



Effects of Coating with Different Doses of Boron Compound on Germination Parameters of Cotton Seeds

Mustafa Yaşar^{1,a,*}, Nurettin Baran^{1,b}, Yusuf Güzel Demiray^{2,c}, Remzi Ekinci^{3,d}, Yaşar Karadağ^{1,e}

¹Muş Alparslan University, Faculty of Applied Sciences, Department of Plant Production and Technologies, Muş, Türkiye

²GAP International Agricultural Research and Training Center Directorate, Diyarbakır, Türkiye

³Dicle University, Faculty of Agriculture, Department of Field Crops, Diyarbakır, Türkiye

*Corresponding author

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ABSTRACT

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In this study, seeds of cotton plants were coated with different boron concentrations and their effect on seedling growth was investigated. The experiment was carried out at Muş Alparslan University, Faculty of Applied Sciences, Department of Plant Production and Technologies, under laboratory and greenhouse conditions, according to the Randomized Plots Trial Design with 4 in laboratory and 3 in greenhouse replications, respectively. In the study, Di-sodium tetraborate decahydrate ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) ($M=381.37 \text{ g mol}^{-1}$) with a minimum purity of 99% boron compound have been used in different doses; 0 (control), 250, 500, 750, 1000, 1250, 1500, 1750, 2000, 2250 and 2500 mg kg^{-1} . According to the results of the regression analysis, it was determined that as the boron dose in the application increased, the root length was positively correlated and other parameters were negatively correlated. In terms of the parameters examined, 250 mg kg^{-1} Kaolin+liquid K-Humate (15% HA) 400 ml kg^{-1} (U3) boron application under laboratory conditions showed better results, and 1000 ml kg^{-1} Kaolin+liquid boron application under greenhouse conditions. It was determined that the application of K-Humate (15% HA) 400 ml kg^{-1} (U6) promoted growth. In addition, it was found that the results were confirmed in the grouping of the relationships between applications and parameters with heat map clustering and PCA graphs. As a result, it was determined that the toxic range of boron applications applied to the seed in the cotton plant was very narrow, and that it promoted seedling development and root elongation in the U6 application, but it was concluded that the study should also be studied in field trials in order to make more decisive decisions.

^a mustafa.yasar@alparslan.edu.tr

^b <https://orcid.org/0000-0001-9348-7978>

^b n.baran@alparslan.edu.tr ^c <https://orcid.org/0000-0003-2212-3274>

^c yusufguzel.demiray@tarimorman.gov.tr

^d <https://orcid.org/0000-0002-4113-5855>

^d renzi.ekinci@dicle.edu.tr ^e <https://orcid.org/0000-0003-4165-6631>

^e y.karadag@alparslan.edu.tr

^e <https://orcid.org/0000-0002-0523-9470>



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Introduction

Cotton (*Gossypium hirsutum* L.) is a fiber plant that prefers warm climates with sufficient humidity and is cultivated in nearly 100 countries around the world. It is cultivated around the world, especially in China, India, USA, Brazil, some African countries and certain regions of the Middle East (Özyiğit and Gözükırmızı, 2009; Datta et al., 2020; FAOSTAT, 2021).

It is used as a raw material in many industrial areas and it is as main source of plant protein, edible oil, textile, feed and paper industries (Özyiğit, 2009; Munir et al., 2020; Zhang, 2019; Yaşar, 2022). In addition, cottonseed oil has found a lot of use in biodiesel production in recent years (Sharma et al., 2020; Sundar and Udayakumar, 2020).

As in all agricultural production, the main purpose of cotton production is to increase the quality of cotton fiber and obtain more and better quality products per unit area (Yaşar et al., 2017). Therefore, cotton breeding studies

focused on cultivars that are resistant to biotic and abiotic stress conditions and have rapid growth ability as well as yield and quality parameters (Gao et al., 2016; Rauf et al., 2019; Chohan et al., 2020; Hocaoglu Özyiğit et al., 2020). However, in addition to the effects caused by global warming, the increase in average temperatures throughout the world and the irregularities in precipitation regimes create serious problems in agricultural production (Malcolm et al., 2012; Gustafson et al., 2014; Yadav and Sharma, 2016; Li et al., 2020; Yaşar and Sezgin, 2022; Yaşar ve Yetişsin, 2023).

One of the important conditions for success in plant production is the use of quality seeds. It covers all of the applications made and developed from seed technology to sowing to harvest, in order to increase the quality of the seed (Aslan and Pirli, 2018).

Seed is becoming a strategic input with increasing importance and commercial value day by day. For this reason, every single seed produced and imported in Turkey should be evaluated in the best way possible and ways to make the best use of seeds in seed production should be investigated.

