Altering Shoot Extension Did Not Affect Bitter Pit Incidence in ‘Honeycrisp’ Apple

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Abstract. *Malus ×domestica* Borkh. cv. Honeycrisp has been widely planted in North America during the past two decades. However, it is susceptible to many disorders that result in high postharvest losses. Excessive vegetative vigor in apple trees can reduce fruit calcium (Ca) concentrations and increase bitter pit incidence in apple fruit. Plant growth regulators are used routinely in tree fruit orchards to control vegetative growth to increase light penetration into the canopy. The objective of this study was to determine whether shoot growth inhibition using the application of prohexadione-calcium (P-Ca; Apogee®) or stimulation via application of gibberellic acid (GA₃; ProGibb®) affected bitter pit incidence in ‘Honeycrisp’ apple. In 2016 and 2017, the experiment was conducted in a commercial ‘Honeycrisp’ orchard with five treatments [untreated control, 62.5 mg·L⁻¹ P-Ca (low P-Ca); 125 mg·L⁻¹ P-Ca (high P-Ca); 16 mg·L⁻¹ GA₃ (low GA₃); and 32 mg·L⁻¹ GA₃ (high GA₃)]. Treatments were applied twice during the growing season. Shoot length and the number of internodes for new growth were measured 4 weeks apart after treatment. Overall yield and fruit quality were assessed at harvest, and bitter pit incidence was assessed after 4 months of storage. Low and high P-Ca rates limited shoot growth extension; high GA₃ increased shoot extension compared with the untreated control. However, the number of internodes did not change substantially for each shoot. The number of internodes is one of the primary factors affecting leaf area and, consequently, the transpiration balance between fruit and leaves. In both years, treatments with either GA₃ or P-Ca did not affect fruit elemental concentration or bitter pit incidence. These results indicate that growth-inhibiting plant growth regulators that reduce shoot extension may not be useful for managing bitter pit incidence in ‘Honeycrisp’ apple.

Bitter pit is a physiological disorder that has long been associated with low fruit Ca concentrations. The symptoms appear as depressed brown lesions on the skin of the fruit, typically located directly below the peel, but are often found scattered in the cortex near the calyx end (Perring, 1986). Bitter pit develops primarily during storage, but the underlying reasons for bitter pit begin during fruit development in the orchard (Ferguson and Watkins, 1983, 1989; Shear, 1980). Previous research has reported that bitter pit is a consequence of localized Ca deficiencies aggravated by genetic and environmental factors regulating tissue Ca content and

cellular Ca distribution, such as alternate dry/wet conditions, excessive vegetative growth, and large fruit size (Bangerth, 1979; de Freitas and Mitcham, 2012; Ferguson and Watkins, 1989; Ho and White, 2005; Saure, 2005; Taylor and Locascio, 2004). Specifically, crop load has been shown to be one of the critical factors regulating the susceptibility to bitter pit (Robinson et al., 2009; Serra et al., 2016). ‘Honeycrisp’ apple is particularly susceptible to bitter pit and, in some years, losses commonly exceed 40% to 50% (Rosenberger et al., 2004).

Tree vigor is an important indicator of tree health and productive balance. Excessive vegetative growth can have a negative effect on fruit quality, yield, and pest control (Greene, 1999; Miller and Tworkoski, 2003). More vigorous tissues can contain higher levels of GA₃-like substances (Kato and Ito, 1962; Saure, 2005) that can have a significant impact on tree growth, productivity, and quality. Schumacher et al. (1978) showed that fruit in the most vigorous parts of a tree are more susceptible to bitter pit. Excessive vegetative growth in apple trees can reduce fruit Ca content compared with trees with reduced shoot growth. The total Ca concentration in fruit tissue is usually much

less than leaves (Saure, 2005). During early fruit development, vigorous shoots are strong competitors for available Ca (Garman and Mathis, 1956; Greene, 1991; Greene and Lord, 1983; Sharples, 1974; Shear, 1980). Correspondingly, this is also the time when much of the Ca deposition occurs in fruitlets, and excessive vegetative growth during this period may result in less Ca deposition in fruitlets.

