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# Effect of gibberellic acid-3 and 6-benzylaminopurine on dormancy and sprouting of potato (Solanum tuberosum L.) tubers cv. Diacol Capiro

Efecto del ácido giberélico-3 y 6-bencilaminopurina sobre el reposo y brotación de tubérculos de papa (*Solanum tuberosum* L.) cultivar Diacol Capíro

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## **ABSTRACT**

The duration of the dormancy period in tubers is a determining factor in the planning of potato planting and production. The effect of two plant growth regulators on the dormancy period and sprouting of cv. Diacol Capiro tubers was evaluated in this study. The experiment was carried out under storage conditions (15°C and 75% RH) using a completely randomized block design with 3×3×3 factorial arrangement. The factors were: gibberellic acid-3 (GA<sub>3</sub>) and 6-benzylaminopurine (6BAP) (0, 25, and 50 mg L<sup>-1</sup>) and the immersion time (iT) (10, 60, and 120 min). The application of GA<sub>3</sub> and iT had an effect on dormancy breakage; the treatments with 25 mg L<sup>-1</sup> GA<sub>3</sub> and 60 min of immersion were enough to reduce dormancy by 18 d (35%) compared to untreated tubers. The factor GA<sub>3</sub> increased tuber weight loss (10.2%), generated sprouts with higher weight (25.6-28.4%), higher length growth rate (42.3%), and lower dry matter content (21.8-28.4%), and it increased secondary sprouting (36.2-57.9%) in comparison with untreated tubers. This way, despite the treatments with 25 mg L-1 GA<sub>3</sub> reduced the dormancy period, this dose generated sprouts more susceptible to mechanical damage. The treatments with 6BAP did not significantly affect the evaluated variables.

**Key words:** tuber potato seed, cytokinins, gibberellins, growth regulators, sprouts.

# RESUMEN

La duración del periodo de reposo en tubérculos es un factor determinante en la planeación de las siembras y la producción de papa. En este estudio se evaluó el efecto de dos reguladores de crecimiento sobre el periodo de reposo y la brotación de tubérculos de papa cultivar Diacol Capíro. El experimento se realizó en condiciones de almacenamiento (15°C y 75% HR) con un diseño de bloques completamente al azar en arreglo factorial 3×3×3. Los factores fueron: ácido giberélico-3 (AG<sub>3</sub>) y 6-bencilaminopurina (6BAP) (0, 25 y 50 mg L<sup>-1</sup>) y el tiempo de inmersión (Ti) (10, 60 y 120 min). La aplicación de AG<sub>3</sub> y Ti tuvo efecto en interrumpir el periodo de reposo y tratamientos con 25 mg L<sup>-1</sup> de AG<sub>3</sub> y 60 min de inmersión fueron suficientes para disminuir el reposo en 18 días (35%) con respecto a los tubérculos no tratados. El factor AG<sub>3</sub> aumentó la pérdida de peso de los tubérculos (10.2%), generó brotes de mayor peso (25.6-28.4%), con mayor tasa de crecimiento en longitud (42.3%), con menor contenido de masa seca (21.8-28.4%) y aumentó la brotación secundaria (36.2-57.9%) con respecto a tubérculos no tratados. De esta forma, a pesar de que los tratamientos con 25 mg L<sup>-1</sup>de AG<sub>3</sub> redujeron el periodo de reposo, esta dosis generó brotes más susceptibles al daño mecánico. Los tratamientos con 6BAP no afectaron significativamente las variables evaluadas.

**Palabras clave:** tubérculo semilla de papa, citoquininas, giberelinas, reguladores de crecimiento, brotes.

## Introduction

The potato (*Solanum tuberosum* L.) is one of the most important horticultural crops in the world and the fourth most consumed source of carbohydrates for humans after rice, wheat, and corn. Potatoes have high nutritional value and they are consumed in both fresh and processed form (Devaux *et al.*, 2014; Navarre and Pavek, 2015). In Colombia, Diacol Capiro is one of the cultivars with the largest planted areas. It is also the main cultivar (cv.) used for processing due to its high productivity and frying quality

(Ñústez, 2011). Dormancy is a factor that can limit seed availability when establishing new crops (Mustefa *et al.*, 2017; Deligios *et al.*, 2020). The most common strategy for breaking dormancy is storage in warehouses (Jansky and Hamernik, 2015). The dormancy period for cv. Diacol Capiro could last up to three months at 15°C and 75% RH (Ñústez, 2011). This affects the costs for seed producers.