Due to reasons such as not being homogeneous, the genetic structure of the seed and being affected by environmental factors, there are problems in germination and emergence rates. In order to eliminate these problems and bring the seeds to the desired quality level, some extra applications are made to the seeds (Gray, 1989; Duman ve Gökçöl, 2018). In order to improve seed quality, seed coating technologies come to the fore, as do many other applications especially before sowing. Seed coating is done to increase the germination and emergence performance of the seeds, to facilitate the sowing, to give the seeds an identity, to shape the seeds and to protect the viability of the seeds against diseases and damages.

A sufficient amount of the boron (B) micronutrient is needed for better plant growth and productivity in plant breeding. However, boron (B) deficiency and toxic limits have a very narrow range. Boron, as a micronutrient, has different roles in many stages of plant development. Boron uptake by plants is related to the plant's water uptake and its movement in the xylem, and differs between plant species. Since boron deficiency affects the growing parts of the plants, growth slows down and with the increase of the deficiency, the growing parts die and growth stops. Therefore, boron is of vital importance in plant cultivation. The lack of sufficient research on boron limits its use in agriculture (Barut et al., 2018).

The aim of this study was to see the effect of the boron (B) micronutrient, which is of vital importance for plants, on the germination and growth parameters of cotton seed, and to contribute to the most effective use of the seed, which is important for the country's economy.

Material and Method

This research was carried out at Muş Alparslan University, Faculty of Applied Sciences, Department of Plant Production and Technologies, under laboratory and greenhouse conditions. The BA 440 cotton variety registered by Pro Gen Seed A.S. has been used in the trial.

In laboratory trial, the randomized plots were designed according to the experimental design with 4 replications. For the germination studies, 48 pieces of 18 cm glass jars

and 160 g and 2 mm sterilized sand in plastic lids with closures were used. 20 seeds with different boron (B) doses were used in each jar. The prepared jar containers were placed in a germination cabinet adjusted to 14 hours of light, 10 hours of darkness, 65% humidity, 8 hours at 30 °C and 16 hours at 20°C temperature conditions according to ISTA rules. Jars were kept in the germination cabinet for 12 days (ISTA, 2020).

In greenhouse trial, the experiment was designed according to the randomized plot design with 3 replications. 3 seedling trays (total of 108 seeds) in which 3 seeds could be planted were used as 3 replications for each application. Sterilized by washing 36 seedling trays (42 mmx40mm mouth diameter; 40 mm deep) were used in the greenhouse experiment. Seedling trays were filled with a mixture prepared to be 50% sand + 50% soil and appropriate amount of water was given. 3 seeds were planted into of each cavity of the seedling tray and the sowing depth was adjusted as 2 cm.

Coating process in seeds: A boiler made of rubber material was used for the electric motor, which was reduced to 40 rpm. Di-sodium tetraborate decahydrate with a minimum of 99% purity ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) ($M=381.37 \text{ g mol}^{-1}$) boron compound, were prepared in different doses; 0 (control), 250, 500, 750, 1000, 1250, 1500, 1750, 2000, 2250 and 2500 mg kg^{-1} (Table 1). Kaolin+liquid K-Humate (15% HA) 400 ml kg^{-1} was used as filling coating raw material in pelletizing the seeds in the coating (Table 1).

Spraying apparatus and a hand compressor were used to spray the paste powders, liquid and plant nutrients onto the seed. When light drying occurred, the pelletizing material, which was prepared in powder form, was left to the rolling orbits of the seeds in the boiler. Thus, while the seeds, which are surrounded by sticky liquid in the cauldron, rotate with the coating material, it is ensured that the seeds are surrounded by pulling the pelletizing powder around them. After the first wrapping process was completed, the seeds were subjected to a light drying process and then the adhesive was sprayed again. After the seeds were covered with a sufficient amount of pellet coating material, the liquid boron compound was sprayed. Finally, after some moisture was removed from the seeds with hot air, the seeds were left to dry at room temperature. In the seed coating study, the average thickness of the material used in the coating of the seeds was 0.1415 mm, the percentage of coating was 40%, the amount of coating was 3.40 g and the weight of 100 seeds was 11.9 g.

Table 1. Boron Application Doses and Coating Filling Material Ratios

Applications	Boron Doses (mg kg^{-1})	Coating filler material (ml kg^{-1})
U1 (Control)	0	0
U2	0	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U3	250 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U4	500 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U5	750 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U6	1000 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U7	1250 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U8	1500 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U9	1750 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U10	2000 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U11	2250 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}
U12	2500 $\text{mg kg}^{-1}\text{B}$	Kaolin+liquid K-Humat (% 15 HA) 400 ml kg^{-1}

Measurements in the Seed Coating Study

Observations and measurements in the seed coating study were carried out in the laboratories of Muş Alparslan University, Faculty of Applied Sciences, Department of Plant Production and Technologies. The following observations were made with 4 replications:

