Sauvignon Blanc Wine: Contribution of Ageing and Oxygen on Aromatic and Non-aromatic Compounds and Sensory Composition : A Review

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Oxidation and the capacity for sufficient ageing potential of white wines are constant problems for winemakers worldwide. In general, it is accepted that certain grape varieties are especially sensitive to oxidation, suggesting that some of the chemical components key to their sensory attributes are strongly modulated by oxygen exposure. Sauvignon blanc is a well-documented example of an oxygen-sensitive wine, and understanding the stability of various compounds is crucial in order to preserve the fresh and fruity characters of Sauvignon blanc wines while preventing the formation of off-odours over a long period of time. Compounds such as the volatile thiols and methoxypyrazines are key aroma compounds responsible for the typical Sauvignon blanc aroma, while other aroma compounds, such as esters, alcohols and acids, can also contribute to the wine aroma. Oxidation-related compounds (such as aldehydes) can occur under certain conditions and it is essential to understand the chemistry behind oxidation to control and manage the development of various characteristics in wine. The sensory interactions occurring between various compounds (both aromatic and non-aromatic) are also important due to enhancing or suppressive effects that can mask certain aroma nuances. This review focuses on the stability of certain compounds of Sauvignon blanc wines during oxidation and ageing and how it affects the aromatic and non-aromatic composition of these wines.

ABBREVIATIONS
3MH: 3-mercaptohexan-1-ol; 3MHA: 3-mercaptohexan-1-ol acetate; 4MMP: 4-mercapto-4-methylpentan-2-one; IBMP: 3-isobutyl-2-methoxypyrazine; GSH: glutathione; GRP: grape reaction product; MeSH: methyl mercaptan; DMS: dimethyl sulphide; 5-HMF: 5-hydroxymethylfurfural; 5-MF: 5-methylfurfural

INTRODUCTION

Aroma is an important factor in the quality of all foods, but in wines the aroma is probably one of the most important aspects contributing to the overall quality. A large number of chemical compounds with different volatilities and polarities are responsible for the aroma of wine (Arrehnius *et al*., 1996) and, during winemaking and ageing, various reactions and interactions occur that can influence the perception of the wine bouquet.

One of the most important oenological problems in winemaking is premature wine oxidation, particularly the oxidative spoilage of young white wines. With the exception of sherry and sherry-like wines, white wine quality, in general, will decrease with oxidation (Singleton *et al*., 1979).

This oxidation can take place in a short amount of time during which a significant loss of fresh and fruity aroma takes place, followed by a colour change as well as the development of unwanted oxidation odours.

Aroma attributes that develop due to the formation of new oxidation compounds have been described as honey-like, farmfeed, hay, woody-like, toasted, dry fruits, caramel, overripe fruit, apple, oxidised apple, acetaldehyde, cooked, aldehyde and liquor (Toukis, 1974; Noble *et al*., 1987; Renouil, 1988; Halliday & Johnson, 1992; Chrisholm *et al*., 1995; Escudero *et al*., 2002; Silva Ferreira *et al*., 2002b). These types of unpleasant descriptors are unwanted in wines, and winemakers go to great lengths to avoid the formation of the chemical compounds responsible for these attributes.

Young wines contain high concentrations of oxygen-reactive species, triggering a chain of chemical reactions and consequently altering the wine content. Various closure permeability studies have been performed to assess the effect of oxygen and ageing on the quality of young white wines (Godden *et al.*, 2001; Brajkovich *et al.*, 2005; Lopes *et al.*, 2005; Skouroumounis *et al.*, 2005a). Generally speaking, the conclusion of these studies is that less permeable closures allow better preservation of the fresh and fruity character of the wine, presumably by preventing the oxidative loss of pleasant aroma compounds (Lopes *et al.*, 2009).

Too little oxygen exposure has been associated with 'reductive' off-odours. Closures not allowing sufficient oxygen ingress could lead to the formation of the compounds contributing to the 'reductive' aroma (Godden *et al.*, 2001; Skouroumounis *et al.*, 2005a; Kwiatkowski *et al.*, 2007; Lopes *et al.*, 2009). Conversely, oxidative loss of aromatic compounds and the evolution of undesirable aroma compounds will occur with excessive oxygen exposure. Fig. 1 shows the evolution of aroma compounds during the ageing of wines exposed to low and high oxygen concentrations.

It can be concluded that a moderate degree of oxygen exposure allows the expression of optimal aroma attributes (Brajkovich *et al.*, 2005; Lopes *et al.*, 2009; Ugliano *et al.*, 2009; Nygaard *et al.*, 2010); however, the actual degree of oxygen exposure that is necessary to achieve this has not been determined. Very few such studies on Sauvignon blanc wine have actually included precise oxygen measurements (Brajkovich *et al.*, 2005; Lopes *et al.*, 2009; Ugliano *et al.*, 2011), thus still making it difficult to define how much oxygen would be beneficial for wine during bottle ageing. The type (variety and composition) and style (winemaking practices such as wood contact) of the wine are also expected to play a crucial role in the amount of oxygen required to achieve the ideal wine.

While the chromatic changes during wine ageing are well documented, little is known about the aromatic deterioration in relation to white wine oxidation, even though it seems to take place prior to discoloration (Li *et al.*,

2008). Other than that, studies investigating the oxidative spoilage of wines were often carried out under conditions accelerating oxidation, such as high oxygen exposure or high temperatures (Ferreira *et al.*, 1997; Escudero *et al.*, 2000a; 2002). Conversely, oxidative processes taking place during ageing are usually rather mild and the significance of such levels of oxidation on a wide range of chemical compounds and sensory profile of the wines remains to be established. The interactions between aroma compounds present at any one time in the wine during bottle ageing could also significantly influence the wine's character by changing the perception of certain attributes and should be investigated. This review will focus on the various wine constituents of a typical Sauvignon blanc wine and subsequent oxidation reactions. Sensory aspects and interactions between aromatic compounds will also be discussed.

WHITE WINE OXIDATION

Oxygen during wine processing

During winemaking, the grape must and wine are exposed to different levels of oxygen. Macro-oxygenation refers to higher amounts of oxygen being dissolved during winemaking processes such as pump-overs during fermentation, while micro-oxygenation occurs during barrel ageing, for instance, and nano-concentrations (even lower concentrations of dissolved oxygen) could be obtained during bottle ageing. Any oenological practices involving air undoubtedly will cause oxygen dissolution and wines saturated with oxygen will contain about 8 mg/L oxygen at cellar temperatures and atmospheric pressure (Singleton *et al.*, 1985). Wine is capable of consuming a considerable amount of dissolved oxygen (wine constituents react with oxygen), which has been ascribed to the total phenol content and explains why red wine can consume more oxygen than white wine (Rossi & Singleton, 1966).

The effect of single oenological processes on the level of dissolved oxygen in wines can be classified as 'high enrichment' and 'low enrichment' treatments (Castellari *et al.*, 2004). A study done on over 500 red and white
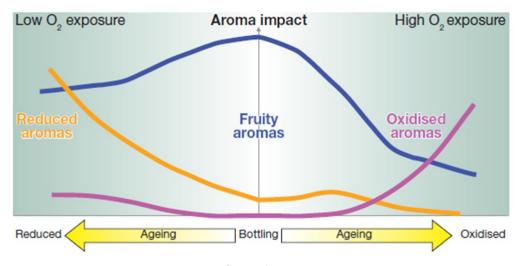

FIGURE 1

The effect of oxygen exposure on the aroma of bottled wine (Ugliano *et al.*, 2010). Reproduced with permission from The Australian Wine Industry Technical Conference Inc.

wines identified 'high enrichment' practices as racking, centrifugation, refrigeration, continuous tartaric stabilisation and bottling (Castellari *et al*., 2004). Dissolved oxygen concentration after these treatments ranged from 1 mg/L to about 8.5 mg/L, with refrigeration and cold stabilisation ranked as the two treatments causing the highest oxygen pickup. 'Low enrichment' treatments are practices like pumping, filtration, heat exchange and electrodialysis, and caused an uptake of up to 0.6 mg/L, with filtration causing the highest oxygen pickup (Castellari *et al*., 2004). During and after bottling, both wine and the gaseous headspace in the bottle will contain a substantial amount of oxygen. The total packaged oxygen, as the sum of these two components, typically can reach 1 to 9 mg/L (Ugliano *et al*., 2013), depending on the specific handling of the wine (care taken to exclude oxygen). In the case of some closures, such as corks or similar products, additional oxygen will be released into the bottle after bottling.

