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Exploring the Impact of Germination on the Macro, Micro and Toxic Element Contents of Kidney Bean and White Bean Seed Germinated at Different Sprout Lenghts

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ABSTRACT

Calcium (Ca) was the least abundant macronutrient in both legumes. Potassium (K) contents in kidney bean and white bean samples ranged from 13,687.08 mg/kg (control) to 20,078.57 mg/kg (6 cm sprouts) and 3,601.90 mg/kg (3 cm sprouts) to 6,459.55 mg/kg (6 cm sprouts), respectively. Calcium contents ranged from 413.91 mg/kg (1 cm sprouts) to 693.78 mg/kg (control) in kidney beans, and from 386.18 mg/kg (1 cm sprouts) to 1,090.84 mg/kg (control) in white beans. The highest phosphorus (P), potassium (K), and magnesium (Mg) contents were found in 6 cm germinated kidney bean (P: 8,095.27 mg/kg; K: 20,078.57 mg/kg; Mg: 1,523.34 mg/kg) and white bean (P: 21,475.71 mg/kg; K: 21,475.71 mg/kg; Mg: 1,169.34 mg/kg) sprouts.

Iron (Fe) contents ranged from 39.60 mg/kg (control) to 55.51 mg/kg (6 cm) in kidney beans, and from 52.22 mg/kg (control) to 80.22 mg/kg (6 cm) in white beans, showing a gradual increase with sprouting. Copper (Cu) was the microelement present in the lowest amounts in both legumes. The highest concentrations of toxic elements were chromium (Cr) and nickel (Ni). Lead (Pb) showed a slight increase with sprouting but remained below tolerable limits.

Keywords: kidney bean, white bean, sprouting, ICP-OES.

INTRODUCTION

Beans (*Phaseolus vulgaris* L.), which represent 50% of grain legumes consumed worldwide, are the main source of protein in the diet of most countries, as well as an excellent source of complex carbohydrates, polyunsaturated free fatty acids (linoleic and linolenic), fiber, vitamins and minerals (Talukder et al., 2010; Ribeiro et al., 2012). Legumes are an excellent source of macro and micronutrients (Zhou et al., 2023). Legumes, which are the second most important product in human nutrition after grains, are from the Leguminosae family and have special importance in meeting the need for plant protein (Siddiq and Uebersax, 2012; Semba et al., 2021). Increasing awareness of the importance of food diversity for human health has led to increased consumption of legumes rich in protein and minerals by consumers (Carbas et al., 2021). Legumes such as chickpeas, lentils and peas, which are grown and consumed worldwide, constitute the source of important protein, minerals and phytochemicals (Hamid et al., 2020; Şengül and Çalışlar, 2020). In developing countries where protein-energy malnutrition is common, legumes are consumed as a staple food and cheap protein source due to their high protein content, dietary fiber, starch, minerals and vitamins (Sozer et al., 2017). In addition, legumes and legume flour products are also used as gluten-free diets to improve the nutrition of the individual in celiac disease (Niewinski, 2008). During the germination process, which is defined as the process of seeds coming out of a dormant

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period and starting to sprout in a suitable environment, various enzymes and phytochemicals are produced to provide the nutrients necessary for the vital activities of the seeds after germination (Lorenz and D'Appolonia, 1980). Germination is a cheap and easy process that does not require complex equipment and provides high yields, making seed sprouts alternative sources of phytonutrients and minerals that are important for human health (Benincasa et al., 2019; Miyahira et al., 2021; Pasko et al., 2021). It has been observed that as a result of the increase in phytase activity with sprouting, the bioavailability of minerals improves, as seen in iron and calcium in germinated millet, green grass, cowpea, lentil and chickpea (Ghavidel and Prakash, 2007; Màrton et al., 2010; Suma and Urooj, 2014). Essential minerals and trace elements are vital for human health due to their direct impact on metabolic and physiological functions, and their deficiencies can cause dysfunction of an organism, contributing to the development of some non-communicable diseases (Gharibzahedi and Jafari, 2017). Therefore, the aim of this study was to investigate the effect of germination on the macro, micro and toxic element contents of kidney bean and white bean seed germinated at different lenghts.

