UNDERSTANDING THE IMPACT OF CLIMATE CHANGE ON ANTHOCYANIN CONCENTRATIONS IN NAPA VALLEY CABERNET SAUVIGNON

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Abstract:

Context and purpose of the study

Climate change is having a significant impact on the wine industry through more regular drought conditions, fires, and heat events, leading to crop loss. Furthermore, these events can reduce overall quality of the fruit, even when crop yields are not impacted. Anthocyanins are considered one of the most important classes of compounds for red wine production and are known to be sensitive to vine water status and heat events. Therefore, they are likely to be impacted by environmental conditions across broad spatial scales. In order to evaluate this, a large-scale project was undertaken in 2015 to look at the differences in berry secondary metabolite chemistry across sub-appellations within the Napa Valley, where there is an exceptional amount of climatic diversity. This study was expanded in the 2021 and 2022 growing seasons, to evaluate impacts of agronomic practices and environmental conditions on crop yields and berry chemistry. The goal of the project is to evaluate the impact of intra- and inter-annual variation in climate on secondary metabolites in Cabernet Sauvignon across a climatically diverse and economically important wine growing region in California; with the aim of increasing the industry's awareness of potential climate change impacts on important grape secondary metabolites.

Material and methods

In 2015, 66 Cabernet Sauvignon commercial blocks were monitored across 10 sub-appellations within Napa Valley. In 2021 and 2022, 50 commercial blocks were monitored across 13 sub-appellations. Berries were collected from veraison until commercial harvest, with each site having 120 berries being collected every 14 days, totaling 3 to 5 sampling dates per site depending on harvest timing. Berry skins were separated from pulp and seed to extract the phenolics in skins. Extractions were conducted using 2:1 acetone:water (v/v) for 24 hours. Acetone was removed under reduced pressure and samples were then analyzed for high and low molecular weight polyphenols by RP-HPLC. Weather stations collecting hourly temperature, precipitation, humidity and radiation data were present in all blocks, and irrigation records, phenology data, and agronomic practices were collated for all sites.

Results

Across all years, trends indicate that warmer sub-appellations had lower overall anthocyanin concentrations when compared to cooler sites, yet there was a substantial amount of variation driven by other factors. Although work has been done within the context of greenhouses looking into the impact of heat on anthocyanin degradation, this work provides a large-scale analysis of the impacts of climate on anthocyanin production across the season in Cabernet Sauvignon.

Keywords: Polyphenols, Flavonoids, Anthocyanins, Climate Change, Winkler Index

1. Introduction

Farming of any kind is reliant on long term climate stability. Current projections on climate change appear to be shaking the foundation of this stability in a way that may make grape growing, as we know it, difficult in areas of the world known for high quality wine. The Intergovernmental Panel on Climate Change (IPCC) reports that by the end of the century California will likely see rises in temperature of 2.0 °C or more (Cubasch et al. 2001). Although some cooler growing regions may benefit from a decrease in frost events and potential to plant warmer-adapted cultivars with higher quality, most planted areas will likely need to endure warmer growing conditions with more extreme climate events. The projected rise in temperatures, in conjunction with greater incidence of heat extremes, may have devastating and unforeseen consequences on grape and wine quality attributes.

Anthocyanins are considered a cornerstone of red wine quality, as they not only provide the red pigment, but also interact with other compounds in the red wine matrix, leading to changes in tannin and sensory characteristics. Work over the last 15 years has shown that anthocyanins are susceptible to degradation at elevated temperatures, leading to the loss of color through the breaking of covalent bonds in the flavonoid (Hiemori, Koh, and Mitchell 2009). Moreover, it has been shown that transcriptionally, grapevines grown in higher temperatures have lower overall biosynthesis of anthocyanins, leading to a net decrease in concentration at harvest (Yan et al. 2020). Combined with increases in peroxidase activity at increased temperatures, these results suggest that climate change may have a detrimental impact on red wine quality in the future (Movahed et al. 2016).

Recently, work conducted in Australia has shown a particularly grim outlook for anthocyanin concentrations in Cabernet Sauvignon grown in Coonawarra (Barnuud et al. 2014). Their results suggest a possible loss of 30% or more of color in that wine production region over the next 40 years due to increases in temperature globally.

