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Title: Evaluate ethephon and 1-aminocyclopropane-1-carboxylic acid (ACC) as ripening aids for blueberry

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Objective: Effect of ethephon and 1-aminocyclopropane-1-carboxylic acid (ACC) on fruit ripening and postharvest fruit quality

Justification and Description:

Blueberry has emerged as a major specialty crop in Georgia over the last decade. It has a farm gate value of over $226 million (2017; Farm gate value report) and is currently cultivated in over 30,000 acres across the state. The two main types of cultivated blueberry in GA are the southern highbush blueberry and rabbiteye blueberry. In both types, during ripening, individual fruit on the branch mature at different rates resulting in a non-uniform ripening period extending over 2-3 weeks (Suzuki et al., 1997), thus requring multiple harvests for each varitey throughout the season. Harvesting is the most labor intensive and expensive aspects of blueberry production, thus harvesting multiple times greatly increases the total cost of production.

We conducted grower surveys in January 2019 at the Annual Blueberry Meeting in Alma, GA and the Southeast Fruit and Vegetable Conference, Savannah, GA where 43 growers participated. This survey indicated that, 37% of the growers spend the highest on manual/machine harvesting among 5 production/management-related costs. In addition, 86% of blueberry growers placed high to moderate value on reducing harvest frequency. Further, growers indicated that ripening aids will help with early blueberry harvests and reduce the number of harvests. *Hence, accelerating ripening of blueberries and ensuring a uniform ripening period by using a ripening aid can potentially reduce production costs and increase profitability thereby increasing the competitiveness of GA and southeastern blueberry production.* Currently there are no well validated ripening aid recommendations for southern highbush and rabbiteye blueberry production in the southeast.

Ethephon is a plant growth regulator (PGR) that releases ethylene after absorption by plant cells. Previous studies have shown that application of ethephon can accelerate ripening in blueberries, but the effects were not consistent across different cultivars (Ban et al., 2007; Dekazos, 1976; Eck, 1970; Wang et al, 2018). Ethephon is approved for applications to concentrate maturity and to obtain early fruit color in blueberries mainly in northern highbush blueberries (US-EPA, 2010). The recommended application rates are between 500-1500 ppm. However, at rates greater than 500 ppm ethephon can have negative effects on fruit quality and may induce fruit drop (Malladi et al., 2012). Currently no recommendations of ethephon as a ripening aid are available for southern highbush and rabbiteye blueberries. However, in some cases, ethephon applications can have inconsistent effects since the rate of evolution of ethylene from ethephon is temperature and pH dependent. Higher temperatures and higher pH of spray solutions increase the rate of ethylene evolution significantly leading to phytotoxic effects on foliage such as leaf yellowing and senescence (Lougheed and Franklin, 1972; Aiken et al, 2015). Our preliminary data demonstrate that ethephon applied at 250 ppm can accelerate ripening without any significant negative effects (Wang et al., 2018), however several follow-up studies need to be performed (described below).

Another potential ripening aid, 1-aminocyclopropane-1-carboxylic acid (ACC), is a precursor of ethylene. ACC has to enter the cell and get converted to ethylene via the action of an enzyme, ACC oxidase, which is not rate-limiting. Thus, slight changes in temperature may not interfere with its activity. Further, as the enzyme may be relatively more abundant in ripening fruit, it is likely that it may allow for selective maturation of the right stages. Therefore, in comparison to ethephon, application of ACC may allow for a more controlled release rate (in communication with Valent Biosciences.) with similar ripening effects (shown by preliminary data below). ACC may take a few years for approval and registration (communication with Valent Biosciences). Thus, there could be potential advantages to developing recommendations for both ethephon and ACC as ripening agents. In this proposal, we plan to evaluate the effect of ethephon and ACC on ripening and evaluate their potential as ripening aids for southern highbush and rabbiteye blueberry.

Preliminary data: In 2019, we demonstrated that both ethephon and ACC applied at 250 ppm promote fruit ripening by increasing the proportion of ripe fruit in 'Miss Lilly' a southern highbush blueberry cultivar, and in 'Premier', a rabbiteye blueberry cultivar. Fruit quality at harvest was not altered in 'Miss Lilly'. In 'Premier', ACC reduced total soluble solids (TSS) content in comparison to the control. These preliminary data support the effect of ethephon and ACC on fruit ripening, but the effects on fruit quality were not consistent. This was probably because the timing of application was different; in Miss Lilly ~20% of the fruit were ripe, whereas in 'Premier' ~50% of the fruit were ripe. In addition, these studies were performed only in one year using a few cultivars. Thus, one more year of study was warranted.

