The Role of Redox Potential Measurement in Oak Barrel Wine Maturation

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Redox potential measurement was efficiently used for monitoring the two-year maturation process of cultivar *Blau Fränkisch* wine in 225-L oak barrels. Redox potential of maturing wine was measured at six levels from the top to the bottom of the barrel. During the process of wine maturation, the formation and heterogeneity of redox layers, as well as the formation of various oxidoreductive zones were indicated. The end of barrel maturation process was represented with homogeneity of all redox zones where no differences in all measured levels were indicated.

Keywords:
wine fermentation monitoring, redox potential, oak barrel wine maturation

Introduction

In chemistry, redox potential represents a sum of the potentials of all oxidoreduction processes, in particular chemical processes. Its indication is influenced by reversible oxidoreduction couples, irreversible reductants, the activity of free oxygen and hydrogen[1]. Its measurement is dependent on pH value, temperature, equilibrium constant, oxidoreduction potentials, and dissolved oxygen concentration[2].

In living organisms, oxidation-reduction systems play such a profound and essential role that life itself might be defined as a continuous oxidation-reduction reaction. In this measurement, the main indication represents the ratio between the sum of all oxidant activities and the sum of all reductant activities of the metabolism in microbial cells. Although wine is a complex redox system, the activity of cell metabolism is a few magnitudes higher than all the other redox complexes and factors. Therefore, in comprehensive bioprocess technology, it was proved that redox measurements and their interpretation represent one of the most significant indications of the fermentation process[3,4]. Thus, it is not surprising that theoretical speculations and experimental studies on oxidation and reduction processes in bioprocesses, animals, and plants have been actively pursued since the isolation of oxygen over 150 years ago[5].

In several aerobic processes with various microorganisms, such as *Pseudomonas*[6], *Actinomyces levoris*[7], and *Corynebacterium glutamicum*[8], redox potential was used as the main bioprocess monitoring parameter. For high-yielding production in *Aspergillus niger* citric acid fermentation, it was found that particular redox potential levels and their fermentation profile are of essential importance[9,10]. Monitoring of redox potential was referred also in fermentations with *Proteus vulgaris, Clostridium paraputrificium* and *Candida utilis*[11,12], *Lactobacillus sanfrancisco*[13], *Lactoccocus lactis*[14], etc.

Redox potential monitoring was also used in several anaerobic processes where the significance of this measurement has been reported[15–17]. In xylitol production by a recombinant *Saccharomyces cerevisiae* containing xyl-1 gene of *Pichia stipatis*, the xylitol yield on substrate increased with increased xylose activity supported by high value of redox potential[18].

Redox potential was successfully used as a main process parameter for *scale-up* in citric acid fermentation from 10-liter laboratory scale to 100- and 1000-liter pilot plant, even in geometrically dissimilar stirred tank reactors[9,10].

Redox potential calculation is defined by Nernst equation (1):

$$E_h = E° + \frac{RT}{nF} \ln \frac{\sum a_{ox}}{\sum a_{red}} \quad (1)$$

This equation expresses the information on redox potential of oxidoreduction reaction. $E°$ is the

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potential of standard hydrogen electrode, a potential where all the activities of the products and the reactants would be equal to 1. Following the Nernst equation, the value E_h of the redox potential (mV), depends on the ratio of the sum of activities of all oxidized and the sum of activities of all reduced parts in the system, on the number of electrons *n,* and on the constant of the system E^o. Its measurement also depends on pH value, temperature, equilibrium constant, oxidoreduction potentials, and dissolved oxygen concentration[2].

Oxidation and reduction reactions in homogeneous systems always take place simultaneously, where free electrons do not exist. In each reaction, the same number of electrons is released, as they are consumed[19].

For more complex redox systems in the presence of a large number of oxidants and reductants that could interact according to the following scheme:

$$\nu_1 S_1 + \cdots + \nu_i S_i + ze^- \rightleftharpoons \nu_j S_j + \cdots + \nu_k S_k \quad (2)$$

Redox potential of such a system could be defined as:

$$E_h = E° + \frac{RT}{zF} \ln \left(\frac{\prod_i a_{ox}^{\nu_{ox}}}{\prod_i a_{red}^{\nu_{red}}} \right) \quad (3)$$

$$\prod_i a_i^{\nu_i} \equiv a_{S_1}^{\nu_1} \cdot a_{S_2}^{\nu_2} \cdots a_{S_i}^{\nu_i} \quad (4)$$

where $\prod_i a_i^{\nu_i}$ is the product of all activities of the oxidants and reductants present in the system.

