Title: Refining the use of fruit abscission agents in muscadine grapes

Progress Report

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Research Proposal

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Objectives: 1) To evaluate the effect of Ethephon dosage and the time of day of application on its efficacy as a fruit abscission agent. 2) To evaluate the effect of methyl jasmonate (MeJA) on muscadine grape fruit detachment.

Justification

Muscadine grapes are among the most commonly grown grape types in Georgia. Muscadine grapes are produced in relatively small bunches with uneven berry numbers and ripeness, are harvested individually, and usually sold in clamshells. Most wild muscadines dehisce fairly rapidly (shatter) when ripe. This trait was heavily selected against during the development of the initial cultivars (Murphy et al., 1938). In the early 1900's, most muscadines were produced for juice and if produced for fresh use, the fruit were sold and used relatively quickly. Thus, initial cultivar development in muscadine grapes involved selection for berries that remained attached to the vine until the fruit could be harvested. With the development of a large fresh market for the muscadines, and the desirability of storing the fruit for up to a few weeks before sale, it has become necessary to develop cultivars with berries that can be picked easily and cleanly from the vine. More recently, interest in mechanical harvesting of the fruit is requiring the development of cultivars which require a lower force for fruit detachment.

Muscadine grape cultivars have been broadly divided into two groups: 1) those which produce berries that separate from the pedicel relatively cleanly with a well defined abscission zone and a breakdown of the vasculature in the junction (dry stem scar); and 2) those in which there is no abscission zone so that when berries are removed, a tuft of the vasculature is pulled

from the berry, leaving an open wound (wet stem scar; Goffinet et al., 2001). Microscopic examination of dry stem scar types reveal a general breakdown of lignified vascular tissue in the abscission zone and the production of a thin, funnel shaped separation layer across the receptacle tissue (Sherman, 1963). Subsequent to fruit detachment, a suberized layer is produced distal to the abscission zone, which helps to seal the fruit against water and juice loss, cracking, and invasion by pathogens. On the other hand, a wet stem scar serves as an entry point for pathogens, increases desiccation of the berry, and enables juice leakage making the fruit commercially undesirable. For example, 'Carlos' fruit with wet stem scars cannot be stored longer than two weeks and had six to ten times greater decay than fruit with dry stem scars (Ballinger and Nesbitt, 1982). Most cultivars typically produce a mixture of wet and dry stem scars, and cultivars are often rated according to the average percentage of berries with a dry stem scar. For example, the vasculature of 'Supreme', the primary fresh market purple cultivar in Georgia, remains in the berry when picked, and thus would broadly be termed as producing a dry stem scar. However, the berry skin in this cultivar is frequently torn or the berry flesh splits, around the picking scar. One major grower (Paulk Vineyards, Ocilla, GA) estimates that they have to juice about 30% of the berries due to this problem. Many other cultivars are not grown commercially because of this issue (Conner, 2009). Hence, the production of reliable and true dry stem scars is vital to the success of any cultivar. The development of new cultivars with thinner more palatable skins is an important goal of the University of Georgia muscadine breeding program, to increase the acceptance of muscadine fruit in the marketplace (Conner, 2010). However, emphasizing this trait will further increase the need for dry stem scars as the thinner skin may make them more vulnerable to damage.

Abscission agents are plant growth regulators that can induce or accelerate organ detachment (abscission) from the parent plant. Several compounds such as Ethephon (2-chloroethylphosphonic acid), Benzyl Adenine (BA) and Methyl Jasmonate (MeJA) have been observed to induce fruit detachment in many fruits such as apple, citrus, table and raisin grapes, and blueberry. In many of the above fruits, abscission agents reduce the fruit detachment force (FDF) and in grape, they also aid in the development of a dry stem scar (Fidelibus et al., 2007; Gonzalez-Herranz et al., 2009). The development of abscission agents would clearly benefit the muscadine grape industry if they are reliable and have no deleterious effects on the vine. In fact, Ethephon has been studied as a potential harvest-aid for muscadines previously. Ethephon treatment reduced the wet stem scar percentage and FDF during initial trials in muscadine grapes (Lane and Flora, 1979; Phatak et al., 1980). The lack of predictability in response to Ethephon due to differences among cultivars, variability in the stage of ripening, and environmental conditions during application, has greatly limited its potential as a harvest-aid (Himelrick, 2003). Ethephon applications have not been evaluated on newer muscadine cultivars and the optimum dosage for its efficacy has not been determined. Also, the sensitivity to abscission agent applications including Ethephon appears to fluctuate diurnally (Pozo et al., 2007; Malladi and Burns, 2008). We hypothesize that Ethephon applied at the optimum dosage and at the optimum time of the day may result in consistent effects on enhancing fruit detachment and increasing the dry stem scar percentage among detached fruit.