Experimental plan: The proposed research has two objectives

Objective 1: To determine the effect of ethephon and ACC applications on the ripening of two rabbiteye blueberry cultivars

Ethephon and ACC applications were performed on two rabbiteye cultivars 'Powderblue' and 'Premier' in summer 2020. The two rabbiteye cultivars used for this study was located at the Durham Horticulture farm at University of Georgia, Athens. We could not perform the application on the two southern highbush cultivars 'Suziblue' and 'Miss Lilly' due to COVID-19 travel restrictions. The two southern highbush cultivars would have required travel to the University of Georgia Blueberry Research Farm near Alapaha, GA. Four, single plant replications per treatment was used in a completely randomized design for this study. Ethephon and ACC was applied at the rate of 250 ppm using a non-ionic surfactant (0.15% Latron B-1956). The control was treated with water and the surfactant. Foliar applications of PGRs was performed when around 20-30% of fruit on the plant were ripe. At the time of application, all ripe and immature fruit were removed and several shoots with around 50-100 fruit each was tagged. After applications, the proportion of green, pink and ripe fruit was determined at 3-5 day intervals for up to two weeks to determine the rate of progression of ripening on three of the tagged shoots. The remaining tagged shoots was used for measurements described in Objective 2.

Objective 2: To determine effects of ethephon and ACC on postharvest fruit quality

For fruit quality analyses, shoots tagged as described previously were used to obtain around 300 ripe fruit from each replicate. Fruit were harvested at 10 DAT in Premier and Powderblue after treatment. Ripe fruit harvested after treatment were randomly split into three groups 1. Postharvest (PH)+3 days; 2. PH+10 days and 3. PH+21 days. Fruit firmness measurements was performed using a Fruit Texture Analyzer (Model GS-15, Güss Manufacturing (Pty) Ltd., Strand, South Africa). For measuring TA and TSS, juice from approximately 20 g of fruit was extracted using a blender and centrifuged using a bench top micro-centrifuge. The supernatant was used to determine TSS using a digital handheld refractometer (Atago USA, Bellevue, WA). To determine TA, the supernatant was titrated using an automatic mini titrator (Hanna Instruments, Woonsocket, RI). Fruit weight was measured from 20 fruit/replicate and averaged to calculate weight/fruit. Data analyses was performed using analysis of variance (ANOVA; = 0.05) in the R package and means separated using Tukey's honestly significant difference (HSD).

RESULTS:

Effect of 250 ppm Ethephon and 1-aminocyclopropane-1-carboxylic acid (ACC) on ripening rate

In 2020, ripening rate results were similar to 2019. Ethephon increased the ripening rate in Premier and Powderblue compared to control, with the exception of Premier at 3 DAT (Figure 1). Overall, ACC increased the rate of ripening in both rabbiteye cultivars. The ripening rate after ethephon and ACC application were the same in Premier, at 7 and 10 DAT; however, in Powderblue, ethephon had a more dramatic effect on ripening rate compared to ACC. At 10 DAT, ethephon had 40% and 32% more ripe fruit in Powderblue and Premier, respectively. ACC had 32% and 23% more ripe fruits in Powderblue and Premier respectively at 10 DAT, respectively.

Effect of 250 ppm Ethephon and 1-aminocyclopropane-1-carboxylic acid (ACC) on postharvest fruit quality

Effect of ethephon and ACC treatement to postharvest fruit quality were inconsistent (Tables 1 and 2). Fruit texture as measured by compression was higher under ethephon treatment by 19% at PH21 in Powderblue. Similarly, ethephon and ACC increased, another fruit texture parameter, skin toughness as measured by puncture at PH3 by 16.7% and 8.3%, respectively, compared to control (Table 1). Based on fruit weight measurements ethephon decreased fruit weight compared to control at PH3 in Powderblue and at all the three time-points in Premier. This trend was not observed with ACC. Only in Powderblue, ethephon decreased the percentage of defect fruit at PH21 compared to control. Higher TSS content was observed in the control treatments at PH3 and PH21 in Powderblue; however, no significantly different in Premier, thus trends were not consistent. Overall no significant trends were noted in titratable acidity between ethephon, ACC, and control (Table 2).

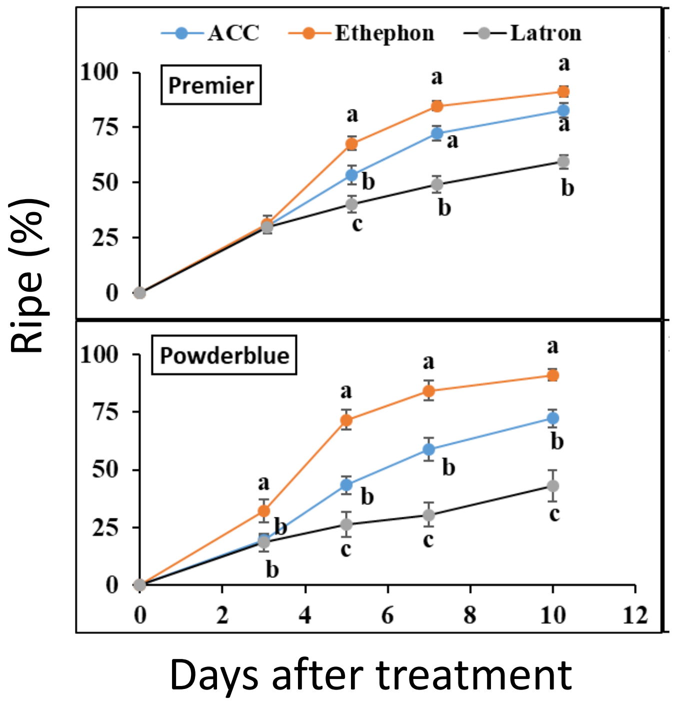

Figure 1: Effect of pre-harvest treatment of Latron, ACC, Ethephon, Water, on blueberry ripening in 2020.

