Development of a new, innovative, specific yeast autolysate to improve the quality of red wine

Anthony Silvano[1], Jose-Maria Heras[1], Julie Mekoue-Nguela[1,2], Marion Schiavone[1,3], Nathalie Sieczkowski[1]
[1] Lallemand SAS, 19, Rue des briquetiers, 31702 Blagnac, France
[2] UMR 1083 Sciences pour l'Œnologie, INRA, Montpellier SupAgro, Université de Montpellier, 34060 Montpellier, France
[3] LISBP, Université de Toulouse, CNRS, INRA, INSA, 135 avenue de Rangueil, 31077 Toulouse, France

Introduction

Consumer demand for fruity red wines with intense colour and good mouthfeel continues to grow. Aging on lees is a widespread traditional winemaking technique aimed in part at reducing astringency and bitterness while increasing body and aromatic length and complexity. Aging on lees can also help stabilize the colour of red wines. During this step, winemakers reap the many well known benefits—including the release of mannoproteins—provided by adding dead or dying autolyzed yeast (Rodriguez et al., 2005). To avoid the inconvenience of traditional aging on lees, a practice has developed over the past 15 years where specific inactivated yeasts are added to promote the release of polysaccharides (Guadalupe et al., 2007, and Rodriguez-Bencomo et al., 2010). Recent scientific advances have provided more precise tools for characterizing wine yeasts and their products, leading to the development of a new yeast autolysate (MEX-WY1) with unique properties based on an innovative combination of a specific inactivation process and a special strain of *Saccharomyces cerevisiae*.

A better understanding of how products derived from yeasts interact with polyphenols in red wine production

The idea that certain polysaccharides can bind with tannins and thereby reduce the astringency of wines has been around for a number of years. Work conducted recently with the joint research unit Sciences for Enology identified the respective roles of compounds in yeast such as proteins, β-glucans, and mannoproteins (Mekoue et al., 2016). Grape skin tannins with an average degree of polymerization of 27 interacted with both intracellular proteins and parietal mannoproteins in yeast. Although the interactions with proteins led to the formation of large aggregates that precipitated within five hours (adhesion-type), interactions with mannoproteins formed finite-size submicronic aggregates that were stable over time and remained in suspension. However, the β-glucan used (laminarin) did not interact with tannins in such as way as to form aggregates.

These findings support the hypothesis that mannoproteins released by specific inactivated yeasts can help improve the taste of red wine by binding with tannins. It is likely that using this type of product at the beginning of the winemaking process will limit aggregation of tannins and anthocyanins early on, thus improving the colour and mouthfeel of red wine.

Identifying the unique properties of a yeast used in red wine production and the product of its specific autolysis

Biochemical and biophysical properties: recent findings

In recent research conducted in partnership with INSA Toulouse, atomic force microscopy (AFM) was used to characterize properties of wine yeast cell walls (Schiavone et al., 2014). Wine yeasts that displayed strong mannoprotein-producing capacity were selected and AFM used to explore the unique properties of the WY1 strain of *Saccharomyces cerevisiae*. Figure 1 shows AFM topographical images of two cells of the WY1 and WY2 strains (Fig. 1A and 1B) and corresponding images of their adhesion (Fig. 1C and 1D). WY1 was particularly adhesive, and due to its high mannoprotein content and the length (average length: 96.9 nm) of its mannoprotein chains stretched on the cell wall (Fig. 1E and 1F), it interacted strongly with the lectin Concanavalin A.

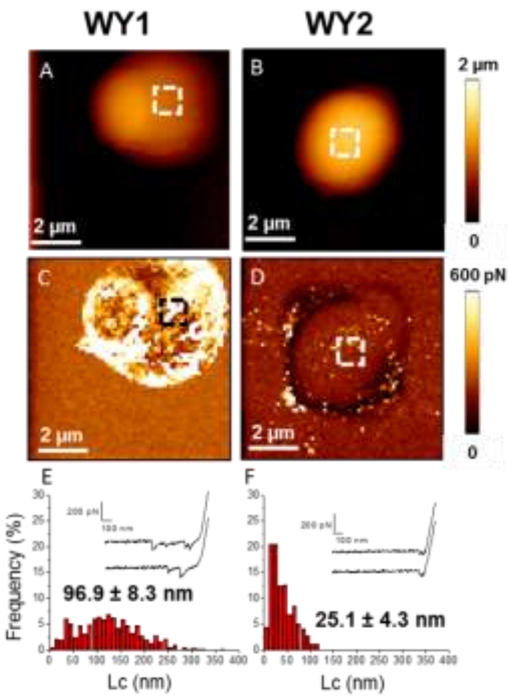


Figure 1. AFM images of the height (A, B) and adhesion (C, D) of strains WY1 and WY2. Distribution and average total length (Lc) of mannoproteins fully stretched on yeast cell walls.

