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Fruit quality, nutraceutical and antimicrobial properties of 58 muscadine grape varieties (*Vitis rotundifolia* Michx.) grown in United States



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ABSTRACT

Fifty-eight muscadine grape varieties were evaluated for their fruit quality, nutraceutical, and antimicrobial properties during two growing seasons (2012 vs. 2013). Fruit quality was significantly different among muscadine grape varieties, with weight ranged from 2.93 to 22.32 g, pH from 3.01 to 3.84, titratable acidity from 0.27% to 0.83%, and °Brix from 10.92 to 23.91. Total phenols for different muscadine juices varied from 0.26 to 1.28 mg GA/mL, skins from 10.13 to 30.02 mg GA/g DM, and seeds from 22.47 to 72.01 mg GA/g DM. Accordingly, the antioxidant activity of grape juices varied from 0.97 to 6.78 mmol Trolox/mL, skins from 83.59 to 221.20 μ mol Trolox/g DM, and seeds from 178.22 to 619.73 μ mol Trolox/g DM. Study demonstratedgrape seed polyphenols (MIC 54.8–60.1 μ g/ml) showed stronger antimicrobial activity against *S. aureus* than skin polyphenols (MIC 70.7–80.2 μ g/ml). This information could be a valuable asset in the research and extension of muscadine grapes.

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1. Introduction

Muscadine grape (*Vitis rotundifolia* Michx.) is native to the southeastern United States and was the first native grape species to be cultivated (>400 years) in North America (Olien & Hegwood, 1990). The natural range of muscadine grapes extends from Delaware to central Florida and occurs in all states along the Gulf Coast to east Texas; it also extends northward along the Mississippi River to Missouri (Andersen, Crocker, & Breman, 2010). Olien (1990) reported there are nearly 100 improved cultivars, and around 5000 acres of muscadine grapes in commercial production in the southeastern United States (*V. rotundifolia* only).

The genus Vitis is divided into two subgenera: Euvitis (commonly referred as "bunch" grapes) and Muscadinia (with a common name as "muscadine" grapes). There are three species within the Muscadinia subgenera (V. rotundifolia, V. munsoniana, and V. popenoei). The genetic make-up of Muscadinia is different from Euvitis since it has two more somatic chromosomes (40 vs. 38). Muscadine grapes are tolerant of insect and disease pests, and homeowners

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can successfully grow muscadine grapes without spraying any pesticides (Huang, Milholland, & Daykin, 1986). Muscadine grapes are consumed as fresh fruit or processed into wine, juice, jam or jelly (Olien & Hegwood, 1990).

Muscadine grapes and wines are noted for their health benefits due to high phenolic contents and other nutritive values. Various studies on the fruit composition and health benefits of muscadine grapes have been reported: 1) grape polyphenols extraction, with organic solvents (Lee & Talcott, 2004) and enzymatic hydrolysis methods (Xu, Yagiz, Borejsza-Wysocki, et al., 2014); 2) grape polyphenols identification, such as resveratrol (Ector, Magee, Hegwood, & Coign, 1996), anthocyanins (Huang, Wang, Williams, & Pace, 2009), and the whole profile of polyphenols (Sandhu & Gu, 2010); 3) grape polyphenols health benefits, such as antioxidation and anti-microbial (Xu, Yagiz, Hsu, et al., 2014), anticancer (Hudson et al., 2007), and anti-cardiovascular diseases (Mellen, Daniel, Brosnihan, Hansen, & Herrington, 2010), 4) grape polyphenols enhancement measures, such as abscisic acid (Sandhu, Gray, Lu, & Gu, 2011), and UV irradiation methods (LeBlanc, 2006); 5) grape wine and juice, such as stability and color (Talcott, Brenes, Pires, & Del Pozo-Insfran, 2003), aroma compounds (Baek, Cadwallader, Marroquin, & Silva, 1997), and processing methods (Leblanc, Johnson, & Wilson, 2008); 6) grape

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fruit and seed oil compositions, such as protein (Mazhar, Basha, & Lu, 2002) and fatty acid analysis (Lamikanra & Lamikanra, 1989).

Although much work has been done on extraction, identification, and health benefits of polyphenols or other nutrients found in muscadine grapes, most of these studies have only looked at a few commonly grown muscadine varieties. With nearly 100 cultivars of muscadine, there is sparse information on the fruit quality, nutraceutical, and antimicrobial properties of these cultivars. Literature searching only can find: Mortensen and Harris (1988) compared the sensory difference among 43 muscadine grape cultivars grown in Florida; yield and fruit quality difference of 48 muscadine cultivars were evaluated between 1969 and 1973 (Mortensen & Balerdi, 1973; Striegler et al., 2005) determined the yield, fruit quality, and general nutraceutical properties of 20 muscadine cultivars grown in Arkansas; Marshall, Stringer, and Spiers (2012) analyzed the stilbene. ellagic acid, flavonol, and total phenols content of 21 muscadine cultivars grown in Mississippi. A more comprehensive look at all the muscadine cultivars would reveal the fruit properties of these less known cultivars. Also, an investigation into the correlation among these grape properties may reveal certain sort of

The objective of this study was to evaluate the fruit quality, nutraceutical, and antimicrobial properties of 58 muscadine grape varieties/breeding lines (varieties still in the breeding but haven't been named) over two consecutive seasons (2012 vs. 2013). This information could be integrated into the current information (e.g. vine vigor, yield, and disease resistance) of muscadine varieties to evaluate the overall performance of a variety. Access to this information, could assist scientists, such as grape geneticists, in breeding varieties with higher nutraceutical value; it also could help growers and consumers to select varieties which offer better quality and health benefit as a table grape.

