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Naphthaleneacetic Acid: A Key Tool for Reducing Preharvest Fruit Drop and Sustaining Apple Red Coloration

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Preharvest fruit drop (PFD) causes significant yield losses in 'Honeycrisp' apples. This study evaluated naphthaleneacetic acid (NAA), aminoethoxyvinylglycine (AVG), and their combined application for PFD management, assessing fruit retention and quality for two growing seasons. All treatments generally reduced PFD compared to controls, with AVG, commercially known as "ReTain" (Valent BioSciences LLC), often showing the greatest reduction. However, ReTain consistently impaired red fruit color development. In contrast, NAA, commercially available as "Refine" (Fine Americas, Inc.), effectively controlled PFD without negatively impacting color or firmness, and it improved fruit sweetness. These findings highlight that while both can reduce drop, Refine provides a better balance for 'Honeycrisp' growers by mitigating PFD while preserving or enhancing key quality attributes like color and sweetness compared to ReTain.

PFD in apple (*Malus* × *domestica* Borkh.) represents a significant challenge to apple production worldwide, characterized by the premature abscission of fruits from the tree before they attain horticultural maturity. Horticultural maturity is defined as the stage of optimal development for the fruit's intended use, whether it's for fresh consumption or processing (Tipu et al., 2025).

The physiological basis of PFD involves the process of abscission, wherein a specialized layer of cells in the pedicel — the stalk connecting the fruit to the tree — undergoes separation, ultimately causing the fruit to detach. This phenomenon typically initiates approximately four weeks prior to the anticipated harvest period, leading to substantial yield reductions in many apple cultivars. Depending on the cultivar and the growing season, yield losses attributed to PFD can range from 20% to more than 50%, directly impacting the profitability of apple orchards (Arseneault and Cline 2018).

Certain commercially important cultivars such as 'Honeycrisp' and 'McIntosh' are particularly susceptible to PFD, making the management of this issue critical for their successful cultivation. These losses not only diminish the overall marketable yield, but they also present challenges in terms of harvest timing and labor management because growers may need to harvest the remaining crop within a compressed time frame to avoid further losses.

Preharvest fruit drop in apples is influenced by a combination of internal and external factors, with plant hormones playing a central role. As apples ripen, there is a natural increase in phytohormones levels, particularly ethylene, which triggers fruit softening and the development of the abscission zone. However, the balance between ethylene and other plant hormones, particularly auxin, is critical in determining the extent of fruit drop (Schaffer et al. 2013). High auxin levels in early fruit development generally suppress the activity of cell-wall-degrading enzymes (Trainotti et al. 2007). As fruits approach maturity, a natural decline in auxin levels relieves this suppression and promotes cell separation and fruit drop (Tipu and Sherif 2024). Therefore, maintaining adequate auxin levels near the fruit-pedicel junction is essential for minimizing PFD.

To mitigate PFD, horticultural practices often involve the application of synthetic auxins, such as Refine. These exogenous auxins supplement endogenous auxin levels, thereby delaying abscission. Another important plant growth regulator employed for PFD management is ReTain, which functions by inhibiting the synthesis of

ethylene rather than interacting directly with auxin (Liu et al. 2022). In several cases, ReTain has demonstrated greater effectiveness than Refine in delaying fruit drop, especially in cultivars like 'Galaxy' with inherently high ethylene production (Schultz et al. 2023). However, ReTain has been associated with reduced fruit coloration in apples (Greene and Schupp 2004; Whale et al. 2008; Liu et al. 2022; Tipu et al., 2025).

This publication highlights the effectiveness of Refine in reducing PFD and red color development in 'Honeycrisp' apples. We explored the application of Refine and ReTain over two consecutive seasons, focusing on their effects on fruit retention and color.

Experimental Methods

Plant Material and Treatments

The study was conducted in 2023 and 2024 at the Alson H. Smith Jr. Agricultural Research and Extension Center in Winchester, Virginia, using 8-year-old 'Honeycrisp' apple trees grafted onto Budagovsky 9 (B9) rootstock. The experimental design followed a completely randomized design, with six randomly assigned adjacent trees per treatment. Of these, three trees per treatment were used for PFD analysis, and the other three were used for biochemical evaluations. Two untreated buffer trees were positioned between trees being treated to prevent cross-contamination.

