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SOCIEDADE DE CIÊNCIAS
AGRÁRIAS DE PORTUGAL

Productivity, quality and composition of soybean seeds in storage as a function of boron doses at different phenological stages

Produtividade, qualidade e composição de sementes de soja armazenadas em função de doses de boro em diferentes estágios fenológicos

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https://doi.org/10.19084/rca.20022
Received/recebido: 2020.05.07
Accepted/aceite: 2020.12.15

ABSTRACT

Soybeans is one of the crops with significant impact on global food supply, so it's important to study those relevant factors affecting its production and even the development of its seeds in the field. The macro and micronutrients are important, and continuously soybeans is concerned, boron is extremely important for biological nitrogen fixation, and participating in the physiological seed formation processes. Since the nutrient is not very mobile in the plant, some authors recommend foliar spraying to improve seed productivity and quality. Dose and timing of applications are important factors inner decisions related to crop production and management. Therefore, the goal of this work was to evaluate effects of times and doses of soil boron application on soybean quality, and its influence on the physiological characteristics after storage. Two experiments were conducted at the State University of Goiás and Federal University of Pelotas. The first in the field, conducted in a randomized block in a 5 x 6 factorial scheme, with five stages (V0, V3, V6, V9 and R1) of boron application and six different doses (0, 1, 2, 3, 4 and 5 kg ha^{-1}) of it, four replications, evaluating leaf boron content, chlorophyll index and seed yield in kg ha^{-1}. The second phase was contrived in the laboratory aimed to evaluate the physiological quality and nutrient titration of seeds after 6 months of storage, with a completely randomized design in a 5 x 6 factorial arrangement with four replications as in the field with 5 stages and 6 doses. The variables analyzed were: germination, root length, shoot length, growth rate index, root development ratio, shoot development ratio and concentration of phosphorus, nitrogen, potassium and boron. The reproductive stage showed the best averages for seed quality variables and concentration nutrients, whereas for doses, the intermediate levels showed positive results for most of the tests used in the study. However, there isn't justification for fertilization with boron to maintain the stored seeds quality.

Keywords: *Glycine max*, Micronutrient, Physiological Quality, Storage, Seed

RESUMO

A soja é uma das culturas de maior impacto no abastecimento global de alimentos, por isso é importante estudar os fatores relevantes que afetam sua produção e até mesmo o desenvolvimento de suas sementes no campo. Os macro e micronutrientes são importantes, especificadamente em relação à soja, o boro é extremamente importante para a fixação biológica do nitrogênio, participando dos processos fisiológicos de formação das sementes. No entanto, o nutriente não é muito móvel nas plantas, sendo assim alguns autores recomendam a pulverização foliar para melhorar a produtividade e a qualidade das sementes. A dosagem e o momento das aplicações são fatores importantes nas decisões internas relacionadas à produção e ao manejo da safra. Portanto, o objetivo deste trabalho foi avaliar os efeitos de tempos e doses de aplicação de boro no solo sobre a qualidade da soja, e sua influência nas características fisiológicas após o armazenamento. Dois experimentos foram conduzidos um na Universidade Estadual de Goiás e outro na Universidade Federal de Pelotas. A primeira fase foi em campo, conduzida em blocos casualizados em esquema fatorial 5 x 6, com cinco estágios (V0, V3, V6, V9 e R1) de aplicação de boro e seis diferentes doses (0, 1, 2, 3, 4 e 5 kg ha^{-1}) dela, quatro repetições, foram avaliandos o teor de boro nas folhas, o índice de clorofila e a produtividade de sementes em kg ha^{-1}. A segunda fase foi elaborada em laboratório com o objetivo de avaliar a qualidade fisiológica e titulação de nutrientes das sementes após 6 meses de armazenamento, em delineamento inteiramente casualizado em esquema fatorial 5 x 6 com quatro repetições, como no campo com 5 estágios e 6 doses. As variáveis analisadas foram: germinação, comprimento da raiz, comprimento da parte aérea, índice de taxa de crescimento, razão de desenvolvimento da raiz, razão de desenvolvimento da parte aérea e concentração de fósforo, nitrogênio, potássio e boro. O estádio reprodutivo apresentou as melhores médias para as variáveis de qualidade das sementes e concentração dos nutriente na semente, enquanto que para as doses, os níveis intermediários apresentaram resultados positivos para a maioria dos testes utilizados no estudo. No entanto, não há justificativa para a fertilização com boro para manter a qualidade das sementes armazenadas.

Palavras-chave: *Glycine max*, Micronutriente, Qualidade Fisiológica, Armazenamento, Semente

INTRODUCTION

Soybeans are a vital crop in human and animal nutrition, contributing to global food security. Its worldwide production in 2018/19 was 359 million tons and a planted area of 125 million hectares (USDA, 2019). By covering the largest area cultivated among agricultural crops, it becomes one of the *commodities* that most demands seeds, pesticides and fertilizers in agriculture; this consumption is distributed over 236,000 rural properties in Brazil (IBGE, 2017).

In order to obtain high quality seeds, one must consider the interaction between the genetic, physical, physiological and health attributes, which can be estimated through the tests of vigor. The effects of seed strength are expressed during the emergence of seedlings in the field by the uniform establishment of the stand, rapid initial development of the plants and consequently in the productive performance (Ruppin *et al*., 2019). The ability to maintain the strength and viability of these seeds is achieved through the optimum conditioning (Alencar *et al*., 2009).

The process of seed classification and storage time has a direct influence on the germination rate and vigor of soybean seeds (Coelho *et al*., 2019). Conservation integrity and effectiveness of seed storage techniques are mainly caused by the genotype of the stored cultivar, as well as when exposed to temperatures below 8°C or above 35°C and relative humidity above 80%, these factors accelerate the deterioration process (Schons *et al*., 2018).

