Title: Evaluation of ethylene emission and fruit detachment characteristics in rabbiteye blueberry varieties

Progress Report for SRSFC Research Project # 2010 - 14 Research Proposal Name, Mailing and Email Address of Principal Investigators: Principal Investigator Anish Malladi Assistant Professor Department of Horticulture, 1121 Miller Plant Sciences, University of Georgia, Athens, GA-30602 Email: malladi@uga.edu

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**Objective**: To evaluate the relationship between ethylene emission and fruit detachment characteristics in rabbiteye blueberry varieties

## Justification

Blueberry is rapidly emerging as a major fruit crop in the southeastern US (Krewer and Nesmith, 2000). Area under blueberry production continues to increase annually in states such as Georgia (exceeding 10,000 acres) and this region is expected to become the leading producer of blueberries in the US over the next few years (Strik and Yarborough, 2005). Harvesting is one of the most labor intensive and expensive aspects of blueberry production. Decreasing availability of labor further complicates harvesting-related issues. Mechanical harvesting (MH) of blueberry can substantially reduce production costs. MH involves vigorous shaking of the bush resulting in fruit detachment, but often causes substantial injury to the fruits and/or bushes. Ground loss in blueberry MH is estimated to be over 20%. Also, MH cannot be used for many of the current cultivars due to poor fruit detachment characteristics or unfavorable plant architecture. Enhancing fruit detachment characteristics may help in reducing the force required for MH thereby minimizing the above losses and increasing the efficiency of MH. Hence, the long-term

goal of our research is to develop a better understanding of factors that determine the ease of fruit detachment.

Plant hormones such as ethylene play an important role in accelerating abscission/organ detachment (Jackson and Osborne, 1970; Roberts et al., 2002). Exogenous application of ethylene or ethylene releasing compounds such as ethephon can decrease the fruit detachment force (FDF) and accelerate abscission in many plants such as citrus and apple. Also, ethephon application can accelerate fruit abscission in rabbiteye blueberries (Malladi and others, *in preparation*). Internal levels of ethylene can strongly influence natural fruit detachment through activation and progression of cell separation processes within the abscission zone (Grumet et al., 1981; Sun et al., 2009). We hypothesize that differences in ethylene emission among rabbiteye blueberry varieties may contribute to variation in FDF, and that an increase in ethylene biosynthesis during fruit maturation may activate cell separation processes in the AZ leading to a reduction in FDF. Therefore, the specific objective of this proposal is to explore the relationship between ethylene emission and the ease of fruit detachment among rabbiteye blueberry varieties/selections.

### Methodology

### **Plant Material**

Mature rabbiteye blueberry plants growing at the Georgia Experiment Station in Griffin, GA, were used for this study. Thirty rabbiteye blueberry genotypes consisting of cultivars as well as advanced selections from the University of Georgia Blueberry Breeding Program were used. Plants were fertilized and irrigated regularly during the growing season. Effective pollination was facilitated by the presence of additional genotypes around the experimental plots and supplemental bees. In addition, mature rabbiteye blueberry plants at the Horticulture Farm in Athens, GA, were also used in this study.

#### Fruit Detachment Force (FDF) Measurement

FDF was measured using a pull force gauge (Wagner Instruments) fitted with an adapter to facilitate fruit removal from the plant. A modification to an earlier protocol (Malladi and NeSmith, 2010) was made to better simulate harvesting conditions. Fruit detachment was directly measured on intact berries on the plant. The fruit was placed in the adapter and the peak force required to detach the fruit from the plant was recorded using the gauge (in KgF). FDF was measured on 40 mature berries (blue to black in color) from 2-3 plants per genotype (30 genotypes). FDF measurements were made when more than 50% of fruit on the plant were mature. After FDF measurement, a 20-fruit sub sample was collected and fruit weight and diameter were measured on these fruit.

#### Ethylene Measurement

Ethylene emission from mature berries was determined using gas chromatography. Two mature fruit from each genotype were placed in a glass vial and the vial was sealed tightly using a rubber stopper (n = 4). Ethylene was collected in the head space of the vial for four hours. Head space gases were extracted using a plastic syringe and injected into a Shimadzu GC-17A equipped with a HP PLOT-Q capillary column. Ethylene standards were used for quantification. Fruit were removed from the vial after ethylene measurement and weighed.

