Production of Free Sulfur Dioxide by Wine Yeasts

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The investigation of free sulfur dioxide (SO_2) produced endogenously during primary fermentation, by differing yeast strains within red and white wines, was performed due to minimal information presented by the yeast manufacturers. A total of 10 yeast strains were tested from two brands, producing 20 wines. Initial SO_2 concentrations, yeast, and yeast nutrient levels were controlled in a stable fermentation environment. The hypotheses were as follows: differing yeast strains produce variable-free SO_2 concentrations, and white wines will contain higher concentrations of free SO_2 as opposed to red wines.The wines produced were analyzed for free SO_2 concentrations, pH and residual sugars. One yeast strain, Red Star Côte des Blancs, was found to produce free SO_2 as well as residual sugars in both wine types. It was also found that endogenous free SO_2 contributions are inadequate for long-term wine protection and appropriate SO_2 levels should be maintained utilizing exogenous SO_2.

INTRODUCTION

Sulfites (SO_2) are known in the winemaking industry for their anti-microbial and anti-oxidant properties. The three sulfite forms in wine are sulfur dioxide (SO_2), bisulfite ion (HSO_3^-), and sulfite ion (SO_3^{2-}), which collectively make up the term sulfite. Sulfites are not naturally occurring in freshly picked fruits, but can be added to wine musts (fruit juice which contains the pomace, e.g. skins, seeds, stems) exogenously as one of two forms: potassium metabisulfite or sodium metabisulfite. In addition, sulfites are produced endogenously in by the fermenting yeasts as a byproduct of the alcoholic fermentation process.[10] Sulfites are known as potential allergens, affecting persons with asthma the greatest.[8] Given the potential sensitivity to sulfites, exogenous sulfite additions to wine have been highly scrutinized worldwide. Under United States law (27 CFR 4.22. 21 CFR 182.3637 (GRAS)), [9] wines containing over 10ppm SO_2 must clarify on the label that the wine contains sulfites.[9] Although organic wines have no exogenous sulfite form added, they do contain traces of sulfites produced endogenously by the fermenting yeasts.[1,10]

Sulfur dioxide (SO_2) is a gas at room temperature and readily dissolves in liquids. Once dissolved into the aqueous form, sulfur dioxide becomes an ion whose form is driven by the solution pH; with general wine pH levels ranging from 2.9 to 3.7, SO_2 and HSO_3^- together, make up the majority of the sulfite levels; about 99.99 % at a pH of 3.41. At higher levels of pH, the sulfite concentration shifts to favor HSO_3^- and SO_3^{2-} (versus SO_2). The following reaction displays the differing forms of sulfites in the must[1]:

$$\mathbf{H2O\ (l) + SO_2(g) \rightarrow H+\ (aq) + HSO_3^-\ (aq) \rightarrow 2H+\ (aq) + SO_3^{2-}\ (aq)}$$

Molecular sulfur dioxide, SO_2, exhibits the most anti-microbial property in wine[1]. This is due to its ability to enter through the cell membrane of microbes, where it then denatures the proteins and enzymes of the microbe, rendering the microbe useless[1]. Furthermore the SO_2 molecule is also capable of binding to oxidative precursors, acetaldehydes, anthocyanins, sugars, and other phenolic compounds in wine as well as slowing the activity of the tyrosinase (polyphenol oxidase) enzyme present in juice.[1] The SO_2 molecule however, is the only sulfite form exhibiting sensory impacts. At high concentrations, SO_2 is sensed by the olfactory system as an overwhelming aroma of a freshly struck match.

Bisulfite, HSO_3^-, is capable of the same binding properties exhibited by SO_2; however its anti-microbial effects are hundreds of times less effective than those of SO_2.[1] Its ability to bind to molecules such as acetaldehydes, sugars, anthocyanins, and other phenolic compounds also gives bisulfite a large role in wine protection.[1] When bound to acetaldehyde, the once unpleasantly fragrant aldehyde becomes odorless and no longer presents itself as a problem, yet problematic binding to anthocyanins strips red wine of its color.