The most common potato propagation method is the vegetative one with seed tubers. The quality of seed tubers is linked to different parameters, such as the physiological

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age of the tuber, its dormancy state and sprouting ability. These are variables that affect the yield potential of the crop (Mikitzel, 1993; Knowles and Knowles, 2006; Struik, 2007; Blauer et al., 2013). Dormancy is the absence of visible sprout growth, even under adequate conditions for it to occur (Suttle, 2007). It is a period that begins simultaneously with the tuber filling at the mother plant and ends with sprouting (Claassens and Vreugdenhil, 2000; Nambara and Marion-Poll, 2005). Dormancy is a plant strategy used to escape from hostile conditions, and this is very important in sessile organisms like plants to ensure their survival (Henis, 1987; Suttle, 2007). In tubers, dormancy is present in the meristems, i.e. buds (Lang et al., 1987; Sonnewald and Sonnewald, 2014), while the rest of the tuber remains metabolically active (Viola et al., 2007; Sergeeva et al., 2012). Viola et al. (2007) establish the presence of at least one sprout of 2 mm length or greater as a criterion to determine the end of the tuber dormancy period. Dormancy breakage begins with the reactivation of the symplastic pathway in the tuber that regulates the distribution of energy reserves towards the meristematic tissues, allowing their activation and sprout development (Viola et al., 2007).

The sprouting process begins with the development of the apical sprout that becomes dominant and inhibits the development of lateral sprouts (Carli et al., 2010). This state is called "apical dominance" or "paradormancy" (Lang et al., 1987; Sonnewald and Sonnewald, 2014). Dominance decreases gradually with the increase of the tuber's physiological age and ends in a state of multiple sprouting that is considered adequate for seed sowing (Krijthe, 1962; Teper-Bamnolker et al., 2012; Blauer et al., 2013; Eshel, 2015). The dormancy period varies according to components, such as genotype, environmental conditions, agronomic management of the crop, storage, and endogenous hormonal dynamics of the tuber (Carli et al., 2010; Sonnewald and Sonnewald, 2014; Eshel, 2015; Christensen et al., 2019; Deligios et al., 2020).

During dormancy the levels of plant growth inhibitors, such as abscisic acid and ethylene, are high. These are the main hormones involved in dormancy initiation and maintenance (Carli *et al.*, 2010; Mani *et al.*, 2014; Sonnewald and Sonnewald, 2014; Deligios *et al.*, 2020). When dormancy is overcome and sprouting begins, the hormonal dynamics change and the concentrations of growth promoters such as cytokinins, gibberellins (Mani *et al.*, 2014) and auxins increase, although the role of auxins is less clear than the other two (Suttle, 2004; Carli *et al.*, 2010).

Different studies have shown that the exogenous contribution of plant growth regulators can increase efficiency of dormancy breakage and potato seed tuber sprouting. The use of exogenous regulators, such as gibberellins (Jansky and Hamernik, 2015; Mustefa et al., 2017; Chindi and Tsegaw, 2019; Christensen et al., 2019; Deligios et al., 2020), cytokinins (Turnbull and Hanke, 1985; Suttle, 2008; Campbell et al., 2014) or their combination (Alexopoulos et al., 2007) have generated positive responses. However, most of the available information is limited to cultivars of the Group Tuberosum, which is not a group of potatoes planted in Colombia (Huaman and Spooner, 2002). The objective of this research was to evaluate the effect of the application of gibberellic acid-3 and 6-benzylaminopurine on tuber dormancy and sprouting in potato cv. Diacol Capiro seed tubers at different immersion times.

# Materials and methods

## Plant material and experiment establishment

The experiment was carried out at the Universidad Nacional de Colombia Bogota campus, using seed tubers under storage conditions with diffuse light, an average temperature of 16°C and 40-80% relative humidity. Potato cv. Diacol Capiro tubers were used 10 d after harvest. The tubers were washed and classified into three categories (I: 90-110 g, II: 70-89 g, and III: 50-69 g). The tubers that showed morphological changes or some type of damage were discarded.

A completely randomized block design with factorial arrangement (3×3×3) was used. The factors were: gibberellic acid-3 (GA<sub>3</sub>), 6-benzylaminopurine (6BPA), at concentrations of 0 mg L<sup>-1</sup>, 25 mg L<sup>-1</sup>, and 50 mg L<sup>-1</sup>, and immersion times (iT) of tubers in the solutions at 10, 60, and 120 min. A control without treatment was used for the variable "dormancy period". The experimental unit (EU) included 20 tubers for the variables: dormancy period, weight loss, number of sprouts and secondary sprouting, and five tubers for the variables apical sprout growth, sprout weight and dry matter. Tuber size was used as a blocking factor.

Seed tubers were placed on plastic meshes and submerged in solutions with the different hormone treatments. The tubers were then removed at different immersion times. Subsequently, the tubers were allowed to dry, randomly arranged inside plastic baskets that were stacked in towers per block. The baskets were randomized per height twice a week and stored until the end of the trial, 100 d after applying the treatments (DAT).

### **Evaluated variables**

## Dormancy period

The tubers that showed at least one bud with a sprout of 2 mm length or greater were counted and recorded twice a week. With this information, the number of days necessary to obtain 80% of sprouting tubers was determined. This number was considered as the breakage of the dormancy period.

