Title: Optimizing texture assessment for muscadine grape breeding

Name, Mailing, and Email Address of Principal Investigator(s):

Principal Investigator

Margaret Worthington, Assistant Professor, Department of Horticulture, 316 Plant Science Building, University of Arkansas, Fayetteville, AR 72701, mlworthi@uark.edu

Co-Principal Investigators

John R. Clark, Distinguished Professor, Department of Horticulture, 316 Plant Science Building, University of Arkansas, Fayetteville, AR 72701, jrclark@uark.edu

Renee Threlfall, Research Scientist, Institute of Food Science and Engineering, 2650 N. Young Ave., University of Arkansas, Fayetteville, AR 72704, rthrelf@uark.edu

Mason Chizk, Graduate Research Assistant, Department of Horticulture, 316 Plant Science Building, University of Arkansas, Fayetteville, AR 72701 tmchizk@uark.edu

Abstract

Muscadine grapes are a popular specialty crop in the southern United States but market expansion is limited by consumer acceptance of certain texture attributes, including thick skins and soft gummy flesh. The development of instrumental techniques that are well correlated with sensory perception of texture attributes and breeders' ratings would allow for more breeding selections to be objectively evaluated for texture each season. In this study we evaluated seven muscadine cultivars and breeding selections of varying texture and a table grape check using trained descriptive sensory analysis, breeders' ratings, and instrumental texture assessment techniques with a variety of probe attachments and protocols. A comparison between sensory and instrumental techniques was conducted to evaluate the role of low-cost instrumental techniques in predicting sensory attributes as they relate to perceived consumer preferences. The muscadine genotypes differed significantly for most instrumental and sensory attributes measured and many correlations were found between the sensory and instrumental measurements. Based on the 2019 data, the best approach for measuring texture on a large number of breeding genotypes appeared to be combining a single breeder rating for texture quality and an instrumental protocol with a 2 mm puncture probe to estimate work to rupture and elasticity. This approach would provide an adequate prediction of sensory characteristics including awareness of skins, crispness, detachability, hardness, and visual separation of skins from flesh that are all important for consumer acceptance.

Introduction

Muscadine grapes (*Vitis rotundifolia* Michx.) are a disease-resistant specialty crop native to the southern United States. Muscadines have a loyal consumer base, but some fruit properties including thick skins, seedy pulp, and unstable aromas and flavors need improvement for successful market expansion. Texture is among the most important quality attributes for fresh-market grapes and has been studied extensively in *V. vinifera* grapes, which have a thin and tender skin that break down easily when chewed and adheres to the firm and meaty pulp (Sato et al. 1997; 2006). In contrast, muscadines typically have a thick, leathery skin, which slips from the soft pulp. While many consumers who grew up eating muscadine grapes discard the skins and/or appreciate the unique texture of this fruit, the soft pulp and leathery skin of many cultivars can be off-putting to consumers unfamiliar with muscadines. In fact, a recent consumer sensory study at the

University of Florida showed that even consumer panelists familiar with muscadine grapes preferred thinner skins and concluded that breeding for thinner skins could increase the marketability of fresh-market muscadine grapes (Brown et al. 2016).

Developing new cultivars with improved flesh and skin texture is a major objective of the University of Arkansas and University of Georgia muscadine breeding programs. Selection for improved texture has already been successful; several newer cultivars such as 'Supreme' and 'Lane' have firmer flesh and more tender skins compared to older cultivars like 'Scuppernong' and processing types such as 'Carlos' and 'Noble' (Conner 2013). Both breeding programs have newer selections in the pipeline with even better texture than can be found in existing cultivars (Conner 2013, Barchenger 2015). Breeders initially characterize most new selections with quick sensory assessments in the field and ratings on a 1-9 scale. While these measures are quick and helpful, they are also subjective. Objective measurements of texture are helpful for supporting cultivar release decisions and choosing parents. Objective, quantitative measurements of fruit texture attributes could also be used to identify quantitative trait loci and molecular markers associated with thin skin or firm flesh that could be used in breeding programs to discard seedlings with poor texture or fast-track parents with superior texture alleles.

Berry texture characteristics are often assessed using universal testing machines (UTM) that produce force deformation curves by taking precise measurements of force, time, distance, and deformation (Harker et al. 1997; Rolle et al., 2012). Penetration and compression tests are the most common tools used to assess fruit texture. In penetration tests, the arm of the texture analyzer moves down the berry to penetrate the skin and/or pulp to a fixed distance, while in compression tests the arm with attached implement compresses the whole berry and seed. Because of their slip-skin texture and large seeds, penetration tests have been used to measure muscadine firmness far more than compression tests.