Combination with an inactivation process for an innovative result

Various autolysis conditions and thermal or physicochemical inactivation procedures were applied to the WY1 yeast. Following an initial screening in the lab, two preparations were selected for more indepth characterization and a few preliminary technological tests. Figure 2 shows transmission electron microscopy (TEM) images illustrating the impact of the autolysis/inactivation process on the visual aspects of the product. For the WY1 yeast, the products obtained through thermal (Fig. 2.A = SWYT-WY1) and physicochemical (Fig. 2.B = MEX-WY1) treatment had very different appearances. Although thermally inactivated WY1 yeasts maintained a certain cellular integrity and were more than 60% insoluble, the WY1 yeasts inactivated using the MEX process broke up and became nearly 80% soluble.

2.A: SWYT-WY1 2.B: MEX-WY1

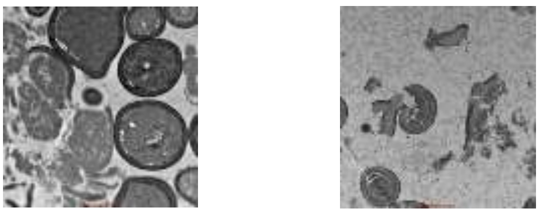


Figure 2: Microscopic (TEM) images of SWYT-WY1 and MEX-WY1 products

Interactions with phenolic compounds in wine

In the lab, the inactivated SWYT-WY1 yeast and the MEX-WY1 autolysate from the WY1 yeast were added at the beginning of fermentation and compared. Fermentation was conducted on a model must medium in the presence of a pool of polyphenols extracted from red must after thermovinification. Bovine serum albumin (BAS) precipitation tests were conducted on the resulting wines using the protocol suggested by Boulet et al. Absorbency differences of

280 and 520 nm (AD 280 and AD 520) between the untreated and BAS-treated wines indicate the amount of tannins and pigments the protein can precipitate. It is important to remember that the capacity of polyphenols to precipitate protein affects the astringency of red wine. Figure 3 shows less precipitation of these compounds in treatments using WY1 yeast products compared to the control. This effect is more marked in the case of the specific autolysate MEX-WY1.

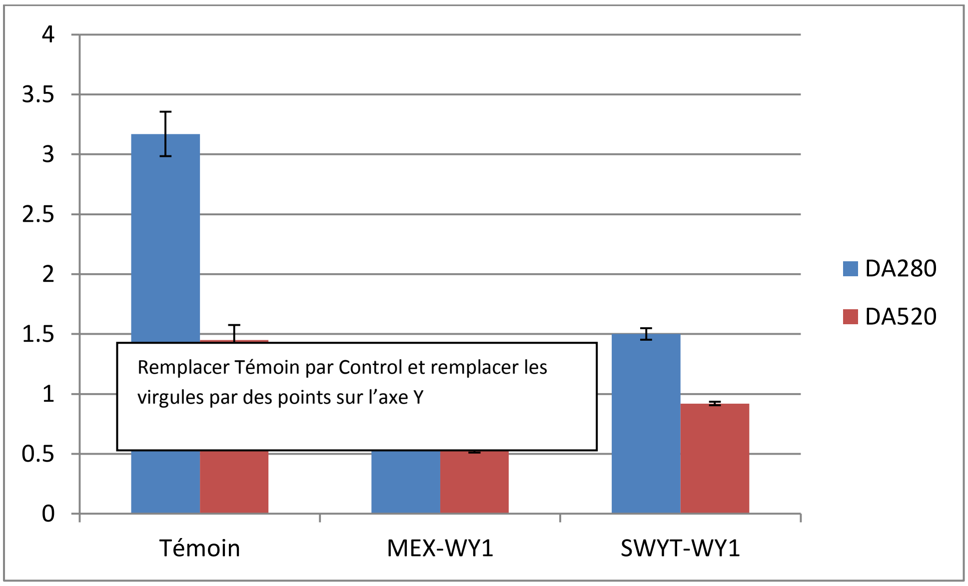


Figure 3: Evaluation of BAS-precipitable tannins (DA 280 nm) and pigments (DA 520 nm) in wines produced through fermentation with or without SWYT-WY1 or MEX-WY1

