BIO Web of Conferences **9**, 02004 (2017)
40th World Congress of Vine and Wine
DOI: 10.1051/bioconf/20170902004

The effect of inactivated yeast-based products on the process of wine aging, phenolic compounds and sensory characteristics of red wine Prokupac

Marko Malićanin[1], Vladislav Rac[2], and Vesna Rakić[2]

[1] Rubin AD, Nade Marković 57, 37000 Kruševac, Serbia
[2] Faculty of Agriculture, University of Belgrade, Nemanjina 6, 11080 Zemun, Serbia

Abstract. Keeping of red wine in bottles is very important for its maturation and quality. However, there are numerous changes that happen during that period, usually caused by oxidative processes and changes in structure and content of polyphenolic compounds. The goal of this study was to determine the effects of Inactivated dry yeast (IDY) products on aging, phenolics content, colour stability and sensory characteristics of red wine Prokupac (Serbian autochthonous variety). The treatment of wine was done by 3 different IDYP (Lallemand, Canada): Optimum White, Opti Less and Noblesse, applied as 0.2 g/L and 0.4 g /L during 15 days. Subsequently, wine clarification was done, followed by filtration and bottling. Untreated wines were used for comparison. Wine was subjected to accelerated aging (10 days at 55°C) and also to normal aging conditions during one year. To determine the influence of IDYP following parameters were monitored: dissolved O_2, free and total SO_2, sugar free extract, content of total phenolics, flavonoids, flavan-3-ols and anthocyanins, percent of polymeric color, color tint, color intensity and sensory characteristics (Panel method). The obtained results clearly show that IDY products are good scavengers of oxygen and have a positive impact on wine quality preservation and its organoleptic characteristics. However, a slight decrease of polyphenols content was detected.

1. Introduction

Inactivate dry yeast (IDY) products are derivatives of yeasts produced from enological variety (Saccharomyces cerevisiae) which have been pre-treated in order to inhibit their fermentative activity. Usually, these products are obtained by growing *Saccharomyces cerevisiae* yeasts in a sugar-rich medium, after which the yeasts are autolysed and dried to obtain final products (in powder form) which, under the generic name of IDY products, may contain inactive yeasts, yeast autolysates, yeast extracts, yeast hulls or walls [1]. In recent years, commercial use of such products in winemaking industry is growing and, according to their composition, IDYP are recommended for various reasons [2,3]. Namely, many bioactive compounds such as peptides, amino acids, gluthation and polysaccharides (mannoproteins) are released during yeast autolysis. Consequently, they can act as: alcoholic and malolactic fermentation enhancers, protective agents to improve active DY rehydration or as organoleptic enhancers. The stabilization of red wine colour is also possible by using mannoprotein-rich IDYPs [3]. Concerning sensory properties and stability, the goal is usually to obtain characteristics similar to those achieved by ageing on lees [2,4].

However, despite the fact that many of these products are currently in the market under different brand names claiming different wine improvements, scientific information about the chemistry behind their application is still insufficient and the mechanisms responsible for the claimed wine's improvements are not completely clear. Moreover, insufficient research on the application of IDYPs in winemaking conditions has been done, since it seems that the application of these products is based mostly on experience acquired during production of wine in small cellar trials. Therefore, well established scientific experiments devoted to the effects that these products induce in wines are needed in order to achieve better understanding of the mechanism of IDYP action.

Having all of previously mentioned in mind, the objective of the present study is to characterise the effect of different products based on yeast autolysates on the wine aging process, phenolic compounds and sensory characteristics of red wine Prokupac during storage in bottles. This work constitutes a primary approach in understanding the action mechanism of IDYP and establishing better criteria for their use in winemaking.

2. Materials and methods

2.1. Materials

Red wine Prokupac (Serbian autochthonous red grape variety), produced from grapes from "Rubin" vineyards, central Serbia wine region and Tri Morave wine sub region, vinified in 2015, was assayed. Catechin, gallic acid and vanillin were from Sigma. Folin–Ciocalteu reagent and sodium bisulfite were from Fluka.

