Southern Region Small Fruit Consortium

Progress Report Research

Title: Characterizing Marketable Attributes of Juice from Noble and a Potential New Processing Muscadine Cultivar

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Public Abstract

In progress

Introduction

Muscadine grapes (*Vitis rotundifolia* Michx.) are a disease-resistant specialty crop native to the southeastern United States. There have been major advances in U.S. muscadine breeding efforts resulting in unique traits emerging for muscadine grapes used for commercial juices. The market presence for the muscadine industry as a southern region crop can be strengthened by understanding what consumers want in a muscadine juice. This research from the University of Arkansas System Division of Agriculture (UA System) will **characterize marketable attributes of juice from Noble and a potential new black-fruited processing muscadine cultivar** by evaluating physicochemical and sensory attributes that drive marketability. **This project is important because it will help establish marketability attributes that drive consumer purchasing of muscadine juice in addition to providing opportunities for a new processing muscadine cultivar for the U.S. southeastern region.**

Growing Muscadine Grapes. Muscadines differ from bunch grapes because they have smaller clusters, the berries abscise (shatter) at maturity, and the tendrils are unbranched. Muscadine clusters contain 6-24 berries and are classified by color, with bronze or black as the two prevalent color types (Conner 2010, Mortensen 2001). These native grapes have been cultivated for over 400 years and have a strong heritage in U.S. viticulture (Olien 1990). Muscadine grape production can be profitable for commercial growers (Noguera et al. 2005), but is dependent on availability of consumer markets. The top commercial muscadine-producing states are North Carolina (2,600 acres), Georgia (1,700 acres), and Florida (1,200 acres) (USDA Agricultural Census 2012).

<u>Muscadine Juice Production.</u> A majority of the commercial muscadine crop is used to produce juice and wine. The two most popular cultivars for processing are Noble, a black

muscadine, and Carlos, a bronze muscadine. Muscadine juice and wine can have poor color, color stability, and cloudiness/sediment. Muscadine grapes and wines contain diglycosidic anthocyanins, which are unable to form stable polymeric pigment complexes (Sims and Morris, 1985). In juice production, extraction of the color from the skins of the grapes is done by applying heat to the must (seeds, skins, pulp, and juice) after the grapes are crushed but before the grapes are pressed. In contrast, cold press is when the must is pressed after the grapes are crushed, yielding lighter-colored and fruiter juice but with less juice yield (Striegler et al., 2005; Threlfall et al., 2005; 2007).

Muscadine Grape Breeding. There are public and private muscadine breeding programs across the southern United States. These continued breeding efforts result in better quality for consumers and increased cultivar options for growers. The southeastern U.S. muscadine breeding efforts are focused on improving traits for fresh-market and processing muscadine genotypes (cultivars and breeding selections), resulting in an expansion of the germplasm base used in muscadine breeding. The UA System Fruit Breeding Program began a muscadine breeding program in 2007 that includes many breeding selections with unique profiles. One of the UA System advanced breeding selections, AM-77, shows great potential for juice and wine production and will likely be one of the first muscadine grape cultivars released from the UA System in 2022-2023. AM-77 is a black muscadine that has larger fruit size than Noble and potentially better color for juice and wine production. To determine whether AM-77 has a potential place in the market as a complement to Noble, we need to evaluate the consumer quality of the juices and wines produced from its fruit.

Physicochemical Components of Muscadines. Muscadines offer a healthy fruit choice for consumers and a marketing opportunity for producers. Muscadine grapes contain many health-promoting phenolic compounds including, resveratrol, ellagic acid, anthocyanins, and proanthocyanidins (Barchenger et al. 2014, 2015a, 2015b, Ector et al. 1996, 2001, Pastrana-Bonilla et al. 2003, Threlfall et al. 2005). Anthocyanins are highest in the skins of dark-colored muscadines (Striegler et al. 2005). Baek et al. (1997) and Baek and Cadawallader (1999) identified the predominant aroma compound in muscadine juice as furaneol (burnt sugar), *o*-aminoacetophenone ("foxy", artificial grape), 2-phenylethanol (honey, floral), diacetyl (butter), and ethyl butanoate (apple). The combination of these volatile compounds gives muscadine grapes, juices, and wines a unique aroma and flavor profile.

