

Abstract

Yeast contribution to the sensory attributes of wine exceeds the conversion of sugars to ethanol and carbon dioxide. Depending on the initial substrate, it synthesizes hundreds of aroma-active compounds, like higher alcohols or esters, while enhancing the release of grape varietal aroma. The aim of this study was to examine the use of two yeasts which produce higher amounts of either of glutathione or varietal thiols, two different organic yeast nutrients (di-ammonium phosphate free yeast autolysates with different amino acids profile) and sulfur beta-lyase enzyme on the aroma production in two different widely used Australian grapes varieties: Sauvignon Blanc and Chardonnay. We measured the different production of thiols, higher alcohols, acetate and ethyl esters in the final wine. Indeed, the nutritional differences of the musts proved to affect yeast metabolism and therefore the production of these odour-active molecules. Among all, amino acids and grape derived precursors proved to be of great importance to the final wine sensory attributes. The results demonstrate how organic nutrient supplementation can increase the complexity of the wine, while the enzyme use is more targeted to specific aromas. Different outputs can be achieved with different combinations of these commercially available products, giving a better idea to winemakers of what they can and cannot pursue.

Material and Methods

A- Fermentation procedures

Sauvignon Blanc (20° brix) and Chardonnay (25° brix) grapes were harvested from McLaren Vale, Australia. Two commercial yeasts (Fermol Sauvignon; GlutafermONE), enzyme (Endozym thiol) and organic nutrients (Fermoplus Dap Free, Fermoplus Tropical) were obtained from AEB Oceania. For each yeast and grape variety combination, eight conditions were established for this study following the producer recommendations: control (1), Endozym thiol 5 mg/L (2), Fermoplus Dap Free 250 mg/L (3), Fermoplus Dap Free 400 mg/L (4), Fermoplus Tropical 250 mg/L (5), Fermoplus Tropical 400 mg/L, Endozym thiol 5 mg/L plus Fermoplus 250 mg/L (7) and Endozym thiol 5 mg/L plus Fermoplus Tropical 250 mg/L (8). Fermentations were carried out in triplicate at the same time using a fit-for-purpose FreedomEVO Robot (Tecan, Switzerland; Figure 1) at the Multi-scale Fermentation Facility at the University of Adelaide. Fermentation volume was 100 ml (Figure 2) and temperature was set to 18°. Fermentations were monitored spectrophotometrically by daily sugar enzymatic assay (Tecan, Switzerland).

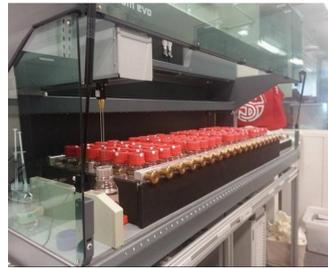


Figure 1: FreedomEVO Robot



Figure 2: Fermentation bottle

B- Aroma compounds analysis

Higher alcohols, acetate and esters were measured by Headspace Solid-Phase Microextraction-Gas Chromatography-Mass Spectrometry (HS-SPME-GC-MS) with an Agilent 7890A GC coupled to 5976C MSD detector following the protocol of Gambetta et al. 2016.

Thiols were measured by Stable Isotope Dilution Analysis by High-Performance Liquid Chromatography – Tandem Mass Spectrometry (SIDA-HPLC-MS/MS) using a ThermoFinnigan Surveyor HPLC and ThermoFinnigan LCQ Deca XP Plus mass spectrometer following the protocol of Capone et al. 2015.

Results

A- Volatile profiles of wines

The two yeasts have very different aromatic profile which changed based on the treatment: organic yeast nutrients resulted the most effective to boost yeast aroma production.

Fermol Sauvignon: in Sauvignon Blanc, ethyl esters (ethyl isobutyrate, C6-10) and acetates concentrations increased significantly (Figure 3A). In Chardonnay, acetate esters were enhanced, followed by 2-phenyl ethanol, ethyl butanoate and ethyl 2-methyl butanoate (Figure 3B).

GlutafermONE: in Sauvignon Blanc, Fermoplus Tropical raised production of acetates, ethyl butanoate and 2-phenyl ethanol (Figure 3C). In Chardonnay, Fermoplus DAP Free enhanced 2-phenyl ethanol, while Fermoplus Tropical increased the content of C8-C10 ethyl esters (Figure 3D).