MATERIAL AND METHODS

Material

Kidney bean and White bean grain samples were provided from Konya provinces in 2025. Before the analysis, the seeds were cleaned. HNO_3 and H_2O_2 are analytical grade and Merck company. The map showing the places where the seeds were collected is shown in Figure 1.

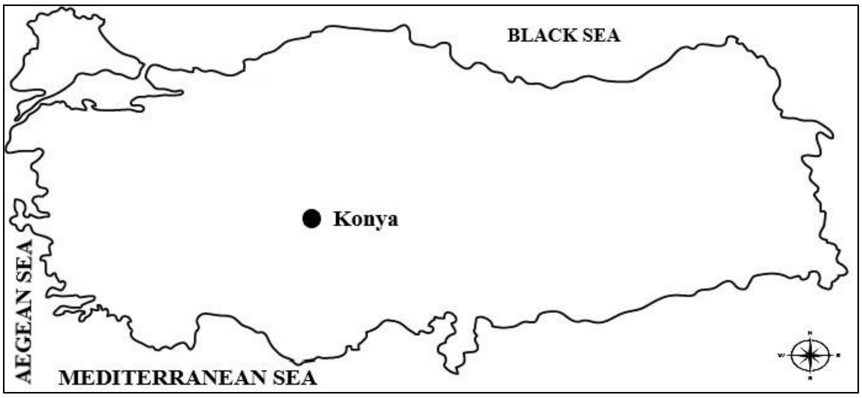

Figure 1. Locations where the seeds used in this study were collected

Method

Germination processing

Legume seeds were selected to be 100 g in equal size and the seed were soaked in 150 ml of pure water for 6 hours. Additionally, legume seeds were placed among cotton in petri dishes for germination. It was germinated by keeping it in 95% relative humidity and 25°C temperature for 10 days. Germination process was followed up to three different lengths (1 cm, 3 cm and 6 cm). Sampling was done when each germination length reached the desired level. After the dried samples were ground, they were filled into colored glass jars for analysis. Ungerminated grains constituted the control group. The elements were determined in the ICP-OES device.

Macro-, micro and toxicmetal contents of kidney bean and white bean seed samples

After 0.2 g seed samples were inginerated in a microwave device at 210ºC and 200 PSI pressure in 5 ml of concentrated HNO_3 and 2 ml of H_2O_2 (30% w/v), the volumes of the dissolved samples were completed to 20 ml with deionized water. Then, toxic metal concentrations in the samples were analyzed with Inductively Coupled Plasma Optical Emission spectroscopy (ICP-OES) (Tošic et al., 2015).

Statistical analysis

The JMP statistical program was used for the statistical analysis of results obtained. Statistically differences were determined by the analysis of variance (ANOVA) procedure in all data ($p<0.01$ and $p<0.05$) (Savaşlı et al., 2019).

RESULTS AND DISCUSSION

Macroelement contents of raw and germinated kidney bean and white bean seeds of different lengths are given in Table 1.

Table 1. Macroelement contents of kidney bean and white bean sprouted at different lengths (mg/kg)

Kidney bean	P	K	Ca	Mg
	(mg /kg)			
Control	3795.41±199.11 C*	11798.10±133.765 C	1090.84±91.937 A	760.52±6.734 C
1 cm	4553.65±20.80 B	14922.21±503.827 B	386.18±28.041 C	895.11±5.915 B
3 cm	3601.90±21.51 C	14218.37±284.575 B	462.17±10.329 C	1144.70±8.562 A
6 cm	6459.55±1.73 A	21475.71±15.409 A	728.43±1.144 B	1169.34±1.275 A
White bean	(mg /kg)			
Control	3532.46±145.15 C	13687.08±387.51 b	693.78±8.40 A	879.46±21.99 b
1 cm	2599.37±146.69 C	13866.62±418.28 b	413.91±32.95 C	1017.05±27.84 b
3 cm	5124.76±12.70 B	16216.31±543.72 b	460.78±28.13 BC	1290.88±176.30 ab
6 cm	8095.27±444.32 A	20078.57±220.59 a	557.49±16.09 B	1523.34±177.56 a

