



CHEMICAL, PHYSICAL AND SENSORY ANALYSIS OF ACTIVITY DIFFERENT YEAST SPECIES ON IDENTICAL SUBSTRATE IN WINE PRODUCTION

*Vladimír Vietoris**, Hana Balková, Tatiana Bojňanská, Lubomír Bennár, Peter Czako

Address: Ing. Vladimír Vietoris, PhD.

¹Slovak University of Agriculture, Faculty of Biotechnology and Food Sciences, Department Storing and Plant Products Processing, Trieda A. Hlinku, 949 76 Nitra, Slovak Republic, phone number: +421 37 6414793.

*Corresponding author: vladimir.vietoris@uniag.sk

ABSTRACT

Rizling vlašský is the second most important variety in Slovakia. The science of wine production includes a summary of knowledge and experience in the field of grape growing and wine making, or the production of different types of wines using specific methods of production. Wine quality is the result of the interaction between yeast, bacteria and microscopic fungi. In this research, we studied the effects of active dry wine yeasts on chemical, physical and sensory parameters in wine production. We have applied five kinds of yeasts (FERMIVIN, FERMIVIN PDV, FERMICRU AR2, FERMIFLOR and FERMICRU VB1). It can be concluded that the application of active dry wine yeasts is beneficial for the production of rizling vlašský. The best showing were yeasts FERMIFLOR and FERMIVIN PDM. In the last sample where they were left the original yeasts the varietal aroma was preserved. It can be noted that the wine was right technologically produced and all wines were harmonious with a pleasant fresh taste.

Keywords: rizling vlašský, active dry wine yeast (ADWY), technology, sensory analysis

INTRODUCTION

In Slovakia, rizling vlašský is the second most important variety in the representation 16.77 % of the vineyard areas, it was registered in 1941 (**Pospíšilová et al., 2007**). Rizling vlašský gives the typical varietal wines with a delicate bouquet and aroma reminiscent bitter almonds or green apples in warm localities (**Pavloušek, 2007**). This variety is mostly used for the production of quality and cabinet wines (**Stevenson, 1998**).

Wine production is one of the oldest world's biotechnological processes. Processing of grapes with fermentation is technically complex process. In order to produce a quality wine with a good taste and aroma, with the optimum ratio of basic components such as sugars, acids, alcohol, wine making throughout the production from the beginning of fermentation to the maturing must be monitored (**Kováč, 2010**).

Many microorganisms that are described as endogenous (original) and exogenous (active dry wine yeasts) participate in wine production (**Robinson, 2000**). Today, the majority of wine production is based on active dry yeast usage, which provides fast, reliable fermentation and it reduces the risk of slowing down or jam fermentation and microbial contamination (**Cordero-Buesco et al., 2011**). Alcoholic fermentation is a dynamic and complex process in which grape cider is subjected to continual transformation and significant change in the environment occurs, which affects the biological activity and viability of microorganisms (**Patrocínio et al., 2008**).

The main feature of ADWY (active dry wine yeasts) is fast invigoration of fermentation under unfavourable conditions such as low temperature, partial decay and so on. ADWY suppresses the original yeast culture, which in part may show weaker fermentation ability and also it produces a number of unwanted products, especially great amount of volatile substances such as ethyl ester (ethyl acetate) and acetic acid (**Minárik, 2004**). The starter of cultures of ADWY belongs to the type of *Saccharomyces cerevisiae* and *Saccharomyces uvarum* (**Farkaš, 2002**).

The main objective of our study was the application of active dry wine yeasts (ADWY) and subsequent observation of chemical, physical and sensory characteristics in wine production.

MATERIAL AND METHODS

The sampling was carried out in Nitra wine region – Šintava in 2011. For experiment we used rizling vlašský variety. In our work, were used the following types of yeasts from firm O.K. SERVIS BioPro, s.r.o.: FERMIVIN, FERMIVIN PDM, FERMICRU AR2, FERMIFLOR and FERMICRU VB1.