Co-PI Conner (2013) used penetration tests with 2mm and 5mm flat cylindrical probes to evaluate a range of skin and flesh texture attributes in 26 muscadine cultivars and selections. Penetration tests of whole berries were used to measure berry deformation at first peak (mm) and berry maximum force (N) and to calculate berry penetration work (mJ). Fruit with a portion of the skin removed and a section of skin placed 1-mm thick polypropylene stage into which a 6-mm hole had been drilled were used to assess flesh maximum force (N) and skin break force (N) respectively. Firm fruit with tender skins are expected to have smaller berry deformation at first peak and lower berry maximum force than soft fruit or fruit with a tougher skin (Sato and Yamada, 2003). Conner (2013) found significant variation in muscadine berry texture for all attributes evaluated. As expected, older cultivars such 'Scuppernong' and 'Thomas' had higher berry deformation at first peak and lower flesh maximum force than firm fleshed cultivars like 'Lane' and newer breeding selections. Some newer selections were identified with a skin break force equivalent to *V. vinifera* (Conner 2013). Barchenger et al. (2015) also found a two-fold variation in berry maximum force among 17 muscadine genotypes in a similar study performed at the University of Arkansas.

Based on these results, Conner (2013) selected berry penetration work and flesh maximum force as the most useful attributes to measure for routine screening in breeding programs. Though skin break force seemed to be a useful measurement, it was too labor intensive to recommend for routine screening. Furthermore, the positive correlation between skin break force and berry

deformation at first peak suggested that whole berry penetration tests were also a useful measure of skin tenderness or friability. Still, the time required to individually measure twenty berries per sample with penetration makes this method impractical for assessments of large segregating populations. Furthermore, other methods may better approximate human chewing of muscadine skins than penetration with a small flat probe.

The Kramer shear cell is the most frequently used method for measuring the shear or extrusion properties of fruit tissue and may be a useful replacement or complement to penetration tests in muscadine grape (Harker et al. 1997). Shear is a strain in the structure of a substance, produced by pressure when its layers are laterally shifted in relation to each other. The Kramer shear cell simulates a single bite and provides information about bite characteristic, crispiness and firmness. The Kramer shear cell is a multi-bladed fixture that can be attached to a universal testing device to measure compression, bulk shear, and extrusion forces for samples with irregular shapes and sizes. The shear cell consists of a small box with a grated base that is filled with a fixed amount of berries or other specimens. As five parallel blades move downward through the box at a constant speed, the berries are first compressed, then extruded, and finally sheared as the blades penetrate the bottom slots. The forces needed for the blades to move through the box relate to berry texture. The Kramer shear cell has been used to characterize fruit crispness in other small fruit including blueberry and raspberry (Sousa et al. 2005; Chiabrando et al. 2009). Shear cell measurements have also recently been adopted by the table grape community. Team members of the USDA-NIFA Specialty Crops Research Initiative funded project VitisGen II are using Kramer shear cells to macerate grape berries twice and calculate gumminess, chewiness, and springiness of each cluster and applying these results for QTL mapping (Naegele, personal communication).

Materials and Methods

Plant Materials and Harvest

Three advanced selections from the University of Arkansas breeding program (AM-9, AM-135, and AM-195), one breeding selection from the North Carolina State University muscadine breeding program (NC67AO15-26), and three commercially available muscadine cultivars (‘Carlos’, ‘Ison’, and ‘Tara’) were used for analysis in 2019 on the basis of their diverse texture characteristics and availability of ripe fruit. Ten 500 g clamshells of each cultivar were harvested from the University of Arkansas Fruit Research Station (FRS) in Clarksville, AR on 11 Sept. 2019. Additionally, ten 500 g clamshells of the bunch-grape cultivar, ‘Red Globe’, were purchased on 10 Sept. 2019 to provide a check representing ideal table-grape texture qualities for reference during the analysis. Depending on availability, analyses of all these genotypes will be repeated in 2020. If unavailable, replacement genotypes will be selected to represent a comparable range of texture qualities.

Randomization

After harvest was complete, fruit was transported from FRS to the Department of Food Science in Fayetteville, Arkansas. Fruit from each genotype was mixed and re-sorted into seven 0.5 kg clamshells which were randomly assigned to the five analytical texture analysis methods and sensory analysis (two clamshells). Immature and overripe fruit and any berries that displayed obvious deformity, wet picking scar, or other damage were discarded during randomization.

Descriptive Sensory Analysis

Descriptive sensory analysis of the muscadine genotypes was conducted at the Sensory Research and Consumer Center in the Food Science Department at the University of Arkansas on September 12, 2019. The descriptive panelists developed a fresh-market muscadine lexicon of sensory terms in 2017 (Felts et al., 2018). This lexicon, with slight modifications to the "thickness of skins" attribute (Table 1), was implemented for sensory analysis in 2019. The panelists (n=9) used a modified Sensory Spectrum® method, an objective method for describing the intensity of attributes in products using references for the attributes. The descriptive panel evaluated each sample for 10 texture attributes (Table 1) using a 15-point scale (0=less of an attribute, 15=more of an attribute). The descriptive sensory evaluation was performed in duplicate with randomized presentation order of each of the eight genotypes within each replication.

