

## RESEARCH ARTICLE

# Productive behavior of strawberry from potted seedlings produced with application of prohexadione calcium in soilless cultivation

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## ABSTRACT

Use of national strawberry seedlings, produced in a system without soil, is an alternative to reduce the dependence of producers on using imported seedlings. The objective of this work was to evaluate the effect of applying different concentrations of prohexadione calcium (ProCa) in the growth control of strawberry seedlings and the productive behavior after planting these seedlings. The experiment was carried out in a greenhouse, with a 4 x 2 factorial scheme, four concentrations of prohexadione calcium (0, 150, 300 and 600 mg L<sup>-1</sup>) and two cultivars (Aromas and Camarosa). In the seedling phase, a completely randomized design was used, with 4 replications, with the experimental unit consisting of 12 seedlings. In the post-planting phase, a completely randomized design was used, with 5 replications and the experimental unit consisted of 4 plants. In the seedlings, the following were evaluated: number of leaves, crown diameter, petiole length, leaf area, chlorophyll content and dry matter of the aerial part. The parameters evaluated in the strawberry plants were: full bloom and fruiting, number of fruits per plant, average fruit weight, total fruit weight, length and diameter of fruit, dry weight of leaves and crowns. The application of 150, 300 and 600 mg L<sup>-1</sup> of ProCa decreased the leaf area and petiole length for both cultivars, thus reduced the vegetative growth of the seedlings. On the other hand the chlorophyll a, b and total contents were increased in relation to the absence of ProCa for the two cultivars evaluated. The ProCa concentrations did not influence the number of fruits and the fruit mass per plant, and the results were related to the cultivar factor, where the cv Camarosa had the higher values.

**Keywords:** *Fragaria ananassa*; Fruiting; National seedlings; Plant regulator; Propagation

## INTRODUCTION

Brazil has become the largest strawberry producer (*Fragaria × ananassa* Duch.) In South America, with about 4.300 hectares cultivated with the crop and where around 155.000 tons of fruit are produced (Antunes et al., 2017), resulting in an average yield of 36 t ha<sup>-1</sup> (Zeist and Resende, 2019). The expansion of the cultivation area, with the exchange of seedlings and new plantings, demands a large number of seedlings per year. It is estimated that the annual demand of the main strawberry producing regions in the country is around 175.000.000 seedlings (Antunes and Peres, 2013).

Expenditure on importing seedlings and payment of royalties from property rights contracts, dependence on genetic materials and foreign nurseries results in increased

costs of strawberry production in Brazil (Fagherazzi et al., 2017). The obtaining of seedlings is the main limiting factor in the culture, since most of the seedlings used in Brazil are imported from Argentina, Chile and Spain (Gonçalves et al., 2016; Barreto et al., 2018).

In addition, the delay for seedlings to arrive in Brazil and delivery to the producer, makes planning and planting difficult, where in some producing regions, climatic conditions would allow the harvests to start in March, in addition to the risks associated with the possibility of introduction of pathogens and pests to seedlings (Zeist and Resende, 2019).

In this context, the production of national strawberry seedlings in an out-of-soil system allows the obtaining of

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seedlings with high vigor, in addition to the strict control of sanitary and nutritional quality, which are essential prerequisites for obtaining high fruit yields (Cocco et al., 2011; Gonçalves et al., 2016).

In the soilless system, it is possible to anticipate planting, as it allows the supply of seedlings to be staggered (Antunes et al., 2015), in addition to maintaining sanitary and physiological quality (by eliminating the incidence of soil pathogens) throughout the period of planting (March-July) (Pereira et al., 2016), which is a demand in the most diverse producing regions of the country.

On the other hand, these seedlings will be produced at the beginning of the summer and planted only at the end of this season or in the autumn of the following year, being necessary to control their growth so that the consumption of reserves does not occur before planting (Pereira et al., 2016; Barreto et al., 2018). As there is an excessive vegetative growth of the aerial part with the decrease of the accumulation of reserves in the crown and roots during this period due to the favorable environmental conditions, reducing the quality of the seedlings (Cocco et al., 2015; Pereira et al. 2016).

In order to guarantee the productivity of the orchards and to improve the quality of the fruit, it is essential in fruit plants to control vegetative growth (Carra et al., 2017), which can be carried out with the use of growth regulators, among them calcium prohexadione (calcium 3-oxo-4-propionyl-5-oxo-3-cyclohexane carboxylate), which has already been used in several crops, such as apple and pear, in the vegetative and productive control of plants (Pasa and Einhornet, 2017), in the production phase of mango (Mouco et al., 2010) and strawberry (Pereira et al., 2016) seedlings, in the flowering of the avocado to control production and fruit size (Brogio et al., 2018) for inhibiting the final stages of gibberellin biosynthesis, as it is a co-substrate of the dioxygenases that catalyze the hydroxylations involved in biosynthesis (Mouco et al., 2010; Kim et al., 2019).

The phytohormone inhibiting the biosynthesis of gibberellins prohexadione calcium (ProCa) belonging to the class of acylcyclohexanediones, which are plant hormones that regulate the plant height. Its levels are generally related to the length of the stem, but they also play an important role in other physiological phenomena, such as in floral initiation and fruit establishment (Taiz et al., 2017).

According to Kim et al. (2019), the control of vegetative growth occurs due to the prohexadione calcium acting on 3  $\beta$ -hydroxylase, an enzyme that primarily catalyzes the conversion of inactive GA 20/GA 9 into highly active GA

1/GA 4. The application of ProCa, in addition to reducing the plant height, provides an increase in the quality and yield of fruit, in addition to reducing space and labor (by reducing the size of the plants, it reduces the time to carry out maintenance pruning) (Ito et al., 2016; Kim et al., 2019).

ProCa delays the vegetative growth of plants, but does not compromise the production of strawberry fruits after planting. Because its short duration allows flexibility to manipulate growth as needed (Reekie et al., 2005a). Thus, the objective of this work was to evaluate whether the effect of different concentrations of prohexadione calcium in the production phase of strawberry seedlings influences the quality of seedlings and the productive behavior of cultivars Aromas and Camarosa after planting.

