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THE EFFECTS OF PHOSPHORUS ON NUTRITIONAL CHARACTERISTICS OF FABA BEAN (*Vicia faba* L.)

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ABSTRACT

This study determines the effects of phosphorus, applied in various doses (0-30-60-90-120 kg ha^{-1}), on the seed yield, protein and amino acid content of the faba bean (Salkim, Filiz and Kitik). The study was conducted in the 2022 and 2023 growing seasons in the experimental field of the Faculty of Agriculture of Aydin Adnan Menderes University. The effect of different interactions of cultivar and phosphorus dose on the seed yield and on the levels of the amino acids aspartic acid, serine, alanine, arginine, threosine and histidine was found to be significant, as was the effect of the phosphorus dose on the protein ratio. The optimum values for the seed yield (2.14t ha^{-1}) and protein (26.4%) were obtained when the phosphorus was applied at 60kg ha^{-1}. Among the cultivars, Kitik achieved a higher yield and protein ratio than the others. Arginine (1.056g/100g) and aspartic acid (1.125g/100g) were the amino acids found in the greatest quantities in the faba beans. However, while methionine (0.087g/100g) and cysteine (0.085g/100 g) were the essential amino acids present in the smallest quantities, the application of phosphorus increased the levels of these amino acids. As a result, the changes brought about in the yield, protein content and amino acid content of the faba bean cultivars by applying various doses of phosphorus were determined, revealing ways in which the nutritional value of the seeds might be enhanced.

Key words: Amino acid, faba bean, phosphorus, protein

INTRODUCTION

The faba bean, is an important legume which is valuable for human nutrition on account of its high amino acid and protein content. The faba bean *(Vicia faba* L) is an important source of nutrition on account of its high protein content. The main faba bean producing countries and regions are Ethiopia, Egypt, China, Afghanistan, India, Northern Europe and North Africa (Rahate et al., 2021).

The amino acid composition and digestibility of the proteins in faba bean seeds are different from other legumes (Drulyte and Orlien, 2019). Nutritional elements such as high (26-33%) protein content, carbohydrates, B-group vitamins and minerals are increasing the importance of faba bean production (Crepon et al., 2010; Labba et al., 2021).

In addition to its nutritional value, the faba bean has the capacity to bind free nitrogen from the air to the soil at a rate of 130-160kg N/ha per year) through rhizobium bacteria (Hoffman et al., 2007). This improves soil fertility and makes atmospheric nitrogen available to plants. The availability of phosphorus is one of the most important factors affecting the nitrogen fixation capacities of legumes (Ho et al., 2005). If insufficient phosphorus is present, nitrogen fixation is restricted because early and healthy root formation is interrupted (Marschner, 1995; Rahman et al., 2008). In addition, the phosphorus required by nodules may not be supplied (Schulze and Drevon, 2005; Sulieman and Tran 2015; Legesse et al., 2016). Phosphorus plays a role in the storage and transfer of energy within the plant cell and in the easy and rapid development of roots (Mucchal et al., 1996). It is also an essential component of cell membranes and nucleic acids (Makoudi et al., 2018). Consequently, a low level of phosphorus in the soil constitutes a limiting factor for legumes. Faba beans may require higher levels of phosphorus than other crops (Haling et al., 2016).

The pH, lime and low organic matter content of soils in Turkey can severely limit the availability of phosphorus. The phosphorus content of 58% of the soils in Turkey has been found to be insufficient (Eyupoglu, 1999). One of the main problems limiting crop production in Mediterranean and West Asian countries, including Turkey, is the low availability of phosphorus in soils (Cooper et al., 1987; Matar et al., 1992).

Applying phosphorus can have a direct effect on seed yield. Phosphorus affects the formation and quality of the seeds in the plant, and when sufficient phosphorus is provided, large and healthy seeds are formed (Faucon et al., 2015). Phosphorus also plays an important role in the processes of flowering and fruit formation, so a deficiency of phosphorus can inhibit flowering and the formation of pods. Insufficient phosphorus during flowering can reduce the number of flowers and the success of pollination, which can in turn affect the formation of seeds. Likewise, a deficiency of phosphorus during the formation of the fruit can have a negative effect on their size and fullness (El Mazlouzi et al., 2020). Phosphorus also promotes root development in the pod, resulting in a healthy root structure and uptake of water and nutrient from the soil, and promotes nodule formation in the pod (Lazali and Drevon, 2021). Incorporating legumes into cropping systems with better phosphorus management may be promising for increasing legume productivity (Mitran et al., 2018).

Phosphorus is needed for the process of protein synthesis in the plant cell. While cellular structures called ribosomes synthesize proteins by assembling amino acids, this process requires energy, and phosphorus plays an important role in transporting this energy. Furthermore, phosphorus regulates cellular functions by controlling the activation of proteins through a process called phosphorylation in the structure of proteins (Raghothama, 2000; Liang et al., 2013; Malhotra et al., 2018).

Although it is one of the main limiting factors in faba bean cultivation, only limited studies have been carried out on the efficacy and optimum doses of phosphorus. The aim of this study was to determine the effects of applying phosphorus on the yield, protein content and amino acid composition of faba beans. The protein content and amino acid content and the relationships between them were also investigated.

MATERIALS AND METHODS

The study was carried out at the Department of Field Crops, Faculty of Agriculture, Adnan Menderes University, Aydin, Turkey (27° 51'E, 37°51'N, altitude 50m) during the faba bean growing seasons in the years 2021/22 and 2022/23.

Table 1. Soil characteristics of the experimental area.

Soil Texture			pH	Organic Matter	Phosphorus	Calcium	Sodium
Sand (%)	silt (%)	clay (%)		(%)	(ppm)	(ppm)	(ppm)
72	16.7	11.3	8	1.2	21	2978	101
Sandy loam			High	Low	High	Low	Low

As can be seen in Table 1, the experimental site has sandy loamy soil with a high pH value. The characteristics of the soil were determined according to the Olsen method (Olsen et al., 1956). By this method, the phosphorus content of the soil of the experimental area was found to be high. However, there are many methods for soil analysis and phosphorus content may be found high by one method and low by another method (Yurtsever and Alkan, 1975; 1976). Moreover, critical phosphorus levels (according to the Olsen method) may vary depending on the plants in question (Dodd and Mallarino, 2005). For example, the critical value has been found to be 24% for soybean and maize but lower for wheat (Mallarino et al., 2013).

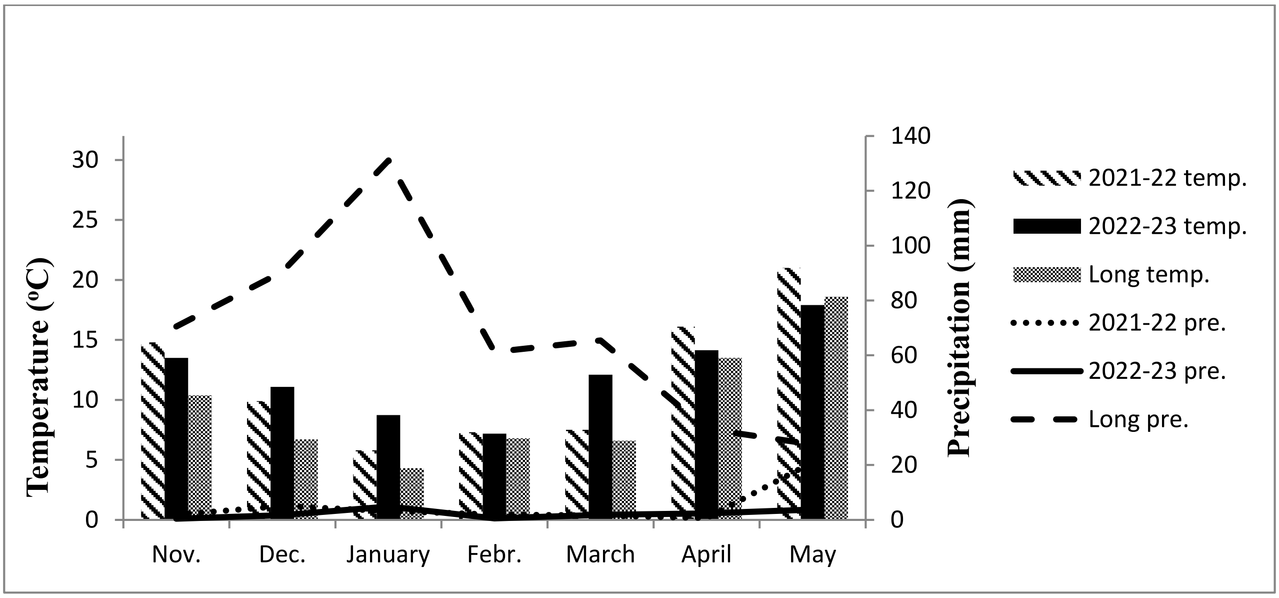

Figure 1. Meteorological data for the trial years (Source: Aydin Provincial Directorate of Meteorology).

