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REGULAR ARTICLE

Effect of glucose on germination performance in two soybean cultivars

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Abstract

Glucose promotes increases in physiological processes related to plant growth and induces cell division and can also act in seed germination. The present investigation has been carried out on the influence of various concentrations of glucose on the germination performance of soybeans seeds of cv. BMX and 66i68. Seeds of soybeans (*Glycine max* (L.) Merrill) were treated with three concentrations of the glucose (0.05, 0.10 and 0.20 mmol L^{-1}), and a control with distilled water. Tests of seed germination and vigor were carried out: it was calculated the germination percentage (first and final counting), percentage of normal seedlings; and speed of germination, length of the primary root and the aerial part of seedlings were recorded. The experiment was arranged in a completely randomized design with three treatments and five repetitions depending on the test. Increasing doses of the plant growth regulator influence the germination and can increase the speed of germination. The use of glucose in the treatment of soybean seeds, increases the rate of germination and emergence speed, growth of the root system and medium of seedlings. Soybean cultivar BMX seeds showed an increase in physiological performance with increasing glucose dosage.

Keywords

Glycine max; phytohormone; root protrusion index; vigor; germination speed index.

Introduction

Brazil is the second largest soybean [*Glycine max* (L.) Merrill] producer in the world, with a planted area of approximately 35,100 million ha^{-1}, with a production of 119,518 million tons with an average productivity of 3,333 kg ha-1, and the 3 largest states producers are Mato Grosso in first place, followed by Paraná and Rio Grande do Sul (Conab, 2020). Worldwide, soybean is the main oil crop produced and consumed. Because soybean is an important source of protein in human food and most of the animals that produce meat, milk and eggs, today it offers a variety of products, forming a very comprehensive production chain, being also considered an important species for the production of biodiesel (Ali et al., 2020).

The germination process depends on a number of factors, which involves several signals and is influenced by both intrinsic and extrinsic factors, including the seed's physical and biochemical integrity and physiological status (Zhou et al., 2020). Other problems observed in the production of seedlings is the rapid development of the aerial part of the seedlings, which can lead to stagnation, with uneven growth, fragile and with few roots. However, new alternatives have been used to improve both the percentage of germination and the development of seedlings, with the encouragement of plant regulators, aiming to regulate the growth and quality of seedlings.

Elicitors can be defined as substances which, when introduced in small concentrations into a living cell system, initiate or augment the metabolites processes of the seeds and plants (Gorni et al., 2020). This practice is a tool used to promote increase of the germination, biomass and improve crop performance and consequently increase productivity (Kim et al., 2020). In this context, glucose (Glc) is one of the most important carbohydrates for plants. This compound is related to several physiological processes of the plant, being able to act as a growth regulator, in addition to having antioxidant properties and is considered a signaling molecule produced in responsible plants for inducing defense responses against biotic and abiotic stresses (Baenas et al., 2019; González-Hernández et al. 2020).

Glc is involved in a series of physiological processes in plants has been implicated to as the primary signal for plant development, including germination, hypocotyl elongation, cotyledon greening and expansion, true leaf development, photosynthesis, carbon and nitrogen metabolism, flowering, mitigation to stress and senescence (Rolland et al., 2006; Zhaid et al., 2018; Baenas et al., 2019; Wang et al., 2019). In addition

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to these effects, studies have showed that pre-soaked with Glc promotes seed germination and growth in Indian mustard under salinity (Sami et al., 2021). Exogenously sourced Glc alleviates drought stress in wheat (Zahid et al., 2018), induced the bioactive compounds contents in almost all combined treatments (priming seeds and spray) in radish sprouts (Baenas et al., 2019) and enhanced root density, primary root length, and lateral root number in tomato seedlings (González-Hernández et al. 2020).

Searching for new alternatives, this research will analyze and describe the effect of increasing Glc concentrations on seed germination and on the development of soybean seedlings in order to improve the quality and productivity of the crop (Zhaid et al., 2018; Baenas et al., 2019). In this context, the application of Glc can improve the germination and seedling quality in soybean. The present investigation has been carried out on the influence of various concentrations of Glc on the germination performance of soybeans seeds of cv. BMX and 66i68.