## MATERIALS AND METHODS

The experiment was carried out in an experimental area belonging to Embrapa Clima Temperado, in Pelotas, RS, Brazil (31°40' S, 52°26' W, altitude 60 m), during the years 2018 and 2019. Seedlings with clod (roots surrounded by substrate) of the cultivars Aromas and Camarosa produced in a production system soilless were used.

The propagules of the stolons were placed in 72-cell polystyrene trays (internal volume of 124 mL), containing commercial substrate Carolina Soil® (compound based on Sphagno peat, expanded vermiculite, dolomitic limestone, agricultural plaster and NPK fertilizer, with pH:  $5.5 \pm 0.5$ ; electrical conductivity:  $0.4 \pm 0.3$  mS cm<sup>-1</sup>; density: 145 kg m<sup>-3</sup>; water holding capacity: 55% and maximum humidity: 50%) in March 2<sup>th</sup>, 2018.

The seedlings remained for 10 days in a misting chamber (irrigation frequency of 10 seconds every 10 minutes with an average flow rate of 36 L hour<sup>-1</sup>), in the initial rooting period. Subsequently, they were transferred to the acclimatization chamber, where they were maintained for 20 days, under controlled irrigation (irrigation frequency of 10 seconds every hour, with an average flow rate of 36 L h<sup>-1</sup>) (development period). At 30 days after planting, the seedlings were subjected to treatments with ProCa (prohexadione calcium) (Viviful® with 27.5% ingredient active) for growth control. The application of the product was carried out by spraying with a manual sprayer (1 L capacity), using a 3 mL syrup volume per seedling.

At 60 days after planting the seedling petiole length and the crown diameter were measured in millimeters (mm) with a 150mm digital caliper (MTX). The fresh leaves were dehydrated in a drying oven at 65 °C until obtaining a constant weight (dry mass) measured with a SF-400

digital scale (YD Tech), expressed in grams per seedling (g seedling<sup>-1</sup>).

The number of leaves per seedling in each plot was determined. The leaf area of the strawberry seedlings was measured with a portable LI-3100C device (LI-COR®, Inc. Nebraska, USA), expressed in square centimeters per plant (cm<sup>2</sup> plant<sup>-1</sup>). The chlorophyll content a, b and total were measured with a portable CFL1030 ClorofiLOG device (Falker, RS, Brazil).

Seedlings were planted on May 28<sup>th</sup>, 2018. Average monthly temperatures during the experiment period were between 12 and 25 °C. 7-L plastic pots containing substrate and drip irrigation tape with a spacing of 20 cm between drippers on the pots were used. The substrate used was carbonized rice husk (CAC). Two seedlings were placed per pot, spaced 10cm apart. The pots were supported on metal tables 90 cm above the ground.

The plants were managed in a closed soil cultivation system (with nutrient solution recirculation and use of water and nutrients), using a commercial solution from the company Samo® With nutrient composition in mg L<sup>-1</sup>: 132.3 nitrogen, 92.3 phosphorus, 36.9 sulfur, 212.2 potassium, 78.0 calcium and 18.5 magnesium. The pH and electrical conductivity were monitored weekly during the evaluation period, with the pH maintained between 5.5 and 6.5 and the electrical conductivity between 1.2 and 1.5 dS m<sup>-1</sup>.

Fertigation was provided to the plants daily using a Bivolt Exatron digital timer in five daily pulses of ten minutes each with intervals of three hours (carried out from 8 am to 8 pm). The nutrient solution system consisted of a ½ CV motor peripheral pump (BPV 375-Vonder) and a 310 L polyethylene reservoir (Fortlev).

The fruit harvesting period began on August 20<sup>th</sup>, 2018 until March 18<sup>th</sup>, 2019, when drastic pruning was carried out on the plants. The second cycle started on June 17<sup>th</sup>, 2019 until November 28<sup>th</sup>, 2019. It was carried out in both cycles, twice a week and standardized based on the color of the fruit's epidermis, being harvested when they had about 75% of the skin's epidermis. red coloring or more.

The number and mass of fruits per plant and the average mass of fruits were evaluated, immediately after the harvest the fruits were weighed on a SF-400 digital scale (YD Tech) and counted. The number and mass of fruits per plant were obtained from the sum of all harvests and divided by the number of live plants in the experimental unit and the average fruit mass was calculated by the ratio between these two variables. Only marketable fruits were considered, discarding those with serious defects and with a mass less than 6 g.

Full flowering and full fruiting, expressed in days after planting (DAP) in 2018 and days after pruning (DP) in 2019, with weekly monitoring of all plants in each plot. They were considered when 100% of the plants, within the plot, had at least one open flower, or a ripe fruit (more than 75% of the red colored epidermis) per plant, respectively.

A sample consisting of 10 fruits per plot in the month of each year was collected, aiming to determine the average fruit size (diameter and length of fruits), measured with the aid of a 150mm digital caliper (MTX), and the results expressed in millimeters (mm).

At the end of the experiment, all plants in each experimental unit were divided into crowns and leaves, placed in an oven at 65°C with forced air circulation until constant mass was obtained. All old and diseased leaves, removed during the cultivation, were evaluated as dry leaf mass, the results being expressed in grams per plant (g plant<sup>-1</sup>).

The experimental design used in the seedling phase was completely randomized, constituted by a factorial 4 x 2, four concentrations of prohexadione calcium (0; 150; 300 and 600 mg L<sup>-1</sup>) and two cultivars (Aromas and Camarosa), with 4 replications, being the experimental unit consisting of 12 seedlings. In the post-planting phase, the experimental design used was in randomized blocks, with the same factorial array used in the seedling phase, with five replications and the experimental unit consisted of four plants.

The data were subjected to analysis of variance by the F test, when the effect was significant, the Tukey test was performed at the level of 5% probability and the variables with significant effect for the quantitative factor were subjected to polynomial regression analysis by the software statistical Sisvar 5.6 (Ferreira, 2014).