The conjugate base of bisulfite is sulfite, SO_3^{2-}. The SO_3^{2-} molecule is capable of the same binding properties exhibited by both SO_2 and HSO_3^-. The concentration of SO_3^{2-} only plays a significant role at pH levels beyond those acceptable for most wines and is rarely a noticeable contributor to wine protection.

Measured SO_2 is found in two different forms: free and bound, with their sums equaling the total SO_2 concentration. Free SO_2 is the portion that provides the anti-microbial and anti-oxidant properties as well as potential to bind to wine molecules.[1] Without proper free SO_2 concentrations, wines may be taken over by spoilage yeasts and bacteria, or suffer from premature oxidation. Bound SO_2 is the portion that has bound itself to a molecule in the wine and is now part of that molecules' structure. Once bound, the molecule no longer provides protection. However, the weak bonds formed by bound SO_2 allow it to become unbound from molecules to once again add to the free SO_2 concentration or to become re-bound to another molecule.

Importance of Sulfites:

There are several methods for testing free SO_2 levels in wine. One method is the aeration oxidation method (AO). The AO method acidifies a sample of wine, shifting the equilibrium to favor SO_2 and HSO_3^-. Air is then aspirated through the acidified wine into a hydrogen peroxide solution containing a

color indicator which loses its color in the presence of free SO_2. This solution is titrated with a base back to the starting point color after the aspiration process. There are also several quick test kits available on the market that give approximated results using different dye complexes which compare color changes, due to the presence of free SO_2, to color change charts.

The free SO_2 content in a wine is very important to the vintner. Too little free SO_2 leads to lack of protection while too much free SO_2 produces negative sensory impacts. Increased free SO_2 levels at the beginning stages of the winemaking process inhibit the fermentation of the must by wild yeasts and bacteria. These wild yeast and bacteria are naturally occurring on the skins of the grapes and may also impart negative sensory impacts in the wine. A winemaker adds a specifically chosen yeast strain to complete fermentation. Each yeast strain provides different qualities to the wine such as body, flavor, bouquet, etc. Depending on the desired product, differing free SO_2 concentrations are necessary. Total SO_2 concentrations should be kept below 110 ppm to avoid unwanted sensory effects[1]. However, there is no single free SO_2 concentration that works for all wines, rather a concentration based on the types and conditions of the wine based off of wine pH, fruit quality, presence of vinegar, picking methods, and many other factors.

The wine making process starts in the vineyard by allowing the fruit to mature to create quality grapes that are free of disease, pest damage and rot. The grapes are tested in the vineyard prior to harvest to determine the Brix (measure of sugar content), pH, volatile and total acidity contents. Once the desired values are reached, the grapes are picked, and transported to a de-stemming machine where the stems are removed. The de-stemmed grapes land on a sorting table where bad grapes, leaves and remaining stems are removed. A crusher can be utilized to break the skins of the grapes before placing them into a tank, or they can be placed straight into a tank as whole grapes. Once in the tank, the weight of the top grapes will crush the grapes below and the juices will be released. For red wines, a cold soak in refrigerated temperatures, to hinder fermentation, can be done to release tannins and anthocyanins from the skins or fermentation can be initiated right away. White wines do not sit on the skins as the color from the skins is avoided to maintain light colored musts low in tannin. To the must, a pure and desired yeast strain is added to out-compete the natural yeasts and also to initiate the alcoholic fermentation process where the sugar is fermented into ethyl alcohol and carbon dioxide.