According to Eq. (3), media having larger product of oxidant activity than of reductant activity, express higher redox potential course (case: citric acid fermentation), and negative redox potential course (case: anaerobic alcohol fermentation).

Redox potential of the measured substance, or substrate, depending on pH, is expressed in Eq. (5)[20].

$$E_h = E_{O_2/H_2O} + RT/4F \ln a_{O_2} - \\ - RT/4F \ln 2.303 \text{ pH} \quad (5)$$

In comprehensive wine technology, the incorrect rH factor is often in use, besides the measurements of redox potential in mV. It was introduced by Clark and Cohen in 1923 in order to eliminate pH_2 dependence of the redox potential by calculation[21].

$$rH = 0.0343\ E_h + 2 \text{ pH}; \quad pH_2 = -\log a_H \quad (6)$$

The rH value is the negative logarithm of the partial pressure of gaseous hydrogen (pH_2): an rH of 0 corresponds to a pH_2 of 1 atm; and rH = 10 corresponds to a pH_2 of 10^{-10} atm, etc. Later it was found that the assumptions made were incorrect in every case. Firstly, the value taken as a basis by Clark, varied if the reduced phase dissociated to yield hydrogen ions on alteration of pH. Although Clark himself recalled this definition as incorrect and useless, in wine technology, this incorrect term is unfortunately still in practice.

Wine, as a product of grape fermentation, represents a complex redox system due to its chemical composition. Many chemical, enzymatic, and biological processes in wine are correlated with the oxidative state of wine. As the value of redox potential is in close relation with a number of reactions taking place in wine, being either spontaneous or induced, it enables one to control the winemaking process[5]. In wine technology, redox potential measurement assesses the ability of the microorganism to grow, as well as its physiological activity in a defined environment. It reflects the rate and intensity of all oxidation and reduction processes that took place in the wine. The addition of sulfur dioxide into the grape juice reduces yeast metabolic activity, so in such an environment, redox potential is lower and the fermentation processes proceed slower[22].

Wine maturation in barrels on yeast sediment (*sur-lies*) is a process used mostly for the maturation of premium white wines. During wine maturation, it is significantly effective in total removal of methanethiol and ethanethiol, and stabilization of proteins and tartrates in wine[23]. Sedimented yeast's strong reductive potential also decreases the influence of oxygen activity very effectively; therefore, it also lowers the need for sulfurization[24,25].

Materials and methods

Microorganism

Selected dry yeast (*Saccharomyces cerevisiae*, Uvaferm SLO) was used in all the experiments. Reactivation of the yeast in a water-diluted must (1 : 1) took 15 minutes at 30 °C. The mix of grape must and mashed grape berries inoculated 45 min after revitalization. The amount of inoculum was equivalent to 0.3 g of dry yeast per liter of must.

Experimental design

Wine fermentation of grape must and mashed grape berries of cultivar *Blau Fränkisch* (Posavje Region, microlocation Bela Krajina, Slovenia) was studied. Fermentation proceeded in 3000-L stainless steel tanks at 28 °C. The head of floated grape berries was immersed three times daily into the fermentation broth. After 24 hours of fermentation,

young wine was separated from the skins, and poured into three 225-L new Slovenian oak wood barrels (Kranjc d.o.o., Slovenia). Maturation temperature of 15 °C was used in all three series of experiments.

In the first part of the maturation experiment, sulfurized wine with 50 mg L^{-1} of 6 % sulfuric(IV) acid was studied. In the second part, unsulfurized wine was studied. For each part of the experiment, three barrels were used. The results of each run were summarized, statistically processed, and presented as the most typical courses.

Redox measurements

For redox *on-line* measurements, the Ingold redox electrode (Pt-4865) (Switzerland), and control unit (Mettler Toledo MFG 509, Switzerland) were applied.

Biomass

Twenty mL of fermentation broth of each sample was centrifuged for 5 min at 4000 rpm, and biomass was determined gravimetrically after 24 h of drying at 105 °C.