The work presented here is a survey of multiple appellations (10+) across Napa Valley which represent a wide range of climatic regions, both cool and warm (**Figure 1**). The goal of this work is to provide an understanding of the impact that Napa Valley growers may face in the future within the context of climate change. The hope is that this will help growers understand the possible problems they may face in the future and develop ways to mitigate the effects of climate change on anthocyanin degradation.

2. Materials and Methods

Berry Sampling, Extraction, and Analysis

Instrumentation and Chemicals

Agilent models 1260 (Santa Clara, CA.) HPLC systems were used to conduct analyses on grape extracts. Chemstation software was used for all chromatographic analysis. All solvents were HPLC grade. Acetonitrile, acetone, methyl cellulose and trifluoroacetic acid were all purchased from VWR (Radnor, PA)., malvidin-3-O-glucoside (≥95% purity by HPLC) was also purchased from Sigma-Aldrich (Extrasynthese, Genay, France) and was used as a quantitative standard.

Berry Sampling

Prior to veraison an area of 100 vines was flagged for berry sampling per block. Both sides of the canopy were sampled as well as the top, middle, bottom and back of sampled clusters. In 2015, 66 commercial blocks were monitored across 10 sub-appellations within Napa Valley. In 2021 and 2022, 50 commercial blocks were monitored across 13 sub-appellations (**Figure 2**). Berries were collected from veraison until commercial harvest, with each site having 120 berries being collected approximately every 14 days, totaling 3 to 5 sampling dates per site depending on harvest timing. All samples were collected in the morning before 10am and the samples were kept at 4 °C before being separated at the University of California, Davis. Berry samples from each site were split into 3 replicates and placed in a -20 °C freezer until extraction.

Extraction

Berry skins were separated from pulp and seed to extract the phenolics in skins. Extractions were conducted using 2:1 Acetone:water (v/v) for 24 hours. Acetone was removed under reduced pressure; all samples were brought back to 50 mL with distilled water and then placed in -20 °C for future analysis.

Primary Chemistry

In both 2015 and 2021 primary chemistry (Brix, pH, TA, malate, glucose and fructose) were analyzed by ETS Laboratory (St. Helena, CA). In 2022 all primary chemistry was analyzed at University of California, Davis. Briefly, a subset of berries was sampled specifically for primary chemistry analysis. These berries were crushed fresh following sampling, the juice was separated from skin and seed, and then it was centrifuged at 4500 × g for 5 mins (5810R, Eppendorf, Hamburg, Germany). Samples were then decanted into another centrifuge tube and analyzed.

HPLC Analysis

Anthocyanin analysis was conducted using a previously published method with modifications using an Agilent 1260 (Santa Clara, CA) with a diode array detector (DAD) (Campbell et al. 2021)**.** Briefly, a Phenomenex Kinetex PFP 150 × 3.0 mm column was used with a particle size of 2.6 µm. Samples were injected at a volume of 10 µL with a column temperature of 50 °C. Mobile phase A consisted of milli-q water with 0.2% v/v TFA; mobile phase B consisted of acetonitrile with 0.2% v/v TFA. Eluting peaks were monitored at 520 nm. Elution conditions were 0.5 mL/min; 12.5% of solvent B for the first 3 min; a linear gradient from 12.5% to 20% from 3 to 14 min; 20% to 27.3% from 14 to 26 min; 27.3% to 70% from 26 to 26.02 min; held at 70% for 2 min; 70% to 12.5% from 28 to 28.02 min. The column was then washed and re-equilibrated for 4 min before the next injection. Anthocyanins were quantified using a malvidin-3-O-glucoside standard curve (Extrasynthese, Genay, France).

Statistics and Graphics

Integration of HPLC data was conducted in R, using the ChromatographR package (Bass n.d.). Visualization of the data was conducted using ggplot2 (Wickham et al. 2019). Maps of the vineyard locations were made using ArcGIS Pro (ESRI 2023. ArcGIS Pro 3.1. Redlands, CA: Environmental Systems Research Institute).