Materials and methods

The experiment was conducted in a seed laboratory at Faculdades Gammon, Paraguaçu Paulista (22º41'76" S, 50º58'33" W, 517 m.a.s.l.), Sao Paulo, Brazil.

The soybean seeds used in the experiment were from the cultivars BMX Potência and DM 66i68 IPRO, which were purchased from commercial establishments, from the 2019/2020 harvest, and were not treated with fungicide. The soybean seeds were previously disinfested with 1% sodium hypochlorite for three minutes. After drying, on sterilized filter paper sheets, these were treated with glucose (Glc) (180.156 g mol-1) at zero concentrations; 0.05, 0.10 and 0.20 mmol L^{-1} and were soaked for 20 minutes. After imbibition, the germination paper was used as a substrate for the germination test (Brasil, 2009).

The seeds were placed in a gerbox [11 x 11 x 3.5 cm] (100 seeds for each glucose concentration, composed of 5 sub-samples of 20 seeds), using blotting paper previously moistened with distilled water (in the proportion of 2.5 times its initial weight). We opted for this methodology instead of germinating the seeds directly in the Glc solutions so that there would be no damage to them due to the longer contact with the reagent. The gerboxes with the seeds were placed on benches with an ambient temperature of 23 ºC ± 3 ºC (Coutinho et al., 2015). The seeds were moistened with distilled water every day, as needed. The seeds were considered germinated when the extruded roots showed a positive geotropic curvature of approximately five millimeters.

Germination test

Counting was carried out on the fifth day after sowing. In the evaluation, normal plants (germination) were considered, data expressed as a percentage, and plants considered abnormal were eliminated.

Evaluation of germination

The results evaluated in the fifth, eighth, eleventh, sixteenth and twenty-first after sowing (DAS) were expressed in percentage of germination (G%) and first count of germination, evaluated in the fifth DAS and expressed in percentage (P%).

The germination speed was determined using an index adapted from the formula of Maguire (1962) in which no daily germination evaluations were performed using the following equation:

$$GSi = G1 / 5 + G2 / 8 + G3 / 11 + G4 / 16 + G5 / 21$$

Where: GSi is the modified germination speed index;

G1, G2, G3, G4 and G5 are the germination counts in the fifth, eighth, eleventh, sixteenth and twenty-first and,

5, 8, 11, 16 and 21 are the counting days.

Root protrusion index

Concomitantly, the root protrusion index was carried out on the germination test, with the number of seeds showing root protrusion 5 mm or more being calculated daily, and the root protrusion index was calculated (RPi) based on the formula of Maguire (1962).

$$RPi = G1 / N1 + G2 / N2 \dots Gn / Nn$$

Where: RPi = root protrusion index;

G1, G2, Gn = Number of normal seedlings to be counted in the first count;

N1, N2 and Nn = Number of days from sowing to first.

Vigor test through seedlet length

It was performed on a paper roll, using four repetitions with ten seeds. The same procedures as described for the germination test were used. The seeds were distributed on a line drawn in the upper third of the blotting paper, in the longitudinal direction. The paper rolls were placed in plastic bags and closed, to avoid dehydration, and kept vertically, at a temperature of 30 ºC, inside the germinator, for five days, in the absence of light (Nakagawa, 1999).

After five days in the germinator, normal seedlings were measured with a ruler. The measurement was taken from the tip of the root to the insertion of the cotyledons. The average length of the seedling or its elected part (s) was calculated as follows:

$$ASL = SL^1 + SL^2 + SLn / Sn$$

Where: ASL = Average seedling length;

SL^1, SL^2, SLn = Normal seedling length or part of it;

Sn = Number of normal seedlings to be measured.

Statistical analysis

The experimental design was completely randomized with 5 replications and arranged in a 2 × 4 factorial scheme, comprising of two soybean cultivars (BMX and 66i68) in combination with five glucose concentrations of [0 (control), 0.05, 0.10 and 0.20 mmol L^{-1}).

Normality and homoscedasticity of the data were analyzed using the Shapiro-Wilk and Bartlett tests, respectively, both at 0.05 probability. Then, the data were subjected to analysis of variance (ANOVA) using the F test ($p \leq 0.05$). When significant, the regression analysis was performed on the quantitative factor (glucose concentrations) and the Tukey test performed on the qualitative factor (soybean cultivars), both at 0.05 probability. All statistical analysis of the data was performed using protocols developed in the R software (R Development Core Team 2019).

