Technical Abstracts

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Viticulture—Climate and Other Environmental Effects on Grapes and Wine Session

Characterizing the Contribution of Topsoil to Mid-Atlantic Grapevine Size and Fruit Composition

Jaclyn Fiola,* Greg Evanylo, Tony Wolf, and Ryan Stewart *Virginia Tech, 342 Smyth Hall, 185 Ag-Quad Ln, Blacksburg, VA 24061 (jcfiola@vt.edu)

The ability of mid-Atlantic states to support high-quality winegrape production can be hindered by excessive plant-available water and nutrients. Such excesses can negatively influence fruit composition and potential wine quality by producing vigorous vegetative growth. A major source of water and nutrients in the soil is the topsoil: the upper horizons of the soil profile that are dark-colored and rich in organic matter. Topsoil management is a potential approach for growers to influence hydrology and fertility of vineyard soils, but very little is known about topsoil's contribution to vine growth and fruit composition, especially in the mid-Atlantic region. We investigated topsoil's contributions at two vineyards in Maryland and Virginia with different topsoil thickness while other factors (climate, vine spacing, training, management, cultivar/ clone/rootstock) were constant. First, we surveyed topsoil thickness along transects in each vineyard. Then, at each panel corresponding to the topsoil measurement locations, we analyzed vine and fruit properties. Results from the 2019 and 2020 growing seasons suggest strong relationships between topsoil and fruit titratable acidity, cluster compactness (berries per length of rachis), and vine size (dormant pruning weights) at both sites. Compared to thick topsoil, vines growing in thinner topsoil had significantly lower vine size, cluster compactness, and titratable acidity. Topsoil effects on fruit pH and Brix varied by site and year but showed some relationship between thinner topsoil and higher pH and Brix. Specific mechanisms for these effects are still being investigated, but it appears that vines growing in thinner topsoil ripen faster than vines in thick topsoil. Results from this project will provide guidance for better site selection and soil preparation in the mid-Atlantic. Our results may also help inform decisions about soil management and harvest timing in established vineyards.

Funding Support: Maryland Wine and Grape Promotion Fund, Virginia Tech Institute of Critical Technology and Applied Science

Investigation of Potential "Rose Taint" Marker Compounds in Cabernet Sauvignon

Scott Frost, Danielle Fox, Tom Collins, Markus Keller, and James Harbertson* *Tufts University, Robinson Hall 212 College Ave, Medford, MA 02155 (scot.frost@tufts.edu)

Several reports from wineries suggest that Cabernet Sauvignon wine produced from fruit harvested after an early frost will have rose-like aromas. These aromas are described as atypical by winemakers and the affected wines are termed "rose tainted" or "frost tainted." Anecdotal evidence suggests that inclusion of freeze-killed leaf material into the fermentation is the taint source. We investigated these claims by studying how adding leaf matter into the fermentation affects the chemical and sensory profile of Cabernet Sauvignon wine. Freeze-killed leaves were hand-collected from Cabernet Sauvignon must at four addition rates: 0.0, 0.5, 2.0, and 8.0 g/kg. A detailed analysis captured the treatment impact on the flavor and chemical profile of the experimental wines. The addition of freeze-killed leaf matter significantly increased the intensity of floral aroma, herbaceous/straw aroma, artificial fruit aroma, and

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floral aftertaste, while decreasing the dark fruit aroma and astringency. Gas chromatography mass spectrometry identified 66 volatile organic compounds (VOCs) that varied by treatment. Statistical analysis was then used to identify which compounds drive the changes in floral aroma, floral aftertaste, and astringency. Partial least squares, correlation, and fold-change analysis were then used to ascertain four marker VOC: 1,1,3,5-tetramethylcyclohexane, 6-methyl-5-hepten-2-ol, p-menth-1-en-9-al, and 6-methyl-3,5-heptadien-2-one. Additionally, the phenolic composition of wines fermented with added freeze-killed material showed reduced concentrations of anthocyanins, tannins, and iron-reactive phenolics. These results clearly show the impact of freeze-killed leaf matter on Cabernet Sauvignon wine and provide convincing evidence of the taint source.

Funding Support: Washington State University, Auction of Washington Wines and all Washington grapegrowers and wineries through the Washington State Wine Commission

Vineyard Row Orientation: Quality and Sensory Effects on Grape and Wine of Cabernet Sauvignon

Xuefei Wang,* Zhumei Xi, and Zhaoxiang Wang *Northwest A&F University, 22 Xinong Rd., Yangling, 712100, China (Wang.xuefei0805@gmail.com)

Different vineyard row orientations and altitudes have been adopted to maximize terroir potential. However, little is known about the impact of climatic determinates on physicochemical and aroma attributes of grapes and wines. Effects of the climate profiles associated with two row orientations on berry ripening, anthocyanins, and volatile compounds of Cabernet Sauvignon planted at two altitudes were investigated. The row orientation mainly changed the canopy climate: soil temperature, canopy temperature, and photosynthetically active radiation. The individual anthocyanins and volatile compounds in grapes were mainly determined by the vineyard locations. Microclimatically, north-south (NS) orientations promoted balanced photosynthetic performance and temperature between the two sides. Grapes from NS row orientation produced more reducing sugar and had lower titratable acidity during berry ripening. Differences in row orientation and associated microclimate manipulated the total flavonoids, individual anthocyanins, and volatile compounds in the grapes. Grapes from east-west oriented rows were more closely associated with the sensory attributes. Results obtained in this study provide new insights into the impact of row orientation in the arid Shangri-La valley and can be applicable to similar terroirs to unlock vineyard potential.

Funding Support: National Key Research and Development Program of China (2019YFD1000102-11), China Agriculture Research System for Grape (CARS-29-zp-6) and Fundamental Research Funds for the Central Universities (Z1090219009)



Viticulture—Rootstocks Session

Evaluating New Traits to Improve Rootstock Drought Tolerance

Megan Bartlett,* Gabriela Sinclair, Gabriela Fontanesi, Thorsten Knipfer, Andrew Walker, and Andrew McElrone *University of California, Davis, One Shields Ave, Davis, CA 95616 (mkbartlett@ucdavis.edu)

Breeding more drought-tolerant rootstocks is a key strategy to reduce irrigation demand and mitigate the impacts of climate change on grape yield and ripening. Identifying traits that confer drought tolerance would accelerate rootstock improvement by providing clear targets for breeding. The living cells in the root are a significant bottleneck for water uptake from dry soil, but the traits that capture the responses of these cells to water stress have never been evaluated as potential drivers of rootstock drought tolerance. We used a greenhouse drought experiment to test the relationships between these traits and vine gas exchange, water transport, and vigor across eight commercial rootstocks that field trials have classified as drought-tolerant (110R, 140-Ru, 1103P, and Ramsey) or sensitive (101-14, 420A, 5C, and Riparia Gloire) grafted to Chardonnay. Root capacitance (CAP) and turgor loss point (TLP) were significantly different across rootstocks and shifted to significantly lower values in water-stressed plants, which we would expect to improve root drought tolerance. The rootstocks with a lower CAP, which indicates the roots retained more water as root water potentials decline, maintained significantly greater stomatal conductance and photosynthesis under water stress ($r^2 = 0.31$ and 0.45, both p < 0.001). The rootstocks classified as drought-tolerant had significantly lower root CAP and TLP values under well-watered conditions, but exhibited significantly smaller shifts in TLP under water stress, indicating these rootstocks would undergo cell collapse at less negative root water potentials, contrary to our expectations. These findings suggest breeding rootstocks for a lower CAP would improve vine water uptake and gas exchange under dry soil conditions, but more work is needed to understand the function of these traits in the deep, highly heterogenous rooting environments in the field, where these rootstocks vary widely in rooting depth and access to deeper, wetter soil layers.

Funding Support: American Vineyard Foundation

Performance of Rootstocks in a Cabernet Sauvignon Vineyard with *Xiphinema index* Nematodes and Grapevine Fanleaf Virus

Rhonda Smith* and M. Andrew Walker

*University of California Cooperative Extension, 133 Aviation Blvd., Suite 109, Santa Rosa, CA 95403 (rhsmith@ucanr.edu)

Grapevine fanleaf virus (GFLV) is the causal agent of fanleaf degeneration, which significantly reduces yield due to poor fruit set. Vine decline is progressive and fruit quality is reduced; thus, diseased vineyards are replanted. *Xiphinema index* nematode acquires GFLV by feeding on roots of infected vines and remnant roots in replanted vineyards. Subsequent feeding transmits the virus, causing disease in future plantings. We evaluated the performance of grapevine rootstocks selected for broad resistance to nematodes by establishing a field trial in a replanted vineyard in 2012, one year after removal of vines which had severe fanleaf degeneration. The UCD GRN series of rootstocks and O39-16—all highly resistant to *X. index*—and medium- to low-resistant rootstocks—RS-3, RS-9, Schwarzmann, 1616C and 1103P—were planted in a randomized complete block design with eight replications of five-vine plots without pre- plant fumigation. Nematode samples were collected by plot. There was no difference in the number of *X. index* nematodes found in O39-16 and GRN rootstock plots; however, more *X. americanum* were found on GRN-1 and O39-16 than in the other

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GRN rootstocks. Grapevine fanleaf virus status was determined by ELISA from shoot tip samples. In 2019, 26 (65%) of the GRN plots and three (37%) of the O39-16 plots tested GFLV-positive and pruning weights were reduced in infected vines. In 2019, only one vine in each of three GRN-1 plots was infected with GFLV; yet from 2015 to 2019, GRN-1 consistently had lower pruning weights than the other GRN rootstocks. In 2019, mean yields of the GRN rootstocks ranged from 8.4 to 10.2 kg/vine and were not statistically different. O39-16 yield averaged 11.8 Kg/vine. At this time, it is still uncertain whether the GRN rootstocks induce tolerance to fanleaf degeneration.

Funding Support: California Grape Rootstock Improvement Commission

The Influences of Rootstock on Performance of Pinot noir (*Vitis vinifera* L.) 1. Grapevine Phenology, Vigor and Nutrients

Yipeng Chen, Pangzhen Zhang,* Yanan Fei, Alexis Pang, Mark Krstic, Peter Clingeleffer, Deli Chen, and Kate Howell *The University of Melbourne, 310/Building 194, the University of Melbourne, Parkville, Victoria, 3010, Australia (The University of Melbourne)

Rootstock grafting is a common vineyard implementation contributing to improved phenology, nutrient uptake, abiotic stress tolerance, and pathogen resistance of grapevines. Nevertheless, few previous studies have focused on how different rootstock genotypes can result in the variation in grapevine performance. In this study, we investigated the impacts of 14 different rootstocks on phenological, physiological, and nutritional performance of scion using Vitis vinifera L. cv Pinot noir MV6 as a model plant. Two commercial vineyards located in the Mornington Peninsula wine region of Victoria, Australia, were selected to reflect two types of meso-climate at the peninsula. At both vineyards, scions of V. vinifera L. cv Pinot noir were grafted onto 14 rootstocks, including 101-14, 1103 Paulsen, SO4, 110 Richter, Schwarzmann, 5C Teleki, 3309C, Merbein 5489, Merbein 6262, Merbein 5512, C20, C29, C113, and C114, with own roots acting as control group. Large canopy size, high pruning mass, and no obvious delay of flowering and veraison were observed in grapevines grafted to SO4 and 5C Teleki compared to the other rootstocks. There were obvious differences in nutrient absorption abilities of different rootstocks and environmental factors such as soil conditions influenced nutrients absorbed by grapevines. Overall, there were clear differences in grapevine performance: in the timing of flowering, progress of veraison, canopy size, pruning mass, and petiole nutrients, which preliminarily reflect the characteristics of the studied rootstocks. The present study helps clarify the impacts of rootstock on grapevine performance, which supports rootstock selection and benefits the wider wine industry.

Funding Support: Wine Australia, The University of Melbourne, The Australian Wine Research Institute Mornington, Peninsula Vignerons Association, Peninsula Vinecare, Yalumba Nursery, CSIRO Wine Victoria



Enology—Microbiology Session

A Consideration of Scale to Define Terroir based on Microbial Biogeography

Di Liu, Pangzhen Zhang, Qinglin Chen, Jean-Luc Legras, Deli Chen, and Kate Howell* *University of Melbourne, Building 194, Grattan Street, Parkville, Victoria 3010, Australia (khowell@unimelb.edu.au)

Microbial biogeography contributes to regional distinctiveness of agricultural products and is important to determine wine quality and marketing. To evaluate the microbial influence on wine characteristics, this study investigated the microbial biogeography of wine, the interplay between microbial patterns and affecting factors, and how these patterns drive wine quality and styles. Samples of soils, plant, grapes, must, and wine were collected in grapegrowing regions across Victoria, Australia. Genomic DNA was extracted for microbial diversity profiling of fungi and bacteria and gas chromatography-mass spectrometry was used to separate and identify volatile compounds in the headspace of wine. At a large scale (~400 km), we found that vineyard ecosystems are structured and distinguished by fungal communities, that the fungal microbiome is an important component of vineyard ecosystems, and that it correlates with regional distinctiveness of wine. Further studies considered a smaller scale of microbiota and investigated the changes in fungal community composition and diversity during the annual growth cycle of the grapevine. We found that fungal ecology is dependent on the grapevine habitat (root zone soil, root, leaf, flower, and grape) and plant developmental stage (flowering, fruit set, veraison, and harvest). We characterized the first core microbiota of grapevines that existed over space and time to drive seasonal community succession in the vineyard ecosystem. The influence of microbial biogeographic patterns decreased during wine fermentation as the fungal populations were dominated by Saccharomyces spp. yeasts. Further investigation of the strain diversity and dynamics of Saccharomyces cerevisiae showed that this yeast can shape geographic patterns at a small scale and determine wine characteristics within a single region. Our findings describe a comprehensive scenario of wine microbial biogeography, providing further evidence for microbial contributions to wine terroir and perspectives for maintaining microbial diversity to produce quality wine.

Funding Support: Wine Australia; University of Melbourne

Stilbenes Can Impair Malolactic Fermentation with Strains of Oenococcus oeni and Lactobacillus plantarum

Fabian Weber,* Sabrina Zimdars, Rita Caspers-Weiffenbach, and Pascal Wegmann-Herr

*University of Bonn, Friedrich Hirzebruch Allee 7, Institut of Nutritional and Food Sciences, 53121 Bonn, Germany (Fabian.Weber@uni-bonn.de)

Malolactic fermentation (MLF) is an important step in red winemaking to improve wine quality by microbiological deacidification, to increase microbial stability, and to modify general wine flavor by increasing red fruit perception. The phenolic composition of the wine influences the growth and metabolism of lactic acid bacteria (LAB) used for MLF. Due to increased emergence of fungus-resistant grape varieties, the present study aimed to analyze the influence of stilbenes on MLF; these compounds play an important role as phytoalexins in plants to counteract fungal pathogens and are therefore believed to accumulate in fungus-resistant grape varieties. It was shown that the grapevine-shoot extract Vineatrol and the stilbenes *e*-viniferin, ampelopsin A, r2-viniferin, r-viniferin, and mixtures of the five stilbenes all decelerated malic acid degradation. This can be attributed only partially to fewer viable cells, so the direct impact of stilbenes on the malolactic enzyme also has an effect. This was deduced

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from a constant viability of LAB while, at the same time, malic acid degradation was reduced. Red wines from fungus-resistant grapes were analyzed and fractionated to reveal the presence of stilbene. These results show that wines made from stilbene-rich grapes like fungus-resistant varieties are at higher risk for an inhibited or incomplete MLF.

Funding Support: This research project was financially supported by the German Ministry of Economics and Technology (via AiF) and the FEI (Forschungskreis der Ernährungsindustrie e.V., Bonn). Project AiF 18645N.

Interactions Between *Oenococcus oeni* and *Brettanomyces bruxellensis* during Malolactic Fermentation

Adam Lauderdale, Aubrey DuBois, Yan Ping Qian, Michael Qian, and **James Osborne*** *Oregon State University, Wiegand Hall 108A, 3051 SW Campus Way, Corvallis, OR 97331 (James.Osborne@oregonstate.edu)

A rapid malolactic fermentation (MLF) initiated by inoculation of Oenococcus oeni is a useful strategy to prevent Brettanomyces bruxellensis spoilage by minimizing the length of time wine is not protected by SO₂. This project investigated an additional benefit of conducting a rapid MLF: the prevention of B. bruxellensis growth due to inhibitory interactions with O. oeni. Pinot noir wine (no SO2, no MLF) was produced and used to test the ability of commercial O. oeni strains to inhibit B. bruxellensis growth at the end of MLF. All 10 O. oeni strains tested inhibited B. bruxellensis UCD2049 growth and volatile phenol production in Pinot noir wine, with some O. oeni strain variation observed. The potential mechanism of this inhibition was investigated by using a dialysis membrane to physically separate O. oeni and B. bruxellensis cells in wine, but to allow free movement of nutrients and other potential inhibitory compounds. Physical separation of O. oeni cells from B. bruxellensis cells relieved the inhibition of *B. bruxellensis* by *O. oeni* that occurred when the two microorganisms were present together. This suggests that inhibition was not due to nutrient depletion by O. oeni and was also unlikely to be caused by an inhibitory compound. Instead, these results provide evidence that the inhibition of *B. bruxellensis* by *O. oeni* was due to cell-cell contact. The sensitivity of additional B. bruxellensis strains to O. oeni was also determined. While B. bruxellensis UCD2049 populations declined rapidly when inoculated into Pinot noir wine that had just completed MLF with O. oeni Alpha, growth of seven other B. bruxellensis strains was not affected. Subsequent experiments showed that ethanol tolerance differences between B. bruxellensis strains may have played a role in their sensitivity to repression by O. oeni.

Funding Support: Oregon Wine Board



Enology—Phenolic Maceration Session

Tannin and Protein Relationship in Low-Tannin Red Wine: Through the Lens of an Optimized Proteomics Method

Alex Fredrickson* and Misha Kwasniewski *University of Missouri, 1453 Wikiup Dr, Santa Rosa, CA 95403 (ajfpvn@mail.missouri.edu)

Using a newly-optimized method to precipitate, quantify, and characterize grape and wine proteins, we identified >1200 unique proteins in juice and 900 in wine of Vitis interspecific hybrid cv. Chambourcin. To modify tannin concentrations, the following treatments were employed in the winery: large exogenous additions of 350mg/L catechin equivalents (CE) of tannin (Tannin+) at (i) crush or (ii) one month post press, or (iii) skin fragmentation (accentuated cut edges or ACE). Mean tannin concentrations at bottling were 36 mg/L CE in the control versus 91 mg/L in ACE. In-crush Tannin+ yielded 187 mg/L. The post-press Tannin+ was nearly 12-fold greater than the control (419 mg/L). Total protein content in wine correlated poorly with final tannin concentration, with an r2 of 0.2146. However, by modeling final tannin content with spectrum counts of individual proteins in juice and wine, we identified numerous proteins with predictive ability of final tannin content (2322 predictors). Overall, these yielded an r2 of 0.8031, indicating a strong inverse relationship between concentrations of certain proteins and tannin retention. Within the model using variable importance projection (VIP) scores, the top 10 individual proteins resulted in eight *Vitis* proteins, including vacuolar invertase, ß-fructofuranosidase, thaumatin-like, and osmotin-like. The other two proteins were yeast-derived. Of the individual proteins, all eight Vitis proteins had a negative logarithmic r2 of 0.8 or better, while the yeast proteins were lower. Proteomics showed many proteins that relate to tannin retention, and they were not limited to pathogenesis-related proteins. Although important, the direct relationship of protein and tannin in wine may not be a simple driving force in the extraction/retention of tannin in red wine.

Funding Support: Grape and Wine Institute (Missouri)

Effect of Polysaccharide Extraction and Addition Strategies on Color Stability and Pigment Formation in Red Wines

Stephan Sommer,* Faeth Anderson, and Fabian Weber *California State University, Department of Viticulture and Enology, 2360 E. Barstow Ave. M/S VR89, Fresno, CA 93740 (ssommer@csufresno.edu)

Dark color is an important quality indicator for red wines and is associated with maximum extraction of grape components, long shelf-life, and increased perceived consumer guality. Increasing anthocyanin concentration alone has only a temporary effect, since large portions of red color are lost during winemaking due to oxidation, polymerization, and additions like sulfur dioxide. Some cultivars present additional challenges during red wine production due to their unique anthocyanin composition, low extractability of anthocyanins, lack of stabilizing factors, or predominant growing conditions like excessive heat. This study was conducted with Barbera as the main cultivar due to observed poor color retention in previous years. In addition to traditional strategies like co-fermentation with other cultivars (Syrah and Touriga Nacional) and extended maceration, the grapes were also subjected to polysaccharide addition (pectin and mannoprotein), cold soak, and ripasso of Syrah and Touriga skins. Color extraction and retention was monitored throughout fermentation and maceration and after bottling. The phenolic composition, astringency, and polysaccharide profiles of all wines were determined using photometric assays and liquid chromatography. Results indicate that the addition of pure polysaccharides prior to fermentation

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affected color quality of red wines differently than extraction of polysaccharides from grape material during and after fermentation. Although the tannin concentration with pure polysaccharide addition was generally lower, the level of polymeric pigments showed no negative effect. Extended maceration clearly creates the most favorable environment for production of stable colored pigments, but also produces wines that lean toward oxidation. A secondary polysaccharide extraction through a ripasso procedure shows that the right cultivar choice can improve the color and pigment composition of challenging grape varieties like Barbera.

Funding Support: n/a

Effect of Structural Transformations on Precipitability and Polarity of Red Wine Phenolic Polymers

Ingrid Weilack,* Christina Schmitz, James F. Harbertson, and Fabian Weber *Rheinische Friedrich-Wilhelms-Universität Bonn - Institute of Nutritional and Food Sciences, Friedrich-Hirzebruch-Allee 7, 53115 Bonn, Germany (weilack@uni-bonn.de)

Condensed tannins and polymeric pigments are essential red wine components since they contribute to color stability, taste, and mouthfeel. Due to the chemical heterogeneity of proanthocyanidin polymers, analytical tools to determine the polymers' structural features are limited. The incorporation of anthocyanins increases the structural complexity even more and leaves it almost impossible to objectively analyze their impact on sensory attributes and quality of red wine. To better understand the structural diversity of red wine polymers, this study used FLASH-fractionation of polyphenolic wine extracts to reveal the relationship between phenolic polymers and two physicochemical properties: polarity and hydrophilicity. Red wine polyphenols were characterized regarding their polarity, octanol-water partitioning coefficient, protein precipitation assay, UHPLC-MS, and color. Tannin concentrations in wines subjected to forced aging decreased while the concentrations remained constant in the corresponding extracts, suggesting an alteration in precipitation behavior. A simultaneous increase in precipitable polymeric pigments gives rise to the assumption that incorporation of anthocyanins into tannin molecules alters interactions with red wine polysaccharides and proteins, resulting in lower tannin readings. Wines produced from differently ripe berries showed a shift in polyphenol polarity from being more polar at early harvest and less polar at the later stage of maturity. This is accompanied by an overall increase in precipitible polymeric pigments and a change in astringency perception. Finding tannins and polymeric pigments in different FLASH-fractions indicates that precipitability of polymers is affected by the physicochemical properties, which in turn depend on the degree of polymerization and degree of pigmentation. The results of this study show that red wine astringency and its sub-qualities may be related to the increase in precipitable polymeric pigments and their putative enhanced interaction with wine polysaccharides and can help to better understand astringency mechanisms.

Funding Support: This research project was financially supported by the German Ministry of Economics and Technology (via AiF) and the FEI (Forschungskreis der Ernährungsindustrie e.V., Bonn); Project AiF 20024N



Viticulture—Irrigation Management Session

Assisting Irrigation Scheduling through Machine-Learning Modeling of Grapevine Water Status in Space and Time

Luca Brillante,* Matteo Ramagli, and Khushwinder Singh *California State University Fresno, 2360 E Barstow Ave, Fresno, CA 93740 (lucabrillante@csufresno.edu)

Modeling grapevine water status on hillslopes is challenging. This research demonstrates for the first time that it is possible to obtain daily estimates of grapevine water status at the estate scale by re-elaborating routine measurements with digital technology. This information can be used to optimize irrigation scheduling, drive selective harvest decisions, and cluster vineyard variability. Data was collected during two consecutive seasons in a ~40 ha (100 ac) wine estate located in Paso Robles, CA. The topography was very diverse, with large variation in slope grade (0 to 30%) and exposure (0 to 359). One hundred experimental units of Cabernet-Sauvignon, Cabernet franc, and Petit Verdot were identified by a maximum dissimilarity sampling algorithm based on environmental attributes derived from a digital elevation model and a soil map. Grapevine water status was monitored by weekly measurements of stem water potential, Ψ_{stem} , and carbon isotope discrimination of musts, δ^{13} C, at harvest. The grape composition during ripening was assessed by measuring primary components and skin phenolic composition with high-performance liquid chromatograph-diode array detection. Vegetation indexes were derived from ~3 m resolution CubeSat satellites. Irrigation amounts were provided by the grower and weather data was obtained from three on-site stations. Ψ_{stem} was modeled from weather data, irrigation amounts, vegetation indicies, topographic attributes, and soil type using a gradient-boosting-machine algorithm. The model predicted plant water status with <0.1 MPa assessed error through several validation methods. External validation of the model was carried out by correlating predictions with $\delta^{13}C$. The model allowed obtaining high-resolution daily mapping of Ψ_{stem} at the estate scale. Time-series of grapevine Ψ_{stem} correlated significantly with the content of total soluble solids of musts, grape anthocyanin amounts, and the ratio of tri-hydroxylated to di-hydroxylated compounds at harvest and were mapped. Spatial clustering of grape anthocyanin composition was obtained from Ψ_{stem} model-estimates and could be used to guide harvest selectively.

Funding Support: Daou Family Estates

Seasonal and Postharvest Canopy Water Use and Dry Matter Production of *Vitis vinifera* L. in a Hot Climate

Vinay Pagay,* Felipe Canela, and Hongkai Pei *University of Adelaide, PMB 1, Glen Osmond, Australia (vinay.pagay@adelaide.edu.au)

Knowledge of grapevine water use is vital for efficient irrigation management in vineyards so that irrigation volumes match crop water demand. Over two growing seasons, 2019-20 and 2020-21, we used custom-built, whole-canopy chambers to estimate the daily water requirement of mature grapevines (*Vitis vinifera* L. cvs. Chardonnay and Shiraz [syn. Syrah]) grown in a hot climate of South Australia. Environmental conditions, soil moisture, and vine water status (midday stem water potential) were measured simultaneously and crop evapotranspiration (ET_c) was calculated using the Penman-Monteith relationship for comparison with the whole-canopy measurements. In the second season, heat pulse velocity (sap flow) sensors were installed in the sentinel vines, calibrated with the canopy chambers, and used to provide continuous vine transpiration estimates from budbreak to postharvest. Our results indicate that mature, fully-irrigated grapevines in the field used 12 to 17 L water per

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day during the peak of the summer. Vine water use measured with the chambers represented 22% and 28% of seasonal irrigation applied on Chardonnay and Shiraz vines, respectively. Vine water use was also estimated at -60% of ET_c . Daily dry matter production of Chardonnay and Shiraz grapevines were -200 g/day and 150 g/day late in the season, between veraison and harvest. Sap flow sensors indicated that vine water use during heatwaves, when daily maximum temperatures exceeded 35°C for three or more days, increased by 13% and 7% (compared to both pre- and post-heatwave periods) in Chardonnay and Shiraz, respectively. Canopy water use efficiency was greatest early in the morning, perhaps due to the relatively greater proportion of diffuse light at that time. Whole-canopy gas exchange chambers coupled with calibrated sap flow sensors have proven to be a valuable tool for quantification of plant water use and therefore, irrigation scheduling in vineyards.

