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Giberellic Acid in the Expansion of the Thermal Range of Lettuce Seeds

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Abstract

Lettuce presents an increasing demand, which makes necessary the expansion of the areas destined to its planting. Temperature is a limiting factor, because it acts directly on germination, where changes and adjustments in the hormonal balance, mainly of gibberellin (GA) and abscisic acid (ABA), occur. The objective of this study was to evaluate if the treatment of seeds with gibberellic acid is capable of mitigating the damages caused by high temperatures. The seeds were submitted to germination test, first count, percentage of abnormal seedlings and analysis of shoot and root growth. The treatments consisted of seven concentrations of gibberellic acid (control—not treated with GA, 25 mg L^{-1}, 50 mg L^{-1}, 75 mg L^{-1}, 100 mg L^{-1}, 125 mg L^{-1}, 150 mg L^{-1}) and three constant temperatures (25, 30 and 35 °C). The germination and first count, when submitted at 25 and 30 °C presented an increase up to 50 mg L^{-1} of GA, while at 35 °C the growth was constant up to 125 mg L^{-1} of GA. Treatment with GA increased root and shoot length. Giberelic acid mitigated the damage caused to lettuce seeds when subjected to temperatures above the optimum.

Keywords: hormonal balance, numbness, temperature

1. Introduction

The lettuce (*Lactuca sativa* L.) is a temperate, belonging to Asteraceae Family, which consists of the most consumed leaves and has an important role in the diet due to its taste, low cost and nutritional quality, as a source of vitamins, minerals and fiber (Santi et al., 2010). It is a short cycle crop, thus enabling, many harvests during the year.

Due to its increasing demand, it is necessary to expand the areas destined to its planting. However the climate conditions are a limiting factor, especially in relation to the temperature, being able to act directly in germination by affecting the dormancy and the viability of the seeds (Deng & Song, 2012). Since most of the cultivars used commercially have their germination suspended above 28 °C (Yoong et al., 2016).

When exposed to high temperatures the lettuce seeds may present a temporary inhibition (thermoinhibition) or complete in germination (thermodormity). The reasons of this inhibition are due to the stiffening of the endosperm, which presents itself as the main barrier of impediment to radicular protrusion and embryo growth (Nascimento et al., 2012).

During the germination, modifications and adjustments in hormonal balance occur, mainly of gibberellin (GA) and abscisic acid (ABA) (Nonogaki et al., 2010), with these exerting important and antagonistic roles in dormancy and germination (Flinkelstein et al., 2008). The GAs interfere in the regulation of division and cell elongation, causing greater embryo growth, besides inducing the synthesis of enzymes that act in the weakening of the micropillary endosperm (Yamaguchi, 2008). The ABA is indicated as the main inhibitor of germination, especially in the early stages of development (Taiz et al., 2017).

Studies have been conducted in order to establish the interactions between GAs and ABA with temperature in the induction and inhibition processes of seed germination in situ (Huarte & Benech-Arnold, 2010; Finch-Savage & Footitt, 2012). Yoshioka, Endo, and Satoh (1998) reported reduction in ABA content in lettuce seeds when they were soaked at optimal germination temperatures but increase at high levels when soaked at elevated temperatures. It is believed that the exougenous application of GA is responsable for the endogenous decrease of

ABA by promoting its catabolism and consequently for the increasing of temperature tolerance for lettuce seeds (Gonai et al., 2004).

Based on the above consideration, the objective of this work was to investigate whether the conditioning of lettuce seeds cv. Grands Rapids with giberelic acid would be able to soften the damages caused by high temperatures both on its physiological potential and germination.

2. Method

The experiment was developed in the Laboratory of Seed Analysis of the Agricultural Sciences Center (CCA) of the Federal University of Ceará (UFC), Campus of Pici-Fortaleza, from April to June 2018 using seeds of crisp lettuce "Grand Rapids".

Before treating the seeds with gibberellic acid (GA3), they were submitted to determination of the water content as described by MAPA (2009).

For conditioning, the seeds were distributed in petri dishes on two sheets of Germitest® type paper moistened with gibberellic acid solutions (GA3) in the proportion of 2.5 times the weight of the dry substrate. Moinstening was conducted for 16 hours at a constant temperature of 15 °C, until seeds reached 40% water content. After this time, the seeds were placed in a dryer for four hours at a temperature of 40 °C, sufficient time to return to the water content it had before conditioning and then submitted to the germination and vigor tests.