#### Weight loss and number of sprouts

Tuber weight was determined using a milligram precision balance (XB220A, Precisa, Switzerland) at 8 DAT (initial weight) and 100 DAT (final weight), and the percentage of weight loss was calculated with these data. The number of sprouts was estimated as the total number of sprouts with sufficient vigor to survive in the field at 100 DAT.

## Apical sprout growth

Five tubers were randomly selected and the sprouts of the apical zone were individually marked, and their length and diameter measured at 16, 24, 32, 39, 52, 60, 73, and 90 DAT. The tuber growth rate in terms of length and diameter was estimated using the obtained data.

## Sprout weight and dry matter percentage

At 100 DAT, all sprouts from five tubers were removed, weighed (fresh weight), and then brought to constant weight in an oven (Thelco Model 16, Precision Scientific Company, Chicago, USA) at 70°C (dry weight). The dry matter percentage was then determined.

## Secondary sprouting

The number of tubers that showed secondary sprouting was quantified. The secondary sprouts are characterized by the presence of small and thin sprouts in the primary sprout (Krijthe, 1962).

#### Statistical analysis

The data were analyzed with the R statistical package (version 3.5.1), using an ANOVA to determine the effect of the factors on parametric variables and the Tukey test for a comparison of means ( $P \le 0.05$ ). For the variable sprout growth (length and width), a generalized linear gamma model with repeated measures was performed based on the methodology of Dobson and Barnett (2008).

# **Results and discussion**

## **Dormancy period**

For this variable, statistically significant differences were observed for  $GA_3$ , iT and the  $GA_3 \times iT$  interaction (Tab. 1).

The response in the dormancy period was different between  $GA_3$  concentrations. Without  $GA_3$  application, the dormancy period was 42.2 d on average, whereas with 25 mg  $L^{-1}GA_3$  it was 32.8 d and with 50 mg  $L^{-1}GA_3$  it was 30.1 d. The three levels were statistically different from each other (Fig. 1A). The data showed that  $GA_3$  promoted the breakage of tuber dormancy in less time, with the dose of 50 mg  $L^{-1}$  as the most effective.

**TABLE 1.** Significance levels of ANOVA results of the variables dormancy period (DP), weight loss (WL), number of sprouts (NS), fresh weight of sprouts (FW), dry weight of sprouts (DW), percentage of dry matter of sprouts (% DM), and secondary sprouting (SS) with the applications of different concentrations of gibberellic acid-3 (GA<sub>3</sub>), 6-benzylaminopurine (6BAP), and immersion time (iT) in *S. tuberosum* cv. Diacol Capiro tubers.

|                          | DP    | WL   | NS    | FW    | DW    | %DM  | SS    |
|--------------------------|-------|------|-------|-------|-------|------|-------|
| Block                    | *     | ns   | ns    | ***   | ***   | ***  | *     |
| GA <sub>3</sub>          | ***   | ***  | ns    | ***   | ***   | ***  | ***   |
| 6BAP                     | ns    | ns   | ns    | ns    | ns    | ns   | ns    |
| iT                       | **    | ns   | ns    | ns    | ns    | ns   | ns    |
| GA₃×6BAP                 | ns    | ns   | ns    | ns    | ns    | ns   | ns    |
| GA₃×iT                   | *     | ns   | ns    | ns    | ns    | ns   | ns    |
| 6BAP×iT                  | ns    | ns   | ns    | ns    | ns    | ns   | ns    |
| GA₃×6BAP×iT              | ns    | ns   | ns    | ns    | ns    | ns   | ns    |
| Variation<br>coefficient | 17.3% | 6.7% | 18.5% | 35.1% | 32.5% | 8.6% | 32.1% |

<sup>\*\*\*</sup>  $P \le 0.1\%$ ; \*\* 1%; \* 5%; ns: non-significant differences.

The dormancy breakage response in the tubers due to the exogenous application of GA<sub>3</sub> has been reported in other studies with different cultivars of the Group Tuberosum, such as 'Red Pontiac' (Guzmán, 1963), 'Agria' (Salimi et al., 2010; Barani et al., 2013; Mohammadi et al., 2014), 'Marfona' (Salimi et al., 2010; Barani et al., 2013), 'Solara' (Hartmann et al., 2011), 'Draga' (Barani et al., 2013), 'Born' (Mohammadi et al., 2014), 'Bubu', 'Bate' (Mustefa et al., 2017), 'Pasat', 'Dorota' (Wróbel et al., 2017), 'Shepody' (Dean et al., 2018a), 'Spunta', 'Monalisa', 'Europe', 'Arinda' (Deligios et al., 2020) and 'Payette', 'Russet' and 'Alturas' (Dean et al., 2018b). However, the available information in Andigenum cultivars is scarce. Alexopoulos et al. (2007) evaluated treatments with GA<sub>3</sub>, 6BAP and their combination at different temperatures in the Andigenum cv. Chacasina, applying the solution directly to the tuber-stolon junction. The authors found that GA<sub>3</sub> treatments and the combination of GA<sub>3</sub> and 6BAP reduced the dormancy period and showed the synergic effect of the two plant growth regulators.