Another factor that reflects the obtaining of high quality seeds is adequate nutrition, the lack of which may jeopardize the yield and quality of soybean seeds, being more evident when the macro and micronutrient deficiency occurs simultaneously in the field (Hansel and Oliveira, 2016). One of the micronutrients important in the formation of the seed and in the biological fixation of nitrogen in soybeans is boron (B), an element that activates the starch phosphorylase enzyme, carries out its synthesis and allocates reserve substances into the seeds (Ceretta *et al*., 2005). The increased accumulation of reserves results in heavy, oblong, high-strength seeds. Soybean seeds from larger sieves result in higher germination rates and longer storage periods than smaller seeds (Coelho *et al*., 2019).

The low mobility of B in the vegetable vascular system makes it difficult to supply the plant, through foliar fertilization, and as a result, the use of soluble formulations via the soil is preferable, since they are easily dissolved and are rapidly available for absorption (Malavolta, 2006). The most widely used formulations are fully soluble or refined solid materials, such as sodium pentaborate ($Na_2B_4O_7.5H_2O$), borax ($Na_2B_4O_7.10H_2O$), sodium tetraborate ($Na_2B_{10}O_{16}.10H_2O$), solubor ($Na_2B_8O_{13}.4H_2O$) and boric acid (H_3BO_3) (Saleem *et al*., 2011).

Various studies relate B with the highest levels of seed and grain production; however, few are those that demonstrate their attributions for plant vegetative growth. (Shireen *et al.*, 2018). Both the deficiency and the toxicity of B lead to a reduction in the photosynthetic rate, interfering with the plant's vegetative performance. (Bolaños *et al.*, 2004). There are several experimental results demonstrating a large variability of responses to the application of boron (Ceretta *et al.*, 2005; Gomes *et al.*, 2017); however, maintaining the appropriate levels of B available for soybeans deserves special attention to ensure and improve the grain and seed yields of the crop.

To this end, it is important to carry out work relating fertilization, plant nutrition, physiological quality of seeds and storage, in order to increase the efficiency of soybean production and in particular to establish guidelines for the application of this micronutrient. Accordingly, this work aims to evaluate the effects of the doses of boron applied via the soil in different phenological stages of soybeans under the productivity and quality of the seeds after storage.

MATERIAL AND METHODS

The first experiment was carried out in the field in the 2018/19 crop as agricultural zone in the experimental area of the State University of Goiás, Ipameri University Unit, located in the municipality of Ipameri-GO, whit geographical coordinates of 17º43'04" S, 48º08'43' W and altitude 794 m. The region's climate, according to the Köppen-Geiger classification, is defined as a tropical climate (Aw) appearing in a dry season in the winter (Cardoso *et al.*, 2014). The soil in the experimental area was classified as a distractive Yellow-Red Latossolo (Embrapa, 2018).

The chemical attributes and granulometric analysis of the soil were determined before the installation of the experiment, according to the methodology proposed by Ribeiro *et al.* (1999), with the following chemical attributes, in the layer 0.0 - 0.20 m: 2.9 mg dm^{-3} P (Mehlich); 15,3 g dm^{-3} from M.O.; 5,1 pH ($CaCl_2$); K, Ca, Mg, H+Al = 0,18; 1.9; 0,5 and 3,0 cmolc dm^{-3}, respectively 46,2 % of base saturation and the B content 0,12 mg dm^{-3} being below the critical level for soybean cultivation. The physical attributes were clay (270 g dm^{-3}), silt (140 g dm^{-3}) and sand (590 g dm^{-3}).

The experimental outline used in the field was the randomized blocks, in a factorial scheme of 5 x 6, with five levels of boron application: stage V0 (at the time of sowing); Stage V3 (third knot, second trifle open); Stage V6 (fifth fully open trifoliate sheet); stage V9 (eighth fully open trifoliate), and stage R1 (beginning of flowering, up to 50% of plants with one flower) and six levels of boron doses (0, 1, 2, 3, 4 and 5 kg ha^{-1}), with four repetitions. Boron was applied via soil using boric acid 17% as a boron source. Each parcel was made up of six rows of five meters in length with spacing of 0.45 m among themselves, and 15 plants per linear meter, making up a total area of 13.5 m^2. The usable area was composed of the three central lines, where 1 m was excluded at the ends of each line, making a total of 4 m^2 of usable area.

The planting system used was no-till, on the corn straw residue. The planting fertilization was carried out according to the soil analysis using 70 kg ha^{-1} P_2O_5 and 100 kg ha^{-1} K_2O, according to Ribeiro *et al.* (1999) recommendation, broadcasting before seeding. The seeding took place in October 2018 with the help of a treated fertilizer drill, with horizontal honeycomb disks covering seven rows, for the distribution of seeds of the Monsoy 7110 IPRO variety.

The boron application was carried out manually, on the planting row, next to the soybean plants, and according to the crop development/treatment stages (V0, V3, V6, V9 and R1), according to the phenological scale of Ritchie *et al.* (1994). The other necessary cultural treatments, such as the application of fungicide and insecticide during the conduct of the experiment, were applied mechanically using a treated sprayer.

For the analyses referring to the physiological quality of the seeds in the laboratory, the experimental outline was randomly, in a 5 x 6 factorial scheme with four repetitions, the five stages of boron application being: V0, V3, V6, V9, R1 and the six boron doses: 0, 1, 2, 3, 4 and 5 kg ha^{-1}, totaling 30 treatments. Before carrying out the laboratory analysis, the seeds obtained from the field experiment were stored for 6 months in a laboratory environment with a temperature of 25-30 °C, in order to maintain the quality of seeds in storage, following the recommendations indicated for maintaining the quality of soybean seeds according to Schons *et al*. (2018).