Funding Support: Wine Australia, Riverland Wine

Effect of Deficit Irrigation and Mechanical Leafing on Yield, Berry, and Wine Chemistry of Cabernet Sauvignon in California

Shijian Zhuang,* Qun Sun, Karl Lund, Matthew Fidelibus, and S. Kaan Kurtural *University of California Cooperative Extension at Fresno, 550 E Shaw Ave Suite 210-B, Fresno, CA 93710 (gzhuang@ucanr.edu)

Cabernet Sauvignon is the second-most crushed variety in CA. The southern San Joaquin Valley is considered not ideal for growing premium Cabernet Sauvignon due to its arid, hot growing climate. However, the demand for high-quality Cabernet Sauvignon is strong and it has been a great interest for local growers and wineries to produce high-quality Cabernet Sauvignon at low cost to stay competitive. A twoway factorial split-plot design was implemented in a commercial vineyard located in Madera County in 2018, 2019, and 2020. Quadrilateral cordon-trained, spur-pruned Cabernet Sauvignon vines grafted onto Freedom rootstock were used under a sprawl system. Two levels of water deficits and three timings of mechanical leafing, replicated five times, were used in this study with six vines designated as an experimental unit. One water deficit was 80% ET_{c} (targeted Ψ of -1.3 MPa) through the growing season with the other of 50% ET_c (targeted Ψ of -1.5 MPa) from berryset to veraison and back to 80% ET, (targeted Ψ of -1.3 MPa) until harvest. The three timings of mechanical leafing were bloom, berry set, and no leafing. Midday leaf water potential (Ψ), fruit-zone photosynthetically active radiation (PAR), and leaf gas exchange were measured during the season. Water deficit significantly decreased yield through reduced cluster number and berry size without changing leaf area. Water deficit reduced TSS, titratable acidity, and 3-isobutyl-2-methoxypyrazine (IBMP) while improving berry anthocyanins. Leafing did not affect any yield component in our study but improved berry anthocyanins. Wine color was improved by water deficit and leafing.

Funding Support: American Vineyard Foundation

*indicates corresponding author



Viticulture—Pest and Disease Session

Evaluation of Grapevine-Derived Microbes for Sustainable Pruning Wound Protection Against Grapevine Trunk Diseases

Robert Blundell, Molly Arreguin, and Akif Eskalen * *UC Davis Dept. of Plant Pathology, UC Davis, Davis, CA 95616-18 (aeskalen@ucdavis.edu)

The grapevine trunk diseases (GTDs) complex, including Esca, Eutypa dieback, and Botryosphaeria dieback, poses a major threat to the economic sustainability of grapevine cultivation, diminishing vineyard longevity and productivity worldwide. Pruning wounds represent the primary entry point for fungal pathogens responsible for these diseases and are the focus of preventative disease mitigation strategies. Our goal was to identify and characterize naturally-occurring potential biocontrol agents from different grapevine tissues/sources, including sap, pith, and wood tissue, and evaluate their antagonistic activity against GTD fungal pathogens. We recovered 11 endophytic fungi (Trichoderma spp. and Aureobasidium spp.) and bacteria (Bacillus spp.) and two epiphytes isolated from the surface of seven-day-old, untreated pruning wounds. Recovered isolates showed inhibition up to 70% against two selected common fungal pathogens responsible for GTDs in vitro. Selected isolates with potential as biocontrol agents were then evaluated in planta in the greenhouse and in three field trials (Yolo, Sacramento, and Kern Counties) and their efficacy was compared with that of commercial pruning wound protectants. In greenhouse and field trials, Trichoderma isolates from this study and Trichoderma-based commercial products were effective in protecting wounds, particularly Biotam, which conferred 88 and 100% mean percent disease control (MPDC) against E. lata and N. parvum, respectively. Some chemical treatments, (Topsin M + Rally and Luna Sensation) were also effective wound protectants, with at least 75% MPDC against both pathogens in Sacramento and Kern County field trials. Trichoderma-based treatments had >80% recovery from treated canes eight months from application, suggesting that effective wound colonization by biologicals is likely tied to sustained pruning wound protection.

Funding Support: Various commercial chemical/biological companies, ASEV traditional scholarship, American Vineyard Foundation

Understanding Fungicide Use Patterns in Vineyards through Historical Records

Charlotte Oliver and Michelle Moyer*

*Washington State University, 24106 N. Bunn Rd., Prosser, WA 99350 (michelle.moyer@wsu.edu)

Development of fungicide resistance is often attributed to practices such as product over-use, product misuse, and poor timing. While applicators assume their practices adhere to recommendations for fungicide resistance mitigation, validating those assumptions can be difficult. One approach for validation is broad-scale evaluation and analysis of regional historical fungicide records across multiple farming entities. The Fungicide Resistance Assessment Mitigation and Extension network has been collecting fungicide records from informed and consenting growers in Michigan (MI), Oregon (OR), and Washington (WA) to take a national look at spray practices and identify potential issues contributing to the selection of fungicide resistance. These records span from 2015 to 2019. The mean of yearly applications was seven; by state, the mean ranged from six (WA) to eight (OR). The mean application interval was 14 days; by state, the mean ranged from 13 (OR) to 16 (MI). Using a subset of data from OR and WA with continuous records from 2016 to 2019, we saw a reduction in FRAC

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3 use (-6% in OR and -9% in WA). There was also a reduction in FRAC 9 (-5%) and FRAC 11 (-4%) fungicide use in WA. We saw an increase in FRAC 7 (SDHI) fungicide use (+1% in OR, +5% in WA), FRAC 50 fungicide use (+4% in OR, +6% in WA), FRAC U06 fungicide use (+1% in OR, +7% in WA), and FRAC M02 fungicide use (+8% in OR, +12% in WA). This shift in product usage is likely related to increased understanding of known resistance issues in FRAC 3 and 11 fungicides. However, the product shift is also placing resistance development pressure on products such as those in the FRAC 50 and U06 classifications, which have seen resistance development due to over-use in Europe.

Funding Support: United States Department of Agriculture—National Institute for Food and Agriculture—Specialty Crop Research Initiative Award No. 2018-03375

The Current Status of *Grapevine Leafroll-Associated Virus 3* and its Genetic Diversity in Washington State Vineyards

Arunabha Mitra, Sridhar Jarugula, Bhanupriya Donda, Emily Jordan, and Naidu Rayapati*

*Washington State University, WSU-IAREC, 24106 N Bunn Rd, Prosser, WA 99350 (naidu.rayapati@wsu.edu)

Among several viral diseases documented in Washington State vineyards, grapevine leafroll disease (GLD) continues to be an economically detrimental threat to vineyard health and productivity. Disease surveys have shown consistently that Grapevine leafroll-associated virus 3 (GLRaV-3, genus Ampelovirus, family Closteroviridae) is the most insidious and prevalent among four GLRaV species reported in Washington vineyards. Since GLRaV-3 occurs as multiple genetic variant groups in several grapevine-growing regions worldwide, this study was undertaken to examine genetic diversity of the virus in Washington vineyards. For this purpose, 2291 samples were collected between 2015 and 2019 from red- and white-fruited winegrape (Vitis vinifera) cultivars from 14 commercial vineyards in four American Viticultural Areas. Samples were tested for the presence of GLRaV-3 by RT-PCR assays, using primers specific to a portion of the heat shock protein 70 homologue (Hsp70h) gene encoded by the virus. The -607 base pair amplicons obtained from 818 samples were sequenced and compared with corresponding GLRaV-3 sequences available in public databases. In addition, total RNA preparations from 81 grapevine samples that previously tested positive for GLRaV-3 were subjected to Illumina high-throughput sequencing to generate near-complete viral genome sequences. These sequences were compared with GLRaV-3 sequences reported from other grapevine-growing regions. A global phylogenetic analysis revealed the presence of several GLRaV-3 variants in Washington vineyards. Some aligned with six genetic variant groups (designated as I, II, III, V, VI, and IX) reported from other grapevine-growing regions. Among them, GLRaV-3 isolates belonging to variant group I were predominant in Washington vineyards. Information on the occurrence of genetically divergent virus isolates is providing insights for robust tracking of genetic variants and better understanding of the epidemiology of GLRaV-3 to improve management of GLD in vineyards.

Funding Support: Washington State University, Auction of Washington Wines, Washington State Wine Commission, Northwest Center for Small Fruits Research, and M. J. Murdock Charitable Trust.



Enology—Impact of Smoke Exposure on Grape and Wine Composition Session

Bucket Fermentations to Predict Risk from Smoke Exposure: Chemical Composition and Sensory Perception

Thomas S. Collins,* Scott C. Frost, Bojana Leonard, Danielle J. Fox, James F. Harbertson, and Matthew Boenzli *Washington State University, 359 University Drive, Richland, WA 99354 (tom.collins@wsu.edu)

Small-scale. "bucket" fermentations have gained acceptance as a way to predict the risk to wine quality from vineyard smoke exposure. However, there is no published data supporting their use. When conducted several days prior to the anticipated harvest date, wines from these fermentations can be evaluated for smoke-related aromas and flavors or analyzed for smoke taint marker compounds. In this study, bucket (5 kg) fermentations and small commercial-scale (362 kg) fermentations were conducted using fruit from a vineyard affected by a wildfire that burned up to one edge of the vinevard. We examined the effects of cultivar and fire proximity by harvesting Cabernet Sauvignon, Syrah, Grenache, and Petite Verdot fruit from blocks that were adjacent to or remote from the most fire-affected edge; for each cultivar a high- and low-impact block was selected. The sensory and chemical profiles of the resulting wines were evaluated using descriptive analysis and solid-phase microextraction-gas chromatography-mass spectrometry. Volatile phenol concentrations were comparable in both large- and small-scale fermentations and were consistent with expectations based on proximity to the fire-affected vineyard edge. However, a significant cultivar effect was also found: Petite Verdot and Syrah had higher concentrations of guaiacol and 4-methyl guaiacol than the Grenache or Cabernet Sauvignon, while the Grenache had more o-cresol and m-cresol than any other cultivar. Sensory evaluation found that the flavor profile of the bucket fermentations was impacted by cultivar and fire proximity. Cabernet Sauvignon wines had more fresh bell pepper aroma and astringent mouthfeel, while the Syrah wines had more canned asparagus character. Proximity to the fire increased the intensity of the peaty/medicinal aromas, but an interaction between cultivar and location was shown for wood smoke/ash intensity.

Funding Support: Washington State Wine Commission

Volatile Phenols as Markers for Smoke Exposure of Pinot noir Wine

Ruiwen Yang, Yanping L. Qian, Armando Alcazar, and **Michael C. Qian*** *Oregon State University, 100 Wiegand Hall, Corvallis OR, Corvallis, OR 97330 (michael.qian@oregonstate.edu)

Guaiacol, 4-methylguaiacol, and o-, p-, and m-cresol are believed to relate to grape and grapevine exposure to smoke generated from wildfires. Both guaiacol and 4-methylguaiacol are widely used as indicators of smoke exposure. In this study, 86 Pinot noir wines were randomly selected from 2013 to 2016 vintages, with -20 samples from each year as control wines. The samples were obtained from industry fermentation without barrel aging. Guaiacol, 4-methylguaiacol, and o-, p-, and m-cresol in the wines were analyzed using solid-phase microextraction-gas chromatography-mass spectrometry with stable isotope compounds as internal standards. Total volatile phenols were also analyzed after the sample was hydrolyzed with HCl for 4 hr at 100°C (pH 1.2). The results were compared with >300 smoke-exposed red wine samples collected during the 2020 wildfire season. T-test, correlation heatmap, individual correlations, principal component analysis, and partial least squares discriminant analysis were used to compare the smoke-exposed and control wines. The results showed that smoke-exposed red wine presented the data's biggest dispersion,

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and o-, p-, and m-cresol exhibited the biggest difference across samples. A similar trend was observed for total phenols, with total p-cresol exhibiting the greatest difference. Except for free 4-methylguaiacol and total m-cresol, all other volatile phenols were significantly more abundant in smoke-exposed wine (p > 0.05). Univariate data analysis suggested that total p-cresol is the most discriminating compound for smoke exposure, followed by free o-, p-, and m-cresol.

Funding Support: American Vineyard Foundation (AVF), Oregon Wine Board (OWB) and Oregon Wine Research Institute (OWRI)

Impact of Amelioration Techniques on the Chemical Composition and Sensory Characteristics of Smoke-Impacted Wines

Anita Oberholster,^{*} Yan Wen, Samnang Hay, Sandra Dominguez Suarez, Jesse Erdmann, Bishnu Neupane, Charles Brenneman, Hildegarde Heymann, Scott Lafontaine, Raul Cauduro Girardello, and Arran Rumbaugh *UC Davis, One Shields Avenue, Davis, CA 95616 (aoberholster@ucdavis.edu)

The increasing incidence of wildfires in winegrape-growing regions poses a significant risk to the grape and wine industry. One risk is the potential of persistent exposure to smoke to compromise the quality and value of winegrapes and adversely affect wines made from smoke-exposed grapes. A wine is seen as smoke-impacted or -tainted when there is an overpowering smoky, medicinal, chemical, burnt, or ashy aroma on the nose and a distinctive retronasal ashtray-like character in the mouth. Several solutions for smoke taint have been promoted, such as treatment with enzymatic enzymes, fining, reverse osmosis, and spinning cone treatments. These amelioration techniques were investigated using smoke-impacted wines made from Cabernet Sauvignon grapes from Napa and Lake Counties (UC Davis Teaching and Research Winery) in 2017 and 2018. The impact on wine composition: specifically, smoke-taint marker compounds (both free and bound volatile phenols as determined by GC-MS and LC-MS/MS) and other key volatile and non-volatile compounds important for wine quality were evaluated to determine their efficacy. Additionally, descriptive analysis was performed on the treated and untreated wines. Results indicated that although most amelioration techniques removed some smoke taint markers (mostly free and some bound) and decreased smoke taint perception, they lacked specificity and impacted the overall character of the wine in a negative way. However, the impact depended on the level of treatment needed and the decrease in mouthfeel could be adjusted using TTB-approved products or blending. Amelioration techniques were not 100% fixes of smoke taint, but significantly decreased smoke-taint perception. Further research could result in recommendations regarding the feasibility of treatment success based on the wine matrix, so that winemakers can make informed decisions.

Funding Support: American Vineyard Foundation



Enology—Sensory and Aroma Compounds Session

Accelerated Bench-Tests for Predicting H₂S Formation in Canned Wines

Rachel Allison,* Austin Montgomery, and Gavin Sacks *Cornell University, 411 Tower Rd., Stocking Hall, Room 253, Ithaca, NY 14853 (rba55@cornell.edu)

Sulfur-like off-aromas (SLOs), also called "reduced aromas," are reportedly responsible for upwards of 25% of faults identified in premium wines. Hydrogen sulfide (H_2S ; "rotten egg" aroma) is often in excess of its sensory threshold (~1 μ g/L) in wines with SLOs. A growing area of concern for H₂S in packaged wines is in the fast-growing category of canned wines. This phenomenon is credited to reaction of SO₂ in wine with aluminum metal and may occur even in the presence of a polymeric liner to prevent direct contact between aluminum metal and the wine. However, considerable variation is observed in H₂S production among canned wines with similar free SO₂ concentrations, such that predicting the suitability of a given wine for canning is still challenging. We report on the development and validation of an accelerated benchtest to predict H_2S formation during canned wine storage. In the assay, a 1 × 2 cm coupon of coated AI is incubated in 25 mL wine under anoxic conditions and elevated temperatures. In initial experiments, 10 commercial wines were incubated with Al coupons coated with one of three different commercial polymeric liners. After three days at 50°C, there was negligible formation of H_2S in red wines (<10 μ g/L) and 15 to 200 μ g/L H₂S in white and rosé wines. Variation in H₂S formation among wines far exceeded variation for replicate samples (average relative standard deviation <30%). Validation against canned wines stored at room temperature for up to eight months is currently underway.

Funding Support: New York Wine and Grape Foundation

Sensory Characterization of Typicity in Australian Cabernet Sauvignon Wines

Lira Souza Gonzaga, Dimitra Capone, Susan Bastian, Lukas Danner, and David Jeffery*

*The University of Adelaide, PMB 1, Wine Central Building, Waite Campus, Glen Osmond, Australia (david.jeffery@adelaide.edu.au)

Wine typicity encompasses the influence of terroir, cultivar, viticulture, and winemaking and enables recognition and characterization of unique varietal wines from a delimited geographical area. Notions of provenance and typicity have traditionally been important indicators for high-quality wines from Old World wine producers but are also significant for countries like Australia that seek to increase the value of regional wines and grow a reputation as a fine wine-producing nation. This project focused on Cabernet Sauvignon wines, Australia's second-most important variety, and used different approaches to identify the sensory attributes responsible for the distinctive profiles of three Australian wine-producing regions: Coonawarra, Margaret River, and Yarra Valley. Cabernet Sauvignon-dominant Bordeaux wines were also included for benchmarking purposes. To begin, content analysis was undertaken with over 2500 online wine reviews from six well-recognized websites, demonstrating an initial regional profile and aiding in selection of wines for further study. Additionally, an expert panel evaluated 86 commercial 2015 Cabernet Sauvignon wines. This involved a sorting task, a short description of each group formed, and a quality rating. This approach evidenced different descriptors for each region and had good agreement with the results of the content analysis, especially in terms of quality (RV coefficient of 0.826), with a number of common descriptors arising such as "green" for lower-qual-

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ity wines and "dark fruits" for higher-quality ones. Subsequently, a subset of 52 wines underwent descriptive analysis (DA) with a trained panel. Between the expert sorting task with descriptions and DA, shared attributes were determined to be important for different regions, such as "savory" and "cooked vegetables" commonly used to describe wines from Bordeaux and Yarra Valley, "mint" being significant for Coonawarra wines, and "floral" for Margaret River wines.

Funding Support: ARC Training Centre for Innovative Wine Production

Tropical Fruit Aroma in White Wines: The Role of Fermentation Esters and Volatile Thiols

Angelica lobbi and Elizabeth Tomasino*

*Oregon State University, 3051 SW Campus Way, Corvallis, OR 97330, Corvallis, OR 97330 (elizabeth.tomasino@oregonstate.edu)

Volatile thiols are impact aroma compounds, well-known for imparting tropical fruit aromas such as passion fruit, guava, grapefruit, and citrus in white wines. More recent evidence suggests that additional aroma compounds also contribute to tropical fruit aromas, such as fermentation esters or the interactions between these volatile families. This study investigated the effects of combining fermentation esters and/or volatile thiols to determine their impact on the fruitiness aroma perception of white wines. Pinot gris wine was produced at the OSU research winery and was dearomatized using Lichrolut EN resin. Combinations of fermentation volatile compounds were added to the wine, forming the aroma base. Treatment wines were composed of additions of different concentrations and combinations of volatile thiols and/or fermentation esters. Samples were subjected to sensory analysis by 46 white wine consumers, who evaluated the orthonasal aroma of the wines and participated in check-all-that-apply (CATA). Following the results obtained by CATA, samples were subjected to a sensory descriptive analysis (SDA) panel, where 13 trained panelists evaluated the intensity of the most-used aroma attributes elicited by consumers. CATA results were analyzed using correspondence analysis and SDA data was analyzed by discriminant analysis. Volatile thiols without esters contributed to earthy and grassy aromas. Overall, tropical fruit aromas were detected in the several treatments containing combinations of fermentation esters and fermentation esters + volatile thiols. Differences in the intensity of the aroma attributes were observed. This study showed that esters and thiols are necessary for tropical fruit aroma causation in white wines. Therefore, grapegrowers and winemakers wishing to enhance the tropical fruit aroma of their wines should adapt viticultural and winemaking conditions to increase the concentrations of both aroma families.

Funding Support: American Vineyard Foundation



Viticulture—Vine Physiology Session

The Effect of Grapevine Age (*Vitis vinifera* L. cv. Zinfandel) on Vine Performance and Fruit Composition

Vegas Riffle, Nathaniel Palmer, L. Federico Casassa, and Jean Dodson Peterson* *California Polytechnic State University SLO, One Grand Avenue, Building 11, Room 218, San Luis Obispo, CA 93407 (jdodsonp@calpoly.edu)

"Old" vines are highly sought after by wine industry professionals and consumers due to their rarity and perceived superior wine quality. To evaluate the validity of this assertion, a two-year study was conducted at a single, interplanted Zinfandel vineyard block in California's Paso Robles AVA. Treatments included Young vines (five to 12 years old), Control (a representative proportion of young to old vines in the block), and Old vines (40 to 60 years old). Vine age had an effect on vine vegetative growth, with Old vines producing shorter internodes (25.5% decrease) and smaller shoot diameters (29.3% decrease) than Young vines. No differences were found in photosynthetically active radiation or leaf area index between Old and Young vines. Old vines developed more quickly during berry formation and more slowly during berry ripening periods. Due to variation in the timing of sugar accumulation, Old vines were harvested 21 days after Young vines in 2019 and nine days after in 2020. Vine age affected yield, with Old vines producing, on average between both seasons, 3.7 kg more fruit per vine than Young vines. Old vines also produced, on average between both seasons, 22.8 more clusters per vine than Young vines (5.41 tons/acre and 2.64 tons/acre, respectively). Larger vine capacity is attributed to Old vines having more arm, spur, and dormant bud positions per vine than Young vines. Additionally, Old vines had larger trunk circumferences and diameters than Young vines. Old vine fruit had higher Brix, pH, and titratable acidity than Young vines at harvest in 2019 only, with no differences found in 2020. No differences in berry color or phenolics were found between Old and Young vines. These results suggest the potential for greater yield and increased growing season length when extending the longevity of Zinfandel vineyards.

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Arbuscular Mycorrhizal Fungi Do Not Enhance Nitrogen Uptake of Pinot noir Grapevines from Organic Nitrogen Sources

Tian Tian* and R. Paul Schreiner

*University of California Cooperative Extension, 1031 South Mountain Vernon Avenue, Bakersfield, CA 93307 (titian@ucanr.edu)

Arbuscular mycorrhizal fungi (AMF) are symbiotic partners of grapevines and contribute to nutrient acquisition from soil. To understand whether AMF enhance vine nitrogen (N) uptake from organic sources of N, mycorrhizal and non-mycorrhizal Pinot noir plants were grown in soil with low N availability. Within each AMF group, vines were assigned to seven different N treatments, including no N addition, NH₄NO₃ applied at 3 or 6 mM N, glycine applied at 3 or 6 mM N, and leaf litter applied at 10 or 20 g. Vines were initially grown for four weeks with moderate N prior to the addition of different types of N. Shoot growth and root colonization by AMF were measured periodically, and vines were destructively harvested after nine weeks to determine biomass and nutrient uptake. Shoot biomass, root biomass, and vine N uptake were

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improved by each of the N sources tested, but AMF did not enhance vine N uptake in any case. Shoot growth was reduced by AMF when NH_4NO_3 was supplied at 6 mM N, but AMF did not alter shoot biomass in other N treatments. Root colonization by AMF increased in response to added N, irrespective of the type of N supplied. However, the frequency of arbuscules in roots declined in both litter treatments at nine weeks but remained high in the NH_4NO_3 and glycine treatments. Results indicate that AMF do not improve vine N uptake from organic N sources and can divert investment to shoot growth when inorganic N is supplied at a moderate rate.

Funding Support: USDA-ARS

Sensitivity of the (*Vitis vinifera* L.) Riesling Volatome to Berry Maturity and Solar Radiation

Josh Vander Weide and Simone Castellarin*

*University of British Columbia, 2329 West Mall, Vancouver, British Columbia, V6T 1Z4, Canada (simone.castellarin@ubc.ca)

Volatile compounds, particularly terpenes and C₁₃ norisoprenoids, are highly subject to modulation during berry ripening. Solar radiation also affects production of grape volatiles. Despite this knowledge, little is understood regarding the specific sensitivity of both free and bound volatiles to berry maturity and solar radiation. To investigate this, a 2 × 2 factorial experiment was established using greenhouse-potted (Vitis vinifera L.) Riesling vines. The first treatment involved altering berry maturation at 50% veraison (~8 Brix) by girdling shoots above and below clusters to estimate the effect of halting berry maturity on volatile production. Second, clusters were covered with light-impenetrable bags at the same point, halting light exposure on volatile production. "Girdling" and "bagging" treatments were compared to vines subjected to a combination of both treatments and an untreated control. In addition to gas exchange and basic fruit quality parameters, free and glycosylated volatiles were profiled from lag phase to harvest maturity stages, including at two (~9 Brix), 10 (~14 Brix), and 30 days (~21 Brix) post-treatment to gauge short-, mid-, and long-term responses to treatments. Solar radiation was reduced to <2 μ mol m²/s, while berry temperature was not altered by either treatment. Photosynthesis was significantly decreased by girdling from one week post-treatment until harvest due to feedback inhibition. Total soluble solids were unaffected by bagging but were significantly decreased by girdling. An immediate decrease in the concentration of select volatiles was observed at two days post-bagging; however, this effect was delayed in response to girdling. Volatiles influencing Riesling varietal character were affected by both treatments. Bagging had a faster and more severe impact on the reduction of free and bound volatiles in Riesling than limitation of berry maturity due to girdling, highlighting the importance of solar radiation in volatile development during berry ripening.

Funding Support: NSERC Discovery



Viticulture—Eastern Viticulture Session

Effect of Sun Exposure on Evolution and Distribution of Anthocyanins in Interspecific Red Hybrid Winegrapes

Catherine Dadmun, Anna Katharine Mansfield,* and Hans Walter-Peterson *Cornell University, 665 W North St, Geneva, NY 14456 (akm87@cornell.edu)

Interspecific hybrid winegrapes are economically important in areas where environmental pressures inhibit traditional Vitis vinifera production. Red hybrid grapes, however, show great diversity in anthocyanin profile, and viticultural and winemaking techniques that optimize color in red V. vinifera wines are often ineffective for hybrid wine production. Because the chemistry of hybrid grape anthocyanins is largely unknown, the reactions they undergo during ripening, wine production, and aging are poorly understood. To clarify the effect of vine microclimate on red hybrid wine color, anthocyanin profiles were assessed for shaded and unshaded fruit from three economically significant cool-climate hybrid cultivars (Vitis spp): Corot noir, Maréchal Foch, and Marguette, Berry samples were collected throughout ripening from triplicate blocks of each cultivar grown in the New York Finger Lakes region, then skin extract anthocyanins were characterized using high-performance liquid chromatography analysis. Tentative anthocyanin identifications were confirmed via mass spectrometry. Light exposure and berry and air temperature were monitored in Corot noir throughout the season to represent generalized vine microclimate. Across all cultivars, the samples that underwent the leaf-pulling treatment (exposed samples) had total anthocyanin concentrations that were not significantly different from the control (shaded samples). However, certain individual anthocyanins within each cultivar demonstrated different concentrations with the exposure treatment. This work is the first step in defining the evolution of anthocyanin profiles during interspecific hybrid grape ripening to allow cool-climate winegrape growers to optimize viticultural production methods for high-quality red hybrid wines.