Figure 1 shows the average temperatures and total amounts of precipitation for the trial years and over the long term. It can be seen that the amounts of precipitation in the trial years were much lower than the long-term average. In the second year of the experiment, however, April and May witnessed higher rainfall and lower average temperatures than the preceding year. This is the period when the pod matures and the seeds fill out. Due to this difference in the climatic conditions, the differences between the data obtained from the experiment for the two years proved to be statistically significant.

Experiment was conducted in Randomized Complete Block Design (RCBD) arranged in split-plots with three replications. The five different doses of phosphorus (0-30-60-90-120 kg ha^{-1}) were arranged at random in the main plots, the sub- plots were assigned random by three cultivars of faba beans (Salkim, Kitik, Filiz). Each plot was 5m long and consisted of six rows with intervals of 45cm between the rows. The total plot area was 13.5 m^2. A distance of 50 cm was left between the areas to which different doses of phosphorus were to be applied.

The phosphorus doses were applied to the soil in the form of triple superphosphate before planting. In addition, base fertilization was applied to all plots including the control sub-plots. Before sowing, potassium nitrate (13-0-46) fertilizer was used as base fertilizer to pure nitrogen and potassium. Planting was done on 02.12.2021 in the first year and on 10.12.2022 in the second year. The first year is indicated as 2022 and the second year as 2023 in the tables. After sowing, urea was applied as nitrogen fertilizer (20 kg N ha^{-1}) when the plants reached 30 cm in height.

Measurements

The seed yield (t ha^{-1}) was calculated from a harvested area of 7.2m^2 (4 rows × 5m × 0.45m) in each plot. The seed protein content (%) was measured using the Near Infrared Reflected Spectroscopy (NIRS) method with the German Bruker Multi-Purpose Analyzer at the Adnan Menderes University Agricultural Biotechnology and Food Safety Application and Research Center (ADU-TARBIYOMER) (Gislum, R.; Micklander, E.; Nielsen, J. Quantification of nitrogen concentration in perennial ryegrass and red fescue using near-infrared reflectance spectroscopy (NIRS) and chemometrics. Field Crops Res. 2004, 88, 269–277).

To measure the seeds' amino acid content (g/100g), the dry seeds were ground after harvesting, and random samples were prepared for each plot. A total of seventeen amino acids were measured. The seed amino acid analyses were performed by means of high-performance liquid chromatography (HPLC) at the Research and Implementation Center for Drug Development and Pharmacokinetics Laboratory at Ege University.

Statistical analysis

The statistical analyses were conducted using JMP Statistical Analysis Software (version Pro 13) for the RCBD (Randomized Complete Block Design) arranged in split plot. The experimental data for each study parameter were subjected to statistical analysis using the analysis of variance (ANOVA) technique, and significance was tested by the "F" test (Steel and Torrie, 1980). When differences were found in the ANOVA, means were compared using Fisher's protected least significant difference (LSD) test at $p \leq 0.05$.

RESULTS AND DISCUSSIONS

According to the results of the analysis of variance (Table 2 and 3), the difference between the years was found to be statistically significant. The average data for each year were therefore evaluated separately. As seen in Table 2 and 3, the interaction of cultivar and phosphorus dose was found to be significant for the seed yield, the protein content and the levels of aspartic acid, serine, histidine, threonine, arginine, alanine, methionine, phenylalanine, isoleucine, lysine, leucine and proline, while the phosphorus dose factor was found to have a significant impact on the protein ratio. The means of the characteristics are presented in tables 4 and 5.

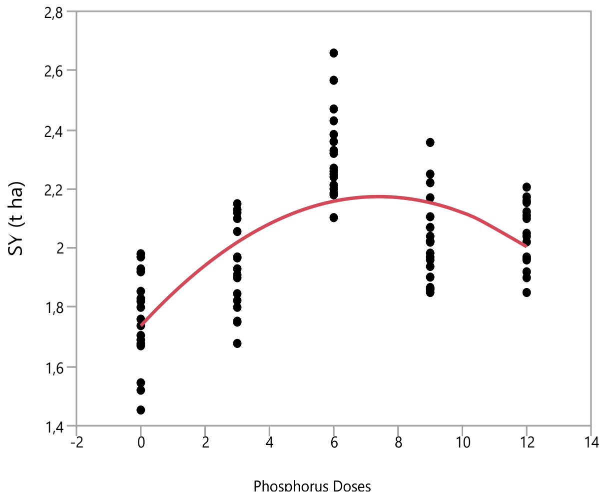

Figure 2. SY (t ha) = 1.886 + 0.021*Phosphorus Doses - 0.006*(Phosphorus Doses-6)^2.

Figure 2 shows that the quadratic relationship between the seed yield and phosphorus doses was found to be significant. The seed yield values in the study varied between 1.55 t ha^{-1} and 2.54 t ha^{-1}. As seen in the graph (fig 2), the seed yield increased from dose 0 to dose 6 kg da^{-1} but started to decrease after dose 6 kg da^{-1}. The effect of excess phosphorus on plants is mostly indirect. On the other hand, since phosphate ions are tightly retained in the soil, it is difficult for plants to take up phosphate ions. Therefore, excess phosphorus in plants is not a common occurrence. In the event of excess phosphorus, deficiencies of micronutrients such as zinc and iron may arise, and calcium, boron, copper and manganese deficiencies may also occur (Aktas and Ates, 1998; Bolat and Kara, 2017). The responses of all the cultivars to the phosphorus doses were similar to each other, although the Kitik had a higher yield potential than the others. The results for the two years of the experiment differed due to the meteorological conditions.

Table 2. Results of analysis of variance for the characteristics examined

Variation Source	DF	Means of Square										
		SY	Protein	ASP	GLU	SER	HIS	GLY	THR	ARG	ALA	ASP
Year	1	9020,01**	188,298**	0,00403**	0,0164**	0,02601**	0,00196**	0,0042**	0,00581*	0,11378**	0,10431**	0,00164**
Cultivar	2	2294,93**	6,39292*	0,01538**	0,03466*	0,00066**	0,00045**	0,00502ns	0,00031 ns	0,01352**	0,05637**	0,03466*
Year*Cultivar	2	1301,22**	0,45456ns	0,0201ns	0,02301ns	0,34083ns	0,00004ns	0,00004ns	0,00014 ns	0,00263 ns	0,04685**	0,06932 ns
Phosphorus	4	5141,27**	32,6044**	0,00029**	0,00884ns	0,00373**	0,00107**	0,00433ns	0,00459**	0,00814**	0,00124 ns	0,00884
C*P	8	96,8539**	3,7232**	0,00662**	0,02387**	0,00114 ns	0,00207**	0,00824**	0,00207**	0,00814**	0,00272*	0,02387*
Y*P	4	772,881**	13,9548**	0,01210ns	0,00172ns	0,05299ns	0,0013ns	0,0058*	0,00001 ns	0,00242 ns	0,00115 ns	0,00237ns
Y*C*P	8	59,5937**	2,40739ns	1,21050ns	0,02310ns	0,00519ns	0,0261ns	0,0483ns	0.0015 ns	0,03254 ns	0,00103 ns	0,00507ns

Y:year; C:cultivar; P: phosphorus; **, * Significant at P ≤ 0.01 and 0.05, respectively, ns – nonsignificant