## RESULTS AND DISCUSSION

The crown diameter, number of leaves and shoot dry matter were influenced only by the cultivar factor. The concentrations of prohexadione calcium (ProCa) did not modify significantly ( $P \leq 0.05$ ) these variables (Table 1). The cultivar Aromas had a larger crown diameter of the seedlings (10.15 mm) and a higher number of leaves per plant (7.27) than the cultivar Camarosa (Table 1). The seedlings used in the work were clod seedlings, for which we still do not have a recommendation for the appropriate crown diameter. For both cultivars, the crown diameter was above 8.0 mm, which according to Cocco *et al.* (2011), is recommended for bare root strawberry seedlings. It can be inferred that the seedlings were ready for transplant

and with accumulation of reserves for post-planting development.

Pereira et al. (2016) working with the cultivar Camarosa, obtained a crown diameter greater than 8 mm only with the application of the 200 mg L<sup>-1</sup> concentration of prohexadione calcium. With concentrations 400 and 800 mg L<sup>-1</sup>, the crown diameter was below of 7.2 mm. In the present study, the results were higher for all concentrations evaluated, which may be related to the time of application of the growth regulator, which was applied at 30 days. In the work by Pereira et al. (2016) the application occurred 12 days after planting in relation to this research, where the seedlings may have translocated crown reserves for the emission of roots and leaves, presenting lower values of crown diameter at the time of evaluation.

The shoot dry mass was higher with the seedlings of cultivar Camarosa (7.70 g), while the seedlings of cultivar Aromas had 6.01 g of shoot dry mass (Table 1). Barreto et al. (2018) when evaluating concentrations and times of application of prohexadione calcium in the growth of strawberry seedlings, verified with the cultivar Aromas and Camarosa, a reduction in the dry mass of aerial parts

of 45.07 and 31.74%, respectively, with the use of the 400 mg L<sup>-1</sup> concentration of prohexadione calcium. It is evident that the cultivar Aromas presented a greater reduction of the aerial part in relation to cultivar Camarosa with the use of growth regulator, the genetic characteristics were decisive when the regulator was used.

The leaf area and the petiole length for both cultivars decreased with increasing ProCa concentration. For the Aromas cultivar, there was a reduction of 40.71% in the leaf area and 40.55% in the petiole length using 600 mg L<sup>-1</sup> of ProCa in relation to the control treatment (Fig. 1). In the cultivar Camarosa there was a reduction of 38.92% in the leaf area and 40.02% in the petiole length using 600 mg L<sup>-1</sup> of ProCa in relation to the absence of the phyto regulator.

During transplantation, seedlings that have a smaller leaf area reduce leaf transpiration, as they are shorter (smaller leaf area and petiole length) suffer less injuries. Minimizing transplant stress, which combined with a good root system of strawberry seedlings and ideal growth conditions help to obtain maximum fruit yield (Reekie et al., 2007).

The cultivar Camarosa showed seedlings with greater vegetative growth than aromas, where in the control treatment they were 8.07 cm in length of the petiole and 208.6 cm<sup>2</sup> per total leaf area change. While the seedlings of the cultivar Aromas in the control treatment were 6.56 cm in the petiole length and 154.17 cm<sup>2</sup> per change in the leaf area.

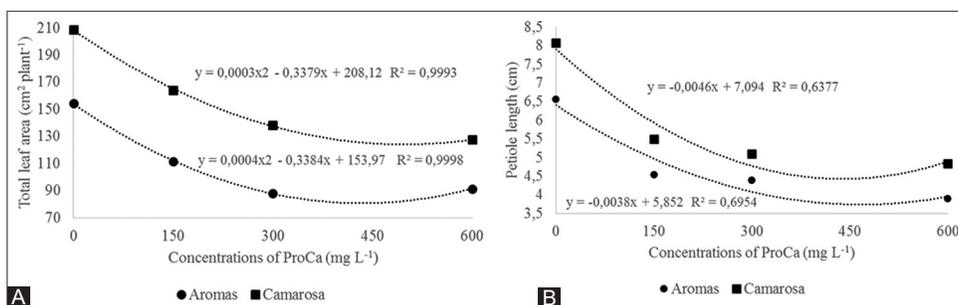
The reduction in the aerial part of the leaves observed in the seedlings with the application of ProCa, where the greatest reductions occurred with the application of the highest concentration (600 mg L<sup>-1</sup>), possibly occurred due to the hormonal regulation exercised by ProCa. That is, when applied to plants, this phyto regulator inhibits cell elongation and leaf growth by inducing a reduction in the levels of active gibberellins (Barreto et al., 2018).

The cultivars and ProCa concentrations influenced the levels of chlorophyll a, b and total. With the increase in

**Table 1: Crown diameter, number of leaves and shoot dry weight of strawberry seedlings of the cultivars Aromas and Camarosa established in the soilless cultivation system under different concentrations of Prohexadione Calcium in 2018**

ProCa <sup>1</sup> concentrations (mg L <sup>-1</sup> )	Crown diameter (mm)	Number of leaves per seedling	Dry mass of aerial part (g seedling <sup>-1</sup> )
0	9,29 <sup>ns</sup>	7,08 <sup>ns</sup>	7,63 <sup>ns</sup>
150	9,82	6,81	6,78
300	9,95	6,45	6,74
600	9,94	6,85	6,28
Cultivar			
Aromas	10,15 <sup>a</sup>	7,27 <sup>a</sup>	6,01 <sup>b</sup>
Camarosa	9,35 <sup>b</sup>	6,43 <sup>b</sup>	7,70 <sup>a</sup>
Mean	9,75	6,84	6,86
CV (%)	8,33	8,97	18,79

Means followed by the same letter in the columns, do not differ statistically from each other by the Tukey test at 5% probability of error. ns: not significant, CV: Coefficient of variation; <sup>1</sup>ProCa: Prohexadione Calcium



**Fig 1.** Total leaf area (A) and petiole length (B) of strawberry seedlings of the cultivars Aromas and Camarosa under different concentrations of prohexadione calcium in 2018.

the application of concentrations, there was an increase in the accumulation of chlorophyll in the leaves. The total chlorophyll content of the seedlings of the control treatment to 600 mg L<sup>-1</sup> of ProCa (higher concentration) increased from 51.26 to 56.99 µg cm<sup>-2</sup> in the cultivar Aromas and from 54.75 to 62.32 µg cm<sup>-2</sup> in the cultivar Camarosa (Fig. 2). Pereira et al. (2016) also found a linear increase in the content of chlorophyll a, b and total in strawberry seedlings in response to the application of ProCa. According to them, the effect of ProCa on the chlorophyll content indicates that in addition to the morphological changes in strawberry leaves, physiological changes also occur.

