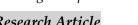


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# The Determination of Agronomic Characteristics of Pea Lines in F4 Generation

□Fatma Zumbul¹, □Nur Koç Koyun¹,\*, □Ahmet Tamkoç¹

<sup>1</sup>Selcuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

# **HIGHLIGHTS**

- Peas are vital for sustainable agriculture, offering high protein content, forage potential, and adaptability, addressing food and forage needs globally and in Türkiye.
- This study evaluated 24 F4 pea lines for seed yield and disease tolerance, aiming to identify superior genotypes for breeding programs.
- Eight genotypes, including G1 (10-1) and G12 (10-1 Ozel), exhibited high yields and disease tolerance, highlighting their potential for developing resilient pea varieties.

#### **Abstract**

Peas, the fourth most produced legume globally, suffer from yield losses due to diseases. Developing disease-tolerant varieties can contribute to sustainable agriculture by meeting protein needs through pea cultivation. For this reason, 24 pea lines, advanced to the F4 generation through hybridization and single-seed descent selection, along with 6 control varieties, were evaluated in 2016 under field conditions in Konya using an Augmented Experimental Design with three replications. Among the genotypes, the highest number of pods per plant (11.35 pods) was observed in the line coded as G1 (10-1). The line coded as G11 (12-1) demonstrated the highest seed yield (229.93 kg da<sup>-1</sup>). Additionally, plants exhibiting natural symptoms of viral infection and powdery mildew under field conditions were scored as 1 (diseased), while those without symptoms were scored as 3 (healthy). To evaluate the tolerance of genotypes under these biotic stress conditions, a path analysis was conducted. The path model revealed that plant height, pod width, and pod height were associated with the presence of viral and powdery mildew pathogens. Furthermore, a cluster analysis based on these five traits grouped the 24 lines and 6 control varieties into four clusters. In the first group, comprising susceptible genotypes, G11 (12-1) and G9 (4) emerged as the most tolerant lines. The second group included genotypes tolerant to both pathogens, with G10 (208) standing out. The third group consisted of genotypes tolerant to powdery mildew, with G1 (10-1), G2 (15), and G18 (10-1 Ozel S) being prominent. The fourth and final group, composed of the most tolerant genotypes, identified G12 (10-1 Ozel) as the most productive type. Based on the findings, we conclude that these eight genotypes, characterized by high tolerance to diseases and superior seed yield, hold potential as genetic resources for developing high-yielding, disease-resistant pea varieties.

Keywords: Augmented trial design; plant breeding; Pisum sativum ssp. Arvense; selection; yield

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\*Correspondence: nurkoc@selcuk.edu.tr

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### 1. Introduction

Pea cultivation today focuses on two main subspecies, *Pisum sativum* ssp. *sativum* (edible peas) and *Pisum sativum* ssp. *arvense* (forage peas), both globally and in Türkiye (Bilgili 2009). Peas, which have diverse uses such as grain, forage, and vegetable production, rank as the fourth most-produced legume worldwide, following soybean, peanut, and common bean (Heuze et al. 2015). According to TUIK data for 2014, forage pea cultivation in Türkiye covered approximately 3,740 hectares, yielding 70,422 tons of production (TUIK 2024). While edible pea cultivation historically covered a larger area compared to forage peas, in recent years, the increasing emphasis on addressing the forage deficit has led to significant growth in the cultivation area and production of forage peas. According to TUIK statistics, forage peas were cultivated over 23,840 hectares in 2023, producing 457,281 tons of green fodder (TUIK 2024). Globally, the average yield of grain peas is approximately 2,000 kg ha<sup>-1</sup>, whereas forage pea yields are generally higher but highly variable, depending on environmental conditions, ranging from 4.9 tons DM ha<sup>-1</sup> to as much as 35.9 tons DM ha<sup>-1</sup> (FAO 2011; Bilgili et al. 2010; FAOSTAT 2024).