Results and discussion

Observing the first germination count (P%), it shows that this trait presented an isolated effect between the factors "glucose concentrations" and "soybean cultivars" (Fig. 1). Glucose application showed significant results between treatments, respectivity compared to control, having a peak point at 87% at a concentration of 0.10 mmol L-1. In compared to cultivars was a higher percentage of germination in cultivar BMX compared to cultivar 66i68.

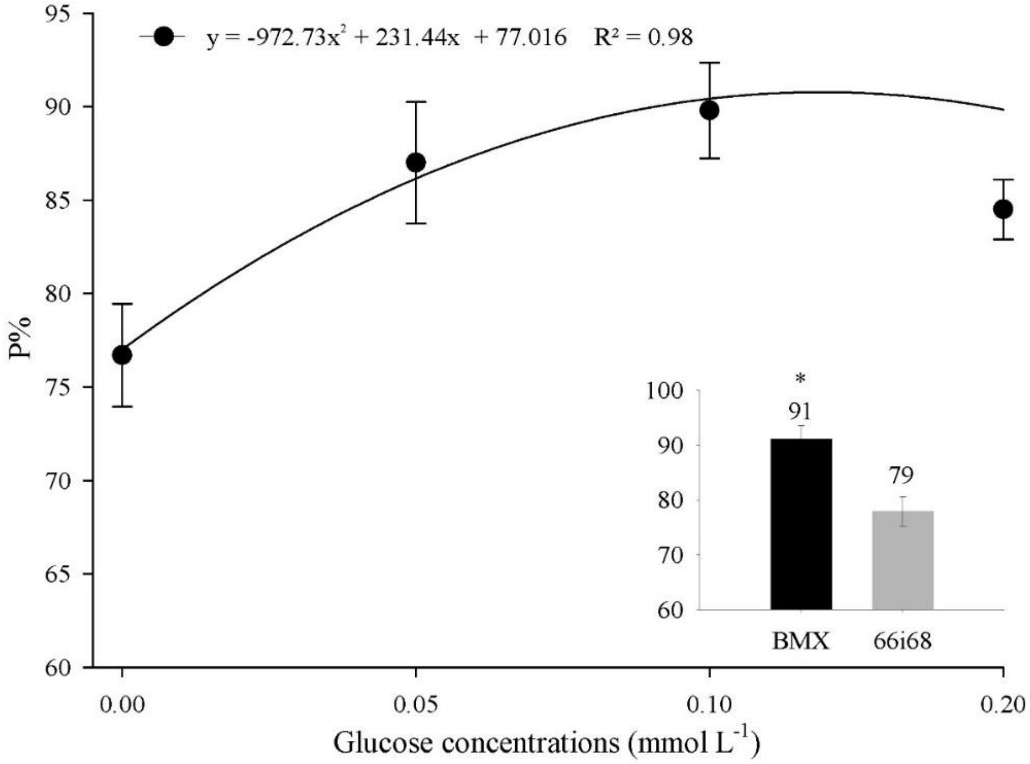

Figure 1. Isolated effect on first germination count (P%) in response to glucose concentrations and soybean cultivars. Vertical bars show the standard error of mean * Significant at the 0.05 probability level.

The data shows that the application of glucose obtain a significant result (Fig. 1), when compared to similar soybean cultivation processes, with or without the use of glucose, in the same period, considering sprouting and degree of germination. Studies show that glucose has a major role in regulating seed germination and modulates both ABA cell concentration and ABA response (Siddiqui et al., 2020).

The germination (%) trait showed an interaction between the factors "glucose concentrations" and "cultivars" (Fig. 2). However, the application of glucose in concentrations of 0.05, 0.10 and 0.20 mmol L-1 showed significant increases of 2.5%, 4.8% and 5.6%, respectively for the cultivar BMX in comparison to control. As for cultivar 66i68, there was a higher percentage of germination at concentrations 0.05 and 0.10 mmol L^{-1}, an increase of 4.7% and 2%, respectively compared to the control.