Analytical methods

Alcohols as well as reducing sugars were determined off-line by gas chromatography according to Dittrich *et al.*[26] AGC 3700 Varian Model, Austria, was used, with *n*-butanol as internal standard. Ethanol and total alcohols (1-propanol, 2-butanol, isoamyl alcohol), as well as the organic acids were analyzed by HPLC in accordance with validated methods proposed by Bio-Rad[27]. Samples were filtered through 0.45-μm membranes, and analyzed using a 300 mm × 7.8 mm Aminex HPX-87H organic acid analysis cation exchange column (Bio-Rad Laboratories). Elution was performed at 65 °C. The mobile phase was 0.005 M H_2SO_4 in milli Q water. The pump was operated at a flow rate of 0.5 mL min^{-1} ($0.008 \cdot 10^{-3}$ L s^{-1}). The injection volume was 20 μL. The eluting compounds were monitored by a fixed wavelength ultraviolet (UV-VIS) detector at 210 nm. This detector was connected in series with a refractive index (RI) detector. The peaks were quantified using external standard calibration. The components were identified by comparing their retention times with those of the standards. Quantification was performed using external standards prepared from pure compounds.

Sensorial analysis

Sensorial analysis proceeded in accordance with the Rules on the Procedure and Method of Assessing Must and Wine (Official Gazette of the Republic of Slovenia). The 20 point Buxbaum method was applied.[280] The results of seven sensorial-analysis wine experts were statistically processed and taken in the final result. The control sample of wine, a standard B, was three-year-old bottled wine from the same cultivar and producer (KZ Metlika, Slovenia).

Statistics

All the cultivation experiments were performed at least in three runs. The results were analyzed for statistical significance by one-way analysis of variance (ANOVA) test using the Statistical Package of the Social Science (SPSS) version 11.0 (SPSS Inc., Chicago, IL, USA). Statistical significance of the test effects was evaluated at $p < 0.05$. Data were expressed as mean ± SD.

Typical measurements of on-line redox potential in fermentation, as well as the biomass accumulation are presented in Figs. 1 and 2.

Results and discussion

In the unsulfurized run, the fermentation started at 200 mV. The initial aerobic phase proceeded for 4 hours, followed by the yeast adoption phase, which lasted 15 hours. In sulfurized substrate, fermentation started at 130 mV and yeast adoption phase proceeded from 166 to 130 mV for 90 hours. Alcohol fermentation of the unsulfurized grape must started after 17 hours at 172 mV and ended after 216 hours at –118 mV. Alcohol fermentation of the sulfurized grape must started after 90 hours at 108 mV and ended after 244 hours at a lower redox potential of –150 mV (Fig. 1).

Sulfurization of grape must also affected the growth of biomass that had to adapt to the more reductive environment. Therefore, the lag phase in this case was prolonged, and the exponential growth phase started after 96 hours. Maximal amount of biomass at 144 hours was 6.08 g L^{-1}, while at the end of fermentation it was 4.88 g L^{-1}. In contrast, with the unsulfurized must, the growth of biomass started much faster, from the very beginning of the fermentation phase. In this process, exponential growth phase started 12 hours after the lag phase. Maximum of 7.45 g L^{-1} biomass was detected after 144 hours, and it was 5.38 g L^{-1} at the end of fermentation (240 hours) (Figs. 1 and 2).

At the beginning of the next step in the process, i.e., barrel maturation, the yeast biomass in the unsulfurized wine was higher by 1.50 g L^{-1} (i.e., by 30.73 %) than in the sulfurized wine.

During the maturation of unsulfurized and sulfurized wines in the barrique oak barrels, the meta-