The bunches were harvested manually and stored in containers, it was followed by processing in the form of grinding. The cider was separated from the solid parts by pressing, to avoid the multiplication of undesirable microorganisms, we added sulfur to the mash. Thus we create the conditions for multiplication of yeast varieties necessary for forming of a variety character of the future wine. Then we measured the sugar content, which was 22.7° ČNM (kg of sugar per 1 hl of cider). Consequently we prepared a yeast starter (50 ml H₂O + 100 ml of cider + 1 g of nutrient salt) so that we found the activity of prepared yeasts. After an hour of yeasts action, the samples showed the following features: sample no. 1, sample no. 2 and sample no. 3 - high foam, sample no. 4 - very low or no foam, sample no. 5 - no foam, but fermentation with the making a noise.

The cider was divided into 6 carboys with a capacity of 5 liters. To the first carboy we applied 2.28 g of yeast FERMIVIN, to the second carboy 2.04 g of yeast FERMIVIN PDM, to the third carboy 2.01 g of yeast FERMICRU AR2, to the fourth carboy 2.09 g of yeast FERMIFLOR and to the fifth carboy 2.05 g of yeast FERMICRU VB1. To the last carboy we didn't add any yeasts, we left there only original yeasts. After fermentation we added 0.3 g Na₂S₂O₅ to the future wine. This was followed by decanting of the wine and adding of the bentonite.

Determination of turbidity

The turbidity of fluid is measured by fully automated machine - nephelometer. The turbidity of liquid is determined by measuring of the light intensity disperse in the sample under the angle 90° and 13°. The intensity of the disperse radiation naturally depends on the intensity of the incoming exciting light beam. The angle of measurement 90° enables to define the turbidity and the amount of proteins. At an angle of 13° the amount of particles larger than 1 micron (yeasts and diatomaceous earth) is established. For comparison of both measured angles, it can be determined the proportion of proteins and yeasts on the turbidity of measured sample (Veverka, 2004).

Determination of chemical parameters

For measurement of chemical parameters in wine was used machine ALPHA FT-IR analyzer that can be utilized for qualitative and quantitative characterization of materials. Molecular structures and components are identified by their characteristic infrared absorption bands.

Pycnometric determination of alcohol in the wine

Principle of the method consists in distillation of a certain capacity of wine in which the same capacity of the distillate is obtained. The density of the distillate is determined by pycnometer. The volume concentration of alcohol is calculated from the density of the distillate according to the table, which presents the relationship between density and composition of ethanol and water solutions (**Hronský, 2006**).

Titration determination of sugars by Schoorl

The principle of the determination is based on the ability of directly reducing sugars (glucose and fructose) reduce the bivalent copper of Fehling reagent on the monovalent. The method is based on the determination of unreacted bivalent copper. Potassium iodide is oxidized by copper sulfate on molecular iodine, the amount of which is determined by titration with sodium thiosulfate (**Benkovič, 2005**).

Determination of total acidity by titration of Bromothymol method

Principle of the method is that from the testing wine the sulfur dioxide is eliminated, for example by heating at the boiling point. The measuring amount of thus modified wine is titrated by solution of alkaline potassium hydroxide to pH = 7.0 (**Benkovič, 2005**).

Titration determination of volatile acids

The principle of the method consists in elimination of carbon dioxide from the testing sample and volatile acids are separated from the measured amount of wine distilled with water vapour. With the volatile acids SO₂ passes to the distillate, therefore the correction on

SO₂ is making in the calculation. The distillate is titrated with a solution of alkali hydroxide (KOH) to the phenolphthalein. In the process, the volatile acids and distilled sulfur dioxide are neutralized. For the calculation of the content of volatile acids, the consumption during the titration with sodium hydroxide is corrected for distilled sulfur dioxide, which was determined by iodometric titration (**Benkovič, 2005**).

Determination of organoleptic parameters of wine

The sensory evaluation was attended by 5 certified assessors from SUA and the State veterinary administration Bratislava. Each evaluator had 6 samples. For the evaluation of wine was used a consensual profile. Parameters selected for profile analysis were evaluated. There were also evaluated the general impression, bitterness, sweetness, acidity and fullness. Intensity scale was determined as follows: 0 - none, 1 - threshold, 2 - weak, 3 - medium, 4 - strong, 5 - very strong. It was possible to use points in the value 0.5.