2. Materials and methods

2.1. Grape materials

Fully ripened Muscadine grapes (53 cultivars and 5 breeding lines) were harvested from the Research Vineyard at the Center for Viticulture and Small Fruit Research (latitude 30.65 N, longitude 84.60 W) at Florida A&M University in 2012 and 2013 season. The ripeness of grapes was confirmed by the grape breeders at the Center, who determined the ripeness based on the grape °Brix, color, previously harvest time for each variety, etc. In each season, grapes of these varieties were harvested separately at three different times during the time of August and September, according to their mature period (early-, middle-, or later-ripe). Three to four clusters per vine and six vines per cultivar were randomly picked and shipped to the University of Florida on the same day and stored in a cold room (4 °C). Grape skins and seeds were separated manually from berries and freeze-dried in a freeze drier (Advantage, The Virtis Company, NY, USA). The freeze-dried samples were stored in vacuum-packaged polyethylene pouches at −20 °C until analyzed. Grape pulp after the removing of skins and seeds was squeezed to make the fresh juice for analysis soon afterwards.

Fifty-eight muscadine grape varieties in this study were: Africa Queen, Alachua, Albemarl, Black Beauty, Black Fry, Early Fry, Carlos, Delight, Dixie, Dixie Red, Doreen, Dixie Land, Cowart, Fry Seedless, Digby, Florida Fry, Farrer, Gold Isle, Darlene, Fry, Hunt, Higgins, Granny Val, Ison, Jumbo, Janet, Janebell, Lommis, Magnolia, Nesbitt, Pineapple, Southern Home, Later Fry, Pride, Sugargate, Noble, Sterling, Senoria, Sugar Pop, Southern Land,

Summit, Pam, Regale, Rosa, Scarlett, Sweet Jenny, Supreme, Tara, Triumph, Welder, Watergate, Majesty, and breeding lines (040-22-9, 026-1-2, 026-1-8, 028-22-5, C1-1-1).

2.2. Chemicals and bacterial strains

Folin & Ciocalteu's phenol reagent (2 N), Gallic acid, Catechin, Epicatechin, Epicatechin gallate, *trans*-resveratrol, Ellagic acid, Quercetin, Cyanidin-3, 5-diglucoside, Nalidixic acid, 2,2-diphenyl-1-picrylhydrazyl (DPPH), and 6-hydroxy-2, 5, 7, 8-tetramethylchroman-2-carboxylic acid (Trolox) were obtained from Sigma–Aldrich (St. Louis, MO, USA). Ampicillin and streptomycin were purchased from Fisher Scientific (Fair Lawn, NJ, USA). All other chemicals and solvents were purchased from Fisher Scientific Co. (Pittsburgh, PA, USA). Three *Staphylococcus aureus* strains (ATCC 12600-U, ATCC 35548, and ATCC 29247) were used for the antimicrobial activity study. All the ATCC bacterial strains tested were purchased from American Type Culture Collection (ATCC, Manassas, VA, USA).

2.3. Analysis of grape physicochemical properties

The surface color of muscadine grapes (n = 10) was measured by a machine vision system (Yagiz, Kristinsson, Balaban, & Marshall, 2007). The machine vision system was calibrated using a standard red plate ($L^* = 48.62$, $a^* = 49.04$, and $b^* = 25.72$) from Labsphere (North Sutton, NH). Average L^* , a^* , b^* values of the grapes surface were calculated using a color analysis program. The weight of muscadine grapes (n = 10) was measured using an analytical balance Mettler PM 400 (Mettler Instrument Corp., Hightstown, NJ, USA). The sizes of muscadine grapes were measured by diameter using a Vernier caliper. The pH and soluble solids (°Brix) were measured using a pH meter AB15 (Fisher Scientific, Pittsburgh, PA, USA) and an ABBE Mark II refractometer (Leica Inc., Buffalo, NY, USA). Titratable acidity was determined using a previous method (Iland, Ewart, & Sitters, 1993) and calculated as tartaric acid content (g/100 mL of juice). The °Brix to acid ratio for each sample was calculated by dividing the °Brix value by% acidity.

2.4. Preparation of grape skin and seeds

Freeze-dried grape skins (20 g) were ground with a stainless-steel grinder (Omni-Mixer 17105, OCI Instruments, CT, USA) for 1 min, and then placed on a sieve (\leqslant 0.25 mm) and the fine powder passing through the sieve was collected (Xu, Zhang, Wang, & Lu, 2010). The powdered samples were stored at $-20\,^{\circ}\text{C}$ and used for subsequent analysis. Freeze-dried grape seeds (20 g) were crushed and then defatted with hexane at a ratio of 1:10 (w/v). After 24 h extraction at room temperature (shaking every 6 h), the hexane extract was filtered using Whatman #4 filter paper (0.45 μ m) (Fisher Scientific, Pittsburgh, PA, USA) under vacuum. The residue was evenly distributed over a tray and kept in the hood to evaporate hexane. The final defatted grape seed powder was ground again in the stainless-steel grinder and the powder through the sieve (\leqslant 0.25 mm) was collected.

2.5. Extraction of phenolic compounds

Powder (0.5 g) from each sample above was extracted with 10 mL of 70% methanol. The extraction flasks were vortexed for 30 s, sonicated for 10 min, kept at room temperature (22 °C) for 60 min (shaking every 10 min), and sonicated for an additional 5 min. The extracts were transferred to tubes, centrifuged at 2820g, 0 °C for 10 min (J-LITE®JLA-16.250, Beckman Coulter Inc.,

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