Peas exhibit high adaptability, enabling cultivation across a wide range of regions in Türkiye, where their planting area has increased over the years. This growth can be attributed to the plant's high crude protein and mineral content, palatability, suitability for early spring grazing, and the nutritional value of pea silage, which is comparable to that of alfalfa silage. Additionally, forage peas play a significant role in agricultural production due to their contributions to soil organic matter preservation and their use as green manure (Bilgili 2009; Heuze et al. 2015). Biotic and abiotic stress factors, particularly diseases, are major contributors to yield losses in pea cultivation, with powdery mildew being among the most critical factors, causing yield reductions of 25% to 50% (Kora and Teshome 2016; Al-Rubaye 2023). Therefore, breeding efforts have increasingly focused on developing disease-tolerant pea varieties. These efforts are expected to make significant contributions to meeting Turkey's protein needs, both by using forage peas for animal protein production and edible peas for plant-based protein consumption (Endes and Tamkoç 2006).

Scientific research in pea breeding has achieved significant progress in developing new varieties with higher nutritional value, improved environmental adaptability, and enhanced stability. Despite these substantial advancements, the ever-changing cultivation conditions and the lack of varieties adapted to these conditions highlight the ongoing need and opportunities for further improvements and the development of new pea varieties. In genetic selection studies, various criteria and analyses, such as correlation coefficients, multiple regression, and path analysis, are employed to examine causal relationships within interconnected systems of traits. Path analysis stands out as an effective tool for evaluating interdependencies between yield and its components, offering more reliable results compared to other methods (Georgieva et al. 2015). However, Kingsolver and Schemske (1991) utilized path models to address topics such as pollination biology, phenotypic integration, and selection on morphometric and ontogenetic traits.

In this study, the morphological traits of pea lines/varieties advanced to the F4 generation through hybridization and single-seed descent selection were examined using an Augmented experimental design, as the available seed quantities were insufficient. Additionally, the study aimed to identify and select genotypes tolerant to naturally occurring viral and powdery mildew symptoms under field conditions by employing a path model. The main hypothesis proposed is that there is significant genetic variation among pea genotypes grown under field conditions in terms of agronomic traits and disease tolerance (viral infection and powdery mildew tolerance). This study seeks to answer the question: Which pea genotypes exhibit both high yield and tolerance to powdery mildew, making them suitable candidate lines for future breeding programs?

### 2. Materials and Methods

## 2.1. Establishment of the Experiment

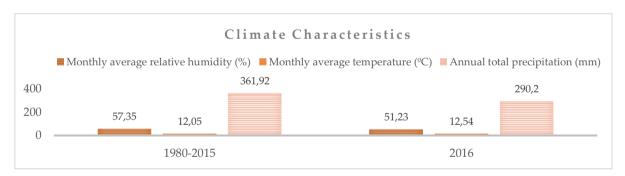
A total of 30 genotypes were used as the material for the study. Among these, 24 pea lines were advanced to the F4 generation through single-seed descent following hybridization. Additionally, three pure lines (Sultan-1, Sultan-2, 8th line) and three registered cultivars (Ozkaynak, Taskent, Furkan) were included as control varieties (Tables 1-2).

Check Variety Code	Name	
C1	Ozkaynak	
C2	Taskent	
C3	Furkan	
C4	Sultan-2	
C5	Sultan-1	
C6	8 <sup>th</sup> line	

Table 1. The codes for the control varieties used in the study

**Table 2**. The codes for the genotypes used in the study

Genotype Code	Name	Genotype Code	Name	Genotype Code	Name	Genotype Code	Name
G1	10-1	G7	1084222	G13	602-1	G19	10-2
G2	15	G8	ALRAQI	G14	12-2	G20	3
G3	17	G9	4	G15	8.38.Row	G21	2
G4	10-1(Ozel-S)	G10	208	G16	602-2	G22	5
G5	B6-Duz	G11	12-1	G17	B6-Renkli	G23	13
G6	16	G12	10-1 Ozel	G18	10-1 Ozel S	G24	7



**Figure 1.** Climate characteristics of the experimental field (Climate data were taken from the Konya Meteorology Regional Directorate)

The research was conducted under irrigated conditions in the Experimental Field of Field Crops at Selçuk University in 2016. According to the analysis of the soil at a depth of 30 cm, the soil was characterized as clay-loam with 2.40% organic matter, an alkaline reaction (pH 8.12), and no salinity problem (EC1:5 0.88 dS m<sup>-1</sup> NaCl). The soil exhibited high levels of lime (20%) and potassium (0.03%) but was deficient in  $P_2O_5$  (18.30 kg ha<sup>-1</sup>) and ferritin content (1.19 mg kg<sup>-1</sup>). The climatic characteristics of the experimental area are presented in Figure 1.