Table 3. Results of analysis of variance for the characteristics examined

Variation Source	DF	Means of Square										
		TYR	CYS	VAL	MET	PHE	ILE	LYS	LEU	PRO	TYR	CYS
Year	1	0,00289**	0,00048*	0,0003**	0,00453**	0,00012*	0,00113*	0,00062*	0,00052**	0,00009**	0,00048*	0,00030**
Cultivar	2	0,00982ns	0,00049*	0,00945ns	0,00042ns	0,00069**	0,00350ns	0,01556**	0,0038*	0,00268*	0,00049*	0,00945ns
Year*Cultivar	2	0,00001ns	0,00017ns	0,0189ns	0,00223ns	0,0015ns	0,00147**	0,00265ns	0,00011ns	0,00235ns	0,00017ns	0,01634ns
Phosphorus	4	0,00728ns	0,00208**ns	0,00708ns	0,00058**	0,00493ns	0,00304ns	0,00017ns	0,00881ns	0,00250ns	0,00208**	0,00708ns
C*P	8	0,00834ns	0,00019ns	0,01348*	0,00049**	0,00272**	0,00176**	0,00669**	0,00679ns	0,00616**	0,00019ns	0,01348*
Y*P	4	0,02913ns	0,00833ns	0,00001ns	0,00258ns	0,01642ns	0,00365ns	0,00053ns	0,01425ns	0,01052ns	0,00723ns	0,00001ns
Y*C*P	8	0,00332ns	0,00006ns	0,00663ns	0,00015*	0,00106ns	0,00121ns	0,00023ns	0,04423ns	0,03731ns	0,00023ns	0,01720ns

Y:year; C:cultivar; P: phosphorus; **, * Significant at P ≤ 0.01 and 0.05, respectively, ns – nonsignificant

Table 4. Means of unit yield, protein ratio and amino acid levels (g/100g) measured in the experiment.

Cultivars	P Doses	SY (t ha^{-1})		Protein (%)		ASP		GLU		SER		HIS		GLY		THR		ARG		ALA	
		2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023
Salkim	0	1.66	1.60	22.0	22.0	1.161	1.172	1.752	1.779	0.397	0.431	0.222	0.230	0.351	0.336	0.342	0.321	1.036	1.111	0.418	0.397
	3	1.77	1.74	22.5	22.5	1.095	1.106	1.739	1.766	0.414	0.448	0.249	0.257	0.386	0.371	0.365	0.344	0.983	1.058	0.396	0.375
	6	2.25	1.92	22.2	25.0	1.128	1.104	1.746	1.780	0.406	0.427	0.236	0.257	0.369	0.408	0.323	0.302	1.010	1.102	0.407	0.384
	9	1.91	1.70	22.2	24.8	1.128	1.099	1.746	1.804	0.406	0.416	0.236	0.234	0.369	0.359	0.359	0.338	1.010	1.151	0.407	0.378
	12	2.09	1.78	22.3	25.7	1.117	1.080	1.733	1.643	0.408	0.442	0.24	0.273	0.374	0.375	0.390	0.369	1.001	1.116	0.403	0.390
Kitik	0	1.87	1.68	23.0	23.0	1.124	1.118	1.745	1.783	0.407	0.427	0.237	0.272	0.370	0.397	0.328	0.307	1.007	1.104	0.406	0.379
	3	2.05	1.81	26.5	26.5	1.123	1.181	1.745	1.901	0.407	0.461	0.238	0.262	0.371	0.406	0.353	0.335	1.006	1.134	0.405	0.397
	6	2.54	2.00	28.0	28.0	1.121	1.140	1.744	1.851	0.407	0.424	0.238	0.280	0.372	0.394	0.369	0.358	1.004	1.156	0.405	0.392
	9	2.23	1.81	26.9	26.9	1.123	1.142	1.744	1.774	0.407	0.429	0.238	0.233	0.371	0.357	0.358	0.347	1.006	1.129	0.405	0.395
	12	2.18	1.80	25.6	25.6	1.122	1.178	1.744	1.791	0.407	0.422	0.238	0.237	0.371	0.364	0.351	0.340	1.005	1.111	0.405	0.405
Filiz	0	1.55	1.68	23.1	23.1	1.122	1.085	1.744	1.766	0.407	0.444	0.238	0.270	0.372	0.409	0.346	0.334	1.005	1.019	0.405	0.330
	3	1.81	1.74	25.4	25.4	1.123	1.108	1.745	1.778	0.407	0.458	0.238	0.263	0.371	0.350	0.355	0.342	1.005	1.077	0.405	0.223
	6	2.17	1.93	28.0	27.3	1.122	1.119	1.744	1.695	0.407	0.400	0.238	0.253	0.371	0.343	0.325	0.312	1.005	1.105	0.405	0.247
	9	1.94	1.83	25.5	25.0	1.122	1.131	1.744	1.789	0.407	0.442	0.238	0.265	0.371	0.404	0.380	0.367	1.005	1.094	0.405	0.235
	12	1.95	1.73	25.6	25.6	1.122	1.122	1.744	1.830	0.407	0.460	0.238	0.244	0.371	0.293	0.379	0.366	1.005	1.122	0.405	0.196
Mean		2.0	1.8	24.6	25.1	1.124	1.126	1.745	1.782	0.407	0.435	0.237	0.255	0.371	0.371	0.355	0.339	1.006	1.106	0.406	0.342
LSDp		0.069	0.051	0.984	1.252	-	0.03	-	-	0.014	0.017	0.009	0.012	-	-	0.01	0.009	0.022	0.016	-	-
LSDc		0.062	-	-	0.888	-	0.004	-	-	-	-	-	-	-	-	-	-	-	0.001	0.008	0.02
LSD p*c		-	4.24	-	-	0.01	1.011	-	-	0.016	0.018	0.016	0.017	-	-	0.031	0.032	0.023	0.023	0.018	0.048

SY:Seed yield, ASP: Aspartic acid, GLU: Glutamic acid, SER: Serine, HIS: Histidine, GLY:Glycine, THR: Threonine, ARG: Arginine, ALA:Alanine.

Table 5. Means for levels of amino acids (g/100g) measured in the experiment.

Cultivars	P Doses	TYR		CYS		VAL		MET		PHE		ILE		LYS		LEU		PRO	
		2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023	2022	2023
Salkim	0	0.267	0.278	0.105	0.095	0.310	0.314	0.096	0.097	0.388	0.390	0.449	0.451	0.782	0.806	0.476	0.485	0.445	0.447
	3	0.277	0.288	0.075	0.065	0.460	0.464	0.076	0.077	0.395	0.397	0.464	0.466	0.783	0.807	0.545	0.554	0.413	0.415
	6	0.272	0.285	0.090	0.066	0.385	0.473	0.086	0.095	0.392	0.444	0.456	0.488	0.782	0.869	0.511	0.543	0.429	0.397
	9	0.272	0.262	0.090	0.081	0.385	0.485	0.086	0.097	0.392	0.437	0.456	0.471	0.782	0.807	0.511	0.462	0.429	0.403
	12	0.274	0.283	0.085	0.094	0.410	0.460	0.083	0.074	0.393	0.378	0.459	0.453	0.783	0.785	0.522	0.518	0.424	0.448
Kitik	0	0.273	0.287	0.088	0.094	0.393	0.494	0.085	0.091	0.392	0.434	0.457	0.507	0.782	0.844	0.514	0.538	0.427	0.436
	3	0.273	0.272	0.088	0.079	0.396	0.473	0.084	0.087	0.392	0.367	0.458	0.447	0.782	0.840	0.516	0.481	0.427	0.455
	6	0.273	0.286	0.087	0.079	0.400	0.485	0.084	0.092	0.392	0.427	0.458	0.487	0.783	0.807	0.517	0.485	0.426	0.449
	9	0.273	0.317	0.088	0.084	0.396	0.446	0.084	0.105	0.392	0.392	0.458	0.470	0.782	0.764	0.516	0.437	0.427	0.452
	12	0.273	0.316	0.088	0.108	0.397	0.470	0.084	0.095	0.392	0.416	0.458	0.467	0.782	0.788	0.516	0.496	0.426	0.388
Filiz	0	0.273	0.265	0.087	0.082	0.398	0.449	0.084	0.076	0.392	0.413	0.458	0.433	0.782	0.742	0.516	0.448	0.426	0.467
	3	0.273	0.298	0.088	0.075	0.397	0.457	0.084	0.092	0.392	0.403	0.458	0.447	0.782	0.757	0.516	0.507	0.426	0.393
	6	0.273	0.417	0.088	0.078	0.397	0.421	0.084	0.078	0.392	0.432	0.458	0.457	0.782	0.724	0.516	0.537	0.426	0.476
	9	0.273	0.297	0.087	0.081	0.397	0.469	0.084	0.097	0.392	0.417	0.458	0.454	0.782	0.776	0.516	0.496	0.426	0.422
	12	0.273	0.298	0.088	0.092	0.397	0.440	0.084	0.089	0.392	0.417	0.458	0.454	0.782	0.792	0.516	0.486	0.426	0.443
Mean		0.273	0.297	0.088	0.083	0.395	0.453	0.085	0.089	0.392	0.411	0.457	0.463	0.782	0.794	0.515	0.498	0.427	0.433
LSDp		-	-	0.016	0.016	-	-	0.005	0.005	0.011	0.010	-	-	-	-	0.007	0.008	-	-
LSDc		-	-	-	-	-	-	-	-	-	-	0.010	0.011	0.014	0.011	0.012	0.012	0.019	0.021
LSD p*c		-	-	-	-	-	-	0.016	0.017	0.026	0.026	0.024	0.024	0.034	0.035	0.031	0.312	0.021	0.022