Preharvest fruit drop and fruit coloration were assessed through three treatments — Refine (NAA, 59 ml/acre), ReTain (AVG, 333 g/acre), and ReTain + Refine — along with an untreated control. Treatments were applied at three intervals: three, two, and one week before the estimated harvest date, except for ReTain treatment, which was applied once at three weeks before the estimated harvest date. Each tree received 2.53 liters of spray solution containing a nonionic surfactant, Regulaid (946 ml/acre), using a utility-vehicle-mounted tank sprayer. All trees, including treated and untreated controls, were subjected to the same orchard and disease management practices.

Assessment of Chlorophyll Degradation and Color Intensity

Fruit background color was assessed on both blushed and unblushed sides using a delta absorbance meter, which measures the index of absorbance difference, indicating chlorophyll levels in the peel. Fruit color intensity was further measured using an NH310 Colorimeter, which evaluated 10 fruits per replicate. Measurements were based on the CIE 1976 color space system using L*, a*, and b* values (McGuire 1992). Lightness (L*) ranges from 0 (black) to 100 (white), a* represents red-green, and b* represents yellow-blue. Hue angle (h°) was calculated to define color nuance, with 0° indicating red-purple, 90° indicating yellow, and 180° indicating bluish-green.

Fruit Drop Monitoring

Approximately 100 fruits per tree were tagged on both sides of the trees about four to five weeks before harvest. Initial counts were recorded at four weeks before estimated harvest date, with weekly monitoring continuing through two weeks after harvest. Fruit drop percentage was calculated using the formula:

$$\text{Fruit drop (\%)} = ((Fi - Fr)/Fi) \times 100$$

where Fi is the initial fruit count, and Fr is the number of fruits retained on the tree at each time point.

Anthocyanin Quantification

Fruit peel tissues from three to five fruits per treatment were collected 24 hours after each application, immediately frozen in liquid nitrogen, and stored at minus 80 C. Anthocyanin concentrations were quantified using a pH differential method with a commercial assay kit (Cosmo Bio, Carlsbad, California) that calculated cyanidin-3-glucoside equivalents based on absorbance at 510 nanometer (nm) and 700 nm.

Fruit Quality Assessment

Ten fruits per replicate were used to measure fruit weight, diameter, flesh firmness, soluble solids, and starch index. Fruit weight, diameter, and firmness were recorded using a fruit texture analyzer on peeled sides of each fruit. Soluble solid contents were measured as brix (%) with a digital refractometer using juice droplets. The starch index was scored using iodine staining, following the Cornell starch-iodine index system (Blanpied and Silsby 1992), with a 1 to 6 scale in 2023 and a 1 to 8 scale in 2024.

Results and Discussion

Effects of Plant Growth Regulators on Reduction of Fruit Drop

Plant growth regulator (PGR) treatments significantly affected the percentage of preharvest fruit drop across all assessment periods in both 2023 and 2024 (fig. 1). In 2023, trees treated with Refine exhibited significantly lower fruit drop at harvest (19.5%) and two weeks after

harvest (27.0%) compared to the untreated control. Notably, in 2023, the combined application of ReTain + Refine did not result in a statistically significant reduction in fruit drop compared to the untreated control at any assessment point. In 2024, all PGR applications — ReTain, Refine, and the ReTain + Refine combination — resulted in significantly less fruit drop than the untreated control at both harvest and two weeks after harvest date. Within the PGR treatments in 2024, ReTain application resulted in the lowest fruit drop percentage, significantly outperforming both Refine alone and the ReTain + Refine combination. There was no significant difference in fruit drop between the Refine and ReTain + Refine treatments at either time point.

These findings align with previous studies demonstrating significant PFD reduction by both Retain and Refine, with ReTain often showing greater efficacy (Greene and Schupp 2004; Arseneault and Cline 2018). The reduction in PFD associated with ReTain appears to be attributed to its inhibition of ethylene biosynthesis, thereby delaying ripening processes (Tipu et al., 2025), while Refine is understood to reduce fruit drop by mitigating cell-wall modifications within the abscission zone (Li and Yuan 2008).