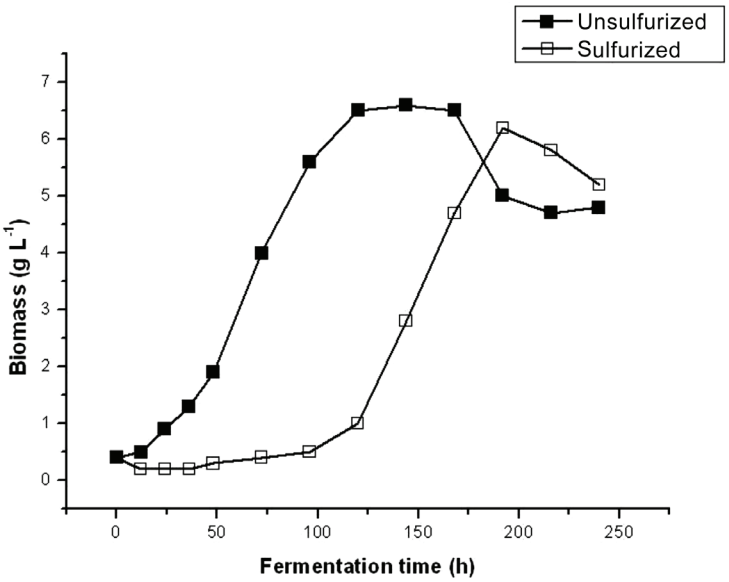

Fig. 1 – *Growth of biomass of unsulfurized and sulfurized fermentation*

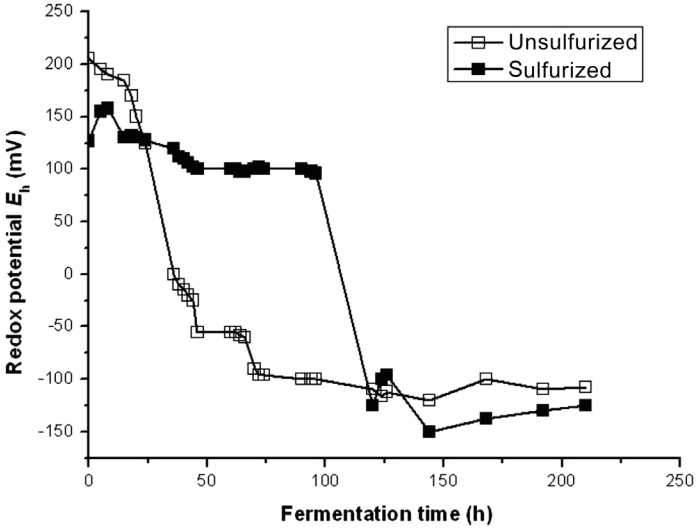

Fig. 2 – *On-line redox potential measurements of unsulfurized and sulfurized fermentation*

bolic activity of yeast biomass was measured by immersing redox potential electrode throughout the depth profile of the barrels positioned horizontally as usual. Over the first and second years, the measurements detected the formation of various redox layers in the barrels. As indicated in the barrel depth line, the highest redox potential levels were measured at the hole at the top of the barrel. The potential decreased with the depth of the barrel, while at both barrel sides, the potential was similar to that close to the entrance hole on the top of the barrel. This data variation indicated the formation of various oxidation/reduction zones as a clear consequence of the maturation process (Fig. 3).

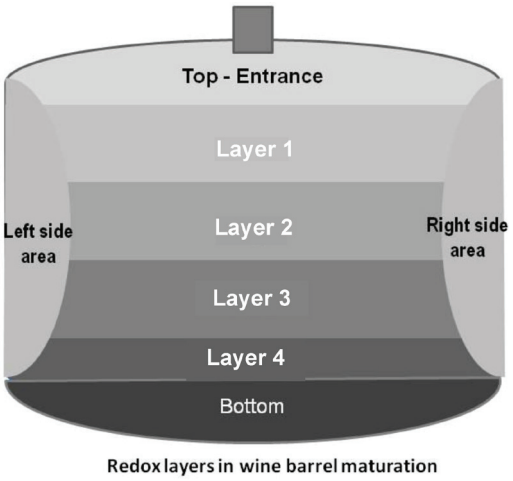

Fig. 3 – *Redox layers in wine barrel maturation*

Off-line measurements during the maturation process actually revealed the residual yeast metabolic activity in both processes. Since in the sulfurized wine the maturation took place in a more reductive environment, the maturation time was shorter. After three years, the total amount of SO_2 at the end of maturation in the sulfurized wine was 46.87 mg L^{-1} and the residual biomass level was 0.87 g L^{-1}. In relation to our measurements, in the first year of maturation, the biomass was found still in the active state. Redox potential measurements in the barrels revealed that, during the maturation process, in spite of the microoxidation areas on both sides of the barrel, the presence of added sulfur induced faster final deactivation of yeast biomass than in was the case with the unsulfurized wine (Table 1 a, b).

Much higher redox potential values were obtained in the maturation process of unsulfurized wine, indicating much more expressed metabolic activity of residual wine yeast in the barrel.

In the case of unsulfurized wine, the total sulfur amount after three years of maturation was 33.57 mg L^{-1} and the amount of free SO_2 was 19.13 mg L^{-1}, i.e., the value was higher by 1.52 mg L^{-1} or 8.6 % in comparison to the 17.61 mg L^{-1} of the final free SO_2 amount in the sulfurized wine (Table 2). In the unsulfurized wine, the presence of various redox layers, still after 30 months of maturation, indicated wine yeast metabolic activity, but after 36 months, all the layers in the barrel exhibited identical redox potential throughout the barrel profile, suggesting the end of yeast activity and thus readiness for the bottling process.