Evaluation of agronomic variables

Leaf boron concentration: 10 leaves have been collected from each parcel, with the 3rd leaf being from the apex on the main rod, with the peculiarity of each parcel during the flowering period (R3) and placed to dry in an oven with a circulation of forced air at a temperature of 65 ºC for about 48 hours (Martinez *et al*., 1999; Malavolta *et al*., 2006). After being dried, the material was ground in a Willey mill, equipped with a 1 mm open mesh sieve and packed in paper bags to determine the foliar boron content (Wikner, 1986; Bataglia, 1991).

Chlorophyll index: at stage V9 of soybean plants, the indirect reading of the chlorophyll concentration of the leaves was carried out using the SPAD index obtained with the portable chlorophylmeter chlorofiLOG CFL1030. The reading was carried out on six leaves of the average third of the plant, being sampled at random six plants per plot, obtaining the average per parcel.

Seed productivity kg ha^{-1}: determined by harvesting and threshing the useful parcel (three central rows). To calculate the productivity after threshing, the water content of the grains was adjusted to 13.0%, also discounting impurities, the result being expressed in kg ha^{-1}.

Laboratory seed quality assessment

1000 seed weight: performed through a precision scale, with 100 seeds in 8 repetitions per treatment (Brasil, 2009).

Water content: conducted by the oven method, at 105 ± 3 °C for 24 hours, using two subsamples for each lot (Brasil, 2009).

Sprocket test: 4 repetitions of 50 seeds were distributed into three papers for each repetition and soaked with distilled water in the proportion of 2.5 times the dry weight of the papers. Next, they were organized in rolls, kept in plastic bags of polyethylene for the maintenance of humidity, and wrapped in B.O.D. type incubator at 25 °C. The evaluations were carried out by means of the normal seedlings count, 8 days after seeding and expressed in percentage (Brasil, 2009).

Root and aerial length: test conducted in a similar way to germination, but in the absence of light, 4 repetitions of 20 seeds per treatment were used that were distributed in two rows with 10 seeds each, over eight days. The length of the normal seedlings (aerial part and root part) was measured with a measuring ruler and expressed in centimeters (Nakagawa, 1999).

Growth rate index: carried out at the same time as the test for the length of the root and the aerial part, which measured the length of the total plant after the root has been protruded, daily measurements being carried out until the end of the test, results expressed per cm of day-1 (8 days). The calculation was carried out by the formula proposed by Maguire (1962) for germination and adapted to the growth of seedlings:

$$IVC = \frac{PN1}{N1} + \frac{PN2}{N2} + \cdots \frac{PNn}{Nn}$$

Where: IVC = growth rate index

PN1, PN2, ... PNn = centimeters of normal seedlings computed on the first, second and last count.

N1, N2, ... Nn = number of days after sowing in the first, second, and last count.

Radical development ratio and aerial part: the results were expressed as a percentage and used the following formulas:

$$Root\ ratio = \frac{root\ length}{root\ length + aerial\ length}\ x\ 10$$

$$Air\ ratio = \frac{aerial\ length}{root\ length + aerial\ length}\ x\ 100$$

Seed nutrient titration

Seed phosphorus concentration: In order to obtain the phosphorus concentration (P) in the seeds, four samples were dried in an oven at 65 °C, followed by sulfuric digestion described by Tedesco *et al*. (1995) and determination of the concentration of P in the extract by the colorimetric method.

Seed nitrogen concentration: made by sulfuric digestion, the N-organic being measured in the extract, using Nessler reagent and the $N\text{-}NO_3^-$ concentration, according to the method described by Cataldo *et al*. (1975).

Seed potassium concentration: were carried out by means of nitroperchloric digestion and subsequent determination of the potassium extract by flame photometry.

Seed boron concentration: the seed has been digested by drought (incineration) and subsequently the concentration of the seed has been quantified by the use of the coloring process by Azomethine H.

The data was subjected to the Shapiro-Wilk normality test and the Bartlett variance homogeneity test, which were normal and then subjected to variance analysis and averages compared by the Tukey test at 5% probability for the phenological stages and for the doses of B. The regression analyses were processed in the R software version 3.1.2 (R core team, 2015).

RESULTS AND DISCUSSION

There was no significant difference in the moisture content of soybean seeds; however, small variation in relation to vegetative stages and boron doses used (Table 1) can be observed. With the lowest water content observed at stage V3, without boron application, with 8,7 % and the highest water content observed at stage R1 for the 5 kg ha^{-1} dose, which presented 9,9 % (Table 1). The seeds tend to lose strength and reduce their germination during storage due to peroxidation of the lipids, so the water content must be less than 14%, which reduces their breathing and consequently their deterioration (Smaniotto *et al*., 2014), so the water levels found for the stored seeds show their physiological integrity.

For the evaluations of the phenological stages, it is observed that the majority of the agronomic variables analyzed did not differ statistically. Finding significant interaction only for foliar boron concentration (Table 2 and Figure 1A), and significant effect of doses on productivity (Table 2 and Figure 1B). All variables showed low coefficient of variation, indicating experimental accuracy and low variation of data collected in the field.

The nutritional requirement of soybeans and the potential for exporting the crop are determined by genetic factors, but there is an influence of the edaphoclimatic conditions and the cultural handling adopted. The amount of boron absorbed and exported by the soybean crop is 20 mg kg^{-1} and 26 mg kg^{-1}, with a recommendation of ground boron application of 0.5 to 1.5 kg ha^{-1} according to the analysis of the B content of the soil (Embrapa, 2014).

