Applying the latest understanding of grape composition

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A wide range of compounds that contribute to wine's appearance, aroma, flavour and texture are derived from compounds present in grapes. Quite a lot is now known about these compounds, their origin, how to measure them and how to influence their concentration through viticultural and/or winemaking practices. This article describes how key grape-derived compounds influence wine and discusses a project that is applying the latest compositional knowledge to improve the objectivity of measures of grape quality and style.

AT A GLANCE

- A large body of research has established the importance of a range of grape-derived compounds in influencing wine sensory properties
- Some of these compounds transfer directly from grape to wine while others are transformed during grape processing and fermentation
- Chemical and spectral measures of compounds in grapes have been used to successfully predict grape allocation grades in an ongoing collaborative study
- Adoption of objective measures when assessing grapes could have benefits for both grapegrowers and winemakers.

The important grape-derived compounds in wine fall into four main categories:

- grape compounds that are directly extracted into wine, without undergoing any changes
- compounds that are formed from grapes during processing steps such as crushing, with minimal yeast involvement
- compounds created by the action of yeast or bacteria on grape compounds
- compounds that are influenced by both processing and yeast effects.

A summary of the key impact compounds that fall under these categories is provided below, with a brief background outlining the points in the wine production process most relevant to their development.

GRAPE COMPOUNDS THAT ARE DIRECTLY EXTRACTED INTO WINE

Compounds in this category include rotundone (black pepper/spicy aroma), methoxypyrazines (green bean, capsicum and asparagus aromas) and the C_{13} norisoprenoid ß-ionone (raspberry, berry and violet aromas). These compounds are present in grapes and end up in wine without undergoing chemical changes.

Rotundone is found primarily in grape skins, but has also been reported in leaves and stalks. It increases during grape ripening and is easily extractable (AWRI publications #1060, #1061, #1356, Caputi *et al.* 2011). The effects of viticultural and environmental factors on rotundone formation are currently being investigated, including fruit exposure, leaf removal time, crop load and vine vegetative vigour. Most recent results suggest that sunlight exposure and the temperature of grape bunches are key factors. Methoxypyrazines are found in Cabernet Sauvignon, Merlot, Sauvignon Blanc and Semillon grapes. They are present in grape stems, skins and seeds (but there is very little in grape flesh) and although they are not found in grapes of Pinot Noir, Shiraz, Chardonnay and Riesling, the stems of these varieties can potentially contribute them to wine. It is generally considered that the concentration in wine depends on berry maturity, climate, fruit exposure and to a lesser extent skin contact during winemaking and pressing. In common with rotundone, methoxypyrazines are also easily extractable and the concentration in free-run wine is principally determined by that in the grapes (Sala *et al.* 2004). Approximately two-thirds of the amount present in grapes is extracted with traditional red winemaking practices (Ryona *et al.* 2009). ß-Ionone is a C_{13}

norisoprenoid present in the grape berry that is also directly extracted from grapes into wine and contributes positive red fruit and floral characters. Norisoprenoids are generally found at higher concentrations in sunlight-exposed bunches.

COMPOUNDS FORMED FROM GRAPES DURING PROCESSING, WITH MINIMAL YEAST INVOLVEMENT

Compounds in this category include some terpenes (e.g. geraniol, linalool, nerol, ß-terpineol and wine lactone), the C_{13} norisoprenoids TDN (aged Riesling characters) and ß-damascenone (sweet, fruity, cooked apple aroma) and the C-6 alcohols (fresh cut grass aroma).

Monoterpenes are a class of approximately 70 identified compounds that generally exhibit floral and citrus aromas. Their aromas are thought to be additive and they have been well-researched and reviewed (Mateo and Jimenez 2000). Free terpenes of particular relevance in very young wines include geraniol and nerol (present in grape skin), while in somewhat older wines linalool (present throughout the berry) and ß-terpineol play a more significant role. Free terpenes can be extracted directly from the grape but enzymes and acid release the glycosylated precursors that are also extracted from grapes into wine. The polyhydroxylated monoterpenes are a class of terpenes that do not contribute aromas themselves but are reactive and can easily rearrange during fermentation to give pleasant and potent aromas. Examples include *cis*-rose oxide which gives the lychee and rose aromas characteristic of Gewurztraminer and wine lactone which gives a coconut/lime aroma commonly observed in Riesling wines. The volatile compound TDN (1,1,6-trimethyl-1,2-dihydronaphthalene) contributes to aged characters in Riesling wines and at high levels can give a kerosene-like aroma. ß-Damascenone is another C_{13} norisoprenoid, which forms during grape berry metabolism through carotenoid degradation. It acts as a general aroma enhancer, lifting the perceived intensity of other molecules. The C6-alcohols give a fresh cut grass aroma in wine and they form through enzymatic oxidation of grape fatty acids. Solids in ferment are considered to increase their concentrations. Characteristic compounds include 1-hexanol and *cis*-3-hexenol.