For the conduction of germination test, four samples of 50 seeds were used, which were seeded in Gerbox® boxes (11 × 11 × 3.5 cm) containing two sheets of Germitest® type paper moistened with diestilled water in the proportion of 2.5 times the weight of the dried substrate. Then, the gerboxes were conditioned in BOD at constant temperatures of 25, 30 and 35 °C and photoperiod of 12 hours. For the evaluation of vigor in the fourth day after the germination test was installed, the first germination count occurs. On seventh day the final germination was observed, determining the seedlings' normal and abnormal percentage in relation to the number of seeds used in the sample, using as a standard of normal seedlings the classification established by MAPA (2009).

The analysis of seedling' shoots and roots growth was performed after the germination test was completed by seedlings per sample with were measured which the aid of a ruler graduated in cm.

A completely randomized design was used in 7 × 3 subdivided plots testing seven concentrations of gibberellic acid (T1: control—not treated with GA, T2: 25 mg L^{-1}, T3: 50 mg L^{-1}, T4: 75 mg L^{-1}, T5:100 mg L^{-1}, T6: 125 mg L^{-1}, T7: 150 mg L^{-1}) and three constant temperatures (25, 30 and 35 °C), with 4 replicates, each of 50 seeds. The data were submitted to analysis of variance at the 5% level of significance, and for the variables that presented significant difference the regression analysis was performed. The statistical program SISVAR® was used (Ferreira, 2000).

3. Results and Discussion

The seeds presenteds 6% of water contente and after conditioning it was sought to maintain the same value by drying them, with the aim that only the exogenous application of gibberellic acid would interfere with the variables analyzed.

A significant effect was observed between the concentrations of gibberellic acid and the temperatures tested for the analyzed variables, except for the percentage of abnormal seedlings (Table 1).

Table 1. Analysis of variance for variables—germination (G), first count (FC), abnormal seedlings (AS), root lenght (RL)

	Variables				
	G	FC	AS	RL	SL
MS	274.71**	195.61**	14.64ns	0.693**	0.071**
Average	82.00	75.00	4.00	4.01	2.05
CV1(%)	4.55	5.38	22.30	5.73	5.01
CV2 (%)	2.66	3.73	16.89	4.67	4.55

Note. CV1 = Coefficient of variation 1; CV2 = Coefficient of variation 2; MS = middle square; (ns): not significant; (*) significant at the level of 5% ($p < 0.05$); (**) significant at the level of 1% ($p < 0.01$).

It was observed an increase in the germination rate of lettuce seeds when submitted to 25 °C and 30 °C until the concentration of 50 mg L^{-1} of GA. While at 35 °C this growth was constant reaching its maximum (84%) at 125 mg L^{-1} concentration of GA (Figure 1).

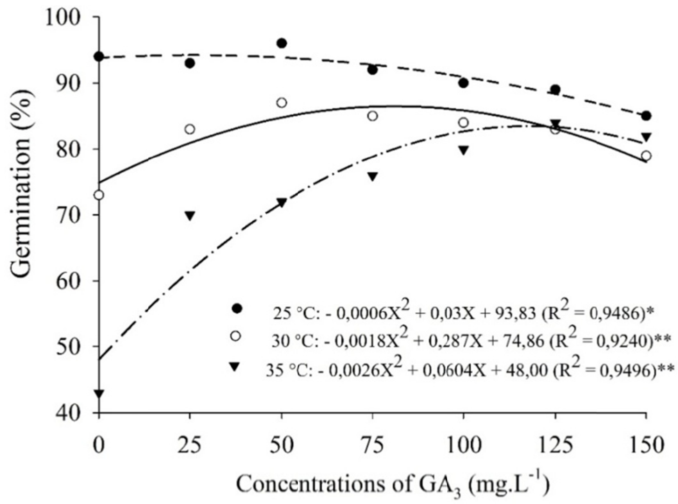

Figure 1. Germination of lettuce seeds treated with gibberellic acid and submitted to temperatures (**: significant at 1%, *: significant at 5%)

The genotype used is classified as intermediate for temperature tolerance which means that even exposed to 28 °C its germination is not impaired. This could be verified from the results obtained for the seeds that were not treated with GA (control), which same, when conditioned at 25 °C the germination percentage was 94%. As the temperature increased, this rate reduced, reaching 73 and 43%, to 30 and 35 °C, respectively, not reaching the minimum value considered for the commercialization of this species which is 80%.

In studies carried by Nascimento et al. (2012), when evaluating the germination of different lettuce genotypes under high temperatures, a decrease in the germination rate from 20 to 35 °C was observed, reaching null germination at 30 °C for the most of the tested genotypes. According to Catão et al. (2014) this reduction may be associated to the thermodormity, because the seeds soak, but the root protrusion do not occur, due to the lower speed of water and oxygem absorption by the seeds of lettuce under high temperatures (Franzin et al., 2004).