Table 1 - Moisture level of soybean seeds as a function of phenological stages and boron doses in the 2018/19 season (Brazil, 2009)

	STAGES				
	V0	**V3**	**V6**	**V9**	**R1**
DOSES (kg ha^{-1})					
0	8,9	8,7	9,1	8,9	9,2
1	9,0	9,1	8,9	9,4	9,1
2	9,1	8,8	8,8	8,9	9,1
3	8,9	9,0	9,1	9,0	9,1
4	9,0	9,4	9,0	9,3	9,2
5	9,3	9,4	9,7	9,8	9,9

Table 2 - Variance analysis for foliar boron concentration (CBF), relative index of chlorophyll (CLOR), mass of 1000 seeds (M1000), productivity per kg ha^{-1} (PROD) in soybean plants as a function of the application periods and boron doses in the 2018/19 crop. Brazil, 2019

FV	CBF	CLOR	M1000	PROD
Stages (A)	4,27**	1,96^{ns}	1,17^{ns}	1,72^{ns}
Boron Doses (B)	15,07**	0,30^{ns}	1,52^{ns}	4,49**
A x B	2,62**	1,97^{ns}	0,78^{ns}	0,76^{ns}
CV (%)	10,14	4,23	8,53	12,42
	mg kg^{-1}	**Spad**	**g**	**kg ha^{-1}**
Stages				
V0	73,92 ab	44,18	18,22	2426
V3	78,1 ab	44,52	17,50	2374
V6	71,17 b	43,91	17,27	2609
V9	79,07 a	42,97	17,94	2389
R1	80,08 a	44,37	18,00	2435
Doses (kg ha^{-1})	---$^{(1)}$	-	-	---$^{(2)}$
0	-	43,90	17,33	-
1	-	43,95	18,26	-
2	-	43,77	17,13	-
3	-	43,84	18,33	-
4	-	44,51	17,86	-
5	-	44,00	17,80	-

Means followed by the same lower case letter in the column do not differ by Tukey test at 1% probability. **= Significant at 1% probability; ns = not significant; (1) Significant regression for application periods and boron doses; (2) = Significant dose regression.

For the chlorophyll index, there was no significant difference, which corroborates the results found by Gomes *et al.* (2017), in studies carried out with different applications of boron via soil in the most diversified phenological stages in soybean crops; however, for a mass of 1000 seeds the results differ from the present research, since it indicates a significant effect for doses 2.8 kg ha^{-1}. This fact is related to the cultivar used, since distinct genetic materials can express a difference in the accumulation of reserves in the grains (Souza *et al.*, 2008).

It can be observed that for the phenological stages V0, V9 and R1, the data presented a square adjustment, whereas the maximum concentrations of foliar boron 79,74, 89,95 and 86,71 mg kg^{-1} were obtained when 2,81, 2,82 and 3,07 kg ha^{-1} of B, respectively (Figure 1A). These results show that soybean plants were well nourished with this element during both periods of boron application, since the ideal boron concentration range was within the limit established by Raij *et al.* (1997), which is 25 to 60 mg kg^{-1}. Stage V3 exhibited positive linear behavior with the increase in boron doses, where for each unit increase of boron applied via the soil an increase of 5,1 mg kg^{-1} in the foliar boron concentration occurs.

Despite the increase in doses, there was no toxic effect and inhibition of the absorption and relocation of B foliar. The deficiency shall be verified when the foliar content is less than the values of 25 to 30 mg kg^{-1} and the toxicity when this content is above 83 mg kg^{-1}, parameters which

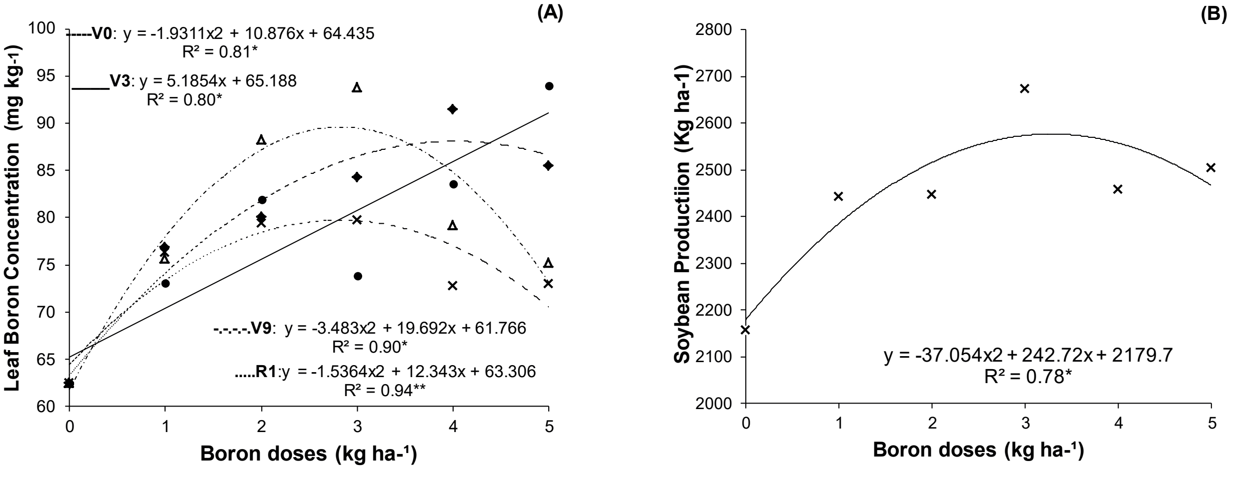

Figure 1 - Return to: (A) Leaf boron concentration (CBF) according to application times and boron doses; (B) Productivity in soybean plants as a function of boron doses. Brazil, 2019. **= Significant at 1% probability; *= Significant at 5% probability.

depend on the cultivar and the environment (Furlani *et al*., 2001). The results found in this work differ from those found by Calonego *et al*. (2010), which, when assessing borate leaf fertilization in soybean crops, found boron leaf content between 94.7 and 131.7 mg kg^{-1}. Urano *et al*. (2007) found lower results with values between 42,7 and 61,0 mg kg^{-1}.