Effects of Plant Growth Regulators on Changes in Fruit Color

Despite its effectiveness in reducing preharvest fruit drop (fig. 1), ReTain application consistently impaired apple peel color development in both the 2023 and 2024 growing seasons compared to the untreated control (fig. 2). This was reflected in chlorophyll degradation rates, measured using the index of absorbance difference (I_{AD}), which varied significantly among treatments at harvest and at two weeks after harvest in both years (fig. 3). Except at the 2023 harvest, ReTain-treated fruits consistently exhibited the highest I_{AD} values (indicating slower chlorophyll degradation) compared to the control. The combined ReTain + Refine treatment generally resulted in the second-highest I_{AD} values (fig. 3). These I_{AD} results suggest that ReTain delays ripening-associated chlorophyll breakdown, and this effect was not counteracted by the application of Refine in the combined treatment.

Conversely, treatment with Refine alone did not negatively impact visual fruit color compared to the control in either season (fig. 2), showing statistically similar I_{AD} values compared to the untreated control, with the exception of two weeks after harvest date in 2023 (fig. 3). This nonsignificant impact on color from Refine application aligns with previous reports (Arseneault and Cline 2018).

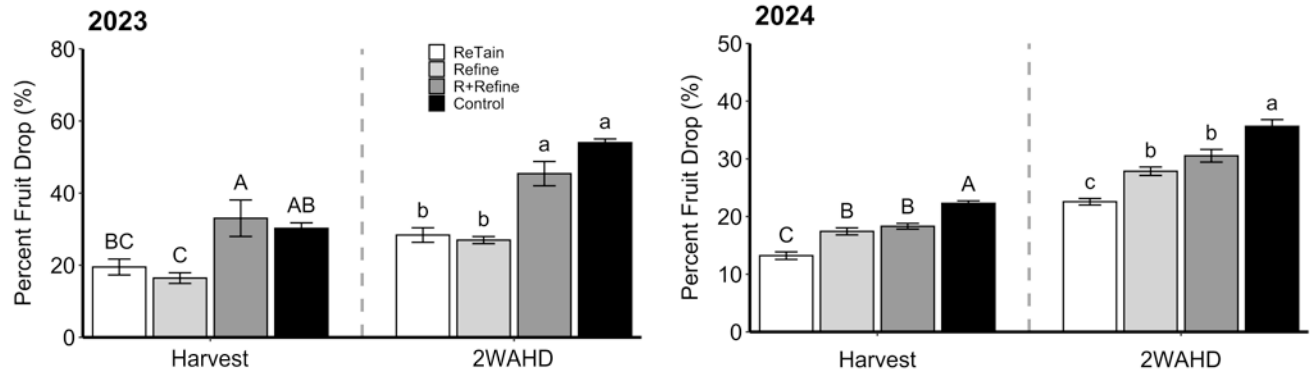

Figure 1. Percentage of fruit drop of 'Honeycrisp' apple treated with ReTain, Refine, ReTain + Refine and an untreated control at harvest (left) and two weeks after harvest date (right) in 2023 (top) and 2024 (bottom). Letters indicate significant differences among treatments at $p \leq 0.05$, using analysis of variance. Uppercase letters denote comparisons among treatments at harvest, while lowercase letters indicate differences among treatments at two weeks after harvest date.