RESULTS AND DISCUSSION

Results of physical analysis (turbidity)

The protein turbidities are one of the most common faults of wines. Adding bentonite to white wines makes the wine delicate and removes these protein turbidities (**Kraus et al., 2008**). The optimum amount of bentonite is determined in wine laboratories. **Pocock and Waters (2006)** detected that the optimal conditions for the application of bentonite are temperatures from 14 °C to 19 °C. **DeBruijn et al. (2009)** found that bentonite positively influences protein turbidities and the quality of the wine.

Sample no. 1 FERMIVIN showed a high level of proteins. After the application of bentonite, the proteins measured at angle 90° have significantly decreased during 48 hours. Thereafter there didn't occur a significant changes in the proteins. Under the angle 13° we observed particles larger than 1 µm. Using the bentonite they reduced significantly on the value 4.92 NTU. Sample no. 2 FERMINIV PDM showed a smaller proportion of proteins than it was in sample no. 1 FERMIVIN. During 336 hours the value decreased from 150.4 to 1.2 NTU. For particles larger than 1 µm, the value has changed significantly from 976 to 3.04 NTU. Wine using bentonite was beautiful cleared. Sample no. 3 FERMICRU AR2 showed the smallest values for proteins and also for particles larger than 1 µm. The main factor that it

influenced was the sulfur. To the sample no. 3 was added sulfur in an excessive amount which resulted a significant reduction of proteins. Sample no. 4 FERMIFLOR showed the second highest amount of proteins after the exposure of bentonite. It was equally in the case for particles larger than 1 µm. It didn't have any negative consequences to the general impression of the wine. Sample no. 5 FERMICRU VB1 showed one of the smallest protein turbidities. Bentonite reduced the amount of proteins significantly. Under the angle 90°, the last measurements have not changed significantly. Under the angle 13° the resulting value of particles larger than 1 µm was the second smallest of all samples. Sample no. 6 (original yeasts) had the highest concentration of proteins. This was due to the late onset of the fermentation process compared to other samples. Sample no. 6 was able to reduce the proteins using bentonite from 1388.0 NTU to 3.52 NTU. At angle 13° we could see minimal changes after 48 hours. This wasn't observed in any other sample. Results of nephelometric determination of the proportion of proteins and yeasts for turbidity of samples FERMIVIN, FERMINIV PDM, FERMICRU AR2, FERMIFLOR, FERMICRU VB1 and the original yeasts are shown in Table 1.

Table 1 Nephelometric determination of the proportion of proteins and yeasts for turbidity of samples FERMIVIN, FERMINIV PDM, FERMICRU AR2, FERMIFLOR, FERMICRU VB1 and the original yeasts

Number of hours	FERMIVIN		FERMINIV PDM		FERMICRU AR2		FERMIFLOR		FERMICRU VB1		THE ORIGINAL YEASTS	
	T 90° (NTU)	T 13° (NTU)	T 90° (NTU)	T 13° (NTU)	T 90° (NTU)	T 13° (NTU)	T 90° (NTU)	T 13° (NTU)	T 90° (NTU)	T 13° (NTU)	T 90° (NTU)	T 13° (NTU)
	432	355.6	150.4	976	40.8	172	300	770	128.4	556	1388	108.8
48	20.28	92.8	10.88	31.68	6.32	25.24	25.04	83.2	6.92	22.72	42.4	108.4
72	15.32	71	6	25.92	4.68	18.12	21.72	71.6	5.6	18.84	35.28	83.6
96	10.2	44.8	4.2	17.16	2.56	12.8	16.12	48.4	3.8	12.16	20.8	50.4
120	7.32	31.2	2.96	12	2.56	9.28	10.76	32.64	2.88	8.68	16.4	33.72
240	2.6	10.48	1.8	5.16	1.6	4.16	3.84	11.12	2.2	4.4	5.24	11
264	2.44	8.96	1.48	4.28	1.56	3.48	3.64	9.76	2.8	3.72	5.04	10.4
336	1.84	4.92	1.2	3.04	1.08	2.24	2.4	5.2	1.68	2.52	3.52	5.92

Legend: T – turbidity, NTU - nephelometric turbidity unit

Results of sensory analysis

Several authors have devoted to sensory evaluation of rizling. **Fischer et al. (1999)** reported an significant differences between commercially available wines of rizling. The difference in a climatic conditions were identified as a major factor. **Douglas et al. (2001)** found that rizling shows citrous, peach and almond taste. Yellow melon was identified as less expressive taste.