Evaluation of the MEX-WY1 specific autolysate during red wine production

To study the effect of adding the specific autolysate MEX-WY1 under large-scale production conditions, numerous tests were conducted at the pilot project (1 hL) and production (50-200 hL) scale on various grape varieties in different grape growing areas in both hemispheres. For each test, the objective was to compare standard red wine production (control) with an identical methodology in terms of quality of the raw materials and the winemaking process. The only difference was that MEX-WY1 was added at a rate of 30 g/hL at the beginning of alcoholic fermentation in one of the treatments. Fermentation kinetics were monitored and the resulting wines were analyzed at different stages (post-alcoholic fermentation, post-malolactic fermentation, and post-stabilization). Batch homogeneity was checked by measuring classic physicochemical parameters. The colour of the wines was evaluated through spectrophotometry and by measuring tristimulus values (CieLab). The wines were subjected to a post-stabilization sensory analysis and the saliva precipitation index (SPI) assay. The results of these experiments are illustrated below using several examples.

The beneficial effect of adding MEX-WY1 on fermentation kinetics was observed in most of the tests, especially those featuring particularly stringent conditions (high alcohol potential). This illustrates this autolysate's contribution to the yeast's nutrition (data not shown). It is important to make sure that the yeasts have all the necessary nutrients, depending on the winemaking conditions and the quality of the raw material. Specific activators can be added, as necessary.

Effect on the colour of red wine

In many cases, adding the specific autolysate MEX-WY1 at the beginning of fermentation was observed to have a positive effect on the colour of the wine once alcoholic fermentation was finished and the wine stabilized. This is illustrated in Figure 4, which shows the colour (parameters L, a, b) measured in Pinot Noir wines from a test conducted at wineries in New Zealand (Marlborough area) in 2016. The wine from the fermentation using MEX-WY1 had a darker, redder colour. The ΔE calculated based on three parameters was 4.7. It is widely recognized that a trained professional is able to detect an average ΔE of 3 in red wine.

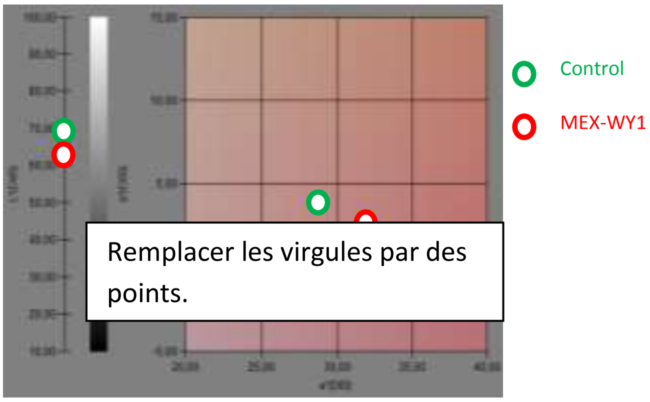


Figure 4: Graph showing L, a, b parameters measured in Pinot Noir wines from MEX-WY1 (MEX-WY1 added at the beginning of fermentation) and control fermentations.

Effect on the sensory qualities of red wine (fruitiness, mouthfeel, overall quality)

Tests using the specific autolysate MEX-WY1 at 30g/hL at the beginning of fermentation demonstrated that several sensory characteristics of red wine can be improved: reduced astringency, better overall mouthfeel, and a riper, fruitier nose.

- Significant reduction in astringency:

The Saliva Precipitation Index (SPI) measures the reactivity of salivary proteins to polyphenols in wine. It has been shown to be a good estimate of astringency (Rinaldi et al., 2012). Figure 5 shows this index in a Grenache wine made with a Thermo Flash process, known to promote significant phenolic extraction, which can lead to pronounced astringency. We can see here that the SPI is 38 for the wine fermented with MEX-WY1 and 52 for the control, reflecting a very significant difference that directly correlates with the reduced astringency in the MEX-WY1 wine.