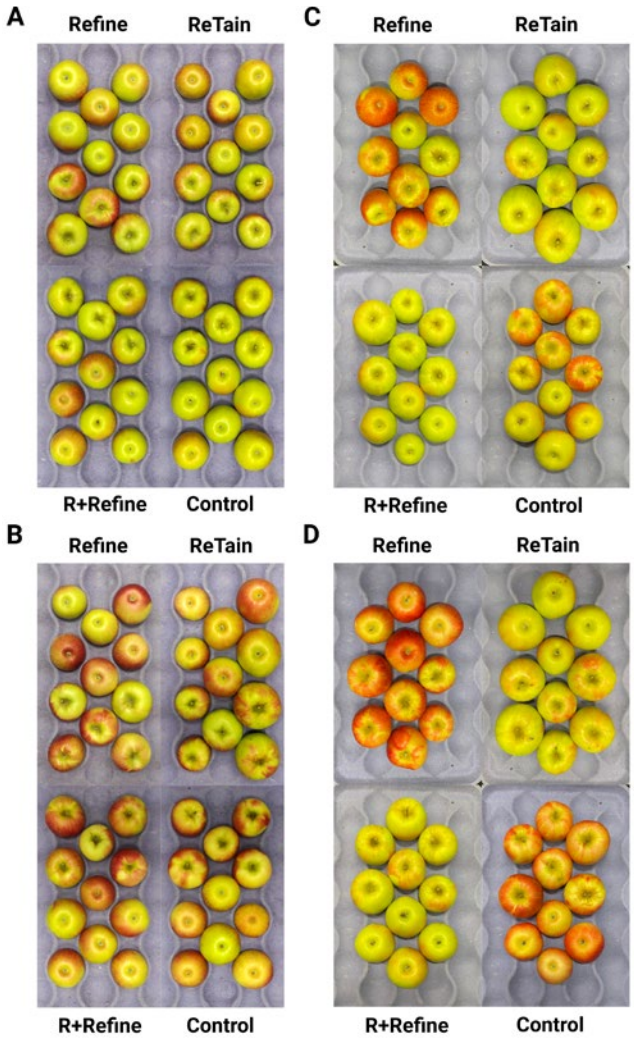

Figure 2. Effect of treatments on 'Honeycrisp' apple coloration. Images of fruits were taken at harvest (top) and two weeks after the harvest date (bottom) in 2023 (first column) and in 2024 (second column).

In agreement with the results for chlorophyll breakdown, color intensity measurements based on hue angle values of apple peel tissues also revealed significant treatment effects (fig. 3). Specifically, both ReTain and the ReTain + Refine combination resulted in significantly higher hue angle values at harvest and two weeks after the harvest date compared to the control, indicative of greener fruit (fig. 3).

In contrast, Refine treatment alone generally resulted in lower hue angle values compared to both ReTain and the ReTain + Refine combination, suggesting enhanced red color development (fig. 3). Although the hue angle value for Refine-treated fruit was the lowest at harvest, it was statistically identical with untreated control. On the other hand, at two weeks after harvest date, the hue angle value for Refine-treated fruit was significantly higher than the control. However, this value still corresponded to a red hue within the CIE 1976 color space (McGuire 1992).

Analysis of anthocyanin content (measured as nanogram per liter; ng/L), a key pigment contributing to red skin color, revealed significant variations emerged by one week before harvest date, as fruits approached maturity (fig. 3). At one week before harvest, the Refine treatment resulted in the highest anthocyanin concentration (199.5 ng/L), which was nearly five times greater than that of the untreated control (fig. 3). In contrast, anthocyanin levels in fruits treated with ReTain or ReTain + Refine were not significantly different from the control, indicating that these treatments suppressed anthocyanin accumulation relative to Refine alone. No significant differences among treatments were observed at two or three weeks before harvest (fig. 3).