The solubility of oxygen in wine is influenced by the wine composition (*e.g.*, ethanol content), but depends primarily on the temperature and the partial pressure of the gas, with greater solubility at lower temperatures and when pure oxygen is used instead of air (Waterhouse & Laurie, 2006). The rate of the oxidation reactions, however, increases with increased temperature (Margalit, 1997; Vivas de Gaulejac *et al*., 2001; Ribéreau-Gayon *et al*., 2006). The contact of wine with oxygen can be minimised by the use of inert gasses such as nitrogen, carbon dioxide and even argon gas, which can displace the air in a tank or barrel.

Oxygen and phenolics

Phenolic compounds are characterised by an aromatic ring containing one or more hydroxyl groups (-OH). The concentration of phenolics in white wine will depend on the variety, cultivation conditions, climate, grape maturity, winemaking techniques as well as ageing conditions. The polyphenol content of white wines is substantially lower than that of red wines due to different winemaking techniques favouring phenolic extraction during red wine production. Phenolics are strong hydrogen-donating species, which makes them ideal oxidation substrates (Wilderandt & Singleton, 1974).

Phenolic molecules originating from grapes can be divided into non-flavonoids and flavonoids. The non-flavonoids are grape-derived and consist, among others, of hydroxybenzoic acids, hydroxycinnamic acids and stilbenes, and are normally the principal phenolic molecules in white wines. Compounds such as the caffeic acid, *p*-coumaric acid and ferulic acid and their tartaric acid esters are examples of hydroxycinnamic acids, and are the main phenolic molecules in white wine that did not receive prolonged periods of skin contact. Of these, *trans*-caftaric acid and the grape reaction product are the predominant hydroxycinnamate esters in grape juice and wine, together with smaller quantities of coutaric and fertaric acid (Singleton *et al*., 1978; 1984; Vrhovšek, 1998). The naturally occurring tartaric esters are susceptible to hydrolysis, liberating the corresponding free hydroxycinnamic acids. The hydroxybenzoic acids represent a minor class of white wine polyphenols, of which gallic acid, vanillic acid and syringic acid are a few examples.

The major classes of flavonoids in grapes and wine are flavan-3-ols, flavonols and anthocyanins. Anthocyanin pigments are significant only in red grape varieties and generally are absent from white wines. The main flavan-3-ols found in grapes are (+)-catechin and (-)-epicatechin, as well as the galate ester (-)-epicatechin-3-*O*-gallate (Tsai Su & Singleton, 1969), while the flavonols consist mainly of quercetin, kaempferol and myricetin, which are found mainly as glycosides in grapes and as aglycones in wine (Monagas *et al*., 2005; Ribéreau-Gayon *et al*., 2006).

In grape must, the enzymatic oxidation of phenolic compounds takes place due to the presence of oxidation enzymes (*e.g.*, polyphenol oxidase), which catalyse the oxidation process, while in wine, non-enzymatic chemical oxidation is the predominant oxidation reaction due to the absence or inhibition of oxidation enzymes. Although polyphenol oxidation is extremely slow (Oszmianski *et al*., 1985), it affects a wide range of polyphenols, depending on their individual redox potentials (Kilmartin *et al*., 2002). The oxidation process will depend on a number of factors, such as oxygen concentration, temperature, presence of catalysts, the nature and composition of polyphenols, pH, ethanol content and the presence of antioxidants (Berg & Akiyoshi, 1956; Cilliers & Singleton, 1989; Kilmartin *et al*., 2001; Waterhouse & Laurie, 2006).

Oxygen in wine is in the unreactive triplet state, and its ability to react directly with most wine components is low (Waterhouse & Laurie, 2006). The presence of a catalyst (particularly iron and copper) increases the reaction speed by donating an electron to oxygen, resulting in a superoxide ion that exists as a hydroperoxyl radical at wine pH (Fig. 2). The radical has relatively low reactivity in the wine environment and will react with strong hydrogen-donating species such as phenolic molecules (Wilderandt & Singleton, 1974). The reaction of the superoxide ion with *o*-diphenols forms H_2O_2 and *o*-quinones at wine pH (Fig. 2).

The H_2O_2 can react with ferrous ions via the Fenton reaction to produce a hydroxyl radical, which is extremely reactive and can react with various wine constituents (*e.g.*, alcohols, organic acids and sugars) in proportion to their concentration, causing the formation of aldehydes and ketones. In this case, ethanol will be oxidised primarily to produce acetaldehyde (Waterhouse & Laurie, 2006). The *o*-quinone is unstable and very reactive and can react further with other molecules with lower redox potentials, such as other phenolic molecules, SO_2 and thiol-containing compounds, including glutathione and amino acids. This array of reactions can cause a substantial change in the composition of wine.

The role of antioxidants

The most common antioxidants (other than phenols) present in wine are sulphur dioxide (SO_2), ascorbic acid and glutathione (GSH). These compounds interfere in the phenol oxidation process, either by removing oxygen from the wine or by reversing or altering the oxidation process.

Sulphur dioxide is an effective and low-cost additive for the preservation of wines and other food products (Doyle & Beuchat, 2007). In wine, it serves as the main preservative to prevent oxidation and decrease microbial activity. Even
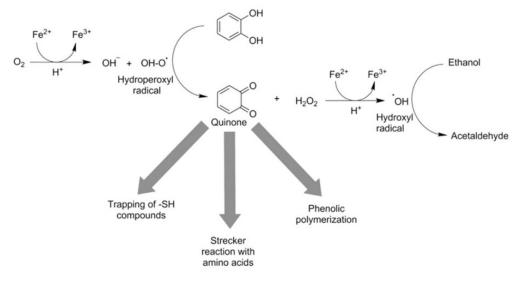
FIGURE 2

Formation of *o*-quinones and hydrogen peroxide and the consequent oxidation of ethanol to form acetaldehyde and other reactions in the wine matrix. Reprinted with permission from Ugliano (2013). Copyright American Chemical Society, 2013.

though SO_2 occurs naturally in all wines as a by-product of yeast metabolism during fermentation (Rankine & Pocock, 1969), it is usually added at several stages in the process of conventional winemaking, such as during crushing, settling or after primary and secondary fermentation (Paul, 1975). Unfortunately, excessive use of SO_2 can be detrimental not only to the organoleptic quality of a wine, but also poses a health risk for sensitive consumers (especially asthmatics), and its presence requires mandatory label warning statements in most jurisdictions (Kleinhans, 1982).

Sulphur dioxide exists in wine in both free and bound form (the sum equalling total SO_2). At wine pH (pH 3 to 4), free SO_2 can exist in three forms: molecular SO_2, bisulphite (HSO_3^-) and sulphite (SO_3^{2-}). The equilibrium is pH dependent and is affected by the presence of wine constituents that bind the bisulphite, as well as by wine temperature (Usseglio-Tomasset, 1992). The molecular form is responsible mainly for antimicrobial properties due to its ability to penetrate the cellular membranes of microorganisms (Beech *et al.*, 1979). However, at wine pH, only a small proportion of the free SO_2 is in molecular form, with the predominant form being bisulphite (94 to 99%), which can bind a large range of wine components, consequently producing bound SO_2 (Zoecklein *et al.*, 1995; Oliveira *et al.*, 2002). The sulphite ion can react directly with oxygen, but is present at extremely low concentrations at wine pH. Direct reaction of bisulphite with oxygen is slow, and the antioxidant activity lies in the ability to reduce the H_2O_2 to water, convert *o*-quinones back to *o*-diphenols and react directly with *o*-quinones to form sulphonic acids (Danilewicz, 2007).