Breeders' Ratings

PI, Margaret Worthington, and Co-PI's Renee Threlfall and Mason Chizk rated all breeding selections and check cultivars for skin texture, flesh texture, and overall texture desirability on a 1-9 scale, with 1 = thick skin, soft, mucilaginous flesh, or undesirable texture, and 9 = thin, crisp skins, firm, meaty, non slipskin flesh, or desirable overall texture respectively.

Instrumental Analysis

All instrumental analyses were performed using a TA.XTPlus Texture Analyzer (Texture Technologies Corporation, Hamilton, MA) with a 5 kg load cell. The specifications for each protocol are recorded in Table 2 and accessories used for each analysis are shown in Fig. 1. Fifteen randomly selected berries were used for each genotype of each type of analysis, except for the Kramer shear cell, which consisted of three runs per genotype and six berries per run.

Penetration rupture force was measured using a 2 mm flat cylindrical probe. Penetrations were made on the equatorial plane of each berry with the stem scar facing the right-hand side at a probe speed of 1 $mm.sec^{-1}$ (Table 2). Rupture force (N) was calculated as the force required to rupture the berry skin. Elasticity was calculated as the distance (mm) the berry was compressed before the skin was ruptured. Berry penetration work (mJ) was calculated as the area under the curve from zero to the point of berry maximum force following Conner (2013).

Skin and flesh analysis. To evaluate skin and flesh properties individually, a small circular section of skin was carefully removed from the equatorial surface of each berry with the stem scar facing the right-hand side using a razor blade. The removed sections of skin were trimmed of any excess flesh clinging to the interior surface and probed using a 2 mm flat cylindrical probe. The distance traveled from first contact to the work surface was recorded as skin thickness. The area under the curve (mJ) was recorded as skin work and the peak force (N) was recorded as skin hardness. The exposed flesh of the entire berry was probed using an 8 mm flat cylindrical probe. The probe traveled 3 mm at a speed of 0.5 $mm.sec^{-1}$ after first contact, and the peak force (N) was recorded as flesh firmness.

Compression. Compression tests were performed using a 10 mm flat cylindrical probe and were conducted on the equatorial plane of each berry with the stem scar facing the right-hand side at a probe speed of 1 $mm.sec^{-1}$ (Table 2). After the probe contacted the berry surface, it traveled

halfway to the work surface, achieving a strain of 50%. Peak force (N) and compression work (mJ) were recorded as maximum force and work to 50% strain respectively. The force (N) and distance (mm) at the first detected peak were recorded as rupture force and elasticity respectively. The elasticity divided by the total berry width, multiplied by 100 was recorded as the percent strain to rupture.

Single blade. Single knife blade tests were performed using a knife blade attachment with a 45 degree chisel end and were conducted on the equatorial plane of each berry with the stem scar facing the right-hand side at a probe speed of 1 $mm.sec^{-1}$ (Table 2). After the probe contacted the berry surface, it traveled halfway to the work surface, achieving a strain of 50%. Peak force (N) and total work (mJ) were recorded as maximum force and work to 50% strain respectively. The force (N) and distance (mm) at the first detected peak were recorded as rupture force and elasticity respectively. The elasticity (mm) divided by the total berry width (mm), multiplied by 100 was recorded as the percent strain to rupture.

Kramer shear cell. Shear tests were performed using a Kramer shear cell (KSC) (TA-91). The box at the cell base was filled with six berries. Each sample was macerated in two cycles with a probe speed of 1 $mm.sec^{-1}$ (Table 2). While the first maceration cycle of the Kramer shear cell yielded data representative of compressing and shearing a berry, the berries of smaller genotypes were extruded through the slots in the base, resulting in a load cell overload during the second cycle. Due to this complication, the second cycle measurement was discarded for the two smallest muscadine genotypes, 'Carlos' and NC67AO15-26. Maximum forces (N) for cycles 1 and 2 were recorded as maximum force (cycle 1) and force (cycle 2) respectively. Similarly, the work (mJ) associated with each cycle was recorded as work (cycle 1) and work (cycle 2). The difference between force and work values between cycles was recorded as change in force and change in work respectively.

Statistical Analysis

Instrumental and breeders' ratings were analyzed using PROC GLM in SAS 9.4 (SAS Institute Inc., Cary, NC) with genotype as a fixed effect. Sensory texture data were also be analyzed using PROC GLM with genotype considered a fixed effect and panelist and genotype x panelist interaction considered random effects. In all analyses, year and year interactions will be considered as random effects. Mean separation for significant factors were estimated using Fisher's F-protected Least Significant Difference and were conducted at the 95% confidence level. PROC CORR was used to conduct Pearson correlations between the instrumental, sensory, and breeders' ratings data.

Results and Discussion

Descriptive Sensory Analysis and Breeder's Ratings

The eight genotypes differed significantly ($p < 0.05$) for all attributes measured in 2019 by breeder's ratings and the descriptive sensory panel, except for moisture release, which was excluded from all further analysis (Table 3). Differences in ratings ($p < 0.05$) existed between panelists who participated in the sensory analysis, but there were no detectable differences between breeders for the breeder's ratings. The *V. vinifera* check, 'Red Globe', was a frequent

extreme, yet expected outlier from among most attributes measured and was, therefore, not included in ANOVA or correlation analyses.