In the unsulfurized wine maturation process, the concentration of the wine yeast biomass was higher by 8.6 %, and its metabolic activity lasted much longer due to the less reductive maturation

Table 1 – *Redox potential layers in the barrels: a) unsulfurized, and b) sulfurized wine*

(a) Unsulfurized must layers	First year E_h (mV)	Second year E_h (mV)	Third year E_h (mV)
Top of barrel, entrance	480	356	192
10 cm below	432	311	186
20 cm below	385	252	185
30 cm below	320	205	185
40 cm below	220	200	184
Bottom of barrel	165	175	184
Left and right side	476	342	190

(b) Sulfurized must layers	First year E_h (mV)	Second year E_h (mV)	Third year E_h (mV)
Top of barrel, entrance	262	202	172
10 cm below	233	182	169
20 cm below	222	188	168
30 cm below	210	183	168
40 cm below	202	176	167
Bottom of barrel	186	172	167
Left and right side	255	198	170

Table 2 – *Results of chemical analysis of sulfurized and unsulfurized wines after three years at the end of maturation*

Parameter	Sulfurized wine	Unsulfurized wine
Biomass (g L^{-1})	1.04	3.46
Total alcohol (vol%)	11.7	12.1
Total extract (g L^{-1})	23.4	26.5
Sugar free extract (g L^{-1})	25.5	22.2
Total reduced sugars (g L^{-1})	2.20	1.55
Total acids (g L^{-1})	5.40	5.80
Volatile acids (g L^{-1})	0.66	0.98
Free SO_2 (mg L^{-1})	17.61	19.13
Total SO_2 (mg L^{-1})	46.87	33.57
Ash (g L^{-1})	1.73	3.23
pH (/)	3.43	3.93
Relative density (/)	0.993	0.994
Tartaric acid (g L^{-1})	3.00	3.00
Malic acid (g L^{-1})	3.60	0.00
Lactic acid (g L^{-1})	0.60	1.30

environment. Spontaneous yeast cells autolysis was pronounced and the concentration of amino acid sulfur released from the dead cells was high enough to protect the wine. Thus, during the maturation no additional sulfurization was needed. The results of chemical analysis of both maturated wines are presented in Table 2.

Comparing the results of chemical analysis (Table 2), the lower amount of total alcohol in sulfurized wine is related to the greater residual amount of insoluble sugar. The higher amount of total alcohol (by 0.4 vol%) in unsulfurized wine can be attributed to a more environmentally friendly fermentation without sulfur inhibitor and earlier initiation of alcoholic fermentation. This results also in a higher residual biomass. The difference of 0.4 g L^{-1} of total acids is attributed to the successful malolactic fermentation in the unsulfurized wine. The higher level of volatile acids (by 0.32 g L^{-1}) in the unsulfurized wine is a consequence of the spontaneity of the process, but is still below the maximum level allowed by the legal norm, although it affects organoleptic properties of the mature wine. The higher free SO_2, by 1.52 mg L^{-1}, in unsulfurized wine at lower total SO_2 of 13.3 mg L^{-1}, points to the effectivity of *sur-lies* maturation. This also indicates the positive effect of autolysis. The higher ash content by 1.5 g L^{-1} in sulfurized wine gives it more consistency and minerality. The difference in relative density of only 0.001 g L^{-1} is evidence of the acceptability and credibility of both processes. The higher lactic acid content of 0.7 g L^{-1} in unsulfurized wine also indicates a softer taste of wine and more smooth tannins that give the wine more softness and velvetiness.

The results of the sensorial analysis are summarized and presented in Figs. 4–6. Fig. 4 presents the visual aspect of control samples of sulfurized wine in the first year of maturation (PL/S), unsulfurized wine in the first year of maturation (PL), sulfurized wine in the second year of maturation (DL/S), and unsulfurized wine in the second year of maturation (DL).

Except for the mark good for PL, in all other samples the limpidity was very good. Color intensity was highest for DL. This was influenced by the more expressed affinity of anthocyanins towards oxygen. The intensity of violet notes in Control, PL, and DLS are lower than in DL. Orange and brownish notes in DL are the consequence of the absence of sulfur.