A number of carbonyl compounds (mainly acetaldehyde, pyruvic acid and α-keto-glutaric acid) can bind with free SO_2 individually to form complex compounds (Jackowetz & De Orduña, 2013). The bisulphite-acetaldehyde addition product normally accounts for the majority of the bound SO_2 in wine. Reactions with carbonyl compounds such as acetaldehyde lead to the formation of bisulphite addition products in reversible reactions; however, other reactions,

such as those between bisulphite and *o*-quinones, form stable adducts and are irreversible (Laurie *et al.*, 2012). Nevertheless, there is a general trend in the wine industry towards minimising SO_2 content, and the use of appropriate substitutes and supplements is a popular topic in research. To date, no single replacement for SO_2 has been found that combines antimicrobial and antioxidant characteristics.

Glutathione is a sulphur-containing tripeptide (L-γ-glutamyl-L-cysteinyl-glycine) and a naturally occurring antioxidant from the grapes and yeast metabolism. The concentration in the must is influenced by the nitrogen uptake of the vine (Choné *et al.*, 2006) and it is accumulated in the berry at the onset of véraison (Adams & Liyanage, 1993). Concentrations of GSH in 28 young Sauvignon blanc wines averaged at 12.5 mg/L (Janeš *et al.*, 2010), and winemaking conditions promoting oxygen exposure led to a decrease in GSH concentrations, while higher concentrations have been observed in juices treated reductively (Du Toit *et al.*, 2007; Maggu *et al.*, 2007).

GSH has an electron-rich nucleophilic mercapto group that can be substituted spontaneously by 1,4-Michael addition to the electrophilic centre of the *o*-quinone formed during oxidation. The product is a thioether, 2-S-glutathionyl-caftaric acid or grape reaction product (GRP). The formation of GRP traps the *o*-quinone, thus preventing any further reactions taking place (Kritzinger *et al.*, 2013a). Glutathione can also react with oxygen species such as H_2O_2 (Anderson, 1998) to be oxidised to glutathione disulphide. It has been argued that the disulphide can also be formed by reducing the *o*-quinone back to the *o*-diphenol (Cilliers & Singleton, 1990). This may explain increasing *trans*-caftaric and coutaric acid concentrations in must after GSH additions, as reported by Penna *et al.* (2001).

Glutathione is also capable of carrying out nucleophilic reactions with other compounds such as aldehydes (Cheynier *et al.*, 1986; Cheynier & Van Hulst, 1988; Sonni *et al.*, 2011) and has protective abilities toward important aroma compounds such as esters, monoterpenes and volatile

thiols (Papadopoulou & Roussis, 2001; Lavigne-Cruège & Dubourdieu, 2003; Roussis *et al*., 2007; Papadopoulou & Roussis, 2008). However, GSH has also been shown to induce the production of H_2S during wine ageing under low-oxygen conditions (Ugliano *et al*., 2011).

Glutathione has been linked to the aroma potential of Sauvignon blanc grapes due to the identification of glutathionylated precursors for 3MH and 4MMP (Peyrot des Ganchos *et al*., 2002). The addition of GSH to must or wine is not legally permitted. However, yeast extracts (which contain GSH) are claimed to preserve the freshness and increase the mouthfeel, as well as the aromatic complexity, of white wines (Kritzinger *et al*., 2013b). These extracts are available commercially.

Ascorbic acid has been considered as a potential replacement for sulphur dioxide due to its ability to scavenge oxygen (Bauernfeind & Pinkert, 1970). The reaction of ascorbic acid with oxygen produces dehydroascorbic acid as well as H_2O_2. It thus is important to have sufficient SO_2 present to react with the formed H_2O_2 in order to prevent further oxidation reactions from taking place (Peng *et al*., 1998).

Ascorbic acid initially functions as an antioxidant; however, over time, the presence of ascorbic acid can lead to enhanced SO_2 consumption and oxidation of phenolic compounds, resulting in browning (Peng *et al*., 1998; Bradshaw *et al*., 2001; 2003; 2004; Clark *et al*., 2008). However, the addition of ascorbic acid to Chardonnay wines at bottling resulted in wines being less oxidised and/or having more fresh fruity aromas compared to wines bottled without ascorbic acid (Skouroumounis *et al*., 2005b).

Effect of oxygen on white wine colour
The colour of white wine is one of the important quality parameters. Dark yellow or brown colour usually indicates the oxidation or spoilage of white wine. A positive correlation between total phenolic content and browning has been reported (Simpson, 1982). However the content of hydroxycinnamic acids in the wines correlated poorly with browning (Fernandez-Zurbano *et al*., 1995). The hydroxycinnamic acids may, however, contribute to the browning through coupled oxidation reactions (Simpson, 1982; Fernandez-Zurbano *et al*., 1995). The monomeric flavan-3-ols and dimeric procyanidins play an important role in white wine colouration, with (-)-epicatechin correlating more strongly with the brown colour when compared to (+)-catechin (Simpson, 1982). Browning in white wine can be due to different mechanisms. Firstly, the oxidation of phenolic molecules to their corresponding *o*-quinones leads to further reactions with phenolic compounds to produce dimers, which appear to be more susceptible to oxidation, and thus accelerate phenol polymerisation and autocatalytic oxidation in wine (Singleton, 1987). The formation of these polymers can result in the formation of more intensely coloured yellow-brown compounds (Es-Safi *et al*., 1999).

The second mechanism is the oxidative degradation of tartaric acid leading to the formation of glyoxylic acid, which can mediate condensation reactions of flavan-3-ols, potentially contributing to browning in white wines.

SAUVIGNON BLANC AROMA AND EVOLUTION DURING AGEING AND OXIDATION
Vitis vinifera L. cv. Sauvignon blanc is a grape variety native to Bordeaux (Graves) and the Loire Valley (Sancerre and Pouilly Fumé) (Adams & Liyanage, 1993). This variety is now widely cultivated in many other wine-growing regions across the globe, including South Africa, Australia, New Zealand, Chile and the United States (Jackson, 2008). The grapes ripen to moderate to high levels of acidity, producing fresh, crisp and dry white wines with pungent aroma and flavour (Cooper, 2008). Generally, South African Sauvignon blanc wines are cold-fermented in stainless steel tanks and not exposed to oak. In this way, the terroir (often delivering fresh and fruity aromas) is reflected in the wine.

Depending on the climate, Sauvignon blanc wines can offer a range of wine styles. On the one hand, the wine can deliver fresh and fruity characters reminiscent of "guava", "grapefruit", "gooseberries" and "passion fruit", which usually originate from grapes grown in a warmer climate. On the other hand, the more "green" style Sauvignon blanc can be produced from cooler grape-growing regions delivering aroma nuances such as "green pepper", "grassy" and "asparagus" (Lund *et al.,* 2009b). The aroma of Sauvignon blanc is known to change dramatically over a period of just a year in the bottle, meaning that the wine should be drunk early to experience the intended aromatic bouquet of the wine (Herbst, 2010).

Various chemical compounds contribute to the aromatic composition of a wine. Sauvignon blanc impact compounds, such as the volatile thiols and the methoxypyrazines, can contribute significantly to the typical character of the wine. Other aroma groups, such as ester, alcohols, acids and monoterpenes, can also contribute to the pleasant wine aroma. Wines developing a 'reductive' aroma will typically have negative aroma descriptors due to low oxygen exposure, while various oxidation-related compounds such as aldehydes, lactones and acetals will contribute to an unpleasant aroma. In the following sections, details are provided of the most important aroma-contributing groups.

Volatile thiols
Thiols (more traditionally referred to as mercaptans) are sulphur-containing compounds possessing a sulfhydryl group (-SH). Various sulphur-containing compounds occur in wine and can contribute to a range of aromatic nuances, depending on the type of compound. Certain thiols typically contribute to the fruity characters of a wine, and attributes such as "grapefruit", "passion fruit", "box tree" and "gooseberry" have been used to describe the odour (Darriet *et al*., 1995; Tominaga *et al*., 1998a). These compounds are referred to as the volatile thiols.

