Technical notes

Choice of yeast influences volatile fermentation-derived compounds, wine colour and phenolic composition – Case study in Australian Cabernet Sauvignon

The colour of red wine provides the first impression of its quality. However, it is the aroma and flavour which will leave the lasting impression. Yeast and bacterial metabolism during fermentation produce a large array of metabolites, volatile and non-volatile, which contribute to the aroma and flavour of wine (Swiegers et al. 2005; Francis and Newton 2005).

Saccharomyces is an important yeast genus within food and beverage industries and *S. cerevisiae* is the most prevalent and important species in wine production (Fleet 1998). However, there are other members of the *Saccharomyces* sensu stricto complex associated with wine production. *S. bayanus*, a cryophilic yeast, is used in winemaking for its ability to ferment at lower temperatures, making it popular in white wine production, and it produces wine with a different chemical composition to *S. cerevisiae* fermented wines (Eglinton et al. 2000). Whilst other *Saccharomyces* species ferment slowly in grape juice most do not tolerate the high ethanol concentrations encountered. Hybrid strains between species within the *Saccharomyces* sensu stricto complex occur naturally in the wine environment or have been generated from crosses between *S. cerevisiae* and *S. paradoxus* or *S. kudriavzevii* (Bellon et al. 2011). Such hybrids have been used successfully in red and white winemaking, and along with *S. bayanus* strains, have recently gained interest in winemaking for their ability to introduce aroma and flavour diversity to wine (Bellon et al. 2008; Eglinton et al. 2000; Bellon et al. 2011).

The role of yeast in the formation of important wine colour components, including anthocyanin adducts and pigmented polymer formation, in addition to aroma and flavour profile is well established. However, most wine compositional studies have focused on a limited number of yeast strains and only examined the effects of yeast metabolism on volatile compounds or wine phenolics and colour individually.

This study was undertaken to investigate the impact of commercial winemaking strains from the *Saccharomyces* sensu stricto group, including *S. cerevisiae* (7), *S. bayanus* (2) and *Saccharomyces* interspecific hybrids (2), on colour and phenolics, wine composition and volatile fermentation-derived compounds to have a comprehensive comparison of yeast influence on overall wine colour and aroma of an Australian Cabernet Sauvignon (sourced from Clare Valley, South Australia). The grapes were handpicked and the trial was conducted on laboratory-scale (1.5 kg) in triplicate at 25°C. At the end of fermentation the wine was sulfured, stabilised and analysed.

Influence of yeast strain on alcoholic fermentation performance and wine composition

All 11 *Saccharomyces* yeast strains were able to complete alcoholic fermentation (AF) to dryness (residual sugar as glucose + fructose < 3 g/L) in the Cabernet Sauvignon grape must. Wine composition and length of AF is summarised in Table 1. The majority of fermentations were completed within 14–19 days; only two strains required much longer (26 and 32 days). All strains completely metabolised glucose and most had < 1.0 g/L fructose remaining in

Table 1. *Saccharomyces* yeast strains used in this study and wine composition of the Cabernet Sauvignon at the end of fermentation.

	AWRI-796 Sc 796	AWRI-838 Sc EC1118	AWRI-1486 Sc BM45	AWRI-1493 Sc 71B	AWRI-1553 Sc UCD522	AWRI-1554 Sc Maurivin B
Length AF* (days)	14	14	14	14	14	32
Ethanol (% v/v)	13.50 ± 0.32	14.09 ± 0.09	14.16 ± 0.54	13.81 ± 0.14	14.13 ± 0.15	13.87 ± 0.48
Glucose (g/L)	0.00 ± 0.05	0.00 ± 0.10	0.00 ± 0.20	0.00 ± 0.22	0.00 ± 0.19	0.00 ± 0.26
Fructose (g/L)	0.51 ± 0.09	0.69 ± 0.05	0.52 ± 0.05	0.87 ± 0.21	0.62 ± 0.01	2.23 ± 0.36
pH	3.39 ± 0.02	3.58 ± 0.01	3.51 ± 0.04	3.65 ± 0.03	3.56 ± 0.02	3.62 ± 0.02
Total Acidity (g/L)	5.77 ± 0.03	4.13 ± 0.08	4.45 ± 0.09	3.86 ± 0.07	4.13 ± 0.04	4.28 ± 0.04
Volatile Acidity (g/L)	0.21 ± 0.01	0.22 ± 0.02	0.15 ± 0.04	0.05 ± 0.03	0.14 ± 0.04	0.70 ± 0.05
Malic Acid (g/L)	2.94 ± 0.05	1.92 ± 0.14	2.14 ± 0.15	1.37 ± 0.08	1.79 ± 0.01	1.50 ± 0.05
Glycerol (g/L)	12.70 ± 0.04	9.54 ± 0.21	10.25 ± 0.13	12.03 ± 0.17	10.08 ± 0.13	9.85 ± 0.12

