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OXIDATION

Learning Outcomes: *This section reviews the three fates of oxygen in winemaking, enzymatic oxidation, non-enzymatic oxidation and microbiological consumption. The reader will understand the factors impacting wine oxidation and reductive strength, oxygen and yeast metabolism, the importance of oxygen management in stylistic wine production and how to identify oxygen degradation*

Chapter Outline

Section 1.

Introduction

Oxygen contact with must and wine is an important production feature throughout the winemaking process. In some instances, such as certain juice processing, micro-oxygenation and during barrel aging, controlled exposure to oxygen may play an important beneficial role in wine quality. Controlled aeration may enhance yeast growth, phenolic polymerization, influencing both color stability and suppleness in red wines. During bottling, oxygen levels are generally controlled and restricted to prevent premature deterioration.

The color of wine is one of its most important characteristics. The potential for browning in wines may be closely tied to grape variety, phenols, and other oxidative substrates, as well as to mold growth that may have occurred. Non-enzymatic oxidation in wines results from direct interaction of susceptible substrates with molecular oxygen. This happens normally during the course of aging including controlled barrel aging. It may be accelerated when wine is exposed to air.

Oxygen can be involved in direct chemical reactions, act as substrate of enzymatic reactions, cause phenol polymerization and effect the growth and metabolite production of wine microorganisms. Oxygen attracts electrons from other molecules changing their chemical nature. In chemical oxidation, intermediates are produced that have a higher affinity for electrons than molecular oxygen. As such, many oxidation reactions do not involve molecular oxygen but merely the transfer of electrons between atoms. Electrons go from compounds of low affinity to ones with a higher affinity. Compounds with the weakest hold on electrons are in the oxidized state having lost electrons to compounds with a higher affinity.

The terms oxidation and reduction are commonly used in chemistry, including wine chemistry. Reduction is the opposite of oxidation. The term reduction is often used (not correctly) to describe a wine with sulfur-like off odors. Reductive strength refers to anti-oxidative power, an important wine concept because it relates to longevity. Reductive strength is measured as the rate at which a given wine can consume oxygen without a resulting buildup in dissolved oxygen (DO). Wine in a reduced state is ‘closed’ in aroma and may produces sulfides. This may not be as defect. A wine in a reductive state may have the ability to age and improve during aging, an important wine quality attribute.

Juice Enzymatic Oxidation

The major enzyme involved in browning reactions of juice and wine is polyphenoloxidase (PPO) also called tyrosinase. This is the same enzyme that turns an apple brown after you have taken a bite. The enzyme activity is greatly inhibited by the presence of SO_2, therefore, moderate SO_2 levels in juice act against enzymatic browning. Important features regarding PPO include the following:

- Oxygen depletion. Left unchecked, PPO can significantly reduce must oxygen levels which can impact yeast growth (Schneider, 1998). See below.
- In juice, oxygen can be consumed as rapidly quickly as it is introduced (Bisson 2013).
- The affinity of uncheck PPO for molecular oxygen in juice effectively prevents chemical oxidation (Bisson, 2013).
- Sulfur dioxide. Lever of 25-75 mg/L in clarified juice led to 75- 97% inhibition of PPO.
- Sulfur dioxide is toxic to most native yeasts. Its use can reduce the microbial populations demand on the juice oxygen supply.
- Temperature. Cool must settling greatly reduces the polyphenoloxidase (PPO) concentration by eliminating suspended grape skin fragments.
- Fining. Addition of bentonite and removal of the bentonite prior to fermentation helps to lower the enzyme concentration.
- The fate of oxygen in juice is dependant on the relative ratios and activities of microbes present, PPO activity and sulfur dioxide concentrations (Bisson 2013).

The enzymatic mechanism of wine browning is believed to follow the following:

- Flavonol phenols react with oxygen in the presence of polyphenoloxidase to produce yellow-colored oxidation products (quinones).

- These, in turn, react with oxygen, polymerize, and produce brown-colored condensation products. The more specific chemistry has been outlined by Bisson et al. 2013:

In juice PPO catalysis the oxidation of caftaric and coutaric acid to caftaric acid quinone. This quinone then under goes coupled oxidations with other components of the juice such that the quinine is reduced back to caftaric acid. Thus, polyphenol oxidase generates the reactive intermediates that the participate in the subsequent electron transfer and coupling reactions. Flavanols oxidation by caftaric acid results in quinines polymerizing and precipitation as brown pigments.

The industry has moved to limit the use of sulfur dioxide. Achieving reductions in the levels of SO_2 used, while maintaining quality, requires an understanding of the vineyard and include variety, climatological conditions, fruit maturity, incidence of fruit rot, and temperature of fruit at harvest, during transport, processing and aging.