Due to the limited availability of experimental material, this study was designed using the Augmented Experimental Design with three replications, including six check (control) varieties in each replication. The number of replications was calculated according to Petersen (1994) (Equation 1).

$$r \ge (\frac{10}{c-1}) + 1,$$
 (1)

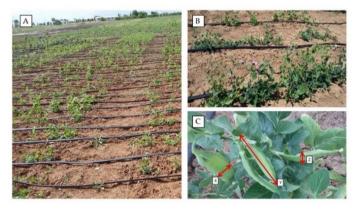
r: replication, c: number of Check

This study was sown on April 10, 2016, with block dimensions of 15 m in length and 2 m in width. The planting was conducted with 50 cm row spacing and 20 cm plant spacing within the rows. Along with sowing, 150 kg ha<sup>-1</sup> of diammonium phosphate (DAP) fertilizer was applied. Seedlings emerged between April 26 and April 29, 2016, and weed control was performed by hoeing when the plants reached the four-to-five leaf stage. The plants were irrigated using sprinkler irrigation for initial establishment, followed by three applications of drip irrigation during the flowering, pod formation, and grain filling stages (Figure 2A).

# 2.2. Data collection and Statistical Analysis

# 2.2.1. Determination of Differences in Morphological Traits Among Genotypes Using Augmented Experimental Design

All genotypes flowered between June 10 and June 14, 2016, and the plants began pod setting between June 12 and June 17, 2016. Observations and measurements were taken during this period (Figures 2B and 2C). This research was organized into two stages. The first phase involved the cultivation of lines according to the Augmented Experimental Design, as well as the collection and evaluation of observations and measurements. In this phase, the following traits were measured: plant height (PH) (cm), number of pods per plant (NPPP) (pods/plant), pod length (PL) (mm), pod width (PW) (mm), pod height (PoH) (mm), seeds per pod (SPP), thousand-seed weight (TSW) (g), and seed yield (SY). Measurements were taken at the harvest stage.



**Figure 2.** (A) The general view of the experiment (May 29, 2016), (B) observation of flowering (June 13, 2016), and (C) measurements taken from pods are presented, where X represents pod width, Y represents pod length, and Z represents pod height (June 17, 2016).

Plant height was measured in centimeters as the natural height of the plant. Pod-related measurements, including pod length, were recorded in millimeters from three pods per plant, as schematized in Figure 2C. The number of pods per plant was determined by counting all pods on each plant within a line and calculating the average reported as the number of pods. The number of seeds per pod was obtained by counting seeds in three pods and calculating the average (Al-Rubaye 2023).

The number of seeds per plant was calculated by multiplying the number of pods per plant by the average number of seeds per pod. Thousand-seed weight (TSW) was determined by weighing 50 seeds and multiplying the result by 20, with the final value reported in grams (Özköse 2012).

Additionally, in the field experiment, symptoms of viral damage and powdery mildew began to appear on the plants 30 days after emergence under natural conditions. Due to the differing responses of the cultivars/lines to these two pathogens, scoring was conducted based on observations: a score of 1 was assigned if virus (VP) or powdery mildew (PMP) damage was observed, and a score of 3 was assigned if the plant was healthy (Figure 3).

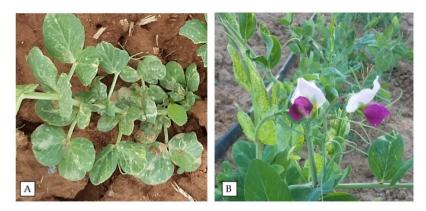


Figure 3. Powdery mildew observed on the plants (A; May 29, 2016) and the presence of the virus (B; June 13, 2016).

In this section, which constitutes the first phase of the study, the data obtained were subjected to statistical analyses using the JMP 7 software package based on the Augmented Experimental Design (Sall et al., 2017). For each trait found to be significant, a Student's *t*-test was performed (Petersen 1994).