***** $p<0.01$

In general, raw and germinated kidney bean and bean samples were found to be rich in macroelement contents. The element established in the highest amounts in raw and germinated kidney bean and bean was K, followed by P, Mg and Ca in decreasing order. It can be seen that Ca amounts were the lowest abundant macro element in both legumes. K contents of kidney bean and white bean samples were found between 13687.08 (control) and 20078.57 mg/kg (6 cm) to 3601.90 (3 cm) and 6459.55 mg/kg (6 cm), respectively. In addition, Ca contents of kidney bean and white bean samples were found between 413.91 (1 cm) and 693.78 mg/kg (control) to 386.18 (1 cm) and 1090.84 mg/kg (control), respectively. The highest P (8095. 27 and 21475.71 mg/kg), K (20078.57 and 21475.71 mg/kg) and Mg (1523.34 and 1169.34 mg/kg) were determined in 6 cm length germinated kidney bean and white bean, respectively. It was observed that macro element contents of legumes (kidney bean and white bean), except Ca, increased with germination length. In general, P, Ca and Mg contents of kidney bean samples were higher than those of white bean samples.

The microelement contents of raw and germinated kidney bean and white bean samples of different lengths are presented in Table 2.

Table 2. Microelement contents of kidney bean and white bean sprouted at different lengths (mg/kg)

Kidney bean	Fe	Zn	Cu	Mn	B
	(mg /kg)				
Control	52.22±1.374 C*	17.75±0.346 B	1.63±0.065 C	18.86±0.031 A	10.76±0.100 C
1 cm	67.20±3.127 B	10.92±1.001 C	0.80±0.005 D	12.52±0.333 C	13.16±0.319 B
3 cm	42.80±0.199 D	3.93±0.013 D	2.93±0.016 B	10.73±0.039 D	9.78±0.059 D
6 cm	80.22±0.329 A	24.26±0.026 A	7.80±0.010 A	14.70±0.004 B	25.11±0.008 A
White bean	(mg /kg)				
Control	39.60±0.57 C*	14.85±0.29 A	4.59±0.16 A	7.99±0.29 b	10.48±0.59 B
1 cm	47.15±0.77 B	7.64±0.56 C	3.18±0.40 BC	9.66±0.36 b	10.08±0.73 B
3 cm	53.79±0.20 A	7.17±0.13 C	3.94±0.04 AB	12.11±1.13 b	15.41±1.95 AB
6 cm	55.51±1.48 A	10.57±0.52 B	2.75±0.13 C	25.50±5.87 a	20.65±1.58 A

***** $p<0.05$

The microelement found in the highest amounts in both legumes was Fe, followed by Mn, B, Zn and Cu in decreasing order. Depending on the germination lengths, the microelement contents of legume samples showed statistical differences when compared to the control ($p<0.01$, $p<0.05$). However, no statistical difference was found between the Mn and B of the 1 cm long germinated part of the kidney bean sample and the control group. Fe contents of kidney bean and white bean samples varied between 39.60 (control) and 55.51 (6 cm) and 52.22 (control) and 80.22 mg/kg (6 cm), respectively. The microelement detected in the lowest amounts in both legume samples was Cu. The Cu contents of kidney bean and kidney bean samples were assessed to be between 2.75 (6 cm) and 4.59 mg/kg (control) and 0.80 (1 cm) and 7.80 mg/kg (6 cm), respectively. The highest microelement contents of kidney bean and kidney bean samples except Cu were determined in the 6 cm long sample. In general, the microelement contents of white bean were higher than those of kidney bean. In addition, Zn, Cu and B contents of kidney bean and kidney bean germinated at 1 and 3 cm length were lower than those of control and 6 cm length germinated ones. A continuous partial increase was observed in the Fe contents of kidney bean and kidney bean with germination.

Toxic element contents of raw and germinated kidney bean and white bean seeds to different lengths are shown in Table 3.