Temperatures of 25 and 30 °C showed an increase in germination rate when treated with GA up to 50 mg L^{-1}, proving that even at temperatures considered as optimal for germination, the treatment was effective. At 35 °C, the maximum germination rate (84%) war reached at the concentration of 125 mg L^{-1}, demonstrating that GA is capable of reducing the damage caused by elevated temperature.

Some authors have also reported the influence of the application of gibberellic acid on the increase of the germination of some crops, *e.g.* in wheat seeds the treatment with gibberellic acid (200 mg L^{-1}) stimulated the overcoming dormancy, improving the seed performance (Tunes et al., 2011). Aragão et al. (2003) found that stored corn seeds, when pre-soaked in a solution of 50 mg L^{-1} of GA_3, showed higher metabolic activity and, therefore, higher germination and vigor when compared to the control (not treated with GA). Braun et al. (2010) evaluating the germination of beet seeds in culture medium, concluded that immersion of these in 1.0 mg L^{-1} of GA_3 provided a higher germination rate when compared to those imbibed only in water. Rego et al. (2018) concluded that the soaking of graviola seeds at a maximum concentration of 140 ppm of gibberellic acid besides helping to overcome dormancy, increases the germination and the rate of germination speed. In brachiaria seeds, Silva et al. (2013) recommend the use of 62 and 57 mg L^{-1} of GA_3 for the cultivars "Marandu" and "MG 5", respectively.

From the concentration of 75 mg L^{-1} of gibberellic acid at temperatures of 25 and 30 °C, reductions in the germination rate of the seeds were observed. Results were similar to those obtained by Santos et al. (2013) who observed a reduction in germination of yellow passion fruit seeds when pre-soaked in solutions with concentrations superior than 100 mg L^{-1} of gibberellic acid.

The first germination count (Figure 2) presented similar pattern to germination, which was negatively affected by temperatures above 25 °C, the highest values of this variable being observed at the temperature of 25 °C, while the lowest values at 35 °C.

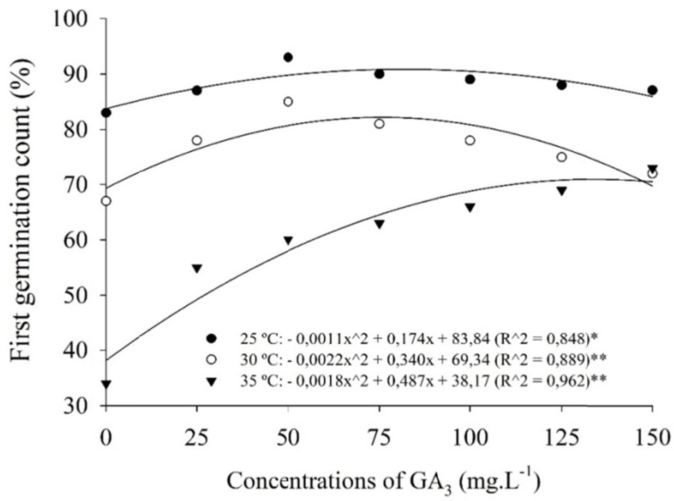

Figure 2. First germination count of lettuce seeds treated with gibberellic acid and submitted to temperatures (**: significant at 1%, *: significant at 5%)

Observing the figure 2 there was an increase of the first count at all temperatures tested from the GA application. At 25 and 30 °C the highest values were found at the concentration of 50 mg L^{-1} and the values decreased as the GA concentrations increased. At the temperature of 35 °C the dose of 150 mg L^{-1} provided the best results but they still lower when compared with those obtained for 25 and 30 °C.

Aragão et al. (2003) observed a reduction in the first count of the germination test and an increase in the number of abnormal seedlings when corn seeds were pre-soaked in gibberellic acid solution of 100 mg L^{-1} concentration. Peixoto et al. (2011) afirm that the use of gibberellin at the dose of 100 μL L^{-1} applied to castor bean (*Ricinus communis* L.) seeds of BRS 188 Paraguaçu cultivar, stimulated the percentage of first count, emergence speed index and percentage of seedling emergence.

The treatment with GA increased the root length of seedlings submitted to temperatures of 25 and 30 °C up to the concentrations of 75 mg L^{-1}. At 35 °C, the concentration of 150 mg L^{-1} promoted a higher growth of the root system providing a increase of 113% in relation to the control (not treated with GA) (Figure 3).