Prefermentation additions of SO_2 carry over into wine as addition compounds such as pigments and acetaldehyde. Aside from having minimal or no activity with respect to oxidative and microbiological control, the reservoir of bound SO_2 creates a need for further additions in order to achieve the desired levels of molecular free SO_2.

High pH musts and wines, such as we have in Virginia, tend to oxidize at a faster rate than low pH lots. Grapes grown in warmer regions tend to darken faster than the same variety grown in cooler areas. Maturity may affect the tendency toward enzymatic browning.

Although a variety of enzymes are present in sound fruit, sulfur dioxide addition, fermentation and processing (including juice fining) reduces their activity substantially. Thus, polyphenoloxidase activity is usually limited to prefermentation and does not play a major role in further oxidative degradation. Winemakers forced to deal with mold-damaged fruit expect to see more oxidation than with sound fruit.

In these cases, use of SO_2 even at relatively-high levels may not be useful in control of further deterioration. For example, *Botrytis*-produced laccase catalyzes phenolic oxidation, with the resultant polymerization responsible, in part, for browning of the fruit (See chapter on Fruit Rots). Perhaps of greater concern is the oxidation of aroma/flavor compounds. Laccase is resistant to sulfur dioxide, cannot easily be removed with bentonite, and is active in the presence of alcohol, including in bottled wines (See chapter on Enzymes).

Green vs. Brown Juice. (Also see sections on hyper-reduction and hyper-oxidation) There is an age-old debate regarding the merits of brown vs. green juice in the white wine fermentor which relates not simply to style, but also variety. Oxygen exposure in white juice increases browning and can lead to an increase in phenol polymerization, lowering the astringency. Because astringency can mask bitterness, this polymerization can unmask bitterness (Nagel et al 1998).

Protecting certain grape-derived volatile compounds from the impact of molecular oxygen makes sense, particularly in a variety such as Sauvignon blanc, which is dependent on volatile sulfur compounds for varietal character. For example, maximizing the passion fruit 'notes' requires the limitation of oxidative degradation. Indeed, this is a primary reason why some add ascorbic acid to the juice. Ascorbic acid is a very good oxygen scalper, acting much more rapidly than sulfur dioxide (See below).

Oxygen and Yeast Metabolism

Oxygen plays important roles in the physiological status of yeast. Molecular oxygen is required in synthesis of lipids (principally oleanolic acid) and steroids (ergosterol, dehydroergosterol, and zymosterol) needed for functional cell membranes. Steroids play a structural role in membrane organization, interacting with and stabilizing the phospholipid component of the membrane.

It has been shown that yeasts propagated aerobically contain a higher proportion of lipids, and up to three times the steroid level of conventionally-prepared cultures. This increase correlates well with improved yeast viability during fermentation.

As fermentation begins, oxygen present in must is rapidly consumed, usually within several hours. After the initial oxygen is utilized, fermentations become anaerobic. Because yeasts are not able to synthesize membrane components in the absence of oxygen, existing steroids must be redistributed within the growing population. Under such conditions, yeast multiplication is usually restricted to four to five generations, due largely to diminished levels of steroids, lipids, and unsaturated fatty acids.

The need for oxygen supplementation may be overcome by addition of steroids and unsaturated fatty acids. Oleanolic acid, present in the grape cuticle (Radler, 1965), has been shown to replace the yeast requirement for ergosterol (the major steroid produced by yeast) under anaerobic conditions (Brechot et al., 1971). There is also evidence that exogenous addition of yeast “ghosts” or “hulls” may overcome the oxygen limitation, possibly by providing a fresh source of membrane components. Several commercially available supplements contain compounds that support yeast cell membrane development (See chapter Controlling Microbial Growth in Wine).

Methodology of starter propagation is important with respect to subsequent requirements for oxygen. Aerobic propagation has been demonstrated to significantly enhance subsequent fermentative activity. Yeast populations reach higher final cell numbers, and fermentations proceed at a faster rate, when starters are prepared with aeration.

Optimum Oxygen Management during Fermentation

Yeast produce membrane lipids only when grown aerobically. In the initial growth phase, proper oxygen management leads to proper production and storage of sterols in the yeast cell, which can be shared with subsequent daughter cells. It is possible to increase yeast ethanol tolerance by promoting synthesis of sterols, by adding oxygen (air) in the starter and during fermentation. This may become very important during seasons where the potential alcohol level is high. Additionally, some yeast-derived commercial products aid in sterol synthesis. Yeast lees deplete the oxygen content and can impact the redox potential and formation of SLO (sulfur-like off odors)(See chapter on lees management). Oxygen management involves an understanding of the following:

- Optimum 8-10 mg/L oxygen during the initial growth phase
- Standard pump-over during active fermentation generally will impart less than 3.0 mg/L oxygen
- Oxidative stress may be a primary cause of early yeast mortality
- Lees are important oxygen consumers, even after yeast cell death
- Lack of oxygen can contribute to sulfur-like off odors
- Oxygen additions may allow yeast to produce more glutathione, and important white wine antioxidant
- Oxygen addition may allow for phenolic polymerization
- Polyphenoloxidase (PPO) enzyme activity can significantly deplete must oxygen levels, and thus impact yeast membrane lipid production
- Aeration of red must is often associated with color loss. Big difference in both effect on color and polymerization of color is usually due PPO activity.