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Effects of Pruning Timing and Leaf Removal on Midsouth Winegrape Quality in South Mississippi

Haley Williams* and Eric Stafne

*Mississippi State University, MAFES South Mississippi Branch Experiment Station, 711 W. North St., Poplarville, MS 39470 (hnw111@msstate.edu)

The hot, humid environmental conditions of southern Mississippi hinder the ability to grow quality winegrapes; however, in other locations, canopy management practices have been shown to improve grape quality. Different pruning times and leaf removal were performed on MidSouth grapevines being grown in southern Mississippi to determine how these practices affect vine development and the quality of resultant grapes. Treatments included early (mid-December) versus normal (late-February) pruning, both with or without cluster zone leaf removal. Cluster temperature, total soluble solids, titratable acidity, pH, crop yield, and growth yield data were collected throughout the growing season and then analyzed statistically. Early pruning, both with and without leaf removal, had negative effects on vine growth and grape quality compared to normal-pruned vines. Normal-pruned vines with leaf removal did not differ significantly from normal-pruned vines without leaf removal. Thus, normal

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pruning, whether with or without leaf removal, was superior to early-pruning treatments in the first year of this study.

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Crop Load Adjustment by Canopy Management and its Effect on Fruit and Wine Quality of Tempranillo in Texas

Pierre Helwi,* Justin Scheiner, Andreea Botezatu, Aaron Essary, and Daniel Hillin *Texas A&M AgriLife, Texas A&M AgriLife Research and Extension Center, 1102 E. Drew, Lubbock, TX 79403 (pierre.helwi@ag.tamu.edu)

Crop load is a common measure of yield relative to the size of the producing grapevine and can be managed by viticulture practices such as pruning and shoot- and fruit-thinning. In this project, the crop load of Tempranillo vines from the Texas High Plains AVA was manipulated using three practices to reflect three different crop load levels: vines pruned to two buds per spur (2B), vines pruned to three buds per spur (3B), and vines pruned to three buds per spur and fruit-thinned using a mechanical harvester at 30 days post-bloom (3BFT). Berry chemistry was monitored during the season, and clusters were harvested when the most advanced treatment reached the desired ripening level. Yield at harvest was lowest for the 3BFT treatment that reflected the lowest crop load, followed by 2B and then 3B. 3BFT treatment had the fastest rate of ripening compared to the two other treatments, with higher soluble solids, sugar content, pH, and tartaric acid at harvest. Wines from this treatment were characterized by higher alcohol, pH, more tartaric and malic acids, greater titratable acidity, and were of deeper color. This study shows that crop load, which has a strong influence on berry chemistry at harvest and wine quality, can be adjusted by the level of pruning and/or by fruit-thinning using a mechanical harvester at 30-days postbloom.

Funding Support: Texas Department of Agriculture



Enology—Mathematical Modeling and Method Development Session

Rapid Alcoholic Fermentation Monitoring Using Raman Spectroscopy

Harrison Fuller, James Harbertson,* and Christopher Beaver *Washington State University, 359 University Drive, Richland, WA 99354 (jfharbertson@wsu.edu)

Raman spectroscopy is presented as a tool to monitor enological fermentations. Unlike the spontaneous Raman effect, fluorescence is the result of light-induced change in molecular electronic state. Fluorescence is a highly efficient process and consequently contributes much more signal than traditional Raman scattering. For this reason, the presence of fluorescence has made many instances of Raman spectroscopy applications impractical, as any spectrum generated from fluorescing compounds in a sample masks the more subtle Raman spectrum. This work employs adsorption methods to mitigate fluorescence while monitoring alcoholic fermentation. Ethanol and total sugars (fructose plus glucose) of wines made from red (Cabernet Sauvignon, Merlot, Syrah, Petit Verdot, and blends) and white (Chardonnay) cultivars were modeled using partial least squares regression (PLSR) and Ridge regression. The results comparing predicted to measured ethanol concentrations were respectable for fermenting and finished wines (R^2_{PLS} = 0.93, R^2_{Ridge} = 0.82) and excellent for samples stripped of phenolic contents (R_{PLS}^2 = 0.99, R_{Ridge}^2 = 0.99). Total sugar models also performed well when comparing predicted to measured values (R2_{PLS} = 0.87, R²_{Ridge} = 0.92).

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Alternative Estimation Routines for Modeling and Predicting Commercial Wine Fermentations

Robert Coleman* and Roger Boulton

*Treasury Wine Estates, Beringer Winery, Saint Helena, CA 94574 (bob.coleman@tweglobal.com)

Dynamic models can provide insights into yeast cell performance, nutritional limitation, and the influence of temperature on the rate and completion of wine fermentations. Such models have been used successfully to fit hundreds of complete fermentation density-time data. These models have also been applied from the onset of fermentation to provide a diagnostic and predictive tool for winemakers to intervene in the case of developing sluggish or incomplete fermentations. The models can also have an application in the estimation of cooling loads and the management of refrigeration loads and electrical energy usage during harvest. The Boulton Fermentation Model was used to evaluate the predictive performance of alternative parameter estimation routines using density-time information at early, middle, and late stages of fermentation (20, 12, and 8 Brix) in five white and five red commercial fermentations. This comprehensive set included fermentations of Chardonnay, Sauvignon blanc (2), Riesling, Muscat, Pinot noir, Syrah, and Cabernet Sauvignon (3) and covered fermentation temperatures between 12.8 and 30.6°C (55 and 87°F). The parameter estimation routines (Bard's, Differential Evolution, Genetic Algorithm, and Particle Swarm Optimization) were compared for their ability to reduce the sum of squares deviation and to converge quickly. Prediction improvement was achieved using correlations with initial fermentation temperature to constrain some parameter boundaries during the fitting process. The most efficient parameter estimation routine for reducing the sum of squares error on this wine fermentation test set was Differential Evolution,

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with acceptable performance by all parameter estimation routines. Bard's routine was most efficient in terms of time; however, the resulting parameter fit was more sensitive to the initial parameter guesses.

Funding Support: Treasury Wine Estates

How Are Wine Lipids Profiled in Different Regions?

Quynh Phan and Elizabeth Tomasino*

*Oregon State University, 3051 SW Campus Way, 100 Wiegand Hall, Corvallis, OR 97331 (elizabeth.tomasino@oregonstate.edu)

Lipids found in wine may alter mouthfeel through interactions with phenolic compounds. However, the lipid composition of completed wine has not been investigated. Lipidomic profiling of different grape varieties has been studied for their impacts on fermentation processes and production of wine aroma compounds, but not for their potential impact on wine mouthfeel. We conducted a comparative lipidome analysis of commercial Pinot noir wines produced from Oregon, California, Burgundy, and New Zealand. Lipids investigated in Pinot noir wines include fatty acids (FA), monoglycerides (MG), diglycerides (DG), triglycerides (TG), phosphatidylcholines (PC), phosphatidylethanolamines (PE), lysophosphatidylcholines (LPC), lysophosphatidylethanolamines (LPE), and cholesteryl esters (CE). Lipid-liquid extraction was done to obtain the total lipid content. Liquid chromatographic-mass spectrometry was used for quantitative analyses of wine lipid composition. Advanced multivariate analysis methods, random forest, and linear discriminant analysis showed clear differentiation among wines based on the region of origin. The Pinot noir wines from New Zealand were characterized by having more FA. Pinot noir wines from Burgundy had less TG than the other regional wines. California Pinot noir wines had an equal balance of the different lipids within each class. California and Oregon wines grouped close together for FA. Thus, potentially, lipidomics could be used in wine research as a tool to obtain insight into a wine's geographic origin. In addition, the regional differences in lipid profiles among Pinot noir wines suggested potential impacts of different concentrations and composition of lipids on wine mouthfeel perception.

Funding Support: E & J Gallo Winery



Enology—General Enology Session

The Impact of Storage Conditions (Light Exposure, Temperature, and Bottle Color) on Rosé Wine Quality

Cristina Medina-Plaza, Aubrey DuBois, Elizabeth Tomasino, and Anita Oberholster* *University of California, Davis, Department of Viticulture and Enology, 595 Hilgard Ln, Davis, CA 95616 (aoberholster@ucdavis.edu)

Bottled wine can be exposed to UV-visible light for relatively long periods of time in retail stores, restaurants, or domestic settings. Exposure to light and elevated temperature can decrease wine quality by causing color change and production of off-odors. The impact of bottle color, light exposure, and temperature on rosé wine ageability and quality was studied. Rosé wines with different organoleptic characteristics (color, phenolic, sugar, and alcohol content) were bottled in clear or green bottles and stored at room temperature (22°C) or cellar temperature (12°C) under three different light conditions (darkness, fluorescent bulb, and cool white LED bulb). Wines were analyzed after 0.3, and 6 months of storage. The color and phenolic composition were determined by RP-HPLC and spectrophotometric analysis. Potential changes in aroma were determined through volatile screening of the wines using SPME-GC-MS. Spectrophotometric analysis showed that color intensity decreased over time. The combination of light type and bottle color had the greatest impact on color (CIELAB color space). For example, the color intensity of wines stored in clear bottles decreased more than those stored in green bottles. Under same temperature and bottle conditions, fluorescent light had a larger impact than LED. Wines stored in the dark, independent of bottle type and temperature, were not affected. Higher temperature and/or clear glass increased the percentage of yellow and decreased the percentage of red color in the wines, potentially due to oxidation. There were significant changes in aroma profiles between the starting wines and the different time points. In this case, bottle color had a smaller impact than storage temperature. Overall, while all variables studied affected rosé wine aging significantly, higher temperature in combination with clear glass bottles under fluorescent light was most detrimental to rosé wine aging.

Funding Support: Lyon-Cisneros Family Research Fund

Investigation of the Impact of Pinot noir Maturity on Grape and Wine Composition and Wine Style

Yu-Te Tseng, Emma Sherman, Charles Brenneman, Claire Grose, and Anita Oberholster*

*University of California Davis, 3146 Robert Mondavi Institute - North, Davis, CA 95618 (aoberholster@ucdavis.edu)

The mouthfeel of red wine is directly related to its phenolic composition, which is affected by grape maturity. Sequential harvesting was used to make different wine styles from the same vineyard and to explore markers that can be used to indicate optimal ripeness for a given wine style. This was achieved by making wines from grape skins and seeds with different maturities while using a common base juice (20 Brix base juice from the same vineyard block chaptalized to 23 Brix). This kept the basic chemical composition (matrix) of the wines (referred to as juice-skins wines) constant, with mainly phenolic ripeness differences. These wines were compared with control wines made using standard winemaking practices. Pinot noir grapes were harvested at 23 and 25 Brix and pressed gently to remove the juice to substitute with the base juice. Basic chemical composition and phenolic and tannin compositions of grapes and wines were determined by the Adams-Harbertson assay, RP-HPLC, phloroglucinolysis, and gel permeation chromatography. Multivariate statistics were

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used to relate compositional data to descriptive analysis. Wines made with more mature grapes contained higher concentrations of monomeric and polymeric phenols; however, no significant differences in monomeric anthocyanins were observed. Juice-skins wines had lower polymeric phenol concentrations than control wines, and this difference increased when wines were made with fruit with greater maturity. Mouthfeel attribute "hot" and "persistence" and "dark fruit" aromas mainly described the wines made from higher maturity fruit. The study showed that wines made with riper fruit, irrespective of must sugar content, were characterized by greater phenolic content and ripe fruit characters.

Funding Support: Bragato Research Institute (New Zealand grape and wine research)



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Does Xiphinema americanum Vector Grapevine Fanleaf Virus?

Cecilia B. Aguero,* Xuyyn Yang, Liang Zheng, Andy Nguyen, Howard Ferris, and M. Andrew Walker

*Department of Viticulture and Enology, UC Davis, Robert Mondavi Institute, 595 Hilgard Lane, Davis, CA 95616 (cbaguero@ucdavis.edu)

Fanleaf degeneration is a severe disease of grapevines due to its effect on fruit set. It is caused by grapevine fanleaf virus (GFLV) and vectored from root-to-root by the dagger nematode. Xiphinema index. The objective of this research was to study the ability of a closely related dagger nematode, Xiphinema americanum, to vector GFLV. X. americanum is common in California vineyard soils. Experiments were performed to determine whether this nematode can vector GFLV among grapevines. Roots of in vitro plantlets of GFLV-infected Chardonnay were inoculated with 10 X. index or X. americanum. After one week, nematodes were transferred to roots of healthy St. George plantlets for another week and then removed. Three weeks later, RNA was extracted from roots and cDNA was synthesized, followed by determination of viral load by qPCR. In a similar experiment conducted under greenhouse conditions, GFLV-infected Chardonnay plants were grown in 4-L pots filled with autoclaved sand. Each pot was inoculated with 1 mL inoculum containing 50 nematodes. Five pots were inoculated with X. index, five with X. americanum, and three with water. Three weeks later, healthy St. George plants were transplanted to each pot, so that every pot contained one GFLV-infected Chardonnay and one healthy St. George. Four months later, gPCR was used to test all St. George plants. In both experiments, only plants inoculated with X. index were clear positives for GFLV, although inoculation efficiency was low, with three positives out of five under in vitro conditions and only one positive in the greenhouse testing. The latter outcome may be due to the low number of nematodes used as inoculum in proportion to the volume of soil. These results, however, suggest that X. americanum does not transmit GFLV, or at least that it is not as effective as X. index.

Funding Support: California Grape Rootstock Research Foundation

Effect of Shoot Trimming on Yield, Fruit Maturity, and Reserves in Malbec Vineyards under a Single High-Wire Trellis System

Gaston E. Ahumada,* Miguel A. Pirrone, Diana Segura, Marcelo J. Belmonte, Carla V. Giordano, and Carina V. Gonzalez

*Grupo Peñaflor S.A., Nueva Mayorga s/n, Coquimbito, Maipu, Mendoza Argentina, 5513, Argentina (gaston.ahumada@grupopenaflor.com.ar)

Argentina is the world's fifth largest wine producer and Malbec is its most emblematic variety. Vineyards producing high-quality winegrapes are mainly trellised to vertical shoot-positioned systems. The single high-wire (SHW) system is a highly productive free canopy trellising/training system used in many world wine regions. We evaluated, for the first time in Argentina, the effect of different shoot-trimming levels on leaf area development, yield, grape maturity, and reserve levels of Malbec (clone 598) from SHW-trellised vineyards in Mendoza (Winkler = V). We imposed different total leaf areas (TLA) by shoot-trimming, varying shoot length at full bloom (0.45 m, 0.80 m, or untrimmed) during three growing seasons (2017, 2018, and 2019). We measured yield and fruit composition at harvest and soluble and structural reserves in trunk and canes at leaf fall. Different shoot-trimming levels produced different total leaf area development. The 0.8 m treatment produced 30% less TLA than the untrimmed treatment, consistently among seasons. The treatments affected yield

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differentially by season. Overall, the untrimmed treatment had the greatest yields. The 0.80 m treatment, while producing high yields in the second season, was not sustained in the third season and declined to similar yield values as the 0.45 m treatment. This drop in yield could be due to the decrease of starch in the trunk during the second season. On the other hand, grape maturity was greater in the 0.80 m and untrimmed treatments. Our results suggest that the untrimmed treatment produces greater sustained yields over time, with good levels of reserves and grape maturity.

Funding Support: Instituto de Biología Agrícola de Mendoza, FCA UNCuyo-CONICET

Metabolomic Profiling of Smoke-Exposed Red Wine by LC-HRMS/MS

Armando Alcazar Magana, Michael Qian,* Ruiwen Yang, and Yanping L. Qian *Oregon State University, 100 Wiegand Hall, Corvallis OR 97331, Corvallis, OR 97331 (michael.qian@oregonstate.edu)

The United States wine industry is facing smoke-exposure issues due to the increasing incidence of wildfires. Exacerbated by climate change, wildfires have increased in both number and size in recent years, intensifying smoke exposure issues in wine. Smoke taint is an off-aroma described as smoky, dirty, burnt, medicinal, and ashy character in the wine. The off-aroma is caused by grape or grapevine exposed to bushfire smoke before harvest. During a wildfire, smoke constituents like volatile phenols can permeate the grape cuticle and bind to sugar molecules, creating volatile phenol glycosides. These glycosides can be hydrolyzed during winemaking, releasing the aglycone volatile moieties such as guaiacol or cresols into the wine. Gas chromatography-mass spectrometry (GC-MS) is frequently used to study volatile phenols in smoke-exposed wine after hydrolyzing the glycosides. Although GC-MS offers a powerful tool to analyze volatile phenols, it lacks the capability to determine the chemical nature of intact phenolic glycosides or other smoke precursor compounds. We employed a state-of-the-art untargeted metabolomics approach using high-performance liquid chromatography combined with high-resolution accurate mass LC-MS/MS and the latest generation of bioinformatics tools to discriminate smoke-related compounds present in wines from 2020 wildfire-exposed and control wines (N = 14 for control wines; N = 26 for smoke-tainted wine). Using this technique, we detected hundreds of molecular features (containing structural and elemental composition) only present in smoke-exposed wine and dozens of volatile phenol glycosides. Univariate data analysis was used to establish differences or similarities between control and smoke-exposed wines. By understanding the chemical nature of phenolic glycosides and other smoke-related compounds, different strategies can be explored to reduce smoke compounds' negative impact on wine quality.

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Fungicide Resistance of *Botrytis cinerea* Populations on Winegrapes in the Central Coast of California

Evelyn Alvarez-Mendoza, Samantha Anderson, Stephen Chang, and Shunping Ding* *California Polytechnic State University, San Luis Obispo, 1 Grand Avenue, San Luis Obispo, CA 93407 (sding01@calpoly.edu)

Botrytis bunch rot, caused by *Botrytis cinerea*, is of major concern to grapegrowers and wine producers in California due to its significant crop losses and reduced fruit and wine quality. The increasing occurrence of fungicide resistance in *B. cinerea* populations poses a major challenge for effective management of this disease in

*indicates corresponding author



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vineyards. In 2020, 35 *B. cinerea* isolates were isolated from grape cluster samples collected from six conventional and two organic vineyards located in Santa Barbara County and San Luis Obispo County of California. The 35 *B. cinerea* isolates were screened against the fungicide Scala, a fungicide commonly used to manage Botrytis bunch rot, to determine their sensitivity to the active ingredient, pyrimethanil. Isolated pure cultures of *B. cinerea* were grown on 0.5% sucrose agar amended with Scala at two discriminatory concentrations, 0.1 µg/mL and 10 µg/mL. Radial mycelial growth was measured following a seven-day incubation period. Measurements were converted to percent relative growth values for four single-colony replicates per isolate. Isolates were classified into four sensitivity categories that consisted of highly sensitive, moderately sensitive, highly resistant, and moderately resistant. Of the 35 isolates screened, 88.6% exhibited high resistance, 5.7% exhibited moderate sensitivity to pyrimethanil. The results from this study can be used by grapegrowers in the Central Coast of California to implement effective fungicide spray programs for control of Botrytis bunch rot in winegrapes.

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Effect of Antiochratoxigenic Compounds on Alcoholic Fermentation by *Saccharomyces cerevisiae*

Gisselle R. Apud, Diego A. Sampietro, and Pedro A. Aredes Fernández* *Facultad de Bioquímica, Química y Farmacia - Universidad Nacional de Tucumán (UNT) - CONICET, Ayacucho 471, San Miguel de Tucumán/Tucumán/4000), Argentina (pedroaredes@hotmail.com)

Ochratoxin A (OTA) is a mycotoxin with harmful effects on health. Its presence in grapes may lead to wine contamination. Aspergillus carbonarius and Aspergillus niger are considered the most dangerous sources, as they have the greatest potential for OTA production in grapes. In previous work, we demonstrated that an antifungal constituent identified as 2-hydroxy-3-(3-methyl-2-butenyl)-1,4-naphthoquinone (lapachol) combined with sodium metabisulfite completely suppressed OTA accumulation at concentrations of 9.8 and 156.3 μ g/mL, respectively. This work evaluated fermentation by the commercial yeast Saccharomyces cerevisiae EC-1118 under vinification conditions in the presence of the mixture of lapachol and sodium metabisulfite. Alcoholic fermentation was performed in flasks with a previously pasteurized commercial white grape juice (22 Brix, pH 4.1) inoculated with S. cerevisiae EC-1118 at a concentration of 7×10^6 cells/mL (OD_{620nm} = 0.5) and supplemented with sodium metabisulfite (156.3 μ g/L) as the control medium, with or without the mixture of lapachol and sodium metabisulfite (9.8/156.3 μ g/L). The flasks were corked with an air trap with sulfuric acid to avoid liquid evaporation and incubated at 22°C. Fermentation was monitored through release of CO_2 by determining weight loss of the system. At the end of fermentation, yeast cells were removed and the ethanol, pH, and sugar consumption (Brix and reducing sugars) were determined in wines. The presence of lapachol and sodium metabisulfite did not affect the yeast, which achieved a complete fermentation. The resulting wines were similar in all parameters evaluated (ethanol 12%, pH = 3.8, 5.2 Brix, and 4 g/L of residual reducing sugars). We conclude that lapachol has potential use as a complement for sodium metabisulfite against OTA production by Aspergillus species without affecting fermentation.

Funding Support: Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET)

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Proteolytic Activity of Strains of *Oenococcus oeni* during Malolactic Fermentation

Irina Kristof, Nancy Roxana Vera, and **Pedro Aredes Fernandez*** *Facultad de Bioquímica Química y Farmacia - Instituto de Biotecnología - UNT, Ayacucho 471, 4000, Argentina (pedroaredes@hotmail.com)

During cider production, malolactic fermentation (MLF) is usually carried out by Oenococcus oeni. Proteolytic activity from O. oeni during MLF releases peptides and amino acids with an important nutritional role for bacterial viability. These released peptides may also benefit human health. The objective of this study was to evaluate the proteolytic activity of three strains of O. oeni against the proteins present in apple juice and the "in vitro" antihypertensive activity of the released peptides. Pasteurized apple juice was fermented with a commercial strain of Saccharomyces cerevisiae. The fermented juice was homogenized and divided into two batches. One batch was centrifuged, filtered, and pasteurized, the other with yeast cells was reserved without pasteurization. Both fractions, with and without yeast lees, were used to perform the MLF with three different strains of O. oeni, Strains RAM10 and RAM11 were isolated from Malbec wine from Tucumán. The VP41 strain was obtained from Lallemand (commercial strain). The strains were separately inoculated at a cell concentration of 106 cfu/mL into each fraction of fermented juice. Proteolytic activity and the concentration of peptides and amino acids were determined in supernatants. The ability of supernatants to inhibit the angiotensin-converting enzyme (ECA) was evaluated by the Cushman and Cheung method. The maximum proteolytic activity of the studied strains was detected in the supernatants after 24 hours of FML, with or without lees. In general, the proteolytic activity was greater in the absence of lees and greatest in the RAM11 and VP41 supernatants. The greatest inhibitory activity on ECA was detected in supernatant without lees fermented by RAM11 and the supernatant with lees fermented by strain VP41. The supernatant of the strain RAM10 fermentation had less inhibitory activity against ECA.

Funding Support: Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET)

Preservation of *Oenococcus oeni* by Different Methods in a New Culture Medium

Jose Martinez Chamas, Maria Cristina Rubio, and **Pedro Aredes Fernandez*** *Facultad de Bioquímica Química y Farmacia - Instituto de Biotecnología - UNT, Ayacucho 471, 4000, Argentina (pedroaredes@hotmail.com)

The preservation of starter cultures for malolactic fermentation (MLF) of wines is necessary for their following use. The conservation is regularly carried out at low temperatures in a suitable medium. The conservation of a starter culture and its fermentation activity depend on the method of conservation selected and the environment in which the conservation is carried out. The *Oenococcus oeni* RAM10 strain was grown in a novel designed medium (M7) to promote biomass production and improve malolactic activity. We evaluated the effect of the medium on cell viability and the loss of fermentative capacity (LFC) after culture conservation by refrigeration, freezing with glycerol, and lyophilization with glutamate, fructose, and grape juice as protective agents. Grown and conserved cultures in M7 showed less LFC than those preserved in conventional MLO (control medium) for refrigeration and freezing conservation methods. The viability did not show significant differences for both media during refrigeration. The conservation of *O. oeni* by freezing in both



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media with glycerol exhibited greater cell viability. However, cultures conserved in M7 induced MLF with lower LFC (17%) than MLO, which had an LFC of 24% until three months of storage. Lyophilization in MOP7 in the presence of glutamate and grape juice maintained high viability until six months of storage, with a LFC of 20 and 25%, respectively. Fructose showed a lower lioprotective effect and a high LFC at the same time. Storage in MOP7 permitted conservation of cultures with the least LFC after refrigeration, freezing, or lyophilization methods. Lyophilization using grape juice as the lioprotector preserved the cultures for at least six months. This protective agent has the advantages of being inexpensive and easy to access.

Funding Support: Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET)

Effect of Planting Stock and Training Strategy on Early Development and Productivity of Pinot noir Grapevines

Larry Bettiga*

*University of California Cooperative Extension, 1432 Abbott Street, Salinas, CA 93901 (lbettiga@ucdavis.edu)

Plant material and training strategies used are critical factors in promoting vine development and production. The objective of this study was to evaluate nursery stock and training strategies for their potential to advance vine development and yield. A trial was established in a Pinot noir vineyard in the Salinas Valley of California to compare standard 30 cm-long dormant benchgrafts to 90-cm tall benchgrafts that were produced by using a longer rootstock cutting from 2015 to 2019. The treatments were: 1) standard field-grown dormant, 30 cm; 2) tall dormant potted, 90 cm; and 3) tall green-growing potted, 90 cm. The tall vines were trained to bilateral cordons in the first year where growth was adequate. Standard vines were trained to a single trunk shoot at the end of the first year, and cordon training started in year 2. The dormant tall benchgrafts at the end of year 1 produced vines with larger diameter trunks and growth was adequate to form the cordons. In years 2 and 3, dormant tall vines had larger trunk and cordon diameters and pruning weights, standard was intermediate, and tall green-growing had the smallest diameters and pruning weights. In year 2, dormant tall vines produced the greatest yield; standard intermediate and green-growing tall vines, the least. In year 3, the dormant tall and standard vines produced similar yield, and green-growing tall vines had lower yields. The results from this trial suggest that both plant material and vine training method in the first year could advance development of the permanent framework of the vine and promote the potential for earlier vine production, especially when either factor improves total vine growth in the year of planting and that growth increase is used to form the vine's permanent framework.

