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Improving feathering in different nursery apple trees by plant growth regulators

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Abstract: Feathering in apple nursery trees is a critical operation used to increase the quality of planting material. Plant growth regulators (PGR) could be used to form feathers in apple nursery trees. The main goal was to study the impact of different PGR and concentrations on feathering promotion in different nursery apple trees. The study had three different trials where the specific aims were to study the impact of: i) different rates of 6-benzyladenine (6-BA); ii) different PGR (6-BA and 6-BA + $GA_{4,7}$) on feathering promotion, and; iii) 6-BA on feathering promotion and prohexadione calcium (P-Ca) on trunk thickening under greenhouse conditions. Both 6-BA and 6-BA + GA₄₁₇ were effective inducing feathering in the different cultivars tested. 6-BA reduced final young plants grafted height only in one out of three seasons, reducing an average of 15 and 10 cm on 'Early Red One' and 'Fuji' apple nursery trees, respectively. P-Ca reduced tree growth rate 4-5 weeks after sprayed, but the difference did not persist until the end of the season. Trunk diameter was not affected by the different plant growth regulators and concentrations tested. 6-BA alone or in combination with GA_{4+7} is an efficient tool to feathering formation in different nursery apple cultivars and conditions.

Index terms: *Malus* × *domestica* Borkh., tree height, 6-benzyladenine, gibberellins 4+7, prohexadione calcium.

Ramificação lateral em diferentes mudas de macieira pela aplicação de reguladores de crescimento vegetal

Resumo: A formação de ramos laterais em mudas de macieira é uma operação crítica utilizada para aumentar a qualidade das mudas e seu potencial para maiores rendimentos, nos primeiros anos após o plantio. Os reguladores vegetais podem ser utilizados para a formação de ramos laterais em mudas de macieira. A 6-benziladenina (6-BA), aplicada isoladamente ou combinada com giberelinas 4 + 7 (6-BA + GA₄₊₇), é o PGR mais frequentemente utilizados para aumentar a formação de ramos laterais em mudas. O objetivo principal do estudo foi estudar o impacto dos reguladores vegetais e concentrações na ramificação lateral de diferentes mudas

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de macieira. O estudo contou com três diferentes experimentos cujos objetivos específicos foram estudar o impacto de: i) diferentes concentrações de 6-BA; ii) diferentes reguladores vegetais (6-BA e 6-BA + GA₄₊₇) na promoção de ramificação lateral, e iii) 6-BA na promoção de ramificação lateral e da prohexadiona cálcica (P-Ca) no diâmetro de tronco em ambiente protegido. Tanto a 6-BA quanto a combinação de 6-BA + GA₄₊₇ foram eficazes na indução de ramos laterais em mudas, nas diferentes cultivares de macieira testadas. A 6-BA reduziu a altura final das mudas apenas em um dos três anos, reduzindo em média de 15 a 10 cm a altura de mudas de macieiras 'Early Red One' e 'Fuji', respectivamente. A P-Ca reduziu a taxa de crescimento das plantas 4-5 semanas após a pulverização, mas a diferença não persistiu até ao final da estação de crescimento. O diâmetro do tronco não foi afetado pelos diferentes reguladores vegetais e pelas concentrações testadas. A 6-BA sozinha ou em combinação com a GA₄₊₇ é uma ferramenta eficiente para a indução de ramos laterais em diferentes cultivares de macieiras e condições.

Termos para indexação: *Malus × domestica* Borkh., altura de planta, 6-benziladenina, giberelinas 4 + 7, proexadiona cálcica.

Introduction

Apple (*Malus* × *domestica* Borkh.) is widely cultivated, with an estimated world production in 2019 of 87.2 million tonnes (FAOSTAT, 2022). Apple cultivation in Uruguay is based mostly in apples from the Red Delicious group, with 1,522 ha planted, which represents about 66% of the total cultivated area, that is 2,303 ha planted (ESCANDA, 2021). New training systems with reduced plant spacing and elevated tree density has been introduced in apple production. However, the implementation costs increased drastically, making it necessary early yields and profitability for quicker returns.