When assessing the doses, it was possible to observe the adjustment of the data of the quadratic regression, where the highest productivity of 2,577.20 kg ha^{-1} was obtained when using 3.27 kg ha^{-1} of B (Figure 1B). Studies found that productivity is reduced with doses of B higher than 4 kg ha^{-1}, indicating toxic effect, and also that the best result found was 2,617 kg ha^{-1} with a maximum point of 3.51 kg ha^{-1} of B (Gomes *et al*., 2017). Results similar to those of Seidel and Basso (2012) show that the applications of calcium and boron based foliar fertilizer, regardless of the stage of development, did not influence the production and productivity components of soybeans.

Application times have influenced all laboratory tests (Table 3). There was no significant interaction between the physiological factors of soybeans and doses of boron, and for this reason, the average data referring to the levels of boron fertilizing was processed.

Table 3 - Analysis of variance and testing of averages for germination (GERM), root length (CPR), aerial part length (CPA), growth rate (IVC), radicular development ratio (RZR) and development ratio of the aerial part (RZA) in soybean seedlings according to application periods and boron doses. Brazil, 2019

FV	GERM	CPR	CPA	IVC	RZR	RZA
Stages (A)	4,67**	6,21**	24,21**	14,56**	5,13**	5,13**
Boron Doses (B)	10,99**	8,92**	11,12**	8,28**	17,37**	17,37**
A x B	2,91^{ns}	3,03^{ns}	4,53^{ns}	3,75^{ns}	3,15^{ns}	3,15^{ns}
CV (%)	9,92	14,87	9,48	10,70	6,19	4,91
	%	**cm**	**cm**	**cm dia^{-1}**	**%**	**%**
Stages						
V0	62 a	5,53 b	6,79 c	1,54 b	45 a	55 b
V3	61 b	5,23 b	6,42 c	1,45 b	45 a	55 b
V6	68 a	6,25 a	7,40 b	1,70 a	46 a	54 b
V9	63 b	6,21 a	7,94 a	1,76 a	44 b	56 a
R1	66 a	5,95 a	7,99 a	1,74 a	42 b	58 a
Doses (kg ha^{-1})	---$^{(2)}$	---$^{(2)}$	---$^{(2)}$	---$^{(2)}$	---$^{(2)}$	---$^{(2)}$

Means followed by the same lower case letter in the column, for each factor studied, do not differ by Tukey test at 1% probability. **= Significant at 1% probability; ns = not significant; (2) = Significant dose regression.

It is noted that boron fertilization showed better results for germination at development stages V0, V6 and R1. The root length on V6, V9 and R1, on the other hand, proved to be higher than the other application stages, as well as stages V9 and R1 for the aerial portion length. The best averages are to be emphasized, for the growth rate and the development ratio of the aerial part, in stages V9 and R1, now for the root development ratio in stages V0, V3 and V6.

Generally speaking, there were greater results of well developed and normal seedlings when boron was applied in R1; the same phenological stage was observed by Berti *et al*. (2019) in seeds without storage. Studies recommend applications of B from stage R1, since B is a not very mobile element in plants, and its application after flowering has a direct influence on seed quality and yield (Calonego *et al*., 2010; Varanda *et al*., 2018a).

There was an average reduction of 21.8% in seed germination after storage for six months when compared to that found by Berti *et al*. (2019), in which the reduction of germination by phenological stage compared to without and with storage was 30.49% in V0, 21.89% in V3, 11.91% in V6, 20.55% in V9, and 30.30% in R1. The reduction of the germinative potential of the seeds can be associated with the storage conditions, by altering the integrity of the membrane of its tegument in high temperature and humidity conditions, reducing the final quality of the product and rapid deterioration of the seeds (Baudet e Villela, 2006).

The regression tests for doses referring to laboratory variables are found in Figure 2. In relation to germination, length of the aerial part, growth rate, radicular development ratio and development ratio of the aerial part, the quadratic model was the one that best adjusted the doses of B with a determination coefficient of 75, 70, 82, 84 and 84%, respectively (Figure 2A, C, D, E and F). For root length, there was an adjustment to the linear model, with a determination coefficient of 51% (Figure 2B).