Funding Support: Various donors

Five-Year Chemical Aging Profiles of Two Red and Two White Wines Stored Under Four Closures

Annegret Cantu, Andrew L. Waterhouse,* Mauri Anderson, Patricia A. Howe, and Nicolas Delchier

*UC Davis Department of Viticulture and Enology, One Shields Avenue, Davis, CA 95616 (alwaterhouse@ucdavis.edu)

Understanding the effect of various closures on wine aging is an ongoing research

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effort globally. Innovative technical closures avoid development of "cork taint" and offer a range of controlled oxygen transfer rates for postbottling storage of different wine styles. This five-year project aimed to compare various closures' effects on aging characteristics of four wines. The wines were all high-quality and commercially available. These included a Sauvignon blanc, a barrel-aged Chardonnay, a Pinot noir, and a Cabernet Sauvignon. All wines came from wineries in Napa and Sonoma, California, and were aged under controlled temperature for five years. The four closures tested were three DIAM controlled-oxygen transfer rate (OTR) closures and a standard 49mm natural cork. We measured free and total sulfur dioxide levels, color, and phenolic profiles. As expected, white wines were more susceptible to the effects of oxidation; thus, their aging profiles were more dependent on the level of oxygen allowed by the different closures we tested. Generally, we observed a difference in oxygen exposure on browning and rates of sulfur dioxide loss. The choice of closure impacted the wine composition over five years but had less impact on red wines than whites. Overall, red and white wines closed with the DIAM technical closures, especially those with lower OTRs, had significantly less variation in the final sampling for sulfur dioxide levels. Furthermore, from these results, we could conclude that red wines were more resilient to oxidation in the bottle and manufactured closures, like DIAM closures. provided a more uniform end product after some time in the bottle. The results also suggest that the choice of a low-OTR closure is more critical for storage and aging of white wines.

Funding Support: DIAM Bouchage

Application of *Hanseniaspora vineae* Hv205 During White and Red Grape Vinification at Diverse Winery Scales

Valentina Martin, Valentina Olivera, Laura Farina, Eduardo Boido, Eduardo Dellacassa, Remi Schneider, Albert Mas, Tomas Roman, Antonio Morata, and **Francisco Carrau**^{*} *Universidad de la Republica, Av. Gral. Flores 2124, 11800, Uruguay (fcarrau@fq.edu.uy)

Vinification is the process in which yeasts ferment grape sugars to produce ethanol, carbon dioxide, and secondary metabolites. Many wine aroma compounds are derived from yeast secondary metabolism, during which different compounds form that contribute to the aroma profile, color, and mouthfeel. The composition of wines associated with these metabolites is key to obtain wines with different character and typicality. Recent laboratory- and pilot-scale research has examined the behavior of native non-Saccharomyces yeasts and their influence on the production of these compounds. At commercial level, winemakers have limited access to yeast diversity, in contrast to the great strain diversity found on grape skin surfaces: the microbial terroir. We focused on Hanseniaspora vineae because of its proven fermentative capacity and low volatile acidity formation when compared to other species of this genus and its contribution to the increased aromatic profile of wines with floral, fruity, and mouthfeel descriptors. The objective of this work was to carry out fermentations with the selected strain Hv205 and with diverse grape varieties in wineries located in Uruguay, Italy, and Spain during two harvests. Chemical and sensory analyses were performed on wines elaborated using Chardonnay, Macabeo, Albillo, Trebbiano, Ugni blanc, and Semillon white grapes and Tannat, Tempranillo, and Pinot noir red grapes. The results allowed us to conclude that the liquid and dry active formulations of H. vineae Hv205 were compatible with fermentation at industrial scale for base sparkling, white, and red wines. There were significant increases in aromatic compounds such as benzenoids and acetates. Furthermore, polysaccharides and other



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compounds derived from yeast cell lysis might explain the increase in roundness perception in mouthfeel compared to *Saccharomyces* strains. Likewise, the wines obtained were preferred and differentiated from those fermented with conventional commercial yeasts.

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Selection of the Best *Saccharomyces* Strain Partner for Vinification under Mixed Cultures with *Hanseniaspora vineae* Hv205

Valentina Olivera, Maria Jose Valera, Valentina Martin, Gabriel Perez, Eduardo Boido, Remi Schneider, Eduardo Dellacassa, and **Francisco Carrau*** *Universidad de la Republica, Av. Gral. Flores 2124, 11800, Uruguay (fcarrau@fq.edu.uy)

Yeasts produce a diversity of secondary metabolites during fermentation that impact the flavor and aroma of wine. Among apiculate yeasts of the genus Hanseniaspora, the species Hanseniaspora vineae has been used at the winery scale with successful results, producing aroma compounds that expand the diversity of wine color and flavor. However, this species tolerates only moderate levels of ethanol (-10% v/v). The implementation of a mixed culture with Saccharomyces cerevisiae under controlled experimental conditions could be a useful strategy to obtain complete fermentations of mature grapes with increased flavor complexity. Co-inoculation with H. vineae strain 205 avoids the risks of undesirable yeasts that might have appeared under spontaneous fermentations but increases strain diversity during fermentation. However, some mixed culture fermentations are not successful due to nutrient competition and may require supplementation with assimilable nitrogen or vitamins. This work focuses on selecting commercial S. cerevisiae strains that can grow and ferment together with Hv 205 to obtain a complete fermentation and on how nutrient addition effects improve mixed-culture performance. Of 23 commercial yeasts evaluated, we present results of the best six S. cerevisiae partners to ferment with Hv 205 under sequential inoculation at 72 hrs. At the end of the fermentation, sensory evaluation and chemical analysis of aromas by gas chromatography-mass spectrometry were performed. Fermentation and sensory analysis in mixed cultures with S. cerevisiae strains 3C and E73 proved the best combinations for wine quality. More intense fruity and floral aromas were found than in conventional, single-strain fermentations. Nutrient additions such as diammonium phosphate, amino acids, yeast extract, the commercial additive Natuferm, and thiamine were evaluated when Saccharomyces was inoculated to improve completion of fermentations.

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White Wines Produced by the Two Main Yeast Species of the Fermentation Clade within the Apiculate Genus *Hanseniaspora*

Maria Jose Valera, Valentina Olivera, Eduardo Boido, Eduardo Dellacassa, and **Francisco Carrau***

*Universidad de la Republica, Av. Gral. Flores 2124, 11800, Uruguay (fcarrau@fq.edu.uy)

Hanseniaspora species are the main yeasts isolated from grapes and grape musts. The genus has two clear technological clusters, the fruit group and the fermentation group, based on genetic and phenotypic characterization. Among the Hanseniaspora species belonging to the latter, *H. osmophila* and *H. vineae* have been found in

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spontaneous fermentations of grape must. These yeasts are characterized by their increased capacity to ferment over other apiculate yeasts. However, there are some differences between these two species that make them interesting from an enological point of view. H. vineae is well known for its ability to improve wine flavor. Genomic, transcriptomic, and metabolomic studies of H. vineae have enhanced our understanding of its utility within the wine industry. H. osmophila has been less characterized in wine production, although its capacity to increase pleasant aromas in red wines was reported recently. The objective of this study was to evaluate differences in wine fermentation performance between these two Hanseniaspora species. Single-strain microfermentations were carried out with Chardonnay grape must inoculated with H. vineae Hv205 and H. osmophila AWRI3579. In addition, H. uvarum AWRI1280, representing the Hanseniaspora fruit clade, and the industrial strain Saccharomyces cerevisiae ALG804 were used as controls for fermentation performance. Wine aromas were characterized chemically by gas chromatography-mass spectrometry and a descriptive sensory analysis was performed by an expert panel of tasters. Both species contributed positive aroma to final wines, while no defects were detected compared to H. uvarum. Wines fermented with H. vineae had significantly greater concentrations of 2-phenylethyl, tryptophol, tyrosol acetates, acetoin, and benzyl alcohol than H. osmophila wines. Sensory analysis showed increased intensity of fruity and flowery notes in wines fermented with H. vineae. Higher gene copy numbers of putative acetyl transferases found in *H.vineae* compared to *H. osmophila* might explain the increased capacity for acetate synthesis.

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Viticultural Performance of Four Petite Sirah Clones and Determination of Their Respective Wine Characteristics

Raul Cauduro Girardello, Anita Oberholster,* Andrew Walker, S. Kaan Kurtural, and Danielle Zaccaria

*University of California, Davis, 1 Shields Ave, Davis, CA 95616 (aoberholster@ucdavis.edu)

Grapevine cultivars have several clones that may vary for specific viticultural and enological characteristics such as yield, cluster and berry size, and berry composition. Vitis vinifera cv. Petite Sirah, also known as Durif, has become an important component of red wines in California. However, little is known regarding the viticultural and enological traits of Petite Sirah clones. This study evaluated phenotypic and enological traits of four clones of V. vinifera L. cv Petite Sirah grapevines. This study was performed in 2019 and 2020 at the University of California, Davis, where four Petite Sirah clones (Durif FPS1, FPS3, FPS4, and FPS5) were planted in 2010 in a complete block design. For each clone, four biological repetitions composed of five grapevines were evaluated for viticultural attributes such as number of clusters/vine, cluster weight, cluster morphology, number of berries/cluster, berry weight, pruning weight, and yield. Grape berries were collected at harvest and analyzed for basic chemical composition (Brix, pH, titratable acidity [TA]), phenolic profile by RP-HPLC and protein precipitation assay, and volatile composition by HS-SPME-GC-MS. Wines were made from each clone and analyzed for chemical and phenolic composition and aroma volatile profiles, while wine sensory attributes were evaluated by descriptive analysis. No major differences were observed in phenotypic traits such as cluster weight and berries/cluster among the clones. However, yield, pruning weight, berry weight, Ravaz index, and berry chemical and phenolic composition were demonstrated to be distinct among the clones. For instance, clones FPS 3 and FPS 5 had significantly

*indicates corresponding author



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more anthocyanin, flavanol, and hydroxycinnamic acid concentrations than FPS 1 and FPS 4. Clonal wines also exhibited unique volatile profiles that correlated well with the sensory differences among the wines.

Funding Support: Bogle Vineyards

Application of Smoke Containing ¹³C Isotopes to Winegrapes for Chemical Characterization of Smoke Influences in Wine

D. Cole Cerrato, Michael Penner, and Elizabeth Tomasino* *Oregon State University, 100 Wiegand Hall, Corvallis, OR 97333 (elizabeth.tomasino@oregonstate.edu)

Smoke exposure to winegrapes is a growing concern for vintners, winemakers, and consumers as many vineyards in the United States and Australia reside in areas where wildfire intensity has been increasing since at least the 1950s. Wine is particularly sensitive to smoke exposure, often acquiring "ashy," "burnt rubber," or "medicinal" flavors or aromas after grapes are exposed to wildfire smoke and then made into wine. To better serve the industry, we have designed a method to perform a more thorough inventory of chemical compounds associated with smoke. A fuel source for smoke, barley, was chemically labeled using the stable isotope of ¹³CO₂. Upon assimilation after 10 days of ¹³CO₂ exposure, the ¹³C is expected to be incorporated into smoke precursor compounds such as lignin. After drying, the barley was burned and the smoke piped "cold" to Chardonnay and Pinot noir grapes grown in the Williamette Valley, Oregon, postharvest in a sealed container designed for this study. Chemical analysis using RP-HPLC, mass spectrometry, and ¹³C-NMR will be used to elucidate the chemicals potentially responsible for smoke flavors and aromas in affected wines.

Funding Support: American Vineyard foundation USDA-NCSFR

Grape Leaves May Be More Vulnerable to Heat Waves in Cool than in Warm Seasons

Ben-Min Chang and Markus Keller*

*Washington State University, 24106 N. Burrn Rd., Prosser, WA 99350 (mkeller@wsu.edu)

Because of the rising atmospheric carbon dioxide concentration, the frequency and scale of extreme weather events is increasing. Grapevines can acclimate to a stable temperature trend and optimize their physiological performance at that condition. The magnitude of temperature change determines how well the vines adapt to the new condition. However, a sudden, large change in temperature can compromise productivity and fruit quality. We developed a mist-type evaporative cooling system to control canopy temperature without compromising deficit irrigation and stimulating vegetative growth. In the current configuration, the threshold to activate misting is arbitrarily and inflexibly set at 35°C. To optimize operation of the canopy cooling system, knowledge about how grapevines acclimate to temperature change is essential. We grew potted Cabernet Sauvignon vines in growth chambers at high (32°C/15°C) and low (27°C/10°C) maximum/minimum temperature regimes for 10 days, simulating hot and cool growing seasons in eastern Washington. Then the vines were exposed to a spectrum of maximum temperatures varying from 40°C to 25°C, maintaining a 17°C difference between daily maximum and minimum temperature. Gas exchange rate was monitored at noon and late afternoon. The results suggested that grapevines are more vulnerable to a sudden temperature spike (heat wave) in a cool season than in a warm season. The photosynthesis rate and stomatal conductance at noon

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were depressed when the cool-acclimated vines were suddenly exposed to higher temperature. In the late afternoon, vines from both acclimation regimes showed gas exchange depression when the vines were exposed to temperatures above the acclimation conditions. Overall, the temperature acclimation experiment suggested the temperature which causes heat stress is flexible and depends on previous environmental conditions. Hence, using a dynamic activation threshold temperature for the cooling system might maintain vine gas exchange capacity better than a fixed threshold temperature.

Funding Support: Northwest Center for Small Fruits Research; USDA Specialty Crop Block Grant Program; Washington State Grape and Wine Research Program

Sodium Chloride Application Decreases Photosynthetic Activity in *Vitis* spp. Regardless of Exposed Rootstock Selection

Christopher Chen,* Nina Romero, and M. Andrew Walker *University of California Davis, 1 Shields Ave, Davis, CA 95616 (codchen@ucdavis.edu)

As our climate changes and temperatures rise, water scarcity and high evapotranspiration has led to a greater buildup of sodium chloride in agricultural soils. Grapevines, like most perennial crops, are glycophytic and lack sufficient responses to high soil salinity in their immediate rooting zone. Salt toxicity can lead to detrimental effects on growth, yield, and overall vine health. There is a need to understand how NaCl affects grapevine photosynthesis and address the potential for different forms of salt tolerance in grapevines. Measurements of transpiration, stomatal conductance, and net assimilation rates were conducted across two trials on grapevine rootstocks exposed to NaCl concentrations of 0, 25, 50, 75, and 100 mM. Because several rootstock selections varied between experiments, five selections were shared across both trials for continuity. The preliminary trial tested the photosynthetic response of eight rootstocks when irrigated with NaCl solution at concentrations of 0, 25, 75, or 100 mM NaCl over 21 days. Differences between selections were only significant at concentrations of 75 mM or greater. A second trial was conducted with nine rootstock selections, all grafted to Cabernet Sauvignon and similarly exposed to 50 mM NaCl. Three measurement time points showed a delineation in photosynthetic response early in the latter trial. However, the observed differences were no longer significant following 21 days of exposure. These trials quantify the response of different Vitis spp. rootstocks under varying NaCl concentrations. More importantly, they explore the potential for translating salt-tolerant qualities of rootstocks to grafted scions in field conditions.

Funding Support: California Grape Rootstock Improvement Commission

How Do Accentuated Cut Edges and Macerating Enzyme Affect Phenolic Compounds Extraction on Marquette Wines?

Yiliang Cheng and Aude Watrelot*

*Iowa State University - Department of Food Science and Human Nutrition, 2567 Food Science Building, 536 Farm House Ln, Ames, IA 50011 (watrelot@iastate.edu)

Accentuated cut edges (ACE) and macerating enzyme are a mechanical and a biological process that breaks grape skins into small fragments and breaks down skin cell wall polysaccharides, respectively. Both winemaking techniques have shown an enhancement of extractability of skin-derived phenolic compounds from *Vitis vinifera* grapes. However, the role of phenolic compounds and cell wall material on phenolic



compound extraction and red wine quality are still unclear. In this study, Marquette red wines were made under three treatments applied at crush: control, ACE, and pectic enzyme (ENZ). The wine collection was processed throughout the winemaking process up to bottling. The wine hue and color intensity were evaluated using a UV-visible spectrophotometer. The composition of monomeric polyphenols was analyzed by RP-HPLC with diode array and fluorescence detectors. The concentration of flavan-3-ols and anthocyanins increased during fermentation and reached the highest concentrations at bottling. Flavonols and hydroxycinnamic acids had the lowest concentration in wines. At bottling, ACE-treated wine showed the greatest color intensity, followed by ENZ-treated wine, and both treatments showed a significant decrease in hue compared to control wine. ACE treatment significantly increased the concentration of flavan-3-ols and hydroxycinnamic acids. There was no significant difference in the concentrations of anthocyanins and total phenolic compounds content among all treatments. The greater color intensity of treated wines suggested that some polymeric pigments were formed during winemaking. To investigate the role of these treatments, ACE and enzymes, on skin tannin extraction, further work will focus on condensed tannin content and chemical structure in control and treated wines.

Funding Support: N/A

Aroma Sensory Interactions Between *cis*-Rose Oxide, Linalool, and α -Terpineol in Gewürztraminer Wines

M. Melina Chigo Hernandez and Elizabeth Tomasino* *Oregon State University, 100 Wiegand Hall, Corvallis, OR 97331 (elizabeth.tomasino@oregonstate.edu)

The aroma of white wine is an important part of wine quality. Traditionally, it was thought that only compounds at high concentrations influenced aroma perception. However, it has been shown that low-impact odorants may act to change the perception of other odorants in a mixture and can significantly impact aroma perception. Monoterpenes are found in their enantiomeric or isomeric forms in grapes and wine; these isomers may have similar odors and thresholds. For other monoterpenes, the isomeric forms present different characteristics and thresholds. Monoterpenes are found at high concentrations in aromatic wines such as Gewürztraminer, with cis-rose oxide the most characteristic odor compound associated with this varietal. We evaluated sensory perception of different ratios of *cis*-rose oxide in mixtures with other monoterpenes and assessed aroma interactions with other monoterpenes. Triangle tests were used to determine whether the ratios of *cis*-rose oxide isomers found in wine alter aroma perception. CATA was used to determine the descriptors associated with wines created to contain different concentrations of cis-rose oxide, linalool, and α -terpineol isomers. Descriptive line scales were used to determine the aroma impact of the different monoterpene combinations. The question being investigated was, is cis-rose oxide the main driver of aroma in Gewürztraminer wines, or are other monoterpenes also important to aroma perception?

Funding Support: Fullbright Scholar Program

Use of VIS-NIR Hyperspectral Imaging to Quantify Anthocyanins, Soluble Solids, and Titratable Acidity in Grape Berries

Paolo De Censi, Khushwinder Singh, Alvaro Sanjuan, and Luca Brillante* *Department of Viticulture and Enology, California State University Fresno, 2360 E Barstow Ave, Fresno, CA 93740 (lucabrillante@csufresno.edu)

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Monitoring ripening is one of the most time-consuming laboratory procedures in grape and wine. The procedures are relatively long and complex and require extensive lab training and important labor expenses. The techniques are destructive and do not allow repeated measurements of the same samples. It is not possible to characterize spatial variability and the ripening process without large numbers of samples, thus incurring large costs. Hence, it is desirable to find new, faster techniques that are less expensive and produce spatial-temporal information to empower precise viticulture approaches. This study assessed grape skin flavonoids, total soluble solids, and titratable acidity in intact berries using a hyperspectral camera. The samples were collected in different vineyards and varieties to ensure a large variability in target compounds. In 2020, we collected ~400 samples of Cabernet Sauvignon and Cabernet franc berries in Madera, Paso Robles, and Rutherford, California, on different dates, but always after veraison. Samples were composed of 20 berries each. Each sample was imaged in a dark room in the lab using a VIS-NIR hyperspectral camera (500 to 900 nm), then peeled. From the flesh, we obtained juice for measuring total soluble solids and titratable acidity; from the skins, we obtained the anthocyanin profile through high-performance liquid chromatography. The reflectance spectra of the camera were related to the content in soluble solids, acids, and anthocyanins through random forest regression, and good correlations were found that predicted grape composition with a small associated error. These models will be deployed under field conditions by mounting the camera on a tripod, an ATV, or a conveyor belt for real-time monitoring of grape composition.

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Tailoring Smart Irrigation Strategies for White Wine Grapes in Eastern Washington

Geraldine Diverres* and Markus Keller

*Washington State University, 24106 N Bunn Road, Prosser, WA 99350 (r.diverresnaranjo@wsu.edu)

Inadequate irrigation management can limit vineyard productivity and fruit quality, diminishing potential revenues and resource-use efficiency. Regulated deficit irrigation (RDI) and partial rootzone drying (PRD) were compared to non-stress irrigation (FULL) in a Riesling vineyard in the Yakima Valley. This project's objective was to determine the usefulness and sustainability of each technique for high-quality white winegrape production in eastern Washington. The results show that canopy growth, canopy architecture, and final yield were significantly affected by the irrigation strategy. RDI vines had smaller canopies with greater sun exposure in the fruiting zone and their harvest yield was lower than those of PRD and FULL vines. On the other hand, PRD vines had denser canopies, less-exposed clusters, and did not show differences in yield compared to FULL. No differences were found between any treatments in terms of fruit composition at harvest (total soluble solids, pH, and titratable acidity). Given these results, PRD may allow considerable reduction in water use for white winegrape production without affecting yield and quality in arid climates like eastern Washington.

Funding Support: NSF/USDA Cyber Physical Systems Program and the Washington State Grape and Wine Research Program

Effect of Forest Fire Smoke and Potentially Mitigating Sprays on Fungal Communities of Grapes in Three Vineyards

Sarah Marie Lyons, Wesley Zandberg, James Favell, and Daniel Durall*



*University of British Columbia (UBC), 1177 Research Rd, Kelowna, British Columbia V4V 1N7, Canada (daniel.durall@ubc.ca)

The increase in forest fires in recent years has had a large effect on North American wineries. When grapes in vineyards are exposed to large amounts of forest fire smoke, volatile phenolic compounds in the smoke absorb into the grapes and result in smoke-tainted wine with ash-like flavors. Wineries want to know more about how smoke exposure will alter their vineyards and wine flavors and how they can mitigate these effects. Understanding how the fungal communities in vineyards change in response to agricultural sprays and forest fire smoke will help wineries to continue producing unique, regionally-specific wines. The objective of this study was to determine the effect of heavy forest fire smoke and agricultural sprays on fungal communities of grapes growing in three different vineyards. Experimental vines in three different vineyards were exposed to either smoke, potentially mitigating agricultural sprays, or a combination of both treatments. Next generation sequencing was used to analyze the composition of the fungal communities on grape bunches before exposure to the treatments, 14 days after exposure, and again immediately before harvest. We found no significant differences between the control and treatment vines at any of the three vineyards; however, we did see differences in the control vines among vineyards.

Funding Support: Natural Sciences and Engineering Research Council of Canada (NSERC)

Developing a Spray-Induced Gene Silencing (SIGS) Method to Control Grape Powdery Mildew (*Erysiphe necator*)

Britt Eubanks,* Bahiya Zahl, Chance Lemon, Madesyn Samples, Satyanarayana Gouthu, and Laurent Deluc *Oregon State University, 2750 SW Campus Way, Corvallis, OR 97331 (eubankbr@oregonstate.edu)

The emergence of fungicide resistance for grape powdery mildew (GPM; caused by Erysiphe necator) suggests the need to develop new strategies for GPM control. Recent research demonstrates that agricultural pests such as insects, nematodes, and fungi can be controlled through exogenous application of RNA molecules to trigger RNA interference (RNAi). This project's primary goal is to develop a spray-induced gene silencing (SIGS) program to induce RNA interference mechanisms targeting both the grapevine and the fungus. The plant genes targeted for RNAi belong to a susceptibility-gene family to Erysiphe necator (Mildew Locus O: MLO genes). We also targeted three pathogen-regulated genes critical to the life cycle of the pathogen. Our first objective was to identify stretches of RNA molecules with maximum interference activity to repress MLO endogenous genes (VitviMLO 3,4,6,9,13,17) and fungal genes (Dicer-like protein 1 and 2 and CYP51). As a first step, we successfully cloned and validated by sequencing all candidate genes' coding regions used to generate dsRNA molecules. Using these clones as a template, we have produced the dsRNA molecules using a commercial kit. We sprayed the MLO-based dsRNA solutions (20 ng/uL) of the MLO candidates combined with a wetting agent (SILWET-L77) on three-day acclimated tissue-cultured disk leaves of grapevine tissue-culture plants. RNA extraction is underway and will be followed by reverse transcription reactions and teal-time PCR experiments. Our second objective is to evaluate uptake and processing of dsRNA molecules from grapevine leaves using a fluorescent tag to assess how systemic the spray can be, and our third objective is to determine the efficacy of clay nanoparticles (layered double hydroxide [LDH]) to prolong dsRNA's lifespan on the surface of grapevine leaves.

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Funding Support: USDA

Propagation Method Quality and Indole Production Evaluation Depend on Yeast Selection in Sparkling Winemaking

Anne Flesch and Etienne Dorignac*

*Fermentis, 137 rue Gabriel Peri, 59170 Marcq-en-baroeul, France (e.dorignac@fermentis.lesaffre.com)

Due to the biologically unfavorable nature of a base wine intended to undergo a Charmat method, the pitching yeast should have optimal viability and be acclimatized to wine conditions prior to inoculation. Three different strains were propagated using two methods. The "direct rehydration" method consisted of simple rehydration of the yeast in a diluted base wine. The "gradual acclimatization" method was multi-stage to acclimatize the yeast to both wine conditions and cooler temperatures through multiple sub-culturing steps. Pitching was determined by cell density. Each strain responded differently to the "direct" preparation method and had optimal viability at different times throughout the procedure. Two strains grew fast and showed their greatest viability early in propagation and before maximum cell counts. They failed to start the subsequent secondary fermentation, suggesting that cell counts alone are not optimal to determine the pitching time. Using the "acclimatization" method, these strains behaved similarly during propagation, but differently at pitching, one being much more affected by wine conditions, but both succeeding to achieve the secondary fermentation. Greater viability of the starting yeast culture not only reduces the risk of failed secondary fermentation, but also decreases the chance that undesirable "off-flavors" are produced, such as indole, which is known to impart plastic aromas. Indole is produced by yeast through metabolism of tryptophan and its production could be linked to sub-optimal yeast vitality that can occur during sparkling wine production. Using the "acclimatization" method, the three strains were used to carry out secondary fermentation in a tryptophan-supplemented base wine. Only one strain, interestingly not showing the lowest viability, produced indole. This suggests that while culture vitality may contribute to indole production, there may also be predispositions in some yeast strains that increase their indole production potential.

Funding Support: Lesaffre-Fermentis

Selection and Hybridization of Yeast Strains to Modulate Key Compounds in Wines

Anne Flesch and Etienne Dorignac* *Fermentis, 137 rue Gabriel Peri, 59170 Marcq-en-baroeul, France (e.dorignac@fermentis.lesaffre.com)

Consumer preferences are trending toward fresher and more expressive wines, combined with greater attention to the environment and to their health. This work offers "blending" tools for winemakers to help them produce intensely fruity and clean wines. Yeast hybridization was followed by a multi-step screening of the strains. It first focused on fermentation ability and sulfite production, then on acetaldehyde and acetate esters, and finally on tasting properties. A specific hybrid produced an average 40% less acetaldehyde and sulfites and two to four times more isoamyl acetate than reference strains. This latter compound imparted distinctive candy flavors and was a strong flavor enhancer, making this strain a powerful tool for blends by enhancing fruitiness and decreasing sulfite concentrations of base wines. Enhancing flavors and improving fermentation by using different species of yeast strains may also



represent a solution. In a second work, we focused on the impact of a species not known in wine: *Saccharomyces pastorianus*. Fermentations were conducted on a Sauvignon blanc must using single strains (two different *S. pastorianus*, one *S. bayanus*, and two different *S. cerevisiae*) and mixtures of them (*S. pastorianus* 1 or 2 and *S. bayanus*) and two different inoculation temperatures: cold (13°C) and classic (17°C). Both *S. pastorianus* strains achieved fermentation, produced almost no acetic acid, and overproduced 2-phenylethanol and its acetate. *S. pastorianus* 2 was of particular interest thanks to its nice tropical fruit notes. Considering its enological properties, *S. pastorianus* species may be of interest for winemaking, alone or in co-inoculation with *S. bayanus* and, in particular, to help decrease wine volatile acidity.