Uruguay is located in a warm region similar to southern Brazil (not a typical temperate climate), which makes the main axis of nurse apple trees grow vigorously and release few or no lateral branches (RUFATO et al., 2019). Feathering in apple nursery trees is a critical operation used to increase the quality of planting material and its potential to bring higher and earlier yields in the first years after planting (SADOWSKI et al., 2007; LAŇAR et al., 2020). High quality apple nursery trees should have dominant straight central leader with sufficient feathers which are induced at desirable height and distributed along the leader at regular intervals, achieving appropriate length and crotch angle. Such type of feathered nursery trees will quickly establish, grow, and fill their allotted space in orchard and consequently improve total light interception in early life of orchard (KUMAWAT et al., 2020). Moreover, feathers form flower buds in the first year of planting and facilitate the tree to bear fruit during second year of planting and will reach to full production potential after few years (SADOWSKI et al., 2007) which help to cover the significant increased cost of establishment of apple high density orchard.

Plant growth regulators proved to be efficient on apple feathering. 6-benzyladenine (6-BA) alone or in combination with gibberellin (GA_{4+7}) sprayed multiple times during the season are reported to improve apple plant quality at nursery by increasing the number and the length of lateral branches as well as the crotch angle (RUFATO et al., 2019; LAŇAR et al., 2020; KUMAWAT et al., 2020).

The production of high-quality trees that meet high standards of plant quality (climatic conditions, pests, and diseases) is important to the fruit industry in Uruguay. In order to prevent some problems with climatic conditions in external environmental, pest, and diseases control, growers in Uruguay are forcing to examinate the alternative to producing apple trees under controlled conditions. Plants are able to modify their growth, development and physiology according to their environment (OZTURK; SERDAR, 2011). The environmental conditions, such as light and temperature are the most important factors affecting plant growth. Koyuncu and Ersoy (2011) in a study with different apple cultivars nursery growing in controlled greenhouse and orchard conditions observed that trees growing under protected conditions present a higher vegetative growth and lower trunk cross sectional area than trees in orchard conditions.

Growers with nursery under protected conditions in south Uruguay are facing problems with excessive growth in height and a low trunk diameter. A possible solution could be the use of the plant growth regulator, prohexadione calcium (P-Ca) to reduce vegetative growth in height and thickening the trunk. P-Ca is a gibberellin synthesis inhibitor, when sprayed during early stages of vegetative growth, reduces the level of active gibberellin, accumulating its precursor (EVANS et al., 1999). Previous studies have demonstrated efficiency in the vegetative growth control in apple (DUYVELSHOFF; CLINE, 2013) and pear shoots (CARRA et al., 2016; 2017).

Despite the necessity for planting well fathered apple nursery trees to reach full production potential after few years in high density orchards, nurseries still produce uneven nursery trees with few feathers in warm areas, resulting in delayed production and recovery the orchard establishment cost. Therefore, our main goal was to study the impact of different plant growth regulators and concentrations on feathering promotion in different nursery grafted apple tree cultivars in a warm area condition. The specific aims of the different trials were to study the impact of, i) 6-BA concentrations (2013-2014); ii) different plant growth regulators (6-BA and 6-BA + GA_{4+7}) (2014-2015) on feathering promotion in different apple

nursery trees and; iii) 6-BA on feathering promotion and P-Ca on trunk thickening under greenhouse conditions (2020-2021).

Materials and Methods

Experiments were carried out in different sites and growing season in the climatic conditions of Uruguay. According to the Köeppen classification, the climate of the region is classified as Cfa (humid subtropical climate). The average temperature for the region is 16.1°C, where the warmest month is January (average temperature of 22.2°C), and the coolest July (average temperature of 10.0°C). The mean annual precipitation is 950 mm.

The different trials were made in collaboration with commercial nurseries. In the first year the aim was to evaluate the effect of different rates of 6-BA on feathering promotion. In the second year, with the results obtained in the first year of study, the objective was to test different products (6-BA and 6-BA + GA_{4+7}) with the same rate (best rate obtained in the first year). Both, experiment 1 and 2 were carried out under field conditions. In the last year of the study, the production of nursery grafted young plants went to a greenhouse (controlled conditions) and nurseries faced problems with excessive growth in height with a low trunk diameter in addition to the problem of feathering. Thus, we decided to evaluate a product to reduce grafted young plant height and trying to increase trunk diameter, besides to use a smaller rate of 6-BA due to the possibility of phytotoxicity (leaf burning or spotting) of higher rates of the commercial source of 6-BA in temperatures above 40°C (personal information). The commercial product MaxCel[®] (2% of 6-BA) already has in its formulation a surfactant and this is one of the reasons that the product may cause leaf damage. During the period of sprays (December and January), inside the greenhouse, temperatures above 40°C were common and we choose to reduce 6-BA rates due to the possibility of leaf burning or spotting decreasing photosynthesis and consequently the quality of the grafted young plants; even having better results in higher rate sprays in field conditions in the previous studied years.