- 100 seed weight (100sw) (g): 3 different 100 seeds randomly selected from each application and control were weighed separately on a 0.0001 gr precision scale, and the average was taken.
- Coating Thickness (CT) (mm): 3 different 10 seeds randomly selected from each application and control were cut separately, and the average of the coating thickness was measured with a digital caliper.
- Amount of Coating (AC) (g toh-1): Calculated by subtracting 100 sw Control (g) values from 100 sw applications (g) measured for each application.

$$AC (g) = 100sw \text{ application (g)} - 100sw \text{ control (g)}$$

- Percent Coverage (PC) (%): The ratio of 100 sw application (g) to 100 sw Control (gr) value measured for each application was calculated as %.

$$AC (\%) = (100 \text{ sw administration (g)} - 100 \text{ sw Control (g)}) / 100 \text{ sw Control (g)} \times 100$$

- Dissolution time of the coated seeds in water (sec): When 10 randomly selected seeds from each application were dropped into the water, the dissolution time of the coating material was measured in seconds with a chronometer and averaged (Doğan et al., 2003).

Observations and Measurements Made in Germination Tests in The Laboratory

In the study, observations taken for 12 days from the 1st day were made at the same hours, and according to the study of Soltani et al. (2012), seeds with a root length of 2 mm were considered germinated. The germination percentage (%) was calculated from the total number of germinated seeds on the 12th day (Scott, 1989). At the same time, 10 plants were taken as samples from the seeds germinated in each jar at the end of the 12th day, and the fresh weights and root-shoot lengths of these samples were measured according to Saboora and Kiarostami (2006). Afterwards, shoots and roots were kept in an oven at 70°C for 12 hours, and their dry weights were measured by weighing them on a precision (0.0001 mg) balance. Shoot and root lengths were measured with a caliper, and the vigor index was calculated with the data obtained (Abdul-Baki and Anderson, 1970; Yaşar, 2023).

Vigor index; was used for the following equation.

$$VI: GP\% \times SL(mm)$$

GP: germination percentage,
SL: seedling length

GP (Germination percentage %): According to the equation by Fang et al. (2006).

$$ÇY\% = n/N \times 100$$

n: seeds germinated on the last count day,
N: total number of seeds used in the test.

MGT (Mean Germination Time): The equation in the study of Ellis and Roberts (1981) was used.

$$OÇS = \sum D.N/n$$

n: the number of germinated seeds on the counting days, N: the total number of seeds used in the test,
D: the number of days counted from the beginning of germination.

SVI (Seed Viability Index): Seed viability index was calculated according to the formula below.

$$TCI = (ÇY \times SL) / 100$$

Rs: Germination rate (%) and SL: Mean shoot length (mm).

Observations and Measurements Made in The Greenhouse Experiment

In the experiment, observations were taken at the same time, with two-day intervals starting on the 4th day and ending on the 28th day. According to Soltani et al. (2012), the seeds whose shoots came to the surface of the soil were considered to have emerged. On the last count day, Rs (germination rate) and GP (Germination percentage) were calculated.

Statistical Analysis

The data obtained for each trait examined were subjected to the F test with the help of the JMP 5.0.1 package statistics package, and the averages were grouped according to the EGF test. A regression analysis was performed in terms of application doses and properties examined. In addition, heatmap clustering (ClustVis) was carried out with the help of a heat map in order to visualize, distinguish, and determine the correlation between the examined traits and application parameters.

Findings and Discussion

Laboratory studies

In the study, according to the results of the analysis of the data obtained under laboratory conditions, it has been determined that there are significant differences at the $p \leq 0.01$ or $p \leq 0.05$ level between the germination test parameters, application and parameters examined by coating cotton seeds with different doses of boron. Except for the mean germination time, all other parameters were found to be significant at the $p \leq 0.01$ level. As a result of this, it is shown that the applications have different effects on germination parameters (Table 2).

Table 2. Variance Analysis of Germination Parameters

Germination Parameters SD	Variation Sources					
	Application	Replication	Error	Total	CV (%)	EGF 0.05
	11	3	33	47		
GP	3957.729**	17.0625	278.1875	4252.9792	3.87	8.49
VI	51073.32**	89.615	457.376	51620.308	2.18	10.9
MGT	0.905192*	0.0456372	0.7932177	1.7440464	2.18	10.9
SVI	4589.724**	2.4373	127.7763	4719.9377	2.65	5.75
RE	4875**	4.1667	270.8333	5150	4.77	8.38
RL	27381.332**	43.105	234.307	27658.745	2.08	7.79
SL	1124.2184**	4.0942	57.8214	1186.134	1.34	3.87
SFW	0.19955011**	0.0017677	0.0307927	0.23211053	3.21	0.08
RDW	0.00052984**	0.0000045	0.0002209	0.00075527	2.82	0.007
SFW	5.3319419**	0.0121095	0.3845551	5.7286065	2.3	0.315
SDW	0.01468926**	0.0000756	0.0038278	0.01859263	2.74	0.031

**p<0,01; *P<0,05, GP: Germination percentage, VI: Vigor indeksi, MGT: Mean germination time, SVI: Seed variability index, RE: rate emergence, RL: radicle length, SL: shoot length, RFW: root fresh weight, RDW: root dry weight, SFW: shoot fresh weight, SDW: shoot dry weight.