Funding Support: Lesaffre-Fermentis

Selection of Yeasts and Fermentation Conditions for Production of Sauvignon blanc

Anne Flesch, Etienne Dorignac,* and Marie-Charlotte Colosio (IFV Nantes) *Fermentis, 137 rue Gabriel Peri, 59170 Marcq-en-baroeul, France (e.dorignac@fermentis.lesaffre.com)

Sauvignon blanc is characterized by particularly strong but also very sensitive varietal flavors such as polyfunctional thiols, which have very low perception thresholds. The choice of yeast strain and fermentation parameters is crucial to achieve winemakers' goals. This work presents a full study, from the screening of a yeast bank collection for genetic (IRC7 gene expression) and phenotypic (H₂S, SO₂, and volatile acidity production) features, to the characterization of the best strains through several laband microscale vinifications (aromatic and sensory attributes), and then extensive field trials. There was a strong correlation between the zygosity of the short or fulllength allele of IRC7 ($IRC7^{\circ}$ or $IRC7^{\perp}$) and the capability of the strains to release thiols, especially 4-mercapto-4-methylpentan-2-one (4-MMP), which imparts box tree and passion fruit aromas. Very different potentials to produce undesirable compounds in addition to acetate and ethyl esters were also observed and led to the selection of a *IRC7*^L/*IRC7*^L strain (A) that primarily promoted all aromatic thiols and complex ethyl esters. The study of this strain in comparison with a *IRC7^s/IRC7^s* strain (B) with a high acetate ester production capability through different fermentation temperature and nutrition regimes was performed. It clearly highlighted a greater release of thiols and lower concentration of acetate esters at high temperature (18°C versus 12°C) and confirmed the importance of the nitrogen catabolite repression regulation system in yeast. The supply of ammonium at yeast inoculation indeed drastically lowered and even blocked release of 4-MMP by strain A when the initial temperature was 18°C, then lowered to 12°C after three days. This latter temperature regime was rated among the best for minimizing defects and maximizing the concentrations of most aromatic compounds and was judged very positively during a preliminary tasting.

Funding Support: Lesaffre-Fermentis Institut Français de la Vigne et du Vin, Nantes - France

Red Wine Fermentation With A Completely Disintegrated Cap: The New Patented Technique with Modulated Air Jets

Giuseppe Floridia* and Fabio Mencarelli

*Parsec Srl, Via J. Nardi 21, Firenze 50132, Italy (sales@parsecsrl.net)

A new method for selective extraction from grapes that doesn't require pumps, délestage, pump-overs, or other bulk inside the tank was developed and tested between

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2015 and the present in Chile, Argentina, Italy, France, Spain, and California. The simplification and optimization of racking and cleaning operations is combined with significant energy savings overall. Because the cap is kept disintegrated, this new technique revolutionizes the traditional management of red wine making through optimized handling of thermal-densimetric stratification, temperature, oxygenation, nutrition, and fermentative kinetics. The advantages are both practical and qualitative, even in tanks with large-scale or complex geometries.

Funding Support: Parsec Srl

Smoke Attribute Carryover and Functionality of Rinse Systems in Sensory Analysis of Wildfire-Affected Wines

Jenna Fryer, Thomas S. Collins, and Elizabeth Tomasino* *Oregon State University, 100 Wiegand Hall, Corvallis, OR 97333 (elizabeth.tomasino@oregonstate.edu)

With developments in climate change, wildfires may increase 15 to 70% by 2050, posing new challenges to the wine industry. Aside from many fires occurring in areas where winegrapes are grown, wildfire smoke can travel vast distances, carrying volatile organic compounds to vineyards that are then adsorbed onto the grapes. Wines produced from smoke-affected grapes contain elevated amounts of smoke-related phenols, which can lead to undesirable aromas and flavors. Glycoconjugated forms of these phenols have also been found to be present after smoke events, which leads to a lingering, ashy aftertaste in wine due to enzymatic breakdown within the oral cavity. When evaluating the sensory profile of many smoke-impacted wines, an additive effect of smoke and ashy flavor carrying over to subsequent wines occurs. We evaluated the extent of the carryover and mitigation of its influence via different rinses and time separation. For evaluation, three red wines with varying amounts of smoke exposure (no smoke, medium smoke, and high smoke) were used across three studies. To determine the best attributes to focus on in future trials, check-all-thatapply (CATA) was performed followed by temporal-CATA (TCATA) to determine the length of time that these attributes were perceived. Finally, the progression of intensities of these smoke-related attributes over a fixed time was determined using a fixed time-point temporal method utilizing different rinse systems – water, pectin, and unflavored antibacterial mouthwash. We hypothesize that the use of an antibacterial mouthwash will decrease the observed amount of smoke attribute carryover due to reduction in enzymatic breakdown in the mouth. Overall, this study will clarify best practices for the most effective analysis of properties associated with smoke exposure, providing a better understanding of these sensory effects and the temporality of these attributes.

Funding Support: USDA-ARS

Evaluating Foliar-Applied Calcium Carbonate on Table Grapes and Observing Cold Storage Potential

Leah Groves, Sonet Van Zyl,* Johan Coetzee, and Moriah Mehlman *CSU Fresno, 2360 E Barstow Ave M/S VR89, Fresno, CA 93740 (svanzyl@csufresno.edu)

California is a leader in producing table grapes and the Southern San Joaquin Valley produces ~85% of the state's table grapes. Postharvest storage is essential and is a common struggle in the table grape industry, where grapes can end up as waste. There have been studies to extend postharvest storage ability by applying preharvest applications like calcium chloride, calcium nitrate, and sodium bicarbonate. Yet no



study indicates calcium carbonate usage, especially in agriculture. Calcium carbonate can be useful in plant physiology, as calcium is important for structural components in the berry cell wall and carbonate can be essential in photosynthesis. However, calcium carbonate is insoluble and due to its large particle size, is not easily absorbed by plants. We evaluated OR-224B, a calcium carbonate-based product with a small particle size. This study is located in the Fresno State Vineyard where Sweet Scarlet, a red mid-season seedless table grape, was used to evaluate calcium carbonate uptake and its effect on grapes after commercial cold storage trials. The cultural and harvest practices followed a conventional operation, with grapes packed into 19-lb. boxes. The experimental layout consisted of eight treatments, including a control, with six replicates. OR-224B was used in 2020 and compared with the individual components and standard industry products containing calcium. The treatments included foliar and soil applications during the growing season. After harvest, grapes were stored for four or eight weeks at a commercial cold storage facility. Measurements of berry texture, size, color, and fungal infection rates were used to evaluate OR-244B as a cheaper and safer calcium-based product to improve postharvest storage quality. This study is ongoing and will continue throughout the 2021 growing season.

Funding Support: Oro Agri A Rovensa Company, Agriculture Research Institute

Understanding the Onset of Systemic Infection of Red Blotch Virus: Toward Practical Solutions in Grapevines

Bailey Hallwachs, Bhaskar Bondada,* Marc Fuchs, Sadanand Dhekney, Benham Khatabi, Alexander Levin, and Patricia Skinkis *Washington State University Tri-Cities, 2710 Crimson way, Richland, WA 99354 (bbondada@wsu.edu)

Red blotch disease, caused by Grapevine red blotch-associated virus (GRBaV), is a recent addition to the extensive repertoire of viruses that infect grapevines. Infection results in red blotches on leaves with pinkish red-colored veins without rolling off the margins observed at the onset of ripening in red cultivars. The objective of this study was to gain an understanding of the onset of systemic infection of the red blotch virus to aid designing antiviral interventions by examining fruit quality attributes and various tissues of healthy and afflicted vines with microscopy. The fruit quality of afflicted vines was compromised, as indicated by reduced Brix, high pH, and reduced vigor. This indicated that the virus blocks the sugar pathway (phloem tubes), impeding translocation of sugars into the berries during ripening. While leaf structural architecture was not affected, the infected chloroplasts developed plastoglobuli, tannins in addition to a massive accumulation of starch. Some chloroplasts were completely dismantled. Furthermore, the infected vines developed clusters of hens and chickens and altered seed morphology. On the other hand, the healthy seeds were pyriform with a distinct beak. The study showed that red blotch virus infection lowers fruit quality by altering the structural integrity of the vine system.

Funding Support: Oregon Wine Board

Influence of Polysaccharides on Extraction of Polyphenols during Maceration at Different Grape Maturities

Jan-Peter Hensen,* Fiona Hoening, and Fabian Weber

*Rheinische Friedrich-Wilhelms-Universität Bonn, Friedrich-Hirzebruch-Allee 7, Bonn/ North Rhine-Westphalia/53115, Germany (hensen@uni-bonn.de)

Red wine sensory properties and quality largely depend on extraction of anthocya-

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nins and tannins during winemaking and their interactions with other wine components. The climatic conditions in cool climate regions make it much more challenging to produce red wines with deep color and a full body, presumably because of the poor extractability of phenolic compounds. The high demand for those types of red wines necessitates the full use of the grape's phenolic potential. There is, however, still a lack of knowledge regarding the mechanisms that change the extractability and the impact of polysaccharides. To further understand the correlation between grape maturity and polyphenol extractability, the influence of grape polysaccharides was analyzed in microvinifications of Cabernet Sauvignon and Pinot noir in 2020. Grape polysaccharides extracted from differently ripe grapes were added to musts from three harvest dates (Cabernet Sauvignon: 17.1, 20.9, 21.8 Brix; Pinot noir: 16.7, 19.7, 21.6 Brix). The added polysaccharides were previously extracted as the alcohol-insoluble residues at three different harvest points in 2019 from the respective berries from the same vineyard. The addition of these polysaccharides during maceration influenced tannin concentration in the resulting wine, while anthocyanin concentration was determined by actual berry ripeness. The observed effects were greater when polysaccharides from riper berries were added. Possible interactions with the polysaccharides stabilized or bound tannins in the must, which led to contrasting results in the different wines. While Pinot noir wines had more measurable tannin concentrations, addition of polysaccharides led to a decreased tannin concentration in Cabernet Sauvignon wines. The stabilizing effects could be based on protective reactions, with other wine components hindering those to bind tannins. The smallest impact of added polysaccharides was found in must from unripe grapes. Varietal and ripeness-specific effects highlight the diverse interactions of polysaccharides with polyphenols during vinification.

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Grapevine Red Blotch Virus Impact on Oregon Pinot noir Wine Sensory Properties

Samuel Hoffman, Elizabeth Tomasino,* James Osborne, Robert Martin, and Patty Skinkis

*Oregon State University, 100 Wiegand Hall, 3051 SW Campus Way, Corvallis, OR 97331 (Elizabeth.Tomasino@oregonstate.edu)

Grapevine red blotch virus (GRBV) poses a potential threat to the United States wine industry. Grapes harvested from GRBV-infected vines often have reduced sugar and anthocyanins, but impacts to wine sensory properties are unknown. This study investigated the sensory impact of viticultural management practices on GRBV in Oregon Willamette Valley Pinot noir. The viticultural practice consisted of application of abscisic acid (ABA) to the fruit zone until dripping. ABA was applied to GRBV (+) vines during the 2018 and 2019 growing season in a randomized complete block design during veraison and compared to an unsprayed control. Grapevine virus status was identified via PCR of leaf tissues from individual vines collected in summer 2018. This resulted in two treatments: GRBV (+) control and GRBV (+) ABA. Wines made from the treatments underwent sensory analysis by red wine consumers. Sensory analysis consisted of preference/liking, aroma check-all-that-apply (CATA), color intensity, and mouthfeel just-about-right (JAR) questions for body, bitterness, astringency, sourness, and heat. No significant differences in preference/liking, bitterness, astringency, heat, or sourness were noted during either year. Wine body was significantly different in 2018, with the GRBV (+) control wines characterized as having less body than GRBV (+) ABA wines. This difference was not noticed in the 2019 wines. There



were significant color differences between both 2018 and 2019 wines. In 2018, the GRBV (+) control sample was rated more purple in color, while in 2019, the opposite occurred. Correspondence analysis was used to visualize the aroma CATA results for both years. CATA results showed aromatic trends in both years for the respective groups; however, none were considered statistically significant. While ABA may impact grape maturation, it does not appear to alter the wine sensory characteristics of wines produced with grapes from GRBV (+) vines.

Funding Support: Oregon Wine Research Institute and Private Industry Donations

Mouthfeel Effects of Oligosaccharides within a Model Wine System

Samuel Hoffman, Elizabeth Tomasino,* and Kim Phan *Oregon State University, 3051 SW Campus Way, Corvallis, OR 97330 (Elizabeth.Tomasino@oregonstate.edu)

Mouthfeel is a critical aspect of the wine sensory experience, yet it is one of the least understood. While much is known about compounds such as phenolics, little work has addressed other wine compositional components such as oligosaccharides. Compared to other food and beverages, saccharides are in very low concentrations within wine and their mouthfeel impacts are currently unknown. This study used two oligosaccharides at two concentrations to investigate and quantify potential impacts to mouthfeel within a model wine system. The two oligosaccharides, fructo-oligosaccharide (FOS) and glacto-oligosaccharide (GOS) were added at a rate of 450 or 900 ppm to a basic wine matrix. These concentrations are based off amounts previously measured in Pinot noir wines. Compounds were added to a simple model wine consisting of 12% ethanol with 4 g/L TA. The four treatments underwent triangle tests against an untreated control, then an additional triangle test was conducted between the low and high concentration of each oligosaccharide. After completion of the triangle tests, all treatments and the control underwent descriptive analysis (DA) using 100 mm line intensity scales. Before conducting DA, panelists underwent training on sweetness, bitterness, viscosity, astringency, and acidity standards. The triangle test showed a significant difference between the FOS 450 ppm sample and the FOS 900 ppm sample. Interestingly, both FOS samples, 450 and 900 ppm, were not found to be different from the control. Additionally, none of the GOS samples were significantly different. DA found no significant difference for any of the five attributes. This indicates that the difference between FOS samples may be above the detection threshold, but not the perception threshold. Future work will look at different levels of oligosaccharides, as well as implement more training.

Funding Support: No specific funding source used for this study.

Impact of Pinot noir Crop Load Metrics on Wine Quality

Ling Huang, Jingwen Li, Yanping Qian, Patty Skinkis, and Michael Qian* *Oregon State University, 3051 NW Campus Way, Corvallis, OR 97331 (michael.qian@oregonstate.edu)

Cluster thinning is a traditional practice used in premium wine production to reduce yield, as it is thought to improve fruit quality. The impact of cluster thinning levels on Pinot noir wine quality was studied in this work. Total monomeric anthocyanin, total phenolic content, polyphenols, and volatile profile of Pinot noir wines from Oregon commercial wineries were analyzed over several vintages. Crop level was adjusted using cluster thinning at the lag-phase of berry development using a cluster number per shoot regime or ton per hectare treatments, and many vineyards included a full-crop

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control (no thinning). Total monomeric anthocyanin was determined with a spectrophotometer based on the absorbance change of anthocyanin at different pH. Total phenolic content was analyzed using the Folin-Ciocalteu colorimetric method. Wine polyphenols were analyzed with high-performance liquid chromatography. Volatile aroma profile of wine was analyzed with solid-phase microextraction gas chromatography-mass spectrometry (SPME GC-MS) headspace-GC-Flame ionization detector and SPME-GC-PFPD according to properties of aroma compounds. Isotope internal standards were used to build standard calibration curves. In general, cluster thinning affected the monomeric anthocyanin, total phenolic content, and anthocyanin levels in wines compared to the no-thinning treatment. Volatile compound concentrations also were influenced by cluster thinning, but the effect was inconsistent among vineyards and vintages.

Funding Support: USDA-ARS

Impact of Abscisic Acid Treatment on Composition of Wine Made from Grapevine Red Blotch Disease-Infected Grapes

Ling Huang, Patty Skinkis, James Osborne, Qin Zhou, Yanping Qian, and Michael Qian*

*Oregon State University, 3051 NW Campus Way, Corvallis, OR 97331 (michael.qian@oregonstate.edu)

Grapevine red blotch disease has a negative effect on the grape berry developing process and decreases sugar accumulation. Endogenous abscisic acid (ABA) is an important plant growth regulator that triggers berry ripening. We examined the impact of exogenous ABA on red blotch-affected vines with regard to wine composition. ABA was sprayed on red blotch-affected grapevines around veraison, while control groups were left unsprayed. Total monomeric anthocyanin was determined with a spectrophotometer by the absorbance change of anthocyanin at different pH. Total phenolic content was analyzed using the Folin-Ciocalteu colorimetric method. Volatile profiles of wines made from grapes of the ABA trial were analyzed using headspace (HS)-GC-FID, stable isotope dilution approach with solid-phase microextraction gas chromatography-mass spectrometry (SPME-GC-MS) and SBSE-GC-MS techniques. Compared to control, ABA treatment led to lower levels of total monomeric anthocyanin and total phenolic content in wines. ABA treatment had no significant effect on important volatile aroma compounds of wines.

Funding Support: Oregon State University

Impact of Early Leaf Removal on Aroma Profiles of Wine Made from Grapevine Red Blotch Disease-Infected Grapes

Ling Huang, Patty Skinkis, James Osborne, Qin Zhou, Yanping Qian, and Michael Qian*

*Oregon State University, 3051 NW Campus Way, Corvallis, OR 97331 (michael.qian@oregonstate.edu)

Grapevine red blotch disease has a large impact on grape development and lightens grape berry color. The typical symptoms are red spots and red veins on leaves. We examined the impact of two different leaf-removal treatments on red blotch-affected vines on wine aroma composition. The leaf-removal trial consisted of an early leaf-removal applied prebloom, in which all leaves in the cluster zone were removed by hand, or mechanical leaf removal at fruit set (control). Early leaf removal increased the levels of monomeric anthocyanin and phenolic compounds in wines.



This suggests early leaf removal may lessen the effect of red blotch disease on grape color. No obvious significant differences in wine volatile profile were found between treatments. Early leaf removal led to higher levels of bound-form C_{13} -norisoprenoids in wines than the control.

Funding Support: Oregon State University

Impact of Grapevine Red Blotch Disease and Irrigation Treatments on Grape/Wine Quality

Ling Huang, Alexander Levin, James Osborne, Yanping Qian, and Michael Qian* *Oregon State University, 3051 NW Campus Way, Corvallis, OR 97331 (michael.gian@oregonstate.edu)

Grapevine red blotch disease is associated with a single-stranded DNA virus (grapevine red blotch disease-associated virus, or GRBV). The disease delays grape berry maturity and sugar accumulation and has a high impact on fruit quality. To better understand the impact of red blotch disease on grape and wine quality, two irrigation treatments (wet and dry) were randomized in two blocks of fields and characterized by varying water application rates on both red blotch-infected and non-infected grapevines. Berry maturity parameters, wine anthocyanins, phenolics, and flavor profiles were investigated. Volatile compounds were quantified by headspace (HS)-GC-FID, stable isotope dilution approach with solid-phase microextraction gas chromatography-mass spectrometry (SPME-GC-MS), and stir-bar sorptive extraction GC-MS (SBSE-GC-MS) techniques. Red blotch disease decreased the total soluble solids of grape berries and the total phenolic content of wines. The impacts of red blotch on volatile compounds were not obvious. Wines from wet treatments had higher concentrations of isoamyl acetate than dry treatments.

Funding Support: AVF, CDFA

Study on Expression of Acid Protease using *Saccharomyces cerevisiae* Surface Display Technology

Rong Huang and Yuyang Song*

*College of Enology, Northwest A&F University, College of Enology, Northwest A&F University, 712100, China (yuyangsong@nwsuaf.edu.cn)

Protein turbidity is a significant defect showing non-biological instability of wine. Acid protease is often used during food fermentation because its hydrolysis process helps avoid bacterial growth and corruption. Expression of acid protease pepA was initiated using the promoter and anchor protein in the surface display system of *Saccharomyces cerevisiae*. The acid protease gene (pepA) from *Aspergillus usamii* was cloned to construct an integrated plasmid and an integrated vector to obtain the recombinant plasmid PUC-GAP- α -factor-pepA-SED1. After the recombinant plasmid is cut with Sma I, it is transformed into the haploid and diploid of *S. cerevisiae* using an electric shock transformation method. Through homologous recombination, the acid protease gene (pepA) of *A. usamii* was integrated into the gene locus of *S. cerevisiae*. Two strains of *S. cerevisiae* that secrete acid protease through a cell surface display were obtained. Their protease activities were 285.71 U/mL and 495.24 U/mL. The research outlines a new method to solve the problem of protein turbidity in wine and lays a theoretical foundation for industrial application of pepA acid protease wholecell catalyst.

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High-Resolution Vine Irrigation: A Low-Cost Proximal Sensing Method for Predicting Single-Vine ET

Matthew Jenkins, Autumn Mannsfeld, Konrad Miller, Jean-Jacques Lambert, Mason Earles, and David Block* *University of California - Davis, 1 Shields Ave, Davis, CA 95616 (deblock@ucdavis.edu)

As California becomes increasingly affected by drought and water use regulations become more restrictive, the demand for increased water use efficiency grows in importance. Currently, the viticulture industry relies on bulk irrigation, which assumes all vines in a given area have the same water demands. This assumption ignores individual plant health, the heterogeneity of soil, canopies, topography, and many other factors that affect plant water use, leading to over and under-watering. To increase water use efficiency, irrigation practices must take this spatial variation into account. Existing methods for estimating single-vine ET, such as sap flow or aerial imaging, are too expensive and/or technically advanced to use in most commercial settings. The purpose of this project was to provide proof of concept that simple ET sensors, along with appropriate, physically-based models, will allow precise control of irrigation down to single-vine resolution. Four mature Zinfandel vines were potted and placed on load cells in a vineyard setting, as simple weighing lysimeters to accurately measure ET. We then developed three mathematical models, two based on first principles and one empirical model, to predict the mass flow rate of water leaving a single vine based on common biometeorological parameters measured on each of the four vines. To evaluate the utility of each model, we compared the predicted mass flow rate of water out of a grapevine to load cell-derived ground truth ET data. We report a strong correlation between all models' predicted water use rate and real water use rate ($r^2 = 0.78$ to 0.88). Predicted water use rates can be used to accurately calculate single vine ET, thereby informing irrigation decisions at the previously unthinkable scale of single vines.

Funding Support: Private gifts; Ernest Gallo Endowed Chair in Viticulture & Enology

Following the Map To Climate Resilience in the Next Generation of Cold **Climate-Adapted Winegrapes.**

Venkateswara Rao Kadium,* Andrej Svyantek, John Stenger, Sarah Bogenrief, Collin Auwarter, and Harlene Hatterman-Valenti *North Dakota State University, 1053 17th Ave N, University Village, UV Unit #152, Fargo, ND 58102 (venkateswara.kadium@ndsu.edu)

Grape production in North Dakota's short growing season is severely limited by winter injury, frost risk, and low fruit quality. Along with climate-driven abiotic challenges, the cultivar options are restricted due to disease risks and demands for early ripening phenology. Due to these needs, the North Dakota State University Grape Germplasm Enhancement Project (NDSU-GGEP) relies heavily on wild Vitis spp. and interspecific hybrids as parental material. To investigate grapevine phenology, ripening dynamics, cold hardiness, and environmental adaptation in the context of North Dakota's challenging climate, an incomplete diallel population of 1064 unique individuals was



planted in 2017 from three core, interspecific NDSU-GGEP breeding lines with diverse grapevine genomic backgrounds, including *V. amurensis, V. labrusca, V. riparia*, and *V. vinifera*. After trunk establishment, detailed phenotypic data for the planting was collected in 2020, with 255 individuals evaluated for fruit composition (soluble solids content, pH, titratable acidity, malic acid content, and yeast assimilable nitrogen). To further investigate climate adaptation, phenology (budbreak, bloom, and fruit ripening time), dormancy acclimation (wood and bud maturation), and cold hardiness (winter survival and differential thermal analysis of dormant buds) traits were monitored for all fruiting individuals. Genetic maps of the population were constructed using GBS and rhAmpSeq markers to perform informative analysis of marker-trait associations. By combining phenotypic and genotypic data, we identified some QTLs covering the genomic regions driving climate-adaptive traits and fruit quality (Brix, pH, and TA) within interspecific cold-hardy grapevine breeding efforts.

Funding Support: Specialty Crop Block Grant Program

Impact of Activated Carbon and Polyvinylpolypyrrolidone Treatments for Amelioration of Smoke-Tainted Cabernet Sauvignon

Henry Kampen* and Benjamin Benitez

*California State University Fresno, 9 Edith Court, Napa, CA 94558 (hnkampen@gmail.com)

The wine industry faces a growing problem of wildfire smoke, causing grapes to possibly be unsuitable for wine production. The volatile phenol compounds (VPs) present in wildfire smoke impart unpleasant sensory notes to the wine, including multiple ashy aromatics and bitter and smoky notes to the palate. This issue impacts wine-producing regions on an international scale, so methods to effectively remove VPs with minimal impact on wine quality are necessary. The goal of this project was to investigate the effect of activated carbon and polyvinylpolypyrrolidone (PVPP) treatments on primary smoke phenols (4-ethylguaiacol, 4-ethyl phenol, guaiacol, and 4-methylguaiacol), pigment density, and wine structure in a smoke-tainted Cabernet Sauvignon wine. Smoke-impacted Cabernet Sauvignon berries were harvested from Napa Valley and processed using conventional wine production methods. The wine was then treated with two amelioration regimens: activated carbon or activated carbon in conjunction with PVPP. Both treatments were used at manufacturer's recommended dosage (50 g/hL for carbon and 2lbs/1000 gal for PVPP). The treatments were followed by sterile filtration. While the overall effects of activated carbon have been shown to strip smoke phenols from wine, it is beneficial to establish parameters for the consequent removal of other, desirable aroma compounds. Spectrophotometry, gas-chromatography mass-spectrometry, and statistical sensory evaluations (triangle test) were used to determine which treatment was more effective at smoke VPs removal. By establishing rates of loss of desirable wine characteristics along with smoke phenols removal, risk/benefit decisions can be made when using activated carbon or PVPP for smoke amelioration.