Other sulphur-containing compounds can contribute to a 'reductive' aroma due to the low oxygen content in certain wines, and the aroma reminiscent of "rotten egg", "garlic" and "cabbage" occurs (Rauhut, 1993; Brajkovich *et al*., 2005). Compounds such as furfural and 5-hydroxymethylfurfural (5-HMF) (also sulphur-containing compounds) can also impart significant aroma to a wine.

Although first identified in Sauvignon blanc wine during the 1990s, the presence of volatile thiols has also

been reported in wines made from other varieties (both red and white), such as Riesling, Colombard, Semillon, Cabernet Sauvignon and Merlot (Bouchilloux *et al*., 1998; Tominaga *et al*., 2000; Murat *et al*., 2001b). Along with methoxypyrazines, volatile thiols are considered to be impact odorants for Sauvignon blanc wines due to the fact that they form part of the typicality of the variety and are highly sought after by consumers (Goniak & Noble, 1987; Rauhut, 1993; Tominaga *et al*., 2004; Lund *et al*., 2009b; King *et al*., 2011). The volatile thiols playing an important role in Sauvignon blanc wine aroma are mainly 4-mercapto-4-methylpentan-2-one (4MMP), 3-mercaptohexan-1-ol (3MH) and 3-mercaptohexan-1-ol acetate (3MHA) (Darriet *et al*., 1995; Tominaga *et al*., 1996; 2000; 2004; Coetzee & Du Toit, 2012). Other volatile thiols have been identified; however, the concentration of these compounds in wines is usually below the perception threshold (Tominaga *et al*., 1998a).

3MHA has been described as "sweet-sweaty passion fruit", "grapefruit", "box tree", "gooseberry" and "guava" (Tominaga *et al*., 1996; Nicolau *et al*., 2006) and has a very low perception threshold of 4.2 ng/L in a model wine solution (Tominaga *et al*., 1996). 3MH has been described as "passion fruit", "grapefruit", "gooseberry" and "guava" (Swiegers *et al*., 2005; Nicolau *et al*., 2006; Van Wyngaard *et al.,* 2014), with a perception threshold of 60 ng/L (Tominaga *et al*., 1998a), while 4MMP has been described as "box tree", "passion fruit", "broom" and "black current bud" (Darriet *et al*., 1995), with a perception threshold of 0.8 ng/L in a model wine solution (Tominaga *et al*., 1998a). The low perception thresholds reported makes these compounds potent aromatic contributors in wine.

In a study done on 24 South African Sauvignon blanc wines (2011 vintage), the average thiol concentrations were found to be 10 ng/L, 158 ng/L and 969 ng/L for 4MMP, 3MHA and 3MH respectively (Van Wyngaard, 2013). These concentrations were in line with those found in other studies investigating Sauvignon blanc wines from all over the world, including New Zealand, Australia, France and Chile (Benkwitz *et al*., 2012b), with the exception of 3MH, for which the concentrations were lower (Lund *et al*., 2009b; Benkwitz *et al*., 2012b).

Unlike the methoxypyrazines, which are present as such in the grapes, the volatile thiols (4MMP and 3MH) are thought to be released in part by the yeast from odourless, non-volatile precursors during fermentation (Darriet *et al*., 1995; Tominaga *et al*., 1996). Some precursors in the juice have been identified as being cysteinylated or glutathionylated precursors (Tominaga *et al*., 1998b; Peyrot des Ganchos *et al*., 2002; Fedrizzi *et al*., 2009). However, studies have shown that these precursors account for only a fraction of the total amount of thiols present in the wine (Subileau *et al*., 2008a; Roland *et al*., 2010a; 2010b), and no direct correlation between precursor concentration and the amount of free volatile thiols in the resulting wines has been observed (Capone *et al*., 2010; Roland *et al*., 2011; Pinu *et al*., 2012). Other than release from precursors, the biogenesis of volatile thiols also has been proposed. The direct addition of H_2S or another sulphur donor to conjugated carbonyl compounds such as *(E)*-2-hexenal and

mesityl oxide, followed by a reduction step, has been shown to contribute significantly to the pool of volatile thiols in Sauvignon blanc wines (Harsch *et al*., 2013). The formation of the volatile thiols is not fully understood, since the main precursor has yet to be elucidated and the production of these volatile thiols needs further investigation.

3MHA is formed by the esterification of 3MH with acetic acid during fermentation. The final concentration of 3MHA (and other fermentative esters) depends on the balance of activities of alcohol acetyltransferase (promoting estrification of the corresponding alcohol) and esterase (promoting hydrolysis). Yeast strains differ in their ability to release the volatile thiols from their precursors, and also in their ability to convert 3MH to 3MHA (Murat *et al*., 2001a; Swiegers *et al*., 2005; 2006; Anfang *et al*., 2009). 3MHA typically amounts to 10% of the concentration of 3MH (Tominaga *et al*., 2000; Dubourdieu *et al*., 2006).

The formation of volatile thiols can be manipulated through both viticultural and oenological operations. Research on the impact of viticultural practices on the production of volatile thiol precursors is limited and under question, due to the main mechanisms of volatile thiol formation from precursors not yet being identified. The effect of factors such as nitrogen fertilisation, water availability and *Botrytis cinerea* infection on certain precursors has been reported (Choné, 2001; Peyrot des Ganchos *et al*., 2005; Choné *et al*., 2006; Sarrazin *et al*., 2007; Thibon *et al*., 2009). During winemaking, various processes can also be adapted to maximise precursor extraction and/or formation, as well the liberation of the thiol from the precursor. Practices such as mechanical harvesting, skin contact, pressure during pressing and oxygen exposure could significantly increase the precursor content in the juice and influence the volatile thiol concentration in the corresponding wines (Maggu *et al*., 2007; Patel *et al*., 2010; Roland *et al*., 2010b; Capone & Jeffery, 2011). The liberation of the volatile thiols from the respective precursors depends on conditions such as yeast strain, nitrogen availability and fermentation temperature (Murat *et al*., 2001a; Masneuf-Pomarède *et al*., 2006; Swiegers *et al*., 2006; Subileau *et al*., 2008b; Anfang *et al*., 2009; Harsch *et al*., 2013). More detail regarding the influence of these processes on the volatile thiol concentrations can be obtained from a review article published in 2012 (Coetzee & Du Toit, 2012).

The volatile thiols are particularly susceptible to oxidation during ageing (Murat *et al*., 2003; Blanchard *et al*., 2004), and various packaging and ageing studies have reported the decrease of the volatile thiols during storage (Murat, 2005; Lopes *et al*., 2009; Herbst-Johnstone *et al*., 2011; Ghidossi *et al*., 2012). The volatile thiols 3MH and 3MHA were also found to mainly decrease in Sauvignon blanc wines that were exposed to repetitive oxidation (Coetzee, 2014). Three mechanisms have been identified through which the thiol content can decrease. Volatile thiols can oxidise easily in the presence of oxygen and iron to form the corresponding disulphides (Jocelyn, 1972; Kotseridis *et al*., 2000). Furthermore, these thiols are nucleophilic and capable of addition reactions with electrophiles such as polymeric phenolic compounds (Ribéreau-Gayon *et al*., 2006), and participate in chemical reactions (1,4-Michael-

type addition) with the products of phenolic oxidation such as *o*-quinones (Herbst *et al*., 2008; Nikolantonaki *et al*., 2010). The formed adducts are non-volatile and would cause a loss in varietal character. Recent observations have reported *o*-quinone trapping as the main mechanism accounting for 3MH loss in wine under oxidative conditions, while other reactions seems to contribute marginally (Kreitman *et al*., 2013).

The rate of these reactions are pH dependent, since the concentration of the thiolate ion (RS⁻), which is more reactive than its protonated form (RSH), is low at wine pH, with pK_a values for thiols typically being between 9 and 12. The greater proportion of thiolate anions at higher pH may explain the consistently lower levels of 3MH under these conditions (Blanchard *et al*., 2004).