# 2.2.2. Selection of Genotypes Tolerant to Naturally Occurring Virus and Powdery Mildew Pathogens Under Field Conditions

In this section of the study, path analysis was performed using multiple regression analysis (Forward MRA) in SPSS 15 software, with the purpose of determining the relationship between the presence of naturally occurring virus and powdery mildew pathogens under field conditions and the examined botanical traits (Ho, 2013). In the multiple regression analysis, the presence of virus and powdery mildew were set as the dependent variables, while botanical traits (plant height, number of pods, pod length, pod width, pod height, number of seeds per pod, thousand-seed weight, and seed yield) were defined as independent variables. To better explain the relationships between the traits, a path model was constructed based on the results of the multiple regression analysis (Forward MRA). In the model, the standardized beta coefficient ( $\beta$ ) obtained from the multiple regression analysis was used as the path coefficient, or direct effect (Anonymous 2021). For a specific residual variable (U), the path coefficient is calculated using the following formula (Equation 2) (Kingsolver and Schemske 1991). Additionally, the correlation between the traits used in the path model was determined using Pearson correlation analysis.

$$U=\sqrt{(1-R^2)},$$

In this section of the study, cluster analysis was performed using the JMP 7 software package based on the results of path analysis, with virus and powdery mildew presence and the associated traits (Sall et al. 2017). Using the SRplot online platform, principal coordinate analysis was performed to identify 30 genotypes in the F4 generation that are both high yielding and tolerant to virus and powdery mildew (Tang et al. 2023).

### 3. Results

### 3.1. Determining the Differences in Morphological Traits Among Genotypes

Analysis of variance was performed based on the Augmented Experimental Design provided in Table 3. For the check, the number of pods per plant, pod length, and pod width were found to be statistically significant at the 5% level, while pod height, seed yield, thousand-seed weight, and virus presence were found to be statistically significant at the 1% level. For the genotypes, only the number of pods per plant, pod height, and virus presence were statistically significant at the 5% level, while the other traits were determined to be non-significant.

Table 3. The table of variation analysis's summary (F value)

Source of Variation	Degree of Freedom	РН	NPPP	SPP	SY	TSW
Block	2	3,754	3,958	0,574	0,862	0,786
Check	6	1,196	5,266*	0,265	6,583**	32,083**
Genotype [Check]	23	0,927	3,555*	1,686	1,954	2,419
Error	10					
Total	41					
CV%		21,83	17,87	15,75	40,49	14,25
Source of Variation	Degree of Freedom	PL	PW	РоН	VP	PMP
Block	2	2,083	1,061	1,893	1,000	0,294
Check	6	3,750*	4,926*	18,125**	12,339**	0,982
Genotype [Check]	23	1,130	2,436	3,444*	4,228*	0,805
Error	10					
Total	41					
CV%		11,90	11,73	7,46	22,50	60,85

\*P<0.05, \*\*P<0.01, PH: Plant Height, NPPP: Number of Pod per Plant, SPP: Seed per Pod, SY: Seed Yield, TSW: Thousand Seed Weight, PL: Pod Length, PW: Pod Width, PoH: Pod Height, VP: Virus Presence, PMP: Powdery Mildew Presence

Table 4. Mean values and standard deviations of the control varieties

Check variety	PH (cm)	NPPP	SPP	SY (kg da-1)	TSW(g)
C1	43,6±11,79	7,7±2,52	4,7±0,67	68,62±23,29	95,6±15,31
C2	44,0±8,19	10,3±1,80	4,4±0,79	87,28±40,02	93,4±19,92
C3	39,3±3,51	7,7±0,88	4,6±0,69	65,78±13,04	92,2±11,50
C4	36,7±5,84	8,9±0,69	4,8±0,19	211,71±33,60	243,3±15,19
C5	46,3±13,07	7,7±1,76	4,4±0,98	137,53±67,17	190,9±24,92
C6	38,1±13,77	8,2±1,17	5,0±0,67	106,86±22,88	129,9±17,88
Mean	41,3±6,62	8,4±1,06	4,6±0,23	112,97±14,41	140,9±6,62
Check variety	PL(mm)	PW(mm)	PoH(mm)	VP(Scor.)	PMP(Scor.)
C1	43,4±2,40	7,3±0,43	4,0±0,22	1,0±0,00	1,0±0,00
C2	43,0±0,49	6,8±0,29	4,1±0,48	1,0±0,00	1,0±0,00
C3	43,2±1,97	7,5±0,14	4,3±0,26	1,0±0,00	1,0±0,00
C4	59,3±4,33	10,3±1,38	6,4±0,36	3,0±0,00	2,3±1,15
C5	54,5±12,19	9,5±0,47	5,5±0,69	2,3±1,15	1,7±1,15
C6	54,4±8,45	8,8±1,91	5,9±0,17	3,0±0,00	1,7±1,15
Mean	49,6±3,49	8,4±0,42	5,0±0,21	1,9±0,19	1,4±0,19