In all experiments, rooted rootstocks from stool beds were planted in the different nurseries previously to be grafted in August with the respective cultivar. Treatment using the different plant growth regulators began after most of the grafted young plants reached 80 cm height.

Treatments with the different plant growth regulators, rates and spray timings are shown

in Table 1. In all years, the sprays were done with a backpack manual sprayer equipped with a full cone tip nozzle. During sprays, it was delivered a single spray directed to the apical meristem of the grafted young plant. The average amount of spray each grafted young plant received each spray timing was 6 mL. The 6-benzyladenin (6-BA), 6-benzyladenin + GA_{4+7} (6-BA + GA_{4+7}) and prohexadione calcium (P-Ca) sources were the commercial products MaxCel[®] (2% of active ingredient w:v), Promalin[®] (1.9% and 1.9% of active ingredient w:w); and Viviful[®] (27.5% of active ingredient w:w), respectively.

Table 1. Treatments (plant growth regulators, rates, and applications time) of the different experiments and growing seasons, Uruguay.

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	Treatments		Sprays data				
Experiment 1:	Control	-	-	-	-		
2013-2014 growing season	500 mg L ⁻¹ of 6-BA	18 Jan., 2014	-	-	-		
	1000 mg L ⁻¹ of 6-BA	18 Jan., 2014	-	-	-		
Treatments			Spray				
Experiment 2: 2014-2015 growing season	Control	-	-	-	-		
	1000 mg L ⁻¹ of 6-BA	25 Nov., 2014	09 Dec., 2014	22 Dec., 2014	-		
	1000 mg L ⁻¹ of 6-BA + GA ₄₊₇	25 Nov., 2014	09 Dec., 2014	22 Dec., 2014	-		
	Treatments	Sprays data					
Experiment 3: 2020-2021 growing season	Control	-	-	-	-		
	300 mg L ⁻¹ of 6-BA	23 Dec., 2021	06 Jan., 2021	20 Jan., 2021	3 Feb., 2021		
	300 mg L ⁻¹ of P-Ca	23 Dec., 2021	-	-	-		

*6-BA: 6-benzyladenin, P-Ca: prohexadione calcium.

To facilitate the understanding, experiments will be presented separately to plant material, design and evaluated parameters.

Experiment 1: Carried out under field conditions, in a nursery spaced 160 cm between rows and 25 cm within the row at INIA Las Brujas, located in Rincón del Colorado, Canelones, Uruguay (-34.670016, -56.341974) during 2013-2014 growing season. Plant material consisted of 'Cripps Pink' apples grafted on 'M9-337' rooted rootstock from stool bed planted in the nursery previously to be grafted. A slow-release fertilizer a week after budding and three additional nitrogen applications

during the season were applied. Grafted young plants were drip-irrigated to field capacity during the whole trial. Grafted young plants height was measured on December 18, 2013; January 21, 2014; February 17, 2014; and July 21, 2014. Grafted young plants were measured from the soil until the top of the tree. At the end of the growing season, moment when the trees were dug out to be planted, the total number of feathers per grafted young plant was evaluated by counting the number of feathers larger than 5 cm. The experimental design was the randomized complete block design with six replications of three grafted young plants per replicate. Blocks were selected by uniformity, then grouped into blocks based on the position in the nursery.

Experiment 2: Carried out in field conditions, in a nursery spaced 160 cm between rows and 25 cm within the row at a commercial nursery, located in Melilla, Montevideo, Uruguay (-34.736835, -56.284239) during 2014-2015 growing season. Plant material consisted of 'Early Red One' and 'Fuji' apples grafted on 'M7' and 'M9-337' rooted rootstock from stool bed, respectively. Fertilization and irrigation were similarly as described in experiment 1. Grafted young plant height was assessed on November 25, 2014; December 09, 2014; and May 13, 2015, similarly as experiment 1. Due to the importance of trunk diameter to the final grafted young plant classification in the nursery, in experiment 2, trunk diameter (mm) was assessed 20 cm above graft union in the end of the growing season on May 13, 2015, with a digital caliper, and expressed in mm. At the same day, the total number of feathers per grafted young plant was counted as experiment 1. The experiment was arranged as in experiment 1.