Funding Support: California State University Fresno

Impact of Yeast-Derivative Treatment on Smoke-Tainted Cabernet Sauvignon Wine

Henry Kampen* and Benjamin Benitez *California State University Fresno, 9 Edith Court, Napa, CA 94558 (hnkampen@gmail.com)

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With increasing risk of wildfires in grapegrowing regions, the wine industry needs methods to remove or mask the unpleasant phenolic compounds present in smoke-tainted juice. The ability of yeast derivatives to mask or balance these undesirable wildfire volatile phenols (VPs; 4-ethylguaiacol, 4-ethyl phenol, guaiacol, and 4-methylguaiacol), especially on the retronasal mid-palate, was explored. These VPs contribute many negative sensory notes to wine, both aromatically and on the palate. We investigated the efficacy of two postfermentation yeast-derivative treatments in varying concentrations. Smoke-impacted Cabernet Sauvignon grapes were harvested from the Napa Valley AVA and processed using a standard winemaking regimen. Two experimental yeast-derivative treatments were compared at varying concentrations (50, 100, 250 and 500g/hL) to establish sensory masking effects. Regular bâtonnage was employed to ensure full contact with the yeast products. VPs were monitored by gas-chromatography mass spectrophotometry. Sensory analysis with a ranking test was used to determine if the levels could be differentiated from one another. The ability of yeast-derivative treatments in conjunction with amelioration methods to minimize the impact of wildfire smoke on future vintages will be established. The potential to mask or reduce the sensory impact of wildfire smoke could be beneficial to the wine production industry.

Funding Support: Laffort Labs

Vineyard Leaf Tissue Survey of Auxin Herbicides within the Texas High Plains AVA

Suraj Kar, Thayne Montague,* Kyle Lauterbach, Trey Ruland, Pierre Helwi, and Edward Hellman

*Texas Tech University and Texas A&M University AgriLife Research and Extension Center, Department of Plant and Soil Science, Texas Tech University, Bayer Plant Science Building, Room 103, 2911 15th Street Mail Stop 2122, Lubbock, TX 79409 (thayne.montague@ttu.edu)

With more than 500 wineries, the state of Texas is the fifth-largest wine-producing state within the United States. The Texas High Plains American Viticulture Area (AVA) produces nearly 80% of all winegrapes (Vitis vinifera L.) in Texas. However, this same area is a major cotton-producing region where new formulations of auxin-based herbicides are being used by the agriculture industry with increasing regularity. It is thought synthetic auxin herbicides (predominantly 2,4-D and dicamba) are applied to nearly 90% of cotton grown on the Texas High Plains. Grapevines are among the most sensitive species to synthetic auxin herbicides, and concentrations lower than 100 times the recommended rate can cause vine injury. Close proximity to cotton fields and climate factors (hot, dry, and windy conditions) found within the AVA have made vineyards particularly vulnerable to herbicide volatilization and drift injury. Multiple injury events, causing as much as 85% reduction in grape yield, have been reported within the Texas High Plains AVA. To evaluate the extent and risk frequency of off-target movement of auxin-based herbicide, a field survey was conducted during the 2020 grapegrowing season within six Texas High Plains AVA vineyards through monthly leaf tissue sampling. Results indicated a single volatilization and drift event occurring early in the growing season, with concentrations ranging between 3.75 to 13.70 ng/g, and 80.41 to 266.55 ng/g for 2,4-D and dicamba, respectively. Although the effect on yield reduction in these vineyards were not studied, a visual appraisal indicated damage to vegetative tissues of vines, including decreased shoot growth, leaf area, internode length, and pruning weight. This survey is a first of its kind study and gives growers crucial field-level information on the extent and frequency of off-target movement of synthetic auxin herbicide within Texas High Plains AVA vineyards.



Funding Support: State of Texas Viticulture and Enology Research Funds, Department of Plant and Soil Science

Comparative Diagnostic Methods for Grapevine Red Blotch Virus (GRBV)

Achala KC,* Joseph DeShields, and Alexander Levin *Oregon State University - Southern Oregon Research and Extension Center, 569 Hanley Rd, Medford, OR 97502 (achala.kc@oregonstate.edu)

Grapevine red blotch disease (GRBD) diagnosis is often challenging due to the nature of symptoms in grapevines and complexity of testing methods. Recently, a loop-mediated isothermal amplification (LAMP) method was developed for grapevine red blotch virus (GRBV) testing, which is purportedly more sensitive, quick, and affordable than PCR-based diagnosis. However, with limited field testing, it is difficult to fully rely on the test results to determine the full impact on vineyard. In 2020, an experiment was conducted to compare the accuracy of three GRBV detection methods in samples collected from a commercial vineyard in southern Oregon. Vines were naturally infected and their GRBV status was predetermined from year-end sampling in 2019. Leaf samples were collected from basal, middle, and apical shoot nodes at berry set, veraison, and harvest. Each sample consisted of two whole leaves (blades with attached petioles) collected from each bilateral cordon. Samples were rated for GRBD symptoms at the time of collection and subsequently tested for GRBV infection using LAMP, PCR, and qPCR assays. All positive vines were consistently positive in 2020; however, their detectability by the assays differed significantly among node positions depending on phenology. At berry set, qPCR was the only assay that detected positive vines with 100% accuracy from basal samples. At veraison, both PCR and qPCR, and at harvest, all methods including LAMP detected positive vines with 100% accuracy from basal samples. When the ability of these methods to detect negative vines was analyzed, both PCR and qPCR test results were 100% negative; however, LAMP test results were positive for 24 and 19% of the samples at veraison and harvest, respectively. Given its specificity, affordability, and simplicity, the LAMP method can be a promising tool in monitoring GRBV. However, its sensitivity to negative vines needs further validation.

Funding Support: American Vineyard Foundation and USDA-NIFA Specialty Crop Research Initiative (SCRI)

Efficacy of XyIPhi-PD for Reduction of Pierce's Disease in Vineyards: 2020 Field Trial Results

Anika Kinkhabwala,* Michael Rupert, and Amy Ritchardson *A&P Inphatec, LLC, 1060 East Meadow Circle, Palo Alto, CA 94303 (accounting@inphatec.com)

Few methods for controlling and treating Pierce's Disease (PD) have been available, with efforts historically focused on controlling the sharpshooter vector (e.g., insecticides) or roguing seriously ill vines, both of which have demonstrated only limited success. However, an option that reduces PD in grapevines is now available, XyIPhi-PD. XyIPhi-PD is a novel biological treatment for PD that is approved for use in organic production. This breakthrough technology was developed exclusively for viticulture. XyIPhi-PD contains a cocktail of viral bacteriophages (bacteria-killing viruses) that enter and destroy *XyIella fastidiosa* bacteria. Multiple university field studies assessed the efficacy of XyIPhi-PD to treat or prevent PD in vineyards when used in accordance with label Directions for Use. In a 2015 Texas A&M study with natural infection exposure, three monthly XyIPhi-PD treatments significantly re-

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duced PD incidence to 44% versus controls. For the 2020 multisite field trials (North Coast California commercial vineyards with high PD pressure), vines were randomly assigned to "2020 Treated," "2019 and 2020 Treated," and two separate, untreated control groups to test the efficacy of XyIPhi-PD. XyIPhi-PD treatment of diverse wine varietals prompted reductions in visual PD symptoms of 60% versus controls, which was confirmed by qPCR. XyIPhi-PD can prevent disease in healthy vines and treat vines early in their infection.

Funding Support: A&P Inphatec

COVID-19 and the Wine Industry: How the Pandemic Changed the Daily Lives of Industry Workers and Influenced Wine Sales

Kendal Koorenny*

*UC Davis, 1543 Drake Drive, Davis, CA 95616 (klkoorenny@ucdavis.edu)

The international wine industry has been altered greatly since COVID-19 was declared a pandemic in March of 2020. Since the pandemic began, the wine industry has faced significant challenges. These challenges include changes in wine sales (earnings, sales methods, and products sold), disruptions to the daily routines of wine industry workers, and adjustments to company marketing strategies. The industry has had to accommodate the changes caused as a result of the pandemic while remaining relevant to consumers; however, while sales and employment figures have provided quantitative data, there has yet to be any qualitative data that demonstrates the effects of the pandemic on this industry. In this whitepaper directed to the wine industry, the stories behind these changes are presented, illuminating exactly how COVID-19 has impacted the wine industry and how these adjustments are projected to influence the industry in the long term.

Funding Support: Not funded, completed as an honors thesis for my BS in viticulture and enology with the UC Davis University Honors Program

Building Wine Appreciation for the New-Generation Consumer

Kathryn Anne LaTour, Joy Annamma,* and Roger Noujeim *University of British Columbia, 3333 University Way, Kelowna/V1V 1V7, Canada (Annamma.Joy@ubc.ca)

For the wine industry, consumer education is both a goal and a challenge. Consumers show interest in learning more about wine, yet also feel intimidated and confused. Making things more difficult is the wine industry's traditional scientific approach to education, with complex terminology that is not appreciated by the younger consumer generation. While the traditional, more technical approach to wine education generated interest among more experienced consumers, little consideration was paid to consumer preferences and likeability of wine. This study examines teaching two Gen Z and millennial consumer groups about wine using a holistic, visual, and verbal analytical approach. Group A had some wine education background and Group B had none. This approach to teaching younger consumers about wine leads to them liking the wine more, especially in Group B, who had no prior wine education. Group A desired an overall more upbeat and happy approach to learning about wine than Group B consumers who were just starting to learn about wine. However, both groups desired a more pleasurable approach to wine education than a purely scientific and technical learning environment. Overall, younger consumers were interested in learning about wine, even though some experts have suggested that the industry is losing touch with the new consumer. As a prerequisite to our follow-on study, we partnered



with Quini, a digital visual wine tasting and rating platform, to examine differences in wine consumer engagement on their platform among age groups. We analyzed three years of data and followed up with an online survey to determine interest in and education about wine. This paper discusses our findings and proposes how industry executives might engage younger consumer groups, especially online.

Funding Support: School of Hotel Administration, Cornell University Quini. This research paper has been accepted for publication by the Cornell Hospitality Quarterly Journal.

Biogenic Amines Production in Mixed Culture of Lactic Acid Bacteria Isolated from Tucumán Wines

Silvana C. Ledesma, María Cristina Rubio, and Pedro A. Aredes Fernández^{*} *Facultad de Bioquímica, Química y Farmacia, Ayacucho 471, San Miguel de Tucumán/Tucumán/4000, Argentina (pedroaredes@hotmail.com)

In a co-culture, microorganisms combine their metabolic activity to degrade and metabolize the substrates present in the environment. Just as there are symbiotic relationships, there is also competition that involves mechanisms of protection and antagonism. Lactobacillus paracasei AT45, isolated from wine, produces histamine and tyramine. These amines cause vasoactive and psychoactive effects in humans. Oenococcus oeni RAM10, a proteolytic microorganism, was selected as optimal to carry out malolactic fermentation. The objective of this work was to evaluate the interaction between O. oeni RAM10 and L. paracasei AT45 in pure and mixed cultures to establish a relationship with production of biogenic amines under winemaking conditions. The assays were carried out in commercial red wine inoculated with pure cultures of O. oeni RAM10 and L. paracasei AT45 at final concentrations of 10⁷ cfu/mL and 10^3 cfu/mL, respectively, and in mixed culture at the same final concentration of both microorganisms. The trials were incubated at 23°C for 96 hr. Bacterial viability and production of histamine and tyramine were evaluated using a colorimetric method developed in our laboratory. In pure cultures, the viability of both microorganisms remained constant for all incubation times. In mixed culture, O. oeni RAM10 maintained viability until 96 hr, while L. paracasei AT45 decreased growth to undetectable values. In pure culture, O. oeni RAM10 did not produce histamine or tyramine, while L. paracasei AT45 produced a concentration of 6.52 mg/L histamine and 3.57 mg/L tyramine at 96 hr. In mixed culture, no amine production was observed. These studies demonstrate that O. oeni RAM10 in mixed culture with a harmful bacterium causes a decrease in the concentration of biogenic amines.

Funding Support: Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET)

Foliar Potassium Application Has Limited Effect on Berry Composition in Grapevine Red Blotch Virus-Infected Grapevines

Alexander Levin,* Joseph DeShields, and Achala KC *Oregon State University, 569 Hanley Rd., Central Point, OR 97502 (alexander.levin@oregonstate.edu)

Delayed ripening is one of the most commonly reported fruit symptoms in grapevine red blotch virus-infected (GRBV+) grapevines and is attributed to reduced sugar translocation. Potassium (K) nutrition is closely linked with berry ripening, and early studies on K nutrition in GRBV+ grapevines indicated low K status in GRBV+ leaves. Thus, it was hypothesized that foliar K application would improve berry ripening

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through improved leaf K status and sugar translocation. A field experiment was conducted in 2020 to test two formulations of foliar K fertilizers (0-0-24) against a water control. The three treatments were applied weekly for four weeks to previously identified healthy (GRBV-) and infected grapevines beginning at 50% veraison. At each application, the equivalent of 1.2 L/ha was applied with a backpack sprayer to eight single-vine replicates. Treatments were arranged in a randomized complete block design with a split-plot treatment structure whereby GRBV status was main plot and foliar application was split-plot. Leaf blades and petioles were sampled for nutrient analyses both before and after treatment application, and berry composition was measured at harvest. There was little to no effect of K application on K status in blades or petioles, with all values in the normal range. Berry fresh weight (FW) was slightly reduced (-10%, p = 0.003) in vines sprayed with K, independent of GRBV status. Concomitantly, berry total soluble solids (TSS) at harvest were increased (+1.6 Brix, p = 0.002) by both K fertilizers independent of GRBV status. However, there were no significant effects of K application on berry phenolic composition. The lack of treatment effects on berry composition suggests that while foliar K application has limited effects on fruit quality in GRBV+ grapevines, the increased TSS may advance harvest date in heavily infected blocks.

Funding Support: USDA-NIFA-SCRI grant number 2019-51181-30020

Fermentation Kinetics and Sensory Analysis of Microbially-Impacted Red and White Wine

Jeremiah Loyd,* Garrett Morales, Sonet Van Zyl, and Stephan Sommer *Fresno State Viticulture and Enology, 2360 E. Barstow Avenue, M/S VR89, Fresno, CA 93740 (jeremiahloyd@mail.fresnostate.edu)

Commercial grape quality is assessed visually. Contamination can potentially lead to loss of quality, yield, and profit. Fungal infections, such as Botrytis, Penicillium, and Aspergillus, can negatively affect the progression of fermentation and the overall flavor profile of the finished wine. The study objective was to use analytical testing of FT-MIR spectroscopy on incoming different levels of sour rot to assess quality, evaluate fermentation kinetics, and perform sensory analysis. Over the past three years, 13 tons of grapes have been collected to facilitate winemaking with grapes from 10 different vineyards, producing a total of 72 wines. Grapes were harvested separately from each vineyard and divided into clean and microbially impacted clusters. The visually clean grapes were dosed by percentages of infected grapes at 0, 5, 10, 15, and 20%, by weight in reds and by volume in whites. Infected grapes initially showing higher levels of volatile acidity did not directly correlate with higher volatile acidity in a finished wine, sometimes even having less. On the other hand, greater sugar levels in impacted juice did lead to more ethanol in the finished wine. 3-AFC difference testing and ranked preference testing were performed with 16 panelists who were tracked over eight groups of wines. Higher levels of impacted juice/must could not be discerned while lower levels could be differentiated.

Funding Support: California Winegrape Inspection Advisory Board Agricultural Research Institute

Testing Nematode-Resistant Rootstocks for San Joaquin Valley Viticulture Karl Lund*

*UC Cooperative Extension, 145 Tozer Street Suite 103, Madera, CA 93638 (ktlund@ucanr.edu)



Plant parasitic nematodes can extensively damage grapevine roots that are not resistant to them, especially in vineyards with sandy soils, as is common in the San Joaquin Valley of California (CA). Fumigation is an effective control measure; however, regulations have restricted the use of fumigants in CA, heightening the importance of nematode-resistant rootstocks. The development of better nematode-resistant rootstocks is an ongoing effort. Documenting the viticultural performance of scions grafted to rootstock selections is critically important for proper selection and commercial acceptance of rootstocks and should be part of the process of selecting new material for potential release. Therefore, the performance of Petit Verdot winegrapes on newer nematode-resistant rootstocks (RS-3, RS-9, GRN1, GRN2, GRN3, GRN4, and GRN5), with 1103P and Freedom as controls, were planted in a replicated trial in a commercial vineyard in Madera county. Data collected over the past two years have shown differences in canopy growth and water stress based on rootstock. Shoot count shows that the entire GRN series along with Freedom start off with more shoots at the start of the season. GRN2, GRN4, and GRN5 maintained the largest canopies throughout the season, while GRN3 stayed in the middle and GRN1 became one of the smallest. Water relations measured as midday leaf water potential showed that despite having one of the largest canopies, GRN2 stays the least stressed of all the rootstocks, while RS-3 and RS-9 are the most stressed. Interestingly, GRN3 normally maintained a midrange water stress, except when the vineyard adds in water stress postveraison. During this drv-down, GRN3 becomes one of the most water-stressed rootstocks. This indicates that GRN3 is much more susceptible to drought stress. This ongoing trial will provide useful data for growers needing nematode-resistant rootstocks.

Funding Support: American Vineyard Foundation California Grape Rootstock Research Foundation

Exploring use of Redox Potential to Predict Fermentation Outcomes in Relation to Initial Juice Conditions

Gita Mallya, Ben Montpetit,* James Nelson, Kimberlee Marinelli, Ron Runnebaum, and Andre Knoesen

*UC Davis, 1 Shields Avenue, Davis, CA 95616 (benmontpetit@ucdavis.edu)

Each year in the wine industry, economic loss occurs due to stuck or sluggish fermentations and the corresponding off-flavors produced. While monitoring by using standard methods such as total soluble solids (density) may reveal a problem, often this occurs after wine quality has already been impacted and remediation techniques are less effective, more intrusive, and costly. The use of redox potential, or oxidation reduction potential (ORP), as a process parameter is being explored to predict fermentation outcomes early in fermentation, even before measurable changes in total soluble solids occur. ORP describes the tendency for molecules or ions to gain or lose electrons in relation to the chemical makeup of a solution being measured. Consequently, ORP values are sensitive to the fermentative activity of the yeast as metabolic products are released and alter the chemical conditions of the solution. This makes ORP a sensitive tool for understanding the state of the fermenting yeast in a must, even before sugar consumption can be measured. This study aimed to monitor ORP under varying nutrient conditions and with different yeast strains to better understand the relationship between ORP and fermentation outcomes. Several wine strains of Saccharomyces cerevisiae (EC1118, Elixir, CY3079, Montrachet, and RC212) were observed, as well as varying pH and nutrient conditions. Preliminary ORP data for stuck fermentations show indications of failure sooner than would a slowing sugar consumption rate.

Funding Support: Wine Spectator, ASEV, Jastro Shields Research Award

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Development of a GMO-Free RNA Interference Approach to Mitigate the Negative Effects of Red Blotch Disease on Grapevine

Christian Mandelli,* Emanuelle Chrysilla, and Laurent Deluc *Oregon State University, 2750 SW Campus Way, Corvallis, OR 97331 (mandellc@oregonstate.edu)

In plant-pathogen interactions, small RNAs (sRNAs) ranging from 21 to 24 nucleotides have a pivotal role in inducing epigenetic and post-transcriptional gene silencing via RNA interference mechanisms. This knowledge is the basis for new crop protection technologies that mostly rely on the production of transgenic plants expressing a double-stranded RNA molecule specific to pathogen sequence(s). This induces in the host the RNA interference mechanism, also termed host-induced gene silencing (HIGS). However, the use of genetically modified organisms (GMOs) raised scientific and public concerns. A new innovative strategy, termed spray-induced gene silencing (SIGS), allows RNAi induction without requiring plant genetic modification. Spraving dsRNAs on the leaf surface results in their cellular uptake and processing into siRNAs. Our group developed a long-term research project aiming to exploit this technology to control grapevine red-blotch disease. As a first step, we aim to characterize the responses of grapevine following grapevine red blotch virus (GRBV) infection by producing i) the landscape of small RNAs produced during the early steps of infection, ii) the methylome targeting the viral genome, and iii) the grapevine transcriptome associated with plant pathogen responses. To achieve these objectives, we will use Agro-drenching and green grafting for the plant infections and next generation sequencing for transcript characterization. Altogether, we hope to identify the "hot spots" for transcriptional and post-transcriptional gene silencing during the early phase of infection and use them to design the dsRNAs necessary for SIGS application.

Funding Support: California Department of Food and Agriculture

Solar Radiation Exclusion Reduced Evapotranspiration and Improved Skin Flavonoid Content of Winegrapes

Lauren Marigliano, Nazareth Torres, Runze Yu, Mark Battany, and Kaan Kurtural* *University of California Davis, 1 Shields Avenue, Davis, CA 95616 (skkurtural@ucdavis.edu)

Climate conditions are expected to increase temperature and water deficits in viticulture regions by the mid-21st century, impacting grapevine physiology and production sustainability. Photoselective overhead shade films (D1, D3, D4, D5) with varying degrees of solar spectra exclusion were installed and compared to an uncovered control (CO) in a randomized complete block design with four replicates. The goal of the experiment was to evaluate the vulnerability of Cabernet Sauvignon grape berry to solar radiation overexposure and optimize the use of shade films for berry development. Results indicated that CO had significantly greater evapotranspiration than shaded treatments. Consequently, the water footprint (m³/tonnes) of CO was significantly greater than the shaded treatments with no difference in yield. Throughout berry ripening and at harvest, total skin anthocyanins (mg/g fresh mass) in the shaded treatments were greater than CO and the molar abundance of kaempferol (%) was lowest in D1 and D3. Relative yield was not different between the control and treatments, although dormant season precipitation was excluded from shaded treatments. Our results indicated that shade films reduced evapotranspiration from the grapevine canopy in the hot climate without reducing yield, while arresting anthocyanin and flavonol degradation relative to exposed grapevines.

Funding Support: University of California Davis Foundation



Investigating the Impact of Oxygenation on Redox Potential and Fermentation Kinetics

Kimberlee Marinelli, Ben Montpetit,* Ron Runnebaum, James Nelson, Gita Mallya, and Andre Knoesen

*University of California, Davis, 1136 Robert Mondavi Institute North, 595 Hilgard Lane, Davis, CA 95616 (benmontpetit@ucdavis.edu)

In the wine industry, tracking redox potential, or ORP, during fermentation is an emerging process parameter. ORP can offer a valuable, real-time indicator of yeast metabolism during fermentation and could be used to make decisions around aeration to prevent formation of unwanted compounds associated with reductive fermentation conditions. The goal of this study was to gain a better understanding of how oxygen introduction, at various times and amounts during the course of fermentation, affects redox status and how this is linked to overall fermentation outcomes. Using juice concentrate as a consistent fermentation medium, the impact of oxygenation on redox potential and fermentation kinetics was investigated at lab scale in triplicate, including experiments set out to evaluate when introduction of air during fermentation is required to improve fermentation performance. Total soluble solids, redox, and cell density were measured in all trials and compared against un-aerated ferments and across those subject to different oxygenation time regiments. Data collected to date shows that aerated ferments with higher redox values have faster fermentation kinetics and reach overall greater cell densities than those that were un-aerated. In addition, the timing of aeration was found to be important for this effect. Future work will investigate the amount of oxygen required, i.e., target ORP values and how these additions may alter wine chemistry.

Funding Support: N/A

Latic Acid Concentrations in Pinot noir Wines from Several Vineyards from Oregon to Southern California

Maisa Martins Monteiro Lima,* Desmon Hernandez, Taylor Reiter, Ben Montpetit, and Ron C. Runnebaum *UC DAVIS, 3127 RMI North Viticulture & Enology, Davis, CA 95616 (mmlima@ucdavis.edu)

The conversion of malic acid to lactic acid is responsible for deacidification, flavor modifications, and microbial stability of wines. The presence of lactic acid in wines is generally through malolactic fermentation (MLF). Lactic acid can also be formed by the presence of microorganisms on grapes or in the winery. The presence of lactic acid in juice at the beginning of fermentation has been shown to have an effect on fermentation outcomes in non-wine fermentations. Gene expression data of Pinot noir wines has shown instances of fermentation outcomes that are consistent with the presence of lactic acid in early fermentation from select vineyards. To better understand the presence and reproducibility across vintages of lactic acid in juice, three vintages (2017 to 2019) of Pinot noir wines from 15 vineyard sites were evaluated. Pinot noir grapes harvested represented eight American Viticultural Areas along the United States West Coast. The wines were produced with a goal of minimizing potential sources of variation, including using a single scion clone of Pinot noir and reproducible and replicated winemaking. Grapes were destemmed and inoculated with Saccharomyces cerevisiae RC212 yeast. MLF was completed by inoculation of *Oenococcus oeni* and the wines were stored in stainless steel vessels until bottling. Malic acid in juices was characterized by enzymatic assay prior. Lactic acid in wines was characterized using high-performance liquid chromatography. Wines from

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specific vineyard sites had more lactic acid concentration than possible after MLF on the basis of initial malic acid concentration (actual and theorical difference in lactic acid concentrations). Such differences suggest that significant levels of lactic acid were present prior to start of fermentation at these sites. These chemical composition results will be compared and contrasted with RC212 gene expression data from those same sites during primary fermentation.

Funding Support: Jackson Family Wines

Comparison of Nano- and Microscale Fermentations for Evaluation of Risks Associated with Smoke Exposure

Caroline Merrell, Bojana Leonard, Samantha Young, Torey Arvik, and Thomas S Collins* *Washington State University, 359 University Drive, Richland, WA 99354 (tom.collins@wsu.edu)

Microscale or bucket-scale fermentations have been used to evaluate the impact of vineyard smoke exposures prior to harvest. While chemical analysis or sensory evaluation of the resulting wines can be used to identify affected blocks, a key limitation for their usefulness is the amount of time required to complete fermentation, along with lack of temperature control and the amount of space required. Using smaller-scale fermentations in glass canning jars, fermentation temperatures can be maintained in ad hoc water baths warmed by sous vide heaters. At 85°F, nanoscale fermentations can be completed in three to four days. Space requirements are also greatly reduced with canning jar fermentations. To evaluate whether the different fermentation scales produce comparable results, 12 lots of smoke-affected Cabernet Sauvignon grapes were fermented in triplicate using both systems. The microscale fermentations completed fermentation in eight days, while the nanoscale fermentations completed fermentation in four to five days. Guaiacol concentrations in the affected wines ranged from not detected to $3.2 \,\mu g/L$ in the microscale fermentations, compared to 1.6 to 5.2 μ g/L in the nanoscale. Concentrations of the other smoke-exposure markers also correlated well between the two fermentation scales. Fifteen commercial vineyard lots were also evaluated using both nano- and microscale fermentations, both of which were compared with the results of the commercial fermentation of those vineyard lots. Concentrations of smoke-exposure markers were similar in both the - and nanoscale fermentations and concentrations of marker compounds in both scales correlated well with concentrations in the production-scale wines. These results suggest that the use of nanoscale temperature-controlled fermentations can provide more timely results, with good predictive ability of concentration of smoke exposure compounds in production-scale wines.