Treatment with iT resulted in statistically significant responses between all the evaluated times as compared to the untreated control (49 d), showing the effect of humidity at

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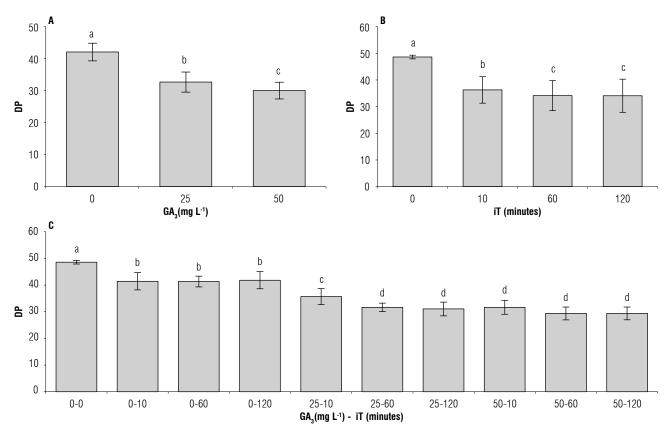
the beginning of the sprouting processes (Fig. 1B). The 10 min iT generated a dormancy reduction of 12 d compared to the untreated control. It also differed statistically from 60 and 120 min (14 d), which did not differ from each other (Fig. 1B). These results suggested that, for the cv. Diacol Capiro, 60 min were enough to generate the dormancy breakage response in the evaluated treatments. This result was similar to that reported by Alexopoulos et al. (2008) in the Andigenum cv. Chacasina, in which 120 min was the optimum time of the treatment. It also underlines that the iT has more influence than the plant growth regulator dose, possibly due to the higher amount of solution that enters the tuber. There were interactions between GA3 and iT (Fig. 1C) in which GA<sub>3</sub>-iT combinations 25-60, 25-120, 50-10, 50-60 and 50-120 reduced the dormancy period by 18 d on average compared to non-treated tubers, and 11 d compared to tubers immersed in water (Fig. 1C).

There were no effects of the treatments with 6BAP on the tuber dormancy period. Different studies have shown that the effectiveness of treatments with cytokinins as dormancy disrupters in potato varies with conditions such as tissue

sensitivity that is greater in less deep stages of dormancy (Turnbull and Hanke, 1985; Suttle, 2001; Rodríguez and Moreno, 2010; Hartmann *et al.*, 2011), genetic component, and type of cytokinins used (Suttle, 2008).

## Weight loss

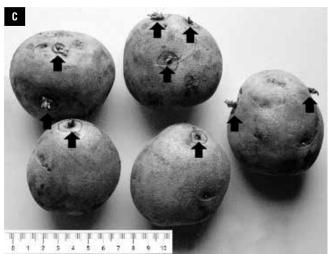
Only the GA<sub>3</sub> treatment showed significant differences in the tuber weight loss (Tab. 1). At 100 d of storage, the treatments with 50 mg L-1 GA3 showed a lower weight loss percentage (4.4%) than the one obtained with 25 and 50 mg L-1 (4.9%). The last two had no differences between them (Fig. 3). Weight loss is a response associated with the degree of tuber longevity that increases when sprouting is higher, due to the greater permeability of the cell membranes of the sprouts compared to other tuber tissues (Carli et al., 2010). The response is similar to that reported in cultivars Andigena of the subspecies andigena, such as 'Chacasina' (Alexopoulos et al., 2007; Alexopoulos et al., 2008) and the Group Tuberosum, such as 'Agria' (Rentzsch et al., 2012), and 'Bubu' and 'Bate' (Mustefa et al., 2017), in which weight loss is related to an increase in the respiratory rate and sprout development in GA-treated tubers.



**FIGURE 1.** Duration of the tuber dormancy period in days (DP) in response to A) gibberellic acid-3 (GA<sub>3</sub>), B) immersion time (iT), and C) the interaction  $GA_3 \times iT$ , in the cv. Diacol Capiro. The data shown is the number of days needed to reach breakage in 80% of the tubers after the application of the treatments. Standard deviations are indicated by vertical bars. Different letters above the bars indicate significant difference at 95% confidence, according to the Tukey test. N: A/B = 27; C = 9.

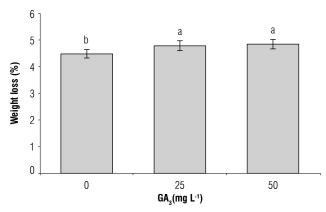








**FIGURE 2.** Sprout development in A) untreated, B) treated with water for 10 min, C) treated with a 25 mg  $L^{-1}$  gibberellic acid-3 (GA<sub>3</sub>) solution for 10 min, and D) treated with a 50 mg  $L^{-1}$  GA<sub>3</sub> solution for 10 min potato (*S. tuberosum*) cv. Diacol Capiro tubers 39 days after treatment. Arrows show the buds that have sprouts.