3. <u>**Results and Discussion**</u>

The initial findings from both 2015 and 2021 suggest that the warmer appellations appear to have lower concentrations of anthocyanins (**Figure 2**). This is particularly evident when examining results from 2021 **(Figure 3b)**. Pope Valley is the warmest area in our experiment based on the growing degree-days calculated from PRISM data over a 30-year span (**Figure 1)**. Moreover, the highest anthocyanin concentrations seen in both 2015 and 2021 came from sites in the coolest regions seen on the map – the southern parts of Napa Valley and the Vaca Mountains.

There is a large amount of anthocyanin concentration variation across locations within the Napa Valley. In 2015 there was a 4-fold difference between the highest and lowest anthocyanin concentrations seen in berries, with the highest value coming from Atlas Peak (1.11 mg/b), which is one of the coolest appellations Napa Valley, and the lowest coming from St. Helena (0.211 mg/b), one of the warmest appellations in Napa Valley. Similar results were seen in 2021 where the highest anthocyanin concentrations per berry were seen on Pritchard Hill (1.21 mg/b) and in the Vaca Mountains, while the lowest values were seen in Pope Valley (0.223 mg/b). This represents a large difference in observed concentration from regions that are within *hundreds* of growing degree days of each other. Although these show that warmer climates negatively affect the production of anthocyanins, there is a large amount of variation within appellations, suggesting that management can mitigate some of the environmental effects seen in this study.

While the project is ongoing, the current data suggests that an expansion of the growing degree model commonly used in the United States (Winkler Index) is required in the context of these results. Although there are minor differences between Pope Valley and Calistoga (<200 GDD), and both are considered Region IV, there is a significant impact on the ability to synthesize anthocyanins and prevent degradation due to small differences in thermal time. Lastly, these small differences in thermal time having such a substantial impact on the formation of anthocyanin production provides a relatively stark picture of red wine production in the context of climate change. Given an expected increase in global temperatures of 2 °C over the next century, representing an increase of 400 GDD or more, it is likely that we will continue to see a downward decline in anthocyanin production in some of the warmer regions where Cabernet Sauvignon is grown, which makes critical the serious consideration of altering growing practices and/or mitigation strategies, planting warmer-adapted cultivars, and potential movement towards regions that were once unsuitable for producing high quality Cabernet Sauvignon (Jones 2021).

4. **Conclusions**

The current survey across various climates suggests significant differences in the synthesis, and degradation, of anthocyanins in Cabernet Sauvignon. Based on previous work in Australia, this will likely be magnified in the future due to impacts of climate change on global temperatures. Moreover, extreme heat events near harvest, such as the heatwaves in 2015 and 2022, can cause considerable decline in anthocyanin concentrations due to high temperatures. This should be an area of more focus, with the aim of preserving anthocyanins produced during other phases of ripening.