Table 3. Toxic element contents of kidney bean and white bean sprouted at different lengths (mg/kg)

Kidney bean	Al	Ba	Cd	Co	Mo
	(mg /kg)				
Control	0.011±0.0003 A	0.015±0.007 A	0.031±0.0006 A	0.096±0.0006 B	0.021±0.0003 A
1 cm	0.008±0.003 B	0.0001±0.000 D	0.014±0.005 C	0.019±0.007 D	0.002±0.001 D
3 cm	0.007±0.003 BC	0.006±0.0003 B	0.017±0.0002 BC	0.140±0.0023 A	0.013±0.0002 B
6 cm	0.004±0.001 C	0.003±0.001 C	0.020±0.0018 B	0.084±0.0007 C	0.006±0.0002 C

Kidney bean	Cr	Ni	Pb	Se
Control	0.200±0.0021 A	0.068±0.0005 A	0.036±0.009 A	0.073±0.0002 D
1 cm	0.127±0.0001 C	0.006±0.0015 D	0.020±0.0001 B	0.129±0.0005 B
3 cm	0.141±0.0027 B	0.046±0.0001 B	0.021±0.0004 B	0.232±0.0003 A
6 cm	0.078±0.0015 D	0.029±0.0001 C	0.012±0.004 C	0.102±0.0009 C

*****p<0.01

Table 3. (continued)

White bean	Al	Ba	Cd	Co	Mo
	(mg /kg)				
Control	0.021±2.35 A*	0.011±8.65 A	0.038±0.0003 A	0.051±1.58 C	0.009±2.94 B
1 cm	0.015±1.93 B	0.008±1.72 B	0.036±0.0003 B	0.049±0.0003 C	0.007±4.41 B
3 cm	0.005±0.001 D	0.006±0.0005 C	0.020±0.0004 C	0.120±0.002 B	0.009±0.001 B
6 cm	0.008±0.0002 C	0.007±0.0001 B	0.013±0.0002 D	0.141±0.0007 A	0.014±0.0004 A

White bean	Cr	Ni	Pb	Se
Control	0.020±0.001 A	0.045±0.012 A	0.029±0.0001 A	0.018±0.005 C
1 cm	0.019±0.0002 B	0.017±0.003 D	0.016±0.006 B	6.021±0.001 A
3 cm	0.010±0.0002 C	0.038±0.001 C	0.010±0.002 C	0.010±0.0003 D
6 cm	0.018±0.0002 B	0.041±0.0002 B	0.025±0.0007 A	0.039±0.0006 B

*****p<0.05

Germination lengths showed differences in toxic element contents of both legumes and these differences were found to be statistically significant. In general, toxic element contents of germinated samples decreased compared to control. The toxic elements found in the highest amounts in both legumes were Cr and Ni. In addition, there was a partial increase in the Pb contents of the samples, but it remained below the tolerable limits. Cr contents of kidney beans and white beans were determined between

0.010 (3 cm) and 0.020 (control) to 0.078 (6 cm) and 0.200 mg/kg (control), respectively. While the Pb contents of kidney beans samples varied between 0.010 (3 cm) and 0.029 (control), the Pb contents of white beans samples were determined between 0.20 (1 cm) and 0.036 mg/kg (control), respectively. The fact that the heavy metal contents in kidney beans and white beans samples are below the limit values is an indication that the seeds were grown and processed in clean environments. It also creates confidence for human health. Bean, pea and chickpea and lentil contained 7, 7.36, 6.96 and 7.50 mg/100 g Fe, 3.0, 3.01, 3.54 and 3.73 Zn, 197, 96, 124 and 71 Ca and 250, 132, 155 and 129 mg/100 g Mg, respectively (Fachmann et al., 2000). The levels of minerals in legumes generally range between 1.5-5.0 µg/g Cu, 0.05-0.60 µg/g Cr g, 18.8-82.4 µg/g Fe g, 32.6-70.2 µg/g Zn, 2.7-45.8 µg/g Al g, 0.02-0.35 µg/g Ni g, 0.32-0.70 µg/g Pb (Cabrera et al., 2003). Beans of Indian origin contained 9-22 Cu, 108-150 Fe and 50-109 µg/g) Zn (Vadivel and Janardhanan, 2000). According to the results of this study, Fe concentrations in the samples are below the WHO standard value of 425.5 mg/kg (WHO, 2001). Macro and micro element contents of different legumes differed according to seed parts depending on seed types and treatments applied. When the amounts of macro and micro elements in seeds are compared with the results of studies conducted by Fachmann et al. (2000), Vadivel and Janardhanan (2000), Cabrera et al. (2003), Muche et al. (2023), our findings are high according to some element amounts. These differences may possibly be due to seed type, germination conditions and duration. It has been reported that increased activities of lipolytic enzymes during germination cause a decrease in the oil content of germinating seeds as they hydrolyze their oils into simpler products that can be used as energy sources for the developing embryo (Abioye et al., 2014). It has been reported that a gradual increase in mineral content with increasing germination hours means that these minerals are synthesized and those in combined form with some anti-nutrient components are made available through the germination process (Abioye et al., 2014). The increase in germination time caused a significant decrease in the total protein, fat and carbohydrate contents of legume seed flours, while it caused an increase in the non-protein nitrogen, ash, minerals (Na, K, Ca, P, Mg, Fe and Mn) and fiber contents (El-Adawy et al., 2003). As a result of germination of Africam Yam bean, calcium, ironi, magnesium and phosphorus contents chaged between 48.33 to 51.67 mg/100 g, 6.23 to 6.43 mg/100 g, 41.67 to 48.33 mg/100 g and 141.67 to 146.67 mg/100 g (Abioye et al., 2014). Köse et al. (2019) determined 6917.3 and 8349.1 mg/kg P, 16145.0 and 14730.0 mg/kg K, 1261.0 and 1275.5 Mg, 1468.0 and 1482.0 mg/kf Ca, 29.56 and 30.55 mg/kg Zn and 56.40 and 53.53 mg/kg in Yanice and Pınarlo bean genotypes.