2.2. Methods

Prokupac grapes were harvested at technological maturity, based on indices of sugar content and acidity established

Table 1. Wine sample labeling and treatment description.

Sample	IDYP used	Concentration of IDYP
BL	/	/
OW20	Optimum White	0.2 g/L
OW40	Optimum White	0.4 g/L
OL20	Opti Less	0.2 g/L
OL40	Opti Less	0.4 g/L
N20	Noblesse	0.2 g/L
N40	Noblesse	0.4 g/L

by the Vine & Wine Institute (Paris, France), destemmed and crushed. Pomace were inoculated with selected yeast strain (S. cerevisiae var cerevisiae) and fermented under controlled temperature (16–21 °C) to dryness (reducing sugar content below 4 g/L). Maceration lasted nine days. After months of wine care, the treatment of wine was carried out by 3 different products based on inactive yeast (from Lallemand, Canada) which are available on the market under the names Optimum White, Opti Less and Noblesse. These products were applied in two concentrations: 0.2 g/L or 0.4 g /L during a period of 15 days, with stirring every third day. Denotations and treatment descriptions are given in Table 1.

After treatment, wine clarification was done (using bentonite and gelatin), followed by filtration and bottling. Untreated wines were used for comparison ("blank"). After bottling, the wine in some bottles was subjected to the accelerated aging while the wine in other bottles was stored in the stock under normal conditions, in dark place, at 15–20°C during one year.

Titratable acidity, density, alcohol, reducing sugars and extract content, pH and free and total sulphur dioxide were determined using well-established, standard methods [5]. Dissolved oxygen content was measured by polarographic oxygen analyzer (YSI model 51A disolved oxygen meter, Yellow Springs Instruments CO). Total polyphenol concentration was determined with the Folin–Ciocalteu assay [6]. Gallic acid was used as standard and results were expressed as grams per litre of gallic acid equivalents. Flavonoids content was determined using $AlCl_3$ spectrophotometric assay [6]. Catechin was used as standard and results were expressed as milligrams per litre of catechin equivalents. Flavon-3-ols content was measured using vanilin spectrophotometric assay [7]. Catechin was used as standard and results were expressed as milligrams per litre of catechin equivalents. Anthocyanins content was measured according to Ribereau-Gayon and Stonestreet [8]. Percent of polymeric colour was determined using bisulfite bleaching method [9]. Colour intentisty and tint were measured spectrophotometrically [10]. All parameters were monitored after each of the experimental phases (Table 2) and measured in triplicates.

Panel method was used for sensory evaluation. A panel of five judges was recruited comprising "Rubin" staff with extensive experience in wine sensory evaluation, of which four are members of the Commission of Ministry of Agriculture, Republic of Serbia; appointed for the organoleptic assessment of wine and similar products. 19 descriptors were evaluated: smell profile (overall odor, spice, color intensity, vegetable, red fruit berries, black fruit berries, floral, complexity, intensity and typicality), and taste profile (taste, harmony, acidity, astringency, fullness, complexity, duration, structure and typicality).

Table 2. Abbreviations and descriptions of experimental phases.

Phase abbreviation	Phase description
W	Basic wine
TR	After IDYP treatment
CFB	After clarification and filtration - after bottling
ACC	After accelerated aging test
NOR	After one year of normal aging conditions

3. Results and discussion

3.1. Oenological analyses

Basic oenological parameters (acidity, density, alcohol, reducing sugars, extract, pH, free and total sulphur dioxide) of Prokupac wine before and after the IDYP treatment, and after one year of aging are presented in Table 3. The results show that the treatment had not induced significant changes, which was indeed expected, considering the relatively short treatment duration.