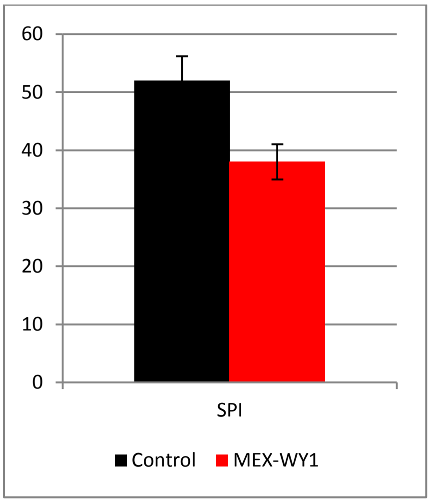


Figure 5: Saliva Precipitation Index (SPI) measured in two Grenache wines where the only variable was the addition of the specific autolysate at 30g/hL at the beginning of fermentation in the MEX-WY1 treatment compared to the control without MEX-WY1.

- Overall improvement in the mouthfeel and structure of red wine:

Apart from the reduced astringency mentioned above, most of the tests showed an overall improvement in the perceived structure and mouthfeel of the wine. Figure 6 shows the results of a sensory analysis on a 2016 Cabernet Sauvignon (Paso Robles, Central Coast, California) where a MEX-WY1 treatment was compared to a control treatment without MEX-WY1. Both wines underwent a blind test by an expert panel trained in wine texture and structure descriptors (La Rioja, Spain, March 2017). The panel found that the addition of MEX-WY1 significantly improved the five criteria they looked at: greater freshness, more volume/roundness, enhanced tannin structure and concentration, and better length. So it appears that the mechanisms and interactions described at the beginning of the article have an impact on not only astringency, but also other taste characteristics related to the wine's mouthfeel and structure.

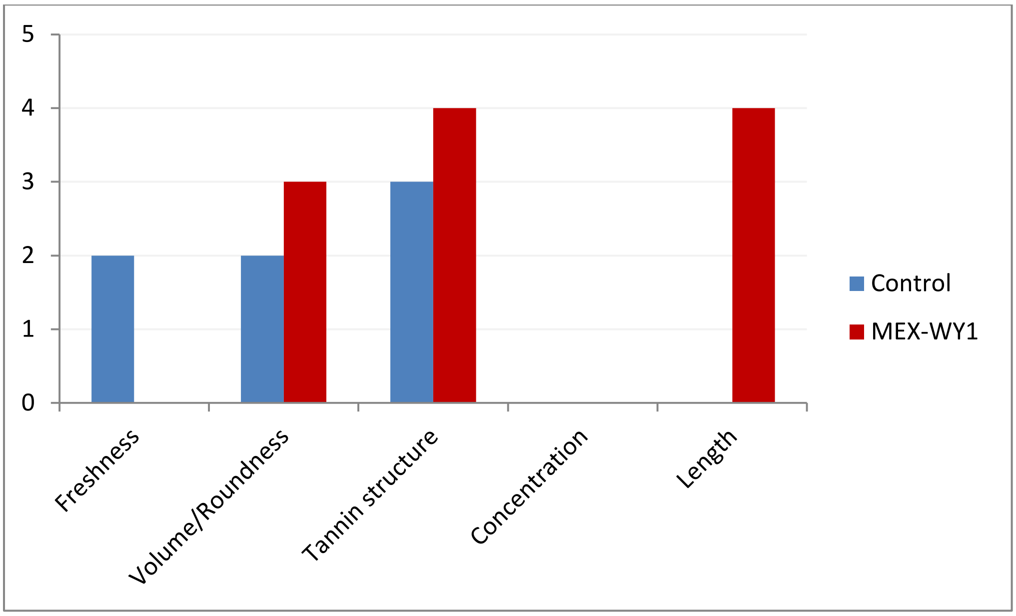
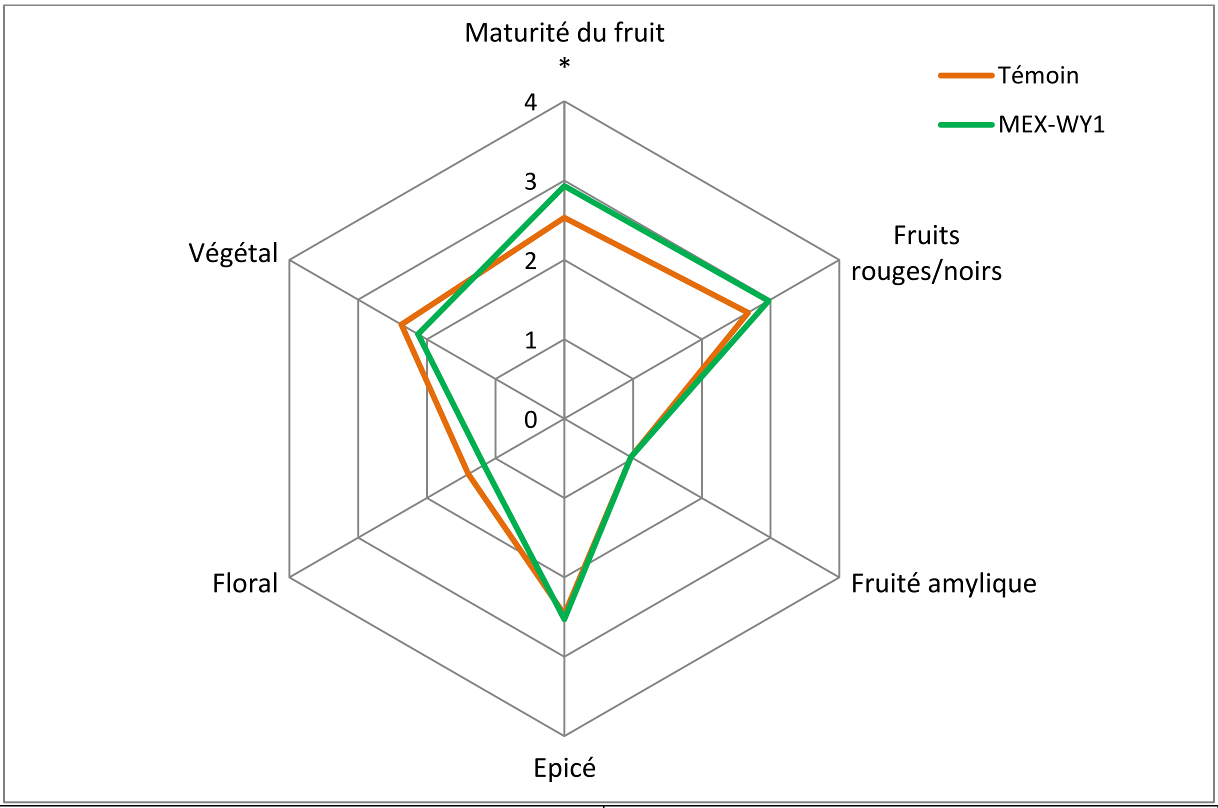