Sensory of Muscadine Grapes/Juice. The evaluation of factors that drive consumer acceptance is critical to the marketing of unique products. A consumer sensory study at the University of Florida showed that consumer panelists familiar with muscadine grapes had overall liking scores of fresh muscadine grapes correlated to muscadine flavor (Brown et al. 2016). For fresh-market muscadines and juice, the initial taste perception of sweetness, in particular the soluble solids/acid ratio, is a key aspect for sensory acceptability. Threlfall et al. (2007) evaluated the descriptive and consumer sensory attributes of juice from eight muscadine cultivars, including five black cultivars and three bronze cultivars plus two commercial juices. Consumers rated Black Beauty, Granny Val, Ison, Southern Home, and Summit juices highest for overall liking. The descriptive panel created a sensory lexicon with major attributes identified as sweet, sour, cooked muscadine, cooked grape, and astringent. Correlation between consumer and descriptive showed overall liking correlated positively to sweetness and caramelized and correlated negatively to sour and green unripe. Consumers showed a preference for juice sweetness with soluble solids $\approx 14\%$ and soluble solids/acid ratios of 26 to 31. Further consumer sensory evaluations of muscadine juice is needed, especially using 100% Noble muscadine

grapes, and although commercial muscadine juices are available these are typically blends of different cultivars.

Objectives

This is a progress report on the research efforts expended. Muscadines were harvested in September/October 2022, and juice was made for the project. We completed the consumer sensory evaluation of the muscadines juices, but have not completed the physicochemical analysis for this project. The equipment for volatile analysis is shared with other researchers, and the equipment was being used for another project. Color and composition analysis will be done in December 2022. Volatile analysis and volatile identification of the muscadines is planned for December 2022 and January 2022.

Objectives:

1. Evaluate the impact of cold versus hot pressing on juice from Noble and a potential new processing muscadine cultivar from the UA System

Measure physicochemical attributes and conduct consumer sensory analysis of juice produced from Noble and AM-77, a potential new processing muscadine cultivar from the UA System Fruit Breeding Program

2. Compare physicochemical and consumer sensory attributes of juice from Noble and a potential new processing muscadine cultivar from the UA System

Identify consumer-driven attributes of Noble and AM-77 muscadine grape juice by comparing the physicochemical and sensory attributes

3. Disseminate information to muscadine grape growers and juice producers on marketable quality attributes

Present results at conferences and host a field day/workshop on muscadine breeding, production, and utilization

Materials and Methods

Harvest

The genotypes were hand harvested in the morning from a commercial vineyard in Altus, AR. About 120 kg of each genotype were harvested (117 kg of AM-77 harvested on September 9, 2022 and 118 kg of Noble harvested on October 4, 2022). After harvest, the grapes were taken to the UA System Food Science Department in Fayetteville. The lugs of grapes for each genotype were randomized into eight lugs prior to processing on the day of harvest.

Grape processing

The grapes were divided into four lots (27 kg each) for each genotype. Each batch of grapes was passed twice through a crusher/destemmer, and 30 mg/L sulfur dioxide (SO2) as potassium metabisulfite (KBMS) was added at crush. After crushing the grapes for each genotype, the must (seeds, juice, skin, and pulp) for each batch was placed into food grade polyethylene containers and assigned into two treatments (Cold Press or Hot Press) in duplicate.

For the cold press treatments, the must was pressed after crushing. For the hot press treatments, the must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed. The musts were pressed

with a 70-L Eno Agricola Rossi Hydropress (Calzolaro, Italy). The juice was collected in 11.4-L glass carboys, cold (2°C) settled overnight, then racked. The juice was placed in a steam kettle, pasteurized (88 °C for 1 minute), and hot filled into 125-mL, 375-mL and 750-mL glass bottles. After sealing the bottles with the caps, the bottles were placed on their sides to cool. The pasteurized juice was stored at 2 °C until physicochemical and sensory analysis.

Statistical design and analysis

There were four treatments (AM-77 Cold Press, AM-77 Hot Press, Noble Cold Press, and Noble Hot Press) in duplicate. Statistical analyses will be conducted using JMP® (version 16.0; SAS Institute Inc., Cary, NC). Tukey's HSD (Honestly Significant Difference) will be used for mean separation. Pearson's correlation will be used to test the relationship between/within attributes.

Methods for physiochemical analysis (*will be done December 2022/January 2023*) The physiochemical attributes (color, composition, and volatile profile) of each juice sample (2 genotypes x 2 press treatments x 2 replications) will be evaluated at the UA System Food Science Department.