The decrease in especially 3MHA can also be attributed to acid-catalysed hydrolysis at wine pH (Tominaga *et al*., 2004). The ester structure of 3MHA makes it susceptible to this type of reaction. Volatile acetate esters are introduced enzymatically to the wine during fermentation by the action of yeast via a combination of acetyl-CoA with an alcohol, which is catalysed by alcohol acetyltransferase. The products of this reaction are 3MH and acetic acid. The hydrolysis reaction is expected to be accelerated by higher temperature, but not directly affected by oxidative conditions (Makhotkina & Kilmartin, 2012).

Acid hydrolysis was found to have a stronger influence than *o*-quinone trapping on the decrease in 3MHA concentrations during bottle ageing of Sauvignon blanc wine under highly hermetic conditions such as screw caps (Herbst *et al*., 2008; Herbst-Johnstone *et al*., 2011). As 3MH has a higher perception threshold than 3MHA, the hydrolysis is expected to result in lower aromatic intensity and possibly a different aroma.

The presence of SO_2 was shown to prevent the loss of volatile thiols due to its ability to recycle the *o*-quinones to the original phenol, bind them directly, or reduce H_2O_2 to water (Danilewicz *et al*., 2008; Nikolantonaki *et al*., 2010; Laurie *et al*., 2012). However, the formation of the *o*-quinone adducts was shown to occur even in the presence of SO_2, although to a lower extent (Nikolantonaki *et al*., 2012).

Methoxypyrazines
Methoxypyrazines are secondary plant metabolites with nitrogen-containing ring structures that are responsible for aroma descriptors such as "green pepper", "asparagus", "grassy", "herbaceous" and "vegetative". Three main methoxypyrazines occur in wines, namely 3-isobutyl-2-methoxypyrazine (IBMP), 3-isopropyl-2-methoxypyrazine and 3-*sec*-butyl-2-methoxypyrazine (Allen *et al*., 1991; Lacey *et al*., 1991; Marais, 1994).

These compounds have a very low odour threshold value of 1 to 2 ng/L in water (Buttery *et al*., 1969; Allen *et al*., 1991; Lacey *et al*., 1991; Marais, 1994), and surveys done on South African Sauvignon blanc wines reported the IBMP (the most important methoxypyrazine) concentrations to be in the range of 1.2 to 40 ng/L (Alberts *et al*., 2009; Van Wyngaard, 2013).

Settling of the juice before fermentation can lead to a decrease in methoxypyrazine concentrations (Kotseridis

et al., 2008), while concentrations have been reported to remain stable during fermentation (Sala *et al*., 2004). The compounds are generally resilient to standard wine fining practices (Pickering *et al*., 2006), and concentrations did not differ when exposed to various ageing conditions, such as light exposure and temperature variations (Blake *et al*., 2010). Even hyperoxidation of the musts and wine did not alter the methoxypyrazine content (Marais, 1998; Coetzee *et al*., 2013). Surprisingly, a weak correlation ($r^2 = 0.37$) was obtained when comparing IBMP concentration with the aromatic intensity of the "capsicum" attribute (Lund *et al*., 2009b). The contribution of other aromatic compounds, such as the varietal thiols, C6-alcohols and dimethyl sulphide, to these types of attributes can be significant in Sauvignon blanc wines and help explain the weak correlation observed previously (Park *et al*., 1994; Marais *et al*., 1998; Lund *et al*., 2009b; Kotseridis *et al*., 2000; King *et al*., 2011; Van Wyngaard, 2013; Coetzee *et al*., 2015a).

Esters, higher alcohols, fatty acids
Esters constitute one of the most important classes of aroma compounds and are largely responsible for the fruity aromas associated with wine (Lilly *et al*., 2000; Swiegers *et al*., 2006). Esters are mainly produced during fermentation through the condensation of an alcohol and a coenzyme-A-activated acid by the action of alcohol acetyl transferase (Lambrechts & Pretorius, 2000). A large variety of esters can be formed, as all of the alcohols (especially ethanol) and fatty acids may react to form esters. Acetate esters of the higher alcohols and the ethyl esters of straight-chain saturated fatty acids are the most significant esters produced in wine (Lambrechts & Pretorius, 2000). Some of the most quantitatively significant esters in wine have been identified to be isoamyl acetate, ethyl hexanoate and 2-phenylethyl acetate (Thurston *et al*., 1981).

Ester concentration can decrease during ageing due to chemical hydrolysis (Marais, 1978; Ferreira *et al*., 1997; Lambropoulos & Roussis, 2007) or oxidation due to direct attack by hydroxyl radicals or by ester interaction with *o*-quinones (Escudero *et al*., 2000a; Patrianakou & *et al*., 2013), which can lead to a loss of the fruity character of a wine. The hydrolysis is favoured at elevated temperatures and low pH values (Ramey & Ough, 1980). The decline in the acetate esters in particular contributes to the loss of freshness and fruitiness in white wines during bottle ageing. Acetate esters of higher alcohols tend to diminish more rapidly during ageing compared to ethyl esters of fatty acids. Ethyl esters are thought to be close to their chemical equilibrium in young wines (Simpson, 1978), and the hydrolysis that occurs during storage happens relatively slowly (Ramey & Ough, 1980).

Higher alcohols are also a product of alcoholic fermentation and can be important precursors for ester formation (Soles *et al*., 1982). At concentrations below 300 mg/L they generally contribute to the complexity of the wine aroma (Rapp & Mandery, 1986). However, at higher concentrations the aroma can become too intense and can contribute to a strong, pungent smell and taste (Nykänen, 1986), thus possibly masking other aroma contributors. Higher alcohols can be anabolically synthesised from

intermediates of the sugar metabolism or catabolically synthesised from branched-chain amino acids through the Ehrlich pathway (Nykänen, 1986; Boulton *et al.*, 1996; Dickinson *et al.*, 1997; 2003). During ageing, alcohols can be oxidised to form aldehydes (Marais & Pool, 1980), causing the concentration to decrease. However, many studies report stable alcohol concentrations during the ageing of wines (Marais, 1978; Roussis *et al.*, 2005; 2007; Blake *et al.*, 2009). Unlike other higher alcohols, the concentration of hexanol can increase during storage, which is probably due to the oxidation of linoleic and linolenic acids (Oliveira *et al.*, 2006).

Fatty acids, of which the most abundant are acetic, hexanoic, octanoic and decanoic acid, contribute to the fresh flavour of wine. However, at very high concentrations of fatty acids, unwanted flavours described as "rancid", "cheesy" and "vinegar" can develop (Schreier, 1979; Lambrechts & Pretorius, 2000). Medium-chain fatty acids, such as hexanoic, octanoic and decanoic acid, are produced by the yeast as intermediates in the biosynthesis of long-chain fatty acids. The hydrolysis of ethyl esters during ageing can lead to an increase in the corresponding acid. However, an increase in these acids has not always been observed, as studies have shown that the stability of volatile fatty acids is not uniform, as some compounds increase while others decrease or remain stable during ageing (Roussis *et al.*, 2005; Câmara *et al.*, 2006; Blake *et al.*, 2009; Lee *et al.*, 2011).

Many factors can influence the formation and degradation of esters, higher alcohols and fatty acids during fermentation, ageing or oxidation. Fermentation conditions such as temperature, juice clarification and yeast strain, as well as other parameters such as oxygen exposure and SO_2 additions, are some of the most important factors (Bertrand, 1968; Daudt & Ough, 1973; Marais, 2001; Garde-Cerdán & Ancín-Azpilicueta, 2007; Coetzee *et al.*, 2013).

Monoterpenes

Monoterpenes are known for their "floral", "fruity", "citrus" and "perfume" odours, usually expressed by geraniol, linalool, nerol and α-terpineol (Marais, 1983). Sauvignon blanc can be classified as a member of an intermediate class between monoterpene-dependent floral grapes and monoterpene-deficient non-floral grapes (Benkwitz *et al.*, 2012a). Most terpenes increase during ripening (Bayonove & Cordonnier, 1971), with some studies reporting a decrease at the overripe stage (Versini *et al.*, 1981). A considerable proportion of these compounds are in the bound form in the juice and are released during fermentation by the yeast; however, some can become aromatic by chemical rearrangement such as the non-enzymatic rearrangement of hydroxylated linalool derivatives to give several volatile terpenes, including α-terpineol (Williams *et al.*, 1980; Loscos, 2007). The oxidation mechanism is known to play a role in some cases in the release and rearrangement of the terpenes (Williams *et al.*, 1982; Marais, 1983).

