**Effect of different ageing techniques on the polysaccharide and phenolic composition and sensorial characteristics of Chardonnay white wines fermented with different selected *Saccharomyces Cerevisiae* yeast strains**

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**ABSTRACT**

Polysaccharides, mainly mannoproteins, play an important role on technological and sensory quality of wines. These compounds are released to the wine during alcoholic fermentation and, thereafter, during yeast autolysis in the ageing process.

The effect of the ageing on lees and other alternative ageing techniques was studied in Chardonnay Chilean white wines previously fermented with different *Saccharomyces cerevisiae* yeast strains (Zymaflore VL2 and Lalvin CY3079).

The Lalvin CY3079 released higher amounts during alcoholic fermentation and during the first stages of the ageing than the Zymaflore VL2, which could produce a positive effect on the colour intensity of wines. Some statistically significant differences on phenolic families and sensory attributes were observed after alcoholic fermentation, during the ageing period and bottle storage but this depended on the yeast strain used, the ageing technique applied and the period of ageing and bottle storage analysed.

**Keywords:** Polysaccharides, phenolic compounds, white wines, *Saccharomyces* *cerevisiae* yeast strains, TDS sensory analysis.

**1. Introduction**

 Over the years, the wine industry has made several efforts in order to continue improving the quality of white wines adapting, on one hand, to the consumers’ demand, whose demand more complex wines with more fruity, varietals and fresh aromas in nose and balanced in mouth and, on the other hand, extending the range of quality wines [1].

In last few years, several scientific studies have postulated that yeast polysaccharides (mainly mannoproteins) play an important role in the technological and sensorial characteristics of wines. They can improve the tartaric [2] and protein stability [3,4], reduce wine astringency and bitterness [5-8], enhance the mouthfeel [5, 7, 9] and improve the aromatic complexity and persistence [9-11]. In addition, they can adsorb phenolic compounds [12-13], preventing the oxidation and, therefore, the formation of browning compounds of white wines [5,14,15]. Mannoproteins are released in the wine during alcoholic fermentation [16] and, thereafter, during yeast autolysis [3,17]. They are synthesised in the cytoplasm of yeasts but they aren’t entirely used in the cell wall synthesis [18], and the excess could be released into the must during the alcoholic fermentation [19]. This liberation occurs during the active growth phase of the yeasts and their final content after alcoholic fermentation depended on the initial concentration of colloids in the must [20]. On the other hand, winemakers have used the ageing of white wines on lees after alcoholic fermentation since several years ago in order to release into the wine higher amounts of polysaccharides (mainly mannoproteins) by the autolytic processes and, thereby, obtaining higher quality wines. This technique is performed, in most of the cases in oak barrels increasing the complexity of wines as the oak wood can provide other polysaccharides (different to the yeast polysaccharides) that can improve the sensorial characteristics of the wines [5]. However, this technique also entails a cost that, in some cases and given the current global wine market, nowadays cannot be maintained [21]. This fact forced winemakers to search alternative techniques, such as the use of oak wood chips with or without toasting, in order to obtain wines with similar characteristics to those produced in oak barrels but reducing costs. The use of oak wood chips with different toasting levels can provide volatile compounds which can modify the aromatic characteristics of wine and increase the complexity.

However, despite the positive effects referred to above, the liberation of the polysaccharides during the ageing of wines on lees is too slow, because the temperature and pH conditions are not the most suitable for this process [22].In addition, these techniques can also involve many disadvantages, such as greater demands on winery resources, namely more staff to perform the ‘batonnage’ and longer wine storage times, which increase the price of the final product, as well as the appearance of reduction notes [23]. The ageing of wines on lees, with or without oak wood chips, may involve some microbiological alterations due to the development of spoilage microorganisms such as *Brettanomyces* [23,24]. Therefore, commercial yeast derivative products from *Saccharomyces cerevisiae*, such as inactive dry yeasts, are being marketed in order to provide similar benefits as the yeast lees but in a shorter period of time and reducing their disadvantages [9]. Thus, the wineries can put the wine in the market in a shorter period of time. Inactivated dry yeast belong to a wide group of yeast derivative products, which presented different structure and chemical composition due to their different inactivation (thermal or enzymatic), extraction and purification processes [25]. Today we still don’t have a complete description of the mechanism of action of yeast derivatives on the composition and quality of wine, but there are recent studies that establish an important basis on their action mode and their effect on the quality of wines [12,13]. These studies explain the potential implications of the use of some yeast derivatives on winemaking due to their capacity to interact with phenolic compounds.