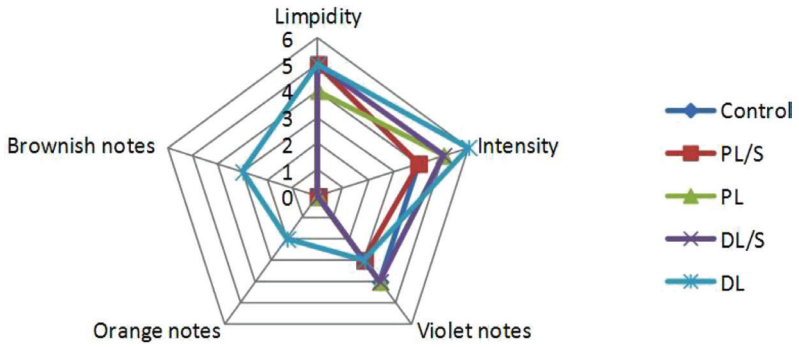

Fig. 4 – *Visual aspect of control sample: sulfurized wine in the first year of maturation (PL/S), unsulfurized wine in the first year of maturation (PL), sulfurized wine in the second year of maturation (DL/S), and unsulfurized wine in the first year of maturation (DL)*

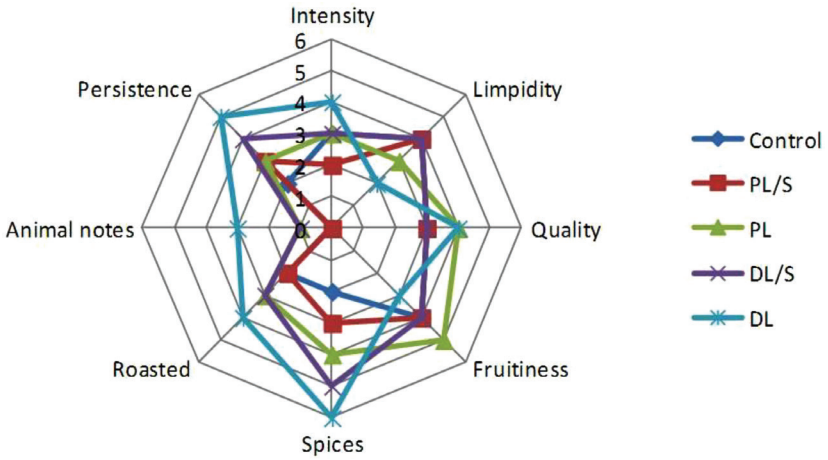

Fig. 5 – *Aroma aspect of control sample: sulfurized wine in the first year of maturation (PL/S), unsulfurized wine in the first year of maturation (PL), sulfurized wine in the second year of maturation (DL/S), and unsulfurized wine in the first year of maturation (DL)*

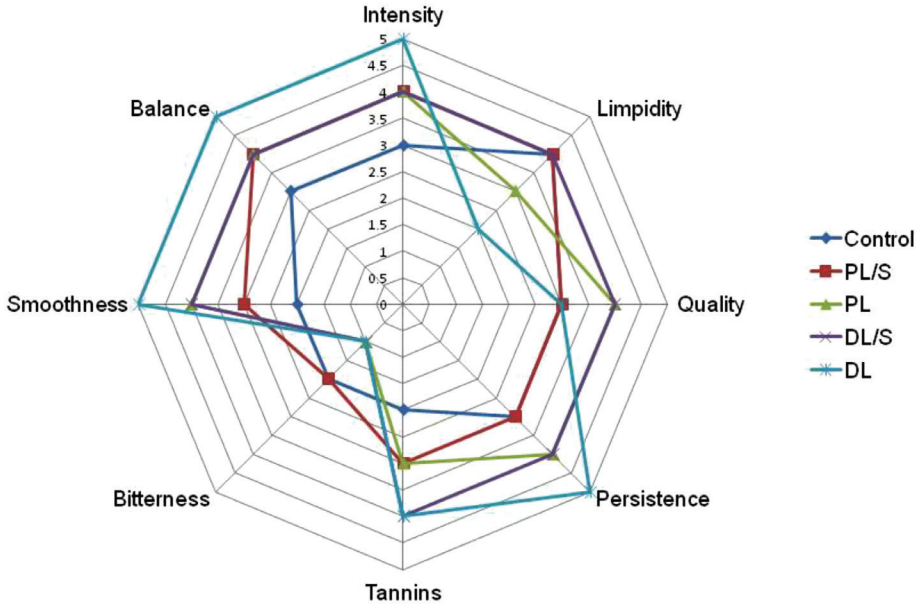

Fig. 6 – *Taste aspect of control sample: sulfurized wine in the first year of maturation (PL/S), unsulfurized wine in the first year of maturation (PL), sulfurized wine in the second year of maturation (DL/S), and unsulfurized wine in the first year of maturation (DL)*

The most intensive wine aromas in DL originated from the faster development of aromatic compounds in sulfur-free environment. Quality of maturation aromas in sulfurized and unsulfurized wines is very comparable. The most intense fruitiness was expressed in PL, while in the next year of maturation, those compounds were covered over with spicy and animal notes. The most intensive spiciness, animal, and roasted notes were detected in sulfur-free wines. They resulted from the micro-oxygen maturation of sulfur-free wines. The highest persistence was detected for DL, where maturation process in the presence of micro-oxygen was more intensive.

