

Prohexadione-calcium and Acibenzolar-*S*-methyl Effects and Interactions I: Vegetative Growth Management in a Young North Carolina Apple Orchard

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Abstract. Young apple (*Malus × domestica* Borkh.) orchards are intensively managed to ensure successful orchard establishment. Use of growth retardants may aid in disease management; however, concerns regarding reduced bearing surface and increased time to full production are disincentives for use in young orchards. Prohexadione-calcium (P-Ca) is a gibberellin synthesis inhibitor used in apple to manage vigor and fire blight (*Erwinia amylovora*). Acibenzolar-*S*-methyl (ASM) is a systemic acquired resistance inducer used to induce an immune response in plants to protect against fungal and bacterial pathogens. The objective of the study was to apply different suggested rates of P-Ca and ASM alone and in combination to determine their impacts on shoot growth rate, tree size, canopy infill, and crop load. In 2021 to 2024, the study was conducted in a newly planted experimental ‘Gala’ orchard with a factorial treatment structure of two factors [P-Ca (0, 42.5, and 125 mg·L⁻¹) and ASM (0 and 37.5 mg·L⁻¹)]. Treatments were applied twice each season, on the same plots each year to determine cumulative effects. As P-Ca rate increased, shoot growth rate and tree height decreased. Even with a decrease in tree size, canopy infill and linear bearing surface were maintained regardless of P-Ca or ASM rates. Fruit set was increased and return bloom decreased as P-Ca rate increased, but the addition of ASM lessened these effects. These results indicate that repeated applications of P-Ca on young orchards, in high-vigor locations, may reduce tree size, but not to a level that limits orchard establishment. P-Ca may increase fruit set, but ASM may have utility in lessening this effect and managing crop load.

Apple (*Malus × domestica* L. Borkh.) orchard production faces growing challenges with the threat of climate change, loss of land and labor, disease pressures, and increasing apprehension around pesticide use. A shift in

orchard production systems has arisen out of need to maximize space (Barritt et al. 2008), increase light interception (Robinson and Lakso 1991), and increase labor efficiency. Orchard production efficiencies have been increased with the adoption of high-density systems (Barritt et al. 2008; Palmer and Warrington 1998). Since the 1980s, tree density increased from 500 to 5000+ trees per hectare in some commercial orchards (Robinson et al. 2006). The goal of these systems is to produce high-quality fruit early in the life of the orchard to recuperate the increased orchard establishment costs of high-density production systems. The increased light interception of high-density systems leads to increased cumulative yield and improved fruit quality (Lordan et al. 2018), ideally translating into future profitability. Adapted management techniques must be developed and

used to maximize efficiency and productivity while also protecting against disease.

Fire blight caused by *Erwinia amylovora* is a serious and economically important pathogen in many areas where apples are grown (Sparks 2001). Warm, humid conditions during bloom lead to outbreaks of blossom blight (van der Zwet 2012), and late spring rain events can increase the spread of secondary infection shoot blight (Wallis et al. 2021). In newly planted, nonbearing orchards, vegetative growth is promoted via cultural practices to fill orchard space and realize full production earlier. This vigorous, vegetative growth is particularly susceptible to shoot blight (Suhayda and Goodman 1981). The traditional approach to fire blight control is the use of agricultural antibiotics (Sundin et al. 2009), which have been restricted in many areas of the world due to perceived adverse effects to human health (Phillips 2007) and increased antibiotic-resistant strains of *E. amylovora* (Chiou and Jones 1995). As a result, identifying alternative fire blight control strategies is a research priority.

Prohexadione-calcium (P-Ca) is an effective growth retardant (Greene 1999) that inhibits gibberellin biosynthesis through the inhibition of dioxygenase enzymes (Davies 2004). Further investigation into the mode of action of P-Ca found that P-Ca mimics a cosubstrate to the deoxygenase pathway in the biosynthesis of active growth gibberellins, called 2-oxyglutaric acid. As a result, the biosynthesis of gibberellins is reduced (Rademacher et al. 2006). Reduced gibberellin results in reduced cell elongation in shoots. Benefits of reduced shoot growth include increased spray coverage and air flow due to decreased growth and decreased susceptibility to fire blight because of the arrest of active shoot growth (Miller 2002; Rademacher and Kober 2003). In young orchards, growth and canopy infill is desirable, so the use of P-Ca is not common. P-Ca also increases fruit set through the interruption of the ethylene biosynthesis pathway (Rademacher et al. 2006), which can lead to decreased fruit size (Greene 2008), increased thinning efforts, and poor return bloom (Cooley 2007). P-Ca resembles ascorbic acid in the plant which can interfere with 1-aminocyclopropane-1-carboxylic acid oxidase, a precursor to 1-aminocyclopropane-1-carboxylic acid and thus ethylene biosynthesis. It is thought to interfere with the action of some chemical thinners because of this interference in the ethylene biosynthesis pathway (Rademacher et al. 2006).

P-Ca is an effective fire blight management aid (Aldwinckle et al. 2000; Norelli and Miller 2004). The primary mechanism of protection is a thickening of cell walls, which prevents bacterial penetration and could slow the progression of bacteria (McGrath et al. 2009). P-Ca has been shown to affect flavonoid pathways, inducing increased production of a flavonoid that could contribute to plant protective qualities. For a while, this flavonoid, leuteoferol, was thought to be the mechanism of plant protection from fire blight, but that has since been disputed (Rademacher et al. 2006). When P-Ca is applied at bloom

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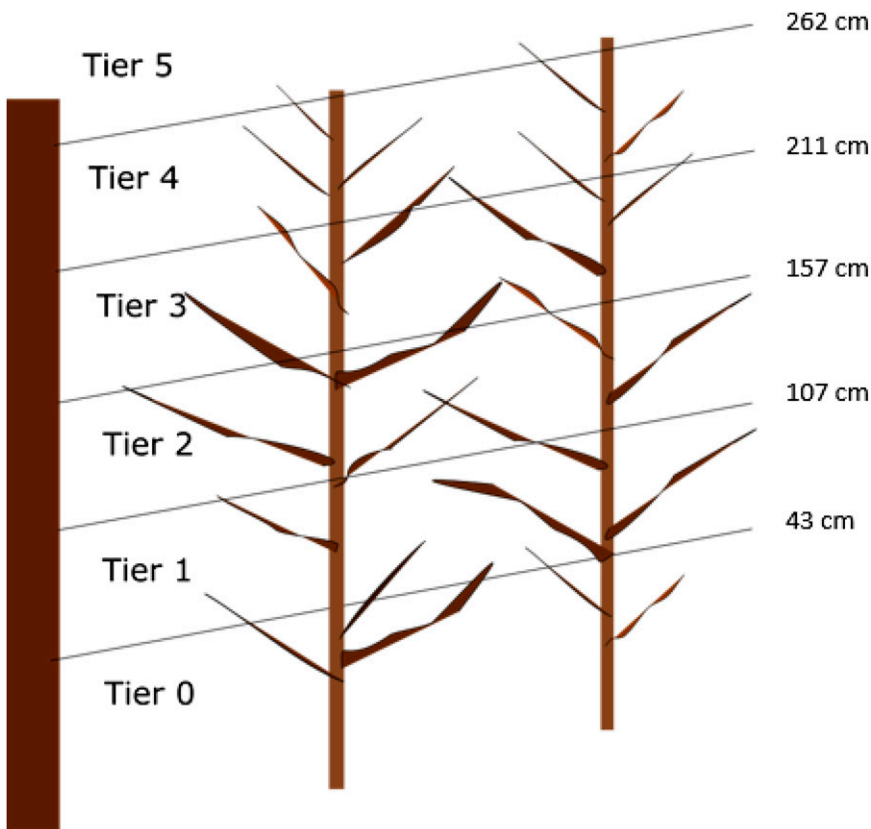


Fig. 1. Total linear bearing surface of ‘Simmons Gala’ apple (*Malus × domestica* Borkh.) measurements were taken in the dormant season. The length and count of all branches were taken on two representative trees from the five-tree plots, and the subsample was averaged. Shoot location was classified by the tiers, which were separated by trellis wires.

or pink stage, it reduces shoot blight spread in the late spring when vegetative tissue is most susceptible (Cox et al. 2019; Yoder et al. 1999). Due to concerns that P-Ca will inhibit canopy infill and reduce productivity of young trees, P-Ca is most frequently applied for disease management in mature orchards under high disease pressure (Aldwinckle et al. 2000; Costa et al. 2004; Norelli and Miller 2004).

An alternative approach to disease control is through the use of chemicals that induce a plant resistance called systemic acquired resistance (SAR). Acibenzolar-*S*-methyl (ASM) is a host defense inducer that aids in increased resistance to fungal, bacterial, and viral pathogens in a range of plants (Maxson-Stein et al. 2002). ASM was first used as a defense activator in annual plants, but the use of ASM to manage fire blight in pear led to the discovery of its efficacy in apple (Ishii et al. 1999). As a SAR inducer, ASM moves systemically to elicit defense responses before plant tissue is exposed to phytopathogens. ASM acts as an analog to a stress hormone, salicylic acid (Kunz et al. 1997). In response, plants produce pathogenesis-related proteins that activate defense systems. Sakata et al. (2020) found that ASM activates the production of reactive oxygen species leading to the closure of stomata and limiting potential infection points. Spring foliar applications of ASM resulted in decreased incidence and severity of fire blight in the spring, but trees became susceptible to

infection later in the summer (Maxson-Stein et al. 2002). ASM should be used as a supplemental product. The combination of ASM with other alternative control measures such as P-Ca could have additive or even synergistic effects, resulting in a management option that could reduce antibiotic use. Unfortunately,

the cost of ASM is currently prohibitive to widespread commercial adoption. Further research needs to be done to understand the effects of cost-effective, low-rate ASM applications on the presence, survival, and evolution of more virulent strains of *E. amylovora*.

The use of PGRs and SAR inducers should be tailored to orchard site, production goals, and disease management strategy and refined for specific cultivars. Further investigation of the main effects and interactions of P-Ca and ASM is required in young orchard systems, which often are susceptible to fire blight but do not typically receive vegetative growth-mitigating products. The present study evaluated the cumulative effects and interactions of P-Ca and ASM on vigor and growth rate, plant productivity and leaf function, and canopy infill and architecture. A concurrent study evaluated disease incidence and severity and bacterial systemic movement (Vogel et al. 2025). We hypothesized that P-Ca at both low and high rates would decrease shoot growth rate and canopy infill and would increase return bloom, while ASM would have limited effects on any horticultural parameters.

Materials and Methods

Experimental orchard and treatments. The experiment was conducted from 2021 to 2024 on ‘Simmons Gala’/‘M.9-T337’, maintained at the Mountain Horticultural Crops Research and Extension Center in Mills River, NC (lat. 35.428079°N, long. 82.563295°W, elevation 649 m). Trees were planted 30 Mar 2021 with 0.9 × 4-m tree-row spacing in rows oriented east to west. On the day of planting, white latex paint with 5000 ppm of 6-benzyladenine [6-BA (Exilis 9.5 SC; Fine Americas, Inc., Walnut Creek, CA, USA)] was applied to the central leader of trees to enhance lateral branching. Trees were trained to tall spindle (Robinson et al. 2006). Commercial

Table 1. Main effects and interactions of acibenzolar-*S*-methyl (ASM) and prohexadione-calcium (P-Ca) rate on tree size in ‘Simmons Gala’ (*Malus × domestica* Borkh.).