As mentioned above, yeast cells can release polysaccharides during the wine fermentation which can contribute to the wine quality. Some studies have been carried out in last years in order to isolate and develop yeast strains that secrete higher amounts of mannoproteins using different genetic methods [3,26]. For this reason, the use of some of these selected yeast strains have gained interest in the wine industry because they are capable of releasing mannoproteins more rapidly into the wine, minimising production costs as well as the technological and microbiological disadvantages involved in the ageing on lees.

No studies have been found relating to the effect of these techniques on the quality of Chilean white wines. For this reason, the objective of this work was to study the effect of the ageing on lees, with or without oak wood chips, with a commercial inactive dry yeast and previously fermented with different *Saccharomyces cerevisiae* yeast strains, which exhibit different capabilities for polysaccharide release, on the quality of Chardonnay Chilean white wines.

**2. Materials and methods**

**2.1 Winemaking and treatments**

All fermentation process was carried out in Santa Carolina Winery. Approximately 100 tons of Chardonnay grapes variety were used and supplied by the winery. The grapes were harvested with 22.6 ± 0.2 ºBrix and the total acidity value was 4.45 ± 0.01 (g L−1 of H2SO4) and the traditional winemaking process in white wines was followed. The alcoholic fermentation was carried out in two different 40000 L. stainless steel tanks which were inoculated with different *Saccharomyces cerevisiae* yeast strains. One tank, was inoculated with Zymaflore VL2 (Laffort, Bordeaux, France) **(VL2)** and the other with Lalvin CY 3079 (Lallemand-South America, Santiago, Chile) **(CY) (figure 1).** Both yeast strains were used in a dose of 30 g HL−1.

Alcoholic fermentation was carried out at a controlled temperature of 16 (± 2) °C and, once it was completed, we followed the methodology described by Del Barrio-Galán et al. (2015) [21]. Briefly, the wines were racked off to other tanks for 5 days to permit the sedimentation of gross lees, and they were then racked off again. Then, 500 L of each type of fermented wine was transported to the pilot plant of the Department of Agro-industry and Enology of Agronomical Sciences Faculty of Chile University and the free SO2 was adjusted to 35 mg L−1. Each type of wine was divided in different 25-L food-grade plastic tanks and stored for 5 days to favour the sedimentation of fine lees. Then, the wines were racked again and the fine lees were collected. Different treatments were performed in triplicate (**figure 1**) and lasted 6 months: control wines (fermented wines without any treatment) **(C)**; wines treated with fine lees (3% v/v) **(L)**; wines treated with fine lees (3% v/v) and French (*Quercus petraea*) oak wood chips (3 g L−1 medium toasted degree of Noble Sweet, l’oenologie du bois (Laffort, France)) **(L+CH)**; which have a length between 7 and 20 mm; and wines with commercial inactive dry yeast added (30 g HL−1 of OPTILEES supplied by Lallemand-South America, Santiago, Chile) **(CIDY)** that were rich in low-molecular-weight polysaccharides, mainly mannoproteins (according to the supplier specifications). All the wines were homogenized with lees, chips and commercial inactive dry yeast through two batonnages per week during the first two months. Over the next four months, only one batonnage per week was performed in order to prevent wine oxidation and microbiological alteration. After the ageing period, the wines were filtered and bottled and were stored for six months.

**2.2. Chemical reagents**

 The standards of gallic, protocatechuic, caffeic, syringic, *p*-coumaric, ferulic, ellagic and caftaric acids, tyrosol, thyptophol, quercetin, myricetin, astilbin, (+)-catechin and (−)-epicatechin, dextrans and pectines were purchased from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA). Polyethylene membranes of 0.45 and 0.22 μm pore size were acquired from EMD Millipore (Billerica, MA, USA). Sodium sulfate (anhydrous), potassium metabisulfite, vanillin (99%), ethyl acetate, diethyl ether, sodium hydroxide, acetic acid, formic acid, sulfuric acid, ethanol, hydrochloric acid and high-performance liquid chromatography (HPLC) grade acetonitrile, methanol and ammonium formiate were purchased from Merck (Darmstadt, Germany). All the reagents were of analytical grade or higher.

