Forcing Spring Bud Break in 'Hass' Avocado

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Abstract

Strategies to increase spring floral bud break of 'Hass' avocado (*Persea americana*) to increase floral intensity hold promise for increasing fruit set, yield and grower income. Such strategies would be especially important during the spring following the heavy on-crop when trees flower poorly. Both apical and lateral buds on 1-year-old bearing spring shoots (shoots that set fruit the previous year with fruit present through bloom in California) excised from on-crop trees in spring (March) underwent minimal bud break and plant bioregulators (PBRs) had no effect on lateral bud break even when the apical bud was removed. In contrast, apical buds on 1-year-old nonbearing spring shoots (shoots that did not set fruit the previous spring) excised from off-crop trees had a greater percent bud break than lateral buds and produced predominantly floral shoots. When the apical bud was removed, 6-benzyladenine (BA) increased lateral bud break of both floral and vegetative buds. In a commercial orchard, removal of the apical bud before the end of March increased bud break of lateral vegetative buds on 1-year-old bearing and nonbearing shoots of on- and off-crop 'Hass' avocado trees, respectively. Early spring (Feb) foliar applications of BA significantly increased bud break of apical buds, which were floral, but not lateral buds, of nonbearing shoots on off-crop trees; PBRs had no effect on apical or lateral bud break on bearing shoots of on-crop trees. A January trunk injection of a proprietary product to increase bud cytokinin concentration combined with the auxin-transport inhibitor 2,3,5-tri-iodobenzoic acid significantly increased bud break of apical and lateral floral buds on both nonbearing and bearing shoots of on-crop trees. In spring, 'Hass' avocado floral buds are dormant due to correlative inhibition of apical and lateral buds caused by mature fruit on bearing shoots of on-crop trees and also due to apical dominance of lateral buds.

INTRODUCTION

Despite problems of low fruit set, small fruit size, and alternate bearing, the Hass cultivar (*Persea americana*) dominates worldwide avocado production (>80%) (http://www.avocadosource.com). Yield of the 'Hass' avocado is proportional to the number of floral shoots and flowers at spring bloom (Garner and Lovatt, 2008). Thus, strategies that increase bud break in general and floral bud break in particular might contribute significantly to increasing yield and grower income.

Davenport (1986) reported that a percentage of lateral buds of the 'Hass' avocado did not undergo bud break and remained dormant (inactive) through spring bloom and subsequently abscised. Later research with the 'Hass' avocado provided evidence that growth of the vegetative shoot apex of a terminal indeterminate floral shoot inhibited bud break of lateral floral and vegetative buds (Salazar-García et al., 1999), consistent with apical dominance of lateral buds (paradormancy). The number of buds that remained dormant through spring bloom was consistently, significantly greater in the spring following the setting of the heavy on-crop compared to the spring following the light off-crop year (Lovatt, 2006). This observation suggests that fruit might cause correlative inhibition of buds for next year's spring bloom (paradormancy). Alternatively, Bangerth (1989) introduced the concept of primigenic dominance to explain the effect of a more

mature sink on a developing organ, including inhibition of lateral buds by fruit through a simpler mechanism than correlative inhibition. Primigenic dominance involves indole-3-acetic acid (IAA) signaling, but not the second signal (typically a cytokinin) operative in apical dominance and correlative inhibition (Bangerth, 1989; Tamas, 1995; Ferguson and Beveridge, 2009). Paradormancy and primigenic dominance can be overcome by prebloom foliar applications of PBRs, such as cytokinins and auxin-transport inhibitors, or removal of the dominant sink, e.g., shoot apex or fruit. Such treatments might successfully increase 'Hass' avocado floral intensity during bloom.

Buds of the 'Hass' avocado do not require a period of endodormancy for transition to flowering or floral shoot (inflorescence) development (Salazar-García et al., 1998), but can be subjected to ecodormancy due to low temperatures (≤17°C) during the winter months (Dec-Jan) in California, and other avocado-growing areas of the world. At the present time, there are few cultural practices (branch girdling and early fruit removal during the on-crop year) that dramatically increase floral intensity of the 'Hass' avocado.