In the initial phase of fermentation, winemakers are usually relatively aggressive in providing oxygen (air), depending on the fruit condition, cultivar and style. Due to the production of carbon dioxide, it is difficult to over-aerate. Punch-down in a carbon dioxide environment does little to get much air in the fermentor. As such, many use pump-over with aerators. Some use air wands to incorporate air below the cap in red wines. Additionally, micro-oxygenation is used during and post-fermentation (see module on Micro-oxygenation).

Aeration of red must is often associated with color loss. Enzymatic oxidation and production of quinines. Difference in both the effect on color and polymerization of color is usually attributed to lack PPO activity (Bisson 2013). Acetaldehyde is not formed in significant concentrations in juice because there is no ethanol is present and because hydrogen peroxide is not a product of PPO.

Post-Fermentation Oxidation

Management of oxygen pickup in aging wine is crucial. Because air is 21% oxygen, it is necessary to reduce this concentration to the lowest point possible during movement or fining of delicate white wines. Transfer lines, pumps, and receiving tanks may be purged with inert gas before use (See chapter on the use of Gases). Commercially-available racking devices use nitrogen gas to pressurize the headspace above the wine, thus facilitating transfer of wine from barrel to barrel without pumps. Controlled air exposure (splash racking, etc.) is frequently useful to help soften and evolve tannins and stabilize color in young red wines.

Acetaldehyde, an Important Product of Oxidation

As wines age, acetaldehyde levels increase due to chemical oxidation of ethanol, and in the case of improperly stored wines, growth of oxidative yeasts and bacteria at the wine's surface. The film (referred to in older literature as "mycoderma") exists as a mixed population of several species including *Pichia, Candida, Hansenula*, and oxidatively growing *Saccharomyces.* Ethanol represents the primary source of carbon in aerobic film growth. In addition to substantial production of acetaldehyde, film yeasts may produce acetic acid and ethyl acetate. Oxidative metabolism may be exploited in production of flor sherry where acetaldehyde levels may exceed 500 mg/L.

Control of film yeast is best accomplished by depriving them of oxygen needed for growth. Thus, minimizing oxygen contact during storage is essential in preventing population buildup (See chapter of gasses). Cellar temperature is also known to impact development of oxidative yeasts. At 8°C to 12°C (47-54°F) negligible film formation can be seen in wines of 10 to 12% alcohol, whereas at higher temperatures, growth can be observed at 14% alcohol.

In that some film-forming species are fairly resistant to molecular SO_2, attempting to suppress growth with SO_2 (once film formation is observed) may not be feasible. In these instances, it is recommended that the wine be transferred (without disruption of the film) to sanitized cooperage (Baldwin, 1993).

Acetaldehyde is also an intermediate in bacterial formation of acetic acid. Under low-oxygen conditions and/or alcohol levels greater than 10% (vol/vol), acetaldehyde tends to accumulate instead of being oxidized to acetic acid. Muraoka et al. (1983) report that aldehyde dehydrogenase is less stable than ethanol dehydrogenase.

Aside from chemical and microbiological formation, winemaking practices influence the level of acetaldehyde present in wine. Timing of SO_2 additions is important. Pre-fermentation additions, as well as additions during the course of fermentation, increase the concentration of acetaldehyde in the wine and lower the concentration of free SO_2. This is a reason why attempting to stop fermentation by the addition of sulfur dioxide may not always be the best approach. Other parameters producing higher levels of acetaldehyde include increases in pH and fermentation temperatures (Wucherpfennig and Semmler, 1973).

The combination of anthocyanin and tannin molecules is promoted by the presence of acetaldehyde produced from ethanol oxidation during barrel storage. The anthocyanin-tannin complex is important to red wine color stability.

Oxygen uptake such as may occur during bottling may result in oxidation of ethanol to acetaldehyde. The muted varietal character of newly bottled wines, may reflect transitory oxidation and accumulation of acetaldehyde. Although it is believed by some that SO_2 additions at bottling will limit short term oxidation, the binding rate of SO_2 is a very slow process.

Sensory Considerations of Acetaldehyde

Immediately after fermentation, table wines generally have an acetaldehyde concentration of less than 75 mg/L. The sensory threshold in wines ranges from 100 to 125 mg/L. Above this concentration acetaldehyde can impart an odor to the wine described as over-ripe bruised apples, sherry, and nut-like.