Recently, several potential new abscission agents have been evaluated for their use in mechanizing the harvesting process in the raisin grape industry (Fidelibus et al., 2007). The most promising of these abscission agents was Methyl Jasmonate (MeJA). Jasmonates are novel plant hormones involved in numerous physiological plant processes including stress responses, senescence, and abscission (Gross and Parthier, 1994). Jasmonate applications appear to increase

internal ethylene concentrations of fruit similar to other abscission agents (Hartmond et al., 2000). Application of 10 mM to 20 mM MeJA reduced the FDF in table grapes within 2 to 3 days after treatment, and promoted abscission from 3-8 days after treatment (Gonzalez-Herranz et al., 2009). Importantly, treated berries also displayed less tearing of the skin around the detachment point than control berries (Fidelibus et al., 2007). Lower levels of MeJA appeared to have little deleterious effects on the vine canopy (Gonzalez-Herranz et al., 2009). In blueberries, application of 20 mM MeJA enhances fruit detachment resulting in extensive fruit drop within 2-3 days after treatment (Malladi and others, *In Press*). To our knowledge, the potential of MeJA as a harvest-aid in muscadine grapes has not been investigated. MeJA has the potential to reduce tearing and increase the percentage of dry stem scars, leading to an increase in the marketable yield and a reduction of postharvest losses in muscadine grape. It may prove to be especially useful in cultivars with a concentrated harvest period such as the newly released cultivar, Lane, by the University of Georgia, in which most berries ripen at same time. Another potential use of MeJA may be to increase the dry stem scar percentage and yield in mechanically harvested wine grape cultivars.

Methodologies

Mature vines of the muscadine cultivar, Fry, were used in this study in 2012. Two abscission agents were evaluated: ethephon and MeJA. Ethephon applications were performed at the following rates: 250, 500, 1000 and 2000 $mg \cdot L^{-1}$. The MeJA applications were performed at the rates of: 2, 5, 10 and 20 mM. All applications were performed with 0.15% of the adjuvant (Latron B-1956). The control vines were treated with the adjuvant only. Four single vine replicates per treatment were used in this study, except for the control (n = 3) and the 2 mM MeJA treatments (n = 3), due to the limited availability of the vines. All applications were performed by 1030 HR using a hand-pump sprayer until run-off. The average daily temperature on the day of the treatment was 25.5 °C and ranged from 23.1 °C to 27.5 °C during the course of the experiment. Four clusters per vine were selected and tagged. These clusters were enclosed in a perforated bag to allow for the collection of detached fruit. The number of detached fruit was counted at 2, 4 and 7 d after treatment and the total number of fruit on the cluster was determined at 7 d after treatment. These data were used to determine the percent fruit detachment. At 2, 4 and 7 d after treatment, the percent dry scar was determined.

Results

The higher rates of ethephon applications resulted in significant fruit detachment in ‘Fry’ (Table 1). The highest rate of ethephon application (2000 $mg \cdot L^{-1}$) resulted in rapid fruit detachment within 2 d after treatment, while the 1000 $mg \cdot L^{-1}$ application resulted in significant fruit detachment only at 7 d after treatment. The highest rate of ethephon application ultimately resulted in up to 73% fruit abscission. The applications of MeJA did not affect fruit detachment in ‘Fry’ (Table 2).