Variable	TCSA (cm ²) ⁱ				Height (cm)		
	2021	2022	2023	2024	2021	2022	2023
ASM ⁱⁱ							
Yes	1.2	1.9	4.9 b	7.1	201	243	291
No	1.1	2.2	5.7 a	7.8	201	245	295
P-Ca (mg·L ⁻¹)							
125	1.2	1.9	4.6	6.5	202	236	281
42.5	1.1	2.1	5.8	7.7	201	239	296
0	1.1	2.2	5.6	8.1	200	256	302
Significance ⁱⁱⁱ	ns	ns	Q	L	ns	Q	L
P value	0.8183	0.1372	0.0229	0.0131	0.9522	0.0085	0.0059
R ²	0.15	0.14	0.24	0.20	0.00	0.30	0.24
F significance ^{iv}							
ASM	0.7578	0.0655	0.0168	0.1993	0.8907	0.7465	0.4952
P-Ca	0.8009	0.1265	0.0125	0.0559	0.9543	0.0113	0.0241
ASM × P-Ca	0.0733	0.6951	0.3483	0.9586	0.4316	0.4805	0.2382

ⁱ TCSA is calculated as $\pi \cdot \left(\frac{\text{Avg circ}}{2\pi}\right)^2$, where Avg circ = average circumference. Height was measured from the ground to the terminal bud of the leader.

ⁱⁱ For the ASM rate, Yes = 37.5 mg·L⁻¹ and No = 0 mg·L⁻¹.

ⁱⁱⁱ L = linear model; Q = quadratic model. P value is for the model ($n = 30$).

^{iv} F significance shows P values from full factorial analysis of variance main effects and interactions. Trunk cross-sectional area (TCSA) and tree height were measured in spring each year. Year was not evaluated as a factor. Instead, the data were analyzed by year. ns = not significant.

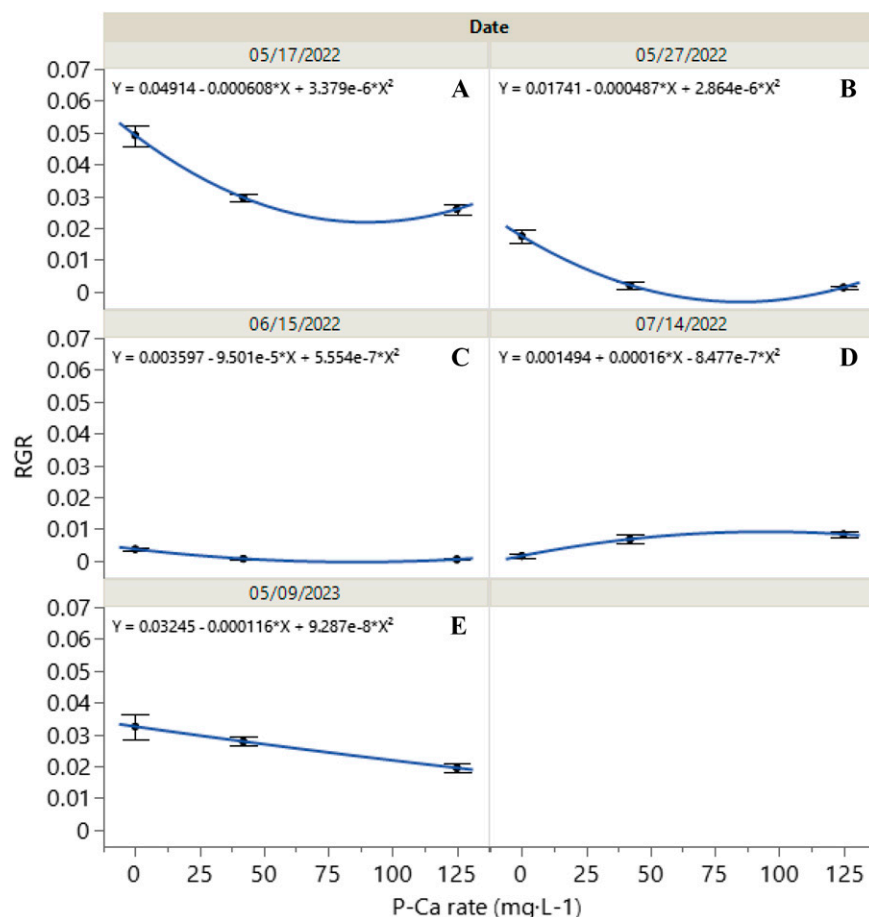


Fig. 2. Relative growth rate (RGR) of ‘Simmons Gala’ apple (*Malus × domestica* Borkh.) shoots was calculated throughout the season by repeated measures of 10 shoot samples from each experimental unit. Dates when the main effect of prohexadione-calcium (P-Ca) were significant are shown. Acibenzolar-S-methyl (ASM) had no effect on RGR. As the P-Ca rate increased, RGR decreased in a negative curvilinear fashion. (A) $P < 0.0001$, $R^2 = 0.692$. (B) $P < 0.0001$, $R^2 = 0.777$. (C) $P < 0.0001$, $R^2 = 0.569$. In 2022, the first application of P-Ca was on 8 May, so the first two dates were the most efficacious time period. The final date in 2022 demonstrates the recovery of treated trees, which respond with a positive curvilinear relationship between P-Ca rate and RGR. (D) $P = 0.0002$, $R^2 = 0.462$. Only one timepoint in 2023 demonstrated a main effect of P-Ca rate on RGR, which was a linear reduction. (E) $P = 0.0040$, $R^2 = 0.336$. In 2023, the first P-Ca application took place on 4 May ($n = 30$).

management practices for fertility, pesticide, crop load management, and herbicide applications adhered to local recommendations in all years of the study. In the years 2021 and 2022, fruit was removed from trees to align with commercial management practices. Specifically, all trees received 150 mg·L⁻¹ 6-BA + 1200 mg·L⁻¹ 1-naphthyl methylcarbamate (Carbaryl 4L; Drexel Chemical, Memphis, TN, USA) + 0.25% (v/v) nonionic surfactant (Regulaid; Kalo, Inc., Overland Park, KS, USA) at ~14-mm fruit diameter to defruit trees chemically. Any persisting fruit were removed manually after June drop.

Experimental units consisted of five-tree plots. Treatments were implemented in a completely randomized design and replicated five times. Single trees served as a buffer between treatments. The treatments were applied twice in May. The first application occurred when shoots were ~2.5 cm long, and the second application occurred between 10 and 14 d later. The application dates were 20 May and 1 Jun 2021, 28 Apr and 8 May 2022, and 4 and 16 May 2023. The factorial treatment structure

was composed of three rates of P-Ca (Kudos; Fine Americas, Inc.) and two rates of ASM (Actigard 50WG; Syngenta, Greensboro, NC, USA). The rates were as follows: P-Ca = 125, 42.5, and 0 mg·L⁻¹; applied with 0.125% (v/v) nonionic surfactant and 0.39% (v/v) water conditioner (Choice Trio; Loveland Products, Inc., Loveland, CO, USA); ASM = yes, no (37.5 and 0 mg·L⁻¹, respectively).

Height and trunk cross-sectional area. Two representative trees were chosen from each five-tree plot for annual measurements. Tree height (cm) and trunk circumference (cm) were measured on 13 May 2021, 13 May 2022, and 1 Jan 2023. In 2024, trunk circumference was measured on 5 Mar. Tree height was a measure of length from the ground to the terminal bud of the leader. Trunk circumference was measured at 30 cm above the graft union, and trunk cross-sectional area (TCSA) was calculated using Eq. [1]:

$$TCSA = \pi \cdot \left(\frac{\text{Avg circ}}{2\pi} \right)^2 \quad [1]$$

Tree height and TCSA were expressed as an average of the two representative sample trees.

Shoot lengths and relative growth rate (RGR). When vegetative shoots were ~2 cm, five shoots on two representative trees from each experimental replicate were tagged and labeled for repeated measures. Primary shoots emerging from 2-year-old wood or terminals of 1-year-old wood were chosen, excluding latent shoots from dormant pruning cuts. Shoot lengths were measured from the terminal bud scar to the growing tip terminus. Over the course of the season, shoot lengths were measured on multiple dates (between seven and eight measurement dates), and relative growth rate was calculated (Eq. [2]) where L1 and L2 represent initial and final length, respectively, and T1 and T2 represent the timings of initial and final, respectively.

$$\text{Relative growth rate} = \frac{[\ln(L2) - \ln(L1)]}{(t2 - t1)} \quad [2]$$

Shoot length measurement intervals were variable. Shoots were measured at frequent intervals (7 to 14 d) up to 49 d after treatment and then monthly until shoot growth ceased for the season. Shoot lengths and growth rates of each shoot were averaged across the two-tree, ten-shoot sample.

Gap fraction leaf area index. Leaf area index (LAI) was measured on 10 Oct 2022 using a plant canopy imager with a hemispherical lens (CI-110; CID Bioscience, Camas, WA, USA). The canopy imager was positioned immediately above the lowest trellis wire (~0.61 m irrigation wire) between two trees. This method captures half of the canopy from two separate trees within the same plot. Two measurements were taken for each five-tree plot. In 2023, the canopy imager was used on 22 May and 26 Jun. From the hemispherical photos taken, the gap fraction, the proportion of visible sky to canopy, was analyzed using the Beer-Lambert law (Monsi and Saeki 2005). LAI is the ratio of leaf area to ground area.

Leaf function. Using a porometer/fluorimeter (LI-600; LI-COR Biosciences, Lincoln, NE, USA), quantum yield of photosystem II (Fv/Fm), stomatal conductance (gs), and electron transport rate were estimated. On one representative tree from each plot, six leaves were measured, three from each row side. In 2023, measurements were taken on multiple dates starting -1 to 63 d after treatment.

Blossom cluster density. In 2022 to 2025, blossom clusters were counted at bloom on two representative trees in each five-tree plot. Blossom cluster density was calculated as the quotient of the blossom cluster count and TCSA.