During ageing, wines are known to lose some of the floral aromas associated with monoterpenes (Rapp & Mandery, 1986; Rapp, 1988). Linalool specifically is known to decrease during storage (Ferreira *et al.*, 1997; Lambropoulos & Roussis, 2007), while α-terpineol initially

increased (probably due to the oxidation of other terpenols) and then decreased at a later stage (Ferreira *et al.*, 1997). Terpenes are sensitive to acidic conditions, storage time and temperature and can be transformed into other compounds, which could contribute to a different aroma and have different perception thresholds (Papadopoulou & Roussis, 2001; Roussis *et al.*, 2005; 2007). The importance of monoterpenes in wines is accentuated by the fact that these compounds act synergistically in a wine medium and thus can influence the aromatic composition of a wine (Ribéreau-Gayon *et al.*, 1975).

REDUCTIVE AROMA

'Reductive' aroma properties have been described as "rotten egg", "cabbage" and "garlic". The compounds responsible for these aroma attributes usually form after a period in the bottle. The occurrence of these odours has been attributed to the presence of low molecular weight sulphur compounds. Hydrogen sulphide (H_2S) and methyl mercaptan (MeSH) have been identified as being primarily responsible for post-bottling reduction (Lopes *et al.*, 2009; O'Brien *et al.*, 2009; Ugliano *et al.*, 2011; 2012). H_2S can accumulate during bottle ageing, and concentrations three to four times higher than its perception threshold have been reported after six months in the bottle (Ugliano *et al.*, 2011).

The mechanisms for the formation of the 'reductive' compounds are not fully understood. However, the formation of H_2S from a cysteine precursor in the presence of a dicarbonyl compound and the direct reduction of sulphate or sulphite have been suggested (Pripis-Nicolau *et al.*, 2000; Lopes *et al.*, 2009). Interestingly, studies have shown an increase in the formation of 'reductive' odours in wines that were treated with copper sulphite at bottling, which is a common practice amongst winemakers to prevent the formation of these odours (Ugliano *et al.*, 2011). The addition of glutathione to Sauvignon blanc wine also led to increased levels of H_2S, although the preservation effect of GSH addition on 3MH concentrations was more favourable (Ugliano *et al.*, 2011).

H_2S also can participate in other reactions, such as the reaction with benzaldehyde to form methyl mercaptan, which can contribute to a 'reductive' aroma in the wine, with "smoky/empyreumatic", "cabbage" and "sewage" nuances. However, this has not been demonstrated conclusively (Tominaga, 2003; Tominaga *et al.*, 2003; Ugliano, 2013).

Keeping oxygen levels low during bottle ageing led to a larger accumulation of H_2S and MeSH (Lopes *et al.*, 2009; Ugliano *et al.*, 2011; 2012), with the accumulation of MeSH occurring mainly in the first six to 12 months of bottle storage (Ugliano *et al.*, 2012). Dimethyl sulphide (DMS) also increases during bottle ageing and can make an important contribution to wine aroma (Segurel *et al.*, 2005; Escudero *et al.*, 2007). The formation of DMS does not seem to be affected by oxygen exposure (Ugliano *et al.*, 2012), although a decrease in DMS has been reported in the presence of excess oxygen (Silva Ferreira *et al.*, 2003c; Fedrizzi *et al.*, 2011). Oxygen exposure also will lead to a decrease in H_2S and MeSH concentrations. The oxidation of mercaptans to the corresponding disulphides has been proposed, but not proven (Limmer, 2005), and

recent data have not found a direct relationship between the mercaptan and the disulphides (Nguyen *et al*., 2010; Ugliano *et al*., 2012). The reaction of 'reductive' compounds with *o*-quinones resulting from phenolic oxidation could explain the decrease of especially H_2S and MeSH in an oxygenated wine due to the high reactivity of H_2S toward *o*-quinones in wine-like solutions (Nikolantonaki & Waterhouse, 2012). The presence of oxygen during the early stages of bottle ageing thus will prevent the formation of these compounds, possibly at a precursor level. However, this needs further investigation (Ugliano, 2013).

OXIDATION AROMA

Early work on the oxidative spoilage of white wines indicated that oxidation brought about sensory characters described as "honey", "farmfeed", "woody" and "cooked vegetables" (Escudero *et al*., 2002; Silva Ferreira *et al*., 2003b). A number of trace aroma compounds have been identified as major contributors to these aroma attributes, including various aldehydes, lactones and acetals (Escudero *et al*., 2000a; 2000b; 2002; Silva Ferreira *et al*., 2002b; 2003b). The aldehydes are especially important because of their possible impact on wine aromatic composition (Escudero *et al*., 2000b; Silva Ferreira *et al*., 2003b; Culleré *et al*., 2007). The various chemical compounds potentially contributing to wine oxidation aromas will be discussed in the following sections.

Aldehydes

Acetaldehyde is one of the most important carbonyl compounds in wine and constitutes up to 90% of the total amount of aldehydes found in wine (Nykänen, 1986). Its organoleptic influence and its ability to combine rapidly with SO_2, even at low temperatures, makes this compound one of the critical markers during winemaking (Burroughs & Sparks, 1973). Acetaldehyde in the free form is a key impact aroma compound for sherry wines, and can reach high concentrations in these types of wines (Martínez *et al*., 1998). Odours associated with the presence of free acetaldehyde have been described as "green apple", "overripe bruised apple", "grassy", "pungent", "nutty" and "sherry" (Margalith, 1981; Henschke & Jiranek, 1993; Miyake & Shibamoto, 1993; Frivik & Ebeler, 2003).

Acetaldehyde is formed by the yeast during alcoholic fermentation (Margalit, 1997) and can also originate from the microbial activity of other microbes, such as lactic acid bacteria and acetic acid bacteria (Drysdale & Fleet, 1988; Liu & Pilone, 2000). However, winemaking practices that enhance acetaldehyde formation are usually post-fermentation, and can lead to moderate to important acetaldehyde increases (Jackowetz & De Orduña, 2013).

The most important production of acetaldehyde is during the oxidation of ethanol (Wilderandt & Singleton, 1974; Ribéreau-Gayon, *et al*., 2006). This reaction is not direct, but rather via the coupled auto-oxidation of certain phenolic compounds (Fig. 2) (Wilderandt & Singleton, 1974), and the formation will depend on the amount of oxygen present (Kielhöfer & Würdig, 1960; Schneider, 2003; 2005).

Acetaldehyde concentration (free and bound) in dry white wines has been reported to range between 7 and 240 mg/L, averaging at 40 mg/L (Lopes *et al*., 2009; Jackowetz & De Orduña, 2013), while the highest concentrations were reported in fortified wines ranging from 12 to 800 mg/L (Lachenmeier & Sohnuis, 2008). Coetzee (2014) found levels to increase gradually, reaching up to 100 mg/L total acetaldehyde in a Sauvignon blanc wine exposed to repetitive oxidation.

Other aldehydes can contribute to "honey", "boiled vegetable" and "rotten potato" nuances in oxidised wines (Escudero *et al*., 2000b; Culleré *et al*., 2007). Of these aldehydes, the Strecker aldehydes, 3-(methylthio)-propionaldehyde (methional) and phenylacetaldehyde, have been identified as major contributors (Escudero *et al*., 2000a; 2000b; 2002; Silva Ferreira *et al*., 2002b; 2003b). The formation of these aldehydes has been proposed to occur via the Strecker reaction of dicarbonyl compounds with amino acids, methionine and phenylalanine to form methional and phenylacetaldehyde respectively (Fig. 3) (Escudero *et al*., 2000b; Pripis-Nicolau *et al*., 2000; Silva Ferreira *et al*., 2002b; Rizzi, 2006).