Vegetative vigor in apple trees is affected by many separate, but often interacting, factors. These factors can be internal, such as variation in tissue water and carbohydrate balance or differences in plant hormonal balance, or external, such as horticultural management and orchard environment (Rom et al., 1991). Scion cultivar (Atkinson et al., 2003; Cohen and Naor, 2002), rootstock cultivar (Webster, 1995), soil type (Raese, 1995), tree spacing (Bianco et al., 2015), irrigation (Leib et al., 2006), and light quality can all affect shoot vigor. Shoot length can also be manipulated through horticultural management by using plant growth regulators (Evans et al., 1997).

Plant growth regulators that limit shoot length extension are used most often to improve the light environment in the canopy for better fruit color development and quality. P-Ca (Apogee®) is a primary plant bio-regulator in pome fruit that inhibits the production of gibberellin (Miller, 2002; Rademacher and Kober, 2003). P-Ca reduces terminal shoot growth by inhibiting 2-oxoglutarate-dependent dioxygenases, which are involved in the formation of growth-specific gibberellins—a group of plant hormones primarily responsible for regulation of shoot elongation in apple trees (Rademacher et al., 2004). Well-timed applications of P-Ca to trees during periods of active shoot growth can significantly inhibit shoot extension. Effective rates for vegetative growth control are typically 63 to 125 mg·L⁻¹, depending on tree vigor. In general, two applications per season are required during early growth immediately after bloom. P-Ca requires about 14 d to slow growth after the first spray application (Cline, 2006).

Because shoot vigor has been linked with Ca concentrations in developing fruit, the objective of this study was to determine whether inhibiting shoot length extension via the application of P-Ca or stimulating shoot length extension via the application of GA₃ (ProGibb®) decreases or increases bitter pit incidence, respectively. This work will provide a greater understanding of the association between shoot extension and bitter pit incidence in ‘Honeycrisp’ apple.

Materials and Methods

Orchard description and experimental design. The experiment was conducted in 2016 and 2017 using Royal Red ‘Honeycrisp’ on the ‘M.9 Nic 29’ rootstock in a commercial orchard near Quincy, WA (lat. 47.163377°N, long. -119.944585°W). The trees were planted in 2014, trained to a tall spindle training system with

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Table 1. Preharvest growth regulator treatments, spray rate, and the two spray applications during the 2016 and 2017 growing seasons.

Yr and FB date	Treatment	First spray		Second spray	
		DAFB	Date	DAFB	Date
2016, FB = 14 Apr.	High GA ₃ (32 mg·L ⁻¹)	14	27 Apr.	47	31 May
	Low GA ₃ (16 mg·L ⁻¹)	14	27 Apr.	47	31 May
	Control	NA	NA	NA	NA
	High P-Ca (125 mg·L ⁻¹)	5	19 Apr.	40	24 May
	Low P-Ca (62.5 mg·L ⁻¹)	5	19 Apr.	40	24 May
2017, FB = 5 May	High GA ₃ (32 mg·L ⁻¹)	14	19 May	47	22 June
	Low GA ₃ (16 mg·L ⁻¹)	14	19 May	47	22 June
	Control	NA	NA	NA	NA
	High P-Ca (125 mg·L ⁻¹)	5	10 May	40	14 June
	Low P-Ca (62.5 mg·L ⁻¹)	5	10 May	40	14 June

FB = full bloom; DAFB = days after full bloom; GA₃ = gibberellic acid; P-Ca = prohexadione-calcium; NA = not applicable.

0.6 × 3.0-m tree spacing (5382 trees/ha), and irrigated using microsprinklers. Horticultural practices and pest management were carried out by the grower using accepted commercial practices, including Ca applications every 7 to 10 d using 2 kg·ha⁻¹ active Ca. Five treatments were tested in the experiment—1) an untreated control, 2) 62.5 mg·L⁻¹ P-Ca (low P-Ca), 3) 125 mg·L⁻¹ P-Ca (high P-Ca), 4) 16 mg·L⁻¹ GA₃ (low GA₃), and 5) 32 mg·L⁻¹ GA₃ (high GA₃)—and applied twice during the growing season (Table 1). Spray timing was based phenologically. Therefore, application dates varied between 2016 and 2017. For both 2016 and 2017, P-Ca was applied at petal fall [5 d after full bloom (DAFB)] and ≈35 d after that (40 DAFB). For both 2016 and 2017, GA₃ was applied at 10 DAFB, when shoot growth had just started, and then again at 47 DAFB. Spray treatments were applied to whole trees at a rate of 935 L·ha⁻¹ between 8:00 AM and 10:00 AM by using a backpack sprayer application to both sides of the row to cover the entire canopy. Different trees were used in 2017 than 2016 to limit the risk of carryover effects on return bloom and yield (no effect was observed; data not shown).