Funding Support: Washington State Wine Commission Jackson Family Wines

Calibration of Microbial Spoilage Indicators via Mycobiota Levels in Winegrapes

Garrett Morales, Sonet Van Zyl,* Jeremiah Loyd, and Stephen Sommer *CSU Fresno, 2360 E. Barstow Avenue, Fresno, CA 93740 (svanzyl@mail.fresnostate.edu)

Within the San Joaquin Valley, daytime temperatures during the peak of the winegrape harvest season can reach well over 37°C. As temperatures increase, biological and chemical reactions also tend to increase. Because of this, winegrapes are typically harvested in the early morning to combat the effects of these biological and



chemical reactions, i.e., spoilage of the winegrapes by microorganisms. As many vineyards and processing facilities are spread out over large distances in the state, longer wait times at winery test stands give grapes in the gondolas the opportunity to be exposed to elevated temperatures. This in turn allows microbial spoilage to occur within the gondola before reaching the test site. With no standard method of measuring the impact of microbial spoilage within the gondola, other than visual observations, absorbance by FT-MIR spectroscopy is being used to create a standard. Fresno State has worked for 10 years on a calibration method using clean winegrapes inoculated with the most common forms of spoilage found in the Central Valley: Aspergillus niger, Rhizopus stolonifera, and Penicillium italicum. These molds are dosed by weight to create a linear trend that can measure the amount of microbial spoilage. Thirty-six samples were taken from nine different varieties during the 2019 harvest as well as 30 samples taken from 10 different varieties during the 2020 harvest to build upon this model. Analytical attributes that were tracked throughout this study, which aid in monitoring the amounts of spoilage, include total soluble solids, tritratable acidity, ethanol, volatile acidity, tartaric acid, gluconic acid, and glucose. Currently, the most predominant indicators of spoilage appear to be ethanol, volatile acidity, and gluconic acid

Funding Support: California Winegrape Inspection Advisory Board, Agriculture Research Institute

Utilizing Non-Saccharomyces Yeast as Bio-Protectants during Prefermentation Cold Soaking

Melanie Nicholls, Michael Qian, and James Osborne* *Oregon State University, Wiegand Hall 108A, 3051 SW Campus Way, Corvallis, OR 97331 (james.osborne@oregonstate.edu)

This study investigated whether adding a non-Saccharomyces yeast culture during prefermentation cold soaking could act as a bio-protectant against Hanseniaspora uvarum. Initial experiments tested the ability of three non-Saccharomyces yeast cultures to suppress H. uvarum and acetic acid production during a simulated cold soak in a model grape juice. Two different initial populations of *H. uvarum* were inoculated to mimic healthy grapes (low H. uvarum population) or damaged grapes (high H. uvarum population). All three non-Saccharomyces yeasts repressed the growth of H. uvarum to varying degrees. Metschnikowia fructicola and Torulaspora delbrueckii were more effective than Lachancea thermotolerans and growth repression was greater if the initial population of H. uvarum was low. The addition of non-Saccharomyces cultures also decreased production of acetic acid by *H. uvarum* by up to 50%. Additional experiments tested the efficacy of the non-Saccharomyces cultures at different temperatures, with or without added SO₂. M. fructicola was still effective at repressing H. uvarum growth and acetic acid production after an addition of 20 mg/L SO₂, but T. delbrueckii was only effective if no SO₂ was added. At 40 mg/L SO₂, H. uvarum populations declined whether M. fructicola or T. delbrueckii had been added or not. However, H. uvarum populations recovered near the end of the cold soak when grown alone, while populations remained low if M. fructicola or T. delbrueckii was added. The greatest reduction in acetic acid occurred when M. fructicola and 20 mg/L SO₂ were added. In these treatments, a 58% reduction in acetic acid was measured. Additional experiments are currently being conducted using Pinot noir grapes.

Funding Support: Northwest Center for Small Fruits Research

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How Do Consumers Treat Wines Post-Purchase?

Daniel Peters and Hildegarde Heymann*

*University of California Davis, 595 Hilgard Lane, Davis, CA 95616 (hheymann@ucdavis.edu)

Consumer preferences are a key component to consider when producing a wine. Understanding preferences helps producers target their market of interest and can solidify consumer satisfaction. Considering how consumers treat wines post-purchase is important, since purchasers who handle wine negligently potentially alter the wine and thus, their perception of it. Knowing consumer preferences and treatment habits post-purchase allow us to modify winemaking practices to fit consumer inclinations and educate consumers on proper care. We surveyed wine consumers across the world to clarify these areas of interest and to possibly identify significant differences in specific locations. This survey was distributed through personal, professional, and social media channels. Possibly the most revealing associations were observed with consumers who prefer red wine, white wine, or who have no preference. Significant values (χ^2 , $\alpha < 0.05$) were observed for consumers who prefer red wine, purchase red wine with a cork closure, and purchase white with a cork closure. Significant values for consumers who prefer white wine include drinking wine monthly, purchasing white wine with a screwcap, indifference to white wine closure, and not owning a wine preservation device. Significant values were obtained for consumers who have no wine preference and are indifferent to both white wine and red wine closures. Temperature at which consumers store red or white wine depends on the wine and consumers consider themselves to be very knowledgeable. Our results suggest that consumers who prefer red wine are more likely to purchase red and white wines with a cork. Consumers that prefer white wine may be less particular, not own a wine preservation device, and gravitate toward white wines with screwcaps. Consumers with no red or white wine preference may be fairly knowledgeable about wine and less biased in their purchasing decisions.

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Weakening Ethanol Synthesis Capacity of *Saccharomyces cerevisiae* with a Randomly Mutated *SPT15* Transcription Regulator

Yi Qin* and Qing Du

*College of Enology, Northwest A&F University, 22 Xinong Rd, Yangling, Shaanxi, 712100, China (qinyi@nwsuaf.edu.cn)

With global warming, over recent decades the alcohol concentrations of wines from warm regions around the world has increased by -2% (v/v). There is significant interest in the wine industry to develop methods to reduce the ethanol content of wine. Generally, microbiological strategies to isolate and/or generate the yeast strains used to make wine, including *Saccharomyces cerevisiae* and non-conventional yeast species, have proved the simplest and most economic methods. We used global transcriptional machinery engineering (gTME) technology, based on the mutation of the *SPTI5* gene, to weaken the capacity of *S. cerevisiae* to produce ethanol and ultimately created a new strain of *S. cerevisiae*, YS59-409, with ethanol-production capacity reduced by 34.9% below that of the control strain. Sequence analysis was performed on the mutated Spt15p, demonstrating that the five mutation sites (Ile 46 Met, Asp 56 Gly, Ser 118 Pro, Tyr 195 His, and Leu 205 Ser) may work collectively, or at least in part, to create the specific characteristics of *S. cerevisiae* YS59-409, including greater CO₂ release, biomass, and glycerol formation. Integration of RNA-Seq and metabolomics analysis showed that the specific phenotype of the *S. cerevisiae* YS59-409 featured



changes in ribosome biogenesis, nucleotide metabolism, glycolysis flux, the Crabtree effect, NAD⁺/NADH homeostasis, and energy metabolism. Furthermore, two genes related to energy metabolism, *RGI1* and *RGI2*, were associated with the weakened ethanol production capacity, although the precise mechanisms involved are still unclear. This study highlights the potential of gTME technology to reduce the ethanol content of yeast for the winemaking industry.

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Effects of Alcohol Removal on the Chemical Composition of Wine Using Low-Vacuum, Thin-Film Distillation

Kelsey Quetsch*

*Conetech, 1450 Airport Blvd, Ste 180, Santa Rosa, CA 95403 (kquetsch@conetech.com)

Alcohol removal is becoming a more commonly used winemaking technique as climate change and consumer trends impact the trajectory of the wine industry. As global temperatures steadily increase due to climate change, grapes are producing more sugar and less acid, resulting in higher-alcohol wines. In contrast, consumer trends are steadily transitioning to healthier lifestyle options such as lower-alcohol wines. One alcohol removal option is low-vacuum, thin-film distillation, a distillation technique that uses reduced pressure to separate alcohol at a lower boiling point than at ambient pressure. Low-vacuum, thin-film distillation has been used for alcohol adjustment and production of high-quality low- and no-alcohol wines, but there is little information detailing how wine is affected during this process. Trials were conducted to better understand how alcohol removal via low vacuum thin film distillation affected the chemical composition of wine by analyzing the same wine before and after alcohol removal for pH, TA, RS, color, and F/T SO2. Trials have shown that the removal of alcohol via this method increases the concentrations of specific chemical components present in wine such as tartaric acid, glucose, and fructose, and compounds associated with color, but reduces volatile compounds such as sulfur dioxide.

Funding Support: ConeTech

Hybrid Trunk Disease Evaluation: A Serendipitous Opportunity

Paul Read,* Benjamin Loseke, and Stephen Gamet

*University of Nebraska - Lincoln, Plant Sciences Hall (PLSH) 377J, Lincoln, NE 68583 (pread1@unl.edu)

Declining yields in Nebraska vineyards and in the Midwest generally are variously attributed to vine aging, winter injury, or other environmental factors. Only recently have trunk diseases become suspect, largely because of concerns raised by Richard Smart in a June 2018 visit. A serendipitous opportunity arose for the University of Nebraska Viticulture Program (UNVP) to investigate trunk disease symptoms when a 20-year-old cultivar research planting was terminated in August 2018. This planting was on a commercial vineyard at Nebraska City, NE, and contained 36 cultivars, with the oldest vines planted in 1999. The vines were "destructively harvested" aboveground and evaluated for grapevine trunk disease (GTD) symptoms—a minimum of two replicates of 21 cultivars of hybrid grapevines, most of which had been in the ground for nearly 20 years. All were trained to a high-wire double cordon

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system. Evaluations were based on observations of both visible staining and dead wood symptoms found in cross-sections at five locations: 15 cm from the distal end of the cordon, at mid-cordon, at the juncture of the cordon and the trunk (crown), and at 90 cm and 10 cm aboveground on the trunk. All of the cultivars evaluated exhibited recognizable symptoms of TD, and some cross-sections also exhibited textbook symptoms of *Eutypa* and/or *Botryosphaeria* wedge-shape cankers. Severity of cross-section symptoms was rated on a scale of 1 (no visible symptoms) to 10 (cross-section completely stained or dead) using visual assessment.

Funding Support: Nebraska Grape and Wine Board

Effect of Contrasting Fermentation Temperature Regimes on Three Dijon Clones of Pinot noir over Two Consecutive Vintages

Josh Reynolds and Federico Casassa*

*Wine and Viticulture Department - Cal Poly San Luis Obispo, 1 Grand Avenue, San Luis Obispo, CA 93410 (Icasassa@calpoly.edu)

Three different Pinot noir clones (115, 777, and 828) grown in California (Santa Maria Valley AVA) were subjected to different maceration temperature regimes (cold: 10°C, hot: 25°C, and variable: 10°C first week/25°C second week) to determine the effect of maceration temperature on phenolic extraction across all three clones over two consecutive vintages (2019 and 2020). Triplicate fermentations were conducted for 14 days and anthocyanins, tannins, polymeric pigments, and total phenolics were followed during winemaking. Temperature across treatments stayed consistently higher in hot trials than in cold trials by at least 12°C throughout maceration, and fermentation reached completion up to four days earlier in hot trials across both vintages. Using aggregate data from all three clones across both vintages, measured at time of pressing, extraction of total phenolic compounds was 723 mg/L for cold treatments, 894 mg/L for variable treatments, and 1031 mg/L for hot treatments. The same data also shows differences among cold, variable, and hot treatments in anthocyanins (208, 265, and 284 mg/L, respectively), tannins (68, 81, and 123 mg/L, respectively), and polymeric pigments (0.40, 0.46, and 0.51 mg/L, respectively). Trends across the three clones were also visible using both vintages, with the lowest phenolic concentrations across the board in 115 and the greatest in 777. Vintage effect was present, with higher concentrations of total phenolics, tannins, and polymeric pigments in 2019 and higher values for anthocyanins in 2020. Color analysis of the averages of all three clones indicates negative correlation with temperature for Cielab lightness values across cold, variable, and hot (88.81, 87.35, and 85.36, respectively), with positive correlation for red values (9.41, 11.14, and 12.10, respectively), similar yellow values (6.42, 6.05, and 6.62, respectively), and positive correlation for color values, 420 + 520 + 620 nm (0.38, 0.44, and 0.52, respectively), measured at 1-month postbottling.

Funding Support: World of Pinot noir

Effects of Grapevine Red Blotch Virus (GRBV) on Grape Metabolic Pathways Through Ripening

Arran Rumbaugh, Blythe Durbin-Johnson, Monica Britton, Raul C. Girardello, Mysore Sudarshana, and Anita Oberholster* *University of California, Davis, One Shields Ave, Davis, CA 95616 (aoberholster@ucdavis.edu)

Grapevine red blotch virus (GRBV), causative agent of red blotch disease, is a single-stranded DNA virus that is part of the *Geminiviridae* family. GRBV causes delayed



ripening with significant decreases in sugar accumulation and color development that alter wine sensory characteristics. We investigated the viral impact on transcription factors and metabolite accumulation across genotypic and environmental factors. Healthy and diseased grapes were collected from Cabernet Sauvignon (CS, grafted on 420A or 110R rootstocks), at preveraison, veraison, postveraison, and harvest in 2016 and 2017. RNA in grapes was sequenced to determine viral gene expression and differences in grape gene regulation. Grape metabolites were measured using UPLC-QTOF-MS, NMR, and HS-SPME-GC-MS. Across all rootstocks and seasons, viral gene expression was greatest at preveraison, steadily decreased through ripening, and was greater in 2016 than in 2017. This correlated with fewer differences in transcription factors between healthy and infected grapes in 2017 than in 2016. Between CS 110R and CS 420A in both seasons, there were 97 common differentially expressed genes due to GRBV infection. These genes are generally responsible for key primary metabolic processes, response to stimulus, and gene silencing that could be related to viral infection. GRBV inhibited the phenylpropanoid pathway, which decreased flavonoid compound concentrations. Some metabolic pathways in postveraison and harvest samples that were induced due to GRBV are normally associated with early fruit ripening. One of these pathways was the lipoxygenase pathway, involved in synthesis of C₆ alcohol and aldehyde compounds. This generally correlated with significantly lower concentrations of these compounds through ripening.

Funding Support: California Department of Food and Agriculture

Reproducibility of Site Contributions to Elemental Composition of Pinot noir Wines over Multiple Vintages

Maisa Lima, Desmon Hernandez, and **Ron Runnebaum*** *University of California - Davis, 595 Hilgard Lane, Davis, CA 95616 (rcrunnebaum@ucdavis.edu)

Correlations between vineyard site and wine are historically limited due to lack of continuity across multiple vintages, lack of uniformity in scion clone, and lack of controlled pilot-scale winemaking conditions. We determined the elemental composition characterizing wines from four vintages (2015 to 2018). The experimental design aimed to minimize sources of potential variation by using a single scion clone and automated 200-L fermentation vessels at the UC Davis Teaching and Research Winery, in which fermentations are highly controlled across vineyard replicates, vineyards, and vintages. Grape clusters were hand-harvested from vineyards that span a distance of >1400 km. American Viticultural Areas (AVAs) represented included Santa Rita Hills, Santa Maria Valley, Arroyo Seco, Carneros, Sonoma Coast, Russian River Valley, Anderson Valley, and Willamette Valley. Fruit was destemmed only and inoculated with Saccharomyces cerevisiae yeast. Upon completion of inoculated MLF, wines were stored in stainless steel vessels until sampling for characterization. Forty-seven elements were profiled in a mass range of 7 to 238 m/z by using inductively coupled plasmamass spectrometry. Principal component analysis was used to characterize vineyards using only significant elements identified by an analysis of variance measuring effects of vineyard. AVAs were generally separated by elemental composition profile, such as Santa Maria Valley and Arroyo Seco. Some vineyard locations, however, were more similar in elemental composition to sites in other AVAs than to those within their AVA. Differences in profiles within an AVA suggest that factors such as unique soil composition or conditions or microclimate may have an effect. Overall, separation and clustering of wines by elemental composition appears consistent across vintages in this experiment. Details to be unraveled include confirming the consistency of

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elemental profile from sites across vintages and correlations with wine chemistry and sensory profile.

Funding Support: Jackson Family Wines

Arbuscular Mycorrhizal Fungi Do Not Enhance Nitrogen Uptake in Pinot noir when Phosphorus Supply is Adequate

Tian Tian and R. Paul Schreiner*

* USDA-ARS Horticultural Crop Research Laboratory, 3420 NW Orchard Avenue, Corvallis, OR 97330 (Paul.Schreiner@usda.gov)

Grapevines form associations with arbuscular mycorrhizal fungi (AMF) that enhance nutrient uptake, especially of phosphorus (P). Studies in other crops indicate that AMF can increase plant nitrogen (N) acquisition, but whether AMF improve N uptake in grapevines is unclear. To understand whether AMF play a role in N uptake, responses of mycorrhizal and non-mycorrhizal Pinot noir grapevines were compared using a range of N and P inputs in the greenhouse. A soil with moderate P was used to ensure vines were not P-deficient, which would mask the ability to test whether AMF increase N uptake. A factorial experiment with four rates of N (0, 3, 6, or 12 mM N as ammonium nitrate), three rates of P (0, 1, or 2 mM P as potassium phosphate), and two AMF treatments (mycorrhizal, non-mycorrhizal) was conducted using potted vines. Shoot growth and mycorrhizal colonization of roots were periodically assessed, then vines were destructively harvested to determine biomass and nutrient uptake after exposure to different N and P treatments for nine weeks. Shoot and root biomass increased with increasing N supply, but shoot length and dry weight were both suppressed by AMF at intermediate levels of N. P supply did not alter vine biomass. Vine N uptake increased with N supply, but N uptake was not improved by AMF at any level of N or P supply. Across all treatments, AMF enhanced uptake of P and zinc. Root colonization by AMF was reduced at the lowest rate of N supply, while arbuscules in roots were more abundant at intermediate rates of N coincident with reduced shoot growth. These findings indicate that AMF do not enhance N uptake when vines receive inorganic N and that AMF may compete with shoots for carbon when N supply is moderate and P is ample for growth.

Funding Support: USDA-ARS

Co-Expression Network-Based Analysis of Genes Associated with Leaf Temperature and Stomatal Conductance in Grapevine

Harshraj Shinde, Yikang Hu, Kiflu Tesfamicael, Penny J. Tricker, Ute Baumann, Everard J. Edwards, and Carlos M. Rodriguez Lopez* *University of Kentucky, 1405 Veterans Dr, 432, Lexington, KY 40506 (cmro267@uky.edu)

Grapevine (*Vitis vinifera* L. cv. Cabernet Sauvignon) is widely used for winemaking all over the world. Quality and yield is challenged increasingly by environmental stresses. Leaf temperature, stomatal conductance, and stem water potential change rapidly in response to abiotic stress. Our study aimed to screen genes regulating these physiological changes in grapevine. We applied weighted correlation network analysis and clustering to associate changes in physiological parameters with gene expression data. Four significant modules containing 3521 genes and six clusters containing 14,143 genes were identified from transcriptome data. The darkmagenta module showed a strong positive correlation with leaf temperature and significant negative correlation with stem water potential and stomatal conductance. Five hub genes



were identified from the darkmagenta module; these genes are involved in reactive oxygen species scavenging, histone methylation, and so on. Gene ontology analyses of modules and clusters highlighted key biological processes as biological regulation, response to stimulus and response to stress. Pathway analyses highlighted key pathways as thermogenesis, plant hormone signal transduction and protein processing in endoplasmic reticulum. The candidate genes and metabolic pathways identified in this study are valuable genetic resources or targets for future breeding programs.

Funding Support: Australian grape and wine authorities grants, Grant ID: VA1503.

Control of Vine Mealybug (*Planococcus ficus* Signoret) in Organic Viticulture

Khushwinder Singh, Kent Daane, and Luca Brillante* *Department of Viticulture and Enology, California State University Fresno, 2360 E Barstow Ave, Fresno, CA 93740 (lucabrillante@csufresno.edu)

The control of vine mealybug (Planococcus ficus Signoret) is complex in conventional viticulture, but even harder in organic production. We conducted a large vineyard trial (Pinot noir) to compare the efficacy of commercially available insecticides for organic production: pyrethrins, neem oil, diatomaceous earth, and Chromobacterium subtsugae. We also controlled for side effects on grapevine physiology and grape composition related to spraying dust and oils on leaves and berries. The experiment was set up as a randomized complete block design with four treatments plus control and four replicates. Each replicate was 1 acre large for a total trial size of 20 acres. The products performed similarly and none were effective in controlling mealybugs on the trunk. Pyrethrins were least effective in controlling mealybugs on leaves, while diatomaceous earth and neem oil were most effective, but not significantly different from the control. An inverse relationship appeared between efficacy on leaves and presence in the cluster. The most effective products had more mealybugs inside the clusters and the least effective products on leaves had fewer mealybugs in the clusters. Plants treated with neem oil had significantly lower stem water potentials. Treatments did not have any significant effect on gas exchange measurements, except on one date, when the neem treatment had cooler leaves and more photosynthesis and stomatal conductance. Diatomaceous earth never significantly reduced photosynthesis or stomatal conductance, even though leaves were covered with dust. Although not statistically significant, we observed a strong tendency in sugar content. Total soluble solids were lower in the control than in all other treatments and reached up to 1.9 Brix difference on the second measurement date. This delay in ripening showed as a significant difference in pH, where the control had a significantly lower pH than most other treatments.

Funding Support: American Vineyard Foundation, Bronco Wine Co.,

Monitoring Grapevine Response to Calcium-Based Soil Amendments through Remote Sensing in the San Joaquin Valley, California

Khushwinder Singh, Paolo De Censi, and Luca Brillante^{*} *Department of Viticulture and Enology, California State University Fresno, 2360 E Barstow Ave, Fresno, CA 93740 (lucabrillante@csufresno.edu)

Alleviation of salt-related problems is crucial to increase soil health in the San Joaquin Valley of California. This study monitored the response of soil physics, grapevine physiology, and fruit composition to different dosages and forms of $CaSO_4$ (anhydrite [CaSO₄] and gypsum [CaSO₄2H₂O]) in synergy with organic matter (biosolids). The

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experiment was performed for two years, 2020 and 2021, in a Merlot vineyard located near Bakersfield with a sodic soil. The experiment was a completely randomized block design with six treatments replicated four times and broadcast in winter 2020 (2.5 t/ac gypsum, 5.1 t/ac gypsum, 10.2 t/ac gypsum, 5.1 t/ac anhydrite, and 5.1 t/ac gypsum + biosolids) after the first season of measurements to ensure no differences across treatments before application. Each experimental unit has a 30 × 30 m surface that overlaps with a pixel from Landsat 8. Biweekly measurements of stem water potential and leaf gas exchange showed moderate to severe water stress but did not evidence significant differences across treatments in plant water status, carbon assimilation, stomatal conductance, or water use efficiency. Treatments had similar values in grape soluble solids, pH, titratable acidity, and anthocyanin profile measured during ripening. The yield was greatest in the 10.2 t/ac gypsum and 5.1 t/ac gypsum + compost treatments. Soil infiltration measurements showed that gypsum treatments increased infiltration more than the control or anhydrite treatments. The Normalized Difference Vegetation Index measured from Landsat 8 decreased during the season and the Crop Response to Salinity Index had a strong but similar drop during preveraison at the time of greatest water stress. We expect more differences across treatments to manifest in year two, as the reaction of soil amendments in the soil is a slow process.

Funding Support: American Vineyard Foundation

Sustainable Replacement Strategies for Bentonite in Wine Using Renewable Fining Agents

Stephan Sommer* and Federico Tondini

*California State University, Department of Viticulture and Enology, 2360 E. Barstow Ave. M/S VR89, Fresno, CA 93740 (ssommer@csufresno.edu)

Protein stability is an important quality attribute in wines, where haze in the bottled product will lead to consumer rejection. Traditionally, stability is achieved by bentonite addition; however, environmental concerns and disposal problems mean that alternatives are required to achieve the same goal. We evaluated Sacharomyces paradoxus, chitosan, polystyrene, carboxymethyl cellulose, and bentonite as such alternatives. Trials in finished wines were agitated for 10 hr overnight and analyzed for turbidity and color characteristics spectrophotometrically. Experiments were conducted with wines that were expected to develop protein instabilities: Muscat Canelli, White Zinfandel, Cabernet Sauvignon blanc de noir, Barbera rosé, and Touriga Nacional. Results indicate that S. paradoxus can help remove proteins from wine. Wines with low protein instability can be stabilized by S. paradoxus and by polystyrene and chitosan to a lesser degree. All fining agents except bentonite showed variability in fining efficiency between white and red wines that can be explained by matrix interferences in red wine and the specific reaction mechanisms of the additive. With an average protein reduction ~50%, none of the alternative fining methods could reach the efficiency level of bentonite. Experiments in a model system confirm the findings and explain some of the mechanisms involved: for example, the specificity of chitosan and challenges related to the use of yeast as a fining agent. Carboxy methylcellulose and polystyrene show promise but need to be evaluated further to optimize the application procedure.

Funding Support: n/a



Evaluation of Cabernet Sauvignon Microbial Diversity during Grape Maturation under Rain-Shelter Cultivation Mode

Yuyang Song* and Rong Huang

*College of Enology, Northwest A&F University, College of Enology, Northwest A&F University, 712100, China (yuyangsong@nwsuaf.edu.cn)

The rain-shelter cultivation model offers protection against heavy rainfall in wine regions affected by continental climates with monsoonal tendencies, as this impacts wine terroir. Here, we investigate the effects of this cultivation mode, associated meteorological factors, and phenological periods on the diversity of grape-related microorganisms. Actinomycetes, Bacteroidetes, and Firmicutes were the primary bacterial phyla in vineyards, while Ascomycota and Anthophyta were the most important fungal phyla. The soil had a more complex bacterial composition than the leaves, branches, and fruit. Fungal diversity between tissues was similar, but not bacterial diversity. Based on the interactive analysis of meteorological factors and microbial diversity, we conclude that air temperature and UV had the greatest impact on community composition and bacterial species variation. The contribution of precipitation and soil humidity on fungal distribution was significant. Overall, the differences in meteorological factors caused by the cultivation modes significantly regulated distribution of microorganisms. Understanding the succession of the microbial community and how the microclimate influences the microbial community structure has direct practical significance in wine terroir and could provide information on sustainable management practices to maintain and enhance microbial diversity.

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Differences in Cabernet Sauvignon Grapes and Wines between Madera AVA and other California Wine Regions

Qun Sun,* George Zhuang, Antonios Marinos, David Garcia, and John Wilson *California State University, Fresno, 2360 E. Barstow Avenue, M/S VR89, Fresno, CA 93740 (qsun@csufresno.edu)

Cabernet Sauvignon is planted widely in California's Madera AVA (crush district 13). However, the price per ton for Cabernet Sauvignon in this region is much lower than in other California wine regions. It is becoming progressively more challenging to maintain economic sustainability and competitiveness to grow Cabernet Sauvignon in this region. The overall goals for this study were 1) to understand the differences in characteristics of Cabernet Sauvignon grapes and wines between Madera AVA and other California wine regions and to determine which fruit traits drive the quality difference; and 2) to investigate whether mechanical leafing and deficit irrigation could achieve the desired fruit quality, then guide Madera AVA grapegrowers to use appropriate farming practice. Cabernet Sauvignon berries were harvested from commercial vineyards in the Madera AVA and five other regions (McFarland, Linden, Livermore, Paicines, and San Lucas). Cultural practices were carried out according to commercial industry standards for that area. In a Madera vineyard, the experiment was a two (deficit irrigation: 50% ET_c and 80% ET_c) × three (leaf removal at bloom or berry set and control) factorial trial with a split block design and five replicated blocks. Wines

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were made in triplicate for each location. Berries from the Madera AVA treated with deficit irrigation (50% ET_c) combined with mechanical removal at bloom had more total berry anthocyanin, total phenolic compounds, and wine color intensity. These quality indicators were better than those from McFarland and close to Linden. Sensory evaluation indicated that the wine made from treated berries in Madera AVA was fruity, peppery, and balanced with sourness, bitterness, and astringency.