TYR: Tyrosine, CYS: Cysteine, VAL:Valine, MET:Metionine, PHE: Phenylalanine, ILE: Isoleucine, LYS: Lysine, LEU: Leucine, Pro: Proline

In the second year, cool and rainy weather during the period of pod tying and seed filling, which coincided with March and April, led to a prolongation of this period, as a result of which higher average yields were obtained (Guevara et al., 2022). This is because faba beans are considered to be sensitive to high temperatures and low soil moisture during the flowering and ripening period (L'opez-Bellido et al., 2005). Previous research has shown that phosphorus increases the seed yield of faba bean crops. The use of phosphorus fertilizers can increase plants' uptake of phosphorus and lead to improved seed yield. Previous study reported that a dose of 40kg ha^{-1} phosphorus is appropriate for optimum yield (Yasmin et al., 2020). In a similar study, it was reported that 46 kg phosphorus resulted in the highest seed yield (Kubure et al., 2016). In a study conducted in Ethiopia, it was determined that doses of 36-72kg ha^{-1} of phosphorus increased yield by between 9.8% and 15.7% compared to the control group (EIAR, 2011). (Masood et al., 2011; Tadele et al., 2016) recorded increases in yield as the doses of phosphorus increased.

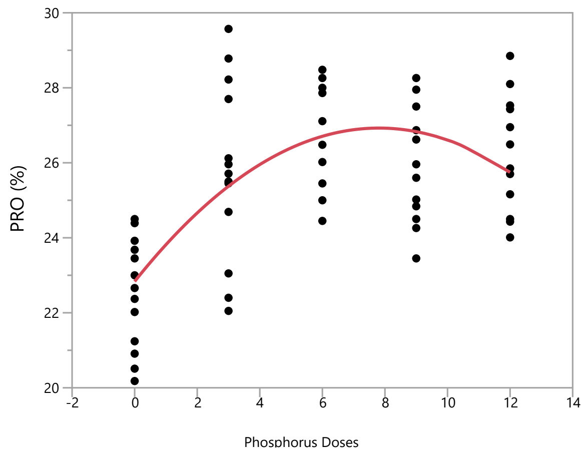

Figure 3. (PRO (%) = 25.251 + 0.243*Phosphorus Doses - 0.067*(Phosphorus Doses-6)^2.

As shown in Figure 3, the quadratic relationship between seed protein ratio and phosphorus doses was found significant. The graph shows that the seed protein ratio increased from dose 0 to dose 6kg da^{-1}, but decreased after dose 6kg da^{-1}. The mean of the protein ranged between 22% and 28%. The cultivar of the faba bean did not have a significant impact on the protein content. Phosphorus participates in the structure of nucleotides (such as ATP), which play an important role in protein synthesis. Stored in the cell as ATP (adenosine triphosphate), it is used in energy transportation, cellular communication and protein synthesis (Bonora et al., 2012). Protein synthesis is the process by which cells combine amino acids to produce new proteins. In this process, ATP provides energy, allowing amino acids to combine and thus contributes to the synthesis of proteins (Kim and Swartz, 1999). In this way, phosphorus makes an indirect contribution to protein synthesis. In previous studies (Crépon et al., 2010; Gucci et al., 2019) reported that protein content increased with increased doses of phosphorus. However, reported that doses of phosphorus had no effect on seed protein content in pods (Henry et al., 1995).

Table 6 shows the relationships between seed yield, protein content and some amino acids, including the essential ones, based on correlation analysis. The protein of Fabaceae seeds is characterised by a lower value than that of animal protein and a higher value than that of cereal protein (Khattab et al., 2009). The amino acid profile of faba bean includes essential amino acids – i.e., isoleucine, leucine, lysine, methionine, tyrosine, phenylalanine, valine, histidine and threonine – and nonessential amino acids, namely: aspartic acid, glutamic acid, alanine, arginine, glycine, proline, and ser-ine (Khalil and Mansour, 1995).

There was a positive and significant correlation between seed yield and protein content (0.5876**). As seed yield increases, protein content will increase because the amount of protein accumulated in the cotyledons increases with the increase in seed weight and yield (Gasim et al., 2015). There is a positive and significant relationship between seed yield and alanine (0.3046**): in this case, as the yield increases, the alanine content is expected to increase too. There is a significant negative correlation between serine and seed yield (-0.5106**). There is a positive and significant correlation between serine and histidine (0.3152**) and arginine (0.3448**), while there is a negative and significant correlation between alanine (-0.4766**), phenylalanine (-0.3694**) and isoleucine (-0.2965**). Serine is an important building block in the process of protein synthesis. In ribosomes, the amino acid serine is recognized by the codons of genetic information on mRNA and incorporated into proteins. Serine plays an important role in the synthesis of other amino acids such as glycine, methionine and threonine in plants (Simola, 1968). For this reason, it is one of the most important amino acids. The relationship between seed yield and lysine and methionine was found to be insignificant. In other words, the increase in seed yield achieved by agrotechnical methods may not lead to an increase in these essential amino acids. It is important to consider this situation in terms of seed nutrition. The positive and significant relationship between lysine, methionine (0.2938**) and isoleucine (0.4438**) is remarkable.

Regarding the amino acid composition, it is observed that the level of lysine is high and the level of amino acids containing sulphur (methionine and cysteine) is low. Similar results were obtained in previous studies (Apata and Ologhobo, 1994; Khattab et al., 2009). Methionine has a positive and significant correlation with arginine (0.3254**) and a negative and significant correlation with histidine (-0.2904**). Leucine has positive and significant correlations with phenylalanine (0.2139*) and isoleucine (0.2772*). This indicates that if leucine tends to increase, phenylalanine and isoleucine will increase.

Some of the amino acid results obtained from the trial are presented in graph form in Figures 4, 5, 6, 7, 8, 9, 10, 11 and 12.

Table 6. Correlation coefficients between yield and yield components.