Flowering, crop load adjustments, and yield. Trunk cross sectional area (TCSA) was determined by measuring trunk diameter at 15 cm above the graft union before flowering, then calculating the area. After fruit set, the total number of fruitlets were counted for each tree, then target crop load was calculated using a crop load density of 4 fruit/cm² TCSA; crop load was thinned to an individual target. Fruit were harvested on 24 Aug. 2016 and 11 Sept. 2017 just before commercial harvest.

Vegetative growth. Shoot growth was measured on five randomly selected non-fruitlet, extension shoots per tree from the beginning of first year wood to the apical meristem. Measurements were made at 40 and 67 DAFB during the growing season in 2016 and 2017. At the time of shoot growth measurements, the number of internodes were counted on the same five shoots per tree.

Fruit elemental analysis. To assess the impact of shoot length extension on elemental balance, Ca, magnesium (Mg), and potassium (K) concentrations were measured for developing fruitlets and fruit at harvest. Ten fruitlets per replication were collected when fruit diameter reached 25 mm (31 May 2016). For fruit at harvest, 15 fruit per replication were collected, a 1-cm-wide equatorial slice

Fig. 1. Vegetative shoot length extension of 'Honeycrisp' trees treated with either high or low rates of gibberellic acid (GA₃) or prohexadione-calcium (P-Ca) measured at 40 d after full bloom (DAFB; light-gray bars) or 67 DAFB (dark-gray bars) in 2016 compared with an untreated control. Vertical bars represent SE (n = 3). Means with the same letter are not significantly different at $P < 0.05$ (Fisher's least significant difference).

was taken from each fruit, and the core was removed. Fruitlet and mature fruit samples were oven-dried at 60 °C for 1 week, ground using a mortar and pestle, then ground to micron size using a high-throughput homogenizer (VWR, Radnor, PA). A dried tissue sample (200 ± 1 mg) was weighed into digestion tubes with an analytical balance (Mettler-Toledo, LLC, Columbus, OH). Samples were then hot plate-digested with 6 mL HNO₃. The digest was filtered with a 0.45 µm polytetrafluoroethylene (PTFE) filter (Thermo Fisher Scientific, Waltham, MA), and then diluted 100× and analyzed at the Washington State University soil chemistry service laboratory using an Agilent 4200 MP-AES (Agilent, Santa Clara, CA) and run in combination with Ca, Mg, and K inductively coupled plasma (ICP) standards that were chosen to bracket the approximate concentrations of the digests.

Fruit quality and bitter pit assessment. In 2016 and 2017, 15 fruit per replication were sampled to determine fruit quality and maturity at harvest. Mean fruit weight and diameter were measured for each fruit. Background color was determined using the Washington Tree Fruit Research Commission (WTFRC) 'Honeycrisp' Background Color Scale 1–4, where 1 = green, 2 = break, 3 = yellow, and 4 = pink or red. The percentage of red color was estimated using the WTFRC Color Classification–Percent Red Scale 1–4, where 1 = 0% to 25%, 2 = 25% to 50%, 3 = 50% to 75%, and 4 = 75% to 100%. The peel was removed from ≈3 cm² on the sun-exposed and shaded side of the fruit to measure fruit firmness using a Fruit Texture Analyzer (Güss Manufacturing Ltd., Strand, South Africa). Firmness [kilograms force (kgf)] was calculated as the average of the two measurements. The fruit

was then cut in half horizontally along the equatorial line of the apple and a 1-cm equatorial section of each fruit was sprayed with a 30-mm potassium iodide solution using a handheld spray bottle. After 30 min, the starch pattern was rated visually by using a 1–6 scale according to the WTFRC Starch Index for ‘Honeycrisp’ (Hanrahan, 2012), where 1 = most starch and 6 = no starch. Juice from a slice from the shoulder of each