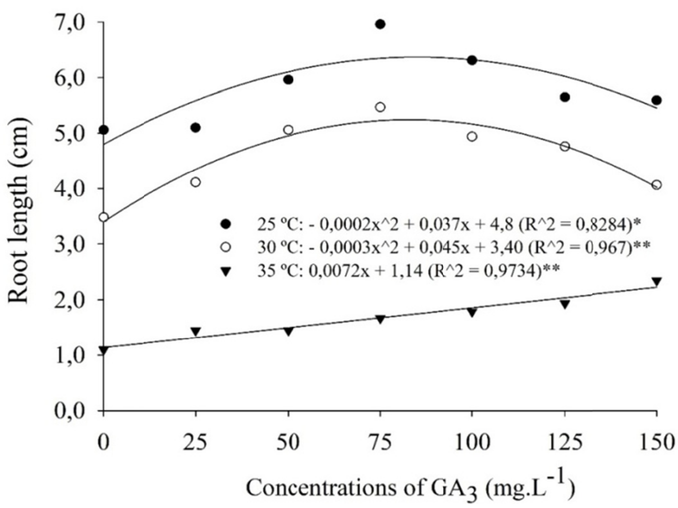

Figure 3. Root length of lettuce seedlings obtained from seeds treated with gibberellic acid and submmited to temperatures (**: significant at 1%, *: significant at 5%)

The efficiency of the use of gibberellic acid in the increase of root size was also found by Lima et al. (2009), who observed shorter lengths when passion fruit seeds were not treated with GA3, while in laboratory conditions the immersion of the seeds in solution with concentration of 500 mg L^{-1} resulted in roots with higher length.

The temperature impaired the root growth of lettuce seedlings. For the temperatures of 25 and 30 °C the root growth presented better results when submitted to the treatments with gibberellic acid, due to its stimulating effect on elongation and cell division. However, the temperature of 35 °C presented the smallest average values for length, even if the treatment with GA promoted the increase in these values, although it did not reach growth similar to that presented for the other temperatures.

Similar behavior was observed in shoot length, where the highest values were obtained at temperatures of 25 and 30 °C, while the lowest values were observed at 35 °C (Figure 4).

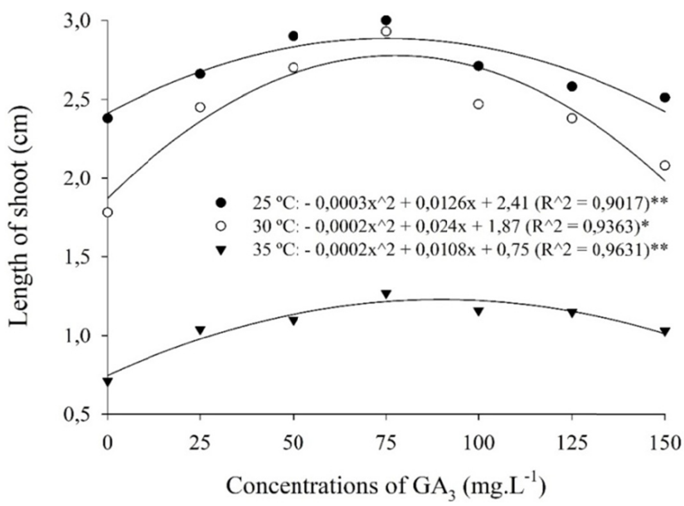

Figure 4. Shoot length of lettuce seedlings obtained from seeds treated with gibberellic acid and submitted to temperatures (**: significant at 1%, *: significant at 5%)

The length of the shoot length had the best values at the temperature of 25 °C, observing an increment for this characteristic after the application of gibberellic acid. At 30 °C it was possible to observe an increase in the values using concentrations of GA obtaining the maximum length when treated with 75 mg L^{-1}. It was verified that high temperatures cause damage to root growth, and when submitted to 35 °C, the use of gibberellic acid was not able to soften these damages.

Linear increases in seedlings growth under the elevation of gibberellin concentrations have been reported by Campos et al. (2015), in biribá seeds for which 1000 mg L^{-1} provided a greater development of both aerial and root parts, with 8.5 and 8.2 cm, respectively. Peixoto et al. (2011) observed increases in root length, shoot size and in the whole plant when castor bean seeds were conditioned in increasing concentrations of gibberellic acid, reaching the maximum when submitted to 100 μL L^{-1}, while declining in higher concentrations.

4. Conclusion

The germination and first count when submitted at 25 and 30 °C presented an increase up to 50 mg L^{-1} of GA, while at 35 °C this growth was constant until 125 mg L^{-1} of GA. Treatment with GA increased root and shoot length. Giberelic acid mitigated the damage caused to lettuce seeds when subjected to temperatures above the optimum.

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