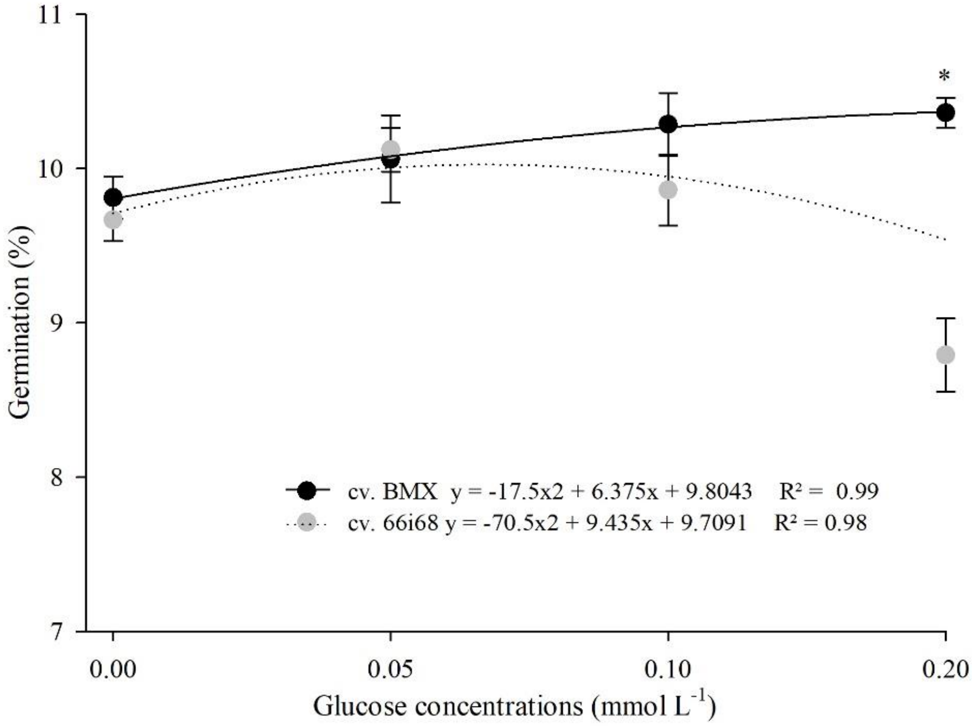

Figure 2. Interaction effect for the germination percentage (%) in response to glucose concentrations and soybean cultivars. Vertical bars show the standard error of mean * Significant at the 0.05 probability level.

A higher percentage of germination was observed in cultivar BMX, when compared to cultivar 66i68, considering the concentrations of 0.10 and 0.20 mmol L^{-1} of glucose (Fig. 2). These results are in agreement with what Vishal & Kumar (2018) affirm, where they affirm that the balance between hormones, promoters and inhibitors plays a fundamental role in the germination and initial seedling growth. Thus, it can be said that glucose may have increased the concentration of ABA, thereby increasing germination in soybean seeds. Studies show that the interaction application of sucrose (30 g L^{-1}) and 0.25 mg L^{-1} of GA3, promoted a higher germination speed in 'Pedro Sato' guava (Menezes et al., 2010).

The greater the role of glucose, the greater the germination percentage, because glucose functions as a growth promoter. Enhance in germination percentage after treatment might be the consequence of breakdown of dormancy in fresh seeds (Hosseini 2020). Alternatively, the earlier and synchronized germination might be attributed to enhanced metabolic activities in treated seeds. This study is still in agreement with the results acquired by Pereira et al. (2006), who, working with murmuru embryos (Astrocaryum ulei), found that the addition of 30 g L^{-1} of sucrose to the medium provided greater seedling development and height.

Analyzing germination speed index, it can be seen that in the increasing concentrations of glucose there was an isolated effect between the factors "glucose concentrations" and "cultivars" (Fig. 3). The different concentrations of glucose did not show significant results. However, in comparison with cultivars it is shown that cultivar BMX had a better vigor compared to cultivar 66i68.