Malolactic Fermentation (Secondary Fermentation):

Malolactic fermentation (MLF) is a secondary fermentation process which converts malic acid into lactic acid through the use of lactic acid bacteria. MLF is important as a method to raise the wine pH, affecting the form that the sulfite ion takes in solution. Once a wine has undergone MLF, it is also more microbiologically stable and the threat from the wild lactic acid bacteria Lactobacillus and Pediococcus is negligible as there is no longer malic acid for their digestion.[1] The most prominent MLF bacterium utilized by winemakers is Oenococcus oeni due to it imparting the most desired MLF sensory profiles. Wines benefiting most from MLF are medium to full bodied reds, as well as high-acid whites requiring de-acidification. MLF reduces the fruity grape profile and gives wine a softer, rounder mouth-feel by adding buttery flavors and aromas due to the production of diacetyl, as well as softening of the tannins. Malic acid is associated with the tart taste of green apples and has a harsher mouth-feel than lactic acid. Lactic acid is a softer acid, similar to yogurt or sour milk in acidity, on the palate and adds complexity to a wine. MLF bacteria are highly sensitive to all forms of SO_2, and for their survival total SO_2 levels must be less than 25 ppm, along with a slightly higher wine pH.

There are many spoilage microbes that present themselves as problems in wine. Four of the most prevalent culprits are: Brettanomyces, Lactobacillus, Pediococcus, and Acetobacter. Brettanomyces, or Brett, is a yeast that grows in high pH and aged wines lacking proper free SO_2 protection. Brett can grow in wines without the presence of oxygen or sugars, imparting a "barnyard" or "horse sweat" flavor.[1] Lactobacillus and Pediococcus are two forms of bacteria that cause unwanted malolactic fermentations in high pH wines containing residual sugars.[1] Lactobacillus favors stuck fermentations where it turns residual sugars into copious amounts of acetic acid. Pediococcus is responsible for fermenting cabbage into sauerkraut and imparts an aroma of sauerkraut and dirty socks in the wine[1]. Acetobacter is a bacterium that produces acetic acid in the presence of oxygen and favors tanks with large headspace and improper free SO_2 levels. Acetobacter is found in the intestines of the fruit fly, which is a major vector for the bacteria to reach the wine. There are no microbes capable of growing in wine that are pathogenic towards humans; however the negative sensory impacts that they may impart make infected wine undesirable for consumption.

Wild yeast strains are non-Saccharomyces yeast strains. These naturally occurring yeasts are microscopic and are widely dispersed via many vectors. Throughout history, winemakers would allow the natural yeasts on the grape skins to spontaneously ferment their musts as they had no exogenous yeast with which to inoculate. During the first couple of days of this natural fermentation, the most prominent yeasts would ferment the must until it contained too high of an alcohol content, ~1-5% alcohol by volume, thus killing the most prominent yeasts. The remaining yeasts that could tolerate the higher alcohol levels were then all that remained to ferment the must to higher, yet varying alcohol contents. Primary fermentation would cease when the must was depleted of digestible sugars or the alcohol content had poisoned the yeasts. The leftover grape skins and pomace from the winemaking process, containing the surviving yeasts, were then spread into the vineyards where the surviving yeasts would then be dispersed via wind, insects, and many other vectors onto the next year's grape crop. These surviving yeasts that were dispersed then became the dominant yeast strain of their particular grape growing region. Given the distances between wine producing regions worldwide, different strains of the same yeast began appearing, and each of these strains provided different flavors, smells, fermentations, alcohol contents, mouth feel, bouquet, complexity and even colors to the finished wines.

The most prominently used winemaking yeast strain worldwide is *Saccharomyces cerevisiae*. It is in the fungi kingdom since it has a cell wall made of chitin, uses a DNA template for protein synthesis, is unicellular and cannot form a fruiting body.[2] *Saccharomyces* is used in both brewing and baking, digesting primarily glucose. The yeast digests the glucose molecules for energy through aerobic and anaerobic fermentation producing ethanol and carbon dioxide as main byproducts. The *Saccharomyces* strains have become specialized to specific styles of wine. *Saccharomyces bayanus* is another widely available hybridized yeast strain used for alcoholic fermentation of wines, beers, and ciders.

Different companies have taken the *Saccharomyces* yeast strain and altered it in ways specific to the preferences of a professional or home vintner. Two companies, Red Star and Lallemand, dominate the market for packaged active dry wine yeasts. Each company has its own product line of different *Saccharomyces* yeast strains of which different styles of wine benefit the most. Red Star's yeast line comes from the research teams at the University of California at Davis. Lallemand offers its Lalvin yeast line from its research teams based in worldwide locations and has since the 1970's.

