The Australian Wine Research Institute

Fact Sheet
ANALYSIS

Measuring grape colour, phenolics and tannins using different analytical methods

Introduction

The WineCloud™ provides the ability to measure tannins, phenolics and colour attributes in grapes, as well as wines. The analysis is based on UV-Visible spectral readings and uses calibrations maintained by the AWRI. Grape measurements made through the WineCloud™ can be significantly affected by the homogenisation method employed, the extraction medium used and its volume proportionate to the amount of grape homogenate. This sensitivity to preparation method appears to be true for all published methods available for the determination of colour and phenolics in grapes. This fact sheet aims to help you understand the significance of tannin, phenolics and colour measurements made using different available methods.

What is being measured?

Phenolics are a chemically diverse, important class of wine molecules that have a significant impact on the colour and taste of red wine.

For grape samples, three phenolic measures are available via the WineCloud™:

Grape Total Phenolics: This is a measure of all coloured and non-coloured phenolic molecules present in grape skin, flesh and seed. Grape total phenolics are measured in absorbance units (AU) per g fruit.

Grape Total Tannin: Tannins are a sub-class of phenolics and are characterised by their ability to precipitate proteins. They contribute to texture, particularly astringency. Grape tannins are present in both the seeds and skins of grapes and begin to accumulate very early during grape development. Grape total tannin is measured in mg/ g fruit in epicatechin equivalents. It's important to note that grape tannin is chemically different from wine tannin. Wine tannin starts with the base molecules of grape tannin, but evolves though a series of complex reactions. The relationship between grape tannin and wine tannin can be influenced by a number of winemaking practices and techniques, including fermenter type and size, cap management, temperature and yeast strain.

Grape Total Anthocyanin: This is a measure of the red colour of a grape sample. Grape anthocyanins are measured in mg/ g fruit in malvidin-3-glucoside equivalents. Most anthocyanins come from grape skin and begin to accumulate after veraison. Anthocyanins are relatively soluble and more freely extracted than tannin.

How do methods differ?

The most widely used grape analysis methods for colour (anthocyanins) and phenolics have been developed by Glories (Saint-Cricq et al, 1998), the AWRI (Iland et al, 2004) and the ITV Standard (Cayla et al, 2004). These methods all use spectroscopic techniques to measure the total anthocyanins and polyphenol content of grape homogenates. However, the extraction media used (pH and ethanol concentration) and the homogenisation processes are different for each method and therefore the extraction efficiency in each case will be different. The original AWRI method is incorporated into the WineCloud™ for the measurement of anthocyanins and phenolics, whilst tannin content is calculated using a predictive algorithm based on the methyl cellulose precipitation (MCP) method developed by the AWRI (Dambergs et al. 2012).

Method	AWRI	Glories	ITV
Homogenisation	Retsch/Ultra-Turrax	Waring	Waring
Acid strength (extraction)	1M	0.1M	0.1M
% Ethanol	50%	-	15%
Incubation time	1hr	4hrs	1hr

How do the results compare?

The AWRI has compared tannin, phenolics and anthocyanin data generated using the Glories (pH 1) method against that generated using the WineCloud™ method. These studies have shown that results generated using the Glories and AWRI methods differ in the following manner:

- Colour values generated using the AWRI method appear to be approximately 2 times higher than those measured using the Glories (pH 1) method (Figure 1).

- Phenolics values generated using the AWRI method appear to be approximately 4 times higher than those measured using the Glories (pH 1) method (Figure 2).

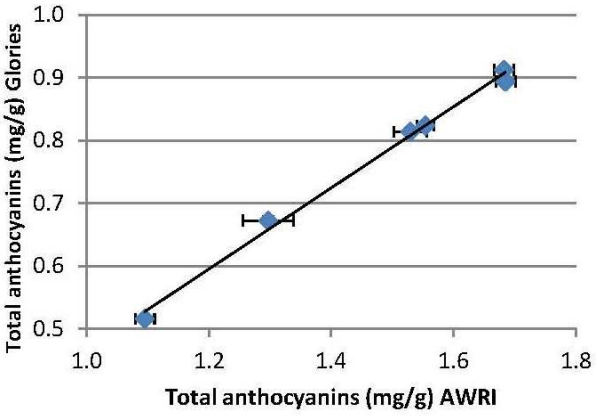

Figure 1. Comparison of total anthocyanins measured using AWRI and Glories methods