	AWRI-1555 Sc BP725	AWRI-1176 Sb 1176	AWRI-1375 Sb 1375	AWRI-1501 Sc x Sp 1501	AWRI 1503 Sc x Sk 1503
Length AF* (days)	18	14	26	19	14
Ethanol (% v/v)	13.97 ± 0.09	12.67 ± 0.86	13.35 ± 0.05	13.94 ± 0.14	13.91 ± 0.23
Glucose (g/L)	0.00 ± 0.08	0.00 ± 0.30	0.00 ± 0.05	0.00 ± 0.21	0.00 ± 0.10
Fructose (g/L)	0.66 ± 0.22	1.32 ± 0.45	2.04 ± 0.14	1.41 ± 0.30	0.56 ± 0.11
pH	3.42 ± 0.02	3.07 ± 0.11	3.06 ± 0.01	3.39 ± 0.03	3.50 ± 0.01
Total Acidity (g/L)	5.14 ± 0.11	8.65 ± 0.28	8.84 ± 0.07	5.54 ± 0.09	4.58 ± 0.09
Volatile Acidity (g/L)	0.12 ± 0.02	0.01 ± 0.06	0.04 ± 0.01	0.03 ± 0.01	0.04 ± 0.01
Malic Acid (g/L)	2.73 ± 0.13	5.57 ± 0.23	5.87 ± 0.18	2.17 ± 0.07	1.91 ± 0.10
Glycerol (g/L)	11.09 ± 0.13	16.03 ± 1.50	15.29 ± 0.37	10.79 ± 0.09	10.98 ± 0.18

Sc x Sk: hybrid between *S. cerevisiae* and *S. kudriavzevii*; Sc x Sp: hybrid between *S. cerevisiae* and *S. paradoxus*. *AF – alcoholic fermentation the wine. *S. cerevisiae* strain AWRI 1554 was the slowest to complete AF (32 days) and had the highest fructose concentration remaining (2.2 g/L). Wine ethanol concentrations ranged 12.7% – 14.2%; *S. cerevisiae* strains produced the highest ethanol concentrations (up to 14.2%), *S. bayanus* the lowest (12.7%) and *Saccharomyces* hybrid strains intermediate concentrations (13.8–13.9%). Malic acid concentration ranged in the *Saccharomyces* interspecific hybrid wines; from 1.91 to 2.17 g/L. As previously observed (Eglinton et al. 2003), *S. bayanus* strains produced malic acid (5.57–5.87 g/L). Glycerol concentrations in the wines varied by up to 6 g/L (9.54–16.03 g/L). *S. bayanus* strains produced the highest glycerol concentrations (15.29–16.03 g/L), whereas for *S. cerevisiae* and *Saccharomyces* hybrids concentrations were much lower (9.54–12.70 g/L).

In this Cabernet Sauvignon study, wine produced with *S. cerevisiae* strain AWRI 1554 had the most distinct chemical composition. *S. bayanus* strains also produced wines which were distinct (Figure 2).

