Influence of gibberellin inhibitors applied during flowering of nonirrigated 'Hass' avocado trees

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Abstract – The objective of this work was to evaluate the effect of different gibberellin biosynthesis inhibitors on shoot growth above the panicle, as well as on fruit yield, size, and shape of nonirrigated 'Hass' avocado (*Persea americana*) trees. The experiment was carried out in a randomized complete block design, with five treatments, four replicates, and 40 trees. From 2013 to 2015, the following treatments were applied at full bloom, as foliar sprayings: water (control), 250 mg L⁻¹ prohexadione-calcium, 2,500 mg L⁻¹ trinexapac-ethyl, 1,750 mg L⁻¹ paclobutrazol, and 350 mg L⁻¹ uniconazole; the first two are acylcyclohexanediones and the last two, triazoles. Fruit yield, yield efficiency, and alternate bearing were not affected by the application of plant growth regulators. However, the gibberellin inhibitors increased fruit size and modified the fruit shape of nonirrigated 'Hass' avocado. Uniconazole reduces shoot growth above the panicle, and this effect may vary depending on environmental conditions, especially water supply.

Index terms: *Persea americana*, acylcyclohexanediones, production, triazoles.

Influência de inibidores de giberelinas aplicados no florescimento de abacateiros 'Hass' não irrigados

Resumo – O objetivo deste trabalho foi avaliar o efeito de diferentes inibidores da biossíntese de giberelinas no crescimento dos brotos acima da panícula, bem como na produção, no tamanho e no formato de frutos de abacateiro (*Persea americana*) 'Hass' não irrigado. O experimento foi conduzido em delineamento de blocos ao acaso, com cinco tratamentos, quatro repetições e 40 árvores. De 2013 a 2015, foram aplicados os seguintes tratamentos, no florescimento, via pulverização foliar: água (testemunha), 250 mg L⁻¹ de prohexadione-cálcio, 2.500 mg L⁻¹ de trinexapaque-etílico, 1.750 mg L⁻¹ de paclobutrazol e 350 mg L⁻¹ de uniconazole; os dois primeiros são acilciclohexanodionas e os dois segundos triazóis. A produção de frutos, a eficiência produtiva e a alternância produtiva não foram afetadas pela aplicação dos fitorreguladores. Entretanto, os inibidores de giberelinas proporcionaram frutos de maior tamanho e modificaram o formato de abacates 'Hass' não irrigados. O uniconazole reduz o crescimento dos brotos acima da panícula, e esse efeito pode variar de acordo com as condições ambientais, especialmente com a disponibilidade de água.

Termos para indexação: Persea americana, acilciclohexanodionas, produção, triazóis.

Introduction

Brazil has excellent soil and climate conditions for avocado (*Persea americana* Mill., Lauraceae) production. However, the expansion of the growing area is limited by several factors, such as: the non-adoption of high planting density and of agronomic managements for improving fruit production and quality (Menzel & Le Lagadec, 2014); the presence of *Phytophthora cinnamomi*, the main pathogen that threatens avocado production worldwide (Acosta-Muñiz et al., 2012); and the cultivation of most of the

local commercial orchards under rainfed conditions. Avocado productivity is also reduced by alternate bearing (Whiley et al., 2013), excessive vigor (Lovatt, 2005), and a low fruit set rate, due to the intense competition of flowers and fruitlets with the new vegetative shoot developing above the panicles (Symons & Wolstenholme, 1990).

In order to manage these limitations and increase fruit yield, the application of phytoregulators has been adopted in the main avocado producing countries (Lovatt, 2005). Several studies conducted in irrigated

orchards of 'Hass' avocado, the main cultivar planted around the world, have shown the effectiveness of applying phytoregulators in the bloom period, with improvements in fruit yield and size, increasing the economic return to growers (Erasmus & Brooks, 1998; Lovatt, 2005). The most widely adopted phytoregulators in irrigated avocado orchards are gibberellin biosynthesis inhibitors, particularly the triazoles paclobutrazol and uniconazole (Menzel & Le Lagadec, 2014). Symons & Wolstenholme (1990) pointed out that the intense competition for resources between the shoot growing above the panicle and the concurrently developing flowers and newly set fruit is mitigated by the application of gibberellin inhibitors.