Experiment 3: Carried out at a commercial nursery, located in Cuatro Piedras, Canelones, Uruguay (-34.627624, -56.283410) during 2020-2021 growing season. Plant material consisted of potted 'Fuji' apples grafted on 'G.213' rooted rootstock from stool bed planted in containers inside a greenhouse (nursery) previously to be grafted. The experiment was carried out inside a greenhouse; grafted young plants were planted in 5 L containers filed with a potting mix with fertilization trough the irrigation water. Plants were spaced 80 cm between rows and 20 cm within the row. Grafted young plants were drip-irrigated to field capacity during the whole trial, and the excess water was drained. Grafted young plant height was measured from the graft union until the top of the tree weekly, starting in the moment of the first spray (December 23, 2020) until the winter period on July 21, 2021. The multiple evaluations in experiment 3 were made to

see how grafted young plants treated with the different PGR's and spray timings behave over time. Using the weekly grafted young plant height evaluation, it was possible to calculate the average grafted young plant growth rate (mm day⁻¹). Grafted young plant diameter (mm) was assessed weekly starting at December 23, 2020 (first spray) until July 21, 2021, with a digital caliper to evaluate the effect of the different PGR's on the trunk thickening (problem faced by the nursery in controlled conditions - greenhouse). The total number of feathers per grafted young plant was evaluated as experiment 1 and 2. Additionally, observing the need of not too long feathers in new orchards (when the feathers are too long growers will need to cut back the feather to regrowth), in experiment 3 the average feathers length was evaluated on July 21, 2021 and expressed in cm. The experiment was similarly arranged as in experiment 1 and 2, except for the number of replications and grafted young plants per replication used in the trial, which was 4 and 5, respectively.

Each experiment (growing season) was analyzed independently as the cultivar, rootstock, treatments, time of application, products and rates were different in each growing season. Statistical analyses were performed using the R software, with package ExpDes (FERREIRA et al., 2013). Data were analyzed for statistical significance by means of F test. Tukey's test at 5% of significance was performed to compare treatments when analysis of variance showed significant differences among means.

Results and Discussion

Experiment 1. In the 2013/2014 growing season only the highest rate of 6-BA (1000 mg L⁻¹) sprayed in 'Cripps Pink' apples increase the total number of feathers per grafted young plant with a total of 9.5 feathers per grafted young plant (Table 2). Furthermore, grafted young plant height was not affected by the different treatments compared to untreated control.

Table 2. Height (cm) and number of feathers of young plants of 'Cripps Pink' apples grafted on 'M9-337' rootstock treated with different concentrations of 6-benzyladenine (6-BA) in the 2013/2014 growing season, Rincón del Colorado, Uruguay.

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Treatment		Height	Height	Number of	
	18 Dec. 2013 (cm)	21 Jan. 2014 (cm)	17 Feb. 2014 (cm)	21 Jul. 2014 (cm)	feathers per plant
Control	94.00	121.58	130.50	135.50	2.25 b
500 mg L ⁻¹ 6-BA	90.17	117.50	125.08	130.00	5.17 b
1000 mg L ⁻¹ 6-BA	90.42	119.00	127.08	132.50	9.50 a
CV (%)	8.38	6.15	4.71	4.56	35.23
p-value	0.6389	0.6356	0.3291	0.3301	0.0003

*Mean separation within columns by Tukey's test at p<0.05; means followed by different letters are significantly different.

Experiment 2. In 2014/2015 growing season, both 6-BA and 6-BA + GA_{4+7} increased the number of feathers per grafted young plant in 'Early Red One' and 'Fuji' apples (Table 3). For 'Early Red One' treatments with 1000 mg L⁻¹ of 6-BA and 6-BA + GA_{4+7} did not vary between them in the total number of feathers per grafted young plant in the end of the season. However, for 'Fuji' apples, 1000 mg L⁻¹ of 6-BA alone showed to be the most effi-

cient treatment with more feathers per grafted young plant than the other treatments, followed by the 1000 mg L⁻¹ of 6-BA + GA_{4+7} . In both cultivars grafted young plants treated with 6-BA were smaller than untreated control grafted young plants, showing a final reduction around 15 and 10 cm compared to untreated grafted young plants on 'Early Red One' and 'Fuji', respectively. Treatments did not affect the final trunk diameter.

Table 3. Height (cm), trunk diameter, and number of feathers of young plants of 'Early Red One' and 'Fuji' apples grafted on 'M7' and 'M9-337', respectively treated with 6-benzyladenine (6-BA) and 6-benzyladenine + Gibberellins 4 and 7 (6-BA + GA_{4+7}) in the 2014/2015 growing season, Melilla, Uruguay.

