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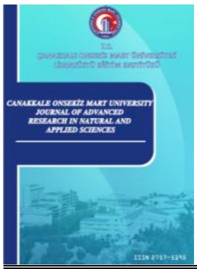

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Evaluating of Bean (*Phaseolus vulgaris* L.) Cultivars for Boron Efficient and Tolerant

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Research Article

Abstract − This study was carried out in a greenhouse in 2021 to determine the boron response of some bean cultivars at increasing levels of boron doses. The experiment established according to the complete randomized design with three replications was applied to 15 registered bean cultivars at 0, 5, and 10 mg B/kg doses using $Na_2B_8O_{13}.4H_2O$ (20.8% B) fertilizer. The above-ground organs were harvested during the blooming period, and the cultivars of B-efficient and B-tolerant were determined in the investigation of their dry weight, B content, and concentrations. Dry weights increased by 1% (Doruk, 10 mg B/kg) and 38% (Kantar-05, 5 mg B/kg) under B conditions, compared with the non-treatment group. Furthermore, insufficient B levels in the soil conditions were evaluated as B-efficient bean cultivars having a dry weight above the average, but cultivars having a dry weight below the average were named B-tolerant cultivars. As a result of the study, it was determined changing depends on the boron application of boron efficient (B-effect) and boron tolerant (B-tolerant) bean cultivars. Cihan, Güngör, Berrak, Elkoca-05, Özdemir, Kantar-05 and Arslan cultivars were confirmed as B-efficient, although Zülbiye, Sururbey, Doruk, Göksun, Karacabey, Özmen, Battallı and Zirve cultivars were determined as B-tolerant cultivars. As a result of the study, it was determined that the efficient boron cultivars were Zülbiye, Zirve and Battallı, while the boron tolerant cultivars Cihan and Arslan.

Keywords *– Bean, B-efficient, boron activity, boron content, B-tolerant*

1. Introduction

Boron (B) is a micronutrient element that affects vegetative and generative growth in plants and, is necessary for plant development. Although the effect of boron on plant growth is still discussed in the literature, it is known that deficiency or excess B intake damages plant growth. (Dhaliwal et al., 2021). Boron deficiency and toxicity prevent the metabolic function of plants and cause damage. Determining the response of plants to boron is important as it affects yield and quality when boron deficiency and toxicity levels are narrow to each other. (Brdar-Jokanovic, 2020). These responses show variation between cultivars and even within-cultivar genotypes (Schnurbusch et al., 2010). It has been determined in the studies that some of the plants develop well in soils that do not contain enough boron and give quality products, while some show average growth in soils with rich boron (Punchana et al., 2012). B efficient plants show optimum plant development by taking enough B to meet their needs in soils that do not contain enough boron and provide optimum yield. Still, B-resistant or tolerant plants are growing at optimum growth by reducing B uptake as much as possible in soils having rich boron (Torun et al., 2006). Harmankaya et al. (2008) reported that wide genetic variation in the study investigated efficiencies of boron utilization in different bean genotypes. Because plants give different morphological and physiological responses to boron deficiency or toxicity, for this reason, this study aimed to determine the B-efficient and B-tolerant cultivars by considering the boron efficiencies of bean cultivars, which have the main legume plant.

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2. Materials and Methods

2.1. Plant Material

This research used 15 registered cultivars of the bean (*Phaseolus vulgaris* L) as plant material (Table 1). The seed of recorded cultivars, being different properties in terms of physiological, morphological characteristics, and resistance to disease and pests, were obtained from the research institutes where they were registered.