An aroma screen takes advantage of the fact that acetaldehyde quickly binds with sulfur dioxide. For the specific methodology for conducting an aroma screen go to www.vtwines.info. Click On-Line Publications or see Zoecklein et al. 1995, 2005. Blending and refermentation are industry practices used to reduce acetaldehyde concentrations.

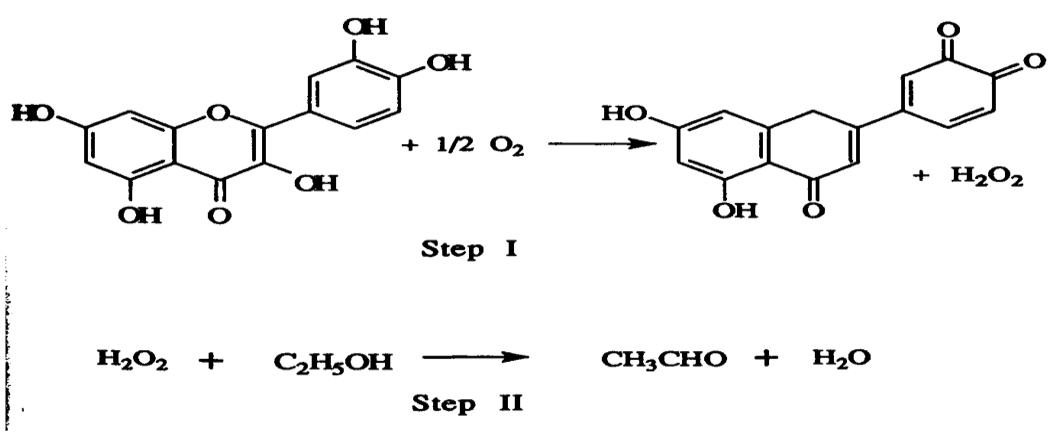
OXIDATION

Section 2.

Coupled Oxidation and Antioxidants

Understanding the mechanisms of oxidation is important for winemakers. As illustrated in Figure 1, wine oxidation can involve the oxidation of a phenol to produce a quinone (oxidation product) and hydrogen peroxide (H_2O_2). In the example below, the hydrogen peroxide generated oxidizes ethanol to acetaldehyde (coupled oxidation).

Figure 1. Coupled Oxidation to Produce Acetaldehyde from a Phenol



Sulfur Dioxide

Sulfur dioxide is an important antioxidant used in grapes and wines, and is discussed in a separate module chapter titled: Sulfur Dioxide. It is important to note that sulfur dioxide additions do not bind the oxygen and, therefore, do not prevent the first step in the coupled oxidation shown in Figure 1.

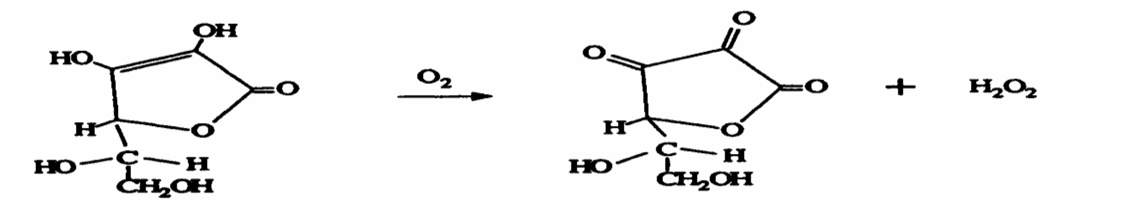
Because of the size of the molecule sulfur dioxide can act as an allergen. The FDA estimates that 1% of American consumers are hypersensitive to inhaled sulfur dioxide. As such, the BATF (Now TTB) in 1986 began to require sulfite labeling on all wines containing 10 mg/L sulfur dioxide or more. That caused to the wine industry to re-think the use of this compound, the quantities added, when to add it, and alternatives.

Ascorbic Acid

It is important to note that sulfur dioxide additions do not bind the oxygen and, therefore, do not prevent the first step in the coupled oxidation shown in Figure 1. Some winemakers use ascorbic acid, or vitamin C, as an antioxidant, particularly in wines with low oxidative buffering capacities, such as white wines low in phenols. Ascorbic acid sometimes protects the fruit and wine, and acts as an antioxidant, while at other times it can act as a proto-oxidant, or oxidative promoter.

The two roles of ascorbic acid are mainly the result of its concentration and the presence of adequate sulfur dioxide. As illustrated below, when ascorbic acid is added to wine, it binds oxygen rapidly to form two reaction products, dehydroascorbate and hydrogen peroxide. If there is not enough ascorbic acid maintained to react with the oxygen, oxidative degradation, including coupled oxidation, can occur. If there is not adequate sulfur dioxide maintained to bind with the hydrogen peroxide formed by the ascorbic acid, wine oxidation can occur.