Experiments were conducted to distinguish the effect of the apical bud on lateral bud break from that of mature fruit on apical and lateral bud break. One-year-old nonbearing shoots (spring shoots that did not set fruit plus the summer shoot that developed at the apex) on off-crop trees and 1-year-old bearing shoots (spring shoots that set fruit; summer shoots did not develop) on on-crop trees were tested in vitro using excised shoots and in vivo using shoots on mature trees in a commercial 'Hass' avocado orchard. The efficacy of the following PBRs to increase bud break was tested: 6-benzyl-adenine (BA), gibberellic acid (GA_3), hydrogen cyanamide (HC), 2,3,5-tri-iodobenzoic acid (TIBA), and a proprietary precursor of cytokinin biosynthesis (PP). With the exception of PP, the PBRs tested are each known for their capacity to stimulate precocious bud break in numerous vine and tree fruit crops both in vitro and in vivo (Lombard et al., 2006; Tworkoski and Miller, 2007; Shimazu-Sato et al., 2009), but their capacity to stimulate spring bud break in avocado was unknown.

MATERIALS AND METHODS

Plant Material

The research was conducted with shoots excised from bearing 'Hass' avocado trees at the Citrus Research Center and Agricultural Experiment Station of the University of California-Riverside (UCR), Riverside, California (33°N, 117°W), and a commercial orchard owned by the Irvine Company, Irvine, California (33°N, 117°W), where the whole tree experiments were also carried out. All trees received standard grower practice; no visual symptoms of water stress, disease or nutritional disorders were observed.

Excised Shoots

Starting in February, spring flush vegetative shoots from the previous year having swollen (active, more advanced) or closed (inactive, less advanced) apical buds were excised. In addition, bearing shoots from on-crop trees (predominantly last year's spring shoots) and nonbearing shoots that did not set fruit from off-crop trees (predominantly last year's summer shoots) were excised. All shoots (leaves and fruit removed) were washed with detergent, sterilized in a 10% bleach solution for 10 min, and rinsed with distilled water (dH_2O). The cut end of the shoots was cut again before placing the shoots in a 125 ml Erlenmeyer flask containing 50-75 ml of the solutions described below to cover the cut end of the shoots for 7 days, when the solutions were replaced with fresh solutions and the shoot was cut again to remove 6-7 mm to facilitate uptake. The shoots were maintained at 22°C, 16-h day/ 8-h night under minimal evapo-transpiration in a growth room in the Plant Transformation Research Center at UCR. At time zero, each shoot was labeled and the number of nodes with buds was counted. Apical and lateral buds were evaluated weekly to determine bud break date and whether a floral or vegetative shoot developed.

1. Experiment 1. One-year-old spring shoots collected at UCR in early spring (Feb) were treated with: (*i)* 136, 68, 34, 17, 8.5 or 4.25 mg/L BA (Maxcel®, Valent BioScience Corp.); (*ii*) 3.5 or 1.75 g/L GA_3 (Progibb®, Valent BioScience Corp.); (*iii*) rinsing for 15 min with dH_2O before transfer to dH_2O; and (*iv*) dH_2O only (control).
2. Experiment 2. One-year-old spring shoots were collected at UCR in early spring (Feb), the apical bud was removed, and the shoots were treated with: (*i*) 136, 34 or 4.25 mg/L BA; (*ii*) 3.5 or 1.75 g/L GA_3; (*iii*) rinsing for 15 min with dH_2O before transfer to dH_2O; and (*iv*) dH_2O only (control).
3. Experiment 3. One-year-old spring shoots collected in Irvine in spring (March), with and without the apical bud removed, were treated with: (*i*) 68, 34, 17 mg/L BA; (*ii*) 1,750 or 875 mg/L GA_3; (*iii*) 1,250, 630 or 320 mg/L HC (Dormex®, Dormex USA); (*iv*) rinsing for 15 min with dH_2O before transfer to dH_2O; and (*v*) dH_2O only (control).