The mean breeder's ratings for skin texture and overall desirability were comparable, with AM-135 and AM-195 both being significantly different ($p < 0.05$) from all other muscadine genotypes (Table 3). These genotypes had skin and desirability ratings closest to 'Red Globe', indicating that their skin attributes may be promising for table use. None of the muscadine genotypes had flesh texture qualities statistically similar to those of 'Red Globe', indicating that all of the muscadines were gummy in comparison to the table grape check. For all three breeder attributes, 'Carlos' and NC67AO15_26 performed similarly and were rated significantly lower than all other genotypes (Table 3). Interestingly, the all breeders' ratings were most correlated ($p < 0.01$) with KSC work in cycle 1, although they were also correlated with four other instrumental methods (Table 4).

Among the nine sensory panel attributes for which genotypic differences were observed, visual separation and detachability displayed the widest ranges of variation between muscadine genotypes according the 15 point scale (Table 3). The genotypic means of these attributes were also correlated to one another ($p < 0.01$), with AM-135 and AM-195 both having skins that were 25-40% less detachable or prone to separation than the other five muscadine genotypes. Even so, the detachability and visual separation scores of these genotypes were twice as large as those of 'Red Globe', suggesting that despite the wide range of skin adherance, all muscadines measured are much more slip-skin than the table grape check. Detachability and visual separation were also both positively correlated with elasticity ($p < 0.01$) and negatively correlated with flesh firmness ($p < 0.01$), indicating that genotypes with softer, gummier flesh tend to slip from skins more easily (Table 4).

Mean seed separation was similar between most genotypes, including 'Red Globe', with 'Carlos' and 'Ison' being the only exceptions. Both of these cultivars had seeds that were approximately 30% more difficult to separate from the pulp of the berry than the other genotypes (Table 3). 'Carlos' is a processing cultivar so the poor seed separation from flesh is less likely to impact consumer acceptance. AM-9 had the largest mean seed size while AM-135 had the lowest. AM-195 and 'Red Globe' both had about one less seed on average than all other genotypes except for AM-9. For berry crispness, hardness, and awareness of skins, the genotypic means of 'Red Globe' were much lower than all muscadine genotypes, indicating that there is large gap between muscadines and table grapes for these attributes. However, 'Carlos' and NC67AO15_26 had lower hardness and crispness ratings than most other muscadine genotypes. The genotypic means of crispness and hardness were generally correlated ($p < 0.01$) with one another, suggesting either that the sensory panel had difficulty distinguishing these two traits, or that this may be an artifact of the particular genotypes surveyed. AM-9 and AM-195 were the hardest and the most crisp genotypes, respectively. Both hardness and crispness reflected strongly positive correlations with compression work to 50% strain ($p < 0.05$) (Table 4). Ignoring 'Red Globe', few significant differences existed between mean ratings for awareness of skins. However, AM-135 did have significantly lower ratings compared to all other muscadine genotypes, suggesting that this genotype may be particularly desirable for table use or parental stock for improved texture. This is somewhat surprising though, as this genotype also had

relatively thick skin, potentially suggesting that firm flesh texture and skin adherence to flesh may impact awareness of skins more than skin thickness directly.

Texture Analysis

Penetration and flesh analysis. Muscadine genotypic means were much larger than those of 'Red Globe' for all attributes measured by the 2 mm puncture probe (Table 5). The thinnest-skinned muscadine, NC67AO15_16, had skin that was about 30% thinner than the thickest skinned, AM-9, but twice as thick as 'Red Globe'. Despite being the thinnest skinned muscadine, NC67AO15_26 had the largest rupture force, skin hardness, elasticity, work to rupture, and skin work. In contrast, AM-195 had the softest skins, as indicated by mean values for both skin hardness and work (Table 5). AM-135 had the lowest rupture force and elasticity, being only 67% and 78% of genotypic mean values for NC67AO15_26 respectively. Penetration elasticity was negatively correlated ($p < 0.01$) with the breeders' rating for skin texture and positively correlation ($p < 0.01$) with visual separation and detachability (Table 4). Skin thickness was positively correlated with berry hardness and the breeders' rating for flesh texture ($p < 0.01$). The correlation between flesh texture and skin thickness was unexpected and may represent a bias presented by skin attributes when breeders rate flesh quality in the field. Additionally, the flesh analysis conducted with the 8 mm probe demonstrated that AM-195 and AM-135 had the firmest flesh, while 'Carlos' and NC67AO15_26 had the softest flesh (Table 5).

Compression and single blade analysis. The means of characters measured in the compression and single blade analysis were similar, as evidenced by multiple high correlations among the attributes measured by these instruments (data not shown). For this reason, the results of the single-blade analysis were excluded from the correlation analysis. In both analyses, 'Carlos' consistently demonstrated the lowest rupture force, maximum force, and elasticity (Table 5). Work, which was tightly correlated ($p < 0.01$) with hardness and crispness (Table 4), was consistently highest for AM-9 and 'Carlos' (Table 5). The genotypic means of maximum force and rupture force were also significantly correlated ($p < 0.05$) with crispness and hardness, indicating that the sensory panel's interpretation these traits was comparable the instrumental compression technique (Table 4).