Table 3. Average Data of Germination Parameters in Different Boron Dose Application

Applications	Çimlendirme Parametreleri										
	GP	VI	MGT	SVI	RE	RL	SL	SFW	RDW	SFW	SDW
U1	91 ^A	208 ^A	7.2 ^D	97.8 ^A	78 ^A	121.5 ^F	107.2 ^A	0.907 ^{CD}	0.084 ^D	5.266 ^A	0.411 ^A
U2	81 ^{BD}	182 ^D	7.6 ^{AB}	84.8 ^B	65 ^C	124.8 ^{EF}	104.4 ^B	1.074 ^A	0.092 ^{BC}	5.020 ^{BC}	0.408 ^A
U3	83 ^{BC}	242 ^A	7.5 ^{AC}	78.3 ^C	60 ^D	194.5 ^A	92.6 ^F	0.934 ^{CD}	0.096 ^A	4.207 ^G	0.369 ^{EF}
U4	77 ^D	188 ^C	7.2 ^D	76.2 ^{CD}	65 ^C	145.8 ^B	98.7 ^{DE}	0.926 ^{CD}	0.094 ^{AB}	4.492 ^F	0.384 ^{CD}
U5	80 ^{CD}	182 ^D	7.4 ^{BD}	73.6 ^{DE}	69 ^{BC}	134.5 ^C	92.6 ^F	0.849 ^E	0.092 ^{AB}	4.185 ^G	0.375 ^{DF}
U6	71 ^E	161 ^E	7.3 ^{CD}	66 ^F	65 ^C	131.7 ^{CD}	93.1 ^F	0.942 ^{BC}	0.096 ^A	4.478 ^F	0.401 ^{AB}
U7	85 ^B	179 ^D	7.3 ^{CD}	78.1 ^C	73 ^B	125.9 ^E	93.4 ^F	0.939 ^{BC}	0.091 ^{BC}	4.524 ^F	0.405 ^{AB}
U8	63 ^F	128 ^H	7.4 ^{AD}	65.4 ^F	48 ^{FG}	105.8 ^H	96.9 ^E	0.947 ^{BC}	0.088 ^{CD}	4.477 ^F	0.360 ^F
U9	61 ^F	123 ^H	7.5 ^{AB}	60.9 ^G	49 ^{EG}	101.4 ^I	99.4 ^D	1.078 ^A	0.092 ^B	5.101 ^B	0.414 ^A
U10	70 ^E	153 ^F	7.6 ^{AB}	71 ^E	46 ^G	116.6 ^G	101.8 ^C	0.915 ^{CD}	0.087 ^D	4.699 ^E	0.377 ^{CE}
U11	70 ^E	146 ^G	7.5 ^{AB}	72.7 ^E	53 ^E	102.2 ^{HI}	104.0 ^B	0.979 ^B	0.091 ^{BC}	4.942 ^{CD}	0.405 ^{AB}
U12	65 ^F	146 ^G	7.5 ^{AB}	63.9 ^F	51 ^{EF}	127.9 ^{DE}	97.8 ^{DE}	0.8949 ^D	0.091 ^{BC}	4.812 ^{DE}	0.391 ^{BC}

GP: Germination percentage, VI: Vigor indeksi, MGT: Mean germination time, SVI: Seed variability index, RE: rate emergence, RL: radicle length, SL: shoot length, RFW: root fresh weight, RDW: root dry weight, SFW: shoot fresh weight, SDW: shoot dry weight

Germination percentage (GP) varied between 61-91% on average. While the highest germination rate was obtained from the Control (U1) application with 91%, the lowest germination percentage was obtained with 61% (U9) (1750 mg kg⁻¹ B, Kaolin+liquid K-Humate (15% HA) 400 mg kg⁻¹) application. It was determined that there was no statistical difference between U8 (63%) and U12 (65%) and therefore they were in the same letter group (Table 3).

Regression analysis results of germination percentage applications: (r² = 88.643939-2.1022727*Applications). It was determined that there is a negative relationship between B concentration and germination percentage. In cotton seeds, the germination percentage decreases as the B dose increases.

Vigor Index (VI) values varied between 123-242%. While the highest vigor index was obtained with a 242% (U3-250 mg kg⁻¹ B- Kaolin + liquid K-Humate (15% HA) 400 mg kg⁻¹) application, the lowest vigor index was 123% (U9-1750 mg kg⁻¹ B), Kaolin+liquid K-Humate (15% HA) and it was obtained from the application of 400 mg kg⁻¹. It was determined that there was no statistical difference between them and U8 (128), so they were in the same letter group (Table 3). At a low boron dose (250 mg kg⁻¹ B), it increased the vigor index compared to the control.