Chlorophyll a is the pigment used to carry out the photochemical phase (the first stage of the photosynthetic process), present in all organisms that perform oxygenic photosynthesis, while chlorophyll b together with other pigments, helps in the absorption of light and the transfer of radiant energy for the reaction centers, and therefore they are called accessory pigments (Taiz et al., 2017).

Pereira et al. (2016) when evaluating the growth of 'Camarosa' strawberry seedlings grown on a commercial substrate with application of prohexadione calcium, found that the chlorophyll content (a, b and total) increased linearly in response to the application of ProCa.

As the increase in the applied ProCa concentrations there was a need for a greater number of days for the plants to reach full bloom in 2018. The cultivar Aromas showed a difference of 10 more days for full bloom with the concentration 600 mg L<sup>-1</sup> in relation to the control. While 'Camarosa' showed a difference of 5 more days (Fig. 3A).

Full fruiting in 2018 as well as full flowering took more days for all plants to produce ripe fruit as the ProCa concentrations increased. Control treatment plants required 106.20 and 103.60 days (Aromas and Camarosa, respectively) to achieve full fruiting while plants subjected to 600 mg L<sup>-1</sup> concentration of ProCa required 120.40 and 115.40 days (Aromas and Camarosa, respectively), that is,

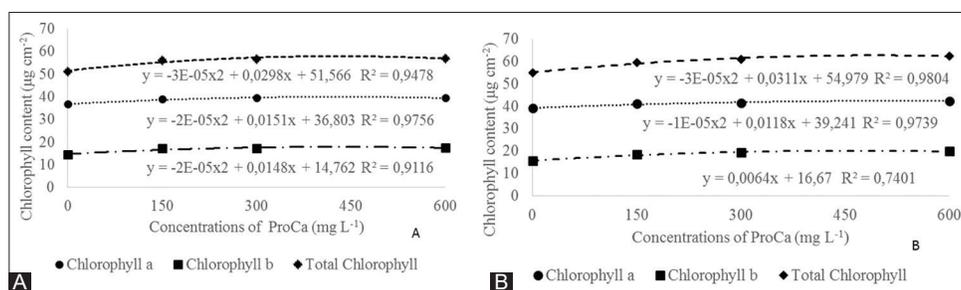
a difference of 17 and 12 days between treatments for cultivars (Fig. 3C).

Methods that inactivate gibberellin (GA) receptors, such as the use of ProCa, impair GA signaling and greatly reduce the delay of GA biosynthesis at flowering. In the process of inactivating gibberellins, changes in the expression of specific microRNAs that represent endogenous flowering pathways may occur, where integrating genes are not transported to the peak of sprouting, where it promotes transcriptional reprogramming of the meristem to initiate flowering (Andrés et al., 2014).

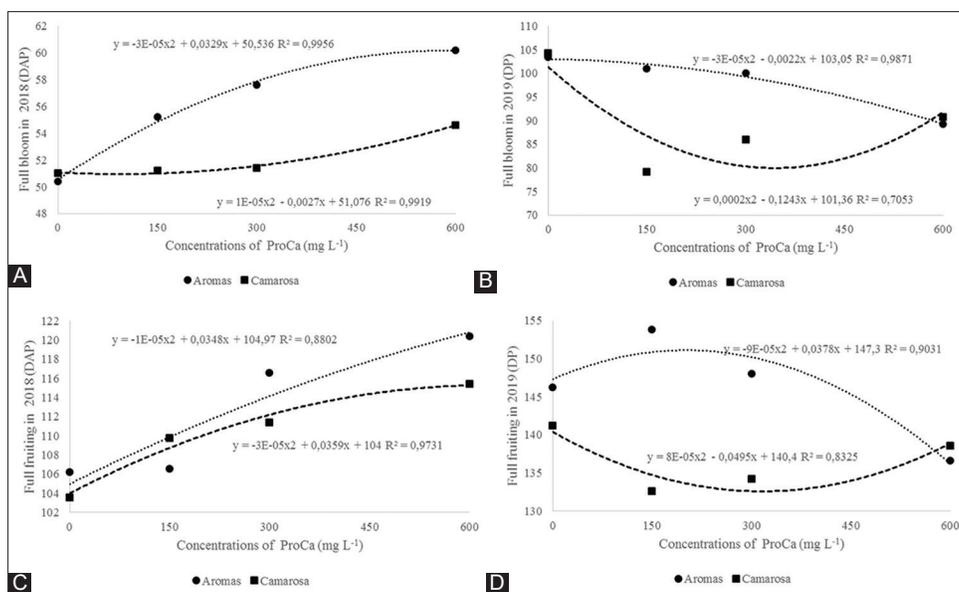
In 2019, the concentrations of ProCa 150 and 300 mg L<sup>-1</sup> for 'Aromas' and 600 mg L<sup>-1</sup> for 'Camarosa' meant that the plants needed fewer days to reach full fruiting compared to the plants control treatment, resuming production faster (Fig. 3D). According to Reekie et al., (2005 b) ProCa has a short effect on plants, after some time, vegetative growth can be resumed and the treated strawberry seedlings can grow quickly.

Tenreira et al. (2017) when evaluating the role of the enzyme GA20 oxidase with application of GA 3 and prohexadione calcium in the stolonation and flowering in strawberry plants, they verified the resumption of plant growth and the resumption of production of stolons at the normal level, five weeks after treatment with application of prohexadione calcium.