**2.3. Analytical methods**

 Classic oenological parameters were evaluated following official analyses methods [27]. The total (TA) and volatile (VA) acidity, pH (Mettler Toledo Sevencompac pH/ion S220, Santiago, Chile), SO2F (sulfur dioxide free) and SO2T (sulfur dioxide total) and alcoholic degree (Aº) were evaluated following the OIV official analytical methods.

The content of total polyphenols [28] and total tannins [29] was expressed as mg L−1 of gallic acid and g L−1of (+)-catechin, respectively. The colour intensity (CI) was evaluated by the absorbance at 280 nm using the method described by OIV 2012 [27]. These measurements were performed using a UV/VIS 1700 Pharmaspec spectrophotometer (Shimadzu, Kyoto, Japan).

The polysaccharides were analyzed using the methodology described by Ayestarán et al., 2004 [30]. 25 aliquot of white wine was centrifuged 3500 rpm during 30 minutes. Then, 10 mL of supernatant was concentrated until 2 mL in a vacuum rotavapour at 35 °C. After that, 10 mL of acid ethanol solution 0.3 M was added to the aliquot of 2 mL of wine in a centrifuge tube and maintained in refrigeration at 4 °C during 24 hours to favour the precipitation of polysaccharides. Then, the sample was centrifuged at 3500 rpm during 20 minutes and de supernatant was discarded. The precipitated fraction was washed three times with 0.35 mL refrigerated ethanol and dried at 50 °C. Finally, it was reconstituted with 1mL of ammonium formiate and filtrated with 0.22 µL membrane in a HPLC vial. HPSEC-RID was performed using an Agilent 1260 Infinity Series liquid chromatograph (Agilent Technologies, Santa Clara, CA, USA) equipped with a G1362A refractive index detector (RID), a G1311B quaternary pump, a G1316A column oven with two Shodex columns, an OHpak SB-803 HQ and a SB-804 HQ (Showa Denko, Tokio, Japan) connected in series (300 mm × 8 mm I.D.), and a G1329A autosampler.The polysaccharides fractions were quantified using dextrans and pectines (*Leuconostoc mesesteroides*) to prepare the calibration curves. The molecular weight distribution of the different polysaccharide fractions was determined by the columns calibration with dextran standards.

Low-molecular-weight phenolic compounds (non-flavonoids and flavonoids) were analyzed using the methodology described in Peña-Neira, Cáceres & Pastenes (2007)[31]. 50-mL aliquot of white wine, which was extracted with diethyl ether (3 x 20 mL) and ethyl acetate (3 x 20 mL) to concentrate the phenolic compounds. The organics fractions were combined, dehydrated with 2.5 g of anhydrous sodium sulfate and evaporated to dryness under a vacuum at 30ºC. The solid residue was dissolved in 2 mL of a methanol/water (1:1, v/v) solution and filtered through a 0.22-μm pore size membrane. Aliquots (25 μL) of the final solution were subjected to reverse-phase chromatographic separation. These compounds were analyzed with a HPLC 1100 Series system (Agilent Technologies, Santa Clara, CA, USA) consisting of a G1315B photodiode array detector (DAD), a QuatPump G1311A quaternary pump, a G1379A degasser and a G1329A autosampler. A reverse phase Nova-Pak C18 column (4 μm, 3.9 mm i.d. × 300 mm; Waters, Milford, MA, USA) was used for HPLC-DAD analysis of individual phenolic compounds at 20ºC. Each major peak in the HPLC chromatograms of the extracts was characterized by retention time, the absorption and the spectrum form (from 210 to 360 nm). The acquisition time was 1 second. The calibration curves at 280 nm were produced by injecting the standard solutions before an extraction under the same conditions as the samples analyzed over the range of concentrations observed (r2 ≥ 0.93). The quercetin glycosides, dimmeric procyanidins, gallates and stilbenes glucosydes, for which no standards are available, were quantified using standard curves for quercetin, (+)-catechin, gallic acid and *trans*-resveratrol, respectively.