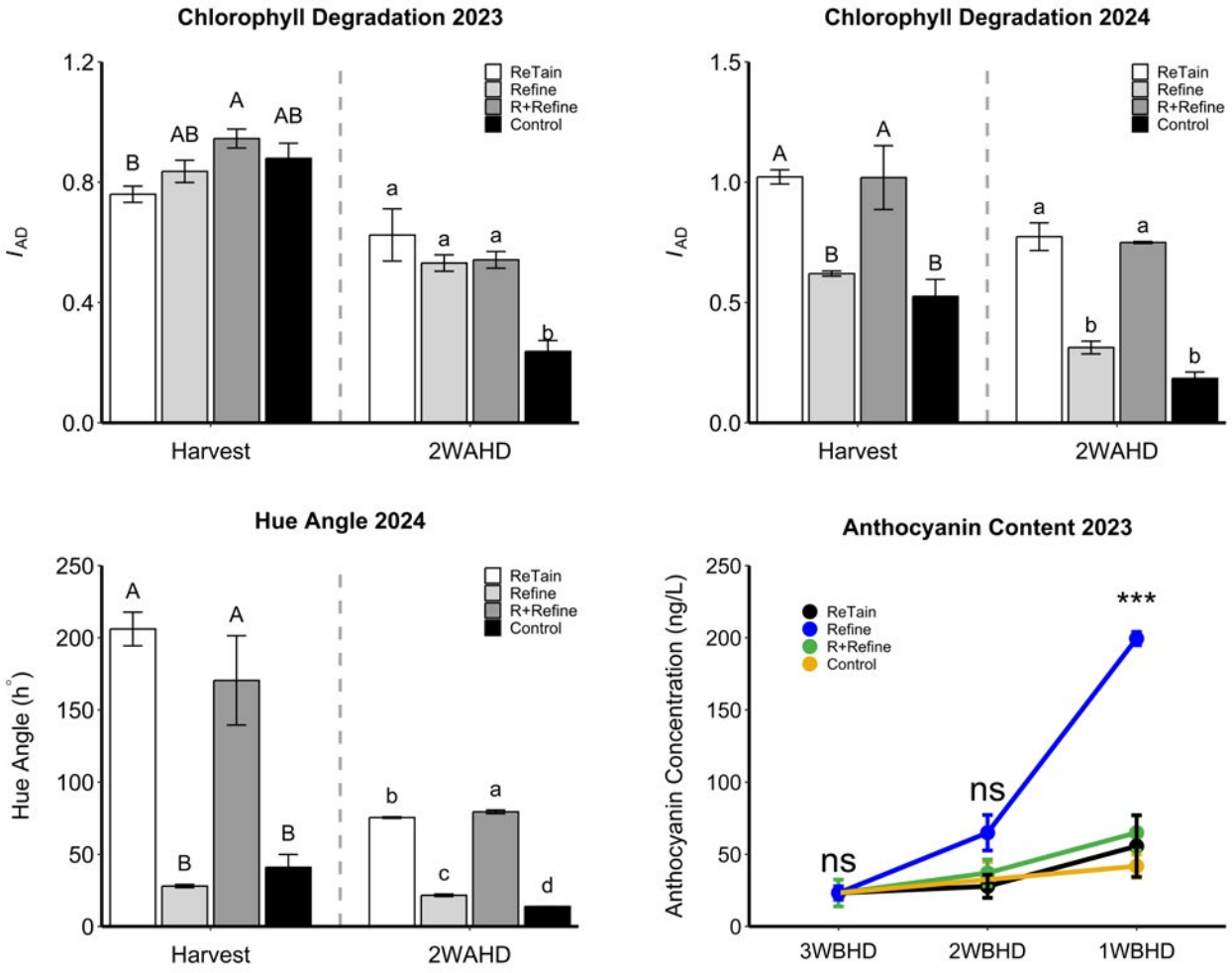

Figure 3. Effect of ReTain, Refine, and ReTain (R) + Refine treatments on chlorophyll degradation, color intensity, and anthocyanin contents of 'Honeycrisp' apple. Chlorophyll degradation was measured at harvest and two weeks after harvest date in 2023 (top-left) and 2024 (top-right). Color intensity was measured at harvest and two weeks after harvest date in 2024 (bottom-left). Anthocyanin contents were measured at three weeks, two weeks, and one week before harvest date in 2023 (bottom-right). Uppercase letters denote comparisons among treatments at harvest, while lowercase letters indicate differences among treatments at two weeks after harvest date, and asterisks represent significant differences among treatments at $p \leq 0.05$, using analysis of variance.

Effects of Plant Growth Regulators on Fruit Quality

The fruit quality parameters such as fruit firmness (measured as pound force; lbf), diameter (millimeter; mm), weight (gram; g), brix (percentage; %), and starch index showed differential variations at harvest and two weeks after harvest date in response to PGR treatments in both growing seasons (tables 1 and 2). As mentioned, Refine treatment reduced fruit drop without any negative effect on fruit color; therefore, we will primarily focus on the effect of Refine on fruit quality in this section.