Figure 6: Taste analysis by an expert panel (La Rioja, Spain, March 2017) of a Cabernet Sauvignon (Paso Robles, California 2016) made either with the specific autolysate MEX-WY1 added at a rate of 30g/hL at the beginning of fermentation (MEX-WY1 treatment) or without (Control treatment)

- Enhanced fruit maturity:

In a number of tests, some unexpected differences in aroma were perceived, including fruit maturity and vegetal and grass characteristics. For a Cabernet Sauvignon (Bordeaux, France, 2016) made from grapes harvested just before maturity, the same must was fermented under strictly identical conditions, except for the addition of the specific autolysate MEX-WY1 at a rate of 30g/hL at the beginning of fermentation in the MEX-WY1 treatment (Figure 7). Both wines were tested by a panel of second-year student enologists (DNO Toulouse, March 2017). The MEX-WY1 treatment produced a significant difference (10% confidence level) in "fruit maturity," i.e. more mature fruit notes, compared to the control. The control wine was considered to be slightly more vegetal and the MEX-WY1 wine to have more red/black fruit notes.



français	**anglais**
Maturité du fruit	Fruit maturity
Végétal	Vegetal
Floral	Floral
Épicé	Spicy
Fruité amylique	
Fruits rouges/noirs	Red/black fruit
Témoin	Control

Figure 7: Aroma analysis by a panel of second-year student enologists (Toulouse, France, March 2017) of a Cabernet Sauvignon (Bordeaux, France, 2016) made with the specific autolysate MEX-WY1 at a rate of 30g/hL at the beginning of fermentation (MEX-WY1 treatment) or without (Control treatment)

Conclusion

Recent research has given us a better understanding of how yeasts and phenolic compounds in red wine interact, enabling us to better characterize the biochemical and biophysical properties of yeast. Thanks to these advances, a specific autolysate with unique properties was developed. A wine yeast with very special characteristics was subjected to a MEX treatment, giving rise to the specific autolysate MEX-WY1. Added early on in the fermentation process, the specific autolysate MEX-WY1 has a positive effect on sensory characteristics such as colour, mouthfeel, and fruitiness in red wine

MEX-WY1 is marketed under the name OPTI-MUM RED™.

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