Influence of yeast strain on wine colour and tannin

Colour is an important red wine attribute and is influenced by numerous chemical and microbial parameters. Fermentation of the same Cabernet Sauvignon grape must by 11 different *Saccharomyces* strains resulted in a broad range of colour densities (13.2–16.0 a.u.) (Figure 1). *S. cerevisiae* strains tended to produce wines with lower colour density (13.2–15.5 a.u.) and *S. bayanus* strains and *Saccharomyces* hybrid (AWRI 1501) produced wines with the highest colour density (15.7–16.0 a.u.). Anthocyanins are important contributors to wine colour. *S. bayanus* wines had the lowest total anthocyanins. Malvidin-3-glucoside, the main

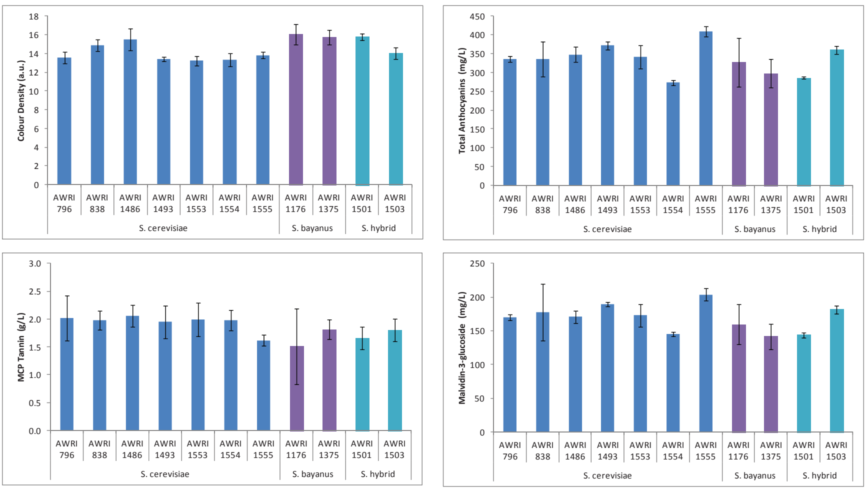

Figure 1. Wine colour density, tannin and anthocyanin composition of Cabernet Sauvignon wines.

anthocyanin in red wine, varied in concentration (141 – 203 mg/L) with no species-associated trend. *S. cerevisiae* strains AWRI 1555 and AWRI 1554 showed the lowest and highest total anthocyanins concentration (273 and 409 mg/l M-3-G equivalents, respectively).

Tannin content of wine contributes to various wine attributes including mouth-feel, which is linked to astringency. In this study, a variation was observed across the tannin content in the 11 different wines (1.51–2.06 g/L). *S. bayanus* AWRI 1176 and *S. cerevisiae* AWRI 1486 wines had the lowest and highest concentrations of tannins (1.51 and 2.06 g/L epicatechin equivalents, respectively).

This study in Australian Cabernet Sauvignon with 11 different yeast strains has highlighted the importance of yeast strain in wine colour and phenolic composition.

Influence of yeast strain on volatile fermentation-derived compounds

Yeast metabolism affects volatile fermentation-derived compounds, including acetate and ethyl esters, higher alcohols and acids, which consequently impact and influence wine aroma (Swiegers et al. 2005). The overall fermentation-derived secondary metabolite pool was influenced by the choice of *Saccharomyces* yeast strain and Figure 2 provides an overview for the 11 different Cabernet Sauvignon wines.

Acetate esters typically have pleasant fruity, perfume and floral aroma descriptors, with the exception of ethyl acetate which is described as nail polish. *S. cerevisiae* AWRI 1554 wine