**2.4. Sensory analysis**

The analysis of the wines was performed by a trained sensory panel of 10 people who were all workers and students at the Department of Agro-Industry and Enology of Agronomical Sciences Faculty of Chile University. The wines were evaluated in individual temperature-controlled tasting booths using a completely randomized order. 20 mL of each wine were served at 18–19 ºC in dark wine-tasting glasses (RCristal, Mendoza, Argentina) labeled with a three-digit code. The dark wine-tasting glasses were used to prevent the interference of visual sensations and to focus the attention of the panellists on gustative sensations. Between each sample, the panellists have a break for 1 minute to chew on a cracker and then rinsed their mouths with water. The FIZZ software (Biosystemes, France) was used. This is a visual tool for delivery of the methodology, automation and data collection [32]. The methodology used was the Temporal Dominance of Sensations (TDS), which allows to observe the progression of the dominance of each attribute studied over time [33]. The attributes evaluated were: sweet, bitter, acid, alcohol and preserved fruits. A gustative protocol was used: the tasters used one hand to bring the sample to their mouth, while using the other to click the “Start” button activating the stopwatch, and checking the dominant attribute throughout. After 12 seconds the sample was spit out. Sample evaluation was over when the taster could no longer perceive any attribute, or after a maximum of 100 seconds [34].

All the analyses were performed after alcoholic fermentation (AAF) and after 2, 4 and 6 months of ageing treatment (2MT, 4MT and 6MT respectively) and after 3 and 6 months of bottle storage (3 MB and 6 MB, respectively).

**2.5. Statistical analysis**

All the data were treated using the analysis of variance (ANOVA). The Tukey’s honestly significant difference (HSD) was used in order to determine statistically significant differences between the means, with a significance level of 95% (*P* < 0.05). All the statistical analyses were conducted using Statgraphics Centurion Version 15.2 (StatPoint Technologies, Inc., Warrenton, VA, USA) and Excel 2007 Version 12.0 (Microsoft Corp., Redmond, WA, USA).

For the sensory analysis with TDS, two different lines are represented in the graphs: the “chance level” indicates that if the dominance rate of a descriptor is below this line, the attribute is considered to have been quoted by chance. The “significance level” (*p*<0.05) indicates that if the dominance rate exceeds this curve, the attribute is considered to be significantly dominant. When the attribute curves are between these two lines, the descriptor is dominant but not significantly.

**3. Results and discussion**

**3.1. Effect on the classic oenological parameters**

 The data ranges of these parameters were: alcoholic degree between 12.5 and 13.6 (% of ethanol); pH between 3.53 and 3.75; SO2F between 35.2 and 12.8; SO2T between 96.0 and 148; volatile acidity between 0.30 and 0.60 (g L−1 acetic acid); and total acidity between 2.85 and 3.43 (g L−1 of sulphuric acid). No statistically significant differences were found between the wines fermented with both VL2 and CY yeast strains. Same results were found between the different ageing treatments applied. Other studies carried out in white and red wines postulated that similar techniques did not produce differences in these parameters [5,9,21].

**3.2. Effect on the different polysaccharide fractions and total content**

 Three different polysaccharide fractions (H, M and L) were obtained and classified according to their molecular weight. H corresponded with the highest molecular weight polysaccharides (320-370 kDa), M with the medium molecular weight (40-65 kDa) and L with the lowest molecular weight (≥10 kDa). The total polysaccharide content corresponded to the summary of H, M and L fractions (**Figure 2)**. Fraction M presented the highest content in VL2 and CY wines (52.1 mg/L and 47.1 mg/L respectively) after the alcoholic fermentation period (AAF), but no statistically significant differences were found. However, the CY wines showed a significantly higher content of H and L fractions than VL2 wines, which resulted in a significantly higher concentration of total polysaccharides (18.1% higher). As occurred in a similar study carried out by our group on red wines but used other yeast strains [21,35], these differences between both yeast strains were maintained after 2MT period, but after 4MT period were lower. The content of total polysaccharides increased using the L, L+CH and CIDY ageing techniques. This content also increased in the control wines, most likely due to a small portion of the fine lees which could pass to the control wine in the racking process during the winemaking, releasing some polysaccharides. However, as expected, this increase was more statistically significant in the wines treated with the different ageing techniques than in the control wines throughout the entire ageing process, being higher after the 2MT period. These differences were maintained during the bottle storage. Similar results were found by other authors using similar ageing techniques in white wines [9,11]. Del Barrio-Galán *et al*. (2010)[11] studied the effect of 6 commercial yeast derivatives with different content on polysaccharides on the quality of Verdejo white wines. It reported that all of these products presented higher content than the control wines after 2 months of ageing. On the other hand, Del Barrio-Galán *et al*. (2011)[9] also study the effect of the ageing on lees and 3 different commercial yeast derivatives rich in polysaccharides lasting 2 months, and then the 6 month bottle storage on the quality of Verdejo white wines. They reported that all of the treated wines presented higher content of polysaccharides than the control wines after the ageing period. However, after 6 months in bottles, only 2 of these commercial yeast derivatives released higher content compared to the control wines.