Whole Trees

In the commercial 'Hass' avocado orchard, bearing shoots on on-crop trees (1-yr-old spring shoots with no summer shoot growth present) and nonbearing shoots on off-crop trees (1-yr-old spring shoots with summer shoot growth present) were randomly selected and used in experiments 4, 5 and 6 to test the effects of foliar-applied PBRs on apical and lateral bud break, removal of the shoot apical bud at progressively later dates on lateral bud break, and trunk-injected PBRs on bud break on bearing and nonbearing shoots on on-crop trees. Treatments were applied to individual shoots of one on- and one off-crop tree (block) replicated on 10 on- and 10 off-crop trees in Experiment 4 and 30 on- and 30 off-crop trees for Experiment 5. For both experiments the design was randomized complete block. The number of apical and lateral buds on spring and summer shoots that underwent bud break and the type of shoot that developed from each bud (floral or vegetative) was determined every 15 days. For Experiment 6, treatments were applied to seven individual trees (replications) per treatment. The number of floral and vegetative shoots that developed was determined for two bearing and two nonbearing shoots in each tree quadrant (southwest, northwest, northeast, and southeast).
1. Experiment 4. Bearing shoots on on-crop trees and nonbearing shoots on off-crop trees were sprayed in early spring (Feb) with the following treatments: (*i*) 25, 50 or 100 mg/L BA; (*ii*) 25, 50 or 100 mg/L GA_3; (*iii*) 10 or 20 mg/L HC; and (*iv*) dH_2O only (control). Solutions contained the wetting agent Silwett L-77 at 0.05%.
2. Experiment 5. Bearing shoots on on-crop trees and nonbearing shoots on off-crop trees were selected, tagged and every 30 days (Feb-May) the apical bud was removed from one set of shoots and compared to control shoots (apical bud not removed) on each data tree.
3. Experiment 6. On-crop trees were trunk injected with 1 g of the following PBRs in 60 ml of ethanol divided into two syringes per tree (BA, GA_3, TIBA [Sigma® Life Science], a proprietary precursor of cytokinin synthesis [PP]) or left untreated to serve as the control.

RESULTS

Excised Shoots

1. Experiment 1. Shoots collected with swollen (active, more advanced development) apical buds produced floral shoots (data not shown). Apical buds on shoots collected with closed (inactive, dormant) buds were exclusively vegetative. BA at 8.5, 17, or 34 mg/L significantly increased bud break of vegetative apical buds on these shoots. Lateral floral shoots developed only on shoots collected with swollen apical buds, but lateral vegetative shoots developed on shoots collected with swollen or closed buds. BA significantly increased bud break of floral (8.5 mg/L) and vegetative (17 mg/L) lateral buds on shoots collected with swollen buds only.
2. Experiment 2. Lateral buds on shoots collected with swollen or closed apical buds at the time of apical bud removal, with or without PBR treatment, produced only vegetative

shoots (data not shown). GA_3 (3.5 g/L) inhibited lateral bud break on shoots collected with swollen or closed buds. BA (34 mg/L) significantly increased lateral bud break on shoots collected with closed buds. Differences in the results obtained in Experiments 1 and 2 are likely due to the fact that 1-year-old shoots were collected indiscriminately without knowing the crop load of the trees or whether a shoot had set fruit because the trees had been harvested. These variables were controlled in all subsequent experiments.