Table 1
Proprietary bean cultivars, sources, and some properties

No	Cultivars	Growth Form	Seed Color	Hundred Seed Weight,g	Registrant Institution
1	Zülbiye	Dwarf	White	49.5-51.5	Black Sea Agricultural Research Institute
2	Arslan	Dwarf	White	27.6-62.1	Mersin Commodity Exchange Seed Research Industry and Trade Joint Stock Company
3	Battallı	Dwarf	beige	26.5-34.7	Mersin Commodity Exchange Seed Research Industry and Trade Joint Stock Company
4	Berrak	Dwarf	Beige	25.7-33.7	Field Crops Central Research Institute
5	Cihan	Dwarf	White	42.1-46.0	Aegean Agricultural Research Institute
6	Doruk	Semi dwarf	White	37.0-39.0	Safgen Seed Agricultural Products Industry and Trade Limited Company
7	Elkoca-05	Dwarf	White	42.4-46.0	Atatürk University, Faculty of Agriculture, Department of Field Crops
8	Güngör	Semi dwarf	White	60.0-65.3	East Mediterranean Transitional Zone Agrıcultural Research of Institute
9	Göksun	Dwarf	White	53.5-55	East Mediterranean Transitional Zone Agrıcultural Research of Institute
10	Kantar-05	Dwarf	Dark red	32.434.3	Atatürk University, Faculty of Agriculture, Department of Field Crops
11	Özmen	Dwarf	White	24.4-34.4	Avesa Agriculture Food and Livestock Limited Company
12	Sururbey	Semi dwarf	White	42.2-54.5	Mersin Commodity Exchange Seed Research Industry and Trade Joint Stock Company
13	Zirve	Drawf	White	37.3-39.7	Taşpınar Agriculture Food and Livestock Limited Company
14	Özdemir	Semi dwarf	Dark red	42.9-51.3	Mersin Commodity Exchange Seed Research Industry and Trade Joint Stock Company
15	Karacabe y	Dwarf	White	38.6-42.0	Transitional Zone Agrıcultural Research of Institute

2.2. Soil Material Used in the Experiment

The soil used in the experiment had a slightly alkaline pH, no salinity problem, high lime content, and low organic matter content. In the soil, Ca (1150-3500 mg/kg), Mg (160-780 mg/kg), K (109-289 mg/kg), Cu (0.2-0.25 mg/kg) and Mn (1-5 mg/kg) were at sufficient level although P (<15 mg/kg), Fe (<2.5 mg/kg), Zn (<0.5 mg/kg) and B (<0.5 mg/kg) were at insufficient level (Gezgin et al., 2002) (Table 2).

Table 2
Some physical and chemical properties of soil tested in greenhouse

Parameters	Results
pH (1:2.5 soil: water)	7.58
EC (1:5 soil: water) (µS/cm)	52
%	
$CaCO_3$	36.6
Organic matter	1.64
Clay	37.60
Silt	20.66
Sand	41.74
Texture class	Clay Loam
Field capacity	22.5
1N NH_4AOC Extractable, mg/kg	
Ca	1622
Mg	231
K	222
Na	9
mg/kg	
P	16.4
İnorganic N (NH_4+NO_3-N)	12.0
Fe	0.70
Zn	0.16
Mn	2.20
Cu	0.80
B	**0.36**

2.3. Setting up and carried out the trial

The trial was carried out in the Computer Controlled Research Greenhouse Department of Soil Science and Plant Nutrition, Faculty of Agriculture, Selcuk University. We provided the greenhouses climate, 25±3 ºC temperature, 1750±50 kcal/m^2 of solar radiation, and 60±10% relative humidity during the experiment. In the greenhouse experiment conducted according to the complete randomized design with three replications, the pots were put through a 4 mm sieve, and 2000 g of soil was added based on the kiln dry weight of the pots.

Essential fertilization was made to ensure the plant's normal development due to nutrient deficiencies in the soil material. As basic fertilization, 200 mg N/kg ($Ca(NO_3)_2 .4H_2O$) in solution, 80 mg P/kg (TSP), 300 mg K/kg (K_2SO_4), 10 mg Fe/kg (Sequestrene), 2.5 mg of Zn/kg ($ZnSO_4.7H_2O$) was applied.

B doses applied in the trial were given in below.

Control (B_0) = 0 mg/kg (Control)

B_5 = 5 mg B/kg

B_{10} = 10 mg B/kg

It was made by using $Na_2B_8O_{13}.4H_2O$ (20.8% B) fertilizer.

In the experiment, which consisted of 135 pots with 15 bean cultivars x 3 boron doses x 3 replications used, each pot was sown with eight seeds, and thinning was made so that four plants after germination. During the experiment, we irrigated the plants with deionized water at the field capacity and changed the pots in the greenhouse every 4-5 days. This trial was harvested separately according to the flowering time of each variety. Harvesting was done by cutting the above-ground parts.