MATERIALS

The selected white grape varietal was Catawba. Classified as *Vitis labrusca L.* (Vitaceae) this grape is of unknown origin and was the most planted grape in the United States until the 1860's.[6] Catawba is a late season ripener with light red grape skins and high natural acidity. The varietal is known as a table grape and is used in jams, jellies, juices and wines. The skins are red but the wines are typically white to pink in color due to low anthocyanin content of the grape skins. The selected red grape varietal was Noiret. Noiret was released in 2006 as a French/American hybrid which was crossbred at Cornell University for use as a wine grape. It was crossbred using Steuben and NY65.0467.08 whose main parenting lines are *Vitis vinifera L.* (Vitaceae) and *Vitis labrusca L.* (Vitaceae). [7] Noiret was bred to be a cold climate grape with late bud break and minimal hybrid sensory effects. The project at hand is examining the concentration of free SO_2 at the end of primary fermentation, in red and white wines, due to different wine making yeast strains. One hypothesis is that the different yeast strains will produce differing amounts of free SO_2. Another hypothesis is that the white wine will contain higher amounts of free SO_2 due to the lack of anthocyanin levels. This project is being performed due to the minimal information presented by the yeast manufacturers on the amounts of the free SO_2 produced. The results will enable both professional and home vintner to be better informed on free SO_2 concentrations post fermentation, increasing the effectiveness of using SO_2 in wines.

METHODS

Juice:

The juice was obtained from Baxter's Vineyards in Nauvoo, Illinois. The grapes were hand-picked and then run through a crusher/de-stemmer machine and pumped through a strainer to remove the stems, seeds, and skins. This free run juice was analyzed for Brix levels and pH.

Musts:

The Noiret grapes were picked on 19 September 2013 at a measured 20 Brix and pH of 3.21. Five gallons of juice were obtained and 1.25 g of potassium metabisulfite were added to obtain a free SO_2 level of 50 ppm. For an alcohol content of 13.5% by volume, 2.5 pounds of C&H pure cane sugar was added and stirred to dissolve. To ensure a healthy and complete fermentation, diammonium phosphate (DAP) was added at a rate of 1g per gallon of juice. The juice was then distributed into 10 half gallon volume containers.

The Catawba grapes were picked on 24 September 2013 at a measured 15 Brix and a pH of 2.97. Ten gallons of juice were obtained and 2.50 g of potassium metabisulfite was added to obtain a free SO_2 level of 50 ppm. For an alcohol content of 12.5 % by volume, 7.5 pounds of C&H pure cane sugar was added and stirred to dissolve. To ensure a healthy and complete fermentation, DAP was added at a rate of 1g per gallon of juice. The juice was then distributed to 10 one gallon volume containers.

One-half of a packet of an individual yeast strain was then added to three-fourths cups of water at 100-105 oF, stirred in, and allowed to sit for 15 minutes to ensure proper yeast activation. Once the activated yeast solution was at room temperature, it was added to its individual container which was then stirred and fitted with a bung and either a 3-piece or S type airlock filled with 80 proof Hawkeye brand vodka (~10 mL) to deter fruit flies. The yeast packet was then taped to the outside of the container to keep record of the particular yeast in each container. The containers were placed in an arched wine cellar with an average temperature of 58°F and allowed to undergo primary fermentation.

Upon completion of primary fermentation, the wines were racked from their respective containers into 355 mL volume containers and fitted with a bung and the same airlock from their previous container which were re-filled with vodka as necessary. The empty yeast packets were transferred from primary fermentation vessel onto the 355 mL vessel to maintain records.