3.2. Oxygen content

Oxygen content values measured in all wine samples after each of the experimental phases are presented in Fig. 1. The results indicate that the three implemented commercial IDYPs exhibit good oxygen scavenging properties, since all treated samples contained oxygen quantities lower than those found in untreated wine. This conclusion holds for all phases of the investigation. Especially low oxygen contents were found after the experimental phase which included IDYP treatment, clarification, filtration and bottling (CFB), regardless of IDYP used. After the accelerated aging test, dissolved O_2 concentrations were also found to be lower in all treated wines than in the blank, which was especially pronounced for the sample treated by "Noblesse", using its concentration of 0.4 g/L (0.81 mg/L of oxygen, compared to 1.23 mg/L for blank). After one year in the bottles, there were no significant differences in O_2 content between treated wine samples and blank.

3.3. Total polyphenol content

Figure 2 presents the total polyphenol contents (TPC) of the investigated wine samples. As a general trend, lower polyphenol concentrations were found for all IDYP treated wines. The observed decrease of TPC can be attributed to partial adsorption of phenolic compounds on yeast cells and yeast walls, which has been observed and discussed in literature [1,2]. Also, the treatment mildly influenced the way TPC changed after clarification, filtering and bottling (CFB). For blank wine, only a slight difference in TPC was observed before and after CFB, while for IDYP treated wine samples these differences were more pronounced: lowering of TPC values was observed in the case of treatment by Optimum White and Opti Less, when amounts 0.4 g/L of IDYP were used, while in the case of Noblesse application, this effect was observed for both 0.2 g/L and 0.4 g/L. After one year of wine aging in normal conditions, relative changes of total

Table 3. Oenological parameters of basic wine and wine samples after treatment and normal aging.

Parameter	Basic wine	After one year of normal aging conditions						
		BL	OW 20	OW 40	OL20	OL40	N20	N40
Density D_{20}^{20}	0.99399	0.99353	0.9936	0.99366	0.9936	0.99396	0.99361	0.99364
Alcohol (%vol)	12.2±0.03	12.17±0.02	12.2±0.015	12.07±0.04	12.22±0.035	12.23±0.03	12.09±0.04	12.22±0.02
Total extract (g/L)	26.00±0.24	24.81±0.31	25.09±0.33	24.86±0.26	25.14±0.35	26.09±0.36	24.77±0.28	24.96±0.21
Total acids (as tartaric) (g/L)	5.6±0.13	5.27±0.15	5.43±0.11	5.37±0.21	5.2±0.18	5.2±0.12	5.37±0.21	5.37±0.22
Volatile acids (as acetic) (g/L)	0.63±0.06	0.54±0.04	0.54±0.045	0.51±0.55	0.51±0.062	0.51±0.33	0.54±0.047	0.51±0.055
pH	3.39±0.03	3.42±0.24	3.43±0.022	3.43±0.015	3.43±0.012	3.43±0.03	3.43±0.026	3.42±0.016
Reducing sugars (g/L)	2.20±0.22	2.42±0.15	2.35±0.16	2.31±0.2	2.25±0.14	2.36±0.23	2.3±0.12	2.37±0.17
Sugar free extract (g/L)	24.80±0.68	23.39±0.86	23.74±0.75	23.55±0.7	23.89±0.55	24.73±0.94	23.48±0.85	23.59±0.69
Free SO_2 (mg/L)	26.88±1.28	26.03±1.21	24.75±0.93	24.75±1.08	25.67±1.12	26.03±1.15	23.04±1.24	24.75±1.15
Total SO_2 (mg/L)	56.32±1.66	46.96±1.93	52.05±1.15	50.64±0.99	52.91±1.42	48.6±1.65	50.01±1.33	49.62±1.55