Pearson correlation (r)

When the macro element contents of kidney bean samples of different sizes were examined. There was a significant and highly positive relationship ($p<0.05$, $r>0.70$) between the P content and the K content (r=0.951**). While no relationship was determined between the micro element contents of the kidney bean samples, positive relationships were determined between their heavy metal contents. Toxic metal contents of red mullet of different sizes show that there are significant and strong positive relationships between the Al contents of the samples and the Cr (r=0.966**) and Pb (r=0.964**) contents, between the Ba contents and the Mo (r=0.980**) and Ni (r=0.958**) contents, between the Mo content and the Ni content (r=0.985**) and between the Cr content and the Pb content (r=0.979**).

The Pearson correlation (r) between the macro (P, K, Ca and Mg) and micro (Fe, Zn, Cu, Mn and B) nutrient elements and heavy metal (Al, Ba, Cd, Co, Mo, Cr, Ni, Pb and Se) contents of white bean plants grown in different lengths are described. When the macro element contents of bean samples grown in different lengths were examined, it was revealed that there were significant and highly strong positive relationships ($p<0.05$, $r>0.70$) between the P content and K content (r=0.983**) and between the K content and Mg content (r=0.963**) of the bean samples. Similarly, while no relationship was determined between the micro element contents of bean samples, positive and negative relationships were determined between their heavy metal contents. The heavy metal contents of bean samples grown in different lengths show that; Significant and highly strong positive relationships were established between the Al contents of the samples and the Ba (r=0.967**) and As (r=0.974**) contents. In addition, significant and highly strong negative relationships were determined between the Cd amounts and Co quantities (r=-0.995**) and between the Ni amounts and Se (r=-0.973**) contents.

The Pearson correlation analysis was conducted to determine the strength and direction of the relationship between the nutrient or heavy metal contents of the kidney bean and bean samples, which are variables. With this Pearson correlation, the best fit line was tried to be drawn on the data of kidney bean and bean samples and nutrient content or kidney bean and bean samples and heavy metal content. Thus, the Pearson correlation coefficient (r) revealed how far all these data points were from the best fit line (Obilor and Amadi, 2018).

CONCLUSIONS

Potassium (K) was the most abundant element in both raw and germinated kidney bean and white bean samples, while calcium (Ca) was the least abundant macronutrient. Among the microelements, iron (Fe) was present in the highest amounts, followed by manganese (Mn), boron (B), zinc (Zn), and copper (Cu) in decreasing order. Overall, white beans contained higher microelement levels than kidney beans. Zinc, Cu, and B contents in kidney beans germinated to 1 and 3 cm were lower than in the control and 6 cm germinated samples. A gradual increase in Fe content was observed with germination in both legumes. Germination length also influenced the concentrations of toxic elements, with statistically significant differences between treatments; generally, toxic element contents decreased in germinated samples compared to controls.

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