Yeasts:

The following Red Star brand yeast strains were chosen: Premier Cuvee, Pasteur Champagne, Pasteur Red, Montrachet, and Côte des Blancs. The following Lalvin yeasts strains were chosen: K1-V1116, EC 1118, Bourgovin RC 212, 71B-1122, and ICV-D47. Each yeast was kept in its package and refrigerated until an hour before being rehydrated. Removal from the refrigerator to achieve room temperature before addition to warm water was done to avoid shocking the yeast.

Chemical Reagents:

All chemicals were of analytical grade and were not further purified. Each wine sample for free SO_2 testing was taken from containers directly before testing to ensure no further deterioration of free SO_2 levels by direct environment. The chemicals used are given in the following procedures.

Free Sulfur Dioxide Tests:

The aeration oxidation (AO) method was employed to test the wine solutions for free SO_2 based on the following net ionic equation:

$$H_2O_2 + SO_2 \rightarrow SO_3^- + H_2O \rightarrow H_2SO_4$$

For the AO method, a 20 mL sample of wine is placed in a round bottom flask and 10 mL of 25 % phosphoric acid (H_3PO_4) is added to the wine. In another flask, three drops of indicator (mixture of methyl red and methylene blue in 50 % ethanol) were added to 10 mL of 3 % hydrogen peroxide (H_2O_2). This second flask is connected to the first by a hose which enters into the hydrogen peroxide solution. The first flask is then hooked up to an air pump which aspirates at a rate of 1 L of air per minute through the wine and into the hydrogen peroxide and indicator solution mixture. The aspiration process runs for 10 minutes. The hydrogen peroxide and indicator solution are then titrated back to their starting color with 0.10N sodium hydroxide (NaOH). To determine parts per million free SO_2, the number of mL of NaOH added is multiplied by a factor of 16. All AO tests were performed at room temperature under fluorescent lighting with fresh samples of the tested wine. ACCUVIN AV-Free SO_2 (AQT) test kits were employed as a secondary measurement technique. The AQT test is based on the reduction in color exhibited by a dye when it reacts with SO_2. The net ionic equation is as follows:

$$SO_2 \text{ (as } SO_3^{2-}) + \text{dye(colored)} \rightarrow SO_3^{2-} + \text{dye complex(colorless)}$$

The test covers two different ranges: a low range of 0-40 ppm, and a high range of 40-130 ppm. The protocol provided by Accuvin was followed and only the low range tests with the green caps were used. For the test, liquid from the tube with the black cap was poured into the powder of the tube with the green cap. The green cap was replaced and shaken to dissolve powder. Using the provided 91µL sampler bulb, a 91µL sample of wine was taken by squeezing the bulb and allowing air to aspirate through wine, then allowing it to expand to contain the desired volume. The green cap tube was opened and the sampler tip was placed into the reagent and the bulb squeezed once to remove wine sample. The sampler bulb was removed, the sample tubes' green cap replaced, the test tube was shaken up and five minutes were allowed for color development. Determination of free SO_2 level in ppm was done by comparison of the color of the reagent with a test strip container. The tube was held 1 inch above a white background under incandescent or natural lighting for comparison. If the color fell between two colors, an intermediate value was assumed. All tests were performed at room temperature under fluorescent lighting using fresh samples of the tested wine.

Residual Sugars Test:

Clinitest® tablets were used to test residual sugar content using the copper reduction method in the following net ionic equation[4]:

$$RCHO + H2O \rightarrow RCOOH + 2H+ + 2e^-$$

The ten drop method was the method employed. Ten drops (0.5 mL) of the sample wine were placed into the Clinitest® test tube. One tablet was dropped into the wine sample and the test tube was flicked as the reaction proceeded. Once the reaction finished, a comparison of the test tube color to the two drop method Clinitest® results that comes with the tablets was performed.[5] If the color falls between two colors, an intermediate value was assumed. All tests were performed at room temperature under fluorescent lighting using fresh samples of the tested wine.