Thinning, fruit set, and yield. In 2021 and 2022, all fruit were removed from trees so that canopy infill would not be influenced by competition of fruit. In 2023, trees were chemically thinned at ~10- and ~20-mm fruit diameter with 600 mg·L⁻¹ carbaryl. In 2024, 75 mg·L⁻¹ 6-BA + 600 mg·L⁻¹ carbaryl was applied at ~10-mm fruit diameter. After June drop, the trees required supplemental hand thinning to achieve an

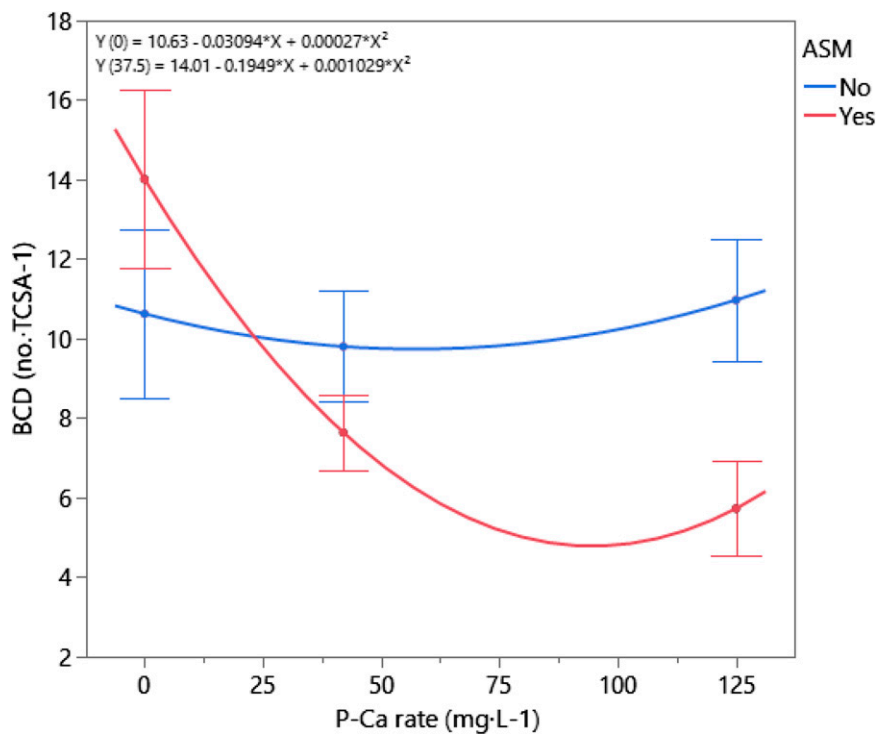


Fig. 3. Interaction of plot illustrating the relationship between prohexadione-calcium (P-Ca) rate and acibenzolar-*S*-methyl (ASM) on blossom cluster density (BCD) of 'Simmons Gala' apple (*Malus × domestica* Borkh.) in 2022 ($P = 0.0445$). ASM was applied at a rate of 75 mg·L⁻¹. P-Ca concentration had no influence on BCD, and the main effects of ASM on BCD were not observed. When ASM was added to P-Ca, as the rate of P-Ca increased, BCD decreased ($n = 30$).

appropriate crop load based on tree size. Any small or damaged fruit were removed, and clusters were singled to prevent branch breakage. All hand-thinned fruit from five-tree plots were harvested, placed in labeled containers, and run through an electronic fruit sorter (Durand-Wayland, Inc., LaGrange, GA, USA) outfitted with a load cell, color and infrared camera system, and full transmittance spectrometer (TrueSort Electronics; Ellipsis, Eindhoven, Netherlands).

At harvest on 14 Aug 2023 and 14 Aug 2024, yield was measured on a per plot basis. Average fruit number, weight (g), and blush (%) were measured by the fruit sorter. A partial economic analysis was conducted using free on board prices from a local packer/shipper. Crop density before and after hand thinning was calculated as total fruit divided TCSA.

Whole tree measurements/total linear bearing surface. During the dormant season in 2022 to 2024 and before pruning, the length and count of all branches were taken on two representative trees from the five-tree plots. The spatial distribution of lateral branches was determined. Specifically, shoot location was classified by tiers, which were separated by trellis wires (Fig. 1). Average shoot length and linear bearing surface were calculated to provide a characterization of productive wood.

Dormant pruning. Trees were pruned with industry standards by removing all branches below the bottom wire (~0.61 m), any larger than half of the trunk diameter, and any with a narrow branch angle. Remaining branches were simplified where shoots were greater

than ~20 cm or if the branch bifurcated. The weight of pruned branches was measured on an experimental unit basis (five-tree plot) using a field scale (WPT 30.FX.K; RADWAG, Radom, Poland) after the 2021, 2022, and 2023 growing seasons. Dormant pruning weights and cuts were then expressed on an average-per-tree basis.

Statistical analysis. Statistical computation was performed using statistical software (JMP Pro version 17; SAS, Cary, NC, USA). A standard least-squares model was used to evaluate the effects and interactions of treatments using two-way analysis of variance [significance ($\alpha \leq 0.05$)] for tree height, TCSA, blossom cluster density, shoot lengths, growth rate, linear bearing surface, LAI, ΦPSII, stomatal conductance, and yield. Where P-Ca was the only significant main effect, generalized linear regression analysis was performed. Where ASM was a significant main effect, a *t* test ($\alpha \leq 0.05$) was performed. All data were analyzed within the time point collected (e.g., "year" was not used as a model effect for data in tables).

Results and Discussion

Height and TCSA. When trees were planted in 2021, before treatments were applied, there were no differences in height or TCSA. This suggests that experimental units at the initiation of the experiment were uniform. In 2022 and 2023, as the rate of P-Ca increased, the height of trees decreased ($P = 0.0085$ and 0.0059 , respectively; Table 1). ASM did not

influence tree height, nor were there any interactions between P-Ca and ASM. In 2023, both P-Ca and ASM had a significant main effect on TCSA, but there was no interaction between factors. P-Ca reduced TCSA in a curvilinear manner, and ASM reduced TCSA ($P = 0.0229$ and 0.0168 , respectively; Table 1). In 2024, P-Ca decreased TCSA linearly as the rate increased ($P = 0.0131$; Table 1). There was no significant effect of ASM.

Previous research has shown that P-Ca hastened terminal bud set when applied in spring (Evans et al. 1997) and midsummer (Owens and Stover 1999) and can decrease tree height (Javeed 2017; Kashirskaya et al. 2021) and TCSA (Byers et al. 2004; Çetinbaş et al. 2015). In North Carolina, the application of P-Ca was effective during the spring flush of growth, ultimately reducing total tree height the following year. By 2024, even with reduced growth due to P-Ca applications, 96% of trees reached the top trellis wire of a tall spindle system (2.62 m). Thus, P-Ca applications to the same plots for 3 consecutive years did not severely impede vertical canopy infill. TCSA was reduced by P-Ca in accordance with some studies (Byers et al. 2004; Çetinbaş et al. 2015), while others found no impact of P-Ca on TCSA (Duyvelshoff and Cline 2013; Javeed 2017). Trunk radial growth can be decreased when crop load is high (Plavcová et al. 2022), and P-Ca increases fruit set (Byers et al. 2004; Cooley 2007). The decrease in TCSA in response to P-Ca in 2023 and 2024 could be due to late thinning and high fruit set, respectively, rather than general reduction in vigor.

Relative growth rate. In 2021, shoot RGR was unaffected by P-Ca, ASM, and P-Ca × ASM except on one time point, between 2 and 8 Jun 2021, when ASM reduced RGR by 53.1% (2021 data not presented; $P = 0.0360$). RGR was not affected strongly by treatments since all trees were likely under transplant stress. Potentially, the addition of stress response induced by ASM compounded with planting stress could have caused the brief cessation in growth. In 2022, P-Ca had a main effect of reducing growth rate from 17 May to 15 Jun 2022. P-Ca had a negative curvilinear relationship with RGR ($P < 0.0001$, $R^2 = 0.692$; $P < 0.0001$, $R^2 = 0.777$; $P < 0.0001$, $R^2 = 0.569$, respectively; Fig. 2A–C). Between 23 Jun and 14 Jul 2022, the effects of P-Ca were reversed as there was a positive curvilinear relationship between P-Ca rate and RGR ($P = 0.0002$, $R^2 = 0.462$; Fig. 2D). The effects of P-Ca typically last a few weeks, and the half-life of the product is ≤ 8 weeks (Carra et al. 2017; Evans et al. 1999). By Aug 2022, the effects of P-Ca had worn off, and trees that were artificially put into paradormancy by P-Ca resumed growth, while others set terminal buds. Notably, by 14 Sep 2022, the average shoot lengths were not significantly affected by either P-Ca or ASM, even though they had been reduced by P-Ca earlier in the season. Norelli and Miller (2004) similarly evaluated shoot lengths at different time periods, but not RGR, and they saw decreased early-season growth as P-Ca rate increased. When two high-dose applications of P-Ca were applied,

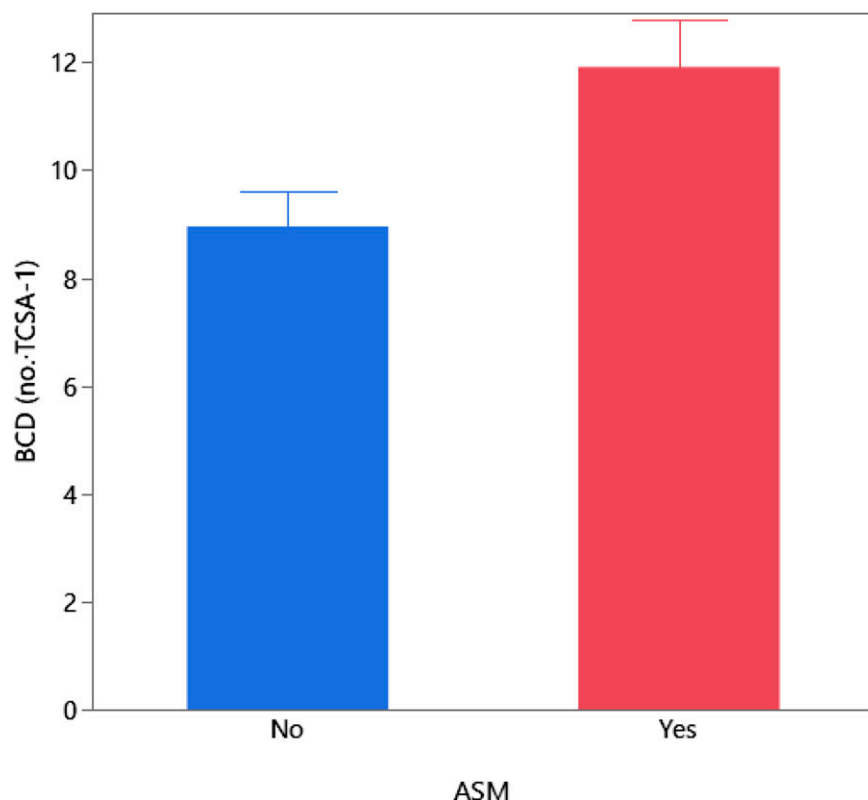


Fig. 4. In 2023, blossom cluster density (BCD) of 'Simmons Gala' apple (*Malus × domestica* Borkh.) was increased by acibenzolar-S-methyl [ASM ($P = 0.0106$)]. ASM was applied at a rate of $75 \text{ mg} \cdot \text{L}^{-1}$. The vertical bars represent the standard error of the mean ($n = 30$).

there was a strong response of late-season growth resulting in no difference in total season growth between two high doses and three

to five low doses. Unrath (1999) observed renewal of terminal budbreak at 60 d after petal fall, when initial application was made.