	Trestment	Height (cm)			Trunk diameter	Number of feathers	
	Treatment	25 Nov. 2014	09 Dec. 2014	13 May 2015	13 May 2015 (mm)	per plant	
	Control	104.08	120.42	174.17 a	15.04	1.20 b	
	1000 mg L ⁻¹ 6-BA	104.8	118.37	158.77 b	15.10	6.27 a	
'Early Red One'	1000 mg L ^{_1} of 6-BA + GA ₄₊₇	102.55	119.83	163.5 ab	14.98	5.67 a	
	CV (%)	4.13	4.04	5.71	3.58	48.81	
	p-value	0.6613	0.7561	0.048	0.9321	0.004	
'Fuji'	Control	102.8	124.20 ab	176.80 a	14.58	0.20 c	
	1000 mg L ⁻¹ of 6-BA	100.6	120.50 b	167.33 b	15.58	9.73 a	
	1000 mg L ⁻¹ of 6-BA + GA ₄₊₇	101.7	129.27 a	172.90 ab	15.43	7.27 b	
	CV (%)	3.49	3.11	2.86	7.2	12.12	
	p-value	0.5791	0.0093	0.0237	0.2806	>0.0001	

*Mean separation within columns by Tukey's test at p<0.05; means followed by different letters are significantly different.

As expected, and observed by other authors, 6-BA alone or in combination with GA_{4+7} increases the number of feathers per grafted young plants in different apple cultivars (RUFATO et al., 2019; LAŇAR et al., 2020; KUMAWAT et al., 2020). This effect on feathers formation might be due the effect of 6-BA on the flow of auxins and temporarily impedes the main trunk growth (SAZO; ROBINSON,

2011), which helps to overcome apical dominance and create favorable environment of feather formation (KUMAWAT et al., 2020).

Experiment 3. In the 2020/2021 growing season, 6-BA and P-Ca did not affect final grafted young plant height and trunk diameter, however, 6-BA 300 mg L⁻¹ increase the number of feathers per grafted young plants

compared to the other treatments (Table 4). The average length of feathers was affected by the different treatments, were untreated grafted young plants showed the largest values (19.7 cm), P-Ca the smaller (5 cm), and grafted young plants sprayed with 6-BA, values around 13.5 cm. P-Ca significantly reduced grafted young plants growth compared to untreated control and 6-BA sprays during nine weeks after spray (Figure 1), after this period, grafted young plants height of P-Ca and 6-BA sprays was similar between

them. A growth flush was observed in grafted young plants sprayed with P-Ca after five weeks of the application (Figure 2) while trees sprayed with 6-BA an untreated control grafted young plants present a growth flush one week after the first spray with a reduction on growth rate from seven to nine weeks after the first spray on trees treated with 6-BA. By the end of the 2020/2021 growing season, grafted young plant height did not differ between treatments (Table 4).

Table 4. Number of feathers, final height, final trunk diameter and average feathers length of young plants of 'Fuji' apples grafted on 'G.213' rootstock treated with 6-benzyladenine (6-BA) and prohexadione calcium (P-Ca) on July 17, 2021 (2020-2021 growing season), Cuatro Piedras, Uruguay.

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Treatment	Number of feathers per plant	Final height (cm)	Final trunk diameter (mm)	Average feathers length (cm)
Control	1.00 b	170.35	9.95	19.77 a
300 mg L ⁻¹ 6-BA	12.00 a	157.65	9.53	13.53 b
300 mg L ⁻¹ P-Ca	0.35 b	157.00	9.80	5.04 c
CV (%)	18.70	5.03	3.30	18.13
p-value	>0.0001	0.1019	0.2468	0.0003

*Mean separation within columns by Tukey's test at p<0.05; means followed by different letters are significantly different.

Grafted young plant diameter during 2020/2021 growing season was reduced by 6-BA 300 mg L⁻¹ sprayed four times compared to other treatments from 2 week after the last spray (February 17, 2021) until 6

weeks after the last spray (March 24, 2021), but this difference did not persist until the end of the growing season, where we did not observe significant differences between treatments (Figure 3).

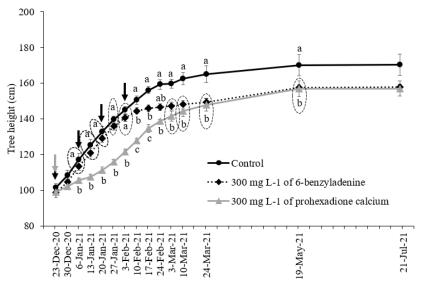


Figure 1. Height of young plants of 'Fuji' apples grafted on 'G.213' rootstock treated with 6-benzyladenine and prohexadione calcium during 2020-2021 growing season, Cuatro Piedras, Uruguay. Black and gray arrows represent 6-BA and P-Ca sprays, respectively. Vertical bars represent ±standard error. Means within figure in each date with different letters are significantly different using Tukey's test at *p*<0.05; All points inside circle did not differ significantly in each date between them. Date without letters = not significant.