In relation to the cultivar factor, with plants of the control treatment, full fruiting in 2019 occurred first in the cultivar Camarosa (141.20 days) and in Aromas around 5 days later (146.20 days) (Fig. 3D). This resumption of plants in the emission of flowers and fruits after the summer period (with high temperatures that favor vegetation and emission of stolons) is extremely important for the producer. The sooner the emission of stolons occurs, the better, as it allows the production of fruit in periods when there is an appreciation of the kilo of fruit. According to Oliveira et al. (2017), the strawberry is subject to the different prices of the seasonality of marketing, which is concentrated in the



**Fig 2.** Content of chlorophyll a, chlorophyll b and total chlorophyll in strawberry seedlings of the cultivars Aromas (A) and Camarosa (B) under different concentrations of prohexadione calcium in 2018.



**Fig 3.** Full bloom (A) and fruiting (C) in 2018, in days after planting (DAP) and full bloom (B) and fruiting (D) in 2019, in days after planting (DP), of plants of strawberry from the Aromas and Camarosa cultivars established in the soilless cultivation system under different concentrations of Prohexadione Calcium.

last months of the year and in this period the prices paid to the producer are lower.

ProCa concentrations did not influence the number of fruits per plant, average fruit mass and fruit mass per plant. These variables were affected only by the cultivar factor (Table 2). The regulator’s temporary action meant that until the beginning of fruit production, it no longer had an effect on these variables. For Reekie et al. (2005a), the short duration of ProCa does not compromise the production of strawberry fruits after planting, it only delays the vegetative growth of the seedlings.

The number of inflorescences produced in strawberry plants was not affected by the application of ProCa in the work developed by Tenreira et al. (2017), where the role of the enzyme GA20 oxidase with application of GA 3 and prohexadione calcium in stolonation and flowering in strawberry plants was evaluated. However, they observed a negative effect, the decrease in the production of inflorescences with the application of GA3 (active gibberellin).

According to these authors, the application of bioactive GAs under conditions of long days (photoperiod greater than 12 h), inhibit flowering, but increase the production of stolons. While the inhibition of GA biosynthesis (prohexadione calcium) has the opposite effect. According to them, the precursors of bioactive GAs synthesized in the leaves change to the axillary meristem to promote stolon induction and when there is inhibition of GA biosynthesis, flowering induction occurs.

**Table 2: Total number of fruits per plant, average fruit mass and total fruit mass per strawberry plant of the cultivars Aromas and Camarosa established in the soilless cultivation system under different concentrations of Prohexadione Calcium in the cycles 2018 and 2019**

ProCa <sup>1</sup> concentrations (mg L <sup>-1</sup> )	Total number of fruits per plant	Average fruit mass (g fruit <sup>-1</sup> )	Total fruit mass (g plant <sup>-1</sup> )
0	61,82 <sup>ns</sup>	18,63 <sup>ns</sup>	652,14 <sup>ns</sup>
150	64,76	17,09	674,92
300	66,2	16,92	692,34
600	57,37	17,1	587,15
Cultivar			
Aromas	50,56 <sup>b</sup>	16,22 <sup>b</sup>	490,68 <sup>b</sup>
Camarosa	74,52 <sup>a</sup>	18,65 <sup>a</sup>	812,60 <sup>a</sup>
Mean	62,54	17,47	651,64
CV (%)	19,82	12,4	20,12

Means followed by the same letter in the columns, do not differ statistically from each other by the Tukey test at 5% probability of error. ns: not significant, CV: Coefficient of variation; <sup>1</sup>ProCa: Prohexadione Calcium

The cultivar Camarosa stood out for presenting 74.52 fruits per plant, with an average mass of 18.65 g per fruit and production of 812.60 g per plant (Table 2). These results demonstrate the greater adaptability of this cultivar in relation to Aromas as to the application of concentrations of ProCa, during the seedling phase and in the resumption of growth and production after planting. ‘Camarosa’ produced 60% more than ‘Aromas’, producing less than 500 g.

Vignolo et al. (2012), when evaluating the production of fruits and functional compounds of four strawberry cultivars in the conventional system (in the soil, with mulching) in Pelotas, observed greater fruit production with the cultivar Aromas (634.90 g and 34.5 fruits per

plant) in relation to cultivating Camarosa with (519.20 g and 26.4 fruits per plant). The divergence in the results of the two studies reveals the adaptability of the cultivar with the environment (different cultivation system and climatic conditions), indicating with which each one adapts.

For both cycles, the cultivar Camarosa showed fruits that were longer than 'Aromas' with all the concentrations used (Fig. 4C, 4D). Indicating the adaptability of the cultivar Camarosa to the seedling system that was carried out and the resumption in plant growth after treatment with the application of ProCa, with more vigorous plants and longer fruits in relation to cultivating Aromas. It is important for producers when choosing a cultivar, to know which one best adapts to the management and climatic conditions of the planting site (Antunes and Peres, 2013).

The fruit length for 'Aromas' in 2018 decreased with the increase in ProCa concentrations, from 35.08 mm with the control to 30.04 mm with 600 mg L<sup>-1</sup> (Fig. 4C). In 2019 there was an increase in the length of the fruits with the increase of the concentrations, going from 30.10 mm with the control to 36.78 mm with 600 mg L<sup>-1</sup> (Fig. 4D). With 'Camarosa there were also variations in fruit length from one year to the next. These values reflect an interaction of the cultivars with the ProCa concentrations and the climatic conditions of each year.