Color analysis

L*, a*, b*, hue angle, and chroma. Juice color analysis will be conducted using a ColorFlex system (HunterLab, Reston, VA). The ColorFlex system uses a ring and disk set (to control liquid levels and light interactions) for measuring translucent liquids in a 63.5-mm glass sample cup with an opaque cover to determine CIELab transmission values of L*=100, a*=0, and b*=0 (CIE, 1986).

Red color and color density. Red color of juice will be measured spectrophotometrically as absorbance at 520 nm, and color density was measured as red color + yellow/brown color (420 nm) (Iland et al. 1993). Absorbance values will be measured using a Hewlett-Packard 8452A Diode Array spectrophotometer equipped with UV-Visible ChemStation software (Agilent Technologies, Inc., Santa Clara, CA). Samples will be diluted with deionized water as needed prior to analysis and measured against a blank sample of deionized water. A 1-cm cell will be used for all spectrophotometer measurements.

Composition analysis

Soluble solids, pH, and titratable acidity. Juice will be used to determine soluble solids, pH, and titratable acidity. Total soluble solids (expressed as %) will be measured using a refractometer. Titratable acidity and pH will be measured with an automated titrimeter. Titratable acidity will be determined using 6 mL of juice diluted with 50 mL of deionized, degassed water by titration with 0.1 N sodium hydroxide (NaOH) to an endpoint of pH 8.2; results will be expressed as g/L tartaric acid. **Sugar and acid analysis.** Organic acids and sugars will be determined using HPLC. Glucose, fructose, and citric, tartaric, malic, succinic and lactic acids of the muscadine juices will be measured using procedures described in Walker et al. 2003. The HPLC will be equipped with a Bio-Rad HPLC Organic Acid Analysis Aminex HPX-87H ion exclusion column (300 x 7.8 mm) and a Bio-Rad HPLC column (150 x 7.8 mm) in series. A Bio-Rad Micro-Guard Cation-H refill cartridge (30 x 4.5 mm) will be used as guard column. The peaks will be quantified using external standard calibration based on peak height estimation with baseline integration.

Volatile profile analysis

Volatile compounds analysis. The volatile aroma compound profile of juices will be determined using solid-phase micro-extraction-gas chromatography (SPME-GC) and SPME-GC-MS. For SPME extraction of volatile compounds, 1 mL of juice will be placed in a 20 mL glass headspace vial with a small glass coated magnetic stir bar, and the vial will be sealed with a PFTE lined silicone septa. The headspace volatiles will be extracted for 30 min at 65°C onto a Supelco 2 cm stable flex 85 μ m CAR/PDMS fiber. The SPME fiber will be manually placed into the injection port of a Varian CP-3800 gas chromatograph. Volatiles will be desorbed from the fiber and separated with a fused silica

capillary column Agilent HP-5 column (30 m length, 0.25 mm id, 1 μ m film thickness). The oven temperature will be held at 28°C for 5 min and then ramped to 250°C at a rate of 8°C/min (total run time 32.75 min). To identify chromatogram peaks, the same procedure will be performed using a GC-MS. Mass spectral data will be done in scan mode with a mass scan range from 20 to 300 m/z. Volatile substances will be identified by comparison of their mass spectra with the Wiley7NIST0.5 mass spectral library and relevant literature data.

Methods for consumer sensory analysis (completed October 2022)

The sensory attributes of the juices were evaluated during the NCCC212 meeting on October 26, 2022 in Fayetteville, AR and at the UA System Food Science Department on October 31, 2022. For sensory analysis, the replications of each treatment were combined (2 genotypes x 2 press treatments) for a total of four juice samples.

Consumers panelists (66) assessed the color, aroma, and taste of the four muscadine juices evaluated for physiochemical attributes. The sample presentation order was randomized. Sample cups were labeled with three-digit codes, and each panelist was served 30 mL of juice. Unsalted crackers and water were provided for palate cleansing between samples. Each consumer evaluated overall impression, color, aroma, flavor, sweetness and sourness on a 9-point verbal hedonic scale (1 = dislike extremely; 9 = like extremely) and a 5-point Just About Right (JAR) scale (1 = not nearly enough 3 = just about right; 5 = much too much). Consumers were also asked a preference question about the samples. Consumers were asked demographic questions. Data was acquired using paper ballots.