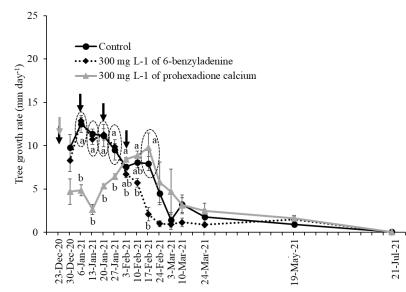


Figure 2. Growth rate of young plants of 'Fuji' apples grafted on 'G.213' rootstock treated with 6-benzyladenine and prohexadione calcium during 2020-2021 growing season, Cuatro Piedras, Uruguay. Black and gray arrows represent 6-BA and P-Ca sprays, respectively. Vertical bars represent ±standard error. Means within figure in each date with different letters are significantly different using Tukey's test at *p*<0.05; All points inside circle did not differ significantly in each date between them. Date without letters = not significant.

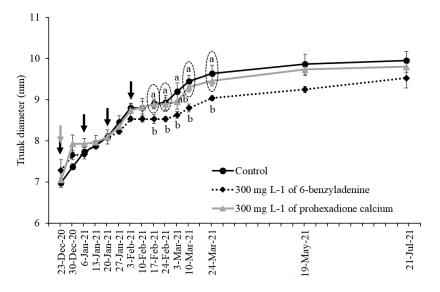


Figure 3. Trunk diameter of young plants of 'Fuji' apples grafted on 'G.213' rootstock treated with 6-benzyladenine and prohexadione calcium during 2020/2021 growing season, Cuatro Piedras, Uruguay. Black and gray arrows represent 6-BA and P-Ca sprays, respectively. Vertical bars represent ±standard error. Means within figure in each date with different letters are significantly different using Tukey's test at *p*<0.05; All points inside circle did not differ significantly in each date between them. Date without letters = not significant.

The temporarily cessation of main trunk growth, was not observed right after 6-BA application in the present study, as observed with the P-Ca application. The reduction of main trunk growth was observed further up the growing season when the feathers were already developed and could be by the competition for nutrients and carbohydrates with the feathers. Vegetative growth control by P-Ca in the present study was consistent with other studies where, P-Ca decreased shoot growth on apples (DUYVELSHOFF; CLINE, 2013) and pears (CARRA et al., 2016; 2017). Furthermore, the growth flush after

five weeks was expected because P-Ca has a relative rapid metabolism in plant tissue (2-3 weeks; EVANS et al., 1999). According to the same authors, when applied during the early stages of vegetative growth, P-Ca reduces the levels of GA₁ (highly active), accumulating its precursor GA₂₀ (inactive) in plant tissues. Our results are similar to those found in 'Le Conte' and 'Smith' pear trees cultivated in warm conditions and treated with P-Ca to reduce vegetative growth of adult trees, when a pronounced second growth flush was observed after the complete metabolism of P-CA in plant tissues (CARRA et al., 2016; 2017), and consequently a growth resumption due to the rapid metabolism of the accumulated GA_{20} (inactive) to GA_1 (active).

The lower height of grafted young plants induced by multiple sprays of the highest rate of 6-BA alone (1000 mg L⁻¹) corroborate with other studies (KUMAWAT et al., 2020; COWGILL et al., 2014). Cowgill et al. (2014) reported that grafted young plants sprayed with higher rates of 6-BA (1000 mg L⁻¹) were significantly shorter than grafted young plants sprayed with lower rates (500 mg L⁻¹). The same authors reported that 6-BA combined with GA₄₊₇ significantly increase grafted young plant height compared to 6-BA sprayed alone and untreated control. In the present study, the difference between 6-BA alone or in combination with GA_{4+7} was not significative, but both plant growth regulators combined did not showed smaller grafted young plants compared to untreated control grafted young plants. This could be a direct effect of the GA_{4+7} , where GA is known as a tree growth promoter (CLINE, 2017).