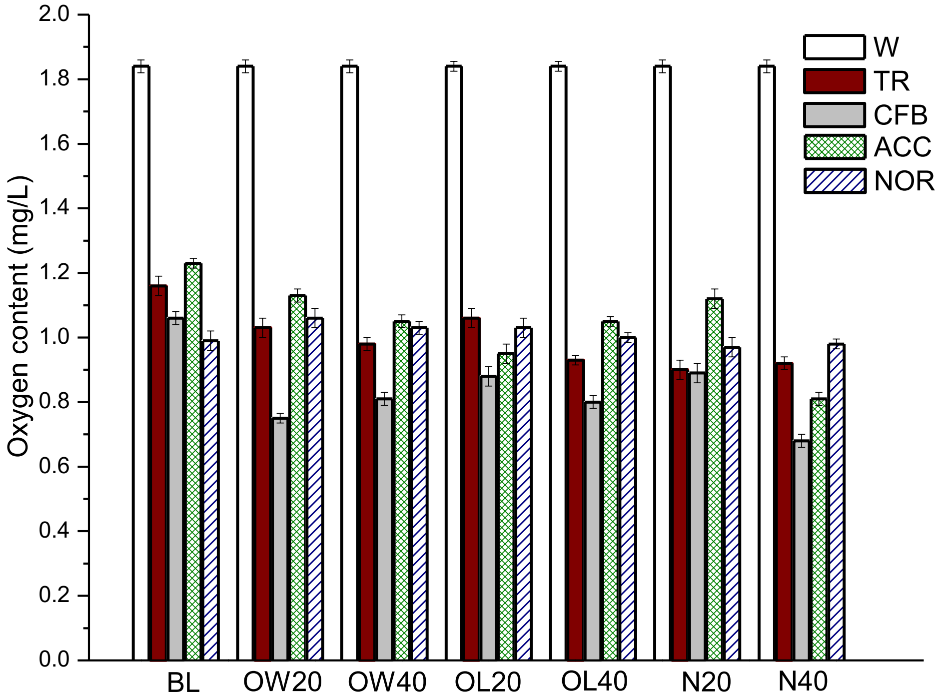

Figure 1. Oxygen content of the investigated wine samples.

polyphenol concentrations (after treatment vs. after aging) were comparable to those observed for blank wine.

3.4. Anthocyanins content

The effects of IDYP treatment on the concentration of anthocyanins in the investigated wines seem to follow a pattern different than the one observed for total polyphenol content. Results presented in Fig. 3 show that similar or only slightly decreased values of anthocyanin contents were detected after the treatment. Again, these changes are probably due to the adsorption of small amounts of anthocyanins on the IDY products used. However, after clarification, filtering and bottling, anthocyanin concentrations decreased to a greater extent in treated wine samples than in blank wine. The largest difference before and after CFB phase was detected when Noblesse IDYP was used. After the accelerated aging phase, values of anthocyanin content in treated wines only slightly differed from the blank wine. However, after one year of aging in normal conditions, higher concentrations were found for the treated samples, compared to the blank, indicating a positive influence of IDYPs on stability of anthocyanins during aging.

3.5. Flavonoids and Flavan-3-ols

The contents of flavonoids and flavan-3-ols in the investigated wines (not presented) showed only slight changes right after treatment (TR) and after clarification, filtering and bottling (CFB). Slightly decreased concentrations were also measured after accelerated aging, for all treated wines. After normal aging, however, noticeably higher values were detected for treated samples, compared to blank wine, which corresponds to smaller relative decreases of flavonoids and flavan-3-ols content during aging, for treated samples.

3.6. Percent of polymeric colour

The percentages of polymeric colour variations in all investigated wine samples, including blank, show a distinct increase after the one year aging period (Fig. 4). This is in accordance with the fact that changes in red wine colour take place as a result of structural changes of the