fruit was collected using a Juiceman Juicer (Juiceman, Middleton, WI) for each replication. Soluble solids concentration (measured as a percentage) was measured from the pooled juice sample using a pocket refractometer (Pal-1; Atago U.S.A. Inc., Bellevue, WA). The juice was frozen immediately after sampling for soluble solids content. Titratable acidity (TA; percent malic acid) was also measured from the pooled juice sample. The juice samples were thawed for at least 1 h before TA assessment, then titration was done with a Metrohm Titrator 815 Robotic USB Sample Processor XL and Titrando 888. After harvest, ‘Honeycrisp’ apples ($n = 45$) were stored in refrigerated air at 2 °C for 4 months and 3 months in 2016 and 2017, respectively. Bitter pit incidence was assessed on individual fruit after removal from storage. Bitter pit was assessed as absent/present with surface lesions less than 5 mm in diameter with underlying brown, corky tissue; lesions were confirmed by cutting the fruit.

Statistical analysis. Data analysis of variance (ANOVA) for a completely randomized design was performed for each variable using the SAS statistical package (SAS Institute Inc., Cary, NC). Shoot length and number of internodes data were tested for homogeneity and skewness, then analyzed using a two-way ANOVA. Interactions were presented when significant ($\alpha = 0.05$); otherwise, significant main effects were presented. Bitter pit incidence expressed as percentages were transformed for analysis when

Fig. 2. Vegetative shoot length extension of ‘Honeycrisp’ trees treated with either high or low rates of gibberellic acid (GA_3) or prohexadione-calcium (P-Ca) measured at 40 d after full bloom (DAFB; light-gray bars) or 67 DAFB (dark-gray bars) in 2017 compared with an untreated control. Vertical bars represent SE ($n = 3$). Means with the same letter are not significantly different at $P < 0.05$ (Fisher’s least significant difference).

Fig. 3. Five representative extension shoots taken at 40, 67, and 100 d after full bloom (DAFB) from ‘Honeycrisp’ apple trees treated with either high or low rates of gibberellic acid (GA_3) or prohexadione-calcium (P-Ca) in 2017 compared with an untreated control. Meter stick marked at 10-cm intervals.

appropriate to ensure a normal distribution of variance. Differences between mean values for each treatment were compared using Fisher's least significant difference test ($P < 0.05$).

Results and Discussion

Shoot length extension is primarily affected by altering the intermodal length when using P-Ca. Shoot length extension was significantly different among treatments (Figs. 1 and 2). Shoot length was significantly reduced in 'Honeycrisp' trees when trees were sprayed at petal fall and then 35 d after with low or high rates of P-Ca. These observations are consistent with previous research for other cultivars (Elfving et al., 2002; Greene, 1999; Lafer and Schröder, 2003; Paulson et al., 2005; Privé et al., 2006). In 2016, shoot length was the greatest for trees treated with low GA₃ and high GA₃, compared with low and high rates of P-Ca (Fig. 1). In 2017, shoot length was significantly less for trees treated with a high rate of P-Ca compared with the rest of the treatments (Fig. 2) ($P < 0.0001$). In contrast to 2016, a low rate of P-Ca did not inhibit shoot length extension compared with untreated control trees or a high rate of GA₃ in 2017. Overall tree vigor was less in 2017 compared with 2016, in which shoot length averaged 30.4 and 33.4 cm, respectively (data not shown).

The difference in response between the 2 years could be caused in part by a second flush of growth that occurred after the first measurement in 2016, but did not occur in 2017. Trees treated with P-Ca did not resume growth after application in either year, but in 2016, trees that were either untreated or treated with GA₃ continued to grow after the first measurement in 2016 but not 2017 (Fig. 3). In 2017, between the first and second set of measurements at 40 DAFB and 67 DAFB, mean shoot length increased by ≈ 6 cm for the two GA₃ treatments and the untreated control. For trees treated with both rates of P-Ca, shoot growth had stopped earlier and there was no significant change in shoot length between 40 DAFB and 67 DAFB ($P = 0.2099$, data not shown). The response to GA₃ was less consistent. Shoot length was significantly greater for trees treated with low rates of GA₃ after at 67 DAFB only, compared with the rest of the treatments. Only those trees treated with the low rate of GA₃ continued to grow after 40 DAFB.