Sample no. 1 showed a strong taste of oxidized apple and it passed to the weak aroma of almond, caramel and peach in the background. Sample no. 2 was showed with a considerable taste of bread and citrus. In the background of prevailing tastes yellow melon and almond tastes were reflected. In the sample no. 3 strongly dominated tastes of sauerkraut and sulfur. Evaluators agreed that behind these major tastes is threshold taste of yellow melon. Sample no. 4 was evaluated as the best, which showed a pleasant taste of peach with mild aromas of almonds, tropical fruits and yellow melon in the background. Sample no. 5 showed the typical taste of rizling vlašký. The tastes of walnut and green apple were dominant. In the background of these tastes, the evaluators captured citrus and peach. Sample no. 6 where the original yeasts were left, fresh tastes of tropical fruits and yellow melon were expressed. Tastes of lime blossom and walnut supplied softness to this sample. Results are shown in the figure 1.

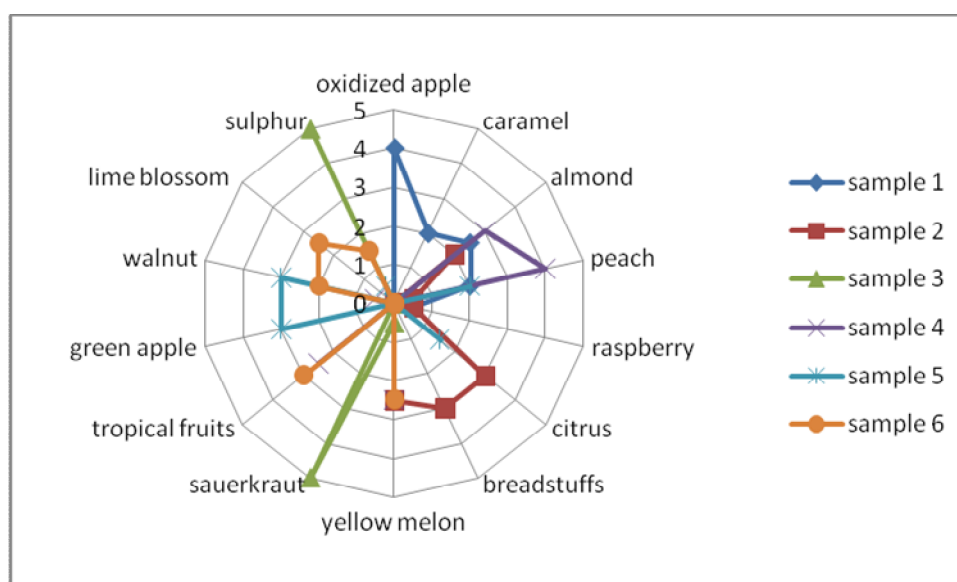


Figure 1 Determination of flavour properties of samples FERMIVIN, FERMINIV PDM, FERMICRU AR2, FERMIFLOR, FERMICRU VB1 and the original yeasts

Further bitterness, acidity, fullness and general impression were evaluated. All samples showed minimal bitterness. Sample no. 4 showed the highest and sample no. 6 the smallest value. In all samples the content of acids was evaluated as slightly increased, which supplied the pleasant freshness to all samples. The fullness of the wine refers to the taste sensation of evaluators which they feel on the surface of the tongue and during the swallowing. The wine is fuller, the massive on the language seems to us. At this scale, the samples no. 4 and no. 6 were evaluated as the best. Results are shown in the figure 2.