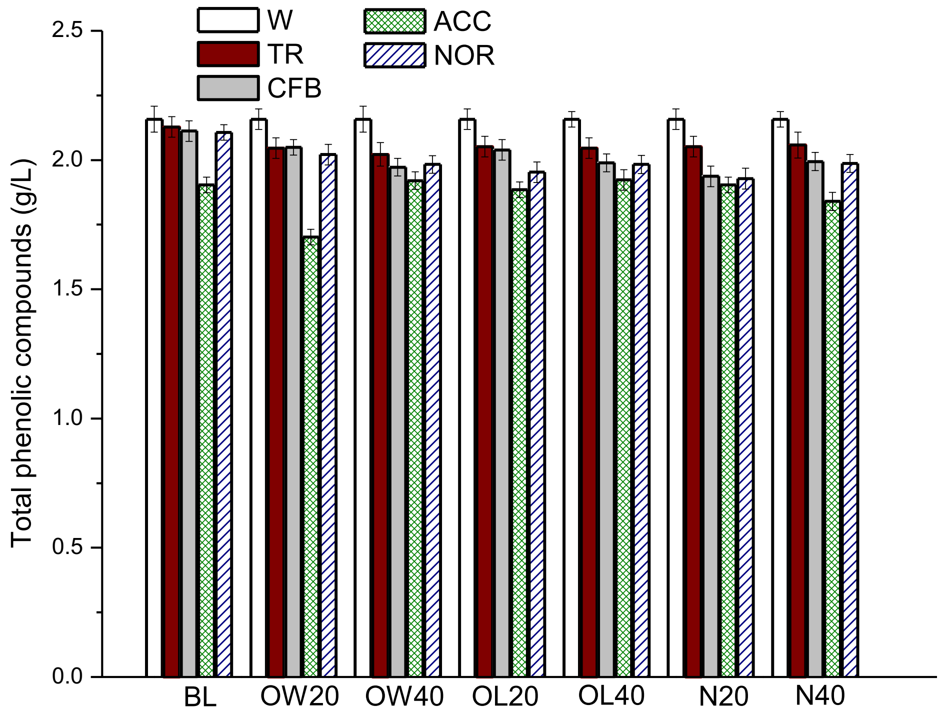

Figure 2. Total polyphenol content of the investigated wine samples.

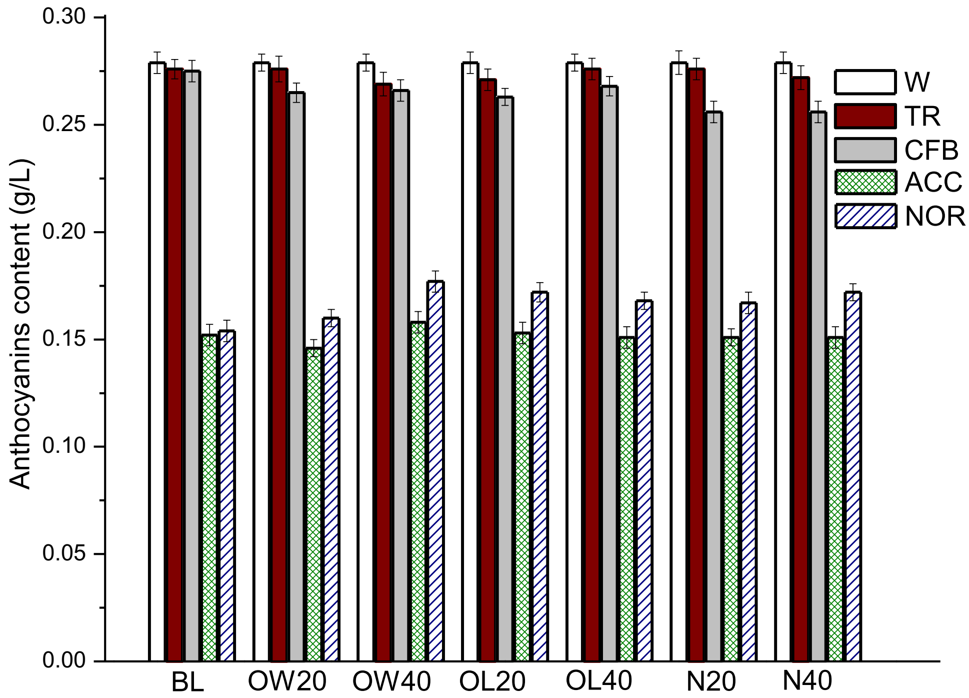

Figure 3. Anthocyanins content of the investigated wine samples.

anthocyanins, from ionized anthocyanins to condensed anthocyanins, which makes the percentage of polymeric colour a parameter of wine ageing. During aging, most of the anthocyanins are subjected to transformations to stable complexes, either with tannins, or between anthocyanins themselves (self-association) or with other colorless cofactors [11] and only a lesser part of them is decomposed under the influence of temperature, light and oxygen or precipitates as colloid fraction. As a result, the absorbance at 520 nm increases in aged wines and, simultaneously, the absorbance at 420 nm decreases, due to the transformation of monomeric anthocyanins to their polymeric forms. The results obtained for IDYP treated samples show that the percentage of polymeric colour slightly increased (compared to blank) after the accelerated aging test and also after normal aging one year period (except for the sample treated with Optimum White at 0.4 g/L). It has been shown [1] that polysaccharides (abundant in IDYPs) can act as protective colloids, preventing precipitation, and therefore lead to an increase in colour stability.