Regression analysis results of vigor index applications: (r² = 218.08158- 7.3501603*Applications). It is seen that the relationship between B dose and vigor index is

negative. It was determined that the increase in boron dose applied to cotton seeds caused a decrease in the vigor index. However, 250 mg kg⁻¹ B, Kaolin+liquid K-Humate (15% HA) and 400 ml kg⁻¹ application were not statistically different from the control group, so they were found in the same group (Table 2). In similar studies, Hacıyusufoglu et al. (2015) stated that there may be decreases in the vigor index in parallel with the germination rate. However, B application did not affect seed germination but increased seedling viability (Muhmood et al., 2014). In the study conducted, the vigor index obtained from the application of (U3) 250 mg kg⁻¹ B, Kaolin + liquid K-Humate (15% HA) 400 ml kg⁻¹ was found to be higher than the control group.

The mean germination time (MGT) values varied between 7.2-7.6 days. While the average mean germination time was obtained from the U4 application with the shortest 7.2 days, the longest mean germination time was obtained from the U10 application (2000 mg kg⁻¹ B, Kaolin + liquid K-Humate (15% HA) 400 ml kg⁻¹), U2 application. It was determined that there was no statistical difference between U9, U11 and U12, therefore they were in the same group (Table 3).

The regression analysis results for mean germination time applications: (r²= 7.2876418 + 0.0212809 ×Applications). It was determined that the relationship between B dose and average germination time was

positive. It was determined that the increase in boron dose applied to cotton seeds caused an increase in the mean germination time. In similar studies, Saba et al. (2012) reported that wheat seeds coated with B solutions shortened the germination time and mean germination time by 50%, but did not affect the final germination. However, in the study, the mean germination time was prolonged in B applications compared to the control group. This is thought to be due to the kaolin application used. Because kaolin dissolution rate in water is slow, it is thought to have affected this time. Parallel to the same study, similar results were obtained in our study as it did not affect the final germination.

Seed viability index (SVI) average values varied between 60.9 and 97.8%. While the highest seed viability index was obtained from the control (U1) application with 97.8%, the lowest seed viability index was obtained from the (U9) application with 60.9% (1750 mg kg⁻¹ B, Kaolin + liquid K-Humate (15%) HA) 400 ml kg⁻¹) (Table 3).

Regression analysis results of seed viability index applications: ($r^2 = 88,215006 - 2.1783782 * \text{Applications}$). It was determined that the relationship between B dose seed and viability index was negative. It was determined that the increase in boron dose applied to cotton seeds caused a decrease in the average seed viability index.

Rate emergence (RE) average values varied between 46-78%. While the highest rate of emergence was obtained from the control (U1) application with 78%, the lowest rate emergence was obtained from 61.46% (U10) application (2000 mg kg⁻¹ B, Kaolin + liquid K-Humate (15%) HA) 400 ml kg⁻¹) (Table 3).

It has been observed that the increase in boron application doses has a reducing effect on the rate emergence. Boron application has a positive effect on some plant seeds and a negative effect on some other plant seeds. In addition, the amount of boron applied can have a positive or negative effect on rate of emergence. In similar studies, they determined that 0.2 ppm B application had a positive effect on the rate of emergence of the BR1 sugar beet variety (Ebrahim Pour Mokhtari et al., 2022).

Root length (RL) average values varied between 101.4-194.5 mm. The longest root length was obtained with 194.5 mm from (U3) application (250 mg kg⁻¹ B, Kaolin + liquid K-Humate (15%) HA) 400 ml kg⁻¹), while the shortest root length was obtained from (U9) application with 101.4 mm (1750 mg kg⁻¹ B, Kaolin+liquid K-Humate (15%) HA) 400 ml kg⁻¹)(Table 3). It is seen that more than 1500 mg kg⁻¹ B, Kaolin + liquid K-Humate 15% HA 400 ml kg⁻¹ B applied to cotton seeds (U8) causes a root length reduction effect.

The regression analysis results of root length applications: ($r^2 = 150,26775 - 3,4707411 * \text{Applications}$). It was determined that the relationship between B dose and root length was positive. It is observed that root length increases in parallel with the increase in boron dose applied to cotton seeds.

It is very important that the root length of the plants develop well in adverse conditions and/or be faster and longer than normal with some interventions in order for the plants to reach the plant nutrients in the soil as soon as possible and benefit from them. It is thought that the appropriate dose for promoting root length in cotton plants may be 250 mg kg⁻¹ B.