In the present study minor differences in the grafted young plant of 'Fuji' trunk diameter were observed during the growing season, but this difference did not persist until the end of the season. Differences in trunk diameter of grafted young 'Gala' apples was observed by DORIC et al. (2015) when 6-BA was sprayed three times in the concentrations of 250 and 350 mg L⁻¹ reduced trunk diam-

eter compared to untreated grafted young plants. Same was observed by STEINER et al. (2013) where 'Golden Delicious' grafted young plants had a lower trunk diameter compared to untreated grafted young plants. Furthermore, ROSSI et al. (2004) found that 6-BA + GA₄₊₇ sprayed once in different concentrations (0, 500, 1000, and 1500mg L⁻¹) decreased trunk diameter of young grafted plants of 'Catarina' apples, but the differences were not statistically significant. Studies with other fruit species, also confirm that BA can decrease trunk diameter to a certain degree, if higher concentrations are applied (MAGYAR; HROTKO, 2002; 2005). On the other hand, some studies found that 6-BA increased trunk diameter of grafted young 'Jonagold' (DORIC et al., 2015) and 'Red Boskoop' (WERTHEIM; ESTANBROOKS, 1994) apples. Based on the present and previous studies, as mentioned by DORIC et al. (2015), the mechanism by which 6-BA affect tree diameter cannot be precisely determined.

Conclusions

Both 6-BA and 6-BA + GA_{4+7} are effective inducing feathering in the different apple cultivars tested. 6-BA (1000 mg L⁻¹) reduced an average of 15 and 10 cm on 'Early Red One' and 'Fuji' young plants grafted height, respectively. P-Ca reduced tree growth 4-5 weeks after sprayed, but the difference did not persist until the end of the growing season in 'Fuji' young plants grafted on 'G.213' rootstock. Trunk diameter was not affected by the different plant growth regulators and concentrations tested by the end of the growing season. So, we can conclude that 6-BA alone or in combination with GA₄₊₇ is an efficient tool to improve young plants grafted apples feathering formation despite the cultivar tested.

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References

- CARRA, B.; PASA, M.S.; FACHINELLO, J.C.; SPAGNOL, D.; ABREU, E.S.; GIOVANAZ, M.A. Prohexadione calcium affects shoot growth, but not yield components, of 'Le Conte' pear in warm-winter climate conditions. Scientia Horticulturae, v.209, p.241-248, 2016. https://doi.org/10.1016/j. scienta.2016.06.036.
- CARRA, B.; SPAGNOL, D.; ABREU, E.S.; PASA, M.S.; SILVA, C.P.; HELLWIG, C.G.; FACHINELLO, J.C. Prohexadione calcium reduces vegetative growth and increases fruit set of 'Smith' pear trees, in Southern Brazil. Bragantia, v.76, p.360-371, 2017. *https://doi.org/10.1590/1678-4499.298*.
- CLINE, J.A. Interactive effects of 6-BA, GA₄₊₇, and prohexadione-calcium on Gala apples. Canadian Journal of Plant Science, v.97, p.632-644, 2017. DO *https://doi.org/10.1139/cjps-2016-0314*.
- COWGILL, W.; BEESE, M.; MAGRON, R.; AUTIO, W.R.; CLEMENTS, J.M.; ROBINSON, T. Studies and Recommendations for Branching Young Apple Trees. Fruit Notes, v.79, p.1-8, 2014.
- DORIC, M.; MAGAZIN, N.; MILIC, B.; KESEROVIC, Z. Enhancing feathering of one-year-old Gala and Jonagold apple trees through application of 6-benzylaminopurine and gibberellins. Bulgarian Journal of Agricultural Science, v.21, p.631-637, 2015.
- DUYVELSHOFF, C.; CLINE, J.A. Ethephon and prohexadione-calcium influence the flowering, early yield, and vegetative growth of young 'Northern Spy' apple trees. Scientia Horticulturae, v.151, p.128-134, 2013. https://doi.org/10.1016/j.scienta.2012.12.002
- ESCANDA, M.C. Fruticultura de hoja caduca. Registro Nacional Frutihortícola 2020. Módulo Manejo regional de plagas. In: CARRA, B.; DINI, M. (Eds.). Seminario de actualización técnica en frutales de pepita. (Ciclo Destacadas INIA 2021). Seminario técnico. Las Brujas, Canelones (UY), 22 y 29 julio 2021. p. 59-66. (Serie Actividades de Difusión; 798 ISSN: 1688-9258), 2021.
- EVANS, J.R.; EVANS, R.R.; REGUSCI, C.L.; RADEMACHER, W. Mode of action, metabolism, and uptake of BAS 125W, prohexadione-calcium. HortScience, v.34, p.1200-1201, 1999. *https://doi.org/10.21273/HORTSCI.34.7.1200*
- FAOSTAT. Products. Food and Agriculture Organization of the United Nations. FAOSTAT (database, Acceded on: Feb. 16, 2022). Available on: https://www.fao.org/faostat/es/#data/QCL>, 2022.
- FERREIRA, E.B.; CAVALCANTI, P.P.; NOGUEIRA, D.A. ExpDes: Experimental Designs pacakge. 2013. R package version 1.1.2. Available on: <<u>http://CRAN.R-project.org/>package=ExpDes</u>>. Accessed on: Jan. 13, 2017.
- KUMAWAT, K.L.; RAJA W.H.; SINGH D.B.; CHAND L.; MIR J.I.; RAI K.M.; KIRMANI S.N. Effects of plant growth regulators applications on induction of lateral branching in Oregon Spur apple nursery trees. Indian Journal of Horticulture, v.77, p.72-79, 2020. https://doi. org/10.5958/0974-0112.2020.00030.4
- KOYUNCU, F.; ERSOY, N. Nursery growing in controlled greenhouse and orchard by using various grafting methods in some apple (*Malus domestica* L.) varieties. Journal of Food, Agriculture & Environment, v.9, p.243-246, 2011. *https://doi.org/10.5897/AJB10.2518*
- LAŇAR, L.; MÉSZÁROS, M.; KYSELOVÁ, K.; NÁMĚSTEK, J.; SUS, J.; BĚLÍKOVÁ, H.; ČONKA, P. Branching of nursery apples and plums using various branching inducing method. Journal of Central European Agriculture, v.21, p.113-123, 2020. https://doi.org/10.5513/JCEA01/21.1.2459
- MAGYAR, L.; HROTKÓ, K. Effect of 6-benzyladenine (BA) and gibberellic acid (GA₄₊₇) application on feathering of plum cultivars in nursery. Acta Horticulturae, v.577, p.345-349, 2002. *https://doi.org/10.17660/ActaHortic.2002.577.59*
- MAGYAR, L.; HROTKÓ, K. Effect of BA (6-benzyladenine) and GA₄₊₇ on feathering of sweet cherry cultivars in the nursery. Acta Horticulturae, v.667, p.417-422, 2005. *https://doi.org/10.17660/ActaHortic.2005.667.60*