**FIGURE 3.** Effect of different of gibberellic acid-3 ( $GA_3$ ) concentrations on the weight loss percentage in potato cv. Diacol Capiro tubers at 100 DAT. Standard deviations are indicated by the vertical bars. Different letters above the bars indicate significant difference at 95% confidence, according to the Tukey test. N = 27.

## Number of sprouts

In this study, none of the treatments affected the number of sprouts per tuber (Tab. 1), confirming Alexopoulos *et al.* (2007). Studies with GA<sub>3</sub> applications at low concentrations (20 mg L<sup>-1</sup>) in cultivars of the subspecies show an increase in the number of vigorous sprouts per tuber (Mustefa *et al.*, 2017). Concentrations between 1 and 2 mg L<sup>-1</sup> GA<sub>3</sub> in tuber seeds increase the number of stems and the percentage distribution of tubers harvested by size in the crop (Herman *et al.*, 2016; Wróbel *et al.*, 2017; Dean *et al.*, 2018a; Dean *et al.*, 2018b).

## Growth in apical sprout length

The applied treatments affected the length of sprouts (Tab. 2). Taking into account the DEVIANCE model used, the differential effect of the DAT (time factor) and their

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interactions with each of the evaluated factors were identified as follow:  $GA_3 \times DAT$ ,  $6BAP \times DAT$  and  $Ti \times DAT$  (Tab. 2). For this reason, the analysis is presented as repeated measures over time.

**TABLE 2.** Significance levels of DEVIANCE analysis of growth variables in apical sprouts expressed in terms of growth rate in length (L) and diameter (D), treated with different concentrations of gibberellic acid-3 (GA $_3$ ) and 6-benzylaminopurine (6BAP) and immersion time (iT), in *S. tuberosum* cv. Diacol Capiro tubers.

|                            | L   | D   |   | L   | D   |
|----------------------------|-----|-----|---|-----|-----|
| Size (Block)               | ns  | *** | DAT                                     | *** | *** |
| $GA_3$                     | *** | *** | GA <sub>3</sub> ×DAT                    | *** | *** |
| 6BAP                       | ns  | ns  | 6BAP×DAT                                | *   | ns  |
| iT                         | *** | ns  | iT×DAT                                  | *** | ns  |
| $GA_3 \times 6BAP$         | *   | ns  | GA <sub>3</sub> ×6BAP×DAT               | *   | **  |
| $GA_3 \times iT$           | ns  | *** | GA <sub>3</sub> ×iT×DAT                 | *   | ns  |
| 6BAP×iT                    | ns  | *   | 6BAP×iT×DAT                             | ns  | ns  |
| $GA_3{	imes}6BAP{	imes}iT$ | ns  | ns  | $GA_3 \times 6BAP \times iT \times DAT$ | ns  | ns  |

GA<sub>3</sub> (Factor): Gibberellic acid-3; 6BAP (Factor): 6-benzylaminopurine; lt: Immersion time; DAT (Time factor): Days after treatment; \*\*\*  $P \le 0.1\%$ ; \*\* 1%; \* 5%; ns: non-significant differences.

The  $GA_3$  treatments affected the growth rate of sprouts (Fig. 4A). Treatments with 0 mg  $L^{-1}$  had the lowest growth rate (0.15 mm/d) compared to treatments with 25 mg  $L^{-1}$  (0.26 mm/d) and 50 mg  $L^{-1}$  (0.26 mm/d) without statistically significant differences between the last two. The sprout length at 90 DAT for 50 mg  $L^{-1}$  (2.2 cm) was 83% higher than that in treatments without  $GA_3$  (1.2 cm), while in treatments with 25 mg  $L^{-1}$  (2.0 cm) sprout length increased by 66% (Fig. 4B). When observing the effect of 6BAP and iT,  $GA_3$  had the greatest effect on the growth of apical sprouts results (Alexopoulos *et al.*, 2008) in the cv. Chacasina.

The treatment with 6BPA did not affect the growth rate or length of sprouts during the evaluation period (Figs. 4C and 4D). The factor iT showed differences in sprout length throughout the evaluation time (iT×DAT) (Tab. 2). The growth rate was higher with 60 min (0.25 mm/day) and differed from treatments of 10 min (0.22 mm/d), whereas treatments of 120 min (0.24 mm/d) did not differ from the previous ones (Fig. 4E). However, the length values at 90 DAT were not very different from each other. They showed the lowest value at 10 min (17 mm), followed by 120 min (18 mm) and 60 min (19 mm), with maximum differences of 11% in length (Fig. 4F).