The taste intensity was also the highest in DL, while the taste limpidity was the most expressed in sulfurized wines PL/S and DL/S. Overall quality of all the samples was good and not related to the sulfurization. The bitterness of the unsulfurized wines was at the lower level, and the smoothness and tannins were at a higher level in comparison to the sulfurized wines, which points to its longer durability in bottles.

Conclusions

In barrel wine maturation, the measurement of redox potential was found to be an excellent tool for monitoring of the maturation process and yeast activity. Redox potential layers were discovered during the maturation process in the oak barrels and interpreted for the first time. The heterogeneity or redox potential indicated that the process of maturation was still active. At constant cellar temperature, the variation of the redox potential between the layers was found to depend on the yeast activity, as well as on the porosity of the barrels. The end of yeast activity was indicated with the homogeneity of redox potential. This may be interpreted in terms of readiness for bottling.

In the sulfurized wine, yeast was deactivated faster, and the bottling point was indicated already after 24 months. In the unsulfurized wine, however, yeast activity was prolonged and cell autolysis was more spontaneous. The autolyzed yeast cells released sulfur compounds that spontaneously protected the wine against oxidation. The process was much longer in comparison to the sulfurized wine, so maturation ended after 36 months and the wine was found ready for bottling. Sensorial analysis also supported the interpretation of redox potential data. In conclusion, the new *sur-lies* process of maturation in the barrels with no additional sulfur is a recommendable process of maturation until the final wine bottling[29,30].

References

1. *Rabotnova, I. L.,* Die Bedeutung physikalisch-chemischer Faktoren für die lebenstatigkeit der Bakterien (in German), Fischer-Verlag, Jena, 1963, pp 46–48.
2. *Ishizaki, A., Snibai, H., Hirose, Y*, Basic aspects of electrode potential change in submerged fermentation, Agr. Biol. Chem. **38** (1974) 2399. doi: https://doi.org/10.1080/00021369.1974.10861537
3. *Berovic, M., Cimerman, A.,* Redox potential in submerged citric acid fermentation, Eur. J. Appl. Microbiol. **16** (1982) 185. doi: https://doi.org/10.1007/BF00505830
4. *Berovic, M., Cimerman, A.,* Redox Potential – an Effective Tool for Scaling-up of Citric Acid Fermentation from Laboratory to Pilot Scale, Ed. A. Nienow, BHRA, 3rd International Congress on Bioreactor and Bioprocess Fluid Dynamics, MEP Publications, Cambridge, United Kingdom, 1993, pp 533–545.
5. *Hewitt, L. F.* Oxidation-Reduction Potentials in Potentials in Bacteriology and Biochemistry, 6th ed., Livingstone, Edinburgh, United Kingdom, 1950, pp 87–89. doi: https://doi.org/10.5962/bhl.title.4516
6. *Tengerdy, R. P.,* Redox potential changes in 2-keto-L-gulonic acid fermentation. Correlation between redox potential and dissolved oxygen concentration, Biotechn. Bioeng. **3** (1961) 255. doi: https://doi.org/10.1002/jbmte.390030305
7. *Sukharevich, V. I., Yakovljeva, E. P., Tsyganov, V. A., Shvezova, N. N.,* The effect of aeration and redox potential of the medium on biosynthesis of levorin A and B. (in Russian) Mikrobiologya **39** (1970) 981.
8. *Radjai, M. K., Hatch, R. T., Cadman, T. W.,* Optimisation of amino acid production by automatic self tuning digital control of redox potential, Biotechn. Bioeng. Symp. **14** (1984) 657.
9. *Berovic, M.,* Redox Potential in Submerged Citric Acid Fermentation. In: Citric acid Biotechnology, Eds. Kristiansen B., Mattey, M., Linden, J., Taylor & Francis, Corp., London, United Kingdom, 1999, pp 85–103.
10. *Berovic, M.,* Scale-up of citric acid fermentation by redox potential control, Biotechnol. Bioeng. **64** (1999) 552. doi: https://doi.org/10.1002/(SICI)1097-0290(19990905)64:5<552::AID-BIT5>3.0.CO;2-2
11. *Jacob, H. E.,* Redox Potential, Methods in Microbiology, in Norris, J. R. and Ribbons D. W. (Eds.), Vol. 2, Academic Press, London, 1970, United Kingdom, pp 91–123.
12. *Jacob, H. E.,* Reasons for redox potential in microbial cultures, Biotechn. Bioeng. Symp. **4** (1974) 781.
13. *Stolz, P., Bocker, V., Vogel, R. F., Hammes, W. P.,* Utilisation of maltose and glucose by *Lactobacilli* isolated from Sour dough, FEMS Microbiol. Letters **109** (1993) 237. doi: https://doi.org/10.1111/j.1574-6968.1993.tb06174.x
14. *Vonktaveesuk, P., Tonokawa, M., Ishizaki A.,* Simulation of the rate of L-lactate fermentation using *Lactoccocus lactis* Io-1 by periodic electrodyalisis, J. Ferm. Bioeng. **77** (1994) 508. doi: https://doi.org/10.1016/0922-338X(94)90119-8
15. *Peck, M., Chynoweth, D. P.,* On-line fluorescent monitoring of the metanogenic fermentation, Biotechnol. Bioeng. **39** (1992) 1151. doi: https://doi.org/10.1002/bit.260391112
16. *Heppner, B., Zellner, G., Diekmann, H.,* Start-up and operation of a propionate degrading fluidized bed reactor, Appl. Microbiol. Biotechnol. **36** (1992) 810. doi: https://doi.org/10.1007/BF00172200