3. Experiment 3. Apical buds on nonbearing shoots from off-crop trees produced significantly more floral shoots than vegetative shoots, with only a few exceptions (Table 1). The PBRs tested did not increase bud break of floral or vegetative apical buds relative to the control. It is noteworthy that rinsing shoots with dH_2O for 15 min before transfer to dH_2O increased bud break of apical floral buds after 1 week, which was significantly earlier than all other treatments, suggesting that buds contain a water-soluble growth inhibitor (data not shown). Apical buds of nonbearing shoots treated with 320 mg/L HC produced significantly fewer floral shoots at bud break than nonbearing shoots treated with 34 mg/L BA, rinsed with dH_2O and transferred to dH_2O, or the control (Table 1).

Apical bud break on bearing shoots of on-crop trees was low and PBR treatment had no effect (Table 1). Similarly, lateral bud break on bearing shoots collected from on-crop trees was low and PBRs had no effect even with the apical bud removed (Table 2). For nonbearing shoots excised from off-crop trees, lateral floral bud break was low but significantly enhanced by 34 mg/L BA when the apical bud was removed. BA at 68 mg/L significantly increased lateral floral bud break in just 3 weeks (data not shown), but after 3 months the control shoots had equal lateral floral bud break (Table 2). Lateral vegetative bud break was not increased by any PBR treatment when the apical bud was present on nonbearing shoots from off-crop trees. Lateral vegetative bud break was greatest on nonbearing shoots after apical bud removal and treatment with 17 mg/L BA. This number was greater than lateral floral bud break for all treatments (Table 2).

Whole Trees

1. Experiment 4. Apical buds on untreated control shoots of off- and on-crop trees produced an equal number of floral shoots at bud break (Table 3). BA (25 or 50 mg/L) significantly increased floral bud break on nonbearing shoots on off-crop trees but not on bearing shoots of on-crop trees. Bud break of apical vegetative buds on untreated control nonbearing and bearing shoots of off- and on-crop trees, respectively, was equal (Table 3). Interestingly, all PBR treatments reduced bud break of apical vegetative buds of nonbearing shoots on off-crop trees, but had no effect on bud break of apical vegetative buds on bearing shoots of on-crop trees. The data provide clear evidence of a physiological difference between apical buds on nonbearing shoots on off-crop trees and apical buds on bearing shoots of on-crop trees. There were no significant effects due to foliar-applied PBRs on lateral bud break (floral or vegetative) for nonbearing spring or summer shoots on off-crop trees or on bearing spring shoots on on-crop trees (summer shoots did not develop on on-crop trees) (Table 3). Thus, there were no significant differences in lateral bud break (floral or vegetative) related to alternate bearing, although lateral buds on nonbearing spring shoots on off-crop trees had numerically, but not significantly, greater floral bud break than bearing spring shoots on on-crop trees.

2. Experiment 5. Lateral floral bud break was low and occurred only on nonbearing shoots on off-crop trees when apical buds were removed by 20 February for summer shoots or as late as 20 March for spring shoots (Table 4). Bud beak of lateral vegetative buds on nonbearing and bearing spring shoots was similar in response to the time of apical bud removal. In both cases, removal of the apical bud before the end of March significantly increased bud break of lateral vegetative buds compared to the untreated control for each shoot type. Removal of the apical bud through 20 April significantly increased bud break of lateral vegetative buds on nonbearing summer shoots on off-crop trees. The June data revealed a high proportion of buds on all shoots that did not undergo bud break (data not shown). Taken together, these results document that fruit inhibit spring bud break and that apical dominance also contributes to bud dormancy in spring.

3. Experiment 6. Trunk injecting PP, a proprietary precursor of cytokinin biosynthesis, combined with TIBA significantly increased bud break of floral buds on both nonbearing and bearing shoots of on-crop trees (Table 5). The results are consistent with overcoming correlative inhibition and apical dominance. TIBA alone increased floral bud break on bearing shoots, consistent with mitigation of primigenic dominance caused by mature fruit on floral buds. No treatment increased vegetative bud break on nonbearing and bearing shoots relative to their respective controls, but GA_3 and TIBA increased bud break of vegetative buds on bearing shoots to a greater degree than several PBRs increased vegetative bud break on nonbearing shoots on on-crop trees or untreated control trees.