In Brazil, previous studies have reported the use of the following gibberellin biosynthesis inhibitors: paclobutrazol (Cardoso et al., 2007; Mouco et al., 2010; Upreti et al., 2013; Chatzivagiannis et al., 2014), uniconazole (Silva et al., 2010), and trinexapac-ethyl in mango (*Mangifera indica* L.) trees (Mouco et al., 2010, 2011, 2013); and prohexadione-calcium in apple (*Malus domestica* Borkh) trees (Hawerroth et al., 2012; Hawerroth & Preti, 2014). However, no studies have been reported on the use of these phytoregulators in nonirrigated avocado trees. In the current scenario of global climate changes and increasing water shortage, it is extremely important to validate managements that may be efficient and beneficial for agricultural production under restricted water availability.

The objective of this work was to evaluate the effect of different gibberellin biosynthesis inhibitors on shoot growth above the panicle, as well as on fruit yield, size, and shape of nonirrigated 'Hass' avocado trees.

Materials and Methods

The experiment was carried out between 2013 and 2016, in nonirrigated 'Hass' avocado trees managed following the recommendations for the crop (Schaffer et al., 2013). The orchard was established in 2009 at 8x6-m spacing, in a Latossolo Vermelho distrófico (Santos et al., 2013), i.e., a Typic Haplorthox, in the southwestern region of the state of São Paulo, Brazil. The climate of the region is Cwa, according to Köppen-Geiger's classification, subtropical, rainy in summer and dry in winter. Meteorological data were collected by an automated weather station installed in the field (Figure 1). The trial was set in a randomized complete block design, with five treatments, four replicates, and two plants per plot, totaling 40 trees under evaluation.

The trees were sprayed with the following treatments: water, as a control; 250 mg L⁻¹ of the acylcyclohexanedione prohexadione-calcium; 2,500 mg L⁻¹ of the acylcyclohexanedione trinexapac-ethyl; 1,750 mg L⁻¹ of the triazole paclobutrazol; and 350 mg L⁻¹ of the triazole uniconazole; the application dates were September 12, 2013, August 5, 2014, and August 16, 2015, at full bloom, when more than 50% of the canopy surface had open flowers. A mean spray mix volume of 4.0 L per plant was applied on the leaves to the drip

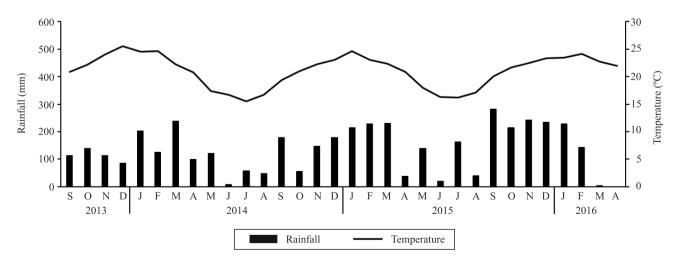


Figure 1. Monthly rainfall and average air temperature during the experimental period from September, 2013, to April 2016, at Fazenda Santa Cecília, located in Bernardino de Campos, in the state of São Paulo, Brazil.

point at the beginning or at the end of the day, when there was no wind or rain. A total of 10 mL L-1 of the Silwet L-77 silicone surfactant (Momentive Performance Materials Inc., Waterford, NY, USA) was added to the spray mix of all treatments to enhance the efficiency of the application. The used doses were defined based on the study of Menzel & Le Lagadec (2014) for triazoles, and of Salazar-García et al. (2007) for prohexadionecalcium. In 2014, due to the intense drought (Figure 1), the concentration of some gibberellin inhibitors was reduced, in order to avoid an excessive inhibition of vegetative growth or an excessive fruit set in the trees, which were already under severe water stress. In that year, the concentration of prohexadione-calcium was reduced to 28%, while the concentrations of both paclobutrazol and uniconazole were reduced to 10%. The concentration of trinexapac-ethyl was not modified, due to the lack of scientific evidences on the use of this inhibitor for shoot growth inhibition in avocado trees.