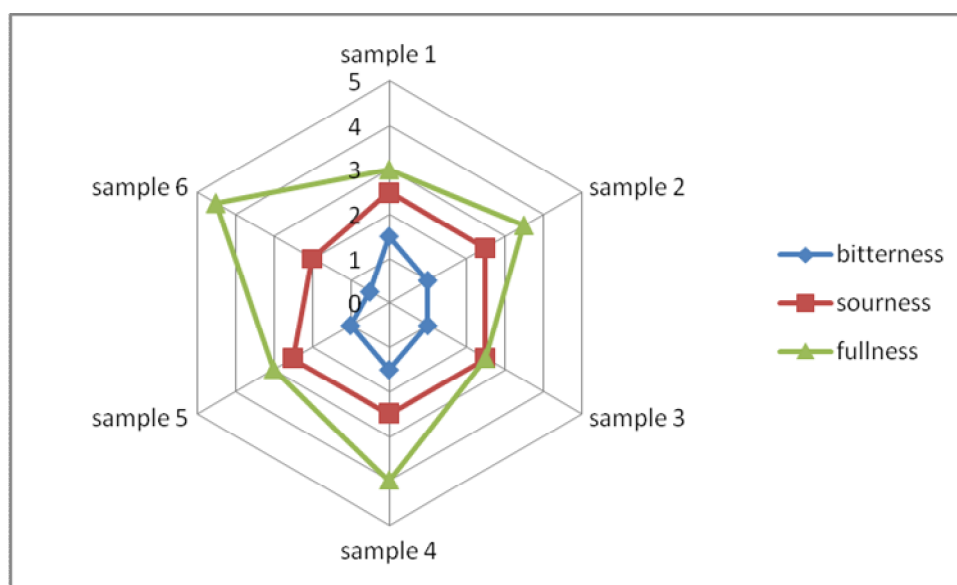


Figure 2 Determination of bitterness, sourness and fullness of samples FERMIVIN, FERMINIV PDM, FERMICRU AR2, FERMIFLOR, FERMICRU VB1 and the original yeasts

The general impression was evaluated by 100 points system. According to this system, gold medal is given to wines that reached at least 90 points, silver medal to wines that rated by 85 points and a bronze medal to wines that scored 80 points. From our samples were placed three samples. Sample no. 4 ended with a silver medal and sample no. 6 and no. 2 ended with a bronze medal.

Results of chemical analysis

Chemical analysis of wine are very important for objective characteristics and it gives information about composition of wine and representation of individual components. **Ferreira et al. (2002)** found that different types of wines are chemically different to each

other. It can be caused by the climate conditions and subsequent technological processes of the wine production.

Using chemical analysis, we found that the alcohol included in all samples was almost the same and the differences were minimal. This content was between 12.0 % and 13.7 % vol. In case of total sugars greater differences from 3.2 to 21.1 g / l were already occurred. It follows that the sample no. 5 is dry (3.2 g / l), the sample no. 2 and no. 4 are the semi-dry (8.5 g / l, 9.0 g / l). Other samples no. 1, no. 3, no. 6 are sweet (13.9 g / l 18.4 g / l 21.1 g / l). Total acids were in the range from 5.6 g / l up to 5.9 g / l. Volatile acids didn't exceed the values of 0.38 g / l which means that there did not appear the production of the acetic acid. Other parameters showed almost identical values.

CONCLUSION

Following the results obtained from the physical analysis, it can be concluded that the application of bentonite in the process of wine production is very effective and it reduces protein turbidities and particles larger than 1 µm. Through the chemical analysis, we found that all wines were harmonious with a pleasant fresh taste. It can be noted that the wine was right technologically produced, because there wasn't determined the occurrence of acetic acid. It based on all the parameters evaluation in sensory analysis sample no. 4 was published as the best, which would receive a silver medal. Bronze medal would receive samples no. 6 and no. 2. According to all these findings, we concluded that the application of active dry wine yeasts is beneficial for the production of rízing vlašský. The best showing were yeasts FERMIFLOR and FERMIVIN PDM.