Wine Condition:

The wine conditions were determined by sight, smell, and taste. Visual inspection was performed under sunlight, fluorescent, and compact fluorescent lighting. Visual wine conditions were inspected by pouring a wine sample into a clean wine glass and tilting the wine glass, holding the wine to be analyzed, slightly to give a shallow area in the wine glass. Visual inspection for color consistency is done here by peering through the shallow area of the sample in the tilted glass, which is held roughly one foot away from the face between one's eyes and the source of light. A color consistent with the red or white varietal indicates good condition while presence of brown color around the edges indicates that the wine has oxidized. Red wines turn a red brick color when oxidized, while white wines tend to turn an amber yellow color, with both styles containing browned color hues around the edges.

Inspection by smell was done by using the same sample from the visual inspection. The wine sample in the clean wine glass is swirled to coat the inside edges of the glass and then by sticking one's nose into the glass and giving a large inhale through the nostrils. Good condition is noted by the olfactory system finding bouquets of aromas consistent with the grape varietal, while oxidized conditions are similar to nutty, sherry-like, or stale olfactory findings.

Taste inspection was performed using the same sample from the visual inspection as well. After the large inhale through the nostrils, a volume chosen by one's own judgment after visual and olfactory inspection is taken in through the mouth. The sample is then rinsed around the mouth taking small inhales through both the mouth and nose to get full flavor recognition. A flavor profile consistent with the grape varietal indicates a good condition, while oxidized wine gives sherry, nut-like, flat, or stale flavors.

RESULTS

A total of 10 commercially available yeast strains were tested: five from Red Star and five from Lalvin. Each strain was tested individually in both a red wine (Noiret) and a white wine (Catawba), for a total of 20 wines produced. The following variables were controlled: initial free SO_2 levels, sugar levels, yeast nutrient levels, and environmental conditions.

The experimental findings showed that one yeast strain, Red Star Côte des Blancs, contained 6.4 ppm free SO_2 by AO and 4 ppm by AQT in the red wine. The same yeast contained 3.6 ppm free SO_2 by AO and 4 ppm by AQT in the white wine. The average end pHs of the red and white wines were 3.30 and 2.91, with standard deviation values of 0.139 and 0.0461 respectively, and relative standard deviation percentages of 3.8% and 1.6% for the red and white

wines respectively. The residual sugar levels measured 0% residual for all yeasts except the Côte des Blancs which produced 0.1 % residual sugars in both the red and white wines, indicating a healthy and complete fermentation in all wines.

Noiret: Red Star Yeast					
Yeast Type	Premier Cuvee	Pasterur Champagne	Pasteur Red	Montra-chet	Cote Des Balncas
Wine Condition	Oxidized	Oxidized	Oxidized	Oxidized	Good
AO (free So2)	0	0	0	0	6.4
AQT (free SO2)	0	0	0	0	4.0
pH	3.70	3.69	3.57	3.56	3.3
Residual Sugars	0.0%	0.0%	0.0%	0.0%	0.1%
Yeast Strain	S. bayanus	S. bayanus	S. cerevi-siae	S. cerevi-siae	S. cerevi-siae

Table 1: Noiret: Red Star Yeast

Noiret: Lalvin Yeast					
Yeast Type	K1-V1116	EC 1118	Bourgovin RC 212	71B-1122	ICV-D47
Wine Condition	Oxidized	Oxidized	Oxidized	Oxidized	Oxidized
AO (free So2)	0	0	0	0	0
AQT (free SO2)	0	0	0	0	0
pH	3.8	3.63	3.62	3.73	3.72
Residual Sugars	0.0%	0.0%	0.0%	0.0%	0.0%
Yeast Strain	S. cerevi-siae	S. bayanus	S. cerevisi-ae	S. cerevi-siae	S. cerevi-siae

Table 2: Noiret: Lalvin Yeast

Table 1 and Table 2 tabulate the results from the Noiret varietal being fermented by the Red Star and Lalvin lines of yeast, respectively. Table 3 and Table 4 tabulate the results from the Catawba varietal being fermented by the Red Star and Lalvin lines of yeast, respectively.