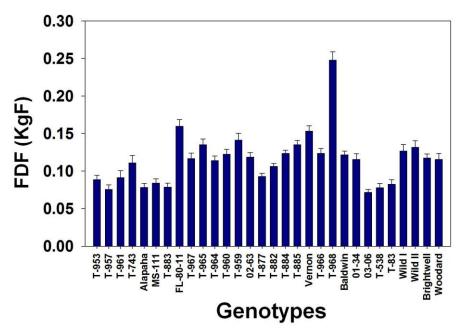
### Expression of Ethylene Biosynthesis-Related Genes

FDF measurements were made on mature as well as immature blueberry fruit from two genotypes growing in Athens, GA. Immature and mature fruit from each of the two genotypes were collected (n = 4). Fruit were frozen in liquid nitrogen and stored at -80°C until further analysis. RNA extraction followed by quantitative RT-PCR will be performed on these samples.

### Results

#### FDF Measurement in Rabbiteye Blueberry Genotypes

FDF was measured in 30 rabbiteye blueberry genotypes. FDF varied by over 3-fold across the 30 genotypes (Fig. 1). T-968 displayed the highest while the 03-06 displayed the lowest FDF (Fig. 1). FDF values were slightly lower in the current study in comparison to that recorded previously (Malladi and NeSmith, 2010), likely due to the modified method of FDF measurement. However, trends in FDF in the current study were largely similar to that in the previous study. For example, the genotype FL 80-11 which displayed the second highest FDF value in the current study, displayed the highest FDF in the previous study (T-968 was not evaluated in the previous study; Malladi and NeSmith, 2010). These data suggest that FDF measurement may be a reliable method for comparing the force required for fruit detachment in blueberry. These data also indicate that considerable variation is present among genotypes in terms of their FDF.



**Figure 1:** Fruit detachment characteristics in rabbiteye blueberry genotypes. Fruit detachment force (FDF) was measured in 30 rabbiteye blueberry genotypes.

#### Fruit Weight and Diameter among Rabbiteye Blueberry Genotypes

Fruit weight was highest in the genotypes, T-953 and T-957 (Fig. 2). These genotypes also displayed the largest fruit diameter (Fig. 3). 'Brightwell' displayed the lowest fruit weight and diameter (Fig. 3). Overall, a five-fold variation in fruit weight and 1.5-fold variation in fruit diameter were observed in this study.

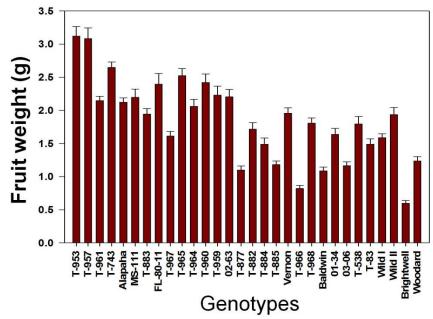


Figure 2: Fruit weight in thirty rabbiteye blueberry genotypes.

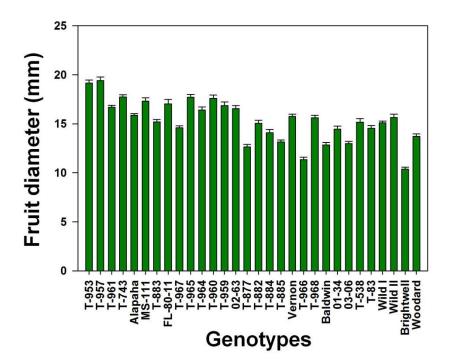
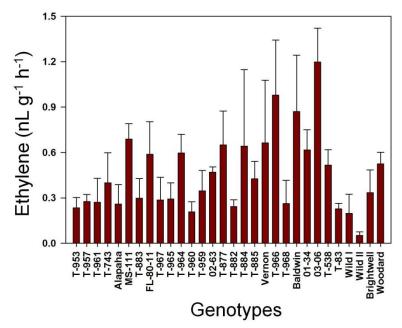


Figure 3: Fruit diameter in thirty rabbiteye blueberry genotypes

## Ethylene Measurement in Rabbiteye Blueberry Genotypes

Ethylene emission from mature fruit was determined in 30 rabbiteye blueberry genotypes using gas chromatography. In general, blueberry fruit displayed lower ethylene emission than

that observed in other fruit such as tomato and apple. Ethylene emission displayed considerable variation among the 30 genotypes (> 22-fold; Fig. 4). While a wild genotype ('Wild II') displayed the lowest, an advanced selection, 03-06, displayed the highest amount of ethylene emission (Fig. 4).