Methods for outreach dissemination

Present results at conferences and host a field day/workshop on muscadine breeding, production, and utilization

Results and Discussion

Physiochemical analysis (In progress)

Consumer sensory analysis (completed October 2022)

Overall, 59% of panelists for the consumer sensory study were female and 41% were male. In terms of age, 20% were 21 years of age or younger, 39% were 22-34, 16% were 35-44, 3% were 45-54, 12% were 55-64, and 10% were 65 or older. Over half (51%) of panelists had a graduate degree, 16% had a four-year degree, 28% had some college education, and 5% indicated "other" for education. Fifty-nine percent of panelists had previously consumed muscadine juice while 41% had not. Twenty-one percent had purchased muscadine juice, while 79% had not. When asked how often they consumed muscadine juice, 49% indicated "never", 28% indicated once per year, 20% indicated 2-3 times per year, and 3% indicated 4-10 times per year.

Consumers panelists (66) evaluated color, aroma, flavor, sweetness, sourness, and overall impression of the juice on a 9-point verbal hedonic scale and a JAR scale. The liking for all the attributes ranged from 4.53 (dislike slightly to neither like nor dislike) to 7.30 (like moderately). For the hedonically rated attributes, the panelists found significant differences in the attributes except aroma which was rated 5.93 (neither like nor dislike to like slightly). The Noble hot press juice (7.30) was scaled the highest for color and was higher than the AM-77 cold press and Noble cold press. In general, the cold press juices were pink/light red, whereas the hot press

juices were dark purple. **AM-77 cold or hot press juice was higher in liking for flavor, sourness, and overall impression than Noble cold or hot press**. AM-77 hot press was scaled highest for sweetness and was scaled higher than Noble cold press.

Seventy-six percent of the panelists found AM-77 too light in color, but 82% found AM-77 hot press JAR for color. The panelists A majority (76%) of the panelists found AM-77 cold press juice JAR for aroma. In terms of flavor, panelists found the AM-77 cold and hold press juice (62-70%) more JAR than the Noble cold and hot juice (42-44%). AM-77 hot press juice (65%) had the highest rating for JAR sweetness. In terms of sourness, panelists found the AM-77 cold and hold press juice (55-56%) more JAR than the Noble cold and hot press juice (32%).

In terms of preference, 42% preferred AM-77 hot press juice, followed by 29% for AM-77 cold press juice, 16% Noble cold press juice, and 13% Noble hot press juice.

Outreach dissemination (in progress)

Conclusions

In progress

Impact Statement

In progress

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Table 1. Composition of cold and hot pressed juice made from muscadine grapes grown in Arkansas and evaluated at the University of Arkansas System Division of Agriculture (2022).

	Press	Soluble solids		Titratable acidity
Genotype z	treatment ^y	(%)	pН	(%) ^y
AM-77	Cold press			
AM-77	Hot press			
Noble	Cold press			
Noble	Hot press			
P-value	1			

^z Genotypes were evaluated in duplicate. Means with different letters for each attribute within location are significantly different (p<0.05) using Tukey's Honestly Significant Difference test.

^y For cold press treatments, must was pressed after crushing. For hot press treatments, must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed.

Table 2. Color attributes of cold and hot pressed juice made from muscadine grapes grown in Arkansas and evaluated at the University of Arkansas System Division of Agriculture (2022).

	Press		Hue		Red	Brown	Color
Genotype z	treatment ^y	L^*	angle $(^{\circ})^{x}$	Chroma	$color^{w}$	$\mathbf{color}^{\mathbf{w}}$	densityw
AM-77	Cold press						
AM-77	Hot press						
Noble	Cold press						
Noble	Hot press						
P value	•						

^z Genotypes were evaluated in duplicate. Means with different letters for each attribute within location are significantly different (p<0.05) using Tukey's Honestly Significant Difference test.

^y For cold press treatments, must was pressed after crushing. For hot press treatments, must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed.

^x Hue angles <90° were subjected to a 360° compensation to account for discrepancies between red samples near 0° and those near 360°.

W Red color calculated as absorbance of wine at 520 nm, Brown color at 420 nm, Color density at 520 nm + absorbance 420 nm.

Table 3. Sugars and organic acids of cold and hot pressed juice made from muscadine grapes grown in Arkansas and evaluated at the University of Arkansas System Division of Agriculture (2021).