PH: Plant Height, NPPP: Number of Pod per Plant, SPP: Seed per Pod, SY: Seed Yield, TSW: Thousand Seed Weight, PL: Pod Length, PW: Pod Width, PoH: Pod Height, VP: Virus Presence (1: diseased, 3: healthy), PMP: Powdery Mildew Presence (1: diseased, 3: healthy)

When examining the mean values of the control varieties presented in Table 4, it can be stated that the Sult-1 variety with code C5, having a natural plant height of 46.3 cm, is the tallest genotype among the controls.

In terms of the number of pods per plant (10.3 pods), the highest yield was obtained from the Taskent variety with code C2. When examining the number of seeds per pod, the highest value of 5 seeds was recorded for the 8<sup>th</sup> line with code C6. In our study, the Sult-2 variety with code C4 stands out in terms of pod length (59.3 mm), pod width (10.3 mm), pod height (6.4 mm), seed yield (211.71 kg da<sup>-1</sup>), and thousand-seed weight (243 g). Additionally, this variety can be considered the healthiest genotype in terms of virus and powdery mildew presence compared to the other controls. Furthermore, the 8<sup>th</sup> line with code C6 was also determined to be the healthiest control variety in terms of virus presence.

The plant characteristics of the 24 pea lines advanced to the F4 generation are presented in Table 5. The highest natural plant height, 50.15 cm, was recorded for the G22 code, from the 5<sup>th</sup> line. The highest number of pods per plant, 11.35 pods, was obtained from the G1 code, 10-1 line. The highest number of seeds per pod, 6.41 seeds, was determined for the 10-1(OZEL-S) line. In our study, the G13 code, 602-1 line, stood out for pod length (64.87 mm), the G16 code, 602-2 line, for pod width (11.91 mm), and the G6 code, 16 line, for pod height (6.39 mm).

When examining the yield components, thousand-seed weight and seed yield, the G10 code, 208 line, had the highest thousand-seed weight (192.56 g), and the G1 code, 10-1 line, had the highest seed yield (229.93 kg da<sup>-1</sup>). The seed yield of the genotypes used in this study ranged from 15 kg da<sup>-1</sup> to 229 kg da<sup>-1</sup>, indicating a large variation among the genotypes.

Among the pea lines advanced to the F4 generation, the G7 (1084222) and G8 (ALRAQI) genotypes, which exhibited the least presence of the virus pathogen under field conditions (score 3.11), can be considered the healthiest lines. Additionally, these genotypes were recorded as the healthiest in terms of powdery mildew presence, with a score of 3.22. Furthermore, the G1 (10-1) and G2 (15) genotypes were also among the healthiest in terms of powdery mildew presence.

# 3.2. Selection of Genotypes Tolerant to Virus and Powdery Mildew Pathogens

The results of the Multiple Regression analysis conducted to identify genotypes that continue to grow and develop under natural field conditions with virus and powdery mildew pathogens are presented in Table 6. Based on the information provided in Table 5, the resulting path model is shown in Figure 5.

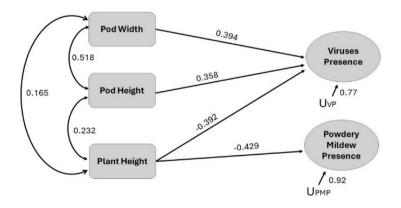


Figure 5. Path model

In the path model presented in Figure 5, the direct effects ( $\beta$ ) of plant traits on the presence of virus and powdery mildew are represented by unidirectional arrows, while correlations between traits are depicted with bidirectional arrows. Plant height has a negative effect on virus presence ( $\beta$  = -0.392), whereas other traits exhibit positive effects. Additionally, pod width has the highest direct effect on virus presence, accounting for 23.9% of the variation in virus presence alone. Similarly, plant height negatively affects powdery mildew presence ( $\beta$  = -0.429) and explains 15.5% ( $R^2$  = 0.155) of the variation in powdery mildew presence (Table 5).