DISCUSSION

Floral shoots developing on excised shoots proved very delicate. They desiccated easily, abscised early and were more sensitive to PBR treatments, exhibiting damage at lower concentrations than developing vegetative shoots, which remained alive for months. Shoots collected with swollen apical buds produced more apical floral shoots than those collected with closed apical buds. BA increased bud break of both floral and vegetative lateral buds on shoots collected with swollen buds. Moreover, BA treatments caused earlier bud break than other PBR treatments.

Apical buds on nonbearing shoots excised from off-crop trees produced more floral than vegetative shoots. These shoots also produce more total floral and vegetative shoots than apical buds on bearing shoots excised from on-crop trees. PBR treatments did not significantly increase bud break, suggesting deeper dormancy or lower viability of apical buds borne on bearing shoots of on-crop 'Hass' avocado trees. BA significantly increased bud break of floral and vegetative lateral buds on nonbearing shoots from off-crop trees but *only* when the apical bud was removed, consistent with apical dominance playing a role in bud dormancy in spring. GA_3 (1.75 and 3.5 g/L) and all concentrations of HC tested caused symptoms of toxicity in excised shoots and failed to stimulate bud break.

In the whole tree experiments, BA (25 and 50 mg/L) increased apical floral bud break on nonbearing shoots compared to the untreated nonbearing (control) shoots on off-crop trees. Removal of the apical bud in late February through late May, demonstrated that lateral bud break of vegetative buds on both nonbearing and bearing spring shoots of off- and on-crop trees, respectively, was significantly increased when the apical bud was removed on 20 February or 20 March, but not later. Removing the apical bud from 20 Feb to 20 April increased lateral bud break of vegetative buds on summer shoots produced by nonbearing spring shoots of off-crop trees. Lateral floral shoots were observed only when the apical bud was removed in February for summer and March for spring nonbearing shoots of off-crop trees. Apical dominance is well known in 'Hass' avocado (Thorp and Sedgley, 1993). Salazar-García et al. (1999) previously demonstrated that growth of the apical vegetative shoot of terminal indeterminate floral shoots inhibited the growth of the lateral floral (and vegetative) buds. The failure of GA_3 applied in January or February to stimulate bud break of shoots was surprising since these same concentrations stimulated bud break of floral buds in a previous study when applied in November through January (Salazar-García and Lovatt, 1998). The effect of fruit on apical bud break was clearly demonstrated in this research. Nonbearing shoots responded to BA by producing more floral shoots, the response of bearing shoots was more vegetative shoots. However, floral bud break was significantly increased on bearing (and nonbearing) shoots of on-crop trees by combining the auxin-transport inhibitor with the cytokinin precursor, consistent with overcoming correlative inhibition of floral buds caused by the on-crop of mature fruit.

CONCLUSIONS

Taken together, the results of this research provide strong evidence that in spring 'Hass' avocado lateral floral buds are dormant due to apical dominance and that apical and lateral floral buds are dormant due to correlative inhibition and/or primigenic

dominance caused by the on-crop of mature fruit. Apical bud removal increased bud break on nonbearing shoots, especially in response to BA, confirming the role of apical dominance in 'Hass' avocado lateral bud dormancy in spring. Increased floral bud break on bearing shoots of on-crop trees required the auxin-transport inhibitor TIBA alone or combined with the cytokinin precursor PP, consistent with overcoming primigenic dominance and correlative inhibition, respectively. PP was clearly active since TIBA plus PP increased floral bud break on nonbearing shoots on on-crop trees significantly more than TIBA alone. With additional research to optimize application time and concentration, it seems likely that PBR strategies could be developed to increase spring floral bud break and floral intensity of 'Hass' avocado to increase fruit set, yield and grower income.