	SY (t ha)	PRO (%)	ASP	SER	HIS	THR	ARG	ALA	CYS	MET	PHE	ILE	LYS	LEU
SY (t ha)	-													
PRO (%)	0.5876**	-												
ASP	0.0178	0.0932	-											
SER	-0.5106**	-0.1743	0.1274	-										
HIS	0.0475	-0.0014	-0.2481*	0.3152**	-									
THR	0.1954	0.3191**	-0.0941	0.0034	0.0230	-								
ARG	0.0025	0.1132	0.4312**	0.3448**	0.0192	-0.1240	-							
ALA	0.3046**	0.0814	0.0747	-0.4766**	-0.2135*	0.0642	-0.2586*	-						
CYS	-0.0047	-0.1187	0.2450*	-0.1964	-0.0973	0.2660*	0.0207	0.1468	-					
MET	0.0145	-0.0627	0.2417*	-0.0536	-0.2904**	-0.0813	0.3254**	0.0634	0.1273	-				
PHE	0.1268	-0.0098	-0.2343*	-0.3694**	-0.0147	-0.2858**	0.0844	-0.1613	-0.0113	0.1038	-			
ILE	0.2319*	0.0302	0.0149	-0.2965**	0.0903	-0.2773**	0.2635*	0.1678	0.0055	0.2382*	0.4771**	-		
LYS	0.0682	-0.2000	0.2089*	0.0952	0.0963	-0.2750**	0.2496*	0.2381*	-0.0229	0.2938**	0.1937	0.4438**	-	
LEU	0.0351	0.0428	-0.1796	0.0108	0.1891	-0.2745**	-0.0485	-0.0789	-0.1678	-0.2052	0.2139*	0.2772*	0.1396	-
PRO	-0.0351	-0.0422	0.0095	-0.0427	0.2058	-0.0388	0.0726	-0.0374	0.1362	-0.0800	-0.1965	-0.1165	-0.2116*	-0.2359*

* Significant at p<0.05, ** significant at p<0.01

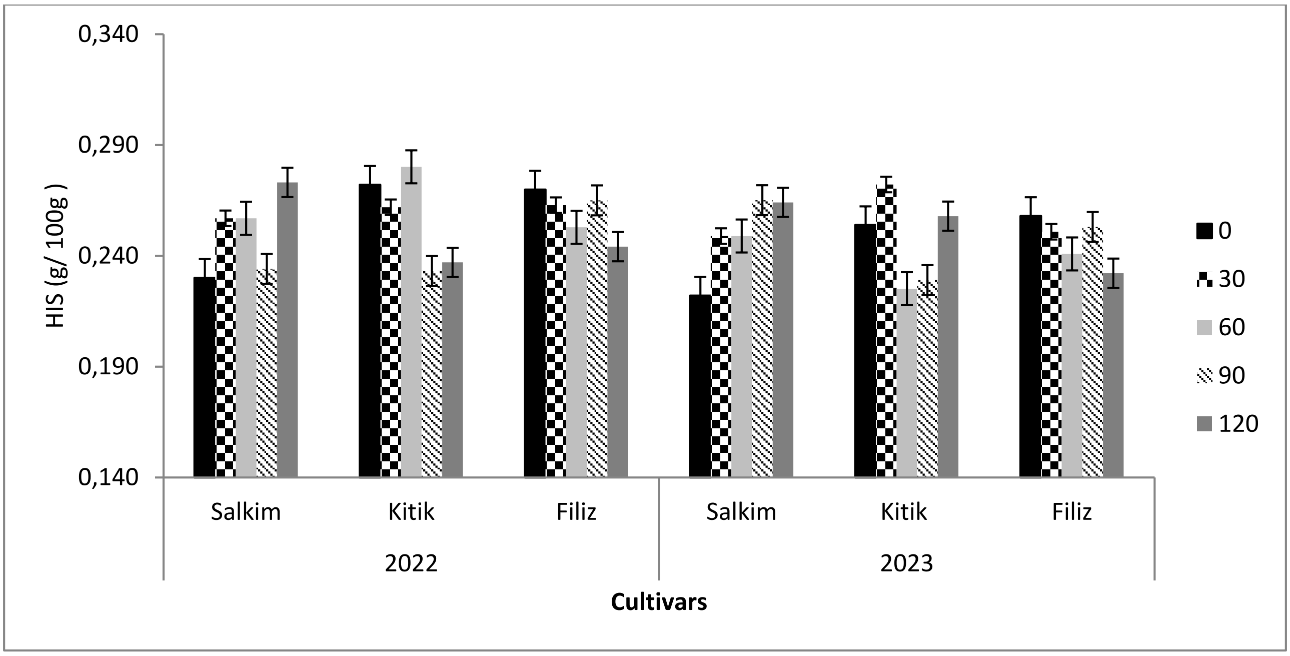

Figure 4. Histidine means obtained in the study

Figure 4 shows the differences in histidine content between the two years. The responses of the cultivars to the various doses are also different. The interaction of cultivar and phosphorus dose was found to be significant, with the highest histidine (0.275g/100g) observed in the Kitik at a dose of 60kg ha^{-1}.

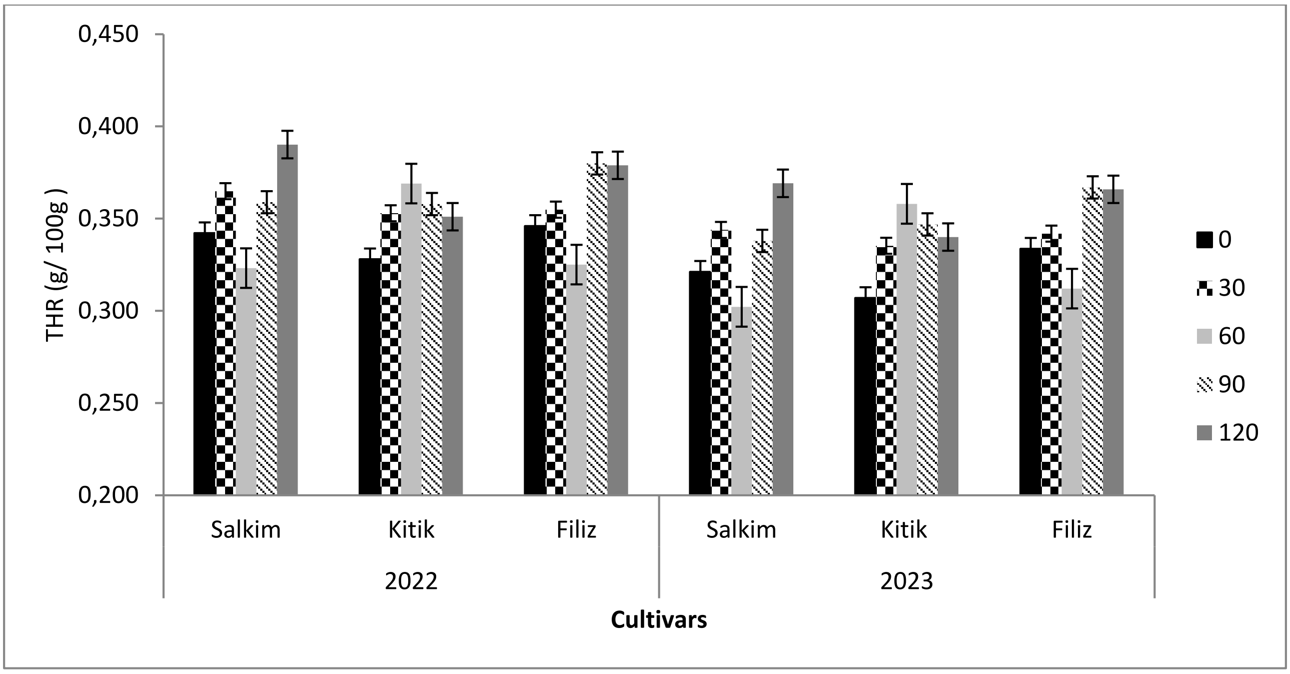

Figure 5. Threonine means obtained in the study

As shown in Figure 5, the interaction of cultivar and phosphorus dose was found to have a significant effect on threonine content, with the highest threonine value (0.390g/100g) observed in the Salkim at a dose of 120 kg ha^{-1}. The level of threonine, which is one of the essential amino acids found in small quantities in pods, was lower in the control groups of all cultivars by comparison with the groups to which phosphorus was applied. It follows that it may be possible to increase the level of this amino acid by applying phosphorus.