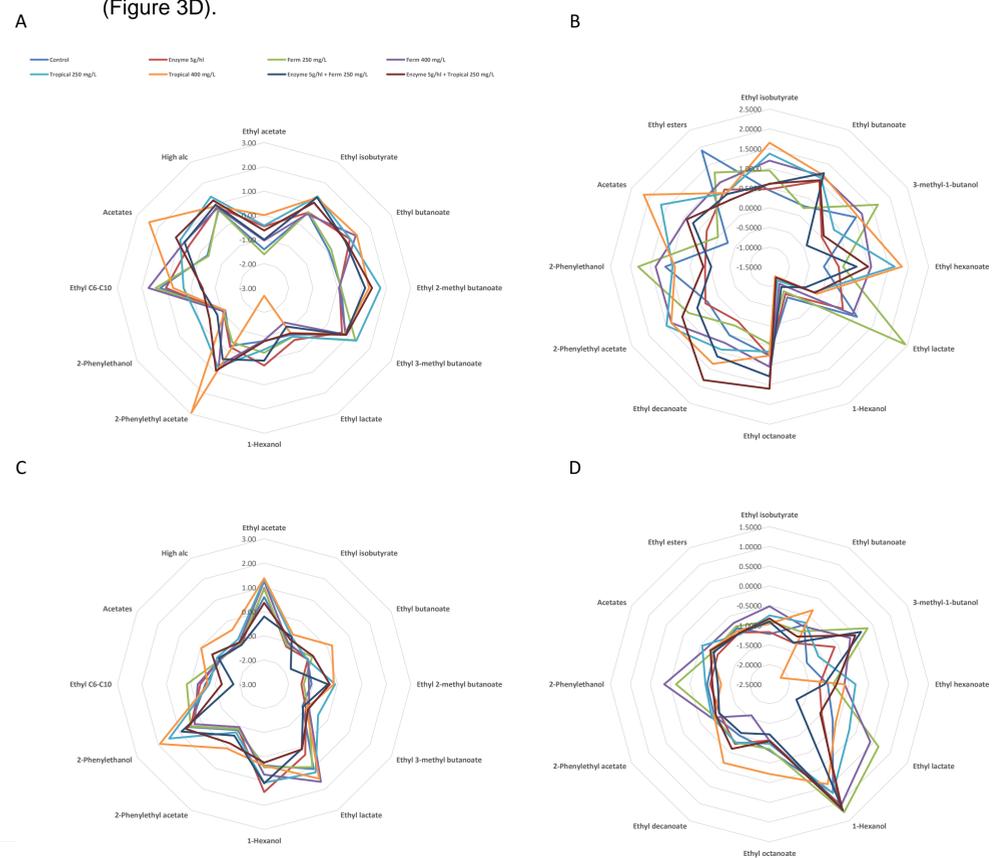


Figure 3: Spider graph of the volatile aromas (normalised values).

B- Thiol profiles of wines

Thiol concentrations were significantly higher in Sauvignon blanc wines than in Chardonnay wines, which was in accord with the fact that Sauvignon blanc is known for its thiols characteristic aromas (Figure 4). The two yeasts produced similar levels of thiols without any addition. Enzyme treatments increased concentrations of 3-MH whereas nutrients additions influenced positively 3-MH and 3-MHA production. In particular Fermoplus Tropical enhanced 3-MHA production in Sauvignon blanc when used in combination with Fermol Sauvignon (Figure 4B).

Conclusion

Yeast selection remains the strongest tool available to winemakers to modulate de-novo aroma production during fermentation. However the combination of one or more oenological products can enhance and modulate yeast impact on the final wine. Enzymes which target grapes aromatic precursor boost varietal aromas which can be further metabolized by yeast. Organic nutrients, on the other hand, enhances de-novo biosynthesis of aromatic compound alongside promoting the endogenous yeast enzymatic expression.

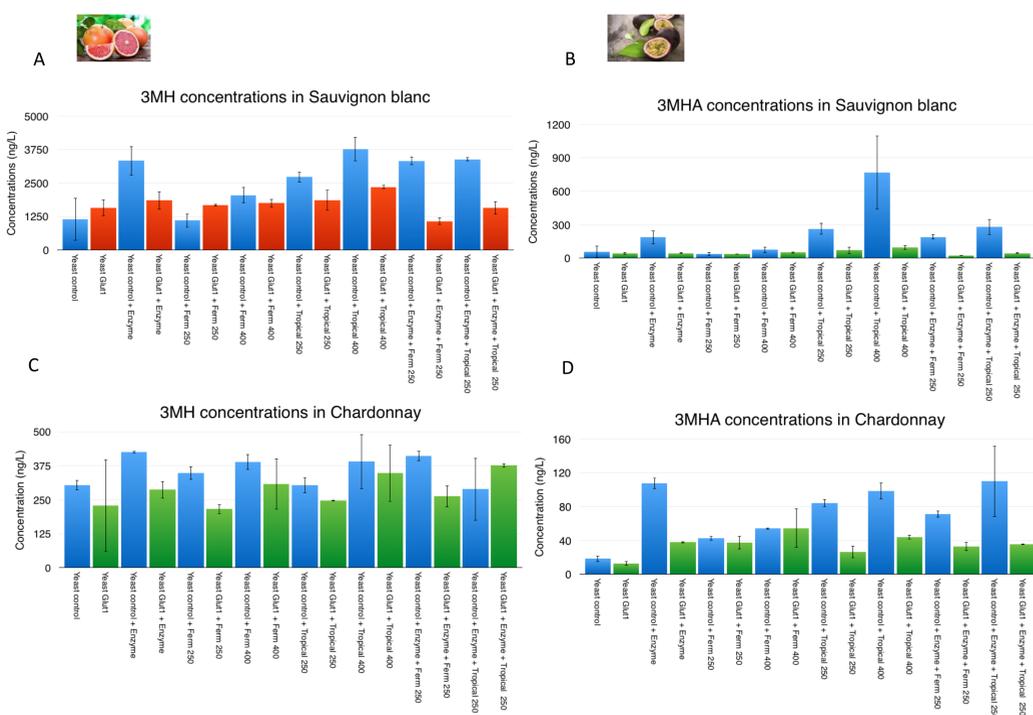


Figure 4: Thiols 3-MH and 3-MHA Concentrations. Fermol Sauvignon (Blue), GlutafermONE (red/green).

References

- Gambetta, J. M., Cozzolino, D., Bastian, S. E., & Jeffery, D. W. (2016). Towards the creation of a wine quality prediction index: Correlation of Chardonnay juice and wine compositions from different regions and quality levels. *Food analytical methods*, 9(10), 2842-2855.
- Capone, D. L., Ristic, R., Pardon, K. H., & Jeffery, D. W. (2015). Simple quantitative determination of potent thiols at ultratrace levels in wine by derivatization and high-performance liquid chromatography–tandem mass spectrometry (HPLC-MS/MS) analysis. *Analytical chemistry*, 87(2), 1226-1231.