17. *Beck, S., Schink, B.,* Acetate oxidation through a modified citric acid cycle in *Propionibacterium freudenreichii,* Arch. Microbiol. **163** (1995) 182. doi: https://doi.org/10.1007/BF00305351
18. *Hallborn, J., Gorwa, M. F., Meinander, N., Pentilla, M., Keranen, S., Hahn-Hägerdal, B.,* The influence of cosubstrate and aeration on xylitol formation by recombinant *Saccharomyces cerevisiae* expressing the xyl1 gene, Appl. Microbiol. Biotechnol. **42** (1994) 326. doi: https://doi.org/10.1007/s002530050258
19. *Bühler, H., Galster, H.,* Redox Measurement– Principles and Problems, Dr. W. Ingold AG, Zürich, Switzerland, 1980, pp 12–18.
20. *Kjaergaard, L., Joergensen, B. B.,* Redox potential as state variable in fermentation systems, Biotechn. Bioeng. Symp. **9** (1979) 85.
21. *Clark, W. M., Cohen, B.,* The determination of hydrogen ions, Publ. Hlth. Rep. Wash. **38** (1923) 666. doi: https://doi.org/10.2307/4576704
22. *Berovic, M., Rošelj, M., Wondra, M.,* Possibilities of redox potential regulation in submerged citric acid bioprocessing on beet molasses substrate, Food Technol. Biotechnol. **38** (2000) 193.
23. *Lavigne, V., Dubourdieu, D.*, De l'aptitude des lies, J. Int. Sci. Vigne Vin (in French) 30 (1996) 201.
24. *Fornairon-Bonnefond, C., Camarasa, C., Moutounet, M., Salmon, J. M.,* New trend on yeastautolysis and wineageing on lees: A bibliographique review (in French), J. Int. Sci. Vigne Vin. **35** (2001) 57.
25. *Vivas, N, Nonier, M. F.,* L'elevage en barrique sur lies des vins rouges. Interet des oxydations en conditions de reduction (in French), Wine Int. Tech. J. **7** (2005) 64.
26. *Dittrich, H. H., Wedler, A., Sponholz, W. R.,* Überseltene Zucker in Mostenausgesunden und *Botrytis*-infizierten Traubenbeeren, wie in den darausgewonnen Weinen. Wein-Wiss. (in German) **35** (1976) 25.
27. Bio-Rad, Guide to Aminex HPLC Columns for Food and Beverage Analysis, Richmond, USA, 1997, pp 24–28.
28. The rules on the procedure and method of assessing must and wine, Uradni list Republike Slovenije. (Official Gazette of the Republic of Slovenia, in Slovenian) 32/00, 99/01.
29. *Killeen, D. J., Boulton, R., Knoesen, A.*, Advanced monitoring and control of redox potential in wine fermentation, Am. J. Enol. Vitic. (2018). doi: https://doi.org/10.5344/ajev.2018.17063
30. Using redox measurements to control an anaerobic yeast fermentation (Application Forum – Sponsored Paper), BioTechniques **45** (2008) 670. doi: https://doi.org/10.2144/000113076