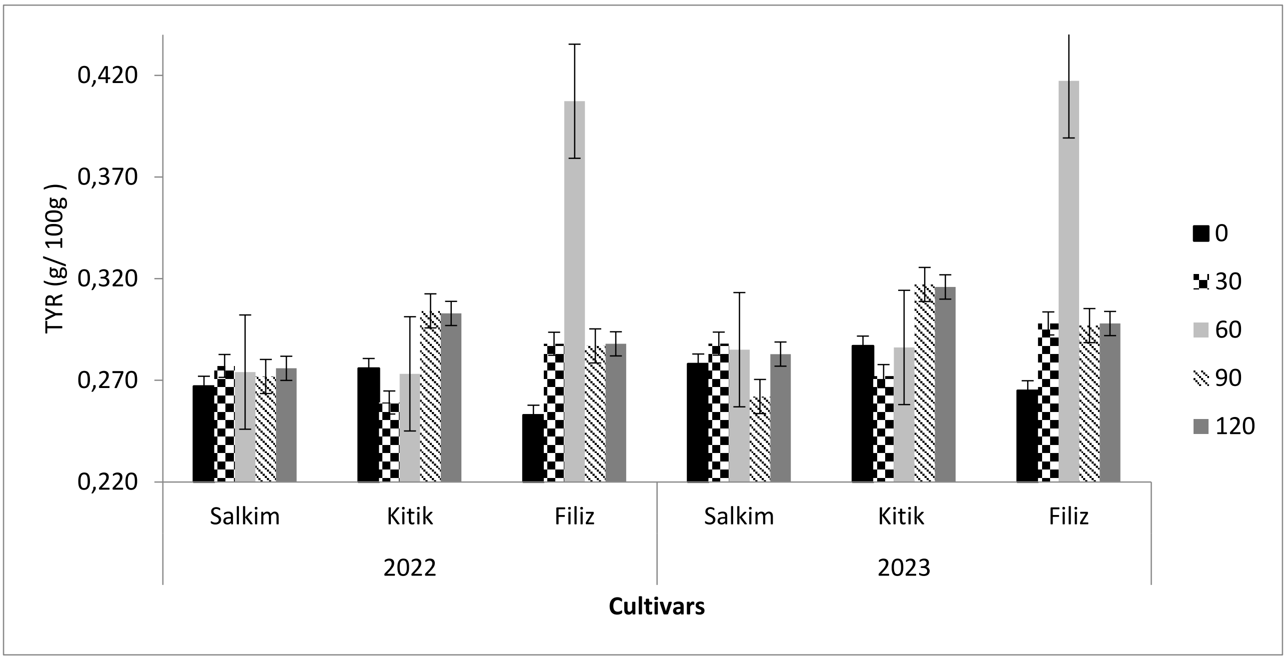

Figure 6. Tyrosine means obtained in the study

Figure 6 shows the tyrosine mean. These values did not display a statistically significant relationship with different cultivars or doses of phosphorus. Tyrosine is one of the non-essential amino acids. However, the response of the Filiz cultivar to the phosphorus dose of 60 kg ha^{-1} is very evident from the graph (Figure 7).

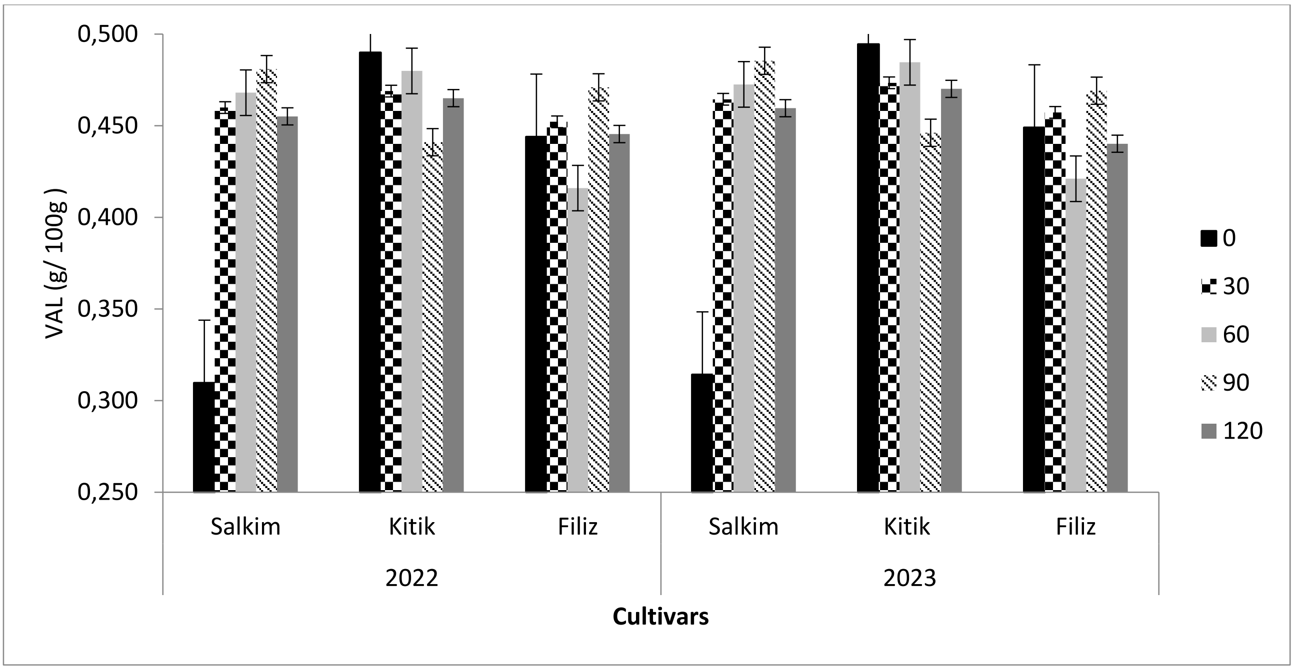

Figure 7. Valine means obtained in the study

The various doses of phosphorus and cultivars were not found to be statistically significant for the mean of valine shown in Figure 7. However, the valine level in the Salkim rose when phosphorus was applied by comparison with the control group. Previous studies have determined that amino acid levels may vary by cultivar (Alghamdi, 2009; Caglar et al., 2017).

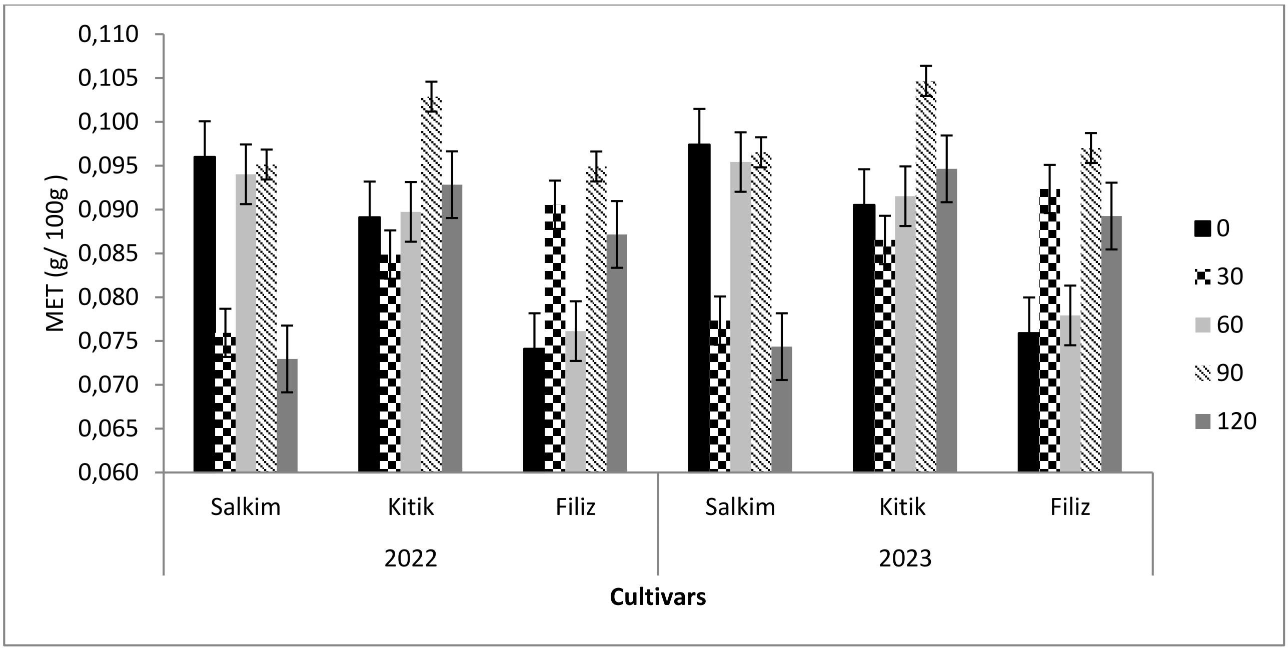

Figure 8. Methionine means obtained in the study

The interaction of cultivar and phosphorus dose was found to be significant for methionine. As shown in Figure 8, methionine levels were found to be higher when phosphorus was applied at 90 kg ha^{-1} than in the control group. Methionine is the essential amino acid present in the lowest quantities (Farzana and Khalil, 1990). Among the different cultivars, Kitik has the highest methionine content. Cultivators might be consider applying phosphorus to increase the level of methionine.