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Tables

Table 1. Effect of plant bioregulators on bud break of floral and vegetative apical buds on excised nonbearing shoots collected from off-crop trees and bearing shoots collected from on-crop trees of 'Hass' avocado in spring (March).

Treatments	Apical buds			
	Nonbearing shoots		Bearing shoot	
	Floral	Veg.	Floral	Veg.
	No. of buds that broke per 10 shoots			
1,750 mg/L GA_3	6 bcd[1]	0 g	0 a	0 a
850 mg/L GA_3	7 abc	0 g	1 a	0 a
1,250 mg/L HC	5 bcde	2 efg	0 a	0 a
630 mg/L HC	6 bcd	1 fg	1 a	0 a
320 mg/L HC	4 cdef	3 defg	1 a	1 a
68 mg/L BA	6 bcd	2 efg	0 a	0 a
34 mg/L BA	8 ab	1 fg	1 a	0 a
17 mg/L BA	6 bcd	4 cdef	0 a	0 a
Rinsed + dH_2O	10 a	0 g	1 a	0 a
dH_2O - control	8 ab	1 fg	0 a	2 a
P value	<0.0001		0.5704	

[1] Values in the paired columns followed by different letters are significantly different at the specified *P* values by Fisher's Protected LSD Test.

Table 2. Effect of apical bud removal and plant bioregulators on bud break of floral and vegetative lateral buds on excised nonbearing shoots collected from off-crop trees and bearing shoots collected from on-crop trees of 'Hass' avocado in spring (March).

Treatments	Lateral buds			
	Nonbearing shoots		Bearing shoots	
	Floral	Veg.	Floral	Veg.
	No. of buds that broke per 10 shoots			
Apical bud present				
1,750 mg/L GA_3	0 f[1]	0 f	0 a	0 a
850 mg/L GA_3	0 f	0 f	0 a	0 a
1,250 mg/L HC	0 f	2 ef	0 a	0 a
630 mg/L HC	0 f	0 f	0 a	0 a
320 mg/L HC	0 f	0 f	0 a	0 a
68 mg/L BA	5 cde	0 f	0 a	0 a
34 mg/L BA	0 f	1 ef	0 a	0 a
17 mg/L BA	0 f	2 ef	0 a	0 a
Rinsed + dH_2O	1 ef	4 cdef	0 a	0 a
dH_2O - control	1 ef	8 bc	0 a	0 a
Apical bud removed				
1,750 mg/L GA_3	0 f	0 f	0 a	0 a
850 mg/L GA_3	0 f	0 f	0 a	0 a
1,250 mg/L HC	0 f	0 f	0 a	0 a
630 mg/L HC	0 f	0 f	0 a	0 a
320 mg/L HC	0 f	0 f	0 a	0 a
68 mg/L BA	3 def	0 f	0 a	0 a
34 mg/L BA	7 bcd	1 ef	0 a	0 a
17 mg/L BA	1 ef	15 a	0 a	0 a
Rinsed + dH_2O	0 f	10 b	0 a	0 a
dH_2O - control	0 f	5 cde	0 a	1 a
P value	<0.0001		0.4746	

[1] Values in paired columns followed by different letters are significantly different at the specified *P*-values by Fisher's Protected LSD Test.

Table 3. Effect of plant bioregulators applied to the foliage of nonbearing shoots of off-crop trees and bearing shoots of on-crop trees of 'Hass' avocado in early spring (Feb) on bud break of floral and vegetative apical and lateral buds.