The interaction between GA<sub>3</sub>xiTxDAT and GA<sub>3</sub>x6BAPx-DAT affected the length and growth rate of tuber sprouts (Tab. 2). These data suggest that the factor that had the

greatest influence was  $GA_3$ , since treatments with  $50 \text{ mg L}^{-1}$   $GA_3$ , regardless of their combination with other treatments, generated 3 of the 4 highest sprout lengths (22.6 mm). The highest  $GA_3$  concentrations together with the combination 25-60 ( $GA_3$ -iT) showed no differences in growth rate (Fig. 5A). The other treatments with 25 mg L<sup>-1</sup>  $GA_3$  (25-120, 25-10) resulted in higher sprout length averages (18.5 mm) and higher growth rates than the treatments with 0 mg L<sup>-1</sup>  $GA_3$  (Fig. 5B). There were no differences in sprout length at 90 DAT or in the growth rate of the treatments with 0 mg L<sup>-1</sup>  $GA_3$ , indicating that the effect of iT requires the presence of  $GA_3$  (Fig. 5B).

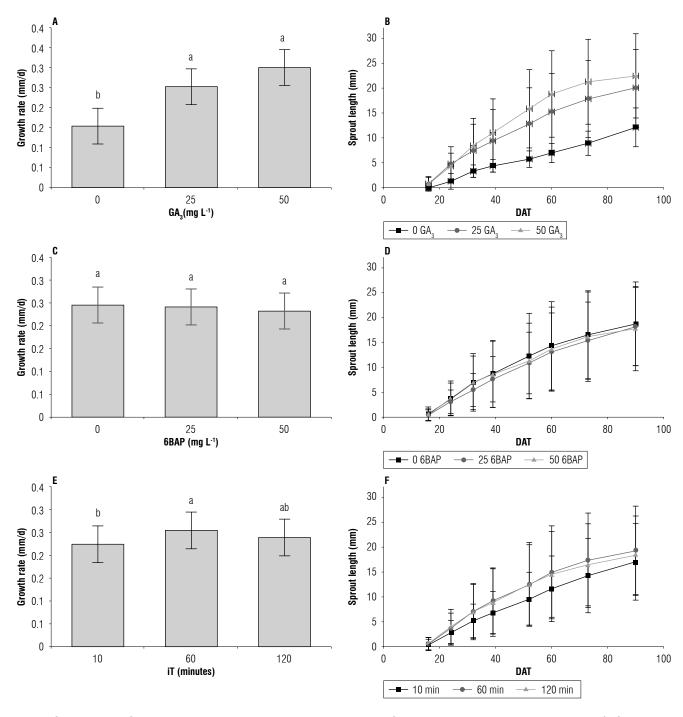
The  $GA_3 \times 6BAP \times DAT$  interaction showed differences in sprout length at 90 DAT. Only the 50-0 treatment ( $GA_3$ -6BAP) differed statistically in the growth rate (Fig. 5C) and showed the greatest length (24.5 mm), 49% higher than the average of the treatments with 0 mg L<sup>-1</sup>  $GA_3$  (12.1 mm) (Fig. 5D).

# Growth in diameter of the apical sprout

With the statistical model used, the effects of DAT and its interaction with GA<sub>3</sub> and the interaction GA<sub>3</sub>×6BPA (Tab. 2) were identified. The GA<sub>3</sub> affected the diameter growth rate of the sprouts. The treatments with 0 mg L<sup>-1</sup> had the highest growth rate (0.073 mm/d), significantly higher compared to 50 mg L<sup>-1</sup> (0.045 mm/d) (Fig. 6A). At the end of the evaluation (90 d), the application of 50 mg L<sup>-1</sup> GA3 generated smaller sprout diameter at levels 25 and 0 mg L<sup>-1</sup>, respectively (Fig. 6B). This coincides with Alexopoulos *et al.* (2008).

We saw no interaction between  $GA_3$  and 6BAP in the sprout diameter, but we observed the growth rate as a product of the interaction of the factors  $GA_3 \times 6BAP \times DAT$  (Tab. 2, Fig. 6C). The treatment with 0 mg  $L^{-1}$   $GA_3$  and with 6BAP did not differ from the treatment with 0 mg  $L^{-1}$   $GA_3$  and without 6BAP in the diameter growth rate (Fig. 6C), but the diameter in length was superior to the other treatments with different  $GA_3$  and 6BAP combinations. This result suggests a negative effect of  $GA_3$  on sprout diameter (Fig. 6D), but a positive effect on length.

Jansky and Hamernik (2015) observed similar results in the growth of sprouts in tubers treated with  $GA_3$ . They found increases in sprout elongation with doses of up to 1000 mg  $L^{-1}GA_3$  and attributed this effect to the role this plant growth regulator plays in cellular elongation processes. The development of long and/or thin sprouts is a characteristic that can affect the quality of the tuber as seed by making sprouts more susceptible to damage by handling during transport and planting (Virtanen *et al.*, 2013).