Shoot growth rate per treatment (millimeters per day) was determined between 2013 and 2015, by measuring the length of 40 shoots growing above the panicle of indeterminate inflorescences. Fruit yield was evaluated annually, on March 28, 2014, April 18, 2015, and April 7, 2016, when a minimum of 23% dry matter content was reached in the fruit pulp (Carvalho et al., 2014).

Fruit number and weight (kg per plant) per tree were recorded annually, and cumulative fruit yield was also calculated. Yield efficiency (YE, kg m⁻³) was obtained as YE = fruit weight (kg) / canopy volume (m³). Canopy volume (V) was calculated according to Mickelbart et al. (2007), using the expression: $V = 4/3 \pi ab^2$, where a is plant height (m) and b is average plant width (m). The alternate bearing index (ABI) was determined according to the equation of Mickelbart et al. (2007): ABI = $(1/n-1) \times \{[(|a_2 - a_1|)]\}$ $/(a_2 + a_1)$] + $[(|a_3 - a_2|) / (a_3 + a_2)]$ + ... + $[(|a_n - na_{n-1}|) / (a_1 + a_2)]$ $(a_n + a_{n+1})$, where n represents the number of years; and $a_1, a_2, ..., a_{n-1}, a_n$ are the yields of the corresponding years. Fruit weight and size were assessed in samples of 100 fruits randomly collected from each treatment at the time of harvest, by obtaining individual fruit mass (g) and equatorial length and diameter (mm). The fruit length/diameter ratio was used to determine fruit shape in the different treatments.

Data were subjected to analyses of variance using the SAS, version 9.0, statistical software (SAS Institute, Inc., Cary, NC, USA). Means were compared by Tukey's test. All statistical analyses were performed at 5% probability. Data that did not meet the assumptions of the analysis of variance were either transformed by the Box-Cox method or subjected to nonparametric analyses by the Kruskal-Wallis or Friedman tests.

Results and Discussion

The use of the gibberellin biosynthesis inhibitors significantly reduced shoot growth above the indeterminate inflorescences in nonirrigated 'Hass' avocado trees. Uniconazole stood out, causing reductions of 10, 17, and 34% in average shoot growth in 2013, 2014, and 2015, respectively, compared with the control treatment (Table 1). This greater reduction induced by uniconazole may be attributed to its inhibitory effect at earlier steps of the gibberellin biosynthesis pathway (Graebe, 1987), even when applied at a 10%-lower concentration, in 2014, and also after an unusual rainy period during the winter months, in 2015 (Figure 1). This change in climate conditions may explain why the inhibitory effect of uniconazole varied so much

Table 1. Average shoot growth rates of 'Hass' avocado (*Persea americana*) trees sprayed with different gibberellin inhibitors from 2013 to 2015⁽¹⁾.

Treatment	Average sho	oot growth rate (r	nm per day)
	2013 (49 DAA) ⁽²⁾	2014 (33 DAA) ⁽³⁾⁽⁴⁾	2015 (44 DAA) ⁽²⁾
T1, water (control)	0.98ab	4.07ab	5.76a
T2, 250 mg L ⁻¹ prohexadione-calcium	1.06a	4.45a	4.36b
T3, 2,500 mg L ⁻¹ trinexapac-ethyl	1.04a	4.00abc	4.39b
T4, 1,750 mg L ⁻¹ paclobutrazol	0.92bc	3.82bc	4.67b
T5, 350 mg L ⁻¹ uniconazole	0.88c	3.39c	3.73b
CV (%)	15.1	8.8	20.7
P-value	< 0.0001	0.0003	< 0.0001

⁽¹⁾Means followed by equal letters, in the columns, do not differ by Tukey's test, at 5% probability. ⁽²⁾The original values were transformed by $\hat{y} = y^{0.5}$. ⁽³⁾The original values were transformed by $\hat{y} = \log_{10}$. ⁽⁴⁾Only 28% of the prohexadione-calcium concentration and 10% of the paclobutrazol and uniconazole concentrations were applied. DAA, days after treatment application.

throughout the experimental period (Moreira et al., 2016).

The gibberellin inhibitors, however, did not affect the alternate bearing index (Table 2) and the production variables average fruit number and weight per plant, and yield efficiency (Table 3).