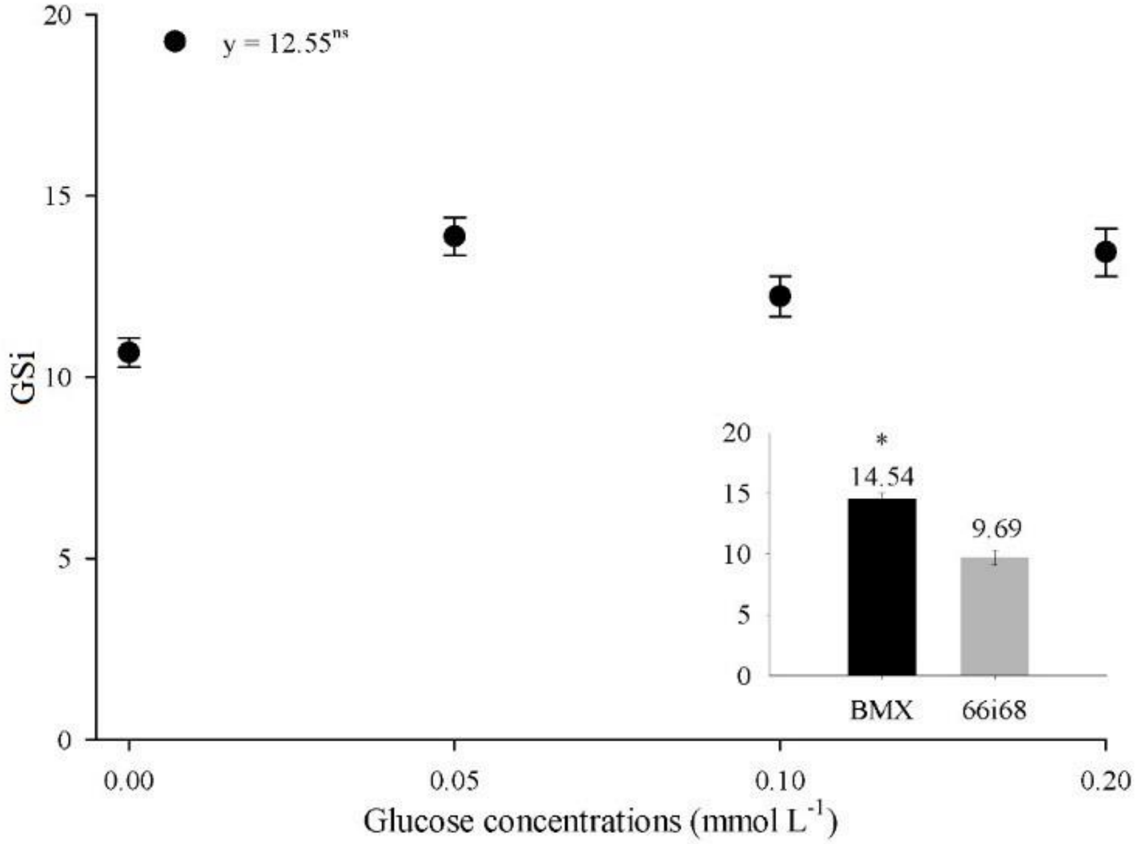

Figure 3. Isolated effect on germination speed index (GSi) in response to soybean cultivars. Vertical bars show the standard error of mean ns Non-Significant at the 0.01 probability level * Significant at the 0.05 probability level.

Observing the root protrusion index shows that this trait did not show any interaction between the factors "glucose concentrations" and "cultivars" (Fig. 4). The applications of 0.05 and 0.20 mmol L^{-1} showed increases of 25.8% and 32.6% for the cultivar BMX, respectively in comparison to the control, whereas for cultivar 66i68 there was an increase of 34.3% in the concentration 0.05 mmol L^{-1}, compared to the control.