**3.3. Analyses of phenolic compounds**

 **Figure 3** shows the trend of the total polyphenols (TP) and total tannins (TT). In general terms, the wines fermented with the CY yeast strain contained a lower content of TP than those fermented with the VL2 yeast strain after the AAF period, throughout the ageing period and bottle storage, with the exception of the wines treated with L+CH. These results could be explained by the adsorption or retention phenomena produced by the higher content of polysaccharides released from CY yeast strain [1,6,9,11,36-38]. The wines assayed with the different ageing techniques presented similar or higher content of these compounds than the control wines throughout the ageing period and the bottle storage. As expected, the higher content was observed in the wines treated with L+CH in both CY and VL2 wines because some of these compounds (mainly ellagic and gallic acids) can be extracted from oak wood [39]. The content of TT was higher in the CY wines than in the VL2 wines after AAF period. All the CY treated wines showed lower content of these compounds than the VL2 treated wines after the ageing period and bottle storage, with the exception of those treated with L+CH which were higher in CY wines. Some differences were also found between the different ageing techniques assayed. After 2 MT period, all the VL2 treated wines and the CY wines treated with CIDY contained lower TT than the control wines, being these wines which presented the lowest values in both types of fermented wines. After the ageing and 3MB periods, only the VL2 and CY wines treated with CIDY showed lower concentration than the control wines. Finally, at 6MB period, all the treated wines presented similar or higher content than the controls.

 **Tables 1 and 2** show the different low molecular weight phenolic compounds identified and quantified. Tyrosol, astilbin, *trans*-caffeic acid and protocatechuic acid were the compounds with the highest concentrations after the AAF period, ageing period and bottle storage. The content of hydroxycinnamic acids (HCA), flavanol monomers, procyanidins and flavonols was lower in CY wines than in VL2 wines after the AAF period. As occurred with TP, there was an important retention/adsorption effect on these compounds by CY yeast strain. In general, this effect was maintained during the ageing period and bottle storage for HCA (mainly due to the effect on the *trans*-caffeic acid) and flavonol compounds, while for the flavanol monomers and procyanidins was only maintained during the aging period. In the case of the phenolic alcohols, both CY and VL2 fermented wines presented similar concentrations after AAF period, but the CY wines had a higher content during the ageing period and bottle storage, mainly due to a higher content in the tyrosol. These compounds are formed by deamination and decarboxylation reactions of tyrosine and tryptophan amino acids, respectively, during yeast fermentation [40].

Regarding the ageing treatments, the wines treated with L+CH showed significantly higher content of hydroxybenzoic acids (HBA) than the rest of treated and control wines throughout the study. As occured with TP, this fact was mainly due to the release of several of these compounds by the oak wood as gallic, ellagic and protocatechuic acids [9,35,41]. No clear differences were found between ageing treatments with lees and control wines. For hydroxycinnamic tartaric ester acids (HCTEA), the most significant differences were found in CY wines, observing that all the treated wines had lower content than the controls after 2MT period and 6MB. Some authors have postulated that, in white wines, this fact implies a lower oxidation risk by reducing the content of easily oxidizable compounds such as these tartaric esters [9,11]. However, this effect was not found in the VL2 treated wines at the time of the study.

In the case of flavanol monomers and procyanidins, only some differences were found at the end of the ageing period. Thus, the VL2 wines treated with CIDY presented lower content of flavanol monomers than the control wines, but both VL2 and CY wines treated with L+CH presented higher content of procyanidins. During bottle storage only the procyanidin B3 was detected and no statistically significant differences were found.