According to Menzel & Le Lagadec (2014), the effects of paclobutrazol, uniconazole, and prohexadionecalcium on fruit yield of 'Hass' avocado trees are less consistent than those on the reduction of vegetative growth, as also observed in the present study. Although these inhibitors retarded shoot growth of avocado plants (Abdollahi et al., 2012), their effect on fruit yield was only indirect or secondary, since it depended on several other factors, such as product concentration and time of application, soil and climate conditions, irrigation, and plant health and nutrition (Whiley et al., 2013; Moreira et al., 2016). It should also be noted that the possible benefits of these inhibitors on yield is limited in plants exposed to environmental stresses, such as water shortage during the dry winter period in the Southeastern region of Brazil (Moreira et al., 2016).

Regarding fruit yield and size, significant increases were reported for 'Hass' avocado trees when sprayed with paclobutrazol (Whiley et al., 2013), uniconazole (Erasmus & Brooks, 1998), and prohexadione-calcium (Salazar-García et al., 2007).

The effect of the gibberellin inhibitors on fruit size varied during the three years of evaluation (Table 4). Paclobutrazol, uniconazole, and prohexadione-calcium applications increased fruit size in 2014, but not in 2015, whereas trees sprayed with trinexapac-ethyl yielded fruit with the highest average weight in 2016.

The average weight of 'Hass' avocados significantly increased in 2014 and 2016 when the gibberellin inhibitors were applied at the same concentrations. It should be highlighted that the trees sprayed with uniconazole during flowering had larger-sized fruit and greater shoot growth reduction (Table 1), which may be an indicative of a consequent dry mass redistribution within the plant (Symons & Wolstenholme, 1990).

The application of gibberellin inhibitors also affected the shape of 'Hass' avocado fruit, although this result was not maintained over the years (Table 4). In 2014 and 2016, all gibberellin inhibitors significantly increased mean fruit diameter. This effect could be a consequence of the steep rise in the number of cells in the fruit equatorial region induced by triazols, causing diameter expansion (Symons & Wolstenholme, 1990). Erasmus & Brooks (1998) reported similar results for 'Hass' avocados trees treated with 500 mg L⁻¹

Table 2. Mean alternate bearing index (ABI) of nonirrigated 'Hass' avocado (*Persea americana*) trees sprayed with plant growth inhibitors during the 2014–2016 period.

Treatment	Mean ABI during 2014–2016
T1, water (control)	0.24
T2, 250 mg L ⁻¹ prohexadione-calcium	0.23
T3, 2,500 mg L-1 trinexapac-ethyl	0.15
T4, 1,750 mg L ⁻¹ paclobutrazol	0.20
T5, 350 mg L-1 uniconazole	0.24
Coefficient of variation (%)	55.43
P-value ⁽¹⁾	0.5162

⁽¹⁾Obtained by the Friedman test.

Table 3. Fruit number and weight, and yield efficiency of 'Hass' avocado (*Persea americana*) trees sprayed with plant growth inhibitors from 2014 to 2016 period.

Treatment(1)		Frui	its per tree	9		Fruit we	ight (kg p	er tree)		Yield eff	iciency (k	g m ⁻³)
-	2014(2)	2015	2016(3)	2014 and 2016 (cumulative) ⁽³⁾	2014(2)	2015	2016	2014 and 2016 (cumulative)	2014(2)(3)	2015	2016(3)	2014 and 2016 (average)
T1	541	493	548	1,089	74	102	84	158	1.8	1.6	0.9	1.3
T2	546	599	374	920	84	121	88	172	2.2	1.7	0.7	1.4
T3	559	547	531	1,090	76	111	85	161	1.7	1.6	0.6	1.1
T4	479	446	404	883	80	96	82	162	1.8	1.3	1.1	1.5
T5	497	493	478	975	74	110	87	161	1.6	1.9	1.0	1.3
CV (%)	40.0	32.7	6.3	4.5	41.6	34.6	33.3	29.1	24.1	31.8	46.33	21.0
P-value	0.9257	0.4389	0.1843	0.6142	0.9639	0.7375	0.9912	0.9765	0.2529	0.2026	0.0950	0.2501

⁽¹⁾T1, water; T2, 250 mg L⁻¹ prohexadione-calcium; T3, 2,500 mg L⁻¹ trinexapac-ethyl; T4, 1,750 mg L⁻¹ paclobutrazol; and T5, 350 mg L⁻¹ uniconazole. (2)Only 28% of the prohexadione-calcium concentration and 10% of the paclobutrazol and uniconazole concentrations were applied. (3)The original values were transformed by \hat{y} =1/y.