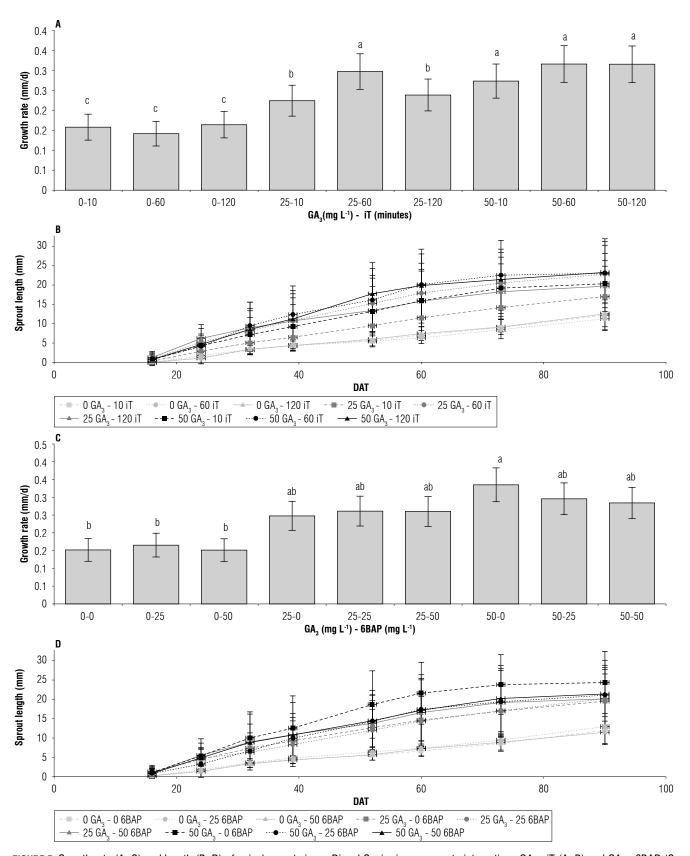
**FIGURE 4.** Growth rate (A, C, E) and length (B, D, F) of apical sprouts in cv. Diacol Capiro in response to factors gibberellic acid-3 ( $GA_3$ ) (A, B), 6-benzylaminopurine (GBAP) (C, D) and immersion time (iT) (E, F) 90 days after treatment (DAT). Data shown in A, C and E correspond to the growth rate during the evaluation period. Vertical bars correspond to the standard deviation. Different letters above the bars indicate a significant difference at 95% confidence, according to the Tukey test. N: A/C/E = 135 per point, B/D/F = 27.

## Sprout weight and dry matter

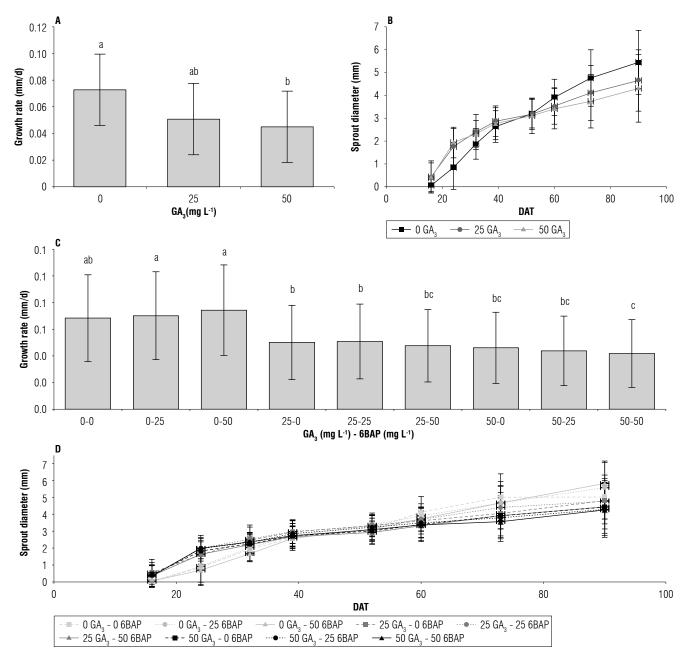
Significant differences were only found for the growth of sprouts (Tab. 1, Fig. 7) represented by fresh weight (FW), dry weight (DW) and dry matter percentage (DM) in response to GA<sub>3</sub> application. The FW and DW of the sprouts were significantly lower for treatments with 0 mg L<sup>-1</sup> GA<sub>3</sub>

(3.02 g, 0.68 g) compared to 25 mg L<sup>-1</sup> (4.06 g, 0.87 g) and 50 mg L<sup>-1</sup> (4.54 g, 0.95 g), but there were no significant differences between these last two (Fig. 8A, Fig. 8B). These results are similar to those observed by other authors in different cultivars (Alexopoulos *et al.*, 2007; Salimi *et al.*, 2010; Mustefa *et al.*, 2017).

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**FIGURE 5.** Growth rate (A, C) and length (B, D) of apical sprouts in cv. Diacol Capiro in response to interactions  $GA_3 \times iT$  (A, B) and  $GA_3 \times 6BAP$  (C, D) 90 days after treatment (DAT). Data shown in A and C correspond to the growth rate during the evaluation period. Vertical bars correspond to the standard deviation. Different letters above the bars indicate significant difference at 95% confidence, according to the Tukey test. N: A/C/E = 45 per point; B/D/F = 9.