2.3 Taking plant samples and preparing them for analysis

After the samples brought to the laboratory were washed with tap water, they were washed with pure water 0.2 N HCl solution, distilled water, deionized water, and dried with coarse filter papers. Then the plant sample was placed in a paper bag. The samples were dried in a circulating air-drying cabinet at 70 ºC until they reached a constant weight, and after their dry weight was determined, they were ground in an agate mill. Then, we weighed about 0.2 g of the dried samples. After adding 5 ml of concentrated HNO_3 and 2 ml of H_2O_2 (30 % w/v), the transferred microwave device (Cem MARSXpress; CEM Corp; Matthews. NC. USA) under high pressure (200 PSI) was dissolved. A blank and certified reference materials were added to the 40-cell microwave set to ensure the reliability of the analysis. The volumes of the dissolved samples were made up of 20 ml of deionized water. The obtained percolates were filtered through blue-banded filter paper. The B concentrations in the percolator were determined in the ICP-OES (Inductively Coupled Plasma-Optical Emission Spectrometer). These values were controlled using reference plant material from the National Institute of Standards and Technology (NIST, Gaithersburg, MD, USA). B contents were calculated by multiplying the dry weights of the cultivars with the B concentrations. Since the sufficiency limit values (20-75 mg B/kg) specified by Jones et al. (1991) are taken into account in the calculation of boron efficiency, it is sufficient for all varieties in B_5 application according to control and B10 application. For this reason, B_5 application was used instead of B_{10} to calculate boron efficiency. In addition, the following equation [(1.1)] was used to determine efficient B (Graham, 1984).

$$\text{B efficient, \%} = (100) \text{x dry weight (B0)} \ / \text{dry weight (B5)} \tag{1.1}$$

2.4. Statistical Evaluation

The data obtained within the scope of the experiment were subjected to statistical evaluation with the JMP 7 statistical package program by the complete randomized design. The variance analysis was performed to determine the differences between the dry weight, boron contents, and concentrations of the samples belonging to each application group. The average values of the applications determined the differences and were grouped according to the "Student's t-test" of significance.

3. Results and Discussion

3.1. B Concentration of Bean Cultivars

The average values of boron concentration of bean cultivars under B treatments are given in Table 3. In addition, according to the studies of variance analysis performed to determine the effects of applications on boron concentration, cultivars (C), boron treatments (BAD) and interactions (C x BAD int.) were found to be statistically significant ($p< 0.01$) (Table1). The C x BAD interaction was found to be a change in the response of boron concentrations of bean cultivars of above-ground organs to the treatment of boron.- (Figure 1). Although the reactions during the flowering period of the cultivars used in the study varied depending on the cultivar and boron doses, we determined that the toxicity symptoms were more apparent at 10 mg B/kg. (Figure 1).

Table 3

Analysis of variance on dry weight, B content, B concentration of cultivars

Sources of Variance	**Mean of Squares**		
	Dry Weight	B Content	B Concentration
Total	330.9	1764185	5366714
Cultivar (C)	105.3**	52142**	592528**
Boron Application Doses (BAD)	114.2**	1644200**	107064421**
C X BAD int.	68.6**	64921**	489685**
Error	42.8	2921	4.84

** $p< 0.01$

The B concentration of dry bean cultivars increased from 4 (Zülbiye) to 8 (Güngör) times at 5 mg B/kg and between 14 (Özmen) and 15 (Battallı) times at 10 mg B/kg when compared to the control group (Table 3).

Figure 1. The responses of different bean cultivars (a: Battallı, b: Battallı c: Battallı d: Doruk, e: Doruk, f: Doruk, g: Arslan, h: Arslan, j: Arslan, k: Berrak, m: Berrak, n: Berrak cultivars) to boron doses (0 mg B/kg dose, 5 mg B/kg dose, 10 mg B/kg dose)

As shown in Table 4, the highest B concentration was obtained at 10 mg B/kg dose with 291.43 mg /kg depending on the average boron dose. In addition to, it was followed by decreasing concentration of 5 mg B/kg (135.78 mg/kg) and 0 mg B/kg (22.20 mg/kg). Jones et al. (1991) reported that the B concentration required to be adequate in bean leaves during the blooming period was about 20-75 mg B/kg. Then, above boron amounts of this level caused toxicity symptoms, which is in line with our study. In research; the non-treatment group was shown that insufficient B in Özmen, Sururbey, and Karacabey bean cultivars. Treatments of 5 and 10 mg B/kg caused B toxicity in all cultivars. Although the B concentration of the above-ground organs for bean cultivars was higher at approximately >75 mg B/kg in 5 mg B/kg, toxicity symptoms were less than 10 mg B/kg. (Figure 1). This situation could be derived from being B-efficient or B-tolerant cultivars. Akoğlu (2013) investigated the effects of increasing doses of B (0, 8, 16, and 24 mg B/kg) applications on boron concentrations of bean cultivars (Eyri Oturak, Ferasetsiz, Şeker bean, and Local genotype) in the greenhouse. Subsequently, increasing boron treatment caused B toxicity symptoms in cultivars.