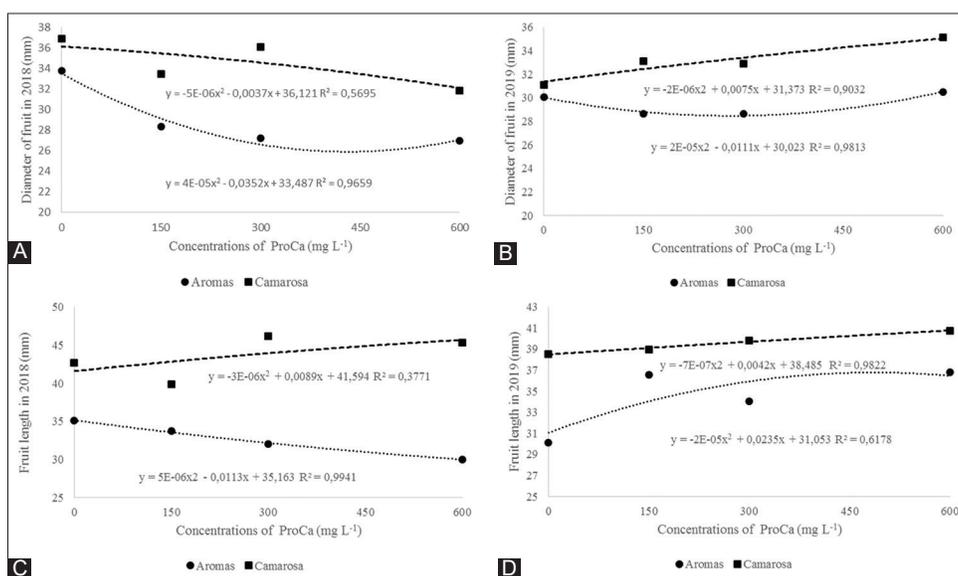
Zhang and Whiting (2011) working with foliar application of ProCa and gibberellins to improve the quality and shelf life of 'Bing' sweet cherry, observed a decrease in fruit size, from 26.13 mm (control) to 24.75 mm (treatment with ProCa). Dong et al., (2019), when verifying the cooperative effects of calcium and gibberellic acid on the calcium

content in the tissue, in the quality attributes and in relation to post-harvest disorders of sweet cherry, observed that the combined treatment in the cultivar Lapins resulted in an increase in fruit size, improved fruit quality, but delayed ripening. According to them, this increase in the size of the fruit occurred due to the greater amount of Ca absorbed by the fruit, which caused elongation of the cell wall and increased demand for Ca in order to synthesize new cell walls.

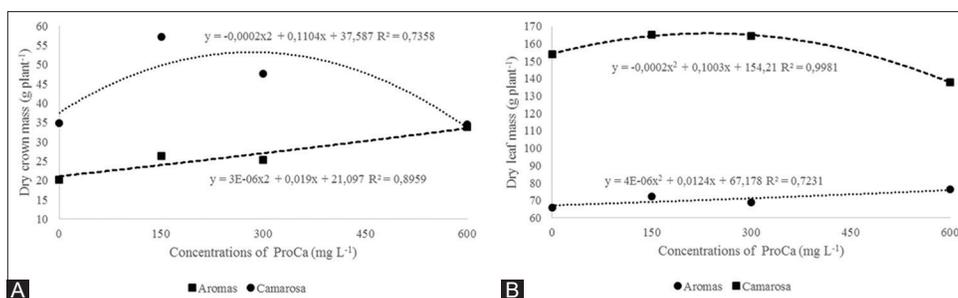
Thus, the results related to the length of fruit such as the diameter and the quality of the same will depend on the concentrations that will be used for ProCa, the frequency that will be applied (once or in installments), the cultivation system, the cultivars that are used and the environmental conditions of the cultivation cycle. It is important to identify which combination will be best for the variable being evaluated.

The fruit diameter was greater in the two years evaluated in the cultivar Camarosa, with the control treatment, 36.88 mm in 2018 (Fig. 4A). and 31.12 mm in 2019 (Fig. 4B). Becker et al. (2017) when evaluating the characterization of strawberry fruits of Italian genotypes, they observed fruits of the cultivar Camarosa with larger diameter (34.64 mm) in relation to the Italian genotypes Garda (28.90 mm), Pircinque (33.81 mm) and PIR 2 (27.66 mm). This shows that plants from the Camarosa cultivars tend to produce larger fruits, regardless of whether or not they use products or hormones to increase fruit size, being a genetic characteristic of the cultivar.

The cultivar Aromas presented fruits with a diameter below 30 mm with the application of concentrations of



**Fig 4.** Fruit diameter in 2018 (A) and 2019 (B) and fruit length in 2018 (C) and 2019 (D) of strawberries from the Aromas and Camarosa cultivars established in the soilless cultivation system under different concentrations of Prohexadione Calcium.



**Fig 5.** Crown dry weight (A) and leaf dry weight (B) of strawberry plants cv Aromas and Camarosa established in the soilless cultivation system under different concentrations of Prohexadione Calcium.

ProCa (Fig. 4A, 4B), values below that obtained with the control treatment. Silveira (2015) observed a decrease in the diameter of apple fruits in relation to the use of prohexadione calcium in the quality of the fruits. Where the diameter went from 78.5 mm in the control treatment to 73.1 mm with the application of 495 mg L<sup>-1</sup> of prohexadione calcium. According to this author, higher doses of ProCa more pronouncedly reduce the levels of active GAs in fruits, compromising the effect of GAs in increasing the size of cells in the apical and median cortical tissue of fruits.

The results of leaf dry matter and crown dry matter were influenced by the factors cultivar and concentrations of ProCa (Fig. 5A, 5B).

Plants, when subjected to the application of some concentration of ProCa, produce a greater crown mass when compared to those without the application. But the increase in concentration may end up decreasing this crown production with the cultivar Camarosa. The concentration of 300 mg L<sup>-1</sup> of ProCa helped in the production of 47.60 g of dry mass of crowns and with 600 mg L<sup>-1</sup> of ProCa the production decreased to 34.60 g of dry mass of crowns (Fig. 5A). This indicates that if the concentration is increased beyond 600 mg L<sup>-1</sup> of ProCa, the plants may produce less crowns or even stop growth and have not recovered after planting. Therefore, the importance of identifying the ideal concentration of ProCa to be applied during the seedling phase.

Hytönen et al. (2008) also obtained an increase in the formation of the number of crowns with plants treated with prohexadione calcium in relation to the plants of the control treatment. The dose of 100 mg L<sup>-1</sup> doubled and the dose of 200 mg L<sup>-1</sup> even tripled the number of crowns of branches compared to the control plants.

Hytönen et al. (2009), when studying gibberellin in the differentiation of vegetative meristems in strawberries, found that the treatment with 50 mg L<sup>-1</sup> of ProCa, quickly reduced the lengthening of stolons and petioles and greatly

improved the branching of the crowns compared to the control plants. When GA 3 (active gibberellin) was used, stolons were formed.