In 2016, the average number of internodes was less for trees treated with low and high rates of P-Ca compared with low and high rates of GA₃ (Fig. 4). Similarly, the number of internodes for trees treated with low and high rates of GA₃ was not significantly different compared with untreated control trees. In 2017, there was a treatment effect ($P < 0.0001$) on the total number of internodes for first-year growth (Fig. 5). The number of internodes was the least for trees treated with the high rate of P-Ca and was significantly different from the other

Fig. 4. The number of internodes for 'Honeycrisp' trees treated with either high or low rates of gibberellic acid (GA₃) or prohexadione-calcium (P-Ca) on first-year growth of 'Honeycrisp' trees in 2016 compared with an untreated control. Vertical bars represent standard SE ($n = 3$). Means with the same letter are not significantly different at $P < 0.05$ (Fisher's least significant difference).

Fig. 5. The number of internodes for 'Honeycrisp' trees treated with either high or low rates of gibberellic acid (GA₃) or prohexadione-calcium (P-Ca) on first-year growth of 'Honeycrisp' trees in 2017 compared with an untreated control. Vertical bars represent SE ($n = 3$). Means with the same letter are not significantly different at $P < 0.05$ (Fisher's least significant difference).

treatments. The number of internodes was the greatest for trees treated with the low rate of GA₃ and was significantly different from the untreated control and low and high rates of P-Ca treated trees, but was not significantly different from trees treated with the high rate of GA₃. Across both years, high rates of P-Ca produced shoots with the least number of internodes. However, the average number of internodes was only 20% less than the control or trees treated with GA₃; therefore, the impact on overall leaf area and transpiration balance was relatively small. Also, most of the additional growth occurred between 40 DAFB and 67 DAFB, which is after the period of cell division and when most of the Ca deposition in fruit occurs (Lakso and Goffinet, 2013).

Trees treated with P-Ca often have been reported to have the same number of shoots as untreated trees (Cline, 2006). Although shoot number was not measured here, there was not an expected difference in overall shoot number in these experiments. P-Ca inhibited shoot length significantly, generally by reducing internode length (Fig. 6) rather than by reducing the number of nodes, regardless of the low or high rate. However, the high rate of P-Ca resulted in significantly fewer internodes in 2017 (Fig. 5). Although leaf area was not measured specifically in this experiment, because P-Ca had a greater effect on the internode length of first-year growth rather than the number of internodes, the total leaf area of the tree was also likely less affected. Because total leaf area is a large

contributor to whole-tree water use, given the results, P-Ca likely did not affect the transpiration balance between leaves and fruit.

Fruit elemental concentrations were unaffected by altering shoot length extension using plant growth regulators. Fruit elemental concentration is a key factor affecting storability and resistance to the development of postharvest disorders (Ferguson and Watkins, 1983). Ca, K, and Mg concentrations were unaffected by either P-Ca or GA₃ applications (Table 2). Similarly, treatments did not affect

leaf elemental concentrations when sampled in either June or September (data not shown). Here, Ca concentrations in the fruit decreased more rapidly from June to harvest than K, which decreased by ≈25% in concentration; or Mg, which decreased only minimally in concentration. During fruit development, fruit expansion can lead to dilution of the Ca pool and Ca concentrations in fruit decrease relative to other more mobile nutrients, such as K, Mg, and nitrogen (Kalsits et al., 2017). Similar patterns were observed

here, where Mg/Ca, K/Ca and (K + Mg)/Ca ratios in fruit were greater at harvest compared with fruit at 47 DAFB. K/Ca ratios measured for fruit at harvest in all treatments suggested that the approximate range in bitter pit should be 10% to 25% (Baugher et al., 2017).

The transpiration rates of individual plant organs have been implicated in the transport of Ca via the xylem stream (Montanaro et al., 2006). For fruit, limiting overall transpiration has been reported to decrease fruit Ca

Fig. 6. The mean internodal length for ‘Honeycrisp’ trees treated with either high or low rates of gibberellic acid (GA₃) or prohexadione-calcium (P-Ca) compared with an untreated control on first-year growth of ‘Honeycrisp’ trees in 2016 (black bars) and 2017 (gray bars). Vertical bars represent SE (n = 3).

Table 2. Calcium (Ca), magnesium (Mg), and potassium (K) concentrations in fruit at harvest for trees treated with high or low rates of gibberellic acid (GA₃) and high and low prohexadione-calcium (P-Ca), and compared with an untreated control.