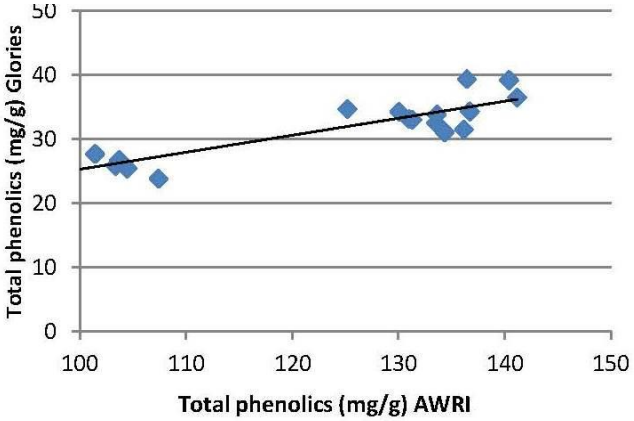

Figure 2. Comparison of total phenolics measured using AWRI and Glories methods

These effects may be due to the higher ethanol concentration used in the extraction medium for the AWRI method.

For the AWRI (WineCloud™) method, the results generated can be significantly influenced by the homogenisation method:

Tannin values generated using the AWRI method appear to be approximately 2-3 times higher (Figure 3) and total phenolics are approximately 50-100% higher (Figure 4) when using a Retsch homogeniser compared with a Waring (kitchen-type) blender. The magnitude of the differences appears to be variety-specific.

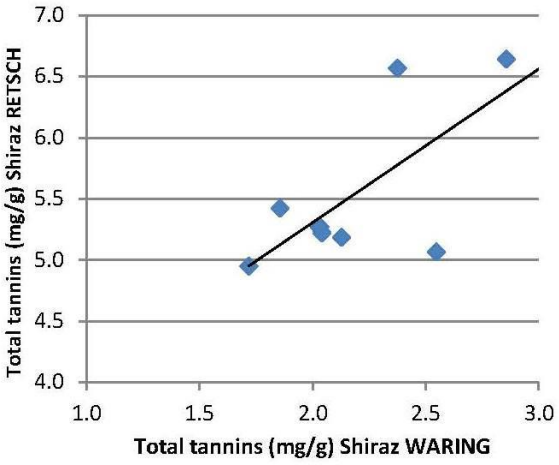

Figure 3. Comparison of total tannin measured following extraction using different homogenisers

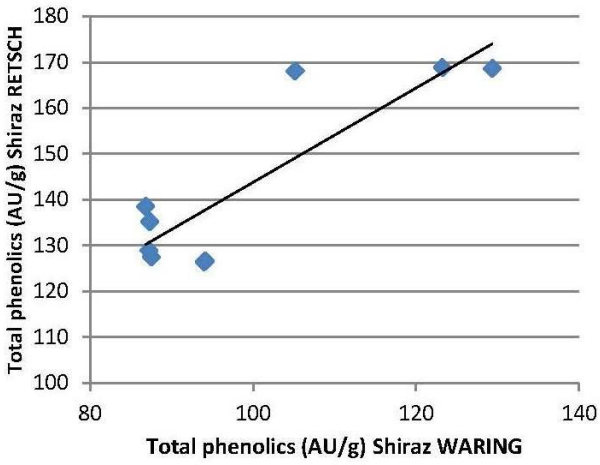

Figure 4. Comparison of total phenolics measured following extraction using different homogenisers

The variability in tannin, colour and phenolics results is higher when a standard kitchen-type blender (e.g. Waring) is used compared with when a higher performance homogeniser (e.g. Ultra-Turrax, Retsch) is used.

Why do the results differ?

Differences observed due to the homogeniser used reflect those seen in previous studies (Cynkar et al. 2004), specifically in relation to the impact on variability of anthocyanin measurements and degree of extraction of phenolic components. It is likely that homogenisation using a Retsch or Ultra-Turrax unit would be powerful enough to break up the seeds and enhance the extraction of tannins.

Factors such as the strength of the acid used and the amount of ethanol present in the extraction matrix, as well as the ratio of homogenate to extraction medium may also be influencing the degree of colour, phenolics and tannins extracted from the grapes. Current studies have shown that the use of 50% ethanol as the extraction medium enhances the extraction of tannins (and colour) to a greater degree than a more wine-like extraction medium (~15% ethanol). With phenolic components, especially tannins, it appears that there is a cumulative effect from both homogenisation method (blender type) AND extraction medium.

Those wishing to develop a better understanding of the anthocyanin, phenolic and tannin levels in their grapes should be aware of the impact that homogenisation method and extraction medium can have in extracting these components from the homogenate.

Reference and further reading

Cayla, L.; Cottereau, P.; Renard, R. Estimation de la maturité phénolique des raisins rouges par la méthode I.T.V. standard. *Rev.Fr. Oenol.* 2002, 193, 10–16.

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