Fable 4. Fruit weight, length (L), diameter (D), and L:D ratio of nonirrigated 'Hass' avocado (Persea americana) trees sprayed at full bloom with different plant growth inhibitors from 2014 to 2016(1)

nent ⁽²⁾		Fruit 1	Fruit weight (g)			Lengt	Length (mm)			Diam	Diameter (mm)			L:	L:D ratio	
	2014(3)	2015	2016	Average 2014/2016	2014(3)(4)	2015	2016(4)	Average 2014/2016	2014(3)(4) 2	2015(5)	2016 ⁽⁶⁾	Average 2014/2016 ⁽⁴⁾	2014(3)	2015	2016	Average 2014/2016 ⁽⁶⁾
	140.0c	40.0c 208.6a 194.6c	194.6c	167.3b	77.7c	90.8a	87.2a	82.5b	59.8c	66.7a	66.7c	63.2b	1.30ab	1.36a	1.30a	1.30a
	163.4ab	203.8a	.63.4ab 203.8a 215.0ab	189.2a	81.6ab	87.3bc	86.3a	85.0ab	63.1ab	67.0a	69.9ab	66.5a	1.29ab	1.31bc	1.24b	1.26b
	151.2bc	51.2bc 199.6a 224.4a	224.4a	187.8a	80.1bc	86.1c	84.1a	83.9b	61.5bc	66.6a	71.4a	66.4a	1.31ab	1.30c	1.18c	1.24b
	176.3a	201.0a	176.3a 201.0a 198.9bc	187.6a	84.6a	89.2abc	85.7a	85.1a	64.5a	66.5a	67.5bc	66.0a	1.31a	1.35ab	1.27a	1.29a
	166.7a	212.3a	212.3a 215.5ab 191.1a	191.1a	81.1b	90.5ab	86.2a	83.6ab	63.8a	67.2a	69.8ab	e8.8a	1.27b	1.35ab	1.24b	1.25b
(%) A:	22.6	22.6 22.3 24.4	24.4	16.9	19.2	8.6	20.6	7.3	16.1	24.3	9.3	6.1	7.4	8.3	7.5	5.3
P-value ⁽⁷⁾	<0.0001	0.2593	<0.0001 0.2593 0.0001 <0.0001	<0.0001	<0.0001	0.0002	0.0360	0.0025	<0.0001 0.8697	0.8697	<0.0001	<0.0001	0.0182	<0.0001	<0.0001	<0.0001

(1) Weans followed by equal letters do not differ, in the columns, by Tukey's test, at 5% probability. (2) Thy water; T2, 250 mg L-1 prohexadione-calcium; T3, 2,500 mg L-1 trinexapac-ethyl; T4, 1,750 mg L¹ paclobutrazol; and T5, 350 mg L¹ uniconazole. (a)Only 28% of the prohexadione-calcium concentration and 10% of the paclobutrazol and uniconazole concentrations were applied. (d)Original Obtained by the Kruskal-Wallis test. values transformed by $\hat{y} = y^2$. ©Original values transformed by $\hat{y} = y^3$. ©Original values transformed by $\hat{y} = \log y$. uniconazole at full bloom. In the same period, all gibberellin inhibitors significantly decreased the length/diameter ratio, except paclobutrazol, which maintained the ratio by increasing fruit length.

It was expected that the reduction in shoot growth above the panicle would increase yield and fruit size. In the present study, average fruit weight was positively affected by the application of gibberellin inhibitors, but not the production variables.

Conclusions

- 1. Uniconazole reduces shoot growth in nonirrigated avocado (*Persea americana*) 'Hass' trees, and its effect may vary depending on environmental conditions, especially water supply.
- 2. The evaluated gibberellin inhibitors do not affect the production variables average fruit number and weight per plant, yield efficiency, and alternate bearing index of nonirrigated 'Hass' avocado trees.
- 3. The application of gibberellin inhibitors at full bloom increases average fruit weight and diameter, and modifies fruit shape.

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