- OZTURK, A.; SERDAR, U. Effects of different nursery conditions on the plant development and some leaf characteristics in Chestnuts (*Castanea sativa* Mill.). Australian Journal of Crop Science, v.5, p.1218-1223, 2011.
- ROSSI, A.D.; RUFATO, L.; GIACOBBO, C.L.; GOMEZ, F.R.C.; FACHINELLO, J.C. Use of promalin[®] on oneyear old trees of the apple cv. 'Catarina'. Acta Horticulturae, v.636, p.145-149, 2004. https://doi. org/10.17660/ActaHortic.2004.636.17
- RUFATO, L.; MARCHIORETTO, L.R.; ORLANDI, J.C.; MICHELON, M.F.; ROSSI, A.D.; SANDER, G.F.; MACEDO, T.A. Lateral branch induction at nursery with growth regulators in 'Maxi Gala' apple trees grafted on four rootstocks. Scientia Horticulturae, v.253, p.349-57, 2019. https://doi. org/10.1016/j.scienta.2019.04.045
- SADOWSKI, A.; MACKIEWCZ, M.; DZIUBAN, R. Growth and early bearing of apple trees as affected by the type of nursery trees used for planting. Acta Horticulturae, v.732, p.447-55, 2007. https:// doi.org/10.17660/ActaHortic.2007.732.68
- SAZO, M.M.; ROBINSON, T. The use of plant growth regulators for branching of nursery trees in NY State. New York Fruit Quarterly, v.19, p.5-9, 2011.
- STEINER, M.; HROTKÓ, K.; VEGVARI, G. Performance of hormonal content and branching of apple nursery trees after BA (6-benzyladenine) application. Acta Horticulturae, v.981, p.419-423, 2013. https://doi.org/10.17660/ActaHortic.2013.981.66
- WERTHEIM, S.J.; ESTABROOKS, E.N. Effect of repeated sprays of 6-benzyladenine on the formation of sylleptic shoots in apple in the fruit-tree nursery. **Scientia Horticulturae**, v.60, p.31-39, 1994. https://doi.org/10.1016/0304-4238(94)90060-4