**Figure 4:** Ethylene emission characteristics in rabbiteye blueberry genotypes. Ethylene emission from mature fruit of 30 rabbiteye blueberry genotypes was measured using gas chromatography.

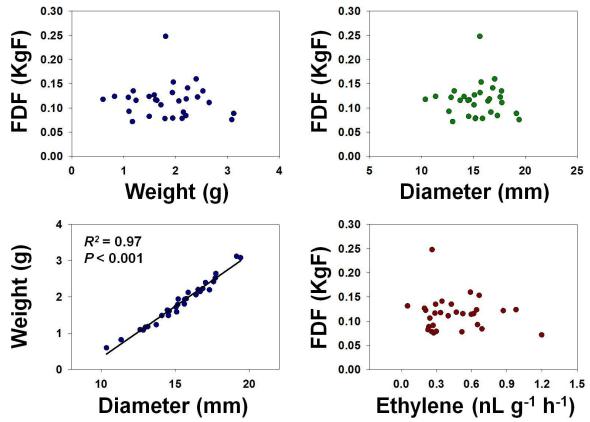
## Relationship among FDF, Fruit Weight, Fruit Diameter and Ethylene Emission

Regression analysis was performed to determine the relationships among the above described parameters. FDF was not significantly related to fruit weight or fruit diameter (Fig. 5). These data suggest that fruit size does not affect fruit detachment characteristics. Interestingly, there was no significant relationship between FDF and ethylene emission across the 30 genotypes. These data suggest that FDF may not be dependent on the extent of ethylene emission from blueberry fruit, indicating that additional factors may influence fruit detachment characteristics in rabbiteye blueberry. Fruit weight and fruit diameter were significantly related across the 30 rabbiteye blueberry genotypes, similar to data reported previously (Johnson et al., 2011).

#### Expression of Ethylene Biosynthesis-Related Genes

Preliminary data suggest that FDF decreases during later stages of fruit development. Such a decrease in FDF may be related to the dynamics of ethylene biosynthesis. The above relationship is being further explored in this study. Although not part of the initial proposal, a quantitative RT-PCR approach is being used for this study to determine the dynamics of the expression of ethylene biosynthesis-related genes. Expression of genes such as amino-cyclopropane-carboxylate (ACC) synthase and ACC oxidase (ethylene biosynthesis-related genes) is often closely related to internal ethylene biosynthesis. Analysis of the expression of these genes during fruit maturation can be an effective tool to explore the relationship between changes in ethylene biosynthesis and fruit detachment characteristics during fruit development. Immature and mature

fruit have been collected from two rabbiteye blueberry genotypes, frozen and stored at -80°C. Analysis of expression of the above genes in immature fruit and mature fruit in the two genotypes using quantitative RT-PCR will be performed shortly.



**Figure 5:** Relationships among fruit detachment, fruit weight, fruit diameter and fruit ethylene emission characteristics in 30 rabbiteye blueberry genotypes. No significant relationships could be determined between: FDF and fruit weight; FDF and fruit diameter; and FDF and ethylene emission. A significant linear relationship was observed between fruit weight and fruit diameter.

## Conclusions

FDF varied widely across the rabbiteye blueberry genotypes. Variation in FDF was not significantly related to fruit weight or fruit diameter indicating that fruit size does not affect FDF. Ethylene emission characteristics were not related to FDF suggesting that ethylene emission may not regulate fruit detachment characteristics across blueberry genotypes.

## **Impact Statement**

This study suggests that fruit detachment is not influenced by fruit size in rabbiteye blueberry. More importantly, this study suggests that variation in FDF across rabbiteye blueberry genotypes is not related to ethylene emission characteristics. These data indicate that additional intrinsic factors may be involved in determining the observed variability in FDF. Analysis of gene expression dynamics in a developmental context may provide insights into the relationship between ethylene biosynthesis and fruit detachment.

# Citation(s)

No publications have yet been developed from the above study.

# References

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