**FIGURE 6.** Growth rate (A, C) and diameter (B, D) of apical sprouts in the cv. Diacol Capiro in response to gibberellic acid-3 (GA<sub>3</sub>) (A, B) and the  $GA_3 \times 6BAP$  interaction (C, D) 90 days after treatment (DAT). Data shown in A and C correspond to the growth rate during the evaluation period. Vertical bars correspond to the standard deviation. Different letters above the bars indicate significant difference at 95% confidence, according to the Tukey test. N: A = 135 per point, B = 27; C = 45 per point; D = 9.

The dry matter percentage in the sprouts showed significant differences for the treatments with  $GA_3$  (Fig. 8C). Treatments with  $0 \text{ mg L}^{-1} GA_3$  obtained greater dry matter percentages (23%) than the treatments with 25 mg  $L^{-1}$  and 50 mg  $L^{-1}$  (21%) with no differences between the last two. These results suggest that sprout growth promoted by  $GA_3$  was caused by cellular elongation rather than by division. Salimi *et al.* (2010) find similar results in response to  $GA_3$ 

treatments. However, partially opposite results are reported by Alexopoulos *et al.* (2007), who find that tuber sprouts treated with GA<sub>3</sub> and 6BAP are larger, with greater dry mass percentages than in untreated tubers.

### Secondary sprouting

Secondary sprouting corresponds to the formation of thin sprouts that grow on the main sprout (Krijthe, 1962). A

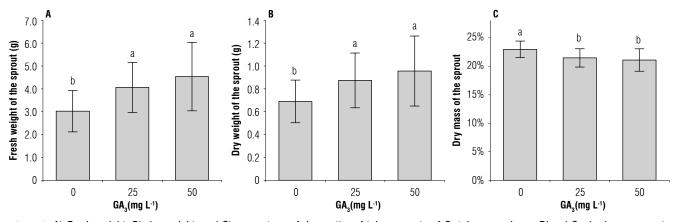
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**FIGURE 7.** Apical sprouts of potato (*S. tuberosum*) tubers cv. Diacol Capiro A) immersed in pure water for 10 min, B) treated with 25 mg L<sup>-1</sup> gibberellic acid-3 ( $GA_3$ ) for 60 min, and C) treated with 50 mg L<sup>-1</sup>  $GA_3$  for 120 min, 90 d after treatment.

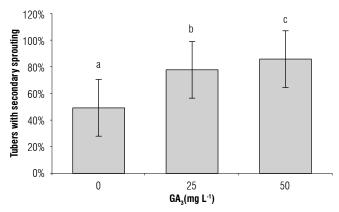


**FIGURE 8.** A) Fresh weight, B) dry weight, and C) percentage of dry matter of tuber sprouts of *S. tuberosum* L. cv. Diacol Capiro in response to different doses of gibberellic acid-3 ( $GA_3$ ). Vertical bars correspond to the standard deviation. Different letters above the bars indicate significant difference at 95% confidence, according to the Tukey test. N: A/B/C = 27.

significant effect of GA<sub>3</sub> was observed in our research (Tab. 1), increasing the percentage of occurrence when the dose was higher, with results of 51% for treatments with 0 mg L<sup>-1</sup>, 80% for 25 mg L<sup>-1</sup> and 88% for 50 mg L<sup>-1</sup> (Fig. 9). Secondary sprouting takes place in tubers with very long periods of storage time and this is associated with tuber senescence (Struik, 2007; Carli *et al.*, 2010) or high storage humidity.

Blocking in the experiment was performed by tuber size and this influenced most of the variables, because size is associated with the physiological age of the tubers (Struik, 2007; Salimi *et al.*, 2010; Mohammadi *et al.*, 2014; Christensen *et al.*, 2019). The dormancy period decreases when the tuber is larger (Knowles and Knowles, 2006). In our research, the largest tubers developed larger sprouts with a higher dry matter content and increased secondary sprouting. These data show that there is a differential response to treatments with growth regulators due to the size of the

tubers, which highlights the importance of taking this factor into account to apply GA<sub>3</sub> as a seed treatment.



**FIGURE 9.** Percentage of tubers with secondary sprouting in S. *tuberosum* L. cv. Diacol Capiro treated with different concentrations of gibberellic acid-3 (GA<sub>3</sub>). Standard deviations are indicated by vertical bars. Different letters above the bars indicate significant difference at 95% confidence, according to the Tukey test. N=27.

## **Conclusions**

The results showed that exogenous  $GA_3$  applications decrease the dormancy period of cv. Diacol Capiro tubers and their effect increases as an effect of prolonged immersion times (60 and 120 min). The treatments with 25 or 50 mg  $L^{-1}$   $GA_3$  and iT 60 min reduced the dormancy period by 35% (18 d). However, 50 mg  $L^{-1}$   $GA_3$  generated long sprouts with smaller diameter and lower DM content. This makes them more susceptible to mechanical damage and increases the occurrence of characteristics associated with advanced physiological age and affects the quality of the tuber. In this study, 6BAP showed no clear effect on dormancy breakage.

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