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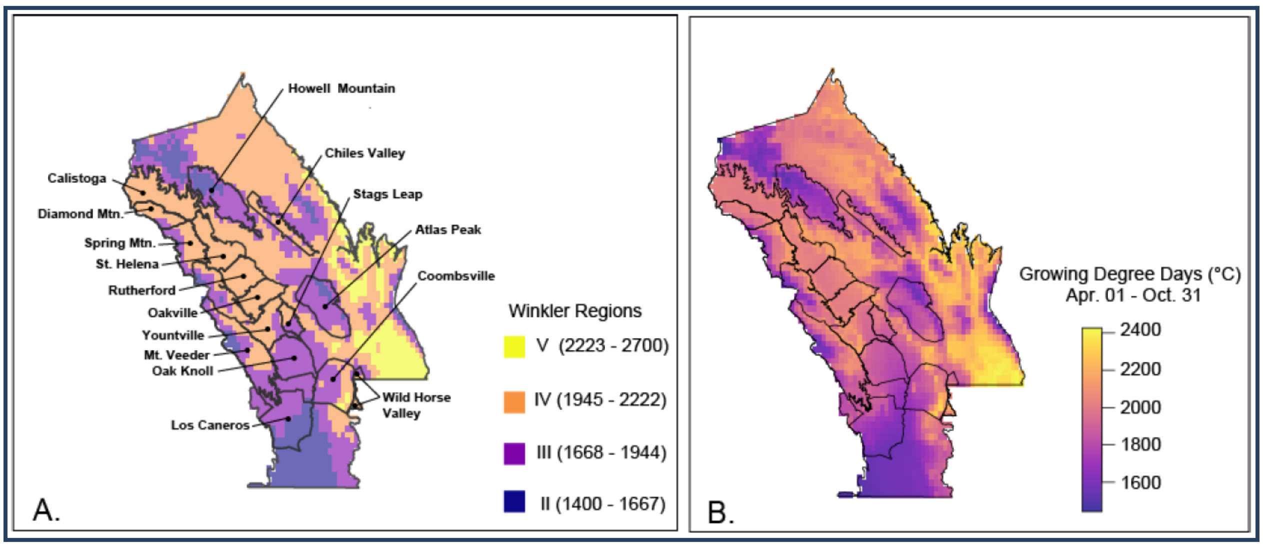
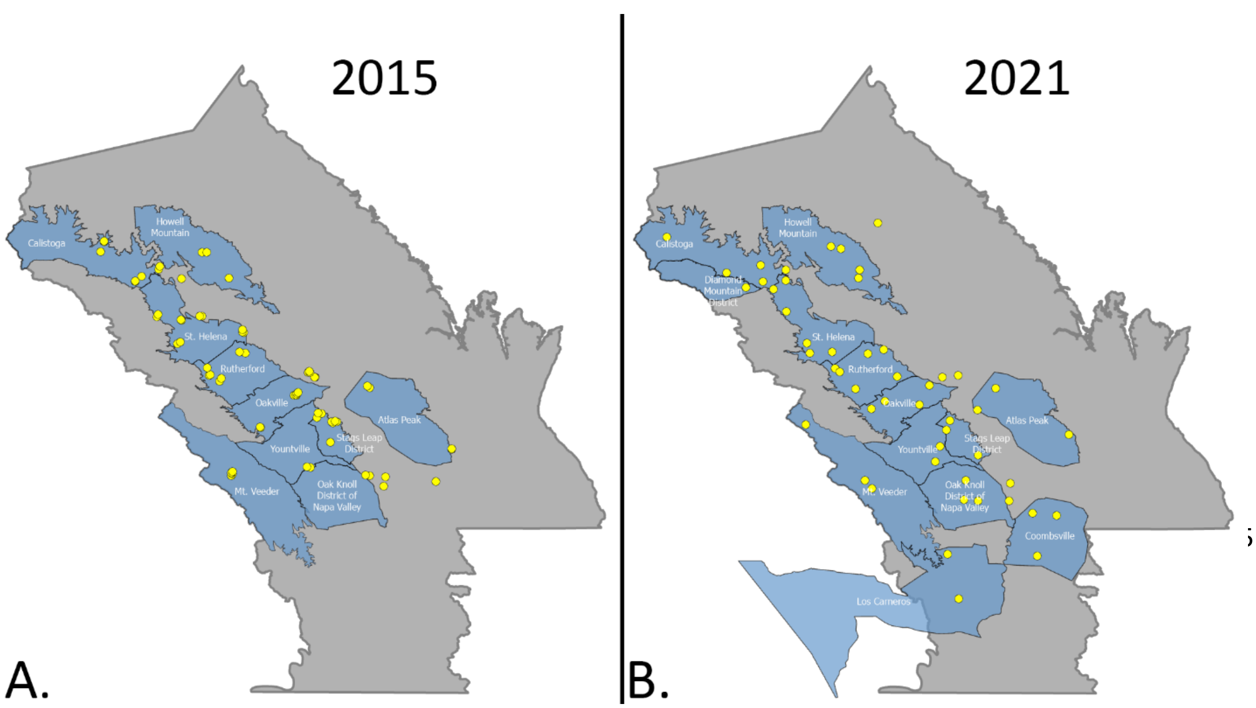


Figure 1: (A) Winkler Regions for American Viticultural Areas (AVA) within the Napa Valley and associated range of growing degree days, and (B) growing degree days above a base temperature of 10° C between April 1 and October 31st calculated according to Winkler & Amerine 1944. Degree days were calculated from the 1980-2010 climate normals at 800-meter grid cell resolution from the PRISM downscaled climate dataset.