Treatment	47 D after full bloom (31 May 2016)					
	Ca (%) ^z	Mg (%)	K (%)	Mg/Ca	K/Ca	(K + Mg)/Ca
High GA ₃ (32 mg·L ⁻¹)	0.200	0.099	1.942	0.501	9.764	10.265
Low GA ₃ (16 mg·L ⁻¹)	0.219	0.108	1.825	0.492	8.337	8.829
Control	0.195	0.110	1.965	0.576	10.356	10.931
High P-Ca (125 mg·L ⁻¹)	0.191	0.099	1.951	0.526	10.370	10.897
Low P-Ca (62.5 mg·L ⁻¹)	0.189	0.101	1.826	0.537	9.722	10.259
Significance	NS	NS	NS	NS	NS	NS
P value (α = 0.05)	0.4476	0.3756	0.5214	0.7137	0.5100	0.5264
Harvest (24 Aug. 2016)						
High GA ₃ (32 mg·L ⁻¹)	0.068	0.118	1.320	1.536	20.386	21.922
Low GA ₃ (16 mg·L ⁻¹)	0.088	0.115	1.245	1.174	15.339	16.512
Control	0.071	0.055	1.257	0.799	18.261	19.059
High P-Ca (125 mg·L ⁻¹)	0.056	0.052	1.336	0.950	24.354	25.304
Low P-Ca (62.5 mg·L ⁻¹)	0.089	0.071	1.305	0.804	14.845	15.649
Significance	NS	NS	NS	NS	NS	NS
P value (α = 0.05)	0.2420	0.6721	0.4010	0.5723	0.0985	0.0757

^zCa, Mg, and K are expressed as percent dry matter.

^{NS}Non-significant.

Fisher’s least significant difference test was used for mean separation.

concentrations in apricot (Montanaro et al., 2010) and kiwi (Montanaro et al., 2014). In this case, if the overall leaf area was unchanged, it could be expected that the transpirational balance would also be unchanged. Because shoot length manipulation with growth promoters (GA₃) or growth retardants (P-Ca) affected internode length more than the total number of internodes, it is not surprising that there were no consistent differences in fruit elemental concentrations among treatments. Here, in young trees with relatively low crop loads and strong shoot growth, fruit Ca concentrations were not consistently improved by using shoot growth-inhibiting plant growth regulators.

Fruit yields and weight were not predictably affected by manipulating shoot length extension. In 2016, mean fruit yield from trees treated with high rates of P-Ca (and thus less shoot length extension) was

significantly greater compared with the untreated control or trees treated with either a low or a high rate of GA₃ (Fig. 7). The average yield per tree from trees that were treated with high rates of P-Ca was not significantly different compared with trees treated with low rates of P-Ca. In 2017 (Fig. 8), there were no significant differences in fruit size among any of the treatments. These results indicate that the growing season environment may contribute to the effect of plant growth regulators on fruit size. Because crop load was carefully controlled, there would not be a large difference in yield unless fruit size was affected significantly by these treatments. Fruit size was associated with overall fruit yield (Figs. 7 and 8). Interestingly, fruit size was significantly greater when trees were treated with high rates of P-Ca in 2016 but not in 2017 (Fig. 8). In 2016, fruit from trees treated with either a

high or low rate of P-Ca had the greatest fruit weight compared with other treatments, with a mean fruit weight of 371 g and 326 g, respectively. However, mean fruit weight from trees treated with a low P-Ca rate was not significantly different from the low or high GA₃ rates (Fig. 8). Untreated trees had the least mean fruit weight, and fruit weight was not significantly different for fruit harvested from trees treated with either high or low GA₃ compared with the untreated control (Fig. 8). Fruit diameter followed a similar pattern as fruit weight and was the greatest in trees that were treated with high-rate P-Ca compared with fruit from trees treated with either GA₃ or the untreated control, indicating that the use of GA₃ or P-Ca did not affect fruit shape substantially (data not shown).