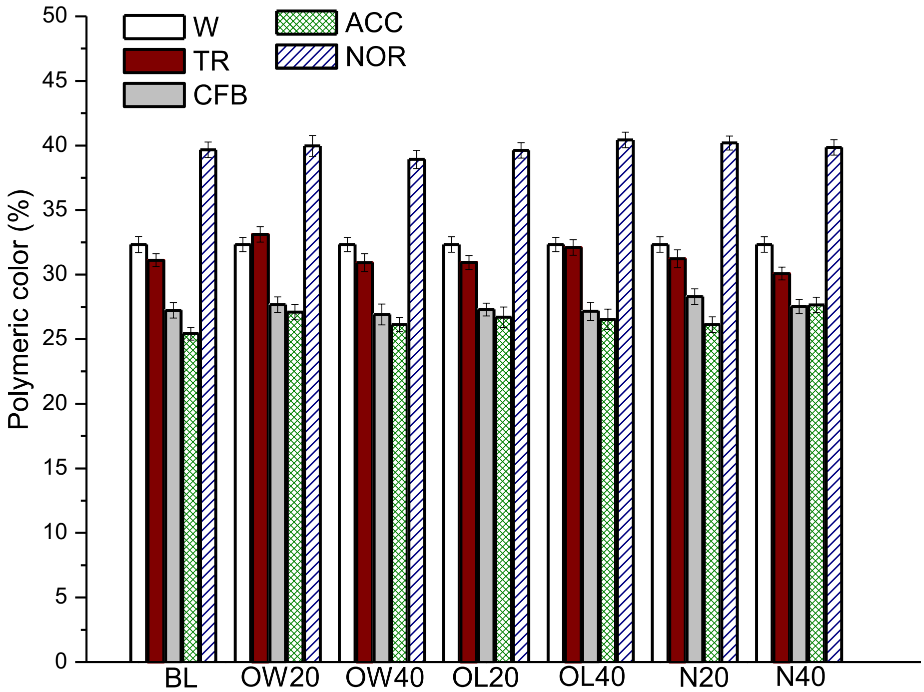

Figure 4. Percent of polymeric colour of the investigated wine samples.