Figure 2. Binding of Ascorbic Acid with Oxygen to form Dehydroascorbate and Hydrogen Peroxide



Ascorbic acid ------------------------> Dehydroascorbate + Hydrogen peroxide

Therefore, the keys to optimizing the performance of ascorbic acid as an antioxidant are to maintain a concentration of about 50 mg/L, and to have adequate sulfur dioxide. The use of ascorbic acid involves the following considerations:

- Reaction between ascorbic acid and oxygen is much more rapid than SO_2.
- SO_2 does not directly react with oxygen, but mainly with reaction products, such as H_2O_2.
- Optimum levels of ascorbic acid (50 mg/L or more) and more SO_2 can prolong the antioxidant phase of ascorbic acid. For example: If 100 mg/L ascorbic acid in wine reacts completely with oxygen, 62 mg/L SO_2 is required to react with the ascorbic acid oxidation product.

The possible use of ascorbic acid should be determined based on the assessment of white wine longevity and oxidation potential. This addition compound may have a place in the production of some delicate, low phenol white wines.

Glutathione, and Important Antioxidant

Glutathione is a polypeptide produced by the grapevine, and by yeast at the end of fermentation. It is a strong antioxidant and helps to stabilize aroma components. The enzymatic oxidation of a simple, but important juice phenol (caftaric acid) forms an oxidation product that can react with glutathione. This binding forms what is termed a grape reaction product (GRP). The grape reaction product terminates the oxidation process and subsequently limits oxidation (Cheynier, 2008).

The effect of this grape reaction product can be dramatic. For example, one molecule of a simple phenol can consume 3.4 atoms of oxygen. When this phenol is combined with glutathione to form the grape reaction product, that increases to 8.5 molecules (du Toit et al., 2007).

Glutathione plays an important role as an oxidative buffer, thus impacting wine longevity. Lees contain glutathione; this is one reason why lees storage keeps wines from undergoing oxidative degradation. The following are important relationships regarding glutathione:

- There is a positive correlation between glutathione and the freshness and longevity of white wines.
- There is a positive correlation between the concentration of glutathione in the must and the concentration in the young wine.
- There is a positive correlation between the juice free amino nitrogen (FAN) concentration and the glutathione concentration at the end of alcoholic fermentation.
- Copper inactivates glutathione. There is an inverse relationship between glutathione and copper concentration in musts and wines.
- Minimizing the loss of glutathione is a key to making white wine and optimizing wine longevity.

The management of native grape and yeast glutathione is an important step for maximizing white wine aroma/flavor and maintaining wine longevity. The following are some generalizations about glutathione (GSH) in white winemaking outlined, in part, by Laffort, Inc. (2008).

- GSH increases with fruit maturation (Adam and Liyanage, 1993).

- The concentration is positively correlated to fruit YAN (yeast assimilable nitrogen) concentration (Dubourdieu and Lavigne-Cruège, 2004), another good reason for the analysis of YAN.
- Grape GSH concentration is easily lowered during processing. Attempts to limit this oxidation spurred the practice referred to as hyper-reduction, or anaerobic grape processing.
- GSH is assimilated by yeast during the beginning of fermentation and released at the end of alcoholic fermentation.
- Maximum release of GSH by yeast occurs only if yeasts have sufficient YAN (Laffort, 2008).
- 30 days-post fermentation, GSH levels can be as high as or higher than in the initial juice (Dubourdieu and Lavigne-Cruège, 2004).
- GSH can slow the decrease of some wine volatiles during aging (Roussis et al., 2007).
- The impact of GSH on limiting the decrease of volatile esters and terpenes during aging is concentration dependent (Papadopoulou and Roussis, 2008).
- It is estimated that 20 mg/L of GSH at the end of the aging period is optimum for aroma protection (Laffort, 2008).
- There is an inverse relationship between GSH and copper concentration in musts and wines (Dubourdieu, 2006).
- When lees are eliminated, the GSH concentration diminishes rapidly. In new barrels, this reduction is even greater due to the oxidation effect of new wood.
- Minimizing the loss of GSH is a key to optimizing white wine longevity.
- Some fermentation adjuncts contain glutathione.

Fermentation Adjuncts and Glutathione. GSH is positively correlated to YAN, an additional justification for the analysis of YAN. Fermentation complements/addition products contain some or all the following:

- inorganic nitrogen (DAP)
- organic nitrogen (alpha-amino acids)
- unsaturated fatty acids
- sterols, thiamine, folic acid, niacin, biotin, and calcium pantothenate
- magnesium sulfate
- inactive yeast cell walls
- peptides
- micro-crystalline cellulose
- other yeast autolysis products, including GSH

One of the possible benefits of some fermentation complements/addition products is that they include GSH. Their use may help resist oxidation. Additionally, optimum oxygen, sulfur dioxide, pH, and lees management can increase the protection of white wine aromatic quality.