The inhibition of GA biosynthesis leads to the formation of the crown branching, and if the activity of GAs in plants is regulated, the induction of cell division in the subapical tissues of the axillary shoots can be controlled, being one of the signals that determine the fate of the shoots. (vegetative or floral).

The plants of cultivar Camarosa had greater vegetative growth than those of cultivar Aromas, being exposed to the genetic characteristics of each cultivar. For example with the control treatment, dry leaf mass was 153.93 g with 'Camarosa' plants and 66.02 g with 'Aromas' plants (Fig. 5B), and the crown dry matter 34.80 g with 'Camarosa' and 20.20 g with 'Aromas' (Fig. 5A).

Strassburger et al. (2011) when evaluating the growth of the strawberry plant in the face of the influence of the cultivar and the position of the plant in the bed, observed results that corroborate those found in this work. The authors found greater vegetative growth (crown and leaves) in the cultivar Camarosa (27.5 and 62.3 g, respectively) compared to cultivars Albion (16.7 and 35.6 g), Aromas (15.0 and 32.6 g) and Camino Real (15.0 and 32.1 g).

This confirms the superiority of the cultivar Camarosa in gains in dry vegetative mass in relation to the other cultivars. Her genetic characteristics in interaction with the environment enable more vigorous plants with greater leaf production, regardless of the cultivation system used. For in the work of Strassburger et al. (2011), evaluated the plants in the soil organic system, and in this work in soilless system.

Vignolo et al. (2011) also observed greater vigor of cultivar Camarosa in relation to cultivar Camino Real, when producing strawberries from alternative fertilizers in pre-planting. They found a dry matter production of the aerial part for cultivar Camarosa of 66.1 g plant<sup>-1</sup>, higher than the 49.6 g plant<sup>-1</sup> presented by cv. 'Camino Real'.

The higher production of dry matter of ‘Camarosa’ according to Strassburger (2010), may be associated with the greater capacity of expansion of the photosynthetic apparatus, with greater interception of solar radiation and greater production of photoassimilates.

## CONCLUSIONS

The application of 150, 300 and 600 mg L<sup>-1</sup> of ProCa in strawberry seedlings favors the reduction of the total leaf area and petiole length of the cultivars Aromas and Camarosa, reducing their vegetative growth. The content of chlorophyll a, b and total of the strawberry leaves of the cultivars increases with applications of ProCa. The cultivar Camarosa is more productive and has a larger fruit size than the cultivar Aromas.

## Authors’ contributions

All authors designed the study. Tais, Andressa, Caroline and Eloi performed the experiment and the data collection. Tais performed the data analysis. Tais, Andressa and Luis Eduardo wrote the manuscript.