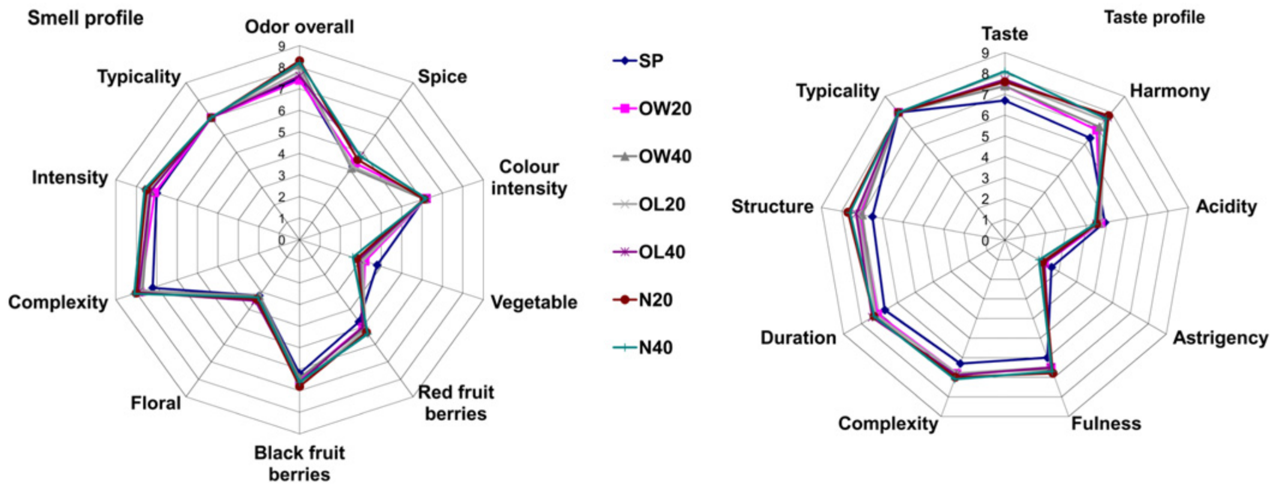

Figure 5. Sensory analysis of the investigated wine samples.

3.7. Colour tint and intensity

The results obtained from colour tint and intensity (not shown) revealed no significant differences between IDYP treated wines and blank

3.8. Sensory analysis

Sensory analysis findings clearly indicate that IDYP treatment had an overall positive influence on the sensory characteristics of the wine. After being bottled and kept in standard storage conditions, the treated wines were found to have improved smell, more complexity, more pronounced black and red fruit berry notes and also less of a vegetable character, which was clearly present in basic wine. Concerning taste, a significant improvement was noticed in terms of fullness, structure, complexity and harmony, accompanied by an astringency decrease.

3.9. Accelerated aging test

Although the evaluation of the accuracy of the accelerated aging test was not amongst primary goals of this investigation, the obtained results should not be left uncommented. Namely, the values of all the monitored parameters presented in Figs. 1–4, measured after the accelerated test and after one year of normal aging, exhibit significant discrepancies, both in terms of absolute and relative values. Hence, our findings indicate that the accelerated aging test results should be used with caution.

4. Conclusion

The results obtained in this investigation show that the use of IDYPs can lead to improved sensory characteristics of red wine after a one year aging period, accompanied by minor changes in wine polyphenol composition and content, decreased oxygen content and slightly enhanced percentage of polymeric colour. However, detailed investigations of treatment parameters, such as quantity of IDYP, temperature and duration of treatment, are yet to be performed in order to reveal the influence of yeast products on wine quality in details.

References

[1] M.Á. Pozo-Bayón, I. Andújar-Ortiz, M.V. Moreno-Arribas, Food Res. Int. **42,** 754 (2009)
[2] J.M. Nguela, N. Sieczkowski, A. Roi, A. Vernhet, J. Agr. Food. Chem. **63**, 660 (2015)
[3] M.A. Pozo-Bayon, I. Andujar-Ortiz, J.M. Alcaide-Hidalgo, P.J. Martin-Alvarez, M.V. Moreno Arribas, J. Agr. Food. Chem. **57**, 10784 (2009)
[4] J.A. Peìrez-Serradilla, M. D. Luque de Castro, Food Chem. **111**, 447 (2008)
[5] International Organization of Vine and Wine, *Compendium of International Methods of Wine and Must Analysis, Vol 1* (OIV, Paris, France, 2017)
[6] V. Ivanova, M. Stefova, F. Chinnici, J. Serb. Chem. Soc. **75**, 45 (2010)
[7] M. Monagas, C. Gomez-Cordoves, B. Bartolome, O. Laureano, J.M.R. Da Silva, J. Agric. Food Chem. **51**, 6475 (2003)
[8] J.C. Rivas-Gonzalo, Y. Gutierrez, E. Hebrero, C. Santos-Buelga, Am. J. Enol. Vitic. **43**, 210 (1992)
[9] T.J. Hager, L.R. Howard, R.L. Prior, J. Agric. Food Chem. **56**, 689 (2008)
[10] M.A. Cliff, M.C. King, J. Schlosser, Food Res. Int. **40**, 92 (2007)
[11] Y. Hayasaka, J.A. Kennedy, Aust. J. Grape Wine Res. **9**, 210 (2003)