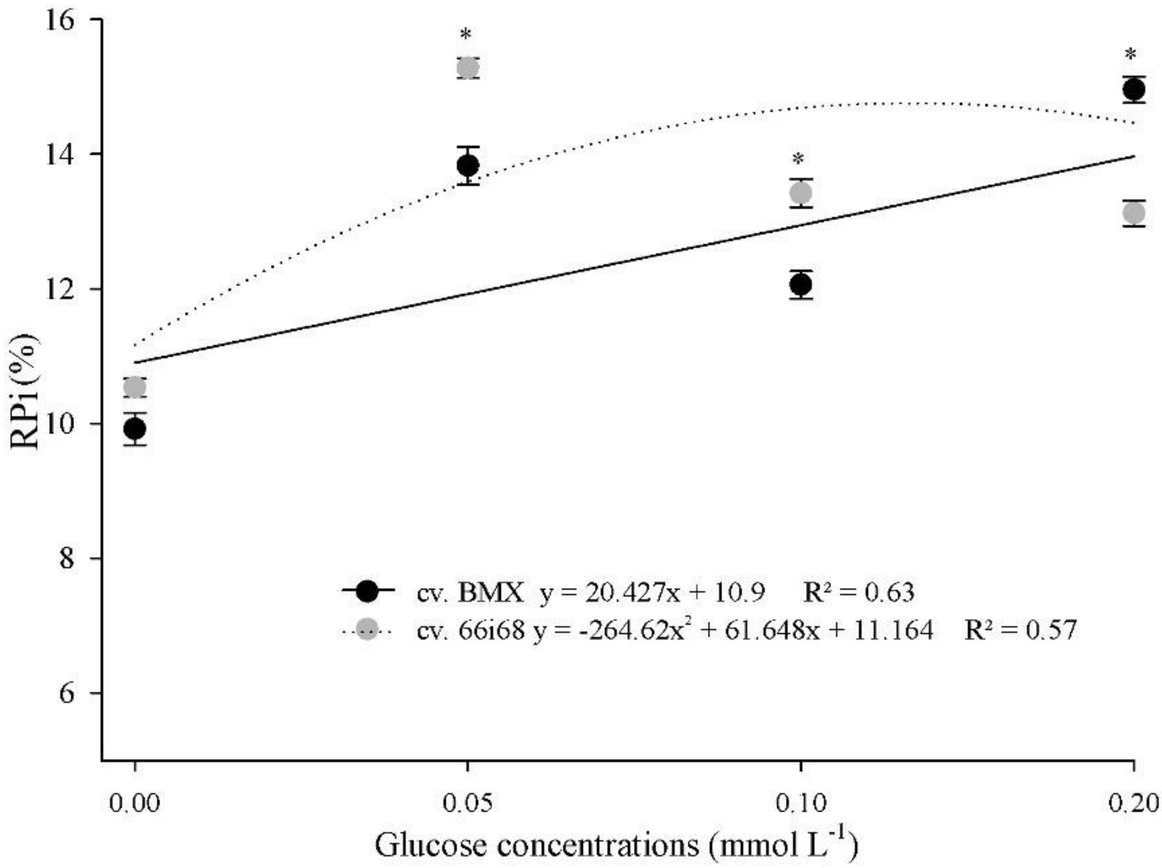

Figure 4. Interaction effect for the root protrusion index (RPi) in response to glucose concentrations and soybean cultivars. Vertical bars show the standard error of mean * Significant at the 0.05 probability level.

There was a significant interaction between the factors "glucose application" and "cultivars" for the trait average seedling length. The BMX cultivar treated with glucose showed increases of 88.8%, 83.8% and 96.1%, respectively compared to the control (Fig. 5). The 66i68 cultivar showed increases of 37.9%, 62.2% and 60.2%, respectively in comparison to the control. In the comparison between cultivars, it appears that only in the 0.05 mmol L^{-1} of glucose concentration there was a longer average seedling length in cultivar BMX.