## REFERENCES

- Andrés, F., A. Porri, S. Torti, J. Mateos, M. Romera-Branchat, J. L. García-Martínez, F. Fornara, V. Gregis, M. M. Kater and J. Coupland. 2014. Short vegetative phase reduces gibberellin biosynthesis at the Arabidopsis shoot apex to regulate the floral transition. *Proc. Natl. Acad. Sci. U. S. A.* 111: 2760-2769.
- Antunes, L. E. C. and N. A. Peres. 2013. Strawberry production in Brazil and South America. *Int. J. Fruit Sci.*, 13: 156-161.
- Antunes, L. E. C., C. Reisser, G. K. Vignolo and M. A. Gonçalves. 2015. Morangos do Jeito Que o Consumidor Gosta. *Campo & Lavoura, Anuário HF*, pp. 64-72.
- Antunes, L. E. C., A. F. Fagherazzi and G. K. Vignolo. 2017. Morangos Tem Produção Crescente. *Campo & Lavoura, Anuário HF*, pp. 96-102.
- Barreto, C. F., L. V. Ferreira, S. I. Costa, A. V. Schiavon, T. B. Becker, G. K. Vignolo and L. E. C. Antunes. 2018. Concentration and periods of application of prohexadione calcium in the growth of strawberry seedlings. *Semin. Ciênc. Agrar.* 39: 1937-1944.
- Becker, T. B., M. A. Gonçalves, S. R. Gomes, G. Baruzzi and L. E. C. Antunes. 2017. Caracterização de frutas de morangueiro de genótipos italianos na região de Pelotas-RS. In: M. Vizzotto, R. C. Franzon and L. E. C. Antunes, (Eds.), *Anais do VII Encontro Sobre Pequenas Frutas e Frutas do Mercosul, Embrapa Clima Temperado, Pelotas*, pp. 12-15.
- Brogio, B. A., S. R. Silva, T. Cantuarias-Avilés, S. F. Angolini, E. G. Baptista and R. V. Ribeiro. 2018. Influence of gibberellin inhibitors applied during flowering of non-irrigated Hass avocado trees. *Pesqui. Agropecu. Bras.* 53: 918-923.
- Carra, B., J. C. Fachinello, E. S. Abreu, M. S. Pasa, D. Spagnol, M. A. Giovanaz and C. P. Silva. 2017. Control of the vegetative growth of “Shinseiki” pear trees by prohexadione calcium and root pruning. *Pesqui. Agropecu. Bras.* 52: 177-185.
- Cocco, C., J. L. Andriolo, F. L. Cardoso, L. Erpen and O. J. Schmitt. 2011. Crown size and transplant type on the strawberry yield. *Sci. Agric.* 68: 489-493.
- Cocco, C., M. A. Gonçalves, L. Picolotto, L. V. Ferreira and L. E. C. Antunes. 2015. Crescimento, desenvolvimento e produção de morangueiro a partir de mudas com diferentes volumes de torrão. *Rev. Bras. Frutic.* 37: 961-969.
- Dong, Y., H. Zhi and Y. Wang. 2019. Cooperative effects of pre-harvest calcium and gibberellic acid on tissue calcium content, quality attributes, and in relation to postharvest disorders of late-maturing sweet cherry. *Sci. Hortic. Amsterdam.* 246: 123-128.
- Fagherazzi, A. F., A. J. Bortolini, D. S. Zanin, L. Bisol, A. M. Santos, F. Grimaldi, A. A. Kretschmar, G. Baruzzi, W. Faedi, P. Lucchi and L. Rufato. 2017. New strawberry cultivars and breeding activities in Brazil. *Acta Hortic.* 1156: 167-170.
- Ferreira, D. F. 2014. Sisvar: A guide for its bootstrap procedures in multiple comparisons. *Cienc. Agrotecnol.* 38: 109-112.
- Gonçalves, M.A., C. Cocco, G. K. Vignolo, L. Picolotto and L. E. C. Antunes. 2016. Comportamento produtivo de cultivares de morangueiro estabelecidos a partir de mudas com Torrão. *Rev. Elet. Cient. Uergs.* 2: 277-283.
- Hytönen, T., K. Mouhu, I. Koivu, P. Elomaa and O. Junttila. 2008. Prohexadione-calcium enhances the cropping potential and yield of strawberry. *Eur. J. Hortic. Sci.* 73: 210-215.
- Hytönen, T., P. Elomaa, T. Moritz and O. Junttila. 2009. Gibberellin mediates daylength-controlled differentiation of vegetative meristems in strawberry (*Fragaria × ananassa* Duch). *BMC Plant Biol.* 9: 18.
- Ito, A., D. Sakamoto, A. Itai, T. Nishijima, N. Oyama-Okudo, Y. Nakamura, T. Moriguchi and I. Nakajima. 2016. Effects of GA<sub>3+4</sub> and GA<sub>4+7</sub> application either alone or combined with prohexadione-Ca on fruit development of Japanese pear “Kosui”. *Hortic. J.* 85: 201-208.
- Kim, H. M., H. R. Lee, H. J. Kang and S. J. Hwang. 2019. Prohexadione-calcium application during vegetative growth affects growth of mother plants, runners, and runner plants of Maehyang strawberry. *Agronomy.* 9: 155-166.
- Mouco, M. A. C., E. O. Ono and J. D. Rodrigues. 2010. Inibidores de síntese de giberelinas e crescimento de mudas de mangueira “Tommy Atkins”. *Cienc. Rural.* 40: 273-279.
- Oliveira, I. P., L. C. Belarmino and A. J. Belarmino. 2017. Viabilidade da produção de morango no sistema semi-hidropônico recirculante. *Cust. Agronegocio.* 13: 315-332.
- Pasa, M. S. and T. C. Einhornet. 2017. Prohexadione calcium on shoot growth of “Starkrimson” pear trees. *Pesqui. Agropecu. Bras.* 52: 75-83.
- Pereira, I. S., M. A. Gonçalves, L. Picolotto, G. K. Vignolo and L. E. C. Antunes. 2016. Controle do crescimento de mudas de morangueiro “Camarosa” cultivadas em substrato comercial pela aplicação de proexadione cálcio. *Rev. Ciênc. Agrár.* 59: 93-98.
- Reekie, J. Y., P. R. Hicklenton and P. C. Struik. 2005a. Prohexadione-calcium modifies growth and increases photosynthesis in strawberry nursery plants. *Can. J. Plant Sci.* 85: 671-677.
- Reekie, J. Y., P. R. Hicklenton, J. R. Duval, C. Chandler and P. C. Struik. 2005b. Leaf removal and prohexadione-calcium can modify “Camarosa” strawberry nursery plant morphology for plasticulture fruit production. *Can. J. Plant Sci.* 85: 665-670.
- Reekie, J. Y., P. C. Struik, P. R. Hicklenton and J. R. Duval. 2007. Dry matter partitioning in a nursery and a plasticulture fruit field of strawberry cultivars “Sweet Charlie” and “Camarosa” as affected by prohexadione-calcium and partial leaf removal. *Eur. J. Hortic. Sci.* 72: 122-129.
- Silveira, J. P. G. 2015. Qualidade e Distúrbios Fisiológicos de Maçãs

- em Resposta a Giberelinas e Proexadiona-cálcio. (Doctoral Dissertation), Universidade do Estado de Santa Catarina, Centro de Ciências Agroveterinárias, Lages, SC, Brazil.
- Strassburger, A. S. 2010. Crescimento, Partição de Massa Seca e Produtividade do Morangueiro em Sistema de Cultivo Orgânico. (Doctoral Dissertation), Universidade Federal de Pelotas, Pelotas, Brazil.
- Strassburger, A. S., R. M. N. Peil, J. E. Schwengber, D. S. Martins and C. A. B. Medeiros. 2011. Crescimento do morangueiro: Influência da cultivar e da posição da planta no canteiro. *Cienc. Rural*. 41: 223-226.
- Taiz, L., E. Zeiger, I. Moller and A. Murphy. 2017. *Fisiologia e Desenvolvimento Vegetal*. ArtMed, Porto Alegre.
- Tenreira, T., M. J. P. Lange, T. Lange, C. Bres, M. Labadie, A. Monfort, M. Hernould, C. Rothan and B. Denoyes. 2017. A specific gibberellin 20-oxidase dictates the flowering-runnering decision in diploid strawberry. *Plant Cell*. 29: 2168-2182.
- Vignolo, G. K., V. F. Araújo, R. J. Kunde, C. A. P. Silveira and L. E. C. Antunes. 2011. Produção de morangos a partir de fertilizantes alternativos em pré-plantio. *Cienc. Rural*. 41: 1755-1761.
- Vignolo, G. K., V. F. Araújo, L. E. C. Antunes, L. Picolotto, M. Vizzotto and A. Fernandes. 2012. Produção de frutos e compostos funcionais de quatro cultivares de morangueiro. *Hortic. Bras.* 30: S3470-S3476.
- Zhang, C. and M. Whiting. 2011. Pre-harvest foliar application of prohexadione-ca and gibberellins modify canopy source-sink relations and improve quality and shelf-life of "Bing" sweet cherry. *Plant Growth Regul.* 65: 145-156.
- Zeist, A. R. and J. T. V. Resende. 2019. Strawberry breeding in Brazil: Current momentum and perspectives. *Hortic. Bras.* 37: 7-16.