Shoot length (SL) values varied between 92.6-107.2 mm. While the highest shoot length was obtained from the control (U1) application with 107.2, the lowest shoot length was obtained from (U3) application (250 mg kg⁻¹ B, Kaolin+liquid K-Humate (15%) HA) 400 ml kg⁻¹) with 92.6. It was determined that they were in the same group statistically with U5 (92.6), U6 (93.1) and U7 (93.4), so there was no difference between them (Table 3). Shoot length is an important criterion that determines resistance of seeds to adverse environmental conditions. Although it was observed that the shoot length decreased as the boron concentration increased, it was observed that the shoot length was close to the control group at the doses after U10 and U11 applications. In similar studies, Esin and Latif (2008) reported that an increase of 47.06% was recorded in the stem length of the soybean plant at the end of the 30th day, compared to the control group in the group with excess boron.

Root fresh weight (RFW) average values varied between 0.849 and 1.078 mg. While the highest root fresh weight was obtained from (U9) and (U2) applications with 1.078 mg, the lowest root wet weight was obtained from (U5) application (750 mg kg⁻¹ B, Kaolin + liquid K-Humate (15%) HA) 400 ml kg⁻¹) (Table 3). In parallel with the increase in the amount of boron applied to cotton seeds, it was determined that root fresh weights decreased in other applications except U5 and U12 application (Table 3).

Root dry weight (RDW) average values varied between 0.084-0.096 mg. While the highest root dry weight was obtained with 0.096 mg (U3) application (250 mg kg⁻¹ B, Kaolin + liquid K-Humate 15% HA) 400 ml kg⁻¹), the lowest root dry weight was 0.084 mg. It was determined that (U1) was obtained from the control application, the application with the highest value (U3) and the application U6 (0.096) were not statistically different, so they were in the same group. In addition, it was determined that the control application with the lowest value (U1) and the application U10 (0.087) were not statistically different, so they were in the same group (Table 3). It was determined that the applied B concentrations caused an increase in root dry weight, depending on the increase. In a similar study by Cömert and Çelik (2017), they reported that there was a negative correlation with plant dry weight as B concentration increased. In the study, it was determined that the dry weight of the root decreased in parallel with the increasing doses of the B concentration, and it is in accordance with the results of above-mentioned researchers.

Shoot fresh weight (SFW) values varied between 4.47-5.27 mg. The highest shoot fresh weight was obtained from the control (U1) application with 5.27 mg, while the lowest shoot fresh weight was obtained from the application (U8) (1500 mg kg⁻¹ B, Kaolin + liquid K-Humate 15% HA) 400 ml kg⁻¹). In the study, although there was a decrease in shoot fresh weight as B concentration increased (U9), positive values were obtained close to the control group in shoot fresh weight in increasing applications after 1750 mg kg⁻¹ B application. In similar studies, Akçam Oluk and Latif (2008) reported that boron excess increases the root and shoot fresh and dry weights in their study, in which they examined the effects of boron excess on the growth and development of soybean. The findings of the researchers support the findings of the study.

Shoot dry weight (SDW) average values varied between 0.36-0.41 mg. While the highest shoot dry weight was obtained from 0.41 mg control (U1) application, the lowest shoot dry weight was obtained with 0.36 (U8) application (1500 mg kg⁻¹ B, Kaolin + liquid K-Humate (15% HA) 400 ml kg⁻¹). The highest shoot dry weight was obtained from (U1) control, U2 (0.41) and U9 (0.41) applications (Table 3). In the study, although there were decreases in shoot dry weight as boron (B) concentration increased (U11), it was determined that shoot dry weight had positive values close to the control group in the 2250 mg kg⁻¹ B application.

Heatmap Clustering and PCA Graph of Germination Parameters

In the research, a heat map graph was created to interpret the relationships between parameters, applications and similarity between parameters. It was determined that two different main groups were formed regarding the parameters examined, and subgroups of each main group were formed. In the 1st main group; MGT (Mean Germination Time), SL (Shoot Length), RFW (Root Fresh Weight), SFW (Shoot Fresh Weight) and SDW (Shoot Dry Weight) parameters were included. In the second main group; Water dissolution rate (sec), Seed coating thickness (mm), Seed coating percentage (%), Seed coating amount (g), 100 seed weight (g), VI (Vigor Index), SVI (Seed Viability Index), RL (Root Length (mm)), RDW (Root Dry Weight (g)) and SFW (Shoot Fresh Weight (g)) (Figure 1). According to the heatmap clustering plot, the subgroups show a high level of correlation (Figure 1 and 2).

It was determined that the effects of the application doses in the same group of different applications applied to cotton seeds coated with different doses of boron compound were similar. In the first main group, while there are U2, U8, U9, U10, U11 and U12 applications, in the second main group,; there are U1, U3, U4, U5, U6 and U7.

The applications in the first main group are divided into 2 subgroups: the first subgroup, U2, U9 and U11. While in the 2nd subgroup, it was determined to be U8, U10 and U12. The applications in the second main group are divided into 2 subgroups, and the first subgroup; U3, U4, U5 and U6. While in the second subgroup, U1 and U7 are being observed (Figure 1).