Acknowledgments: This work was carried out with the financial support of the Slovak Ministry of Education under the program: KEGA 015SPU-4/2011.

REFERENCES

- BENKOVIČ, M. 2005. Evaluation of quality characteristics of vineyard and wine production in the conditions of Nitra wine region: thesis. Nitra: SPU, 2005. p. 12 - 13.
- CORDERO-BUESO, G. - ARROYO, T. - SERRANO, A. - VALERO, E. 2011. Remanence and survival of commercial yeast in different ecological niches of the vineyard. In *FEMS Microbiol Ecology*, vol. 77, 2011, no. 2, p. 429-437.

- DE BRUIJN, J. - LOYOLA, C. - FLORES, A. - HEVIA, F. - MELÍN, P. - SERRA, I. 2009. Protein stabilisation of Chardonnay wine using trisacryl and bentonite: a comparative study. In *International Journal of Food Science & Technology*, vol. 44, 2009, p. 330-336.
- DOUGLAS, D.- CLIFF, M. A.- REYNOLDS, A. G. 2001. Canadian terroir: characterization of Riesling wine from the Niagara Peninsula. In *Food Research International*, vol. 34, 2001, p. 559-563.
- FARKAŠ, J. 2002. All about wine. Martin: Neografia, 2002, 171 p. ISBN 80-88892-47-3.
- FERREIRA, V. - ORTÍN, N. - ESCUDERO, A. - LÓPEZ, R. - CACHO, J. 2002. Chemical characterization of aroma of Grenache Rose Wines: Aroma Extract dilution analysis, Quantitative determination, and Sensory reconstitution studies. In *Journal of Agricultural and Food chemistry*, vol. 50, 2002, no. 14, p. 4048-4054.
- FISCHER, U. - ROTH, D. - CHRISTMANN, M. 1999. The impact of geographic origin, vintage and estate on sensory properties of *Vitis vinifera* cv. Riesling wines. In *Food Quality and Preference*, vol. 10, 1999, p. 281-288.
- HRONSKÝ, Š. 2006. The Viniculture. Nitra : SPU, 2006, 104 p. ISBN 80-8069-774-4.
- KOVÁČ, K. 2010. Good wine in the extreme vintage. In *Vine and wine*, vol. 5, 2010, p. 3.
- KRAUS, V. – FOFFOVÁ, Z. – VURM, B. – KRAUSOVÁ, D. 2008. The New Encyclopedia of Czech and Moravian wine. Praha: Praga mystica, 2008, 306 p. ISBN 978-80-86767-09-3.
- MINÁRIK, E. 2004. Effect of yeast and bacterial flora to wine production. In *Vine and wine*, vol. 4, 2004, no. 6. ISSN 1335-7514.
- PATROCINIO, G. - PILAR, S. - LÓPEZ, R. 2008. The occurrence of fungi, yeasts and bacteria in the air of a Spanish winery during vintage. In *International Journal of Food Microbiology*, vol. 125, 2008, p. 141-145.
- PAVLOUŠEK, P. 2007. Encyclopedia of wine grape. Brno: Computer press, 2007, 320 p. ISBN 978-80-251-1704-0.
- POCOCK, K.F. – WATERS, E. J. 2006. Protein haze in bottled white wines: How well do stability test and bentonite fining trials predict haze formation during storage and transport? In *Australian journal of Grape and Wine research*, vol. 12, 2006, no. 3, p. 212-220.
- POSPÍŠILOVÁ, D. – SEKERA, D. – RUMAN, T. 2007. Ampelography of Slovakia. Modra: Research and breeding station winemaking and viticultural, 2007, 368 p. ISBN 80-969350-9-7.
- ROBINSON, RICHARD, K. 2000. Encyclopedia of Food Microbiology. SPCK Publishing, 2000, 2372 p. ISBN 978-0-12-227070-3.

STEVENSON, T. 1998. Wines: 101 practical advices. Bratislava : Ikar, 1998, 72 p. ISBN 80-7118-653-8.

VEVERKA, J. 2004. Determination of turbidity in wines nephelometry. In *Wine horizon*, vol. 11, 2004, p. 520.