Kramer shear cell analysis: For the KSC analysis, change in work was the only character measured that did not differ across genotypes and was thus excluded from correlation analysis (Table 5). In addition, 'Carlos' and NC67AO15_25 were removed from the analysis of maximum force (cycle 2), work (cycle 2), and change in force due to cell overloads resulting from small fruit size. These three characteristics were removed from subsequent correlation analysis due to the resulting imbalance. Work in cycle 1, which was correlated with crispness, hardness, and skin thickness ($p < 0.01$), was shown to be highest in AM-135, but lowest in 'Carlos' (Tables 4 and 5). Additionally, AM-135 was had the largest negative change in force between cycles, suggesting that it was also one of the easiest muscadines to macerate (Table 5).

Conclusions

The present work demonstrated that all sensory attributes measured, excluding seed attributes, could be related to at least one instrumental characteristic. Additionally, an apparent relationship

between breeders' ratings and the sensory attributes of hardness, crispness, and detachability suggests that breeders may adequately assess these characteristics without need for sensory panels or instrumental techniques. However, the lack of any distinguishable relationship between these characteristics and skin awareness highlights the necessity of instrumentation in accurately assessing this trait. Because this trait is thought to be important to shaping consumer opinion, it may be necessary to screen for awareness of skins with the 2 mm puncture probe (work to rupture), with which it was most highly correlated ($r = 0.79$).

Another potential drawback to using breeders' ratings exclusively is that they are all highly correlated with one another and the ratings for flesh appear to be biased by the qualities contributed by the skin, such as thickness and hardness. This limitation suggests that a single breeder's rating variable may be sufficient for estimating skin attributes instrumental techniques should be adopted for measuring flesh attributes. If one was already screening for awareness of skins with the 2 mm probe, as previously recommended, the puncture elasticity would be an excellent predictor of flesh texture. Puncture elasticity was strongly correlated with flesh firmness measured by the 8 mm probe ($r = -0.97$).

Lastly, quantification the slip-skin nature of muscadines, which was assessed by the sensory attributes of detachability and visual separation, could be accurately assessed using the 2 mm puncture probe as well. Both of these characteristics appear to be linked with flesh characteristics and were highly correlated ($p < 0.01$) with the elasticity determined by the 2 mm probe.

Of all muscadine genotypes included in this study, AM-135 appeared to have characteristics that are most desirable for table use. This cultivar was easily macerated by the KSC, required little work to rupture, and had flesh that was firm. AM-135 also received the lowest sensory panel ratings for awareness of skins, despite having relatively thick skins. These qualities contrast heavily with those of 'Carlos' and NC67AO15-26, which appear to be much more suited to processing than table-use.

In summary, for a more holistic characterization of fruit texture in muscadine breeding programs, we recommend the adoption of a combined approach that utilizes a single breeder rating for texture quality and an instrumental protocol that implements the 2 mm puncture probe to estimate work to rupture and elasticity. This approach would provide an adequate prediction of sensory characteristics including awareness of skins, crispness, detachability, hardness, and visual separation. An estimation of fibrousness would require implementation of another attachment such as the 3" cylinder, but because this trait alone is not considered to be of critical importance, inclusion may not be justified. Furthermore, additional work may be done to develop a more predictive, cost-effective method for assessing seed separation, which is likely to affect consumer acceptance.

Literature Cited:

Barchenger, D.W., J.R. Clark, R.T. Threlfall, L.R. Howard, and C.R. Brownmiller. 2015. Evaluation of physiochemical and storability attributes of muscadine grapes (Vitis rotundifolia Michx.). HortScience 50(1):104-111.

Brown, K., C. Sims, A. Odabasi, L. Bartoshuk, P. Conner, and D. Gray. 2016. Consumer acceptability of fresh-market muscadine grapes. J. Food Sci. 81.

Chiabrando, V., G. Giacalone, L. Rolle. 2009. Mechanical behaviour and quality traits of highbush blueberry during postharvest storage. J. Sci. Food Agric. 89: 989–992.

Conner, P.J. 2013. Instrumental textural analysis of muscadine grape germplasm. HortSci. 48:1130-1134.

Felts, M., R.T. Threlfall, J.R. Clark, and M.L. Worthington. 2018. Physiochemical and descriptive sensory analysis of Arkansas muscadine grapes. Hortscience 53:1570-1578.

Harker. F.R., R.J. Redgewell, I.C. Hallett, and S.H Murray. 1997. Texture of fresh fruit. Horticultural Reviews. 20:121-224.

Rolle, L., R. Siret, S. Rio Segade, C. Maury, V. Gerbi, and F. Jourjon. 2012. Instrumental texture analysis parameters as markers of table-grape and winegrape quality: A review. Amer. J. Enol. Viticult. 63:11–27.