Table 5. Adjusted means and LSD values of the examined traits according to the augmented experimental design

Cada	PH	NIDDD	SPP	SY	TSW	PL	PW	РоН	VP	PMP
Code	(cm)	NPPP	SPP	(kg da-1)	(g)	(mm)	(mm)	(mm)	(Scor.)	(Scor.)
G1	29,43	11,35	5,74	229,93	175,08	55,44	8,29	6,05	1,11	3,22
G2	37,76	9,68	4,74	107,22	113,75	53,01	8,60	6,24	1,11	3,22
G3	37,60	8,18	4,74	85,77	108,41	46,33	8,56	4,06	1,11	1,22
G4	31,43	4,35	6,41	53,39	106,41	54,22	6,15	4,74	1,11	1,22
G5	33,76	8,01	4,41	71,60	102,08	41,93	6,48	4,29	1,11	1,22
G6	48,43	5,01	5,74	84,85	155,75	59,55	9,45	6,39	1,11	1,22
G7	28,10	5,68	3,24	35,27	85,91	48,94	9,33	5,02	3,11	3,22
G8	18,10	4,68	2,74	28,68	106,41	44,80	8,59	4,15	3,11	3,22
G9	37,42	7,11	5,44	101,24	127,73	51,38	8,19	5,45	0,78	0,89
G10	42,25	9,78	5,10	189,89	192,56	52,87	10,89	5,71	0,78	2,89
G11	46,08	11,11	5,77	160,45	118,40	48,26	7,78	4,94	0,78	0,89
G12	21,42	4,78	5,44	71,14	129,06	47,22	7,13	4,70	2,78	2,89
G13	46,75	3,78	5,44	41,24	96,40	64,87	11,77	5,27	0,78	2,89
G14	41,75	7,28	5,10	85,64	114,06	44,21	9,24	5,31	2,78	2,89
G15	23,08	3,78	3,44	15,44	86,09	53,50	11,19	5,54	0,78	2,89
G16	38,42	1,78	3,44	15,42	140,06	45,76	11,91	5,13	2,78	2,89
G17	33,65	5,54	3,66	45,84	98,77	36,08	6,29	4,33	1,11	0,89
G18	36,82	9,54	4,82	134,91	151,52	54,88	7,29	4,93	1,11	2,89
G19	32,15	5,21	4,82	53,65	114,52	56,71	9,47	5,13	1,11	2,89
G20	34,15	4,04	3,16	25,59	67,52	45,85	8,64	3,66	1,11	0,89
G21	35,65	6,04	5,16	64,23	99,02	49,09	7,38	5,45	1,11	0,89
G22	50,15	6,21	4,16	77,95	139,19	54,99	10,43	6,06	1,11	2,89
G23	40,15	7,21	4,49	71,36	110,52	44,60	7,25	4,73	1,11	2,89
G24	40,82	5,54	4,49	61,39	112,74	43,86	7,19	4,74	1,11	2,89
SE	8,817	1,377	0,771	40,089	19,27	6,246	1,053	0,397	0,497	0,92
$LSD_1$	-	2,376	-		-	-	-	0,686	1,220	-
$LSD_2$	-	3,497	-		-	-	-	1,010	1,796	-
$LSD_3$	-	4,115	-		-	-	-	1,188	2,113	-
LSD <sub>4</sub>	-	4,445	-		-	-	-	1,283	2,282	-
1st BCT	5,903	-0,681	0,259	-2,521	2,588	1,930	0,467	0,129	-0,222	-0,111
2 <sup>nd</sup> BCT	1,250	1,222	-0,102	15,509	4,936	2,101	-0,349	0,115	0,111	0,222
3rd BCT	-7,153	-0,542	-0,157	-12,988	-7,523	-4,031	-0,118	-0,244	0,111	-0,111

BCT: Block Correction Term, SE: Standard Error, LSD1: Comparison among the controls, LSD2: Comparison between the controls and genotypes, LSD3: Comparison among genotypes within the same block, LSD4: Comparison among genotypes across different blocks, Scoring 1: disease, 3: healthy

Based on the path model results, the standout plant traits—plant height, pod width, and pod height—were identified as the key features to be used in the selection process of our study concerning virus and powdery mildew presence. The clustering analysis conducted using these traits is presented in Figure 6. As a result of the clustering analysis, 24 pea lines and 6 control varieties were divided into four groups.