Genotype ^z	Press treatment ^y	Glucose	Fructose (%)	Total sugars (%)	Tartaric acid (%)	Malic acid (%)	Citric acid (%)	Total organic acids (%)
AM-77	Cold press							
AM-77	Hot press							
Noble	Cold press							
Noble	Hot press							
P value	_							

^z Genotypes were evaluated in duplicate. Means with different letters for each attribute within location are significantly different (p<0.05) using Tukey's Honestly Significant Difference test.

^y For cold press treatments, must was pressed after crushing. For hot press treatments, must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed.

Table 4. Volatile compounds (µg/kg) of cold and hot pressed juice made from muscadine grapes grown in Arkansas and evaluated at the University of Arkansas System Division of Agriculture (2022).

Compound ^z	Measured retention index	Aroma category	Aroma description	AM-77 cold press ^y	AM-77 hot press	Noble cold press	Noble hot press
Totals							

² Genotypes were evaluated in duplicate. Relative peak area percent Compounds were identified by comparison of mass spectra with NIST14 (National Institute of Standards and Technology, Gaithersburg, MD, USA), Flavors and Fragrances of Natural and Synthetic Compounds (FFNSC3, John Wiley & Sons, Inc., Hoboken, NJ, USA), and Adams Essential Oils (Adams 2007) mass spectral libraries and comparison of calculated Kovats retention indices (Kováts 1958) with previously reported values

^y For cold press treatments, must was pressed after crushing. For hot press treatments, must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed.

Table 5. Attributes evaluated by a consumer sensory panel using a nine-point hedonic scale ^z for cold and hot pressed juice made from muscadine grapes grown in Arkansas and evaluated at the University of Arkansas System Division of Agriculture (2022).

	Press						Overall
Genotype y	treatment ^x	Color	Aroma	Flavor	Sweetness	Sourness	impression
AM-77	Cold press	5.06 c	5.71 a	6.09 a	5.83 ab	5.61 a	6.03 a
AM-77	Hot press	7.02 ab	6.14 a	6.05 a	6.05 a	5.61 a	6.17 a
Noble	Cold press	6.20 b	5.76 a	5.02 b	5.08 b	4.53 b	4.92 b
Noble	Hot press	7.30 a	6.11 a	4.66 b	5.29 ab	4.59 b	4.97 b
P value		<0.0001	0.2842	<0.0001	0.0043	<0.0001	<0.0001

^z Wines were evaluated by 66 consumer panelists using a nine-point hedonic scale (1=dislike extremely, 2=dislike very much, 3=dislike moderately, 4=dislike slightly, 5=neither like nor dislike, 6=like slightly, 7=like moderately, 8=like very much, and 9=like extremely).

y Means with different letters for each attribute within location are significantly different (p<0.05) using Tukey's Honestly Significant Difference test.

^x For cold press treatments, must was pressed after crushing. For hot press treatments, must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed.

Table 6. Percent (%) of responses for consumer sensory analysis using a collapsed five-point just-about-right (JAR)^z scale for cold and hot pressed juice made from muscadine grapes grown in Arkansas and evaluated at the University of Arkansas System Division of Agriculture (2022).

Genotype	Genotype	Press treatment y		Color			Aroma			Flavor		Sv	veetne	ss	S	ournes	SS
		Not enough	JAR	Too much	Not enough	JAR	Too much	Not enough	JAR	Too much	Not enough	JAR	Too much	Not enough	JAR	Too much	
AM-77	Cold press	76	23	1	35	56	9	18	70	12	11	45	44	36	56	8	
AM-77	Hot press	4	82	14	15	74	11	15	62	23	24	65	11	8	55	37	
Noble	Cold press	48	49	3	46	49	5	23	44	33	11	46	43	42	32	26	
Noble	Hot press	2	63	35	36	55	9	11	42	47	27	38	35	32	32	36	

^z Wines were evaluated by 66 consumer panelist using a five-point JAR scale (1 = much to low; 2 = too low; 3 = JAR; 4 = too much; 5 = much too much) collapsed to Too low, JAR, and Too much.

^y For cold press treatments, must was pressed after crushing. For hot press treatments, must was placed in a 76-L (20-gal) steam kettle, heated to 38 °C, held for one minute, placed back into the polyethylene container, allowed to cool, and then pressed.