Sato, A. and M. Yamada. 2003. Berry texture of table, wine, and dual-purpose grape cultivars quantified. HortScience 38:578–581.

Sato, A., H. Yamane, N. Hirakawa, K. Otobe, and M. Yamada. 1997. Varietal differences in the texture of grape berries measured by penetration tests. Vitis-Geilweilerhof 36:7-10.

Sato, A., M. Yamada, and H. Iwanami. 2006. Estimation of the proportion of offspring having genetically crispy flesh in grape breeding. J. Amer. Soc. Hort. Sci. 131:46-52.

Sousa, M.B., W. Canet, M.D. Alvarez, and M.E. Tortosa. 2005. The effect of the pre-treatments and the long and short-term frozen storage on the quality of raspberry (cv. Heritage). Eur. Food Res. Technol. 221:132-144.

Table 1. 2019 Lexicon Muscadine Grapes for Texture (each panelist receives 5 berries).

Term	Definition	Technique	Reference	
Appearance (pulp of one berry cut in half)				
Visual separation	Detachability of pulp from skin of berry	Squeeze half of berry and observe the extent of which the pulp detaches from the skin. (None=*does not detach* to Much=*completely detaches*)	None=0 Much=15.0	
Amount of seeds	Number of seeds in the whole berry	Count the number of seeds in the whole berry.	Number of seeds	
Seed size	Visual size of the seeds	Observe the seeds and determine the overall size. (Small to Large)	Photo reference of size A=12 (5.3 x 8.5 mm) B=7 (4.9 x 7.1 mm) C=3 (3.9 x 6.1 mm)	
Texture (whole berry for 4 berries)				
Berry hardness	Force required to compress the sample.	Place the sample in the mouth. Compress or bite through the sample one time with molars or incisors. (Soft to Hard)	Cream Cheese[1] Egg White Am Cheese Beef Frank Olive Peanut Almond	1.0 2.5 4.5 5.5 7.0 9.5 11.0
Berry crispness	Unique, strong, clean, and acute sound produced in first bite of the food with incisors and open lips.	Place the sample in the mouth. Compress or bite through the sample one time with molars or incisors. Evaluate the sound intensity produced at the first bite. (None=*not crisp* to Much=*extremely crisp*)	Ripe Banana[2] Granny Smith Apple Carrot	0.0 7.5 15.0

[1] Philadelphia cream cheese, cut into ½" cubes (Kraft, Chicago, IL); Egg White, jumbo eggs, boiled for 5 minutes, cut into ½" cubes; American cheese, cut into ½" cubes (Boars Head, Brooklyn, NY); Hebrew National beef frank, boiled for 5 minutes and cut into ½" slices (ConAgra Foods, Indianapolis, IN); Great Value queen olives, with pimentos removed (Walmart, Bentonville, AR); Planters peanuts, whole pieces (Kraft, Chicago, IL); Almonds were not used for this evaluation

[2] Ripe banana, cut into ½" cubes; Granny smith apple, peeled and cut into ½" cubes; Carrot, peeled and cut into ½" cubes

Moisture release	Amount of wetness or moistness felt in the mouth after one bite or chew.	Compress the sample with molars one time only. (Dry to Wet)	Banana[3] Carrot Mushroom Snap Beans Cucumber Apple Honeydew Orange (Chew refs 5 times)	1.0 2.0 4.0 7.0 8.0 10.0 12.0 15.0
Awareness of skins	How aware are you of the skins during mastication of the sample?	Place sample in mouth and chew 3-5 times. Can also be evaluated in first bite stage. (None=*cannot tell skins are there* to Much=*extremely aware of skins*)	Baked Beans[4] Medium Lima Beans Edamame	4.0 8.0 15.0
Detachability	Ease with which the pulp separates from the skin of the berries	Place the sample in the mouth. Compress or bite through the sample one time with molars or incisors. Evaluate the ease that the pulp separates from the skin. (None=*does not detach* to Much=*completely detaches*)	None=0.0 Much=15.0	
Fibrousness between teeth	Amount of grinding of fibers required to chew through the sample (not including skins)	Place sample between molars and chew 3-5 times. Evaluate during chewing, but ignore the skin. (None=*not fibrous at all* to Much=*extremely fibrous*)	Apple[5] Apricot Salami Celery Toasted Oats Bacon Beef Jerky	2.0 5.0 7.0 9.0 10.0 12.0 20.0

[3] Ripe banana, cut into ½" cubes; Carrot, peeled and cut into ½" cubes; Button mushrooms, destemmed and cut into ½" cubes; Snap beans were not used for this evaluation; Cucumber, peeled, deseeded, and cut into ½" cubes; Pink lady apple, peeled and cut into ½" cubes; Honeydew, peeled and cut into ½" cubes; Dole mandarin orange piece (Dole Foods, Westlake Village, CA)

[4] Bush's baked beans (Bush Brothers and Company, Knoxville, TN); Medium lima beans; Edamame in pods

[5] Pink lady apple, peeled and cut into ½" cubes; Mariani apricots, sliced in half (Mariani, Vacaville, CA); Hard salami, cut into ½" cubes (Boars Head, Brooklyn, NY); Celery, cut into ½" pieces; Oats, toasted for 5 minutes at 350 F; Bacon and beef jerky were not used for this evaluation

Seed separation	The ease with which the seeds separate from the pulp of the berry	Manipulate the pulp in the mouth for ease to separate seeds from pulp. (None=*hard to separate seeds from pulp* to Much=*seeds easily separate from pulp*)	None=0.0 Much=15.0

Table 2. TA.XTPlus texture analyzer muscadine project specifications.