The formation of aldehydes from the oxidation of related fermentation-derived alcohols (phenylethanol and methionol) has also been suggested (Escudero *et al*., 2000a; 2000b; Pripis-Nicolau *et al*., 2000; Silva Ferreira *et al*., 2002b; Rizzi, 2006; Loscos *et al*., 2010; Fedrizzi *et al*., 2011; Nikolantonaki & Waterhouse, 2012). Methional and phenylacetaldehyde have perception thresholds of 0.5 µg/L and 1 µg/L in a synthetic wine medium respectively (Escudero *et al*., 2000b; Culleré *et al*., 2007), and concentrations in a young Sauvignon blanc wine just after bottling were found to be below 0.5 and 5 µg/L respectively. After 18 months ageing at 20°C, the concentrations increased to about 5.5 and 34 µg/L respectively (Ghidossi *et al*., 2012). Higher concentrations of these compounds in various white wines have been reported in more extreme storage conditions, such as higher dissolved oxygen concentrations and higher storage temperatures (Silva Ferreira *et al*., 2003b).

The formation of methional and phenylacetaldehyde are greatly affected by temperature and by the concentration of dissolved oxygen (Silva Ferreira *et al*., 2002b). However, both factors are not required for the formation of the aldehydes, as it was found in beer that the aldehydes formed in the absence of oxygen (< 0.2 mg/L) (Soares da Costa *et al*., 2004). The oxidation of the alcohols, as well as the Strecker degradation reaction, will be prevented by the presence of SO_2 (Silva Ferreira *et al*., 2003b), probably due to its ability to reduce H_2O_2 to water, convert the *o*-quinone back to the *o*-diphenol and also reversibly reacting with the microbial-derived dicarbonyls. Thus, SO_2 should effectively be able to prevent the formation of these aldehydes.

Furfural and other furanic aldehydes, such as 5-methylfurfural (5-MF) and 5-hydroxymethylfurfural (5-HMF), usually originate from the heating process of oak during barrel toasting (Moutounet *et al*., 1989). The thermal degradation of polysaccharides produces these furanic aldehydes from carbohydrate polymers. The formation of furfural due to the non-oxidative decay of ascorbic acid has also been reported (Bauernfeind & Pinkert, 1970; Yuan & Chen, 1998; Wallington *et al*., 2013). A good correlation between ageing time and the concentration of these furanic
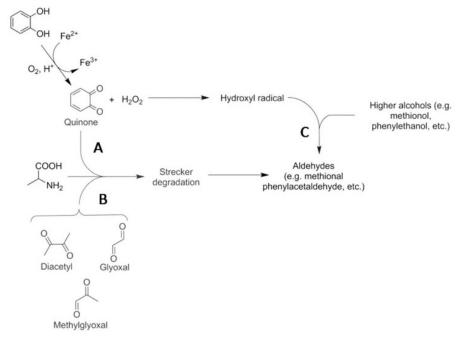
FIGURE 3

Mechanism of formation of branched-chain aldehydes in wines via (A) Strecker degradation of amino acids involving *o*-quinones derived from phenolic oxidation, (B) Strecker degradation involving dicarbonyl compounds of microbial origin, and (C) oxidation of higher alcohols by hydroxyl radical. Reprinted with permission from Ugliano (2013). Copyright American Chemical Society, 2013.

compounds was found in Port and Madeira wines (Silva Ferreira *et al*., 2003a; Câmara *et al*., 2004), and a significant increase is observed in Sauvignon blanc wines aged without oxygen, and even more so in Sauvignon blanc wine to which oxygen was administered at bottling (Coetzee, 2014). The concentrations of furfural and 5-MF in a range of white wines oxidised for one week were found to be between 16 and 342 µg/L and 10 and 60 µg/L respectively (Escudero *et al*., 2002). Perception thresholds of 150 mg/L, 20 mg/L and 100 mg/L have been reported for furfural, 5-MF and 5-HMF respectively (Meilgaard, 1975; Câmara *et al*., 2004), which would suggest minor organoleptic contribution due to the concentrations being below the perception threshold (Ribéreau-Gayon *et al*., 2006). However, the contribution of these compounds to "woody" or "maderized" nuances during the oxidation of white wines has been reported and could explain the perception of these odours in wines that did not receive any wood contact (Escudero *et al*., 2002; Campo *et al*., 2008).

Benzaldehyde is always present in wine and is produced by alcoholic fermentation in concentrations up to 0.5 mg/L. During wine ageing, benzaldehyde formation has been attributed to phenylalanine oxidation (Loyaux *et al*., 1981), while other authors have reported its origin from amygdalin (Nykänen & Suomalainen, 1982). The concentration of benzaldehyde in a range of white wines oxidised for one week was found to be between 20 and 313 µg/L (Escudero *et al*., 2002), and the compound correlated well with the term "liquor" (Escudero *et al*., 2002). The odour detection threshold for benzaldehyde is 0.35 mg/L in water (Belitz *et al*., 2009).

Other carbonyl compounds and acetals

Sotolon is a chiral furanone (lactone) and a very powerful odorant that has been described to have the odour of "curry" and "myrrh" at high concentrations, with "roasting", "maple syrup", "burnt sugar" and "caramel" at lower concentrations (Escudero *et al*., 2000a; Ghidossi *et al*., 2012). Sotolon has been demonstrated to be a key odorant in sherry, port, "vin jaune", and botrytised and "Tokai wines" (Masuda *et al*., 1984; Martin & Etiévant, 1991; Guichard *et al*., 1992; Martin *et al*., 1992; Silva Ferreira *et al*., 2003a; Collin *et al*., 2012), but can have a detrimental effect when present in dry white wines (Escudero *et al*., 2002; Silva Ferreira *et al*., 2003b; Lavigne *et al*., 2008).

The mechanism for the formation of this compound is still not fully understood, although a few pathways have been suggested and the connection between oxidation and sotolon formation seems to be evident (Pham *et al*., 1995; Cutzach *et al*., 1999; Silva Ferreira *et al*., 2003a; 2003c; Escudero *et al*., 2011). The formation of sotolon can occur via the enzymatic or chemical deamination of threonine, followed by aldol condensation with acetaldehyde in various wine media (Takahashi *et al*., 1976; Pham *et al*., 1995; Cutzach *et al*., 1998). Sotolon can also be formed from the oxidative degradation of ascorbic acid in the presence of ethanol (König *et al*., 1999; Pons *et al*., 2010). This mechanism is most likely to occur in dry white wines, especially considering the common practice of adding ascorbic acid to Sauvignon blanc must/wine during winemaking.

The concentrations of this compound in dry white wines are much less compared to other alcoholic beverages (fortified wine, port, sherry) (Salmon *et al*., 1999). Concentrations in

young Sauvignon blanc wines just after bottling were found to be below 0.5 µg/L, while Sauvignon blanc wine stored under various types of closures during a 24-month period had a sotolon content ranging from 0.1 to 1.1 µg/L (Lopes *et al.*, 2009; Ghidossi *et al.*, 2012). Another study measured concentrations of up to 10 µg/L for Sauvignon blanc wines aged between two and 32 years (Lavigne *et al.*, 2008), while a recent study found concentrations below 0.5 µg/L in a Sauvignon blanc wine to which 30 mg/L of oxygen was administered during the ageing process (Coetzee, 2014). The formation of this compound in white wines seems to be somewhat dependent on temperature, and a concentration of around 8 µg/L was obtained during a forced aged experiment conducted at 60°C (Silva Ferreira *et al.*, 2003b). The odour threshold for this compound has been determined to be 15 µg/L in flor sherry (Martin *et al.*, 1992), 2 µg/L in model wine (Pons *et al.*, 2010) and 8 µg/L in dry white wines (Lavigne *et al.*, 2008).

Acetals are formed by the condensation reaction between glycerol and acetaldehyde in an acid medium. Four isomers can be formed via this reaction: *cis-* and *trans*-5-hydroxy-2-methyl-1,3-dioxane, and *cis-* and *trans*-4-hydroxymethyl-2-methyl-1,3-dioxolane. In Port wine, isomers of glycerol and acetaldehyde acetals have been found at total concentrations ranging from 9.4 to 175.3 mg/L (Silva Ferreira *et al.*, 2002a). During the oxidation of wine, large amounts of the dioxanes and dioxolanes are expected to be produced due to the fact that an ample quantity of substrates is available. Dioxane and dioxolane content in an oxidised Sauvignon blanc wine increased significantly during a seven-month ageing period (Coetzee, 2014).