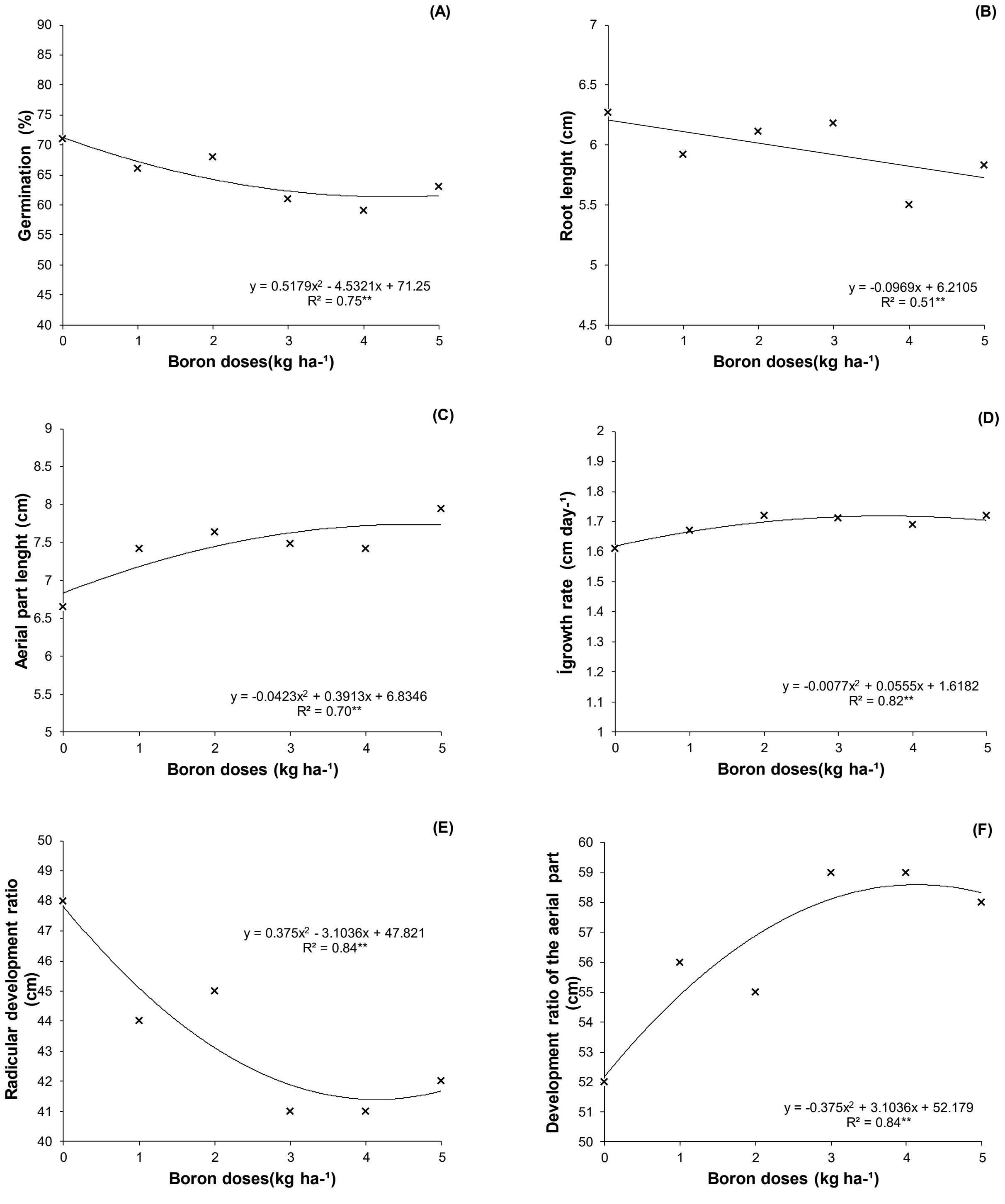

Figure 2 - Analysis of regression for: (A) Germination, (B) root length, (C) aerial part length, (D) growth rate, (E) radicular development ratio and (F) development ratio of the aerial portion of soybean seedlings according to boron application doses. Brazil, 2019. **= Significant at 1% probability.

The application of B doses did not increase germination and root length, as the levels of borate fertilization increased (Figure 2A and 2B). Damage caused to seeds caused by some type of injury or stress can be immediate (visible) or latent (not apparent), manifesting only after a certain time (Heberle *et al*., 2019), that is, after a period of storage, the physiological quality of the seeds is reduced and lost vigor.

The application of doses of B did not give rise to an increase in germination and in the root length, as the levels of the borated fertilization (Figure 2A and 2B) increased. Seed damage caused by some type of injury or stress can be immediate (visible) or latent (not apparent) effect, only manifesting itself after a certain period of time (Heberle *et al*., 2019), i.e., after a storage period, the physiological quality of the seed is reduced and lacks strength.

The application of 4.5, 3.4 and 4.1 kg ha^{-1} of B resulted respectively in a greater length of the aerial part (7.7 cm), growth rate (1.71 cm day^{-1}) and development ratio of the aerial part (58.6%) (Figure 2C, D and F), evidencing a more adequate level for borated fertilization, aiming at the quality of the seeds after a storage period. The radicular and aerial development ratio, because they are inversely proportional variables, showed an inverse trend line; whereas the root development ratio decreased until the 4 kg ha^{-1} of B dosing, and the ratio of the aerial part showed a positive growth in development up to the same dosage of borated fertilization.

This shows that the dose of 4.2 kg ha^{-1} boron will make it possible to plant soybean plants, the largest increase in the vegetative mass of the aerial part, even after six months of storage. As Taiz *et al*. (2017) reports, the boron participates in the regulation of the production of the auxiliary hormone in the plant, which is responsible for the elongation, division and growth of the plant; this justifies the significant results for the part of the area, when submitted to fertilization with boron.

In Table 4, the laboratory variables for the titration analyzed varied statistically when the doses of boron were evaluated. Significant interaction was found for nitrogen (N), potassium (K) and boron (Figure 3B, C and D); and significant effect for phenological stages in the titration of phosphorus (P) (Table 4 and Figure 3A). Phosphorus concentration in the seeds was higher in stage V9 followed by R1, whereas the maximum concentrations in the final stages can be explained by Schoninger *et al*. (2013) who observed the low use of P by soybeans in its initial development is independent of the phosphate source; thus the low metabolic demand reduces absorption, displacement and incorporation of the phosphorus into the root tissue.