Manipulating shoot length extension in a newly productive orchard did not substantially affect fruit quality. Color development in apples is a function of light exposure to maturing fruit. Therefore, excess vigor can reduce red color formation in bicolor varieties like ‘Honeycrisp’. In contrast, in 2016, red overcolor was the least for fruit from trees that were treated with high rates of P-Ca (Table 3). In 2017, there were no differences among treatments for red color development in fruit (Table 4). Even in young trees, inhibition of vegetative growth from P-Ca can improve the light environment for developing fruit (Schupp et al., 2001). However, these patterns were not observed in this experiment, in which color development was unaffected by altering overall shoot length extension. Background color is a function of maturity of the apple in which chlorophyll degradation during ripening leads to fruit losing greenness and becoming more yellow (Song et al., 1997). Background color of fruit was not significantly different among treatments in 2016 (Table 3). However, in 2017, the background color was more yellow for fruit from trees treated with low rates of GA₃ compared with other treatments, although it was not significantly different from the control (Table 4).

Mean fruit firmness was less for fruit from trees treated with low rates of P-Ca compared with the untreated control, but was not significantly different compared with the other treatments in 2016 (Table 3). In 2017, fruit firmness was the greatest for fruit from trees treated with a high rate of P-Ca, although it was not significantly different from the control (Table 4). Fruit firmness at harvest in 2017 varied by less than 0.3 kg pressure among all treatments. These differences appeared to be related to overall fruit size. In 2016, the greatest soluble solids content was in fruit treated with high rates of GA₃, high rates of P-Ca, and the control (Table 3). Soluble solids content was greater in these three treatments compared with fruit from trees that were treated with low rates of P-Ca. Although these differences may have been statistically significant, the differences among treatments were relatively small and did not result in lower quality fruit among treatments. TA was not significantly different

Fig. 7. Fruit yield per tree for ‘Honeycrisp’ trees treated with either high or low rates of gibberellic acid (GA₃) or prohexadione-calcium (P-Ca) compared with an untreated control on first-year growth of ‘Honeycrisp’ trees in 2016 (black bars) and 2017 (gray bars). Vertical bars represent SE (n = 3).

Fig. 8. Mean fruit mass per tree for ‘Honeycrisp’ trees treated with either high or low rates of gibberellic acid (GA₃) or prohexadione-calcium (P-Ca) compared with an untreated control on first-year growth of ‘Honeycrisp’ trees in 2016 (black bars) and 2017 (gray bars). Vertical bars represent SE (n = 3).

Table 3. ‘Honeycrisp’ fruit quality at harvest in 2016 from trees treated with high or low rates of gibberellic acid (GA₃) and high and low prohexadione-calcium (P-Ca), and compared with an untreated control.

Treatment	BC (1–4)	% RC (1–4)	Firm (kgf)	SSC (%)	TA (% MA)	Starch (1–6)
High GA ₃ (32 mg·L ⁻¹)	1.3 ± 0.1	3.2 ± 0.1 a	6.8 ± 0.1 ab	14.2 ± 0.2 a	0.360 ± 0.085	4.3 ± 0.2 ab
Low GA ₃ (16 mg·L ⁻¹)	1.4 ± 0.1	3.1 ± 0.1 a	6.6 ± 0.1 ab	13.7 ± 0.2 b	0.356 ± 0.070	4.5 ± 0.1 ab
Control	1.3 ± 0.1	3.0 ± 0.1 a	6.9 ± 0.1 a	13.8 ± 0.1 ab	0.439 ± 0.025	4.8 ± 0.2 a
High P-Ca (125 mg·L ⁻¹)	1.4 ± 0.1	2.8 ± 0.1 b	6.6 ± 0.1 ab	14.0 ± 0.1 ab	0.442 ± 0.035	4.2 ± 0.2 b
Low P-Ca (62.5 mg·L ⁻¹)	1.5 ± 0.1	3.2 ± 0.1 a	6.5 ± 0.1 b	13.1 ± 0.1 c	0.378 ± 0.045	4.6 ± 0.2 ab

Values are means ± SE (n = 3). Different letters denote statistical differences among means ($P < 0.05$) using a Fisher’s least significant difference means comparison test.

BC = background color (1–4); % RC = percentage of red color (range, 1–4); Firm = fruit firmness [measured in kilograms force (kgf)]; SSC = soluble solids content; TA (% MA) = titratable acidity (measured in percent malic acid equivalents); Starch = starch pattern index (range, 1–6).

Table 4. ‘Honeycrisp’ fruit quality at harvest in 2017 for trees treated with high or low rates of gibberellic acid (GA₃) and high and low prohexadione-calcium (P-Ca) and compared with an untreated control.