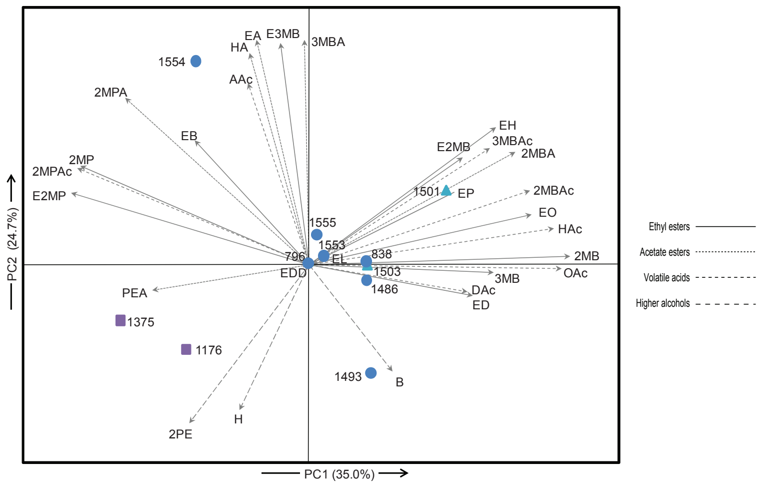

Figure 2. Principal component analysis (PCA) of volatile fermentation-derived compounds of Cabernet Sauvignon wines produced with different *Saccharomyces* strains.

had the highest concentration of this group of fermentation-derived compounds. *S. bayanus* AWRI 1176 tended to exhibit overall lowest acetate ester concentrations.

Short chained ethyl esters (up to C6) exhibit pleasant fruity, berry and green apple aromas, whereas longer chained ethyl esters (C8-C12) have pleasant to soapy aroma. Production of these ethyl esters varied substantially with choice of yeast. *S. bayanus* strains produced the lowest concentrations of long chained ethyl esters (C8-C12) and yeast variation was evident in production of short chained ethyl esters.

Volatile fermentation-derived acids produced by yeast during AF range in aroma descriptors such as cheesy, sweaty and rancid. *S. bayanus* strains generally produced the lowest levels of volatile acids of the 11 yeast strains studied.

Summary

The ability to produce wines with different compositions from the same grape must is valued by winemakers who want to have the ability to tailor their wines to suit diverse consumer markets. There are various options available, with yeast strain selection being one of the simplest. This study, in Australian Cabernet Sauvignon, provides insight into how 11 different commercially available *Saccharomyces* strains influence wine composition, wine colour phenolics profile, and volatile fermentation-derived compounds, all of which play an important role in the colour, aroma and flavour of wine.

When wine components, phenolic and volatile fermentation-derived compounds, are examined together, it is immediately apparent that different yeast have the capacity to make wines with different composition. Cabernet Sauvignon wines produced with *S. bayanus* were very distinct from the *S. cerevisiae* and *Saccharomyces* hybrid wines. Of the seven *S. cerevisiae* strains used, there are two, AWRI 1554 and AWRI 1493, which produced wines that were compositionally very different from the other wines. In addition, the *Saccharomyces* hybrid AWRI 1501 wine had a unique profile, differentiating it from the other wines.

S. bayanus, a cryophilic yeast, is used mainly to make white wine, however, more recently it has also been used with red grape varieties (Eglinton et al. 2000; Bellon et al. 2008). In this study, *S. bayanus* strains produced wines with lower ethanol and higher glycerol concentrations, as well as high concentrations of components which contribute to the wine colour density; all consistent features of this species. Volatile fermentation-derived compound composition was distinct for both *S. bayanus* strains. Red wines prepared with AWRI 1176 and analysed by electronic nose based gas chromatography were distinct from wines made with *S. cerevisiae* NT 116 (Antoce and Namolosanu 2011), which concurs with the results obtained at the AWRI.

The two interspecific hybrid strains used in this study completed AF efficiently, within 19 days, taking only a few days longer than their *S. cerevisiae* parent (AWRI 838). Of the two, AWRI 1501 wine was most different to the *S. cerevisiae* parent wine (Figure 2). There is little available information on the performance of *Saccharomyces* hybrids in relation to wine phenolics and influence on wine colour. The two *Saccharomyces* hybrids used in this study produced wines which had higher colour density than most *S. cerevisiae* wines.

This study of Australian Cabernet Sauvignon wines produced with 11 different commercially available *Saccharomyces* strains has highlighted the diversity that yeast can offer to red sensory properties and the importance of yeast strain selection to achieve the desired wine style.

This article is a summary of a recent publication and further information can be found about the study in Blazquez Rojas et al. (2012) *Influence of* Saccharomyces *species and hybrid yeasts on volatile fermentation-derived compounds, colour and phenolics composition in Cabernet Sauvignon wine. World Journal of Microbiology & Biotechnology 28, 3311–3321.*

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