As mentioned above, astilbin was the most important flavonol detected, and several astilbins derivatives were also found. Quercetin-3-galactoside, quercetin-3-glucuronide, quercetin-3-glucoside were only detected after the AAF period. The most important differences between the treatments applied were found after 3MB period, observing that wines treated with L+CH showed higher content than the control wines.No clear effects were found in the content of total stilbenes. Few differences were also found between the ageing treatments applied in the phenolic alcohols content during the ageing period. After bottle storage, the VL2 wines treated with L and CIDY as well as the CY wines treated with L showed lower content of these compounds than the control wines. However, the CY wines treated with CIDY presented higher content than the control wines. For this reason it is difficult to establish a correlation between the different ageing treatments and evolution of these compounds during bottle storage. In a study carried out by Del Barrio-Galán *et al*., (2011)[9] using similar techniques, no statistically significant differences were found during the same bottle storage in Verdejo white wines.

Recent studies about the interaction/adsorption of polyphenols by yeast, inactive yeast and yeast cell walls hypothesized that not only do the mannoproteins of yeast and yeast derivatives interact with phenolic compounds in solutions, but also some polyphenols can be adsorbed and interact directly with the cytoplasmic membrane lipids of the yeasts. This could be have a protective effect on the oxidation of white wines [12,13]. Moreover, in vitro experiments proved that yeast membrane sterols could be likely involved in the yeast’s ability to adsorb polyphenolic compounds and mainly the colorless intermediate compounds of the browning reactions [42].

**3.4. Colour intensity**

The colour intensity (CI) parameter is a good indicator of the oxidation degree of white wines, which tend to change colour to brown tones due to the oxidation of their phenolic compounds increasing their CI value. As showed in **figure 3**, the wines fermented with CY yeast strain showed statistically significant lower values of CI than those fermented with VL2 after AAF period. This result was maintained throughout the ageing and the bottle storage periods. The result could indicate an important yeast strain effect on the prevention or reduction of the browning of white wines. However, the different ageing treatments assayed did not produce this effect during the ageing period in both types of fermented wines. Only during bottle storage some statistically significant differences were found. Thus, VL2 wines treated with L and CIDY presented lower values of CI than control wines throughout all the bottle storage. This result is well correlated with the higher amounts of polysaccharides released during the ageing period. Similar results were found by some authors using the same ageing on lees technique and other yeast derivatives products in white wines after three months of bottle storage [1,9]. They proposed to use these techniques as fining agents to prevent the browning of white wines [9]. However, this result was not found in the CY treated wines. Finally, both VL2 and CY wines treated with L+CH presented higher CI values than the control wines throughout the ageing period and bottle storage. This result could be explained due to the higher release of TP of oak wood chips which were probably oxidized producing a higher browning of the wines. The objective of combining lees and chips was to prevent this oxidation process through a higher release of polysaccharides from the autolysis of lees, however this effect was not found.

