
- Yimam, K., Yilma, G., Abo, T., Tesfaye, D., Achenef, G., 2024. Effect of plant types on lodging resistance and yield of field pea (*Pisum sativum* L.) and lodging impact on yield and ascochyta blight severity. International Journal of Bio-resource and Stress Management 15(9), 01–15. [HTTPS://DOI.ORG/10.23910/1.2024.5540](https://doi.org/10.23910/1.2024.5540).
- Yirga, H., Tsegay, D., 2013. Characterization of dekokko (*Pisum sativum* var. *abyssinicum*) accessions by qualitative traits in the highlands of Southern Tigray, Ethiopia. African Journal of Plant Science 7(10), 482–487.