Method	Part #	Part Description	Test Speed (mm/s)	Target Distance	Trigger Force (N)	Tare Height (mm)
Puncture Analysis	TA-52	2 mm puncture	1	9 mm	0.07	35
Skin Thickness	TA-52	2 mm puncture	0.2	100% strain	0.07	5
Flesh Firmness	TA-58	8 mm puncture	0.5	3 mm	0.07	35
Compression Analysis	TA-30	3” cylinder	1	50% strain	0.07	35
Single Blade Shear	TA-42	45° chisel knife	1	50% strain	0.07	35
Kramer Shear Cell (2 cycles)	TA-91	Kramer Shear Cell	1	35 mm	2 N	Above the cell

Table 3. Least square means of breeders' ratings and descriptive sensory panel attributes in 2019.

Genotype	Breeders Ratings			Descriptive Sensory Panel (Visual)			Descriptive Sensory Panel (Texture)					
	Flesh Texture	Skin Texture	Overall Desirability	Visual Separation	Amount of Seeds	Seed Size	Awareness of skins	Berry crispness	Detachability	Fibrousness	Berry hardness	Seed separation
	---------- 1-9 scale ----------			-0-15 scale-	--- # ---	0-15 scale	----------0-15 scale ----------					
AM-135	6.67	8.00	8.00	9.08	3.50	3.96	12.06	7.00	8.50	4.27	6.81	8.78
AM-195	6.00	8.00	7.67	7.33	2.72	4.38	13.17	7.47	8.17	4.11	7.14	9.64
AM-9	6.00	5.33	5.67	12.36	3.00	6.11	13.82	7.08	13.81	4.92	7.36	9.30
CARLOS	2.00	2.33	2.00	13.41	3.66	5.31	13.21	5.36	13.66	4.36	5.91	9.31
ISON	5.00	5.67	5.33	12.80	3.66	5.72	13.81	7.33	14.11	5.64	7.01	6.31
NC67AO15_26	2.33	2.00	2.33	14.06	3.99	4.25	13.78	6.11	14.03	5.49	6.06	6.75
RED GLOBE†	8.00	9.00	8.67	3.75	2.78	4.28	6.29	2.92	4.11	2.72	3.72	9.75
TARA	5.33	6.00	6.00	12.94	3.33	4.75	13.51	6.58	13.47	5.01	6.81	9.08
LSD	1.22	1.22	1.32	2.14	0.51	0.90	0.84	1.16	2.16	0.71	0.44	2.17

† Excluded from significance testing and LSD calculations due to bias.

Table 4. Pearson (r) correlation table of instrumental and sensory texture analysis methods used to survey 7 muscadine genotypes in 2019.

			Breeder ratings			Sensory panel (visual)			Sensory panel (texture)					
Trait			Flesh	Skin	Overall	Vis. Sep.	Seed number	Seed Size	Aware. Skin	Crispness	Detachabiliy	Fibrousness	Hardness	Seed Sep.
			1-9 scale			0-15 scale	#	0-15 scale	0-15 scale					
2 mm puncture probe	Rupture F.	---N---	-0.57	-0.54	-0.55	0.24	0.09	0.10	0.68	-0.16	0.31	0.32	-0.27	-0.31
	Skin hardness		-0.66	-0.75	-0.73	0.71	0.89 **	0.16	0.25	-0.51	0.60	0.60	-0.60	-0.72
	Elasticity	--mm--	-0.72	-0.89 **	-0.86 *	0.96 **	0.53	0.53	0.75	-0.62	0.96 **	0.65	-0.47	-0.36
	Skin thickness		0.93 **	0.81 *	0.83 *	-0.47	-0.69	0.29	-0.20	0.75	-0.32	-0.21	0.91 **	0.39
	Work to rup.	--mJ--	-0.78 *	-0.83 *	-0.82 *	0.63	0.32	0.29	0.79 *	-0.48	0.66	0.48	-0.47	-0.33
	Skin work		-0.67	-0.76 *	-0.74	0.72	0.89 **	0.17	0.26	-0.51	0.62	0.61	-0.60	-0.73
7.6 cm cylinder compression	Rupture F.	---N---	0.72	0.52	0.56	-0.32	-0.66	0.49	0.25	0.81 *	-0.11	0.09	0.92 **	0.09
	Max F.		0.87 *	0.77 *	0.80 *	-0.65	-0.79 *	0.19	-0.04	0.92 **	-0.47	-0.18	0.95 **	0.24
	Elasticity	--mm--	0.56	0.29	0.35	0.11	-0.41	0.61	0.46	0.58	0.30	0.40	0.78 *	-0.02
	Work (50% strain)	--mJ--	0.91 **	0.83 *	0.85 *	-0.64	-0.82 *	0.19	-0.05	0.93 **	-0.45	-0.18	0.98 **	0.28
	Comp. to rup.	---%---	-0.17	-0.46	-0.38	0.70	0.30	0.52	0.76 *	0.00	0.77 *	0.84 *	0.10	-0.52
8 mm probe	Flesh firmness	---N---	0.70	0.85 *	0.84 *	-0.98 **	-0.60	-0.58	-0.65	0.65	-0.97 **	-0.63	0.48	0.37
Kramer Shear Cell	KSC max F. (cycle 1)	---N---	0.77 *	0.66	0.68	-0.37	-0.29	0.26	-0.11	0.85 *	-0.24	0.16	0.80 *	-0.23
	KSC work (cycle 1)	--mJ--	0.96 **	0.88 **	0.90 **	-0.59	-0.54	0.07	-0.29	0.91 **	-0.47	-0.10	0.90 **	0.09