Table 4

Effects of B treatments on B concentration (mg B/kg) of registered dry bean cultivars

	B concentration (mg B/kg)			
Cultivars	Control (B_0)	B_5	B_{10}	Average
Zülbiye	29.11 q	102.19 p	330.58 c	153.96 de
Arslan	21.40 qr	140.71 jkl	289.98 d	150.70 e
Battallı	24.97 qr	146.79 ıj	373.98 b	181.91 b
Berrak	22.69 qr	145.79 ıjk	323.54 c	164.00 c
Cihan	21.91 qr	133.96 klm	288.32 d	148.06 e
Doruk	25.35 qr	153.14 hı	295.67 d	158.05 cd
Elkoca – 05	21.15 qr	145.37 ıjk	235.08 fg	133.87 f
Güngör	21.42 qr	164.75 h	391.59 a	192.59 a
Göksun	21.25 qr	146.08 ıjk	326.14 c	164.49 c
Kantar – 05	23.25 qr	117.99 no	231.19 g	124.14 g
Özmen	16.64 r	113.20 op	230.61 g	120.15 g
Sururbey	18.50 qr	132.62 lm	259.25 e	136.79 f
Zirve	24.57 qr	130.21 lmn	244.77 f	133.18 f
Özdemir	21.06 qr	121.88 mno	265.35 e	136.10 f
Karacabey	19.67 qr	142.07 ı-l	285.38 d	149.04 e
Average	22.20 C	135.78 B	291.43 A	--

In variance analysis was determined to be statistically significant in the differences between the responses of the cultivars to boron. Therefore, we obtained the maximum B concentration from Güngör cultivar with 192.59 mg/kg and established the minimum concentration from Özdemir (136.10 mg/kg), Elkoca-05 (133.87 mg/kg), Zirve (133.18 mg/kg), Kantar-05 (124.14 mg/kg) and Özmen (120.15 mg/kg) cultivars (Table 3). However, previous study reported that boron concentrations varied between cultivars and species in all legumes and bean cultivars (Harmankaya, 2008).

3.2. B Content in Registered Bean Cultivars

Boron treatment on bean cultivars was changed of B content and it was determined significant statistically at the 1% level cultivars, boron doses, and their interaction (Table 5). The boron content increased along with increasing in boron doses applied to the cultivars, and we determined the maximum content at the highest B_{10} dose. The significant interaction could prove that boron treatments influenced the development of cultivars and plant dry weight, which showed this effect difference between boron application dose and cultivars. The B contents of the cultivars increased from 4 (Zülbiye) to 10 (Güngör) times at the B_5 dose and from 12 (Elkoca-05) to 14 (Zülbiye) times at the B_{10} dose when compared to the control group (Table 5).

Table 5

Effect of B treatments on B content (µg B/pot) of registered dry bean cultivars

	B Content (µg B/pot)			
Cultivars	Control (B_0)	B_5	B_{10}	Average
Zülbiye	163.08 p	581.02 o	2354.40 a	1032.84 ab
Arslan	114.60 p	1083.31 fgh	1686.93 c	961.62 bc
Battallı	112.51 p	780.63 lmn	1746.01 c	879.72 de
Berrak	107.78 p	914.71 ı-l	2027. 23 b	1016.58 b
Cihan	111.55 p	832.93 j-m	1498.17 d	814.22 efg
Doruk	147.84 p	1001.09 ghı	1733.13 c	960.67 bc
Elkoca – 05	98.62 r	916.91 ı-l	1136.36 fg	717.30 hı
Güngör	111.89 p	1102.35 fgh	2121.71 b	1111.99 a
Göksun	108.52 p	835.98 j-m	1723.26 c	889.25 cde
Kantar – 05	116.30 p	810.54 klm	1313.92 e	746.92 gh
Özmen	84.03 s	661.17 no	1222.07 ef	655.76 ı
Sururbey	99.73 r	757.90 mn	1521.72 d	793.12 fgh
Zirve	148.09 p	921.53 ıjk	1528.89 d	866.17 ef
Özdemir	94.45 r	737.28 mn	1502.63 d	778.12 gh
Karacabey	116.30 p	968.78 hıj	1774.35 c	953.30 bcd
Average	115.72 C	860.41 B	1659.39 A	