COMPOUNDS CREATED BY THE ACTION OF MICROORGANISMS ON GRAPE COMPOUNDS

Examples of compounds in this category include acetic acid (VA), fatty acids (short and medium-chain acids that give rancid, sweaty, cheesy aromas), esters (fruity aromas) and alcohols (rose, lilac, solvent). Yeast produce a wide range of volatile compounds through metabolism of amino acids, with the amino acid profile of a juice considered to be important to the pattern of aroma compounds produced in wine.

Acetic acid is the main volatile acid of relevance and, as with fatty acids, derives from the action of yeast and bacteria. Although commonly found in wine, both are generally only of major sensory relevance in microbially spoiled wine. Esters are very significant in defining the sensory profile of a wine and are generally absent from grapes. Important classes of esters include the acetate esters derived from acetic acid and fusel alcohols, and the ethyl esters derived from ethanol and fatty acids. Characteristic compounds include 3-methyl butyl acetate responsible for banana aroma and ethyl hexanoate which gives a pineapple aroma.

Rotundone, responsible for black pepper/spicy aromas, is present in grapes and ends up in wine without undergoing chemical changes. Rotundone is found primarily in grape skins, and increases during grape ripening.

Choice of yeast and the level of yeast assimilable nitrogen (YAN) in must can significantly influence both the profile and concentration of esters that are formed during fermentation. A range of alcohols also contribute to the aroma profile of wines and are formed primarily through yeast metabolism of amino acids and sugars. Characteristic compounds include 2-phenylethyl alcohol (rose, lilac) derived from the amino acid phenylalanine, isobutanol (solvent, harsh) and isoamyl alcohol (whiskey, malt, burnt).

COMPOUNDS INFLUENCED BY BOTH PROCESSING AND YEAST EFFECTS

Examples of compounds in this category include the so-called varietal thiols (passionfruit, grapefruit, box hedge), colour compounds in red varieties, tannin (astringency, texture, mouthfeel) and polysaccharides (texture, weight).

Varietal thiols are important impact odorants in Sauvignon Blanc wines, and are major contributors to the tropical flavour of other varieties such as Chardonnay and Riesling. They are released from flavourless cysteine and glutathione conjugates by yeast. The conjugates are themselves formed as a result of complex changes that occur after the crushing of the grape berry. Red wine colour has long been known to have a positive relationship with quality and flavour. Grape colour comes from anthocyanins which are mostly located in grape skin. These are found in grape must, ferments and young wines, but are not very stable under wine conditions so their contribution to wine colour decreases quite rapidly as wine ages. Stable red wine colour comes from the formation of pigmented tannins, which are coloured compounds formed through the reaction of anthocyanins with tannins during fermentation and wine storage. Choice of yeast strain has been shown to influence the formation of stable red wine colour. The tannins (also known as proanthocyanidins) in red wine strongly influence mouthfeel, particularly with respect to astringency (Gawel 1998). Tannin concentration has been shown to correlate strongly with perceived astringency intensity (AWRI publication #1086, Kennedy *et al.* 2006) and both total tannin concentration and aspects of tannin composition have both been shown to be positively associated with wine allocation gradings from major commercial wineries (AWRI publications #1254, #1323). Grape tannins are present in both the seeds and skins of grapes and are chemically different from wine tannin. The relationship between grape tannin and wine tannin has not yet been fully established and can be influenced by a number of winemaking practices and techniques, including choice of yeast, fermenter type and size, cap management and temperature. Polysaccharides can also influence wine style, but significant knowledge gaps exist in the understanding of the magnitude of their influence on sensory aspects of wine. They derive from both grapes and the action of yeast and are generally considered to contribute fullness in wines and to modulate the perception of astringency (AWRI publication #763). More research is needed in this area to understand the roles they play in defining wine style and their relevance to consumers. ►

DEVELOPING OBJECTIVE MEASURES FOR GRAPES

The body of knowledge about the most important compounds in grapes that relate to wine appearance, aroma and flavour has been applied in a project working to develop objective measures for grapes that relate directly to attributes that confer value to wine. For both grapegrowers and winemakers, such measures could provide specifications that would allow the most value to be achieved from grapes. The project, funded by Wine Australia, has been carried out in partnership with Accolade Wines and FABAL vineyard managers.

The project aimed to measure a range of chemical compounds in multiple grape batches of different grades and determine which compounds, independently or in combination, could differentiate between grape grades. The objectives were to determine how variable the chemical measures were across a wide range of fruit grades; if there was a relationship with fruit grade; and if the fruit could be classified based on similarity of chemical composition. A further aim was to assess the practical application of grape compositional grading measurements and to support wine producers who intend to apply these measures in their systems.