**Table 6.** Multiple regression analysis of morphological traits in pea in relation to virus and powdery mildew presence (Forward MRA)

		Coef	Model Summary			
Dependent variable	Predictors	Standardized Beta Coefficients (β)	t	P	R	$R^2$
Viruses Presence	Pod Width	0,394	2,351	0,027*	0,515(a)	0,239
	Plant Height	-0,392	-2,662	0,013*	0,614(b)	0,330
	Pod Height	0,358	2,110	0,045*	0,684(c)	0,406
Powdery Mildew Presence	Plant Height	-0,429	-2,512	0,018*	0,429(a)	0,155

### Viruses Presence

- a. Predictors: (Constant), Pod Width
- b. Predictors: (Constant), Pod Width, Plant Height
- c. Predictors: (Constant), Pod Width, Plant Height, Pod Height

### Powdery Mildew Presence

a. Predictors: (Constant), Plant Height

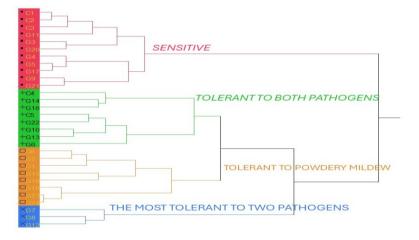


Figure 6. Results of the cluster analysis

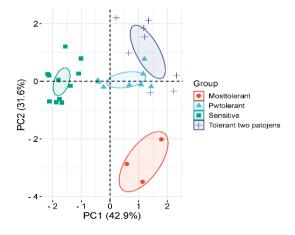


Figure 7. Results of the PCoA analysis using SRplot online platform

The first group consists of sensitive genotypes, including C1, C2, C3, G3, G4, G5, G9, G11, G17, G20, and G21. The second group comprises genotypes tolerant to both pathogens (Tolerant to Two Pathogens), including C4, C5, G6, G10, G13, G14, G16, and G22. The third group includes genotypes tolerant only to powdery mildew (PM-tolerant), comprising eight genotypes: C6, G1, G2, G15, G18, G19, G23, and G24. The fourth and final group, identified as the most tolerant (Most Tolerant), consists of three genotypes: G7, G8, and G12. The clusters obtained from the Principal Coordinate Analysis (PCoA) corroborate the results of the Cluster Analysis, indicating that the fourth group, the most tolerant, forms a distinctly separate cluster from the other three groups (Figure 7).

### 4. Discussion

In genotype selection for plant breeding, lines that are less affected or unaffected by disease pathogens are significant (Al-Rubaye, 2023). In studies related to disease pathogen presence, Al-Rubaye (2023) found that 164 out of 242 lines were unaffected by any disease, and in their scale evaluation, where 1 = diseased and 5 = healthy, the average value was determined to be 3.69.

Singh and Srivastava (2015), in their study on peas with different leaf morphologies, reported that plant height ranged from 110 cm to 140 cm, and thousand-seed weight ranged from 24 g to 38 g. The semi-leafless plant type (LeLe AfAf TITI StSt) exhibited the highest thousand-seed weight compared to normal plant types. On the other hand, the "pleiofila" type with reduced stipule leaves (afaf tltl stst) had the smallest thousand-seed weight. They also found that seed yield per plant was highest in semi-leafless plant types in both tall and dwarf groups. Furthermore, they stated that the most suitable plant height for effectively suppressing weeds and preventing lodging was between 60 cm and 100 cm (Olle, 2015; Olle, 2017).

Avci and Ceyhan (2006), using the Line x Tester method to determine general and specific combining abilities in peas, investigated the pod characteristics of 12 hybrids created from 4 lines and 3 testers. In their study, they found that the number of pods per plant ranged from 45 to 91, the number of seeds per pod ranged from 4 to 6, pod length ranged from 51 mm to 84 mm, pod width ranged from 8 mm to 12 mm, and thousand-seed weight ranged from 110 g to 190 g. If we calculate the seed yield per plant by multiplying the number of pods per plant and the number of seeds per pod, the range was between 200 and 360 seeds per plant. Furthermore, the path coefficient analysis conducted in the study showed that the highest direct effect was recorded for the number of pods per plant.

Ouafi et al. (2016) cultivated pea genotypes obtained from ICARDA in Algeria from 2013 to 2016 and studied the agro-morphological characteristics of the plants based on UPOV criteria. In their study, they reported that the average pod length of the plants was 5 cm, and the pod width was 0.5 cm.