Ethephon applications of 500 $mg \cdot L^{-1}$ and higher resulted in significantly higher percent dry scar development in ‘Fry’, within 2 d after treatment (Table 3). The vast majority (up to 100%) of the fruit displayed a dry scar in response to these rates of ethephon application. At 4 and 7 d after treatment, ethephon applications at 250 $mg \cdot L^{-1}$ also resulted in significantly higher dry scar development than that in the control. The applications of MeJA did not affect the extent of dry scar development in ‘Fry’ at 2 and 7 d after treatment (Table 4). At 4 d after treatment, the

highest rate of MeJA application (20 mM) resulted in higher dry scar development than that in the control and the 5 mM MeJA treatments.

Conclusion

The data from this study demonstrate that ethephon applications at 1000 $mg \cdot L^{-1}$ or higher were effective in inducing fruit abscission and significant fruit detachment in the muscadine cultivar, Fry. The lower rates of application were largely ineffective in inducing fruit detachment. Interestingly, ethephon applications of 500 $mg \cdot L^{-1}$ or higher resulted in a dramatic increase in the occurrence of dry stem scars, often within 2 d after treatment. Together, these data suggest that ethephon applications have the potential to aid in dry stem scar development and may be suitable as harvest aids in muscadine. However, these applications need to be evaluated across multiple genotypes and in different years to determine if the effects are consistent. Also, it would be important to determine the effect of ethephon applications at different times of the day. Although such an experiment was planned in this year of the study, it could not be performed due to inclement weather.

MeJA applications up to 20 mM were ineffective in inducing fruit abscission. Additionally, only the highest rate of application increased the extent of dry stem scars in 'Fry'. The rates of application evaluated here are within the range described earlier for table grapes and blueberries (Fidelibus et al., 2007; Malladi et al., *In press*). Hence, the data from this study suggest that, unlike in table grapes and other fruit, this abscission agent does not induce significant fruit drop or consistent dry scar development in 'Fry'. While evaluations in additional cultivars are necessary to rule out the possibility of MeJA as a harvest aid for muscadines, the results from this study do not support such an application for this compound.

Impact statement

The data from this study suggest that ethephon applications may be useful as harvest aids in muscadines to induce fruit detachment and to increase the extent of dry stem scar development. Future studies should be aimed at evaluating this compound across multiple genotypes. Additionally, time of day of application and post-harvest effects of the application of this compound will need to be evaluated to determine if it can be applied in commercial practice.

Citations

No publications have been made from the above research

Table 1. Percent fruit detachment in response to ethephon application in 'Fry'.

Ethephon ($mg \cdot L^{-1}$)	Fruit detachment (%)		
	2 DAT[zy]	4 DAT[zy]	7 DAT[zy]
0	0 b	0 b	0 c
250	0 b	0 b	1 c
500	0 b	6 b	9 c
1000	10 b	22 b	30 b
2000	32 a	63 a	73 a

[z]DAT: days after treatment.
[y]Numbers followed by the same letter are not significantly different (Duncan's multiple range t-test after ANOVA).

Table 2. Percent fruit detachment in response to MeJA application in 'Fry'.

MeJA (mM)	Fruit detachment (%)		
	2 DAT[z]	4 DAT[z]	7 DAT[z]
0	0	0	0
2	0	0	0
5	0	0	0
10	0	0.5	1.1
20	0	0	1.4

[z]DAT: days after treatment.

Table 3. Percent dry scar in response to ethephon application in 'Fry'.

Ethephon ($mg \cdot L^{-1}$)	Dry scar (%)		
	2 DAT[zy]	4 DAT[zy]	7 DAT[zy]
0	14 b	27 c	18 c
250	30 b	78 b	38 b
500	92 a	73 b	88 a
1000	94 a	98 a	88 a
2000	99 a	100 a	100 a

[z]DAT: days after treatment.
[y]Numbers followed by the same letter are not significantly different (Duncan's multiple range t-test after ANOVA).

Table 4. Percent dry scar in response to MeJA application in 'Fry'.

MeJA (mM)	Dry scar (%)		
	2 DAT[z]	4 DAT[zy]	7 DAT[z]
0	14	27 b	18
2	22	40 ab	33
5	23	21 b	31
10	19	32 ab	32
20	53	48 a	30

[z]DAT: days after treatment.
[y]Numbers followed by the same letter are not significantly different (Duncan's multiple range t-test after ANOVA).

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