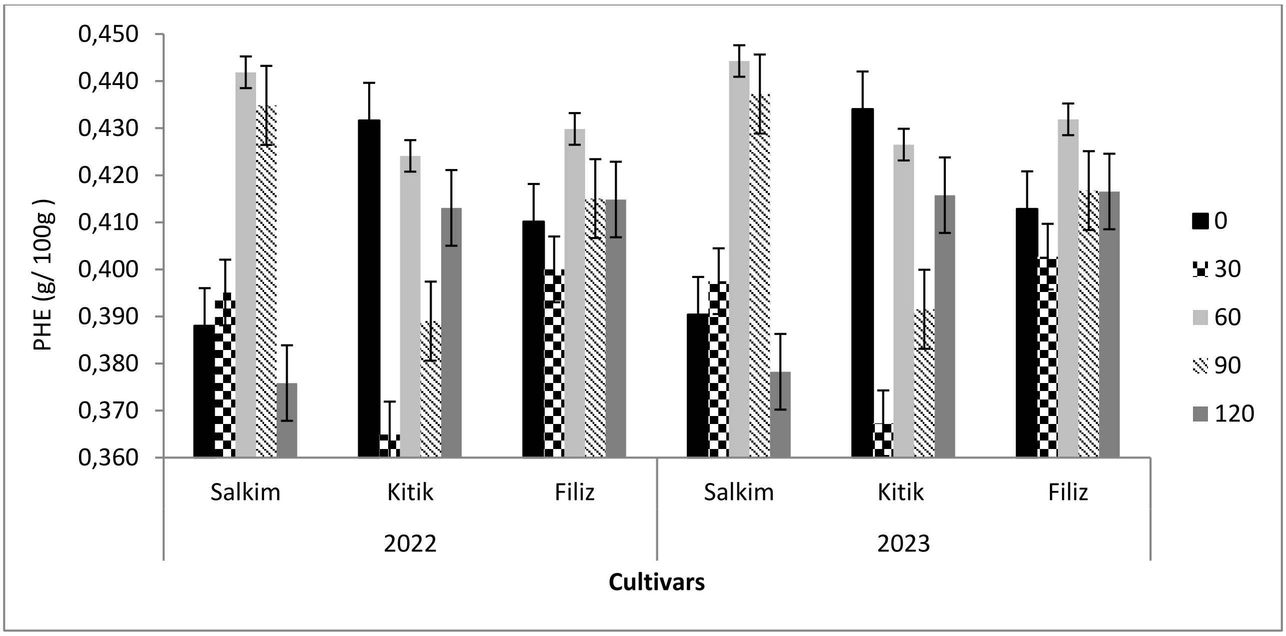

Figure 9. Phenylalanine means obtained in the study

The interaction of cultivar and phosphorus dose was found to be significant for phenyalanine. The mean phenylalanine values obtained are presented in Figure 9. In the Salkim and Filiz, the highest values were obtained at a dose of 60 kg ha^{-1}. However, the phenylalanine levels decreased in the Filiz when phosphorus was applied, by comparison with the control group. Phenylalanine regulates many physiological functions in plants such as their colour, aroma, defence and responses to environmental stresses.

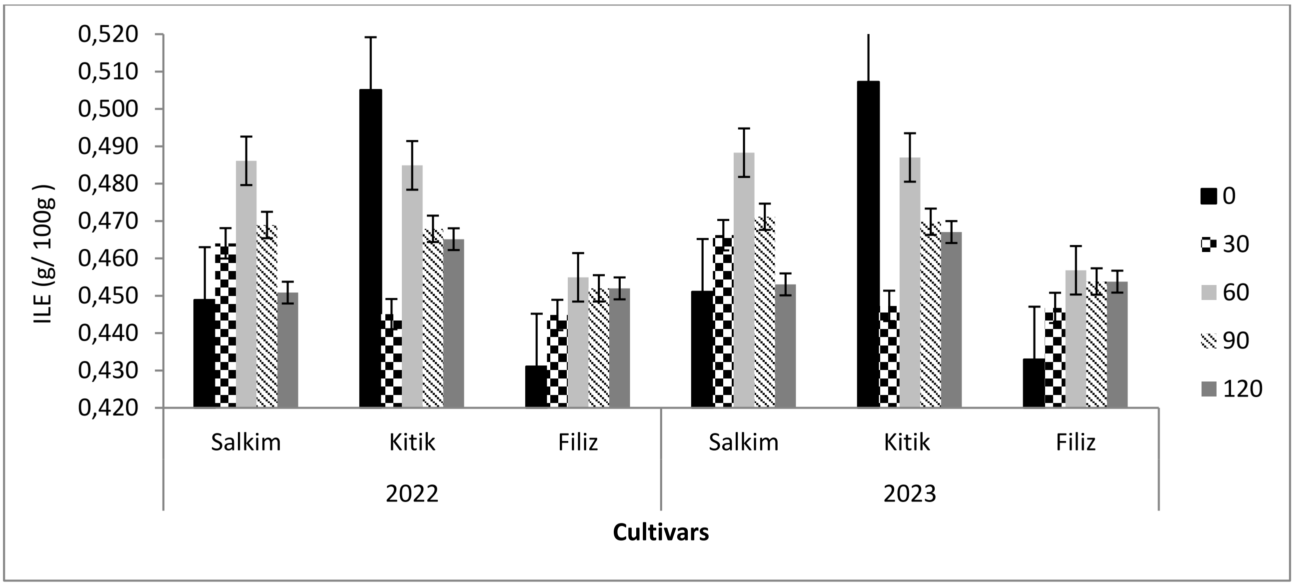

Figure 10. Isoleucine means obtained in the study

The interaction of cultivar and phosphorus dose was found to be significant for isoleucine levels. As seen in Figure 10, the responses of the cultivars to the application of phosphorus differed. Applications of phosphorus decreased the isoleucine content in the Kitik. Filiz and Salkim, a dose of 60kg ha^{-1} was observed to result in the highest isoleucine content. Isoleucine is an intermediate in the synthesis of cytokinins, which are important hormones in plants. Cytokinins regulate many physiological processes in plants such as growth, flowering, fruit formation and root development (Santner et al., 2009).