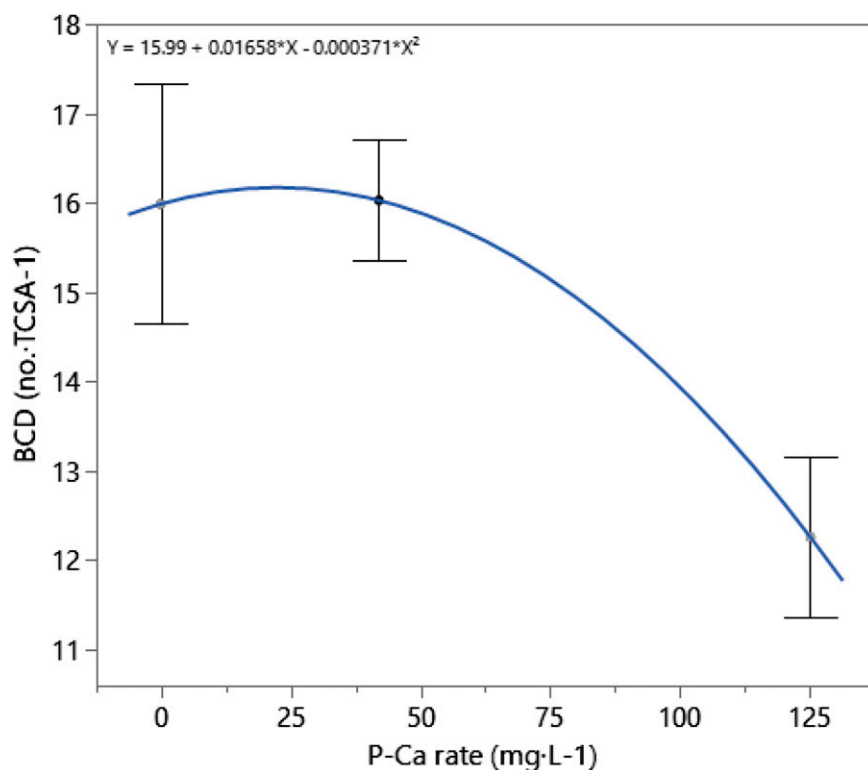


Fig. 5. In 2024, blossom cluster density (BCD) of 'Simmons Gala' apple (*Malus × domestica* Borkh.) had a negative curvilinear relationship with the rate of prohexadione-calcium [P-Ca ($P = 0.0267$)]. The vertical bars represent the standard error of the mean ($n = 30$).

Ultimately, even when P-Ca reduces growth rates during the season, in a humid subtropical climate like North Carolina (Köppen 1884), treated trees can recover and produce canopies similar to those that are not treated.

Between 20 Apr and 9 May 2023, P-Ca had a main effect on RGR, and there was a negative linear relationship between P-Ca rate and RGR ($P = 0.0040$, $R^2 = 0.336$; Fig. 2E). Applications of P-Ca were made 19 Apr and 2 May 2023, so this early time point aligns with when P-Ca should be most active. All other dates in 2023, there was no significant effect of P-Ca or ASM on shoot RGR. Furthermore, 2023 was the first year the trees retained a crop load to harvest. Consequently, vegetative growth rate was affected by the competition of fruit. Overall, P-Ca reduced growth during the critical period of shoot blight, but season long shoot growth was not affected. ASM did not further reduce shoot RGR alone or in combination with P-Ca.

It is unsurprising that RGR was reduced as the rate of P-Ca increased since it is well recorded that P-Ca is effective at limiting shoot growth in apple (Byers and Yoder 1999; Greene 1999; Miller 2002; Norelli and Miller 2004; Unrath 1999). Our study also showed a reduction in shoot lengths when P-Ca was applied (data not shown). However, relative growth rate as a measure gives a better view into the efficacy of the product over time when compared with shoot length measurements alone. RGR accounts for the initial size of shoots and the seasonal changes that take place as terminal buds set naturally before a potential flush of growth in late summer. P-Ca had a greater impact in the early season and when there was no competition between fruit and vegetative growth. Privé et al. (2006) also found that P-Ca is most effective earlier in the season when the natural RGR of shoots is higher (Rademacher and Kober 2003) and thus more sensitive to P-Ca. In general, it is recommended to apply P-Ca at petal fall with 10- to 14-d intervals between applications (Rademacher and Kober 2003). For fire blight control, P-Ca use can be limited to two early season applications when growth is most vigorous. Our research confirms that when competition between vegetative growth and fruit is minimal, P-Ca can have a more significant effect on RGR.

Gap fraction leaf area index. Gap fraction LAI, an expression of how much light is able to penetrate a tree's canopy, did not differ across treatments. P-Ca and ASM had no effects or interactions when measurements were taken 10 Oct 2022. Since shoot lengths and LAI were equal across all factors at the close of the 2022 season, there is evidence that canopies were similar regardless of any level of P-Ca and ASM. In 2023, LAI was measured at multiple time points, and there were no main effects or interactions of P-Ca and ASM. Javeed (2017) investigated the physiological impacts of applications of salicylic acid (SA) and P-Ca at numerous rate combinations. Only very high rates of P-Ca ($600 \text{ mg} \cdot \text{L}^{-1}$) affected leaf area, measured by sampling leaves from the center of the tree and using a leaf area meter. ASM, as an

Table 2. Main effects and interactions of acibenzolar-S-methyl (ASM) and prohexadione-calcium (P-Ca) rate on components of crop in ‘Simmons Gala’ (*Malus × domestica* Borkh.).

Variable	Fruit removed at hand thinning ⁱ				Harvest					
	Fruit no.	Mean fruit wt (g)	Yield (kg)	Crop density ⁱⁱ (fruit/cm ²)	Fruit no.	Mean fruit wt (g)	Yield (kg)	Blush (%)	Crop density ⁱⁱ (fruit/cm ²)	Estimated crop value (US dollars/ha) ⁱⁱⁱ
ASM ^{iv}										
Yes	104 b	59.2	6.1 b	12	183	141.4	26.0	72.4 b	8	\$17,511
No	148 a	60.3	8.6 a	13	198	147.1	28.9	78.5 a	7	\$20,971
P-Ca (mg·L ⁻¹)										
125	172	54.6	9.3	16	191	132.5	25.2	78.7	8	\$16,315
42.5	110	63.3	7.0	10	190	151.6	29.0	72.9	7	\$21,454
0	95	61.4	5.8	11	191	148.7	28.1	74.9	7	\$19,954
Significance ^v	Q	Q	L	Q	ns	Q	ns	ns	Q	ns
P value	0.0005	0.0087	0.0017	<0.0001	0.9805	0.0245	0.4016	0.1812	0.0384	0.1021
R ²	0.43	0.30	0.30	0.56	0.00	0.24	0.07	0.12	0.21	0.09
F significance ^{vi}										
ASM	0.0186	0.6624	0.0100	0.5235	0.2875	0.3768	0.3530	0.0175	0.4806	0.0764
P-Ca	<0.0001	0.0087	0.0078	<0.0001	0.9990	0.0245	0.1998	0.1812	0.0445	0.0900
ASM × P-Ca	0.0227	0.1009	0.0888	0.1976	0.0338	0.0160	0.0884	0.0305	0.4855	0.0964

ⁱ Thinning involved removing damaged and excessively small fruit and singling clusters.

ⁱⁱ Crop density is calculated as the quotient of total number of fruits that set (sum of fruit number at thinning and harvest) and trunk cross-sectional area. Harvest crop density is the number of fruits harvested divided by TCSCA.

ⁱⁱⁱ Estimated crop value based on 2778 trees per hectare; 0.9 × 4-m spacing.

^{iv} For the ASM rate, Yes = 37.5 mg·L⁻¹ and No = 0 mg·L⁻¹.

^v L = linear model; Q = quadratic model. P value is for the model ($n = 30$).

^{vi} F significance shows P values from full factorial analysis of variance main effects and interactions.

Hand thinning occurred on 5 Jul 2023, and harvest occurred on 14 Aug 2023. Harvest was a total tree harvest on a per plot basis. Fruit number, weight, and yield were measured per five-tree plot at the hand-thinning stage to evaluate effects on efficacy of a chemical thinner applied at ~10 and 20 mm. On 14 Aug 2023, the plots were harvested, and fruit number, weight, yield, and blush were measured. ns = not significant.

analog to SA, may also have some ability to influence leaf area, as high rates of SA (150 to 200 mg·L⁻¹) were able to increase leaf area compared with low rates and SA + P-Ca

(Javeed 2017). In our study, the rates of P-Ca and ASM used did not reach a threshold that would change leaf area index. Even if trees may be smaller by some metrics, the amount of light

intercepted by canopies does not significantly differ through the season. Therefore, applications of P-Ca and ASM twice at the start of the season do not affect the canopy enough to restrict its use in a humid subtropical climate (Hernandez-Ochoa and Asseng 2018) like that in North Carolina, even early in the life of an orchard.

Leaf function. When treated with ASM, leaves showed a reduction in ΦPSII in 2022; however, measurements were only taken on one date (3 Aug), due to the availability of the equipment. There were no main effects of P-Ca rate nor an interaction with P-Ca and ASM. Stomatal conductance and electron transport rate were not affected by P-Ca or ASM. In 2023, measurements with the porometer occurred on four dates: 3 May, 17 May, 5 Jun, and 6 Jul. Variable and inconsistent responses were observed, suggesting that both factors evaluated (P-Ca and ASM) did not have a meaningful impact of stomatal conductance and ΦPSII of leaf tissue at the rates and timings tested.

Blossom cluster density (BCD). In 2022, following treatments in 2021, P-Ca and ASM interacted to reduce blossom cluster density ($P = 0.0445$; Fig. 3). When ASM was used alone, BCD was maintained; however, when P-Ca and ASM were used together, there was a negative curvilinear relationship between the rate of P-Ca and BCD. P-Ca alone had no significant impact on BCD. The combination of these two factors results in a lower potential fruit set and thus higher BCD the subsequent year. In 2023, there was a main effect of ASM, which increased BCD relative to no ASM ($P = 0.0106$; Fig. 4). The previous year, the combination of ASM and P-Ca reduced BCD, potentially allowing for an increased BCD in trees treated with ASM, in 2023. In

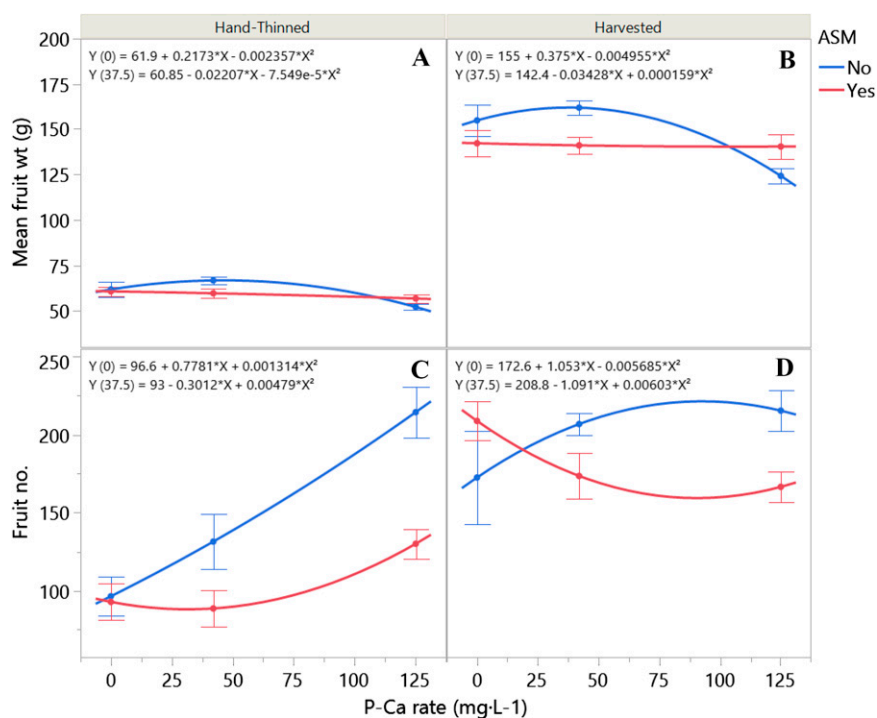


Fig. 6. The effects and interaction of prohexadione-calcium (P-Ca) rate and acibenzolar-S-methyl (ASM) on ‘Simmons Gala’ apple (*Malus × domestica* Borkh.) fruit number and weight at two timings in 2023. ASM was applied at a rate of 75 mg·L⁻¹. Fruit was collected on a per-plot basis. The hand-thinned fruit (A, C) was collected after “June drop” to quantify the necessity for further thinning. Hand-thinned fruit weight decreased ($P = 0.0087$) and number increased ($P = 0.0005$) as P-Ca rate increased. When ASM was applied with P-Ca, the increase in fruit number due to P-Ca was suppressed ($P = 0.0227$). At harvest on 14 Aug 2023, the interaction between P-Ca rate and ASM resulted in maintenance of fruit weight [B ($P = 0.0160$)] and decrease in fruit number [D ($P = 0.0338$)], even with high rates of P-Ca. Vertical bars represent the standard error of the mean ($n = 30$).