Iron and Copper: Pro-Oxidants

Metals, such as iron and copper, have the disadvantage of being strong oxidizers, possibly impacting wine longevity. The potential oxidizing effect is illustrated by the Fenton-type reaction:

$$H_2O_2 + Cu^{+2} \rightarrow Cu^{+3} + OH^{-} + OH^{*}$$

The OH*, or hydroxyl radical, is the most oxidative species. This is a potentially large problem, notably in white wines with relatively low concentrations of oxidative buffers such as phenols. As such, copper in the must, from late-season Bordeaux mix vineyard sprays or any other source, can lower the longevity of white wines.

Enological Tannins: Oxygen Buffers

Tannins are essentially oxidation buffers. There are basically two chemical classes of tannins, hydrolysable and condensed. These two groups differ in nearly every characteristic, other than their ability to bind with proteins. Hydrolysable tannins bind with proteins by hydrophobic interactions. Condensed tannins bind proteins through hydrogen bonding. Grape skins and seeds contain only condensed tannins. Hydrolysable tannins are derived from oak wood or as an additive to wines.

Enological tannins available on the market may differ in a number of respects, including the following:

- extraction method
- purity
- processing method
- source, including wood, grape skins, and seeds
- toasting variation
- degree of oxidation

Tannins for wine addition can be derived from oak, chestnut, seedpods, etc. Most are water or steam extracted, dried, and milled. Different products undergo hydrolysis, pH and color adjustment, and sulfite addition, and may be finished by spray or freeze-drying.

There is a vast array of tannins on the market, and many are tailored to perform different tasks. Tannins are added for the following purposes and problem corrections:

- redox buffer
- raisined fruit
- sun-damaged fruit
- unripe grape tannins
- structural/textural, mouthfeel modification
- increased substrate for micro-oxidation
- limit the activity of laccase

- help to precipitate proteins
- help to modify aromas, including vegetative aromas
- help increase aging potential
- possibly to help stabilize red wine color

Many believe that an addition of both condensed and hydrolysable tannins is best, depending upon the purpose. Regardless, post-fermentation use should involve careful laboratory fining trials. The actual tannin content of commercially-available enological tannins varies significantly.

The timing of tannin addition may be important, depending upon the purpose. Like many agents, their negative impacts are usually less with earlier addition. Adding tannins before or during the early stages of fermentation allows for integration with the other structural elements.

It should be noted that during pre-fermentation or fermentation addition, grape proteins might precipitate a portion of the added tannin. The degree of precipitation is dependent upon the grape variety and the season, among other factors. This is one reason why some use multiple additions during fermentation.

Aroma Compounds and Oxidation

Wine longevity can be defined as the time period that the product conforms to stylistic goals. Crafting fine wine requires a holistic understanding of winemaking. Winemaking goals usually include the following:

- no excess of SLO (sulfur-like off odors) impacting aromas and mouthfeel
- a stable and concentrated colloidal matrix
- no excess of volatiles contributing to “chemical” and “mineral” aromas
- no or limited herbaceous aromas
- no excess of harsh or “green” tannins

- management of desirable aromas/flavors

Management of desirable aroma/flavor is among the universal winemaking goals. Wine oxidation is characterized by the transformation of aroma/flavor compounds, leading to qualitative and quantitative changes resulting in a loss of wine-like, fruity, and estery aromas. On occasion, faulty aromas can develop, reminiscent of wax and naphthalene, as a result of ATA (atypical aging).

Table 1. Theoretical Sensitivity of Aroma Compound Descriptors to Oxygen Exposure (adapted from Nomacorc, 2008)

Passion fruit	Citrus	Candy	Sherry
Gooseberries	Lime	Earthy	Honey
Grapefruit	Peach	Petrol	Horse
Black currant	Geranium	Flowery-fruity	Band-aid
Cat pee	Lychee	Tobacco	Smoky
Tomato leaf	Orange blossom	Violet	Vinegar
Coffee	Rose	Tropical fruit	Vanilla
Earthy	Cucumber	Peach	Nutty
Rotten eggs	Pine	Banana	Oak
Rubbery	Coconut	Melon	Bread
Wet dog		Wax & honey	Caramel

Oxygen Exposure

No Oxygen<-->More Oxygen

Some aroma compounds are not impacted by oxygen. For example, descriptors such as bell pepper, grassy, chocolate, potato, earthy, flinty, and mushroom generally are not impacted by oxygen exposure (Nomacorc, 2008).

OXIDATION

Section 3.

Hyper-Reduction

Although much of the following is directed toward Sauvignon blanc, issues regarding grape-derived aroma/flavor, and methods to maximize aroma/flavor, apply to other white varietals, as well.