Özdemir and Tamkoç (2019) conducted a study in Konya under irrigated conditions during the 2017 summer growing season to determine which lines could be used as second crop cultivars. The study involved 22 different F6 generation pure forage pea lines and 4 registered forage varieties (Ozkaynak, Taskent, Furkan, Bilgehan). According to the results, plant height ranged from 43.3 to 105.0 cm, and the study concluded that the lines 4/3 and 5/1 could be grown as second crops under irrigated conditions in the Konya region.

Kadıoğlu et al. (2020) examined some morphological characteristics of 18 pea genotypes under Erzurum conditions in 2011, 2012, and 2013. The study reported that plant height varied between genotypes, ranging from 76 cm to 110 cm, with the number of pods per plant ranging from 5 to 10, and the thousand seed weight varying between 159 g and 303 g. In this study, as well as in our research, the Ozkaynak and Taskent varieties were used, with plant heights recorded as 90 cm and 93 cm, respectively, the number of pods per plant as 10, and the thousand seed weight as 199 g and 198 g.

Al-Rubaye (2023) studied the winter hardiness of 242 F<sub>2</sub> generation lines obtained from the hybridization of wild pea genotypes collected from nature with improved varieties and lines. As a result of the study, winter damage was not observed in 174 lines, with plant height varying between 35 and 150 cm, the number of pods per plant ranging from 4 to 81, and the number of seeds per pod varying between 2 and 9.3. The differences

between the findings of this study and those of other researchers may primarily be attributed to ecological variations and differences in the varieties or lines used.

Selvi et al. (2016) conducted a path coefficient analysis using morphological data from 24 pea varieties. Their study revealed that plant height during vegetative and flowering stages, the number of days to 50% flowering, pod circumference, single pod weight, single seed weight, leaf area, the number of pods per plant, and shelling percentage had a positive direct effect on pod yield.

Powdery mildew in peas leads to significant yield losses by reducing total biomass, the number of pods per plant, seeds per pod, plant height, and the number of nodes (Fondevilla and Rubiales 2012). In our study, a negative relationship was observed between plant height and the appearance of virus and powdery mildew symptoms. This could be attributed to the increased surface area of taller plants, which provides more space for pathogen adherence. However, Singh et al. (2015) reported that dwarf varieties were more susceptible to powdery mildew compared to medium or tall varieties. Contrary to this, Hammarlund (1925), in the earliest studies on the genetics and physiology of powdery mildew resistance in peas, identified a single resistant plant in the dwarf William Hurst variety, which was exposed to various races of the powdery mildew fungus (Sharma, 2015). The findings indicated that resistance to powdery mildew is governed by a recessive gene, *er-1* (Timmerman et al. 1994; Vaid and Tyagi 1997; Janila and Sharma 2004; Katoch et al. 2010). Different studies have localized genes conferring resistance to powdery mildew on LG I, LG III, LG V, or LG VI. Additionally, a clear genetic linkage between resistance to powdery mildew (designated as *er-1*) and the morphological marker "Gritty" (Gty) has been demonstrated within linkage group 6 (Timmerman et al. 1994). Therefore, the relationships observed among these traits in the F4 generation hybrids examined in our study could be influenced by linkage effects.

# 5. Conclusions

Due to the insufficient quantity of seeds obtained from the 24 pea lines advanced to the F4 generation through hybridization and single plant selection, this study was established and evaluated using an Augmented experimental design. Among the genotypes, G1 (10-1) and G11 (12-1) emerged as the most productive in terms of seed count per plant. However, based on path analysis conducted to determine the tolerance of genotypes to naturally occurring virus and powdery mildew symptoms under field conditions, it was observed that plant height, pod width, and pod height were significantly associated with the presence of these pathogens. Using these five traits, a cluster analysis categorized the 24 lines and six control varieties into four distinct groups. In the first group (sensitive genotypes), G11 (12-1) and G9 (4) stood out as the most tolerant genotypes. In the second group, tolerant to both pathogens, G10 (208) was identified as a promising genotype. In the third group, characterized as productive and tolerant to powdery mildew, G1 (10-1), G2 (15), and G18 (10-1 Ozel S) were notable genotypes. The fourth and final group, comprising the most tolerant genotypes, identified G12 (10-1 Ozel) as the most productive type. Based on the findings of our study, these eight genotypes demonstrate strong potential as genetic resources for the development of cultivars with high disease tolerance and grain yield.

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