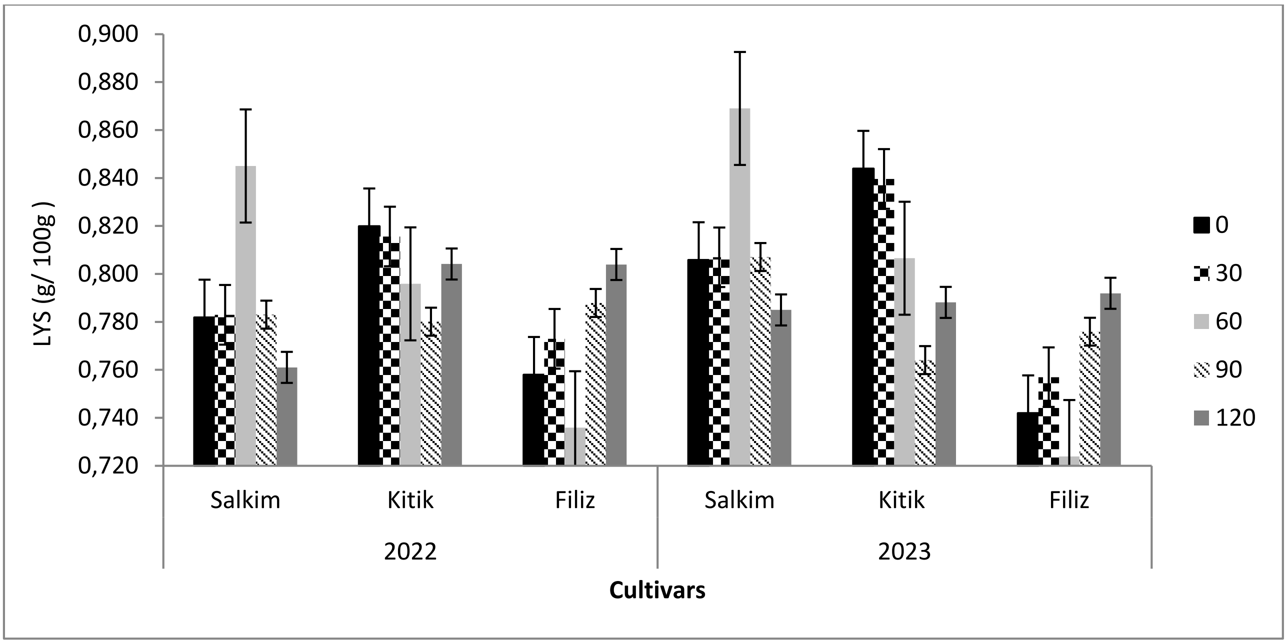

Figure 11. Lysine means obtained in the study

The interaction of cultivar and phosphorus dose was found to have a significant effect on lysine levels. Although lysine is found in lower amounts in cereals, it ranks third (0.788g/100g) among the essential amino acids in seed. As seen in Figure 11, the highest lysine content was obtained in the Salkim at a dose of 60kg ha^{-1}. While the application of phosphorus caused the level of lysine in the Kitik to decline, it positively affected the level of lysine in the Filiz by comparison with the control group. Lysine is used to increase plant resistance against abiotic and biotic stresses, for the production of glutamate amino acid and especially for the production of glutamate, which plays a leading role in the preparation of plant defence systems (Azevedo and Lea, 2001).

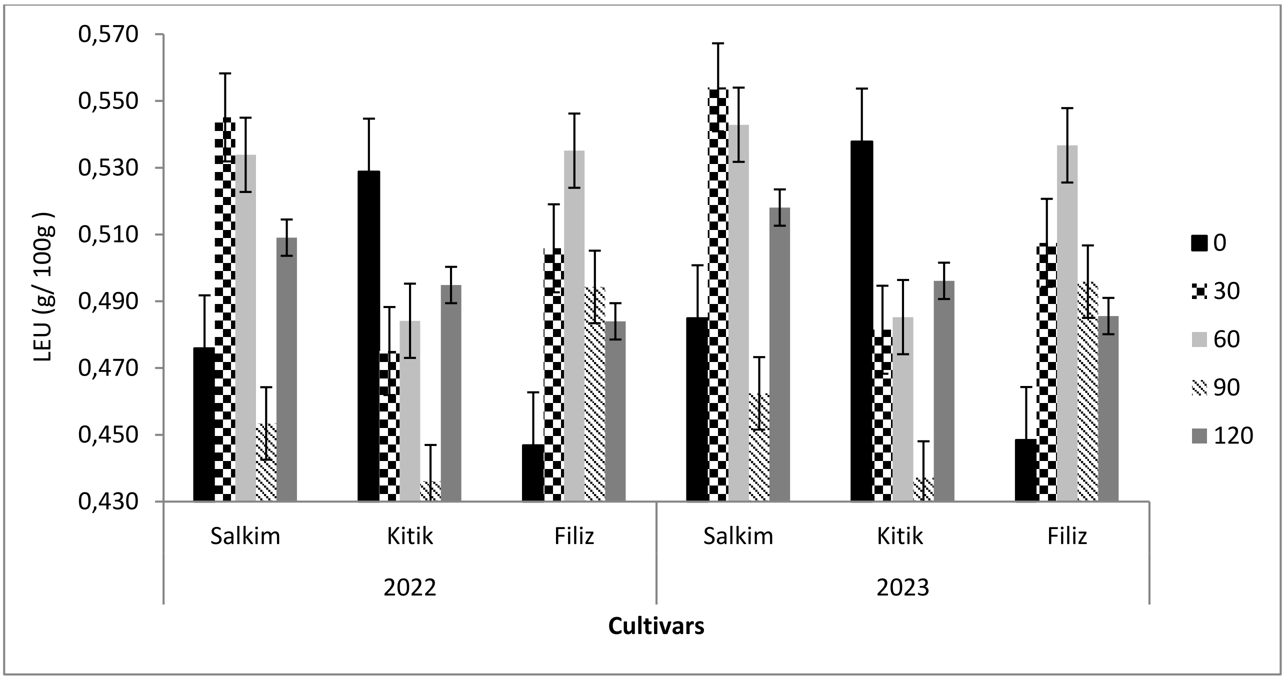

Figure 12. Leucine means obtained in the study

The interaction of cultivar and phosphorus dose was found to have a significant effect on leucine content. As can be seen in Figure 12, the leucine content in the Salkim and Filiz increased by comparison with the control group when phosphorus was applied, but applying phosphorus caused the leucine content in the Kitik to decrease. Thus the results of applying phosphorus differed depending on the cultivar. While a dose of 30 kg ha^{-1} produced the highest leucine level in the Salkim, a dose of 60 kg ha^{-1} produced the highest level in the Filiz. Leucine plays an important role in reducing damage from oxygen free radicals, especially in arid conditions, and in reducing the levels of free oxygen-based radicals in cells (Mikkelsen and Halkier, 2003).

CONCLUSION

The average seed yield obtained in the trial was 1.89t ha^{-1} was obtained. Among the cultivars, the Kitik had the highest average seed yield at 1.99t ha^{-1}. Seed yields showed a significant increase with the application of phosphorus. Meanwhile, correlation analysis revealed a positive and significant relationship between the seed yield and the protein ratio. The protein ratio was found to be relatively high in the Kitik (25.8%) and Filiz (25.4%). The orthogonal relationship between phosphorus doses and the protein ratio was also significant, but the protein ratio showed a declining trend for doses ın excess of 60 kg ha^{-1}. The levels of seventeen different amino acids in the seeds were measured. Among these, histidine, phenylalanine, threonine, tyrosine, valine, methionine, isoleucine, leucine, lysine are essential amino acids and cannot be synthesized by the human body. These amino acids are amino acids which the body needs but which must be obtained externally through food. More and more attention is being paid to the levels of these amino acids in foodstuffs, and to work on raising these levels. Legumes contain higher levels of essential amino acids than cereals. In legumes, the levels of sulphur-containing amino acids such as methionine, tryptophan and cysteine is generally low, while lysine content is higher than in cereals. Agronomic practices which increase the share of essential amino acids in the interaction of amino acids are of great value. In the study, applying doses of phosphorus was found to have a significant effect on the levels of amino acids containing sulphur such as methionine and cysteine, which are present in lower quantities in legumes than in cereals. In addition, levels of essential amino acids such as histidine, lysine, phenylalanine, threonine and leucine were affected significantly by the application of phosphorus. Phosphorus doses of up to 60kg ha^{-1} or 90kg ha^{-1} increased the amino acid content. The differences between the amino acid contents of the different cultivars were also found to be significant. The essential amino acid values obtained from the Kitik and Salkim cultivars were relatively high. The phosphorus content of amino acids can vary depending on their structure. Amino acids with a high phosphorus content include cysteine, threonine and tyrosine. However, the levels of the amino acids measured remain quite low. When the amino acid values obtained were analyzed, the cultivar and the doses of phosphorus applied were found to have an effect on the amino acid concentration. In conclusion, yields and levels of protein and amino acids vary considerably on the plant species, the cultivar and the growing conditions.

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