2024, BCD had a negative curvilinear relationship with P-Ca rate ($P = 0.0267$; Fig. 5). The high fruit set due to P-Ca in 2023 resulted in decreased BCD in 2024. ASM had no main effect. In 2025, BCD was not affected by P-Ca rate, ASM, or the interaction.

As a gibberellic acid (GA) inhibitor, it is expected that P-Ca would affect flowering and yield (Elfving et al. 1991; Greene 1999; Owens and Stover 1999). Byers et al. (2004) and Cooley (2007) report that high rates of P-Ca would increase fruit set and reduce the following year's return bloom. In some cases, high rates of P-Ca did not increase fruit set but still decreased return bloom the following year (Greene 2008). When fall applications of P-Ca are applied, flowering in the spring could be increased, depending on cultivar (Owens and Stover 1999).

In 2022–23, fruitlets were removed from trees in the spring to encourage orchard establishment. In any given growing season, the next year's buds will be induced and begin differentiation as early as 3 to 6 weeks after full bloom (Buban and Faust 1982). Flower bud formation is an incredibly complex process and can be influenced by multiple factors (Campbell and Kalcsits 2024; Elsysy and Hirst 2019; Milyaev et al. 2022). During flower bud formation, there is antagonism between fruitlets and the forming buds (Tromp 2000), possibly due to the presence of GA in apple seeds (Faust 1989; Hoad 1978), although promotion of flower formation originates in tissues beyond spur level (Elsysy and Hirst 2019). The inhibitory effects of endogenous GA on apple flower bud development remains unsettled as additional phytohormones and their interactions have significant impacts on signaling and gene

expression (Milyaev et al. 2022). Thinning fruit >30 d after full bloom will result in decreases in return bloom relative to earlier timings (Koike et al. 2003). While even a short interval of maintaining fruit could influence the fate of future buds, the removal of fruit before a harvest timing may have minimized the effects of P-Ca and ASM on return bloom in 2022–23. In contrast, maintaining a crop through harvest in 2023 began to express the full effect of P-Ca on fruit set and the ensuing return bloom. Since fruit set increased as the rate of P-Ca increased, return bloom decreased in response. The consistency of BCD across treatments in the spring of 2025 is likely a result of consistent crop load management the previous year.

Crop density and thinning. Trees were chemically thinned in 2023 and 2024; however, additional hand thinning was required to achieve consistent crop load and minimize limb and tree breakage. Average number, weight, and yield of hand-thinned fruit were calculated to determine main effects and interactions of P-Ca and ASM on volume of undesirable persisting fruit. In 2023, fruit number at the hand-thinning stage was affected by the main effects and interaction of P-Ca and ASM (Table 2). The relationship between P-Ca and fruit weight was negative curvilinear ($P = 0.0087$; Fig. 6A). The main effect of P-Ca rate on the number of thinned fruits was a positive curvilinear relationship ($P = 0.0005$; Fig. 6C). The high rate of P-Ca resulted in smaller fruit but higher yield because of the increased hand-thinning effort required. The main effect of ASM reduced fruit number and yield of hand-thinned fruit by 29.7% and 29.1%, respectively (Table 2).

Because P-Ca increased fruit number and ASM decreased it, when used together, ASM lessened the positive effect of P-Ca on fruit number (Fig. 6). P-Ca rate had a positive curvilinear relationship with crop density after June drop, before hand thinning (Table 2; $P < 0.0001$).

In 2024, fruit number and yield decreased by 35.3% and 35.2%, respectively, when ASM was applied compared with no ASM (Table 3). Average fruit weight was affected by the interaction of P-Ca rate and ASM when the combination of a high rate of P-Ca and an application of ASM had increased fruit weight compared with when ASM was applied alone. Fewer fruits required removal after chemical thinning when ASM was applied. There were no significant effects of P-Ca rate or ASM application on crop load after chemical thinning and before June drop (Table 3). Hand-thinning efforts should therefore be proportional to tree size across the orchard, so the minimal difference in fruit number, size, and yield of removed fruit is fitting.

P-Ca reduces ethylene formation because of its interference in the ethylene biosynthesis pathway (Rademacher et al. 2006), resulting in increased fruit set (Cooley 2007). Some studies confirm an increased fruit set when high rates of P-Ca are applied (Basak and Rademacher 2000; Greene 2007, 2008), while others found that fruit set was not affected by P-Ca (Byers and Yoder 1999; Costa et al. 2004; Miller 2002). Although chemical thinning becomes more difficult when P-Ca is applied, it is still possible to thin effectively or even overthin (Greene 2007). In 2023, in North Carolina, high rates of P-Ca increased fruit set and thus the amount of time and labor

Table 3. Main effects and interactions of acibenzolar-S-methyl (ASM) and prohexadione-calcium (P-Ca) rate on components of crop in 'Simmons Gala' (*Malus × domestica* Borkh.).

Variable	Fruit removed at hand thinning ⁱ				Harvest					
	Fruit no.	Mean fruit wt (g)	Yield (kg)	Crop density ⁱⁱ (fruit/cm ²)	Fruit no.	Mean fruit wt (g)	Yield (kg)	Blush (%)	Crop density ⁱⁱ (fruit/cm ²)	Estimated crop value (US dollars/ha) ⁱⁱⁱ
ASM ^{iv}										
Yes	167 b	28.0	4.6 b	17	410	119.4	48.3	69.7 b	12	\$24,508
No	258 a	27.7	7.1 a	19	453	122.9	56.4	75.8 a	12	\$34,934
P-Ca (mg·L ⁻¹)										
125	217	28.7	6.1	17	342	121.8	41.7	76.4	11	\$23,940
42.5	199	28.5	5.7	17	423	129.3	55.3	71.2	12	\$34,638
0	221	26.4	5.9	19	528	112.4	60.1	70.6	13	\$30,586
Significance ^v	ns	ns	ns	ns	L	ns	L	ns	ns	ns
P value	0.7711	0.0727	0.7447	0.4016	0.0031	0.0928	0.0236	0.1344	0.0532	0.4265
R ²	0.02	0.18	0.00	0.03	0.27	0.16	0.17	0.08	0.13	0.06
F significance ^{vi}										
ASM	<0.0001	0.6540	<0.0001	0.1670	0.3437	0.5029	0.2177	0.0280	0.8150	0.0950
P-Ca	0.6084	0.0455	0.8483	0.5435	0.0095	0.0412	0.0683	0.1491	0.1666	0.3551
ASM × P-Ca	0.1459	0.0245	0.1538	0.7144	0.2929	0.0047	0.1942	0.0020	0.8620	0.0642

ⁱ Thinning involved removing damaged and excessively small fruit and singling clusters.

ⁱⁱ Crop density is calculated as the quotient of total number of fruits that set (sum of fruit number at thinning and harvest) divided by trunk cross-sectional area. Harvest crop density is the number of fruits harvested divided by TCSA.

ⁱⁱⁱ Estimated crop value based on 2778 trees per hectare; 0.9 × 4-m spacing.

^{iv} For the ASM rate, Yes = 37.5 mg·L⁻¹ and No = 0 mg·L⁻¹.

^v L = linear model; Q = quadratic model. P value is for the model ($n = 30$).

^{vi} F significance shows P values from full factorial analysis of variance main effects and interactions.

Hand thinning occurred on 25 Jun 2024, and harvest occurred on 14 Aug 2024. Harvest was a total tree harvest on a per plot basis. Fruit number, weight, and yield were measured per five-tree plot at the hand-thinning stage to evaluate effects on efficacy of a chemical thinner applied at ~10 mm. On 14 Aug 2024, the plots were harvested, and fruit number, weight, yield, and blush were measured. ns = not significant, TCSA = trunk cross-sectional area.

allocated to hand thinning. Higher fruit set resulted in lower fruit weights even at hand thinning, which translates to smaller fruit at harvest. In 2024, crop density before hand thinning was equivalent across treatments. Minimal differences in number and weight of thinned fruit were present. Observing no effects of P-Ca on crop load after chemical thinning could be related to the decreased blossom cluster density experienced after the 2023 season.

Harvest. At harvest in 2023, the main effect of P-Ca rate on fruit weight was a negative curvilinear relationship ($P = 0.0245$; Table 2; Fig. 6B). The main effect of ASM reduced blush by 7.8% (Table 2). The interaction of P-Ca rate and ASM affected fruit number, fruit weight, and blush ($P = 0.0338$, 0.0160, and 0.0305, respectively; Table 2). Fruit number was decreased in a negative curvilinear manner when P-Ca and ASM were used in combination ($P = 0.0338$; Fig. 6D). There was an interaction between P-Ca and ASM when P-Ca alone at a low rate improved crop value, but when ASM was added, crop value decreased. A partial economic analysis seemed to project a threshold at which the improved fruit set of P-Ca was beneficial, but when the rate surpassed this threshold, P-Ca had a negative impact on crop value, largely due to reduced fruit size. The mediation of crop load by ASM along with reduced blush could be responsible for the difference in economic value.

At harvest in 2024, fruit weight was affected by an interaction between P-Ca rate and ASM, where at the low rate of P-Ca alone, fruit weight was increased relative to when combined with ASM, but at the high rate of P-Ca, fruit weight was increased with the addition of ASM ($P = 0.0047$; Fig. 7A; Table 3). Fruit number had a negative linear relationship with P-Ca rate ($P = 0.0095$; Fig. 7B; Table 3). As P-Ca rate increased, fruit harvested decreased. However, there was no effect on crop load. In the spring of 2024, P-Ca rate had a negative curvilinear relationship with blossom cluster density (Fig. 5), which could lead to the difference in final fruit number at harvest. Blush was affected by the main effects of ASM and the interaction between P-Ca rate and ASM. The main effect of ASM decreased blush by 8.0% (Table 3). However, when ASM was used with P-Ca, there was a positive curvilinear relationship ($P = 0.0014$; Fig. 7C). As the rate of P-Ca increased, there was an increase in percentage blush. In 2024, there were no statistically significant effects of P-Ca rate or ASM application on estimated crop value (Table 3).