As seen in Table 5, the maximum B concentration was obtained at 1659.39 µgB/pot from the B_{10} dose within an treatment of boron doses. Then, it was followed by decreasing amounts of B_5 (860.41 µgB/pot) and 0 mg B/kg (115.72 µg B/pot) doses. The highest B content was determined in Güngör (1111.99 µg/pot), and the lowest B content was determined in Özmen (655 µg/pot) within cultivars means. However, Ceyhan et al. (2006) stated that using six dry bean cultivars in their field trial, the B contents of the types were diverse due to the difference in B concentrations and the cultivar yield variance. In particular, he reported that cereals have a wide content of B levels on plant species and cultivars of the same species (Topal et al., 2002). Therefore, the genetic diversity of plants could cause this difference.

3.3. Dry Weight and Efficiency B in Registered Bean Cultivars

As seen in Table 6, boron treatments on bean cultivars impact dry weight and boron efficiency (%). It was determined that the effects of variety, B application doses and interactions on the dry weight of boron applications of variance analysis were statistically significant at the 1% level. This critical interaction confirms that the effect of boron applications on the dry weight of plants varies depending on the cultivars. For example, the dry weight of the cultivars increased from 2% (Zülbiye) to 37% (Kantar-05) at 5 mg B/kg dose and between 1% (Doruk) and 38% (Zülbiye) at 10 mg B/kg dose when compared to the control group (Table 6).

The maximum dry weight was obtained from the Zirve cultivar with 12.92 g, while the minimum value was determined from the Battallı (9.66 g) cultivar among the averages of the cultivars. When considering boron application doses, boron applied increased in dry weight according to the control group, except at 10 mgB/kg, which was a high boron dose. Some research reported that the yield in wheat (Taban and Erdal, 2000), chickpeas (Hakkoymaz et al., 2006; Ceyhan et al., 2007), and bean (Sadiq and Mohammed, 2022) increased with boron applications when compared to the control group. However, high-dose B applications decreased the yields of plants. It is thought that the decrease in the dry weight of plants may result from the reduction of biomass production due to photosynthetic pigment loss or inhibition of photosynthesis due to damage to photosynthetic membranes (Gunes et al., 2006; Sahin, 2009).

Table 6

The effects of B application on dry weight and B activity of registered dry bean cultivars

	Dry weight (g/pot)				B Efficient (%)
Cultivars	Control (B_0)	B_5	B_{10}	Average	B_0/B_5*100
Zülbiye	11.19 f-k	11.39 f-k	15.39 ab	12.27 ab	98
Arslan	10.71 g-n	14.22 a	11.64 e-j	12.58 a	75
Battallı	9.01 o	10.64 g-n	9.34 mno	9.66 g	85
Berrak	9.49 l-o	12.55 c-f	12.53 c-f	11.52 b-e	76
Cihan	10.18 j-o	12.44 c-f	10.39 ı-o	11.03 def	82
Doruk	11.62 e-j	13.07 b-e	11.72 e-ı	12.14 abc	89
Elkoca – 05	9.33 no	12.62 c-f	9.66 l-o	10.54 f	74
Güngör	10.45 ı-o	13.38 bcd	10.84 g-l	12.56 b-e	78
Göksun	10.19 j-o	11.45 f-k	10.57 j-o	10.73 ef	89
Kantar – 05	10.01 k-o	13.76 bc	11.38 f-k	11.72 bcd	73
Özmen	10.09 k-o	11.68 e-ı	10.59 h-n	10.79 ef	86
Sururbey	10.82 g-m	11.43 f-k	11.74 e-ı	11.33 c-f	95
Zirve	12.06 d-h	14.20 ab	12.49 c-f	12.92 a	85
Özdemir	8.98 o	12.10 d-g	11.33 f-k	10.80 ef	74
Karacabey	11.87 e-ı	13.64 bc	12.44 c-f	12.65 a	87
Average	10.40 C	12.65 A	11.39 B		83