Findings from this project based on data from Cabernet Sauvignon, Shiraz and Chardonnay grape lots from the 2014 vintage are summarised below.

WHICH CHEMICAL COMPOUNDS WERE ASSOCIATED WITH HIGHER VALUE FRUIT?

Grapes from a range of quality grades were sourced by representative sampling of vineyards from multiple regions across Australia, and a wide range of chemical analyses were

Compounds influenced by both processing and yeast effects include anthocyanins, which form grape colour and are mostly located in grape skin. These are found in grape must, ferments and young wines, but are not very stable under wine conditions so their contribution to wine colour decreases quite rapidly as wine ages. Stable red wine colour comes from the formation of pigmented tannins - coloured compounds formed through the reaction of anthocyanins with tannins during fermentation and wine storage.

performed to determine the concentration of compounds known to affect wine style and key sensory properties. The grading data were supplied by the grower or winery contracted to make wine from those grapes. Accolade Wines grades grapes and wines on a scale from one (highest value) to nine.

Using the chemical data, statistical models based on discriminant analysis were developed to predict the grade of the fruit samples. The models were successful, able to classify samples correctly to a high degree (85-93%). Almost half of the incorrect predictions were less than two grades away. Using an alternative statistical approach (partial least squares regression) also gave models with a good ability to predict grades, with overall R^2 values of 0.65 to 0.78 (65-78% of the variance in the grade was explained by the chemical measures), which is a promising result. The regression models can be used to determine which aspects of grape composition are most important in differentiating between grades. Coefficients can be positive or negative depending on whether higher values for particular measures are associated with better grade or poorer grade.

Breaking the 2014 vintage dataset down by variety, for Cabernet Sauvignon there was a moderately successful prediction of grape grade (R^2 of validation = 0.71). The significant variables that were higher in higher grades were some specific amino acids, tannin, glycosyl glucose (GG) and absorbances at some specific UV-Vis wavelengths. The concentration of C6 compounds, YAN and some amino acids was higher in lower grades. Discriminant analysis was 90% correct for grade prediction.

For Shiraz there was also a moderately strong prediction of grade (R^2 of validation = 0.65). The significant variables that were higher in higher grades were some amino acids, UV-Vis absorbances, tannin, GG and unlike for Cabernet, also C6 compounds and YAN. Only the amino acid aspartic acid was higher in lower grades. Discriminant analysis was 85% correct for grade prediction.

For Chardonnay there was good prediction of grades (R^2 of validation = 0.78). The significant variables that had higher values in higher grades were GG, precursors to the varietal thiol 3MH, malic acid, ammonia nitrogen, two C6 compounds and ammonia. The measures of UV-Vis absorbance at 370nm (flavonols, a sun exposure marker) and proline concentration were higher in lower grades, and higher pH and a C6 compound were also associated with lower grades. Discriminant analysis was 93% correct for grade prediction.

Grape spectral data only (a combination of UV-Vis spectra of grape extracts, MIR spectra of juice and NIR spectra of grape homogenates) was able to predict grade better than 90% for Cabernet Sauvignon, Shiraz and Chardonnay. Spectral tools used to obtain such data are rapid and would be more readily accessible to industry than some of the more complex and slow chemical measurements.

In summary, a number of important chemical measures were identified as being related to grade across the 2014 dataset for the different varieties. Measures that were positively associated with higher value grade included total soluble solids (TSS), total phenolics, red colour, hydroxycinnamic acids, a range of amino acids (some of which may be precursors to aroma compounds), glycosyl glucose (GG, aroma precursors), tannin and thiol precursors. Compounds that were negatively associated with grade included nitrogen measures, several other amino acids and the two C6 'green', 'grassy' compounds Z-3-hexenol and E-2-hexenol. Tannin, GG, TSS, total phenolics and red colour have all been previously demonstrated to be generally positively associated with grape and wine quality in Australia and the results from this dataset are consistent with those previous observations. Across two seasons studied, total phenolics, tannin and colour were consistently positively associated with grade in Cabernet Sauvignon. Many of the identified chemical measures can be accessed through commercial laboratories or can be implemented with low to moderate technical investment depending on the facilities available to the winery or grower.

The project has continued in 2015 and wines have been made from the grape samples under standardised conditions to allow the assessment of wine sensory properties and to relate these to the grape compositional measures.

CONCLUSION

Developing an understanding of the relationships of available objective measures to well-established subjective grading systems is one application of the extensive research that has been done across the world on grape and wine composition. The application of such measures has the potential to significantly reduce production costs and increase value by ensuring that fruit is used in the most efficient production stream and that maximum value is returned from the end-product. It could also lead to significant savings in the costs of assessing vineyards through more effective application of resources and clearer understandings of geographical, viticultural and climatic drivers.

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