**3.5. Sensory analysis**

The **figures 4, 5 and 6** show the results obtained with TDS sensory analysis for the different attributes evaluated. In general, it was observed that, in all the wines evaluated, the panel tasters detected the attribute of sweet in first place when they put the wines in their mouth. It is important to note that, according to the supplier specifications, the use of oak wood chips with a medium toasted degree could produce a sweetness sensation (compared to other treatments) but this sensation was not found by the tasters. Then, when they spat out the wine (at 12 seconds) they detected mainly the acid, alcohol and bitter attributes. These 3 attributes were dominant, but their dominance was different in function of the yeast used, the treatment performed, and the ageing and bottle storage periods studied. Thus, the most significant differences were found after 2MT, 6MT and 6MB periods. In this way, after 2MT period, the alcohol attribute was significantly more dominant than the rest attributes in the VL2 control wines, and it was higher than the CY control wines. However, the CY wines treated with L and L+CH showed higher values of alcohol than the same treatments in VL2 wines. The higher differences were found in the treatments with CIDY. Thus, the VL2 wines had a higher percentage of dominance rate of bitter, alcohol and acid than the CY wines. Furthermore, it was observed that following the trend curves for each attribute, the CY wines treated with CIDY were more balanced than VL2 wines because their attributes trend curves were found in a more similar dominance range. After 6MT period, the tasters note that the VL2 control wines had a higher dominance rate of the sweet and bitter attributes than the CY control wines, but they had a higher acidity and preserved fruits than the VL2 control wines. The most important differences were found in the wines treated with L and CIDY. Thus, VL2 L and CIDY wines showed higher dominance of bitter (around 70% and 55% respectively) than the CY L and CIDY wines (around 44%). Conversely, CY CIDY wines had higher notes of preserved fruit than the VL2 CIDY wines. The VL2 L+CH wines presented a higher dominance rate of acid than the CY L+CH wines. In view of this data, it can be said that the CY wines were, in general terms, more balanced in the mouth than VL2 wines because the trend curves of the attributes evaluated were in a more similar dominance range. The differences found between CY and VL2 wines after 6MB period were lower. The CY control wines had higher dominance rate of bitter than VL2 control wines and, this attribute was more prolonged over time. In addition, these wines had higher dominance rate of alcohol attribute. Similar results were found in wines treated with CIDY but only for the alcohol attribute, showing both types of fermented wine similar dominance for the bitter. Comparing the different ageing treatments assayed in both VL2 and CY wines with the controls, an improvement in the sensory quality was only seen in a few cases. Thus, after 2MT period, the tasters observed that the VL2 control wines presented a higher alcoholic character than different treated wines with the exception of VL2 CIDY. In addition, these wines presented higher bitter character. In the case of the CY wines, L+CH wines had higher alcoholic character than the control and the rest of treated wines. After 6MT period, the dominance rate of bitter was higher in the VL2 L and CIDY wines than in the rest, mainly in L wines. Finally, the wines treated with L+CH presented higher acid character respect to the rest of wines. In the case of CY wines, the control and CIDY wines presented higher dominance of acid and preserved fruit attributes than the L. After 6MB period, the VL2 control wines presented an important acid character respect to the treated wines which it was manifested during the first seconds of the tasting. Contrary that we expected find, all the treated wines had higher bitterness than the controls and it was more persistent in the time. On the other hand, the control CY wines had higher bitter and alcohol dominance than the treated wines, with the exception of those treated with CIDY. Besides, the bitter attribute persistence was higher in the control wines than in the treated wines.

**4. Conclusions**

CY yeast strain permitted a faster release of polysaccharides (probably mannoproteins), mainly of low molecular weight, during alcoholic fermentation. The use of its lees for the ageing treatments permitted a faster release of the yeast polysaccharides (probably due to a better autolytic capacity) during the early stages of ageing (2MT) than VL2, but this one released higher amounts during the later stages (4MT) than CY. These results suggested that VL2 lees released the polysaccharide content more slowly than CY.

It can be say that, under our study conditions, CY yeast strain had a significant adsorption/retention effect on the phenolic compounds, which could have a prevention effect of the browning of white wines.

Based on the results obtained with TDS sensory analysis, in general terms, there was an important yeast strain effect on the modulation of some attributes, mainly bitter, acid and alcohol after 2MT and 6MT periods, but it was depended of the treatment and the ageing period studied. This is the first time that Chilean white wines have been evaluated with this technique, and more studies should be carried out for a better understanding of the effect of these techniques on the sensory quality of white wines.

**ACKNOWLEDGMENTS**

The authors are grateful to CONICYT-Chile (FONDECYT Postdoctoral Project Nº 3130482, FONDECYT grant 1140882) for funding this study. The authors are also grateful to Lallemand-Chile for providing the *Saccharomyces cerevisiae* CY3079 yeast strain and the commercial inactive dry yeast (OPTILEES), and to the Santa Carolina winery for providing the wine for the study.

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**FIGURE CAPTIONS**

**Figure 1**. Scheme of the experiences carried out.

**Figure 2.** Total polysaccharide (A) and different polysaccharide fractions concentration (B: fraction H; C: fraction M and D: fraction L) (mg L−1) of wines. AAF (after alcoholic fermentation), 2MT (2 months of treatment) and 4MT (4 months of treatment), 6MT (6 months of treatment), 3MB (3 months of bottle storage) and 6MB (6 months of bottle storage). Values with different letters indicate statistically significant differences for *p* < 0.05.

**Figure 3.** Total polyphenol (TP) (mg L−1) and total tannins (g L−1) concentration, and colour intensity (units of absorbance) of wines. Values with different letters indicate statistically significant differences for *p* < 0.05.

**Figure 4.** Sensory analysis graphics of the different wines after 2 months of treatment (2MT).

**Figure 5.** Sensory analysis graphics of the different wines after 6 months of treatment (6MT).

**Figure 6.** Sensory analysis graphics of the different wines after 6 months of bottle storage (6MB).