There is an age-old debate regarding the merits of brown versus green juice in the fermentor. As research continues, we find the dichotomy between green versus brown relates not simply to style, but also to variety and longevity issues. For example, maximizing passion-fruit notes in Sauvignon blanc requires the limitation of oxidative degradation of fruit volatiles, whereas other varieties may not be quite so labile.

Sauvignon blanc juice has a simple aroma that develops considerably during fermentation, and is dominated by relatively few volatile compounds. The volatiles that contribute to the varietal character are present in the grapes, and formed during winemaking from specific aroma/flavor precursors. Very potent pyrazine compounds are involved in the vegetative, herbaceous, or capsicum-like aromas. Additionally, a number of important thios, or sulfur-containing compounds, contribute to Sauvignon blanc varietal character (Dubourdieu, 2006).

Some sulfur-containing compounds contribute to reductive odor defect, while others are important contributors to varietal character. Those thios responsible for varietal character, and the important pyrazines, are listed below.

Table 2. Threshold, concentrations and olfactory descriptions of the methoxypyrazines and volatile thiols in Sauvignon blanc wine (Tominage et al., 1998; Ribéreau-Gayon et al., 2000)

Compound	Perception Threshold * (ng/L in water) ** (ng/L in model wine)	Sauvignon blanc Wine (ng/L)	Olfactory Descriptions
2-methoxy-3-isobutyl pyrazine (MIBP)	2 *	5-50	capsicum/earthy
2-methoxy-3-isopropyl pyrazine (MIPP)	2 *	<10	capsicum/earthy
2-methoxy-3-*sec*-butyl pyrazine (MSBP)	1 *	<10	capsicum
4-mercapto-4-methyl-pentan-2-one (4MMP)	0.8 **	4-40	cat's pee/broom
3-mercaptohexan-1-ol (3MH)	60 **	200-5000	grapefruit/ passion fruit
3-mercaptohexanol acetate (3MHA)	4.2 **	0-500	broom/passion fruit

In Sauvignon blanc, thio-based aroma/flavor precursors are not evenly distributed in the grape. Approximately 80% of the 4-MMP precursors are found in the juice, while 50% of the 3-MH precursors are in the skins. Therefore, initial processing impacts the extraction from the fruit. The thio concentration changes during Sauvignon blanc fermentation.

Fermentation converts a portion of the non-odorous precursors to their odor-active form. For this conversion to occur, thio-based precursor compounds must enter the yeast cell, be cleaved (converted to their odor-active form), and excreted from the yeast cell into the surrounding environment. Factors impacting these steps include must composition, yeast strains, and oxygen.

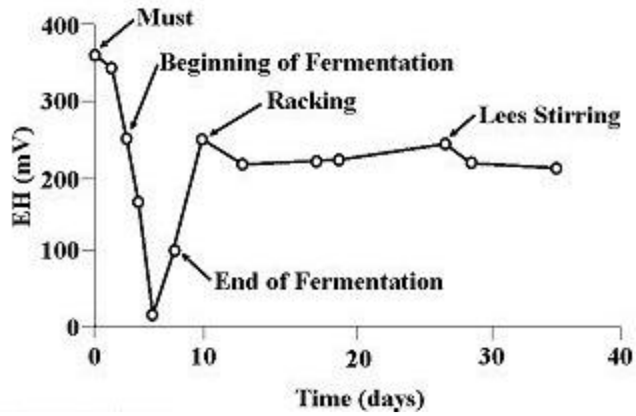
It is believed that thios, like 4-MMP and others, are hydrophobic enough to get inside the cell without an active transport system. Yeasts differ notably in their ability to convert thios to their odor-active form. Different strains have different genes for cleavage enzymes, and the enzymes for precursor cleavage of compounds, such as 4-MMP and 3-MH, may be different.

This may suggest the merit of fermentations conducted by multiple versus single genus or strains of yeast. Currently, processing is a very inefficient means of optimizing aroma/flavor. Regardless of yeast, it appears that only about 1-5% of the precursor compounds get converted to their odor-active form. New efforts regarding hybridized yeast may increase the conversion to the odor-active form.

Thios are easily oxidized, resulting in loss of aroma/flavor. After excretion, thios must be stabilized in order to impact aroma/flavor. As such, phenolic extraction and oxidation must be adequately controlled. Phenols can oxidize to form quinones (brown phenols) and produce hydrogen peroxide, in coupled oxidation.

As a gross generalization and for winemaking purposes, redox can be thought of as the quantity of oxygen. As Figure 3 indicates, after fermentation begins, the redox potential declines significantly. During this period, wine oxidation is limited. Oxidation can occur both before and after fermentation, when the higher redox potential allows for juice or wine oxidation.

Figure 3. Oxidation-Reduction Potential in Dry White Wines (Vivas, 1999)



Many of the volatile sulfur compounds important to Sauvignon blanc varietal character, for example, are easily oxidized. Therefore, protection from the impact of molecular oxygen at berry breakage makes sense. The same is true with many other white varieties.