According to the PCA graph, it was determined that the results of the heatmap clustering were confirmed and that the parameters were in 2 different main groups (Cluster A and Cluster B). Also, the variation between parameters PC1; 38.3% and PC2; 23.9% explained it. A high level of correlation between the parameters in main group A (SL (Shoot Length) and SFW (Shoot Fresh Weight)) and in main group B (VI (Vigor Index) and SVI (Seed Viability Index)) is visually presented (Figure 2).

Germination Parameters Examined in Greenhouse Conditions

According to the variance analysis results of the data obtained under greenhouse conditions in the study, it was determined that there were statistically significant differences at p≤0.05 level according to the germination parameters, germination percentage and germination rate according to the treatments (Table 4). Boron is an element that is required by plants and its toxicity limits are very close to each other (Brown et al., 2002).

It is thought that U3 (250 mg kg⁻¹ Kaolin+liquid K-Humate (15% HA) 400 ml kg⁻¹) stands out in terms of the properties examined and that it promotes root elongation, it will be advantageous in the early period of plant growth and boron can be applied at this dose.

The germination percentage varied between 75-98 and the highest germination percentage was obtained from U6, the lowest germination percentage was obtained from U12 application, and the germination rate varied between 53-92 and the highest germination rate was obtained from U6 and the lowest germination rate was obtained from U1 application. When the investigated properties are evaluated, it is thought that the application of U6 (1000 mg kg⁻¹ Kaolin + liquid K-Humate (15% HA) 400 ml kg⁻¹) can be applied under greenhouse conditions (Table 5).

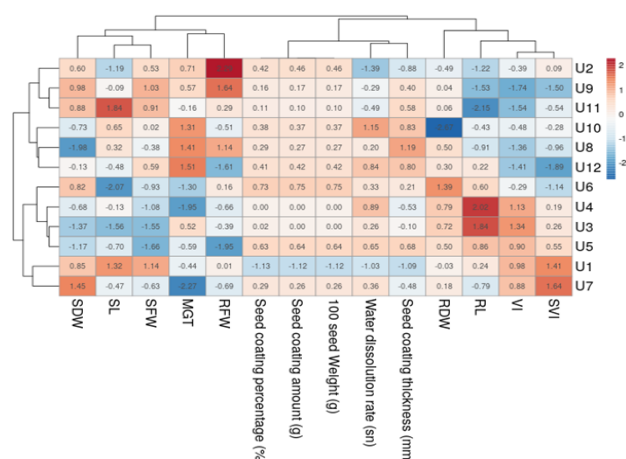


Figure 1. Application and parameter relations with the heat map of the investigated parameters

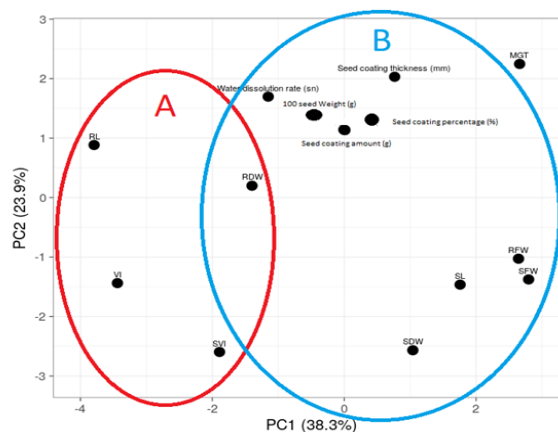


Figure 2. Relationships between the examined features and the resulting clusters

Table 4. Variance Analysis Germination Parameters in Greenhouse Conditions

Variation Source	Germination Parameters	
	GP	RE
Application	1366.012 *	6388.5378 *
Replication	12.4204	0.3045
Error	154.8059	172.8066
Total	1533.2383	6561.6389
CV (%)	2.90	3.54
EGF 0.05	9.31	9.84

GP: Germination percentage, RE: Rate emergence *p≤0.01

Table 5. Averages of Germination Parameters in Greenhouse Conditions

Application	Germination Parameters	
	GP	RE
U1	86 ^{EF}	53 ^F
U2	90 ^{DE}	56 ^F
U3	95 ^{AC}	90 ^{AB}
U4	92 ^{CD}	88 ^{BC}
U5	93 ^{BD}	89 ^{AC}
U6	98 ^A	92 ^A
U7	94 ^{AC}	85 ^{CD}
U8	92 ^{CD}	84 ^{CD}
U9	84 ^F	82 ^D
U10	95 ^{AC}	87 ^{BC}
U11	97 ^{AB}	81 ^D
U12	75 ^G	61 ^E