There is a well-established relationship between crop load and fruit size (Jones et al. 1992; Robinson and Watkins 2003; Schupp 2003; Wright et al. 2006). Achieving an appropriate crop load early in the season improves fruit size (Embree et al. 2007; Serra et al. 2016). Thinning timing ultimately influences final fruit size (McArtney et al. 1996) because of the determination of cell number occurring so early in fruit development

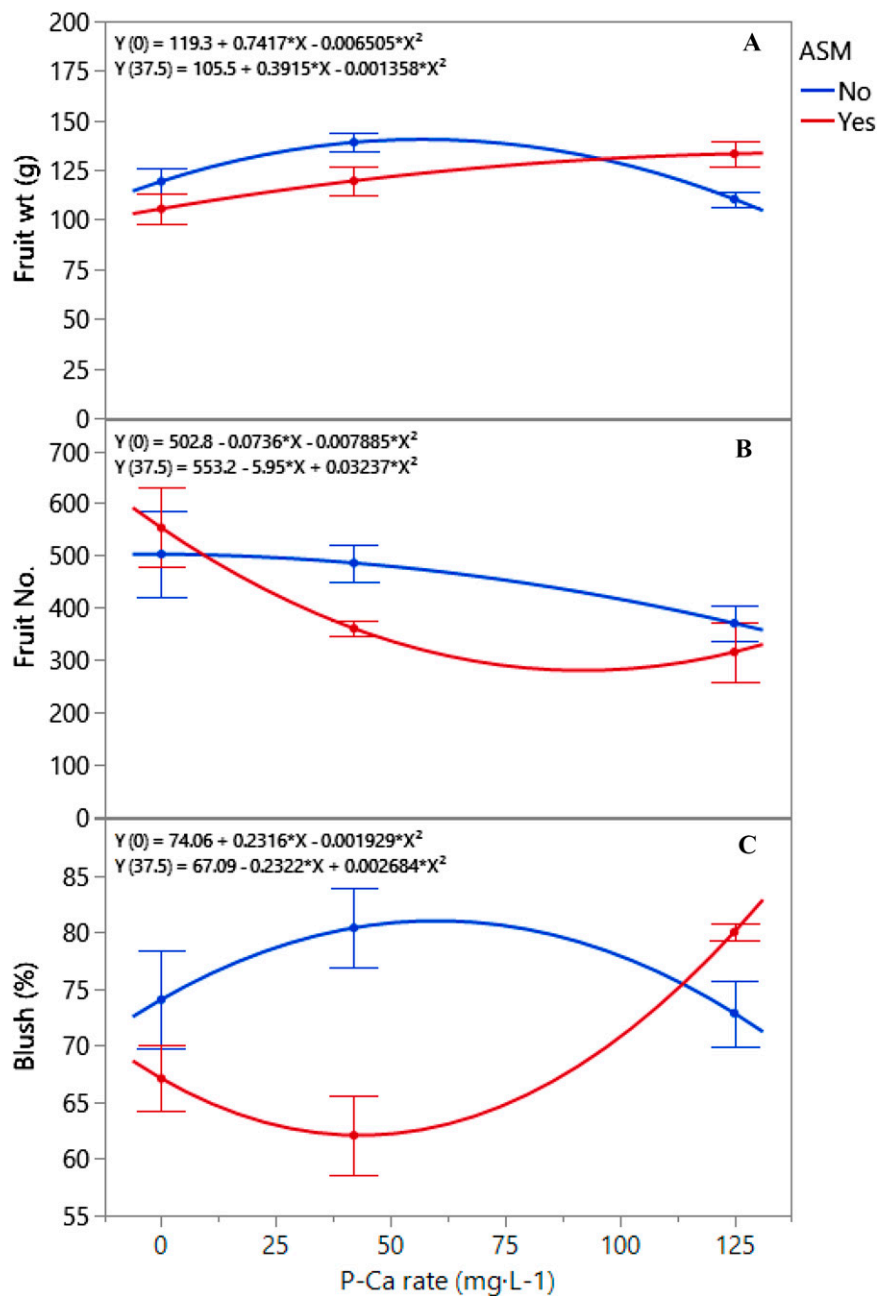


Fig. 7. The effects and interaction of prohexadione-calcium (P-Ca) rate and acibenzolar-*S*-methyl (ASM) on ‘Simmons Gala’ apple (*Malus × domestica* Borkh.) fruit number, weight, and blush at harvest on 14 Aug 2024. ASM was applied at a rate of 75 mg·L⁻¹. Fruit was collected on a per-plot basis. Vertical bars represent the standard error of the mean. At harvest, the interaction between P-Ca rate and ASM resulted in maintenance of fruit weight [A ($P = 0.0047$)] and blush [C ($P = 0.0014$)] at high rates of P-Ca. P-Ca reduced fruit number [B ($P = 0.0095$)]. Vertical bars represent the standard error of the mean ($n = 30$).

(Pearson and Robertson 1953). The late timing of hand thinning in 2023 was a factor in decreased fruit size at harvest. Trees treated with P-Ca retained higher fruit numbers for a longer period, until hand thinning, and when fruit experiences competition during the cell division phase, growth potential cannot be recovered. Higher rates of P-Ca resulted in lower fruit weight, even though number and yield remained insignificant. In 2024, hand thinning occurred 10 d earlier, and trees retained similar numbers of fruit, which could account for the maintenance of fruit weight at harvest. Notably, fruit was generally smaller in 2024 when

compared with 2023. Because hand thinning was implemented, crop density at harvest was consistent across treatments, with a slight increase in density due to high rates of P-Ca in 2023 (Table 2) but no difference in 2024 (Table 3). Since fruit number was unaffected at harvest in 2023, the difference in crop density can be attributed to a reduction in TCSCA as P-Ca rate increases. A reduction in fruit number in 2024 could be attributed to reduced blossom cluster density in the spring.

The effect of ASM when combined with P-Ca helped maintain fruit weight and decrease fruit number relative to when ASM

was not present (Figs. 6B, 6D, 7A, and 7B). The increase in reactive oxygen species in response to ASM could lead to decreased crop productivity (Mansoor et al. 2022). While stomatal closure helps reduce infection points in leaves, it can also influence the assimilation of carbohydrates, limiting availability to growing sinks (Liao et al. 2022). Some studies have found an increased or maintained yield of tomatoes (Imonmion et al. 2023) and melons (Kenney et al. 2020), respectively, related to application of ASM, but this increased yield is attributed to decreased disease and thus a higher number of marketable fruit. Stress induction of ASM may have increased tree sensitivity to chemical thinning and may have potential to increase efficacy of chemical thinning. Use of ASM in the context of an apple crop load management program merits additional research.

Whole-tree measurements/total linear bearing surface. There were no practical or significant differences in total linear bearing surface, branch numbers per tier, or average branch length across treatment factors in 2022. However, in 2023, between the top two wires of the trellis [211 to 262 cm (tier four)], P-Ca reduced branch number and linear bearing surface in a negative curvilinear fashion (Fig. 8). There was no effect of ASM nor an interaction between P-Ca and ASM. Even though tree height was reduced by P-Ca,

linear bearing surface on the whole tree level was unaffected by P-Ca rate. Canopy infill was therefore similar across all trees, even shorter trees, but as discussed previously, trees still experienced sufficient growth to reach the top trellis wire by 2024. The effect of P-Ca on growth could differ across cultivar, rootstock (Crassweller and Smith 2014), application timing, and location (Unrath 1999). To control growth effectively, vigorous cultivars and rootstocks may require higher rates of P-Ca (Byers and Yoder 1999) or more applications at lower rates (Crassweller and Smith 2014; Unrath 1999). Cultivars such as ‘Honeycrisp’ exhibit low vigor and are more susceptible to permanent stunting when compared with higher vigor cultivars like ‘Fuji’. Location effects, such as elevation and climate, affect the growth patterns of apple shoots. Unrath (1999) found that North Carolina has an extended season compared with New York, where shoot growth reached 85% of total growth at 65 d after petal fall in North Carolina compared with 32 d in New York. The extended season of North Carolina resulted in similar tree size and canopy infill by the end of the season in North Carolina. In locations where the season is shorter, the inhibition of growth induced by P-Ca may be more significant and detrimental in young orchards. Even though northern states experience shorter seasons, higher rates of P-Ca after bloom may be required to suppress

growth because of extended daylength during bloom compared with the southeast United States.

Dormant pruning weights. In 2022, there were no significant main effects or interactions of P-Ca and ASM rates on pruning (Table 4). Pruning that was conducted in Winter 2022 was minor and corrective. In 2023, ASM had a main effect on pruning weight, resulting in a 17.7% decrease relative to no ASM ($P = 0.0500$). P-Ca rate also had a negative linear relationship with pruning weight ($P = 0.0323$; $R^2 = 0.15$). In 2024, P-Ca rate and pruning weight again had a negative linear relationship [Table 3 ($P = 0.0070$, $R^2 = 0.0232$)]. P-Ca is known to decrease tree size and vigor (Byers and Yoder 1999; Byers et al. 2004). Winter pruning efforts decrease when shoot growth (Byers and Yoder 1999; Evans et al. 1999) and TCSA (Reig et al. 2020; Robinson et al. 2006) are reduced. Therefore, in orchards where a season-long P-Ca program is used, pruning efforts should decrease (Miller 2002). The effect of ASM on dormant pruning remains undetermined. The first years of orchard establishment often have less significant pruning requirements, leading to insignificant differences in treatments; however, the cumulative effects of subsequent years may reveal more long-term impacts. Because P-Ca inhibits shoot extension while thickening cell walls (McGrath et al. 2009; Wallis and Cox 2020), there could be differences in wood density and therefore pruning weights. In addition, more dormant wood must be removed from trees with higher vigor, which directly affects the time spent pruning (Miller 2002).

Table 4. Main effects and interactions of acibenzolar-S-methyl (ASM) and prohexadione-calcium (P-Ca) rate on weight of pruned wood in ‘Simmons Gala’ (*Malus × domestica* Borkh.).

Variable	Pruning wt (g) ⁱ		
	2022	2023	2024
ASM ⁱⁱ			
Yes	172	299 b	248
No	254	364 a	261
P-Ca (mg·L ⁻¹)			
125	167	277	178
42.5	191	357	270
0	282	361	315
Significance ⁱⁱⁱ	ns	L	L
P value	0.1022	0.0323	0.0070
R ²	0.09	0.15	0.23
F significance ^{iv}			
ASM	0.1141	0.0500	0.7662
P-Ca	0.1621	0.0654	0.0335
ASM × P-Ca	0.3849	0.3299	0.4024

ⁱ Pruning weight measured on a per plot basis and expressed on a per tree basis.