Table 1: Effect of pre-harvest treatment of ACC, Ethephon, and Latron on blueberry compression, puncture, and fruit weight in 2020.

	Compression			Puncture			Fruit weight (g)		
Treatment/Variety	PH3	PH10	PH21	PH3	PH10	PH21	PH3	PH10	PH21
Powderblue									
ACC	0.22	0.21	0.22 b	0.26 b	0.27	0.27	0.94 ab	0.85	0.90
Ethephon	0.24	0.23	0.25 a	0.28 a	0.28	0.28	0.90 b	0.80	0.84
Latron	0.23	0.21	0.21 b	0.24 c	0.24	0.25	1.02 a	0.97	1.00
P-value	0.4970	0.1540	0.0221	0.0050	0.1090	0.5730	0.0469	0.0956	0.0674
Premier									
ACC	0.24	0.25	0.22	0.19	0.19	0.20	1.74 a	1.63 a	1.63 a
Ethephon	0.26	0.27	0.25	0.22	0.24	0.22	1.41 b	1.33 b	1.31 b
Latron	0.24	0.25	0.20	0.19	0.22	0.21	1.87 a	1.66 a	1.74 a
P-value	0.0750	0.2490	0.0649	0.0546	0.0749	0.0649	0.0003	0.0496	0.008

Table 2: Effect of pre-harvest treatment of ACC, Ethephon, and Latron on blueberry defect (%), total soluble solid content (%), and titratable acidity (TA) in 2020.

	Defect (%)			TSS (%)			TA (%)		
Treatment/Variety	PH3	PH10	PH21	PH3	PH10	PH21	PH3	PH10	PH21
Powderblue									
ACC	2.50	1.25	8.75 a	15.15 b	15.93	15.05	0.55	0.53	0.53
Ethephon	1.25	0.00	3.75 b	15.75 b	15.10	14.95	0.58	0.59	0.55
Control	0.00	3.75	12.5 a	17.18 a	16.25	16.73	0.51	0.49	0.48
P-value	0.244	0.0723	0.012	0.0054	0.447	0.0012	0.164	0.113	0.0563
Premier									
ACC	3.75	13.75	18.75	12.95	12.63	12.15	0.48	0.40	0.34
Ethephon	3.75	7.5	13.75	12.28	12.15	11.88	0.51	0.47	0.39
Control	5	6.25	20	13.48	12.70	12.30	0.48	0.43	0.35
P-value	0.863	0.148	0.617	0.15	0.637	0.654	0.808	0.236	0.068

Conclusions:

Both PGRs, ACC and ethephon can accelerate ripening, minimally affecting postharvest fruit quality and has a strong potential to be developed as ripening aids for the blueberry industry.

References:

Aiken MG et al. (2015) Substrate pH impacts efficacy of ethephon drenches on growth of herbaceous perennials. HortScience 50:1187–1191

Ban T et al. (2007). Effect of ethephon (2-chloroethylphosphonic acid) on the fruit ripening characters of rabbiteye blueberry. Sci. Hortic. (Amsterdam) 112:278–281

Dekazos ED (1976) Effects of preharvest applications of ethephon and SADH on ripening, firmness and storage quality of rabbiteye blueberries (cv 'T-19'). Proc. Fla. State Hort. Soc. 89:266–270

Eck P (1970) Influence of ethrel upon highbush blueberry fruit ripening. HortScience 5:23–25

El-Agamy S et al (1982) Fruit maturity as related to ethylene in "Delite' blueberry. Proc. Florida State Hortic. Soc. 95:245–246.

Farm gate value report (2017) https://www.caes.uga.edu/content/dam/caes-subsite/caed/publications/annual-reports-farm-gate-value-reports/2017-farm-gate-value-report.pdf

Frenkel C (1972) Involvement of peroxidase and indole-3-acetic acid oxidase isozymes from pear, tomato, and blueberry fruit in ripening. Plant Physiol. 49:757–763.

Lougheed EC and Franklin EW (1972) Effects of temperature on ethylene evolution from ethephon. *Can. J. Plant Sci.* 52:769-773

Malladi A et al. (2012) Ethephon and methyl jasmonate affect fruit detachment in rabbiteye and southern highbush blueberry. HortScience. 47: 1745-1749.

Suzuki A et al. (1997) Changes of ethylene activity evolution, ACC content, in fruits ethylene forming blueberry enzyme and respiration of highbush blueberry. J. Japan Soc. Hort. Sci. 66:23–27

USDA-EPA, (2010) https://www3.epa.gov/pesticides/chem_search/ppls/000264-00267-20100629.pdf

Wang Y-W et al. (2018) The effect of ethephon, abscisic acid, and methyl jasmonate on fruit ripening in rabbiteye blueberry (*Vaccinium virgatum*). Horticulturae 4:24