GP: Germination percentage, RE: Rate emergence

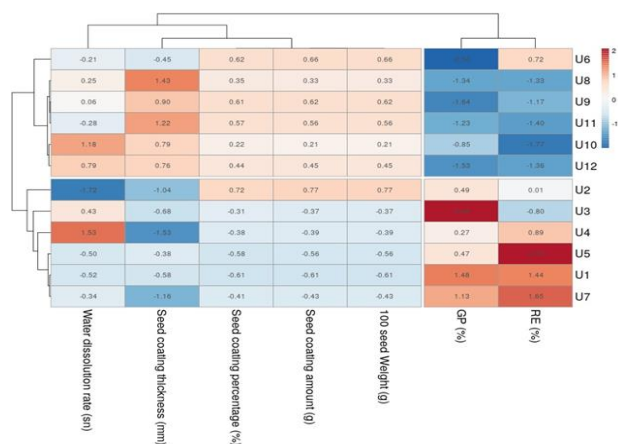


Figure 3. Heatmap relationships of the parameters studied in the greenhouse

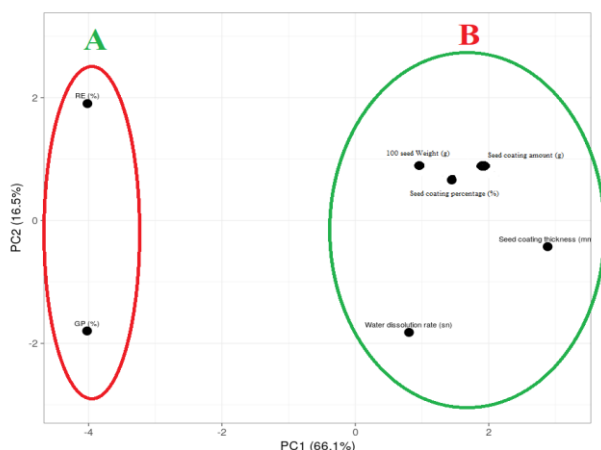


Figure 4. Relationships between the examined traits and the resulting clusters

Heatmap Clustering and PCA Graph of Germination Parameters in Greenhouse Conditions

In the research, two different main groups were formed regarding the parameters examined and subgroups of each main group were formed. In the 1st main group; Water dissolution rate (sec), Seed coating thickness (mm), Seed coating percentage (%), Seed coating amount (g) and 100 seed Weight (g) parameters were included. In the second

main group; RE (Germination Rate (%)) and GP (Germination Percentage (%)) were included (Figure3 and 4).

It has been determined that the level of relationship between the different applications applied to cotton seeds and the applications in the same group is important. Applications consisted of two main groups and subgroups were formed. In the first main group; While U6, U8, U9, U10, U11 and U12 applications are included, in the second main group; there are U1, U2, U3, U4, U5 and U7.

The applications in the first main group are divided into 2 subgroups: the first subgroup, U6 and in the 2nd subgroup, U8, U9, U10, U11 and U12. Applications in the second main group are divided into 2 subgroups, and the first subgroup; it is U2 and in the 2nd subgroup, U1, U3, U4, U5 and U7(Figure 3).

According to the PCA graph, it was determined that the parameters are in 2 different main groups (Cluster A and Cluster B). Besides these, the variation between parameters PC1; 66.1% and PC2; 16.5% is explained. In main group A, RE (Rate Emergence (%)) and GP (Germination Percentage (%)) and in main group B, Water dissolution rate (s), Seed coating thickness (mm), Seed coating percentage (%), Seed coating amount (g) and 100 seed Weight (g) parameters were visually found to be highly correlated (Figure 4).

Conclusion

In cotton plant cultivation, agricultural practices are very important for homogeneous germination and the successful emergence of seeds. In this study, seeds of ton plants were coated with different boron concentrations. and their effect on seedling growth was investigated. In the seed coating study, the average thickness of the material used in the coating was determined to be 0.1415 mm, the percentage of coating was 40%, the amount of coating was 3.40 g and the weight of 100 seeds was 11.9 g. It has been determined that it has a positive effect on strong root and plant growth, as well as showing faster and more complete plant emergence with seed coating. According to the results of the regression analysis, it was determined that as the boron dose in the application increased, the root length was positively correlated and the other parameters were negatively correlated. In laboratory conditions, it was determined that 250 mg kg⁻¹ and Kaolin+liquid K-Humate (15% HA) 400 ml kg⁻¹ (U3) application came to the fore and promoted root elongation. In greenhouse conditions, it was concluded that the most appropriate boron dose for germination percentage and rate was 1000 mg kg⁻¹ and Kaolin+liquid K-Humate (15% HA) 400 ml kg⁻¹ (U6). In the study, significant positive and negative effects were found between the applied Boron doses and the properties examined. In addition, it was found that the results of the heatmap and PCA graphs were confirmed in grouping the relationships between applications and parameters. As a result, it was determined that the toxic range of boron applications applied to the seed in the cotton plant was very narrow, and that it promoted seedling development and root elongation in the U6 application, but it was concluded that the study should also be studied in field trials in order to make more decisive decisions.

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