*, **, Significant at 0.05, and 0.01 probability levels respectively. 'Red Globe' excluded due to expected bias. 45 ° chisel knife measurements excluded due to high correlation with compression measurements.

Table 5. Least square means of instrumental texture attributes in 2019.

	2 mm Puncture Probe						7.6 cm Cylinder					8 mm Puncture Probe
Genotype	Rupture force	Skin hardness	Elasticity	Skin thickness	Work to rupture	Skin work	Rupture force	Maximum force	Elasticity	Work to 50% strain	Percent strain to rupture	Flesh firmness
	----------N----------		----------mm---------		----------mJ----------		----------N----------		--mm--	---mJ---	---%---	----N----
AM-135	7.22	30.77	6.28	1.43	19.47	6.32	27.79	35.57	7.97	212.40	32.32	2.12
AM-195	10.03	24.25	6.30	1.36	27.82	5.00	34.31	44.27	8.08	276.33	31.13	2.35
AM-9	9.09	30.48	7.71	1.50	29.59	6.29	44.48	44.87	11.22	272.65	41.15	1.28
CARLOS	9.26	33.15	7.84	1.14	31.25	6.85	17.11	17.11	6.71	82.83	33.00	1.00
ISON	9.23	33.53	7.43	1.36	28.22	6.95	34.33	35.65	9.50	220.61	38.25	1.28
NC67AO15_26	10.67	36.43	7.99	1.04	35.55	7.53	22.27	22.34	7.77	103.19	40.11	1.18
RED GLOBE†	2.89	6.28	5.50	0.51	7.64	1.34	15.82	15.91	10.30	86.34	46.70	1.92
TARA	8.33	27.03	7.52	1.40	26.82	5.61	25.85	28.55	9.49	189.78	36.52	1.35
LSD	0.83	3.29	0.55	0.17	4.44	0.67	9.33	9.41	1.16	39.59	4.86	0.28

† Excluded from significance testing and LSD calculation due to bias.

Table 5. (Continued).

	45 ° Chisel Knife					Kramer Shear Cell				
Genotype	Rupture force	Maximum force	Elasticity	Work to 50% strain	Percent strain to rupture	Maximum force (cycle 1)	Maximum force (cycle 2) ‡	Change in force ‡	Work (cycle 1)	Work (cycle 2) ‡
	----------N----------		---mm---	---mJ---	----%----	----------N----------			----------mJ----------	
AM-135	23.57	23.96	9.82	127.44	40.19	225.20	180.86	-44.34	3350.07	1377.85
AM-195	27.17	28.17	9.53	160.47	38.69	200.17	209.01	8.84	2946.89	1502.95
AM-9	31.25	31.25	12.96	165.83	46.75	221.59	297.85	76.26	3151.29	2180.39
CARLOS	13.20	13.20	7.69	60.59	38.02	154.78	-	-	1705.11	-
ISON	32.72	32.72	11.75	160.62	48.00	241.43	234.14	-7.29	3086.12	1993.53
NC67AO15_26	17.30	17.30	8.77	77.19	43.49	169.11	-	-	1941.13	-
RED GLOBE†	10.51	10.51	10.64	54.43	47.44	82.26	42.58	-39.68	1309.93	508.40
TARA	20.70	21.00	11.04	126.32	42.63	181.29	227.34	46.06	2665.49	1823.82
LSD	5.09	4.96	0.87	24.86	3.69	44.66	57.93	65.69	534.56	524.66

† Excluded from significance testing and LSD calculation due to bias.

‡ Carlos and NC67AO15_26 excluded due to complications from small size.

Figure 1. TA.XTPlus Texture Analyzer attachments including (a) 2 mm puncture probe, (b) 8 mm puncture probe, (c) 3” cylinder, (d) 45 ° chisel knife, and (e) Kramer shear cell (KSC).