ⁱⁱ For the ASM rate, Yes = 37.5 mg·L⁻¹ and No = 0 mg·L⁻¹.

ⁱⁱⁱ L = linear model; Q = quadratic model. P value is for the model ($n = 30$).

^{iv} F significance shows P values from full factorial analysis of variance main effects and interactions. Year was not evaluated as a factor. Instead, the data were analyzed by year. ns = not significant.

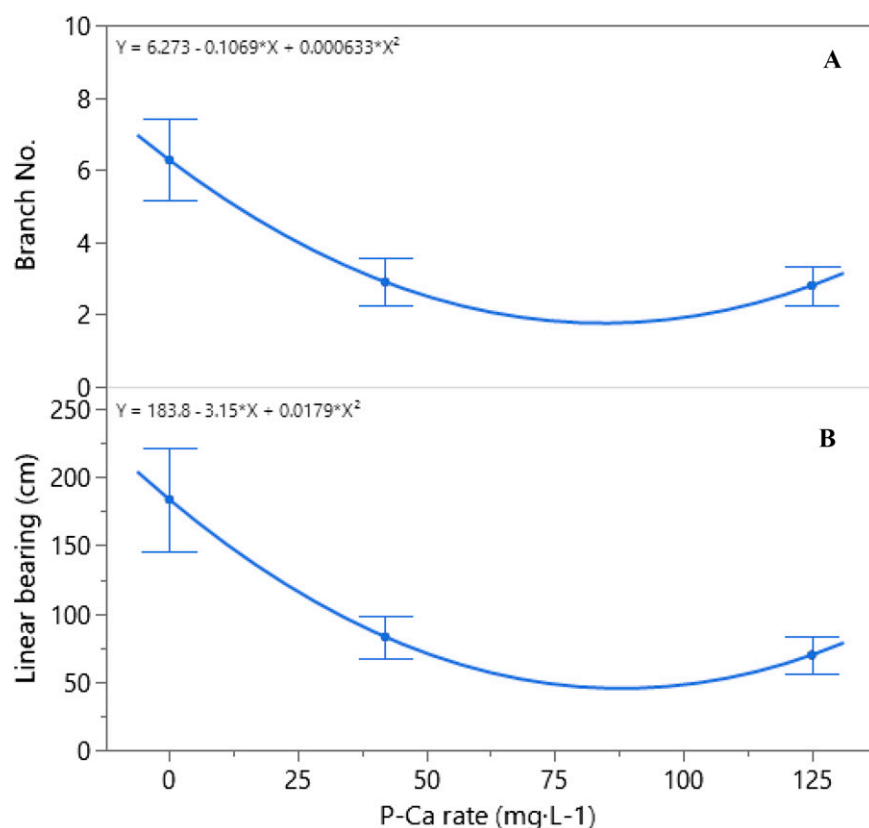


Fig. 8. In 2024, prohexadione-calcium (P-Ca) rate and branch number (A) and bearing surface (B) in the upper portion (between 211 and 262 cm from the ground) of an apple ‘Simmons Gala’ (*Malus × domestica* Borkh.) tree has a negative curvilinear relationship. (A) $P < 0.0084$, $R^2 = 0.289$. (B) $P < 0.0077$, $R^2 = 0.294$. The total linear bearing surface was quantified by counting and measuring all limbs on two representative trees per five tree plot and averaging the subsample. Vertical bars represent the standard error of the mean ($n = 30$).

P-Ca effectively reduced dormant wood and thus pruning efforts in North Carolina.

Conclusions

When ASM and P-Ca were applied to a newly planted orchard for 3 consecutive years, tree vigor, size, and components of crop were influenced. As P-Ca rate increased, relative growth rates and tree size decreased; however, the magnitude of vigor reduction may not preclude the use of P-Ca in climates that experience fall flushes of growth. Even with reductions in growth, canopy infill and linear bearing surface were maintained regardless of treatment rate and combination. P-Ca is therefore not ultimately inhibitory to orchard establishment in NC. P-Ca and ASM reduced dormant pruning efforts, a significant benefit when labor is limited. Crop density increased with the rate of P-Ca, and fruit weight decreased as a result. When ASM was used with P-Ca, hand-thinning efforts were reduced, fruit weight was maintained, and fruit number was reduced relative to use of P-Ca without ASM. With sufficient, early thinning strategies, P-Ca can be used without negatively affecting fruit size. In areas with high vigor and fire blight potential, the use of P-Ca with ASM could be beneficial not only for vigor and disease control but also crop load management purposes.

References Cited

- Aldwinckle H, Robinson T, Norelli J, Momol T, Reddy B. 2000. Controlling shoot blight with 'Apogee'. NY Fruit Q. 8:4.
- Barritt BH, Konishi B, Dilley M. 2008. Performance of four high density apple orchard systems with 'Fuji' and 'Braeburn'. Acta Hortic. 722:389–394. <https://doi.org/10.17660/ActaHortic.2008.722.67>.
- Basak A, Rademacher W. 2000. Growth regulation of pome and stone fruit trees by use of prohexadione calcium. Acta Hortic. 514:41–50. <https://doi.org/10.17660/ActaHortic.2000.514.4>.
- Buban T, Faust M. 1982. Flower bud induction in apple trees: internal control and differentiation. Hortic Rev. 4:174–203. <https://doi.org/10.1002/9781118060773.ch6>.
- Byers RE, Carbaugh DH, Combs LD. 2004. The influence of prohexadione–calcium sprays on apple tree growth, chemical thinning, and return bloom. J Am Pomol Soc. 58:111–117.
- Byers RE, Yoder KS. 1999. Prohexadione–calcium inhibits apple, but not peach, tree growth, but has little influence on apple fruit thinning or quality. HortScience. 34(7):1205–1209. <https://doi.org/10.21273/HORTSCI.34.7.1205>.
- Campbell T, Kalcits L. 2024. Strategies to overcome biennial bearing in apple—A review. Eur J Agron. 158:127213. <https://doi.org/10.1016/j.eja.2024.127213>.
- Carra B, Spagnol D, Abreu ES, Pasa MS, Silva CP, Hellwig CG, Fachinello JC. 2017. Prohexadione calcium reduces vegetative growth and increases fruit set of 'Smith' pear trees, in Southern Brazil. Bragantia. 76(3):360–371. <https://doi.org/10.1590/1678-4499.298>.
- Çetinbaş M, Butar S, Atasay A, İşçi M, Koçak H. 2015. Reduction of apple vegetative shoot growth cv. Starcrimson Delicious/MM 111 with prohexadione calcium application does not decrease fruit quality. J Appl Bot Food Qual. 88(1):259–263. <https://doi.org/10.5073/JABFQ.2015.088.038>.
- Chiou C-S, Jones AL. 1995. Molecular analysis of high-level streptomycin resistance in *Erwinia amylovora*. Phytopathology. 85(3):324–328. <https://doi.org/10.1094/Phyto-85-324>.
- Cooley D. 2007. New England tree-fruit pest management guide. University of Massachusetts, Amherst, in collaboration with the Land Grant Universities of New England and Cornell University. <https://netreefruit.org/apples>. [accessed Jan 2021].
- Costa G, Sabatini E, Spinelli F, Andreotti C, Bomben C, Vizzotto G. 2004. Two years of application of prohexadione-ca on apple: Effect on vegetative and cropping performance, fruit quality, return bloom and residual effect. Acta Hortic. 653:35–40. <https://doi.org/10.17660/ActaHortic.2004.653.3>.
- Cox KD, Wallis A, Carroll J. 2019. Managing fire blight in 2019. Scaffolds Fruits J. 28(6):1–6.
- Crassweller RM, Smith DE. 2014. Influence of rootstock on response of 'Golden Delicious' apple trees to applications of prohexadione calcium. Acta Hortic. 1177:107–110. <https://doi.org/10.17660/ActaHortic.2017.1177.12>.
- Davies PJ. 2004. Plant hormones: Biosynthesis, signal transduction and action (3rd ed). Kluwer Academic Publishers, Dordrecht, The Netherlands, p 775.
- Duyvelshoff C, Cline JA. 2013. Ethephon and prohexadione–calcium influence the flowering, early yield and vegetative growth of young 'Northern Spy' apple trees. Sci Hortic. 151:128–134. <https://doi.org/10.1016/j.scienta.2012.12.002>.
- Elfving DC, Lougheed EC, Cline RA. 1991. Daminozide, root pruning, trunk scoring, and trunk ringing effects on fruit ripening and storage behavior of 'McIntosh' apple. J Am Soc Hortic Sci. 116(2):195–200. <https://doi.org/10.21273/JASHS.116.2.195>.
- Elsysy MA, Hirst PM. 2019. Molecular basis of flower formation in apple caused by defoliation and gibberellins. J Am Soc Hortic Sci. 144(6):414–419. <https://doi.org/10.21273/JASHS04760-19>.
- Embree CG, Myra MTD, Nichols DS, Wright AH. 2007. Effect of blossom density and crop load on growth, fruit quality, and return bloom in 'Honeycrisp' apple. HortScience. 42(7):1622–1625. <https://doi.org/10.21273/HORTSCI.42.7.1622>.
- Evans RR, Evans JR, Rademacher W. 1997. Prohexadione calcium for suppression of vegetative growth in eastern apples. Acta Hortic. 451:663–666. <https://doi.org/10.17660/ActaHortic.1997.451.78>.
- Evans JR, Evans RR, Regusci CL, Rademacher W. 1999. Mode of action, metabolism, and uptake of BAS 125W, prohexadione–calcium. HortScience. 34(7):1200–1201. <https://doi.org/10.21273/HORTSCI.34.7.1200>.
- Faust M. 1989. Physiology of temperate zone fruit trees. John Wiley & Sons, New York, NY, USA.
- Greene DW. 1999. Tree growth management and fruit quality of apple trees treated with prohexadione–calcium (BAS 125). HortScience. 34(7):1209–1212. <https://doi.org/10.21273/HORTSCI.34.7.1209>.
- Greene DW. 2007. The effect of prohexadione–calcium on fruit set and chemical thinning of apple trees. HortScience. 42(6):1361–1365. <https://doi.org/10.21273/HORTSCI.42.6.1361>.
- Greene DW. 2008. The effect of repeat annual applications of prohexadione–calcium on fruit set, return bloom, and fruit size of apples. HortScience. 43(2):376–379. <https://doi.org/10.21273/HORTSCI.43.2.376>.
- Hernandez-Ochoa IM, Asseng S. 2018. Cropping systems and climate change in humid subtropical environments. Agronomy. 8(2):19. <https://doi.org/10.3390/agronomy8020019>.
- Hoad GV. 1978. The role of seed derived hormones in the control of flowering in apple. Acta Hortic. 80:93–104. <https://doi.org/10.17660/ActaHortic.1978.80.14>.
- Imonmion JE, Popoola AR, Afolabi CG, Ganiyu SA, Uzoemeka IP, George J. 2023. Effect of acibenzolar-S-methyl on severity of Fusarium wilt and fruit yield of two tomato cultivars. Nig J Biotechnol. 40(1):86–91. <https://doi.org/10.4314/njb.v40i1.10>.
- Ishii H, Tomita Y, Horio T, Narusaka Y, Nakazawa Y, Nishimura K, Iwamoto S. 1999. Induced resistance of acibenzolar-S-methyl (CGA 245704) to cucumber and Japanese pear diseases. Eur J Plant Pathol. 105:77–85. <https://doi.org/10.1023/A:1008637828624>.
- Javeed K. 2017. Effect of foliar application of salicylic acid and prohexadione–calcium on growth, quality, and storability of apple cv. Red Delicious (PhD Diss). Sher-e-Kashmir University of Agricultural Sciences and Technology, Srinagar, Kashmir, India. <https://krishikosh.egranth.ac.in/handle/1/5810182129>.
- Jones KM, Bound SA, Koen TB, Oakford MJ. 1992. Effect of timing of hand thinning on the cropping potential of 'Red Fuji' apple trees. Aust J Exp Agric. 32(3):417–420. <https://doi.org/10.1071/EA9920417>.
- Kashirskaia N, Kuzin AI, Kochkina AM. 2021. Effect of prohexadione calcium on vegetative growth, yield, and scab development on 'Sinap Orlovskii' apple trees. E3S Web of Conf. 254(3):02011. <https://doi.org/10.1051/e3sconf/202125402011>.
- Kenney JR, Grandmont ME, Mauck KE. 2020. Priming melon defenses with acibenzolar-S-methyl attenuates infections by phylogenetically distinct viruses and diminishes vector preferences for infected hosts. Viruses. 12(3):257. <https://doi.org/10.3390/v12030257>.
- Koike H, Tamai H, Ono T, Shigehara I. 2003. Influence of time of thinning on yield, fruit quality and return flowering of 'Fuji' apple. J Am Pomol Soc. 57(4):169–173.
- Köppen W. 1884. The thermal zones of the Earth according to the duration of hot, moderate and cold periods and of the impact of heat on the organic world (translated and edited by Volken E, Brönnimann S). Meteorol Z. 20:351–360.
- Kunz W, Schurter R, Maetzel T. 1997. The chemistry of benzothiadiazole plant activators. Pestic Sci. 50(4):275–282. [https://doi.org/10.1002/\(SICI\)1096-9063\(199708\)50:4<275::AID-PS593>3.0.CO;2-7](https://doi.org/10.1002/(SICI)1096-9063(199708)50:4<275::AID-PS593>3.0.CO;2-7).
- Liao Q, Ding R, Du T, Kang S, Tong L, Li S. 2022. Stomatal conductance drives variations of yield and water use of maize under water and nitrogen stress. Agric Water Manage. 268:107651. <https://doi.org/10.1016/j.agwat.2022.107651>.
- Lordan J, Francescato P, Dominguez LI, Robinson TL. 2018. Long-term effects of tree density and tree shape on apple orchard performance, a 20-year study—Part 1. Agronomic analysis. Sci Hortic. 238:303–317. <https://doi.org/10.1016/j.scienta.2018.04.033>.
- Mansoor S, Ali Wani O, Lone JK, Manhas S, Kour N, Alam P, Ahmad A, Ahmad P. 2022. Reactive oxygen species in plants: From source to sink. Antioxidants. 11(2):225. <https://doi.org/10.3390/antiox11020225>.
- Maxson-Stein K, He S-Y, Hammerschmidt R, Jones AL. 2002. Effect of treating apple trees with acibenzolar-S-methyl on fire blight and expression of pathogenesis-related protein genes.