Boron-efficient (B-efficient) plants are identified as the plants that show the optimum growth and produce the yield at a decent level in the soil, having boron content insufficient by uptaking as B their needs. However, boron-tolerant (B-tolerant) plants are the plants that show the optimum growth in the soil with boron at sufficient or excess levels by decreasing B uptake (Punchana et al., 2012). Wang et al., (2005) reported that the efficiency of plants at low and high boron concentrations was calculated. In the same study, the researcher determined B-efficient, above 85% boron efficiency, while B-tolerant, below this value. Güneş et al., (2006) reported other boron efficiency evaluation methods. In this method, B activity was made by considering the average of the cultivars. Then, the types below the average were evaluated as B-tolerant cultivar and had above the average as B-efficient cultivar. Therefore, we selected the latter method to determine boron efficiency because the average B efficiency of the cultivars was 83%. Thus, our study considered that B-tolerant cultivars were below the average while B-efficient cultivars were above the mean.

Boron efficiency on cultivars showed varies from 70% (Arslan) to 98% (Zülbiye) (Table 5). It was determined that B-tolerant cultivars were Cihan, Güngör, Berrak, Elkoca-05, Özdemir, Kantar-05 and Arslan cultivars having below 83%. However, B-efficient cultivars were Zülbiye, Sururbey, Doruk, Göksun, Karacabey, Özmen, Battallı and Zirve cultivars above the mean. B-efficient cultivars produced more dry weight than other cultivars under low boron conditions. Also, B-tolerant types showed optimum growth than different cultivars in high or excess boron conditions. Some previous studies reported that B-efficient was a criterion for determining significant cultivars of bean (Harmankaya et al., 2008), wheat (Taban and Erdal, 2000; Hamurcu and Gezgin, 2007), barley (Atalay et al., 2003), and maize (Güneş and Alpaslan, 2000). It was determined that B efficiency varied depending on the increasing dry weight of the plant with boron treatment (0, 5, and 10 mgB/kg) to the soil. There are notable variations in response to boron of cultivars in the studies on beans (Paul et al., 1988; Huang and Graham, 1990; Nable and Paull, 1991; Ceyhan et al., 2006; Hamurcu and Gezgin, 2007). Also, it has been stated that other cultivated plants, except for beans, show wide genetic variation in response to boron (Taban and Erdal, 2000; Topal et al., 2002; Torun et al., 2021). According to their findings, the primary reason for this could be caused that plants show various responses as physiological and morphological to B toxicity and deficiency (Dordas et al., 2000; Dordas and Brown, 2001).

4. Conclusion

The limit range between B deficiency and toxicity in plants is very narrow. However, it is observed that yield and quality decrease significantly in case of B deficiency and toxicity. We applied increasing doses of boron (0, 5, and 10 mgB/kg) to B-efficient and B-tolerant registered dry bean cultivars to increase yield and quality. As a result, we comprehended a wide variation among cultivars against B activity. We could state that the cultivars determined as B-efficient had dry weight values above average in soil conditions at toxic boron levels. In contrast, the cultivars with low B-activity occurred with less dry weight. We determined that the most critical difference between the cultivars having low and high B activity was the dry weight values and B contents at the low B application. We determined among the cultivars used in the study that Zülbiye, Sururbey, Doruk, Göksun, Karacabey, Özmen, Battallı, and Zirve cultivars had the highest B activity, in other words, the B-efficient cultivars. However, Cihan, Güngör, Berrak, Elkoca-05, Özdemir, Kantar-05, and Arslan cultivars showed the lowest B efficiency. In other words, we stated a B-tolerant cultivar for these cultivars. In bean agriculture under soils conditions having sufficient or excess boron content, using Zülbiye, Sururbey, Doruk, Göksun, Karacabey, Özmen, Battallı, and Zirve cultivars could provide more yield per unit area by reducing the yield losses due to excess B in the soil. In addition, there is extremely important regarding both economy and using less labor to use B-efficient plant cultivars in agricultural production.

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Author Contributions

Fatma Gökmen Yılmaz: Designed the experiments and performed the statistical analysis and wrote the manuscript.

Ayşegül Korkmaz: Applied the experiment, analyzed the plant sample, and wrote the manuscript.

Sait Gezgin: Designed the experiment and investigation.

Conflicts of Interest

The authors declare no conflict of interest.

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