Table 1a. Effects of different treatments on fruit quality parameters on 'Honeycrisp'/B9 apples at harvest in 2023.

Treatment	Firmness (lbf)	Diameter (mm)	Weight (g)	Brix (%)	Starch (1-6)
ReTain	17.02 ab	68.70 ab	117.93 b	12.25 a	5.60 a
Refine	16.23 b	70.67 a	133.53 ab	11.69 ab	5.67 a
ReTain + Refine	17.19 a	67.73 b	121.80 ab	12.23 a	4.67 b
Control	16.35 ab	69.93 ab	136.80 a	10.76 b	4.67 b

Different letters within a column indicate significant differences at P ≤ 0.05.

Table 1b. Effects of different treatments on fruit quality parameters on 'Honeycrisp'/B9 apples at two weeks after harvest date in 2023.

Treatment	Firmness (lbf)	Diameter (mm)	Weight (g)	Brix (%)	Starch (1-6)
ReTain	15.68 a	73.37 a	162.13 a	12.13 ab	5.20 b
Refine	16.70 a	69.60 a	130.33 b	12.62 a	5.67 ab
ReTain + Refine	16.30 a	70.13 a	137.53 ab	11.07 c	5.67 ab
Control	16.00 a	69.30 a	133.33 ab	11.39 bc	5.87 a

Different letters within a column indicate significant differences at P ≤ 0.05.

Table 2a. Effects of different treatments on fruit quality parameters on 'Honeycrisp'/B9 apples at harvest in 2024.

Treatment	Firmness (lbf)	Diameter (mm)	Weight (g)	Brix (%)	Starch (1-8)
ReTain	18.39 a	52.20 a	202.37 a	10.81 b	3.13 b
Refine	17.70 a	49.90 a	169.80 a	12.85 a	6.33 a
ReTain + Refine	18.66 a	48.52 a	150.83 a	11.25 b	3.83 b
Control	17.63 a	48.78 a	155.17 a	12.80 a	6.93 a

Different letters within a column indicate significant differences at P ≤ 0.05.

Table 2b. Effects of different treatments on fruit quality parameters on 'Honeycrisp'/B9 apples at two weeks after harvest date in 2024.

Treatment	Firmness (lbf)	Diameter (mm)	Weight (g)	Brix (%)	Starch (1-8)
ReTain	15.67 a	53.25 a	208.90 a	11.43 c	5.40 b
Refine	15.35 a	50.90 b	181.87 ab	12.72 ab	7.43 a
ReTain + Refine	16.83 a	49.55 b	165.53 b	11.72 bc	5.07 b
Control	15.96 a	49.43 b	164.40 b	13.30 a	7.67 a

Different letters within a column indicate significant differences at P ≤ 0.05.

Refine application did not significantly affect fruit firmness or fruit size across both growing seasons compared to the untreated control, indicating it had no adverse effects on these physical attributes of the apple fruits (tables 1 and 2). As an added benefit, Refine positively influenced soluble sugar content, resulting in higher brix values compared to untreated fruits at harvest and two weeks after harvest date in 2023 and at harvest in 2024, which suggests improved sweetness of apple fruits. The starch breakdown as a result of applying Refine was not statistically different from the control at two weeks after harvest date in 2023, at harvest, and two weeks after harvest date in 2024. However, Refine promoted starch degradation with the significantly highest starch index observed only at

harvest in 2023 compared to the untreated control (table 1). These results suggest that Refine may maintain fruit maturity and improve the sweetness of apple fruits while maintaining other desirable quality traits.

Conclusion

Overall, this study demonstrates that while both ReTain and Refine significantly reduced preharvest fruit drop, their effects on fruit quality differed in 'Honeycrisp' apples. Refine effectively decreased preharvest fruit drop without compromising fruit color or firmness and even enhanced fruit sweetness. However, the reduction in PFD achieved with ReTain was slightly more consistent and pronounced than that observed with Refine.

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