The aromatic impact of these compounds could contribute significantly to the oxidised odour perceived in some wines as "sweet" and "old port-like" (Silva Ferreira *et al.*, 2002a).

SENSORY SCIENCE
Evaluating the chemical composition of a wine can deliver valuable information such as the type and quantity of specific aromatic compounds present. However, the use of the chemical composition only is not adequate to predict the aroma profile and wine quality. This is important in a complex medium such as wine, where interactions between all compounds present could influence the sensory perception of the aroma. Sensory analysis (especially descriptive analysis) of wines thus is of critical importance for the evaluation of wine aroma (Stone & Sidel, 1993; Lawless & Heymann, 1998). The combination of the chemical and sensory data could deliver valuable information concerning the aroma profile of the wine and how various compounds interact. Descriptive analysis has been used successfully as a tool to differentiate between Sauvignon blanc wines from different vintages and countries (Lund *et al.*, 2009b; Green *et al.*, 2011). These techniques have also been used to investigate interactions between various compounds in Sauvignon blanc wine (King *et al.*, 2011; Benkwitz *et al.*, 2012a; Coetzee *et al.*, 2015a; 2015b), as well as to identify off-flavours in oxidation-spoiled white wines (Silva Ferreira *et al.*, 2003b).

Sensory interaction studies
Wine is a complex medium containing various types of aroma compounds that can all have an effect (differing degrees of suppression and masking) on each other (Francis & Newton, 2005). This complicates the investigation of interactive effects between compounds. Over the years, researchers have been trying to understand the interactions of aroma compounds in various media, as different compounds can manifest differently in various media (Maga, 1989; Marais & Swart, 1999; Campo *et al.*, 2005; Escudero *et al.*, 2007).

Interactive effects between aroma impact compounds that are found especially in Sauvignon blanc wines have been investigated. Combinations containing high concentrations of 4MMP, 3MH and 3MHA not only delivered high intensity ratings for the "cat urine/sweaty" attribute, but also for the "cooked green vegetal" descriptor, thus demonstrating the contribution of the thiols to the 'green' characteristics of a wine (King *et al.*, 2011). Synergism between IBMP and 4MMP has also been reported. When added individually at low concentrations (2 ng/L for IBMP; 0.2 ng/L for 4MMP), "dusty" was the only attribute generated. However, when added together at the same concentration, the amount of attributes generated by the panel increased to "dusty", "grassy" and "herbaceous" (Marais & Swart, 1999). When added at higher concentrations, a mutual suppression between the volatile thiols and the methoxypyrazines was observed (Marais & Swart, 1999; Campo *et al.*, 2005; King *et al.*, 2011; Van Wyngaard *et al.*, 2014). The addition of esters to the base wine increased the perception of the "confectionary" attribute and also increased the ratings of "overall fruit aroma", "tropical" and "cooked green vegetal" attributes (attributes also associated with thiols). Esters are known to be particularly important in the bouquet of young white wines, including Sauvignon blanc, and interact in an additive manner (Van der Merwe & Van Wyk, 1981; Campo *et al.*, 2005). High thiol concentrations in the presence of an ester combination decreased the ratings for "confectionary", indicating a suppressive effect of the thiols on the esters when present at high levels. In contrast, in the presence of moderate to high thiol concentrations, the esters actually enhanced some thiol-related sensory attributes (Ferreira *et al.*, 2002; Campo *et al.*, 2005; King *et al.*, 2011; Benkwitz *et al.*, 2012a). Overall, the addition of IBMP had a greater impact on the aroma of a wine when compared to the addition of the thiols. The effect of IBMP has been described as being dominant and, in wines containing both thiols and IBMP, the aroma is often driven by IBMP (Hein *et al.*, 2009; King *et al.*, 2011; Van Wyngaard *et al.*, 2014).

Lund *et al.* (2009a) investigated the interaction of non-volatile wine compounds, such as polyphenols and volatile aroma compounds, in a diluted wine medium. They found that the perception of IBMP, 3MH and ethyl decanoate was largely suppressed by the addition of polyphenols ((+)-catechin, quercetin and caffeic acid). On the other hand, the addition of specifically caffeic acid enhanced the perception of 3MH. This could be due to the suppression of other aroma compounds that initially masked the 3MH aroma (Lund *et al.*, 2009a). The aroma perception of 3MHA was least affected by the polyphenol additions when compared to the other aroma compounds tested in the study.

The presence of the ester functional group is thought to make it less susceptible to interaction with the polyphenol when compared to the alcohol group of 3MH (Lund *et al*., 2009a).

Very few studies have investigated the interactive effect of unpleasant aroma compounds, such as compounds related to oxidation. The addition of branched aliphatic aldehydes and (E)-2-alkenals generated attributes such as "sweet orange", "dried fruit", "fusel", "closed room", "dirty", "mouldy" and "rancid", and an additive effect was observed between these aldehydes (Culleré *et al*., 2007). An in-depth investigation by Coetzee *et al*. (2015a) on sensory interactions between 3MH, IBMP, methional and phenylacetaldehyde when combined at different levels yielded interesting results. Methional, especially at certain levels, had a strong suppressive effect on the "grapefruit" and "guava" attributes linked to 3MH. Methional and IBMP, however, enhanced the intensity of "cooked potato" and "cooked beans". In a similar study, Coetzee *et al*. (2015b) found acetaldehyde at lower levels to enhance the fruity aromas associated with 3MH, but suppression occurred at higher levels of acetaldehyde. Higher levels of 3MH might also suppress the "oxidised green apple" often associated with acetaldehyde. Acetaldehyde can also supress the green pepper attribute associated with IBMP. The need to preserve the positive aroma compounds, such as "guava", "passion fruit", etc., while controlling the formation of negative aroma attributes associated with oxidation derived compounds in Sauvignon blanc, thus is of the utmost importance to the wine producer.

The sensory interactive effect of reductive compounds such as H_2S could have important implications for Sauvignon blanc wines. Sauvignon blanc wines are often made in a reductive manner, with the aim being to preserve the fruity aroma as much as possible. However, due to the exclusion of oxygen, reductive compounds could contribute negative odours to the wine. The suppressive effect of some of these reductive compounds needs further investigation, seeing that it would exist in the wine medium together with some the typical Sauvignon blanc impact compounds, which should, at this stage, be present in high concentrations.

CONCLUSIONS

Oxygen management-related issues are estimated to compromise the quality of approximately 3% of all wines before the bottles reach the retailer (Nygaard, 2010). Consumers are sensitive to wine faults, such as 'reductive' and oxidative aromas, and this will decrease the liking scores and purchase intent (Nygaard, 2010). The management of oxygen during winemaking thus is generating considerable interest in the wine world. If oxygen exposure is too high or too low, wines can develop defects that can compromise their sensory quality. Too little oxygen exposure could lead to a wine with 'reductive' odours, while too much oxygen can result in an oxidised wine displaying other unpleasant aromas, together with a decrease in pleasant fruity aromas. The challenge thus is to identify the right amount of oxygen needed for a specific wine to maintain the perfect balance between chemical composition and sensory perception. Other than that, research focusing on the evolution of the compounds during the oxidative ageing of wines is needed to

better understand the relationships between compounds and mechanisms. Such research should also be conducted under the conditions that the wine might encounter in a commercial cellar situation, and not only be performed under conditions of enhanced oxidation.

The complexity of a wine as a medium greatly complicates research attempts. Sauvignon blanc wine, especially, is known for its large diversity of flavours, which adds to the complexity. The volatile thiols and the methoxypyrazines are the main compounds involved in the unique character of Sauvignon blanc wines; however, by themselves these key molecules do not account for the aromatic complexity of high-quality wines. Various interactions occur between both volatile and non-volatile wine constituents, and these interactions can change as the concentrations change. Studies have attempted to identify some of these interactive effects. However, much more research is needed to successfully address this issue.

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