- Plant Dis. 86(7):785–790. <https://doi.org/10.1094/PDIS.2002.86.7.785>.
- McArtney S, Palmer JW, Adams HM. 1996. Crop load studies with ‘Royal Gala’ and ‘Braeburn’ apples: Effect of time and level of hand thinning. NZ J Crop Hortic Sci. 24(4):401–407. <https://doi.org/10.1080/01140671.1996.9513977>.
- McGrath MJ, Koczan JM, Kennelly MM, Sundin GW. 2009. Evidence that prohexadione–calcium induces structural resistance to fire blight infection. Phytopathology. 99(5):591–596. <https://doi.org/10.1094/PHYTO-99-5-0591>.
- Miller SS. 2002. Prohexadione–calcium controls vegetative shoot growth in apple. J Tree Fruit Prod. 3(1):11–28. https://doi.org/10.1300/J072v03n01_02.
- Milyaev A, Kofler J, Moya Y, Lempe J, Stefanelli D, Hanke M, Flachowsky H, Wirén N, Wünsche J. 2022. Profiling of phytohormones in apple fruit and buds regarding their role as potential regulators of flower bud formation. Tree Physiol. 42(11):2319–2335. <https://doi.org/10.1093/treephys/tpac083>.
- Monisi M, Saeki T. 2005. On the factor light in plant communities and its importance for matter production. Ann Bot. 95(3):549–567. <http://doi.org/10.1093/aob/mci052>.
- Norelli JL, Miller SS. 2004. Effect of prohexadione–calcium dose level on shoot growth and fire blight in young apple trees. Plant Dis. 88(10):1099–1106. <https://doi.org/10.1094/PDIS.2004.88.10.1099>.
- Owens CL, Stover E. 1999. Vegetative growth and flowering of young apple trees in response to prohexadione–calcium. HortScience. 34(7):1194–1196. <https://doi.org/10.21273/HORTSCI.34.7.1194>.
- Palmer JW, Warrington IJ. 1998. Underlying principles of successful apple planting systems. Acta Hortic. 513:357–366. <https://doi.org/10.17660/ActaHortic.1998.513.42>.
- Pearson JA, Robertson RN. 1953. The physiology of growth in apple fruits: IV. Seasonal variation in cell size, nitrogen metabolism, and respiration in developing Granny Smith apple fruits. Aust J Biol Sci. 6:1–20. <https://doi.org/10.1071/B19530001>.
- Phillips I. 2007. Withdrawal of growth-promoting antibiotics in Europe and its effects in relation to human health. Int J Antimicrob Agents. 30(2):101–107. <https://doi.org/10.1016/j.ijantimicag.2007.02.018>.
- Plavcová L, Mészáros M, Šilhán K, Jupa R. 2022. Relationships between trunk radial growth and fruit yield in apple and pear trees on size-controlling rootstocks. Ann Bot. 130(4):477–489. <https://doi.org/10.1093/aob/mcac089>.
- Privé JP, Cline J, Fava E. 2006. Influence of prohexadione calcium (Apogee®) on shoot growth of non-bearing mature apple trees in two different growing regions. Can J Plant Sci. 86(1):227–233. <https://doi.org/10.4141/P05-031>.
- Rademacher W, Kober R. 2003. Efficient use of prohexadione–Ca in pome fruits. Eur J Hortic Sci. 68:101–107. <https://doi.org/10.1079/ejhs.2003/10271>.
- Rademacher W, Spinelli F, Costa G. 2006. Prohexadione–Ca: Modes of action of a multifunctional plant bioregulator for fruit trees. Acta Hortic. 727:97–106. <https://doi.org/10.17660/ActaHortic.2006.727.10>.
- Reig G, Lordan J, Hoying S, Fargione M, Donahue DJ, Francescato P, Acimovic D, Fazio G, Robinson T. 2020. Long-term performance of ‘Delicious’ apple trees grafted on Geneva rootstocks and trained to four high density systems under New York State climatic conditions. HortScience. 55(10):1538–1550. <https://doi.org/10.21273/HORTSCI14904-20>.
- Robinson TL, Lakso AN. 1991. Bases of yield and production efficiency in apple orchard systems. J Am Soc Hortic Sci. 116(2):188–194. <https://doi.org/10.21273/JASHS.116.2.188>.
- Robinson TL, Hoying S, Reginato GH. 2006. The tall spindle apple production system. NY Fruit Q. 14(2):21–28.
- Robinson TL, Watkins CB. 2003. Cropload of ‘Honeycrisp’ affects not only fruit size but many quality attributes. NY Fruit Q. 3:7–15.
- Sakata N, Ishiga T, Taniguchi S, Ishiga Y. 2020. Acibenzolar–S–methyl activates stomatal-based defense systemically in Japanese radish. Front Plant Sci. 11:565745. <https://doi.org/10.3389/fpls.2020.565745>.
- Schupp JR. 2003. Effects of chemical thinners on fruit set, yield, fruit size, and quality of ‘Honeycrisp’ apple. NY Fruit Q. 3:3–5.
- Serra S, Leisso R, Giordani L, Kalcsits L, Musacchi S. 2016. Crop load influences fruit quality, nutritional balance, and return bloom in ‘Honeycrisp’ apple. HortScience. 51(3):236–244. <https://doi.org/10.21273/HORTSCI.51.3.236>.
- Sparks B. 2001. Fire blights destructive path. Am Fruit Grower. 121(3):11–16.
- Suhayda CG, Goodman RN. 1981. Early proliferation and migration and subsequent xylem occlusion by *Erwinia amylovora* and the fate of its extracellular polysaccharide (EPS) in apple shoots. Phytopathology. 71(7):697–707. <https://doi.org/10.1094/Phyto-71-697>.
- Sundin GW, Werner NA, Yoder KS, Aldwinckle HS. 2009. Field evaluation of biological control of fire blight in the eastern United States. Plant Dis. 93(4):386–394. <https://doi.org/10.1094/PDIS-93-4-0386>.
- Tromp J. 2000. Lower-bud formation in pome fruits as affected by fruit thinning. Plant Growth Regul. 31(1/2):27–34. <https://doi.org/10.1023/A:1006342328724>.
- Unrath CR. 1999. Prohexadione–Ca: A promising chemical for controlling vegetative growth of apples. HortScience. 34(7):1197–1200. <https://doi.org/10.21273/HORTSCI.34.7.1197>.
- van der Zwet T, Orolaza-Halbrecht N, Zeller W. 2012. Fire blight: History, biology, and management. American Phytopathological Society, St. Paul, MN, USA. <https://doi.org/10.1094/9780890544839.fm>.
- Vogel AR, Villani SM, Kon TM. 2025. Prohexadione calcium and acibenzolar–S–methyl effects and interactions II: Fire blight management in a young North Carolina apple orchard. HortScience. <https://doi.org/10.21273/HORTSCI18806-25>.
- Wallis AE, Cox KD. 2020. Management of fire blight using pre-bloom application of prohexadione–calcium. Plant Dis. 104(4):1048–1054. <https://doi.org/10.1094/PDIS-09-19-1948-RE>.
- Wallis A, Miranda-Sazo M, Cox K. 2021. Assessing and minimizing the development and spread of fire blight following mechanical thinning and pruning in apple orchards. Plant Dis. 105(3):650–659. <https://doi.org/10.1094/PDIS-06-20-1324-RE>.
- Wright AH, Embree CG, Nichols DS, Prange RK, Harrison PA, Delong JM. 2006. Fruit mass, colour and yield of ‘Honeycrisp’™ apples are influenced by manually-adjusted fruit population and tree form. J Hortic Sci Biotechnol. 81(3):397–401. <https://doi.org/10.1080/14620316.2006.11512079>.
- Yoder KS, Miller SS, Byers RE. 1999. Suppression of fire blight in apple shoots by prohexadione–calcium following experimental and natural inoculation. HortScience. 34(7):1202–1204. <https://doi.org/10.21273/HORTSCI.34.7.1202>.