Funding Support: American Vineyard Foundation

Differentially Expressed Genes in Grapevine Associated with Epigenetic Changes Identified Under Combined Stress

Jia Tan, Yikang Hu, Harshraj Shinde, Kiflu Tesfamicael, and Carlos Rodriguez-Lopez* *University of Kentucky, 1405 Veterans Drive, 105 Plant Science Building, Lexington, KY 40546 (carlos.rodriguezlopez@uky.edu)

Grapevine cultivar Vitis vinifera. L. cv. Cabernet Sauvignon is used for winemaking all over the world. Drought and heat stresses are two primary abiotic stresses that reduce grape quality and yield. Drought and heat tolerance are poorly characterized in perennial crops such as grapevine. In this study, RNA-sequencing analysis of leaf tissues of Cabernet Sauvignon plants under drought, heat, and combined stress was carried out. Physiological responses such as the stomatal conductance, stem water potential, and leaf temperature were also measured. Physiological data suggested a significant difference in stomatal conductance, leaf temperature, and stem water potential during stress, where no significant difference was observed during recovery. Data analysis with the criteria of a fold change ≥ 2 and a P_{adj} value < 0.05 resulted in the identification of 533, 112, and 1117 differentially expressed genes (DEGs) for drought, heat, and combined stress, respectively. The combined stress resulted in significantly more differentially expressed genes than individual stress. Gene ontology analysis revealed DEGs associated with epigenetic changes during stress and after stress removal, such as histone modification, protein methylation, and protein alkylation. Altered expression of those epigenetic change-related genes suggests potential establishment of epigenetic memory after stress removal.

Funding Support: Australian grape and wine authorities grants, Grant ID: VA1503

Evaluating Harvest and Postharvest Potential of Fresh-Market Muscadine Grapes Grown in Arkansas

J. Cody Rawls, **Renee Threifall**,* Margaret Worthington, and Luke Howard *University of Arkansas, 2650 N Young Ave, Fayetteville, AR 72704 (rthrelf@uark.edu)

Advances in muscadine grape (*Vitis rotundifolia* Michx.) breeding efforts have resulted in unique traits emerging with commercial, fresh-market potential. Harvest and postharvest attributes of muscadine genotypes (cultivars and breeding selections) grown in Arkansas in 2020 were evaluated at the University of Arkansas System Division of Agriculture (UA System). Two cultivars (Summit and Supreme) and seven UA System selections were hand-harvested from the UA System Fruit Research Station, Clarksville. Approximately 1.8 kg of each genotype were harvested into clamshells and evaluated for physical attributes (berry weight, seed number, stem scar tear, and berry firmness) and composition (soluble solids, pH, titratable acidity, and soluble solids/titratable acidity ratio) at harvest and for postharvest attributes (weight loss and unmarketable berries) after 0, 14, or 28 days at 2°C. Genotype impacted all harvest attributes except seed number (3.11/berry). Berry weights at harvest ranged from 4.76 g (AM 77) to 12.50 g (AM 70), stem scar tear ranged from 0% (AM 26)



to 30.61% (Supreme), and berry firmness ranged from 6.53 N (AM 135) to 10.75 N (Summit). For composition attributes at harvest, Supreme (17.43%) had the highest soluble solids, and AM 195 (3.80) had the highest pH. AM 77 had the highest titratable acidity (1.06%) and lowest soluble solids (11.37%), pH (2.81), and soluble solids/ titratable acidity ratio (10.75). AM 70 had the highest soluble solids/titratable acidity ratio (10.75). AM 70 had the highest soluble solids/titratable acidity ratio (35.17) and lowest titratable acidity (0.43%). There was a significant genotype × storage interaction for both postharvest attributes. During storage, weight loss and unmarketable berries increased. AM 102 had the largest weight loss at 14 days (4.53%) and 28 days (24.56%). The ideal physical and composition attributes and high storage potential demonstrate potential for muscadines as a fresh-market crop.

Funding Support: none

Determining an Effective Concentration of Post-Malolactic SO₂ Addition in Cabernet franc and Cabernet Sauvignon

Kirsty Harmon and Joy Ting*

*Virginia Winemakers Research Exchange, P.O. Box 555, Ivy, VA 22945 (VaWrex@gmail.com)

Despite all that is known about the chemistry of SO₂ in wine, many questions about its practical use remain. This study explored the effects of the concentration and timing of SO₂ additions at the end of malolactic fermentation in Cabernet franc and Cabernet Sauvignon produced in the Monticello AVA of Virginia. In 2018, wine from a single lot of Cabernet franc was racked after completion of malolactic fermentation, then transferred to three sets of barrels (one set of comparable new barrels, two sets of comparable older barrels). One barrel from each set received 30 mg/L SO $_{2}$ ("low dose") while the other received 75 mg/L SO $_2$ ("high dose"). In each pair, the wine receiving the higher dose maintained free SO₂ levels at or near the target of 0.5 mg/L molecular SO₂. Wines given the lower initial dose required many subsequent small additions of SO₂ over five months to reach the same target. The high-dose wines had higher concentration of anthocyanins at the end of five months, with no differences in polymeric anthocyanins or tannins. In year two, the effect of "high" (75 mg/L) and "very high" (100 mg/L) doses of SO₂ were tested in Cabernet Sauvignon. Wine receiving the "very high" dose maintained free SO₂ levels above the 0.5 mg/L target for molecular sulfur throughout aging, with no need for additional doses. Modified sensory analysis by an untrained panel of 23 winemakers found the wines were significantly different in a triangle test, with significantly higher descriptive scores for aromatic intensity in the "very high" dose wine. Adding a larger initial dose of SO₂ better maintains molecular SO₂ levels associated with wine protection than many smaller doses toward the same target.

Funding Support: The Virginia Wine Board

Impact of Prebloom Leaf Removal on Winegrape Production and Quality Parameters – A Systematic Review and Meta-Analysis

Josh Vander Weide and Paolo Sabbatini*

*Michigan State University, 426 Auditorium Road, East Lansing, MI 48824 (sabbatin@msu.edu)

Winegrape (*Vitis vinifera* L.) is the most widely cultivated fruit crop in the world. However, climate characteristics in some growing regions are suboptimal for grape production, including short season length and excess precipitation. Grapegrowers can

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utilize an array of methods to mitigate these issues, including "early leaf removal," a management practice involving removal of leaves from selected basal nodes along shoots around bloom. This meta-analysis reviews the extensive literature on this practice, with specific regard to application at "prebloom" (PB). One hundred seventy-five publications on the topic of "early leaf removal" were identified using key terms and subsequently narrowed via eight data curation steps. The comparison between treated (PB) and control plants in these studies revealed two important results. First, PB lowered bunch rot disease (-61%), partially through reducing the compactness of clusters. Second, PB promoted a significant increase in fruit total soluble solids (Brix, +5.2%), which was related to the increase in leaf-to-fruit ratio. Furthermore, cultivar and rootstock were found to have a large influence on the success of PB, while the contribution of climate was smaller. PB significantly lowers yield and bunch rot disease and increases Brix, both of which improve grape and wine quality.

Funding Support: AgBio-Research at Michigan State University (Project GREEEN) and the Michigan Craft Beverage Council

Remote Sensing-Assisted Scouting of Virus Infections in Vineyards

Kaylah Vasquez, Marc Fuchs, Monica Cooper, and Luca Brillante* *Department of Viticulture and Enology, California State University Fresno, 2360 E Barstow Ave, Fresno, CA 93740 (lucabrillante@csufresno.edu)

The cost of virus infections per acre in vineyards with an average lifespan ranges from \$10,000 to \$16,000. With 880,000 acres, California has ~80% of the total vineyard acreage in the U.S. Early detection and eradication of infected plants are crucial to managing pathogen spread, as there is no cure for viruses in the field. Chemical control of insect vectors of viruses is of limited efficacy and often not an option in organic agriculture. Supported by preliminary data, our project used remote sensing coupled to machine learning to detect major grapevine diseases (red blotch and leafroll) more efficiently than traditional methods. In 2020, we monitored grapevine infections in two vineyards located in Madera and Rutherford, planted with Cabernet Sauvignon and Cabernet franc, respectively. Around 400 plants in each vineyard were imaged twice with a VIS-NIR hyperspectral, thermal, and high-resolution RGB camera mounted on a tripod, an ATV, or a UAV. Leaves were collected for further imaging in a dark room in the laboratory and then used for molecular analyses to assess virus presence. All plants in the vineyard blocks were also scouted to assess the presence of the infection and compared to high-resolution satellite imagery. The imagery was used to train machine-learning algorithms to predict grapevine infection, which show promising performances and will be further optimized in the second season of measurements.

Funding Support: CDFA Specialty Crop Block Grant Program

Exploration of Oxidation Reduction Potential (ORP) as a Process Parameter to Track Pinot noir Fermentations

Gordon Walker, James Nelson, Desmon Hernandez, Thomas Halligan, Andre Knoesen, and Ron Runnebaum*

*UC Davis, 1 Shields Avenue, Davis, CA 95616 (rcrunnebaum@ucdavis.edu)

As part of a larger effort to characterize site-specific contributions in Pinot noir wines, four sites were selected to examine the relationship between vineyard, must composition, and fermentation dynamics. These vineyards were selected for their replicable fermentation kinetics and consistent organoleptic properties. The wines were pro-



duced from the same clone of Pinot noir and were made using the same enological process. To gain insight into real-time dynamics of fermentation, oxidation reduction potential (ORP) probes were used to measure redox potential during alcoholic fermentation. Redox potential determines the type and frequency of chemical half-reactions in solution, including speciation and reactivity of metals. ORP is a quantifiable metric that responds to changes in pH, temperature, dissolved oxygen, enzymatic activity, and yeast metabolism in real time. ORP probes are standard for anaerobic fermentation industries but are not widely used for wine. For a comprehensive view of the characteristics of these wines, phenolics extraction and elemental composition were also analyzed. The vineyards were selected to represent faster- and slower-fermenting musts. While vintage variation exists, these sites have displayed robust trends over five years of vinification. The working hypothesis was that placing ORP probes into the replicate fermentations would provide insight into yeast metabolic activity and determine differences in kinetics. The observed fermentation profiles matched previous vintages, consistent with our expectations. We also observed typical differences in phenolics, anthocyanin extraction, and the elemental profile among wines (specifically the Fe:Mn ratio). The ORP profiles followed similar trends for all fermentations, a sign that the temperature-based process control worked effectively. By examining the ORP profiles in detail, deviations in ORP revealed previously undetectable differences between sites and replicates. ORP is exquisitely responsive to small perturbations, making it ideal to use a process parameter for monitoring and controlling fermentations.

Funding Support: Jackson Family Wines, Silver Oak Cellars, WineXRay

Valorization of Cold-Hardy Grape Pomace Using an Infrared Drying Method

Zeinab Mohammadi Shad, Emily Kuelbs, Chandrasekar Venkitasamy, Lucas Buren, and **Aude Watrelot***

*Department of Food Science and Human Nutrition, Iowa State University, 536 Farm House Lane, 2567 Food Sciences Building, Ames, IA 50011 (watrelot@iastate.edu)

The winemaking process produces ~20% of the weight of the fresh grapes as grape pomace, a solid by-product composed of skins, seeds and some part of stems and rice husks. This by-product is rarely re-used in winemaking in spite of its polyphenol content and biological properties. The goal of this study was to optimize a sustainable drying method for cold-hardy grape pomace to make a microbiologically safe powder that contains polyphenols with antioxidant properties. Two drying methods, hot air (HA) and catalytic infrared (IR) heating using three temperatures, were applied to Edelweiss and Marquette pomaces. The moisture content of the pomaces was evaluated prior to chemical analysis. The color was evaluated using a chromameter (Hunter Lab ColorFlex EZ), the phenolic compounds were extracted using 70% acidified acetone, and the total iron-reactive phenolic compounds content in the extracts was measured. The initial moisture content of grape pomace ranged from 50 to 70% (wet basis) and was reduced to a moisture content of <10% by IR heating for 30 min, while HA drying took ~3 hr. Edelweiss white pomace dried by IR was significantly darker, with lower L* and b* values than when dried by HA. The color of Marquette pomaces was not affected by drying method. Between 10 to 13% of polyphenols were extracted from the dry and ground pomaces. No effect of drying method was observed on the phenolic compounds and condensed tannin content of Edelweiss pomace extracts. The antioxidant activity of pomace extracts will be measured using the DPPH free radical assay to determine the effect of IR drying on the biological properties of grape pomace and to valorize this by-product.

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Funding Support: USDA-AMS lowa Department of Agriculture and Land Stewardship Specialty Crop Block Grant Program

Effects of B-Glucosidase Additions Prior to Fining with Activated Carbon in Smoke Taint Remediation

Brian Wayne* and Lauren Blomberg *Tastry, 3450 Broad St., Suite 101, San Luis Obispo, CA 93401 (brian@tastry.com)

Activated carbon fining products can decrease the concentration of free smoke-taint markers in smoke-affected wines. Treating with ß-glucosidase enzymes prior to the addition of activated carbon has been proposed as a method to increase treatment efficacy. However, the effect of enzyme treatments prior to activated carbon fining has not been fully evaluated. A large sample of 2020 vintage wines that had been exposed to smoke taint were treated with a combination of ß-glucosidase enzymes (LAFAZYM AROM) and an activated carbon product (OENOVEGAN EXTRA). Smoke taint markers and major volatile aroma compounds were measured by gas chromatography-mass spectrometry. Color and total tannin were analyzed using UV-Vis. Sensory analysis was performed using a trained panel. A moderate reduction in smoke taint markers was observed with the activated carbon product. B-Glucosidase enzymes significantly increased the concentrations of free smoke taint markers. The combination of treatments, however, did not result in a net decrease of free smoke taint markers. The activated carbon product significantly reduced concentrations of ethyl esters. The ß-glucosidase enzyme significantly increased the concentration of some aroma compounds. Color was reduced by both treatment products. Total tannin was reduced by the activated carbon product.

Funding Support: Tastry

Relationships of Brix, Titratable Acidity, and pH with Time during Ripening above and below Temperature Thresholds

Jeremy Weiss* and Jesse Noble

*University of Arizona Cooperative Extension, 1064 East Lowell Street, Tucson, AZ 85721 (jlweiss@arizona.edu)

As temperatures continue to trend higher, winegrape growers and winemakers in warm-climate viticultural regions will increasingly be challenged by the many effects temperature has on fruit composition. Warmer ripening conditions in recent years for such growing regions around the world have resulted in higher sugar concentrations, less acidity from declines in malic acid, higher pH, and changes in aromatic and phenolic compounds, all of which can be particularly affected by daytime maximum or nighttime minimum temperatures. Information that helps identify and minimize these impacts and aids production of well-balanced wines in these regions is needed. Here, we evaluate fruit composition at harvest in the context of time spent during ripening above and below temperature thresholds to test our hypothesis that duration and degree of heat and cold exposure correlate to composition values. For analysis, we use measures of Brix, titratable acidity, and pH for several varieties across multiple vintages from a warm-climate vineyard in southeastern Arizona and accumulated hours of temperatures greater than values between 30 and 40°C (86 to 104°F) and less than values between 10 and 20°C (50 to 68°F), ranges encompassing thresholds reported in previous studies. We calculate temperature accumulations over the 30 days leading up to harvest, basing them on data collected at a meteorological station located approximately one kilometer (0.6 miles) from the vineyard. By determining how exposure to heat and cold during ripening potentially has influenced fruit



composition in recent vintages, our work provides an initial model for growers and winemakers in this and other warm-climate viticultural regions with which to predict impacts of temperature conditions in future growing seasons and to consider vineyard and winery adaptations for production of quality fruit and balanced wines under continued regional warming.

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Investigation of the Absorption Mechanism of Smoke Taint Markers onto Winegrapes

Yan Wen, Cristina Medina Plaza, Andrew Waterhouse, and Anita Oberholster* *UC Davis, One Shields Avenue, Davis, CA 95616 (aoberholster@ucdavis.edu)

The increased frequency of wildfires on the West Coast of the United States are seen as a significant risk for the grape and wine industry. High levels of smoke-related compounds can accumulate in winegrapes, which can result in smoke-impacted wines. Among hundreds of volatile compounds reported in wildfire smoke, volatile phenols and their corresponding precursors are widely considered the main markers of smoke taint. However, understanding of the absorption mechanism of smoke-taint markers onto grapes is limited, making it challenging to propose strategies to mitigate or prevent smoke impact. Another challenge is measuring smoke-taint markers in the air generated by wildfires, and how that relates to the amount absorbed by grapes. In this study, different strategies were applied to trace absorption of volatile phenols (VFs) from air into grapes. A well-sealed system containing known concentrations of vaporized VFs was designed to investigate their absorption efficiency onto grape clusters. Experimental factors were optimized to reach sufficient sensitivity and reproducibility, including the headspace sampling conditions, equilibrium of VFs in the vapor phase, and adsorption time. After VFs exposure (absorption), grape berries were extracted to determine the concentrations of all selected markers using a proposed LLE-GC-MS method. The glucoside precursors of VFs were analyzed using a well-developed SPE-UHPLC-QTOF-MS method. Isotopic standards were used as indicators during the whole process. The results show the transfer potential of smoketaint markers from air to grapes and their glycosylation, expanding our knowledge of the formation of smoke-taint flavor. This controlled system is a first step toward understanding the absorption process that will lead to developing preventative strategies during wildfire events. In the next steps, different grape barrier sprays will be evaluated in this contained atmospheric system. Those applications that prove successful will be included in a larger field study.

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Volatile Phenols in Oregon Wines as Indicators of Smoke Exposure during the 2020 Wildfire Season

Ruiwen Yang, Yanping L. Qian, Armando Alcazar, and Michael C. Qian^{*} *Oregon State University, 100 Wiegand Hall, Corvallis OR, Corvallis, OR 97330 (michael.qian@oregonstate.edu)

Climate change has increased the incidence of forest and bushfires in the western United States. When grapevines are exposed to wildfire smoke, the grapes and grapevine can absorb smoke compounds and convert them into the corresponding glycosides or other bound forms. The amount of smoke compounds absorbed by

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the grapes depend on smoke intensity, smoke duration, grape variety, and many other factors. Guaiacol, 4-methylguaiacol, syringol, 4-methylsyringol, and o-, m-, and p-cresols have been suggested as indicators for smoke exposure. A rapid method based on solid-phase microextraction gas chromatography-mass spectrometry with isotope compounds as internal standards was developed to analyze guaiacol, 4-methylguaiacol, 4-ethylguaiacol, o-cresol, m-cresol, p-cresol, and 4-ethylphenol in 30 min. Using this rapid analytical method, over 370 smoke-exposed red wines and 90 smoke-exposed white wines (including Rosé wine) were analyzed during the 2020 wildfire season. Total phenols (after acid hydrolysis, pH 1, 100 C/4 h) were also analyzed for all samples. Out of 370 smoke-exposed red wines, 84 samples had guaiacol concentrations of 0 to 5 μ g/L and 159 samples had guaiacol concentrations of 5 to 10 μ g/L. Most samples had less than 3 μ g/L 4-methylguiacol. In smoke-exposed red wine, the average ratio of free guiacol to 4-methylguaiacol was 4.5. However, the average ratio of total guaiacol to total 4-methylguaiacol was 6.5, higher than the free forms. Except for m-cresol, levels of other total volatile phenols were approximately two to five times greater than the corresponding free volatile phenols. Distribution frequency was used to investigate the relationship between these smoke-related compounds. The highest frequency of total guaiacol/free-form guaiacol ratio was 2:4; the average was 3.3:1. The highest frequency of guaiacol/4-methylguaiacol ratio in free-form and total was 4:6, the average for free-form was 4.5:1, and the total was 6.6:1.

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Extraction, Purification, and Characterization of Polysaccharides from Pinot noir Grape

Danye Zhu, Armando Alcazar Magana, Yongsheng Tao, and Michael C. Qian* *Oregon State University, 100 Wiegand Hall, Corvallis, OR 97331 (michael.qian@oregonstate.edu)

Mouthfeel is an important quality attribute of red wine. It is postulated that polysaccharides from grapes and yeasts could play an important role in wine mouthfeel. Understanding the structural characterization of polysaccharides from different sources is extremely important for wine organoleptic quality. In this study, Pinot noir grapes were broken and then extracted with hot water (90°C, 2 hr) twice to extract free sugar and highly water-soluble polysaccharide (named G1). The residues were then extracted with alkali solution (0.5 M sodium hydroxide solution with 10 mM NaBH₄, 80°C, 2 hr) (named G2). The obtained supernatants were neutralized, concentrated, and centrifuged, then absolute alcohol was added to precipitate polysaccharide overnight. Ultraviolet scanning showed a unique absorption peak between 190 and 200 nm, indicating that the extracts were polysaccharides. The grape polysaccharides were methanolized by 0.5 M HCI/MeOH at 85°C for 18 hr and derivatized by 1-(trimethylsilyl)imidazole (TMSI) for gas chromatography-mass spectrometry (GC-MS) analysis. Glucose, xylose, mannose, arabinose, galactose, rhamnose, fucose, glucuronic acid, and galacturonic acid were methanolized and silylated to confirm sugar identification. Methyl-alpha- and methyl-beta-galactopyranoside and methyl-alpha-mannopyranoside were used to verify sugar anomer distribution. GC-MS analysis revealed that G1 was composed mainly of glucose, arabinose, rhamnose, galactose, mannose, galacturonic acid, and xylose. G2 was composed primarily of arabinose, galactose, glucose, rhamnose, galacturonic acid, xylose, and mannose.

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Physicochemical and Structural Characterization of Polysaccharides from *Saccharomyces cerevisiae*

Danye Zhu, Armando Alcazar Magana, Yongsheng Tao, and Michael. C. Qian* *Oregon State University, 100 Wiegand Hall, Corvallis, OR 97331 (michael.qian@oregonstate.edu)

Polysaccharides are the main macromolecule components in wine. Polysaccharides are of interest because they can interact with wine aroma and flavor compounds to affect sensory quality. The primary polysaccharides in wine are derived from yeasts and grapes. Understanding the structural characterization of polysaccharides from different sources is extremely important for wine organoleptic quality. In this study, the sugar composition, molecular weight, and other physicochemical properties of polysaccharides were characterized systematically. Polysaccharides from Saccharomyces cerevisiae (RC 212) were sequentially extracted with hot water (20% yeast cells aqueous solution, 121°C, 3.5 hr) and alkali (3% sodium hydroxide solution, 80°C, 6 hr). named Y1 and Y2. The mixtures were centrifuged and the precipitates were extracted again. The obtained supernatants were neutralized, concentrated, and centrifuged again to remove any precipitates. Absolute alcohol was added to the supernatants overnight to precipitate polysaccharide. The polysaccharides were harvested and dried under vacuum. Sugar purity was analyzed. The polysaccharides were measured using a phenol-sulfuric acid assay and the results showed high purity for isolated yeast polysaccharides. Size exclusion liquid chromatography was performed using an Agilent 1100 liquid chromatography coupled to a RID detector operated in positive polarity. Separation was performed at 30°C in a Superde 200 increase 10/300 (10mm × 300 mm) GL column. Certified polysaccharides standards (1, 5, 25, 50, 150, 270, 410, and 670 KDa) were used for molecular weight calculation. The sugar composition of polysaccharides was analyzed by gas chromatography-mass spectrometry after metholysis (0.5 M HCI/MeOH at 85°C for 18 hr) and silylation 1-(trimethylsilyl) imidazole (TMSI). Structural characterization results showed polysaccharides vary greatly in composition. Y1 was composed mainly of mannose, glucose, and xylose; Y2 was composed mainly of mannose and glucose.

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Impact of Mechanical Leafing on Yield and Berry Composition of Ruby Cabernet Grown in the San Joaquin Valley of California

Shijian Zhuang,* Qun Sun, Matthew Fidelibus, and Kaan Kurtural *University of California, 550 E Shaw Ave, Suite 210-B, Fresno, CA 93710 (gzhuang@ucanr.edu)

Ruby Cabernet is a cross between Cabernet Sauvignon and Carignan that was bred by Dr. Olmo to produce wine with more color and tannin in the arid, warm climate of the San Joaquin Valley. Local growers and wineries use this variety primarily as a blender to improve color and tannin structure of other premium red varieties like Cabernet Sauvignon or Merlot. High berry anthocyanins and tannin and high production per acre are desired for producing Ruby Cabernet. A randomized complete block design was implemented in a commercial vineyard located on the western side of Fresno County in 2018 and 2019. Quadrilateral cordon-trained, spur-pruned Ruby Cabernet vines grafted on Freedom rootstock were used under a sprawl system. Four timings of mechanical leafing, replicated five times, were used in this study, with five vines designated as an experimental unit. Water deficit was kept at 80% ET_c (targeted Ψ of -1.2 MPa) from berry set to veraison and decreased to 60% ET_c (targeted Ψ of -1.4

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MPa) from veraison to harvest. Four timings of mechanical leafing included bloom, berry set, veraison, and no leafing. Midday leaf water potential (Ψ) and fruit-zone photosynthetically active radiation were measured during the season. Leafing reduced yield by 8% in 2018 and by 11% in 2019. Leafing had minimal effect on berry primary metabolites and leafing at bloom, and berry set improved berry anthocyanins by 16% in 2018 and 5% in 2019, with no benefit of leafing at veraison.

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No-Till Systems and Permanent Cover Cropping Mitigate Plant-Available Water in Vineyards

Maria Zumkeller, Nazareth Torres, Runze Yu, and Kaan Kurtural* *University of California Davis, 1 Shields Avenue, Davis, CA 95616 (skkurtural@ucdavis.edu)

Anthropogenic climate change has left no viticulture region untouched. The San Joaquin Valley of California is the leading irrigated viticulture region in the world. There is little information on how best to preserve plant-available soil water before in-season irrigation is initiated. We studied a grass (*Poa bulbosa* hybrid), barley (*Hordeum* spp.), and native vegetation as cover crops under till versus no-till system in a Ruby Cabernet/Freedom (*Vitis* spp.) vineyard. Our results indicated that grass under no-till preserved plant-available water, resulting in 30% greater midday stem water potential in grapevine. The net carbon assimilation of grass under no-till was also greater than barley and natural vegetation until cover crops went dormant or were terminated. There were no adverse effects of cover cropping or tillage in components of yield or primary or secondary metabolism of grapevine. Under the no-till system, net carbon assimilation of vineyard system was greatest with grass species. Our results will provide a greater understanding of dynamics of the soil-grapevine-atmosphere continuum in the greater San Joaquin Valley under anthropogenic climate change.

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