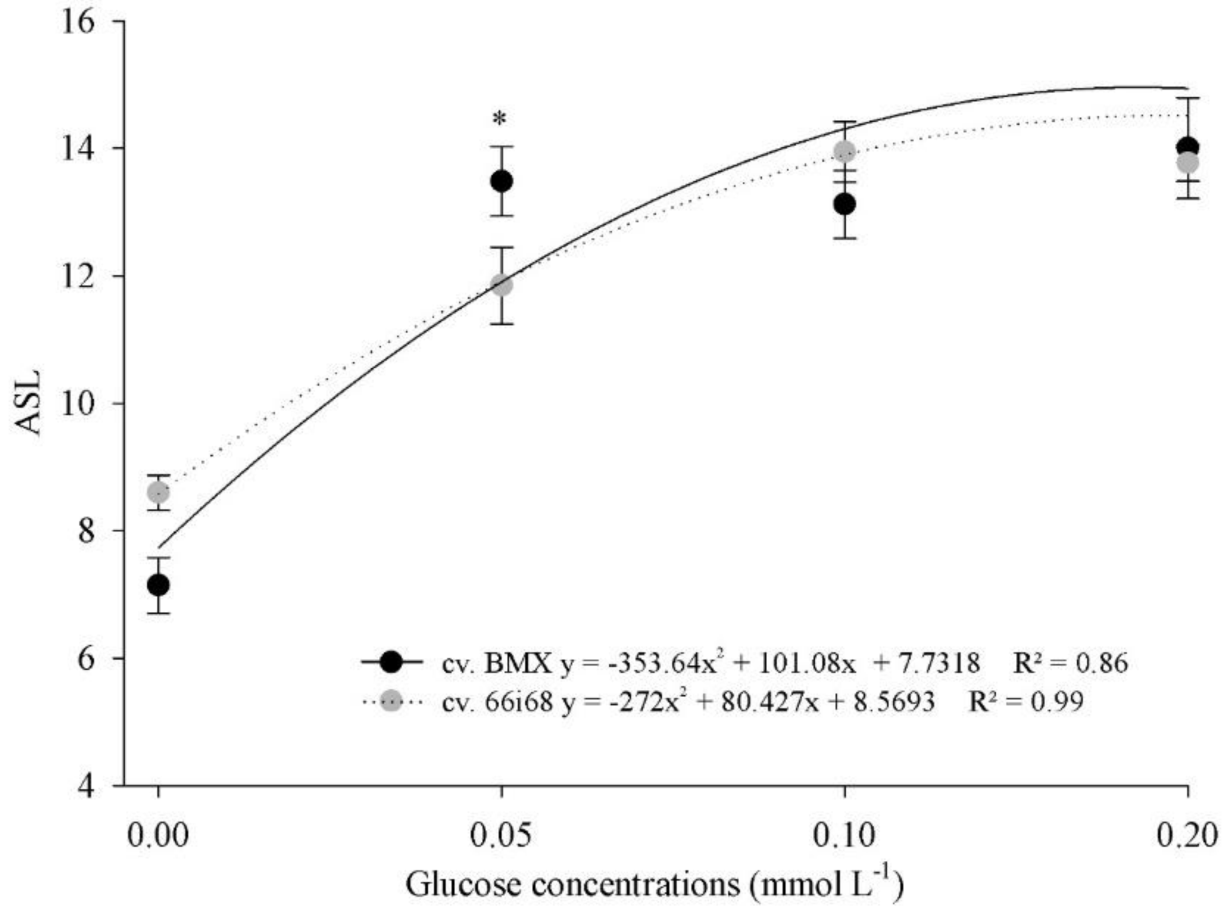

Figure 5. Interaction effect for the average seedling length (ASL) in response to glucose concentrations and soybean cultivars. Vertical bars show the standard error of mean * Significant at the 0.05 probability level.

The GSi (Fig. 3), RPi (Fig. 4), and ASL (Fig. 5) of the seeds of cultivar BMX and 66i68 were higher due to the treatment with glucose. However, the application of phytohormones can alter ethylene levels, they observed an increase in the physiological performance of seeds, protrusion of the radicle and the development of seedlings (Colli & Purgato, 2012). Based on these observations, glucose and CKs are likely to operate antagonistically at different steps of the ABA biosynthesis and signaling pathways (Wang et al., 2021). At lower concentrations, glucose stimulates germination by inducing ABA catabolism (Wang et al., 2021).

Similar results were also observed by Rezende et al. (2009), who verified that the in vitro cultivation of Cattleya loddigesii sp. it can be carried out with a concentration of 30 g L-1 of sucrose or with half that concentration, obtaining positive results in relation to the root length. Ribeiro et al. (2009) reinforces that sugar is essential for the development of roots in vitro, which was observed positively in this work. Sugar application has the positive effect of improving seed germination and seedling establishment, making plant propagation more efficient (Kushwah & Laxmi, 2014; Yusuf et al., 2021). The low concentration of glucose increases the growth of coleoptiles and radicals and contributes to the success of seed germination, thus, increasing the respiratory rate in the seeds and, consequently, the speed of seedling emergence (Hu et al., 2012).

The use of glucose in the treatment of soybean seeds, increases the rate of germination and emergence speed, growth of the root system and medium of seedlings. The BMX percentages were higher than that observed in 66i68, however there was a longer average seedling length in cultivar 66i68. At the end of the experiment, it can be considered feasible to use the experiment technique for practical crops.

Conclusions

The glucose (0.2 mmol L^{-1}) may be more effective for enhanced germination and seeds viability of soybean.

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