The regression test for doses referring to the boron titration variable is found in Figure 3A, the quadratic model being the one that best adjusted the doses of B with a coefficient of determination of 72%. The application of 3.3 kg ha^{-1} of B associated with phosphorous fertilization obtained the highest concentration of phosphorous in the seeds of 3.8 g kg^{-1}, below that identified by Schoninger *et al*. (2013) of 4,65 g ha^{-1} and Krueger *et al*. (2013) of 5,58 g ha^{-1}. However, it stands out that there is an association of boron and phosphorus in the better incorporation of nutrient (P) into seed.

It can be observed that for the phenological stages V0 and V9, the data showed a square adjustment and for R1 linear adjustment, for the maximum concentration of nitrogen in the seeds of 61,71;

Table 4 - Variance analysis and testing of averages for nitrogen (N), phosphorus (P), potassium (K) and boron (B) titration in soybean seeds according to application periods and boron doses. (Brazil, 2019)

FV	N	P	K	B
Stages (A)	25,94 **	9,547 **	1,578 ^{ns}	5,289 **
Boron Doses (B)	4,030 **	3,073 *	5,480 **	30,41 **
A x B	4,343 **	1,470 ^{ns}	1,819 *	8,250 **
CV (%)	3,38	10,13	2,54	8,86
	g kg^{-1}	**g kg^{-1}**	**g kg^{-1}**	**mg kg^{-1}**
Stages				
V0	59,980	3,527 c	21,734	42,960
V3	58,646	3,526 c	21,490	46,528
V6	59,710	3,961 cb	21,288	42,339
V9	59,530	4,122 a	21,423	47,066
R1	54,287	4,018 ab	21,489	45,902
Doses (kg ha^{-1})	---(1)	---(2)	---(1)	---(1)

Means followed by the same lower case letter in the column, for each factor studied, do not differ by Tukey test at 5% probability. **= Significant at 1% probability; *= Significant at 5% probability; ns = not significant; (1) = Significant back to application times and boron doses; (2) = Significant dose regression.

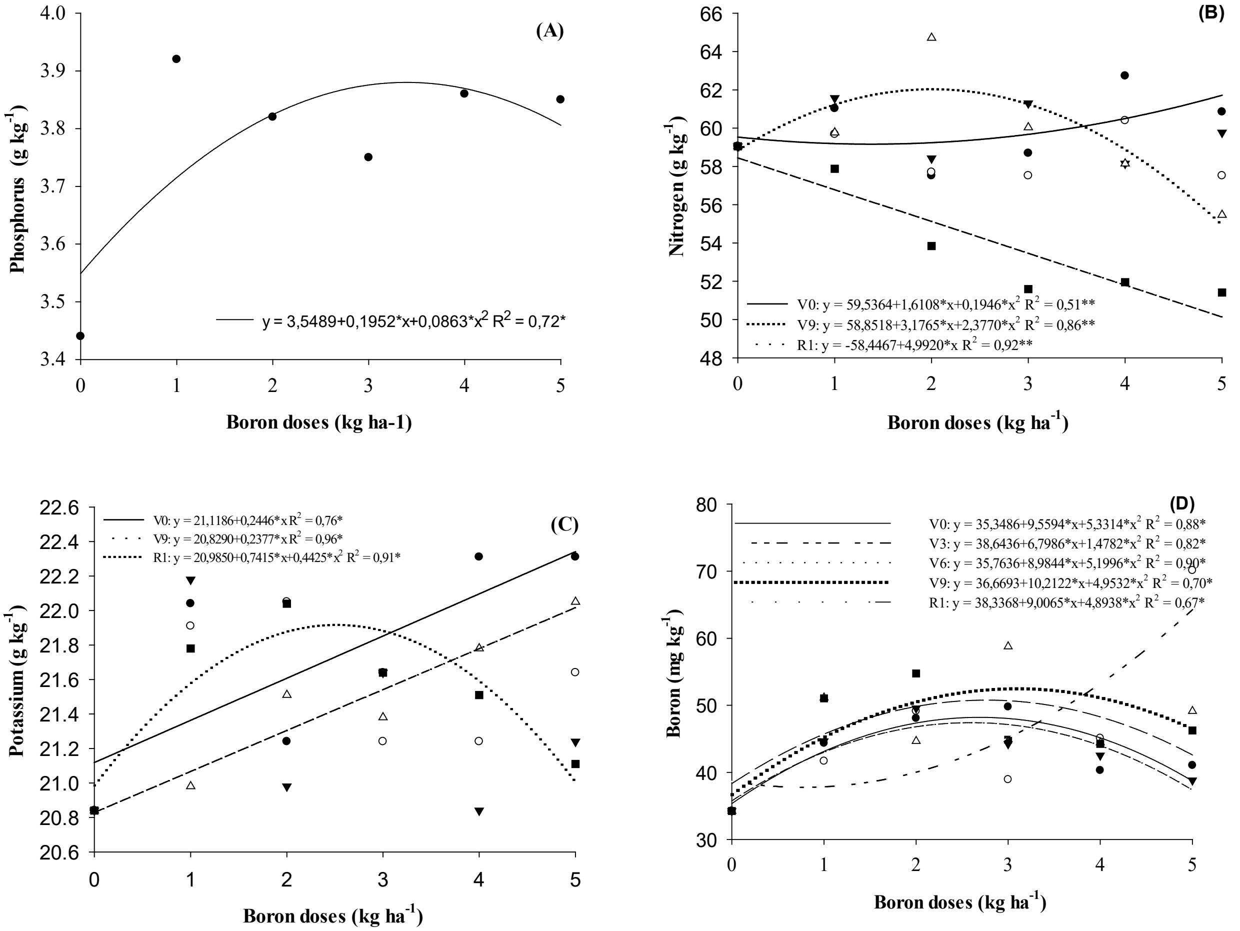

Figure 3 - - Return to: (A) Phosphorus concentration as a function of boron doses; (B) Nitrogen concentration, (C) Potassium concentration and (D) Boron concentration according to application times and boron doses. (Brazil, 2019). **= Significant at 1% probability; *= Significant at 5% probability.