Lowering the molecular oxygen exposure (and the addition of antioxidants, such as ascorbic acid, glutathione-containing products, and sulfur dioxide) helps to lower the redox potential and limit oxidative degradation. This is a primary reason why some winemakers blanket juice with carbon dioxide at the press. Ascorbic acid addition helps to bind molecular oxygen. Ascorbic acid is a very good oxygen scalper, acting much more rapidly than sulfur dioxide.

Blanketing juice with carbon dioxide during pressing may help to keep the juice green, and keep the oxidation-reduction potential low, thus limiting aroma/flavor degradation. This can assist in minimizing oxidative degradation of certain labile aroma/flavor compounds. The debate regarding the importance of green versus brown juice relates both to style and variety. The importance of fermenting green juice with limited oxidative degradation is referred to as hyper-reduction.

Because oxidative degradation results in the loss of aroma/flavor, and because thios are easily oxidized, the concept of hyper-reduction has developed. As reported in Enology Notes #102, hyper-reduction involves processing steps to help minimize oxidative degradation by keeping wines in a reduced or low-oxygen state. How well the varied procedures used in hyper-reduction, such as carbon dioxide blanketing, work has not been fully evaluated or documented.

However, there are some important steps known to have a positive impact on minimizing oxidation and the loss of aroma/flavor. These include the following:

- no copper
- low concentration of phenols
- protection from oxidation via sulfur dioxide, glutathione, and/or lees storage

As previously reported, copper is a strong inorganic oxidative catalyst. Copper in the must from late-season Bordeaux mix vineyard sprays, or any other source, can lower the longevity of wines like Sauvignon blanc.

Copper inactivates glutathione. The role of glutathione as an oxidative buffer is receiving considerable attention. Glutathione is a polypeptide produced by the grapevine and by yeast at the end of fermentation. It is a strong antioxidant. Research conducted by Dubourdieu's group has shown that grape glutathione concentration declines at the beginning of fermentation, and then increases again at the completion of fermentation.

The following are important relationships regarding glutathione:

- There is a positive correlation between glutathione and the freshness and longevity of Sauvignon blanc wines.
- There is a positive correlation between the concentration of glutathione in the must and concentration in the young wine.

- There is a positive correlation between the juice free amino nitrogen concentration and the glutathione concentration at the end of alcoholic fermentation.
- Minimizing the loss of glutathione is the key to white winemaking.

Dubourdieu has conducted trials with the addition of 10 mg/L glutathione at bottling to help control oxidative degradation. Such additions, and/or the management of native grape and yeast glutathione, may be important steps for maximizing white wine aroma/flavor.

Hyper-Oxidation

Hyper-oxidation is a procedure which encourages oxidation at the juice stage. The purpose of hyper-oxidation is to oxidize phenolic substrates prior to fermentation, resulting in a more oxidatively-buffered wine post-fermentation. This procedure is not used on floral or delicate varieties. As a result of PPO activity oxygen can be consumed in a juice as rapidly as it is introduced if not inhibited. Rather than adding air, in many cases one would get the same effect by not adding sulfur dioxide and allowing unlimited PPO activity. In hyper-oxidation it is important to rack the juice off the brown sediment. Failing to do so may help to explain the variable results from this process (Schneider 1998).

Bottle Shock

Bottle shock is a rather strange phenomenon where a wine's aromatic intensity is reduced after bottling, only to return sometime later. The cause(s) of bottle shock are unknown, but widely debated, and are likely the result of oxidative effects. Generally, if wines are tasted immediately during, or shortly after, bottling, there is no noticeable effect.

However, within the course of a few hours or days, the oxygen present can react with wine components, causing a reduction in the aromatic intensity. Traditionally, this loss has been attributed to the production of aldehydes, a product of ethanol oxidation. Others suggest that bottle shock is the result of another oxidation product, hydrogen peroxide.

Untypical/Atypical Aging

Grape nitrogen appears to be related to a sensory phenomenon known as untypical (UTA) or atypical (ATA) aging. This topic has been reviewed in editions of *Enology Notes* (#14, 77, 107). Some Virginia vineyards experienced moisture stress conditions that may have increased the incidence of ATA in wines produced from those vines.

Wines with this taint lose varietal character very early, and develop atypical aromas and flavors, described as naphthalene (moth balls), dirty dish rag, wet towel, linden, floor polish, etc., and are characterized by an increase in a metallic-like bitterness.
First reported in Germany, ATA has been identified in other European wine-producing regions, the Pacific Northwest, California, and the eastern US. This sensory problem has been associated with vine stress, impacting nitrogen metabolism.

A simple screening test for white wines is provided in *Enology Notes* # 77 at www.vtwines.info.

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