Treatments	Apical buds		Lateral buds			
			Spring shoots		Summer/fall shoots	
	Floral	Veg.	Floral	Veg.	Floral	Veg.
	No. of buds that broke per 10 shoots					
Nonbearing shoots						
25 mg/L BA	8 a[1]	1 cd	6 a	0 a	0 a	2 a
50 mg/L BA	7 a	1 cd	5 a	0 a	0 a	3 a
100 mg/L BA	5 ab	2 bcd	3 a	2 a	1 a	3 a
25 mg/L GA_3	5 ab	1 cd	3 a	0 a	0 a	8 a
50 mg/L GA_3	5 ab	0 d	2 a	1 a	1 a	0 a
100 mg/L GA_3	5 ab	1 cd	0 a	0 a	1 a	0 a
10 mg/L HC	3 bc	1 cd	0 a	0 a	4 a	3 a
20 mg/L HC	5 ab	1 cd	2 a	0 a	0 a	3 a
dH_2O - control	3 bc	4 abc	0 a	0 a	5 a	4 a
Bearing shoots						
25 mg/L BA	3 bc	5 ab	0 a	2 a	-[2]	-
50 mg/L BA	1 c	5 ab	0 a	0 a	-	-
100 mg/L BA	1 c	6 a	0 a	0 a	-	-
25 mg/L GA_3	3 bc	2 bcd	0 a	1 a	-	-
50 mg/L GA_3	0 c	5 ab	0 a	0 a	-	-
100 mg/L GA_3	1 c	4 abc	1 a	0 a	-	-
10 mg/L HC	1 c	2 bcd	0 a	0 a	-	-
20 mg/L HC	0 c	2 bcd	0 a	1 a	-	-
dH_2O - control	3 bc	4 abc	0 a	1 a	-	-
P-value	0.0002	0.0194	0.6708	0.5581	0.4227	0.7004

[z] Values in columns followed by different letters are significantly different at the specified *P*-values by Fisher's Protected LSD Test.

[2] Bearing shoots on on-crop trees did not have summer shoots.

Table 4. Effect of removing the apical bud monthly from February through May from nonbearing shoots on off-crop trees and bearing shoots on on-crop trees of 'Hass' avocado on bud break of floral and vegetative lateral buds through June.

Treatments	Spring shoots			Summer shoots		
	Floral	Veg.	Nodes	Floral	Veg.	Nodes
	No. of lateral buds that broke per 30 shoots					
Nonbearing shoots						
20 February	0 a[1]	16 bcd	85 bc	4 a	36 a	90 b
20 March	5 a	21 ab	98 b	0 b	36 a	83 b
20 April	0 a	8 cde	74 c	0 b	29 a	126 a
20 May	0 a	0 e	68 c	0 b	0 b	126 a
Control	0 a	0 e	73 c	0 b	0 b	109 ab
Bearing shoots						
20 February	0 a	20 abc	152 a	-[2]	-	-
20 March	0 a	29 a	133 a	-	-	-
20 April	0 a	6 de	135 a	-	-	-
20 May	0 a	0 e	147 a	-	-	-
Control	0 a	0 e	147 a	-	-	-
P-value	0.1491	<0.0001	<0.0001	0.0269	<0.0001	0.0239

[1] Values in columns followed by different letters are significantly different at the specified *P*-values by Fisher's Protected LSD Test.

[2] Bearing shoots of on-crop trees did not have summer shoots.

Table 5. Effect of plant bioregulators (1 g/tree) injected into the trunks on-crop 'Hass' avocado trees in winter (Jan) on bud break of floral and vegetative buds on nonbearing and bearing shoots.

Treatments	Floral	Vegetative
	No. of buds that broke per 100 nodes	
Nonbearing shoots		
BA	8 bcd[1]	0 c
GA_3	8 bcd	1 abc
PP	10 abc	1 c
TIBA	8 bcd	1 bc
TIBA + PP	14 a	1 abc
Control	6 cd	1 bc
Bearing shoots		
BA	7 bcd	1 bc
GA_3	6 cd	3 a
PP	7 bcd	1 abc
TIBA	10 bc	3 a
TIBA + PP	11 ab	2 ab
Control	5 d	2 abc
P-value	0.0009	0.0501

[1]Values in columns followed by different letters are significantly different at the specified *P*-values by Fisher's Protected LSD Test.