Catawba: Red Star Yeast					
Yeast Type	Premier Cuvee	Pasterur Champagne	Pasteur Red	Montra-chet	Cote Des Balncas
Wine Condition	Good	Good	Good	Good	Good
AO (free So2)	0	0	0	0	3.6
AQT (free SO2)	0	0	0	0	4.0
pH	2.81	2.84	2.85	2.88	2.91
Residual Sugars	0.0%	0.0%	0.0%	0.0%	0.1%
Yeast Strain	S. bayanus	S. bayanus	S. cerevi-siae	S. cerevi-siae	S. cerevi-siae

Table 3: Catawba: Red Star Yeast

Catawba: Lalvin Yeast					
Yeast Type	K1-V1116	EC 1118	Bourgovin RC 212	71B-1122	ICV-D47
Wine Condition	Good	Good	Good	Good	Good
AO (free So2)	0	0	0	0	0
AQT (free SO2)	0	0	0	0	0
pH	2.78	2.78	2.79	2.80	2.78
Residual Sugars	0.0%	0.0%	0.0%	0.0%	0.0%
Yeast Strain	S. cerevi-siae	S. bayanus	S. cerevisi-ae	S. cerevi-siae	S. cerevi-siae

Table 4: Noiret: Lalvin Yeast

DISCUSSION

This project investigated the concentration of free SO_2 available at the end of primary fermentation due to a variety of winemaking yeast strains within both red and white wines. The findings show that endogenous free SO_2 contributions produced by yeast strains are inadequate to protect wines from premature oxidation throughout fermentation and aging. Extra focus needs to be placed on maintaining proper levels of pH, container headspace, sanitation, and yeast nutrition at all stages of the winemaking process. In order to maintain appropriate free SO2 levels, an exogenous form of SO_2 should be utilized in the form of potassium metabisulfite. The hypothesis of variable free SO_2 production by each yeast strain was proven to be correct as the Côte des Blancs yeast strain produced residual free SO_2 while none of the others produced any concentration of residual free SO_2. The hypothesis of higher free SO_2 concentrations in the white wine was inconclusive even though the red wine contained higher SO_2 concentrations for the Côte des Blancs yeast strain.

The hypothesis cannot be rejected due to the fact that all but one red wine had oxidized completely and none of the white wines oxidized at all. The 6.4 ppm free SO_2in red versus the 3.6 ppm free SO_2 in white could be due to the free run juice obtained and its low maceration time, and thus lower than normal anthocyanin levels in the resulting red wine. The SO_2 content bound to anthocyanins could also have been released and falsely resulting in slightly higher readings for the red wine.

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REFERENCES

1. Clark, J. "Oxidation of Aldehydes and Ketones." *Chemguide*. N.p., 2004. Web. 12 February 2014.

2. "Clinitest Color Chart." *Residual Sugar Test Using CLINITEST Tablets.* N.p., n.d. Web. 14 February 2014.

3. "Production of Wine." *Title 21 Code of Federal Regulations*.

4. Grotheer, P., Marshall, M., & Simonne, A. "Sulfites: Separating Fact from Fiction." *University of Florida, IFAS*, April 2005. Web. 13 September 2014.

5. Henderson, P. "Sulfur Dioxide: Science behind This Anti-microbial, Anti-oxidant, Wine Additive." *Practical Winery & Vineyard Journal. January 2009*. Web. 18 February 2014.

6. Lorch, W., Kavanagh, D., Gray, W. B., Adamson, C., & Reeve, J. "Catawba Wine." *Wine-searcher*. 2014. Web. 15 March 2014.

7. Madigan, M., & John M. "Biology of Microorganisms" (11th ed.). Upper Saddle River, NJ: Pearson Education. 2006.

8. Robin, S. "The Health Risks of Sulfur Dioxide in Dried Fruits." *Healthy Eating*. N.p., n.d. Web. 17 April 2014.

9. Lisa S. "Noiret." I*owa State University*, 2008. Web. 13 September 2014.

10. Tebeau, R. "Noiret" *Fringe Wine.* N.p., 2012. Web. 15 March 2014.

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