62,55 and 58,45 g kg^{-1} were obtained when 5, 2,1 and 0 kg ha^{-1} of B were used, respectively (Figure 3B). These results show that the periods of boron application interfere with the dose to be used for higher nitrogen concentration in the seeds, with the optimum nitrogen concentration range being 58 g kg^{-1}.

Marin *et al.* (2015) pointing out that this low variation in N concentration in soybean seeds is caused by the high N requirement of soybeans throughout their biological cycle. Symbiotic with atmospheric nitrogen fixing microorganisms. The non-variation of the genetic material in the work also interferes with the concentration of N in the seeds, since the quantity of nitrogenous elements absorbed by the plants coming from the biological fixation is the same between the different treatments studied.

For the concentration of potassium in soybean seeds, the V0 and V9 data showed linear adjustment and in R1 quadratic adjustment, with this the maximum identified potassium titrations were 22,31, 22,01 and 21,42 g kg^{-1} obtained when 5,5 and 2,45 kg ha^{-1} of B, respectively (Figure 3C). As occurred in the N concentrations, potassium obtained maximum concentrations when applying the highest doses of B at the initial stages, reducing the doses at the final stage (R1).

Krueger *et al.* (2013) obtained K values lower than identified in this work, ranging from 16.5 to 17.8 g kg^{-1}, even using larger K doses (132 kg ha^{-1}), as well as found that high doses of potassium fertilizer interfere with the quality of the seeds. The depositing of nutrients in the seed is related to the genetic capacity of the cultivar to metabolize and store them, and it is possible that by using the best dose of B associated with the handling, whether at the beginning or at the end of the crop cycle, it provides adequate K concentration in the seed.

In Figure 3D, on the other hand, boron titrations for all the phenological stages showed significant regression with a coefficient of variation of 67 to 90% and a quadratic adjustment. Soon, the maximum boron concentrations in the seed were 48.24, 64.12, 47.37, 52.50 and 50.74 mg ha^{-1}, obtained with 2.70, 5.0, 2.78, 3.05 and 2.93 kg ha^{-1} of B via the ground, at stage V0, V3, V6, V9, and R respectively. The result differs from the other titrated nutrients, in which the doses of boron are independent from the phenological stage used and the mean of 3.29 kg ha^{-1}.

Ross *et al*. (2006) and Bellaloui *et al*. (2010) yielded different results when using doses of 0,28 to 1,12 and 0.45 to 1.8 kg ha^{-1} of foliar boron, respectively, and obtained the best concentrations of boron in the seeds when applied at stage R2 from 20 to 47 mg kg^{-1}. Due to the low mobility via the phloem, the leaf application can be less efficient compared to the soil path. Therefore, the use of higher doses via soil is justified and also respond to a better absorption, so it is observed that the values found for the titration of boron were higher in seeds with application via soil with an average of 52.58 mg ha^{-1}.

The viability and the vigor of the seeds, which define its productive potential, is determined by the germination test. In this sense, a well-nourished plant, carries out the translocation of nutrients from the leaves to the reserve organs, allowing for a good formation of the embryo and of the cotyledons, and therefore this directly impacts the physiological quality of the seeds, since the nutrients help the formation of reserve substances such as proteins, carbohydrates and lipids (Teixeira *et al*., 2005).

In this context, it is observed that there has been a reduction in germination as the level of fertilization borated up to a certain point (Figure 2A) has been raised, however in the largest dose (5 kg ha^{-1}) there is an increase in germination. The same was observed when using boron leaf fertilizer, aiming at the quality of bean seeds, where the percentage of germination data also resulted in a square adjustment of the function, where with an increase of boron doses up to a certain level, there was a decrease in germination and afterwards an increase in the doses (Abrantes *et al*., 2015).

At the practical recommendation level, aiming at a higher productivity of soybeans, the present work indicates the dosage of 3 kg ha^{-1}, also taking into consideration the conditioning of these seeds over a period of 6 months, without any loss of quality beyond this. It is, therefore, possible to recommend the fertilization borated at stage R1, which showed significant averages for the seed quality variables, and also corroborated with the findings of Varanda *et al*. (2018b), which obtained better responses from soybeans with the application of B in the R2 and R4 stage, demonstrating important functions of the bordered fertilization interconnected with the physiological stage of the crop.

CONCLUSION

The application of boron in the different phenological stages did not affect the productivity of the Monsoy 7110 IPRO cultivar, but it did increase the physiological quality of the seeds. This study recommends the dose of 3.24 kg ha^{-1} boron via soil at the beginning of the reproductive stage, since the vigor and the concentration of boron in the seeds increases.

ACKNOWLEDGMENTS

The State University of Goiás, Ipameri University Unit (Universidade Estadual de Goiás, Unidade Universitária de Ipameri) and the Phytotechnics Study and Research Group (GEPFi); To researchers at the Federal University of Pelotas (UFPel); To the National Council for Scientific and Technological Development (Conselho Nacional de Desenvolvimento Científico e Tecnológico - CNPq) for the financial assistance for the acquisition of equipment, Universal Call MCTIC/CNPq No. 28/2018. Process: 436837/2018-8.

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