Treatment	BC (1–4)	% RC (1–4)	Firm (kgf)	SSC (%)	TA (% MA)	Starch (1–6)
High GA ₃ (32 mg·L ⁻¹)	1.5 ± 0.1 b	2.8 ± 0.1	6.7 ± 0.1 b	13.7 ± 0.1 a	0.573 ± 0.027	5.4 ± 0.1
Low GA ₃ (16 mg·L ⁻¹)	1.8 ± 0.1 a	2.6 ± 0.2	6.7 ± 0.1 b	13.3 ± 0.2 ab	0.585 ± 0.025	5.4 ± 0.1
Control	1.7 ± 0.1 ab	2.8 ± 0.1	6.9 ± 0.1 ab	13.5 ± 0.2 ab	0.585 ± 0.009	5.7 ± 0.1
High P-Ca (125 mg·L ⁻¹)	1.5 ± 0.1 b	2.7 ± 0.1	7.0 ± 0.1 a	13.1 ± 0.2 b	0.614 ± 0.017	5.2 ± 0.1
Low P-Ca (62.5 mg·L ⁻¹)	1.5 ± 0.1 b	2.8 ± 0.1	6.7 ± 0.1 b	13.3 ± 0.2 ab	0.591 ± 0.027	5.4 ± 0.1

Values are means ± SE (n = 3). Different letters denote statistical differences among means ($P < 0.05$) using a Fisher’s least significant difference means comparison test.

BC = background color (1–4); % RC = percentage of red color (range, 1–4); Firm = fruit firmness [measured in kilograms force (kgf)]; SSC = soluble solids content; TA (% MA) = titratable acidity (measured in percent malic acid equivalents); Starch = starch pattern index (range, 1–6).

Table 5. The effects of high and low rates of gibberellic acid (GA₃), and high and low prohexadione-calcium (P-Ca) on bitter pit incidence in ‘Honeycrisp’ apple after postharvest storage in 2016 and 2017, respectively. Each value represents the mean bitter pit incidence per treatment (n = 3).

Treatment	High GA ₃ (32 mg·L ⁻¹)	Low GA ₃ (16 mg·L ⁻¹)	Control	High P-Ca (125 mg·L ⁻¹)	Low P-Ca (62.5 mg·L ⁻¹)
Bitter pit (%)					
2016	12.5	18.1	4.4	16.3	15.6
Treatment significance			NS		
P value			0.3747		
Bitter pit (%)					
2017	18.1	14.6	11.1	12.5	25.0
Treatment significance			NS		
P value			0.5034		

^{NS}Nonsignificant ($P > 0.05$).

among treatments for either 2016 or 2017 (Tables 3 and 4).

Bitter pit incidence was not related to shoot growth extension. In 2016 and 2017, bitter pit incidence was not significantly different among treatments for fruit stored in air (Table 5). Although bitter pit incidence was not significantly different among treatments, there appeared to be a trend in which bitter pit incidence was more related to fruit size rather than mean shoot extension. Previous literature shows that there is an inverse curvilinear relationship between fruit Ca and fruit diameter, which shows that fruit Ca decreases as fruit size increases (Oberly and Kenworthy, 1961; Perring and Jackson, 1975). This could explain the relationship of bitter pit incidence related to fruit size observed here.

Conclusion

Excessive vegetative vigor in apple trees can be problematic because of its competition with fruit growth (Basak and Rademacher, 2000; Byers and Yoder, 1999; Costa et al., 2002), and can also affect fruit quality, yield, and pest control (Forshey et al., 1992; Greene, 1999; Miller and Tworowski, 2003). Excessive vigor has been linked to low levels of Ca in the

fruit and the occurrence of bitter pit. This study demonstrated that shoot extension could be manipulated with plant growth regulators in the field, but did not have a significant effect on fruit mineral composition. Manipulating vegetative vigor did not change the number of internodes substantially, which is one of the critical factors affecting overall leaf area and the balance between vegetative growth and fruit. Either accelerating or inhibiting vegetative growth did not significantly affect bitter pit incidence. Growth inhibition using gibberellin inhibitors to decrease shoot length extension was not effective at controlling bitter pit in ‘Honeycrisp’. Further studies that identify horticultural strategies that reduce overall leaf area for high-vigor trees are likely to be more successful in reducing bitter pit incidence in apple.

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