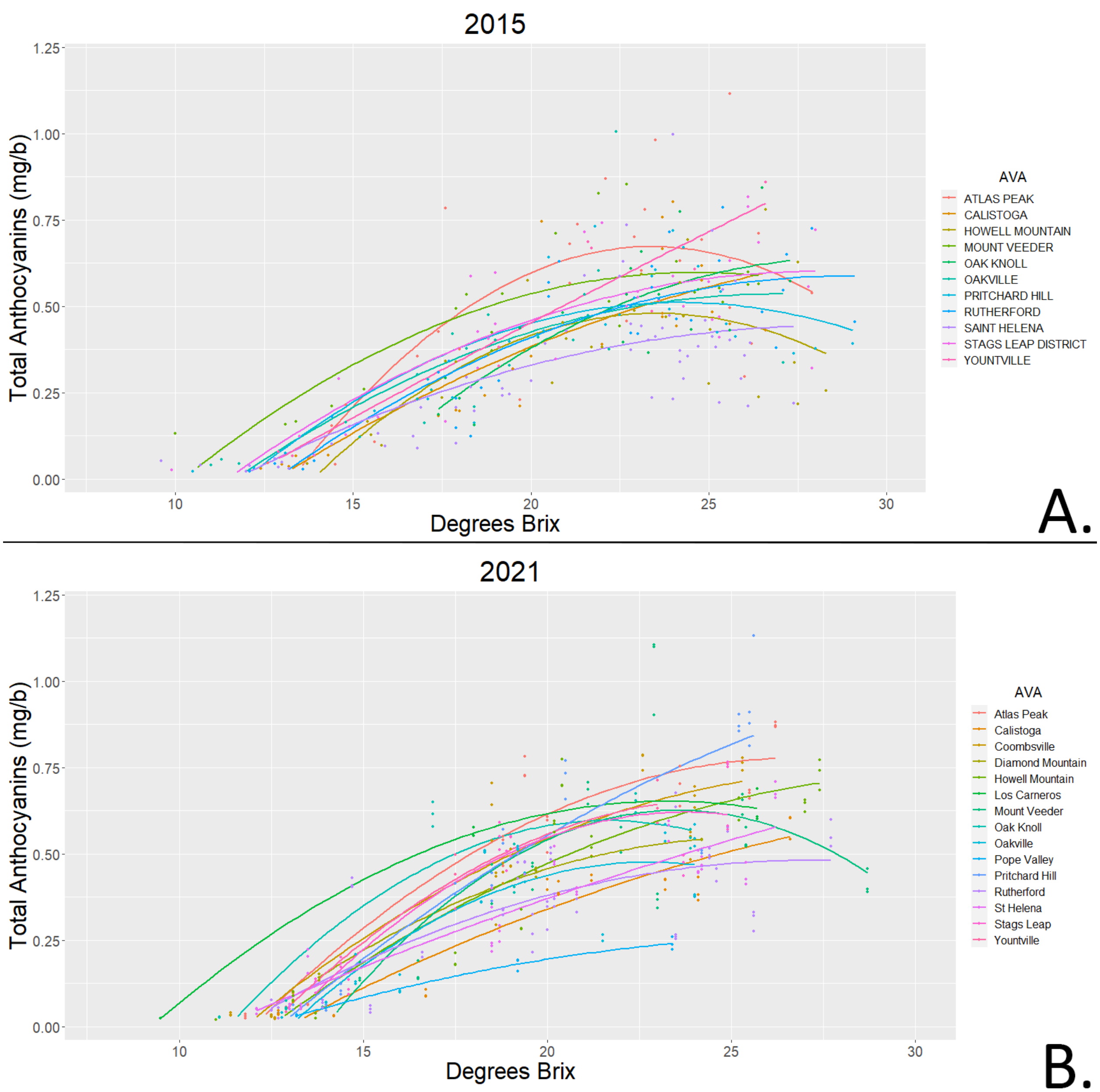


Figure 3: (A) Anthocyanin data and polynomial curves for 66 Cabernet Sauvignon blocks in 2015, representing 10 appellations and (B) Anthocyanin data and polynomial curves for 50 Cabernet Sauvignon blocks in 2021, representing 13 appellations. In both the plots there is a trend toward cooler sub-appellations having higher anthocyanin concentrations at, or near, commercial harvest.