

Original Research Article

<https://doi.org/10.20546/ijcmas.2019.811.235>

Antioxidant Protection during Processing of Red Grapes and Processing of Red Wine

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ABSTRACT

Currently, demand for light red wines has risen sharply. But the color of the wine loses its color and luster during technological treatments associated with the presence of oxygen. Therefore, the study of antioxidants, antioxidant protection system in the processing of red grapes and processing of wines in order to ensure the quality and presentation of red wines is relevant. The work investigated the system of antioxidant protection (AOD) of products of primary and secondary winemaking and describes the results of research on the determination of enzymes of the AOD system during all technological methods: crushing, dripping, sludge, fermentation, pasting, sludge, heat treatment, cold red wine and wine processing. Before and after each technological operation, the physicochemical parameters, the activity of the enzymes in the AOD system were determined, a cascade of oxygen reduction reactions and oxygen reduction reactions in wine are given. As a result, on the basis of the obtained results, the technological canons of winemaking were corrected, for example, during the processing of red grapes, additional measures are clearly needed to provide antioxidant protection. Moreover, cold treatment of dry red wines leads to an increase in oxygen concentration, which threatens to oxidize the phenol - coloring matter in wine and causes a change in color and taste of wine, then this technological method for red wines is recommended in exceptional cases. Consequently, the treatment with heat of red wines provides an increase in the concentration of oxygen and at the same time activates all the component systems of AOD, there is a dismutation and the formation of organic peroxides. Over time, heat treatment enhances AOA, with that, AOA increases more intensively during heat treatment, slightly lower than AOA during technological processing with cold.

Keywords

Oxidation, Oxygen, Enzyme, Antioxidants, Antioxidant defense system

Article Info

Accepted:

17 October 2019

Available Online:

10 November 2019

Introduction

Oxygen plays a key role in the metabolic and chemical reactions of the winemaking process. Wine cannot be completely protected from oxygen throughout the process of making

wine. In the works of W.J.du Toit, the effect of oxygen on both the composition and quality of wine is noted either positively or negatively (1) (Fig. 1 and 2). The oxidation can be enzymatic and non-enzymatic, which is also called auto-oxidation. In wine, usually non-

enzymatic, i.e. by chemical oxidation. In the case of enzymatic oxidation, enzymes such as polyphenol oxidase and superoxide dismutase catalyze oxidation reactions.

The spectrum of antioxidants that make up the wine is represented by phenolic (2) components of a mono-, oligo- and polymer structure, anthocyanins, ascorbic acid, catechins, tannin, etc., which are involved in the processing of all forms of aroma oxygen and taste, determining the quality of wines (3).

Antioxidants are a shield against environmental toxins (four). The antioxidant level depends on the grape variety, the place of growth and the method of its processing (5). Subsequently, we studied the antioxidants of the antioxidant protection system during technological operations for processing grapes (crushing, dripping, pressing, sludge, fermentation) and processes that contribute to the production of wines of stable quality and high biological value that occur in secondary winemaking (pasting, heat treatment, low temperature and heating). The antioxidant defense system is divided into primary and secondary, in which enzymes and vitamins are the antioxidant, respectively. Therefore, the study of the influence of technological methods, with various oxygen saturation and the presence of reactive oxygen species on the behavior of AODs stained wines is relevant.

Since all easily oxidized components of wine are antioxidants, the possible presence of radicals and reactive oxygen species predetermines their interaction. Red wines are characterized by a high content of antioxidants mainly of a phenolic nature, differing in both qualitative and quantitative composition.

The aim of the research is to determine the state of enzymes of the antioxidant defense system in the process of processing red grapes and technological processing of red wine

material. Primary winemaking techniques were carried out on grape processing lines of primary winemaking factories; pasting, cold processing and heat treatment were carried out according to the approved technological instructions in the secondary winemaking enterprise mode.

Physico-chemical characteristics of grapes and wine material were determined by methods common in winemaking.

The object of the study was red Pinot black grapes with a sugar content of 18% and an acidity of 5 mg / dm³. Indicators of red wine material: specific gravity - 0.990 g / cm³; fortress - 11.2% vol; titratable acidity - 5.6 mg / dm³; volatile acidity - 0.59 mg / dm³; SO₂-100mg / dm³ content; Fe-14 mg / dm³ content.

The activity of enzymes was determined: according to a method based on the ability of superoxide dismutase to inhibit the reduction reaction of nitrotetrazolium blue. Catalase activity was determined by reaction with ammonium molybdate, and peroxidase activity was determined by the method based on the oxidation of pyrogallol in the presence of hydrogen peroxide to purpurogallin. The antioxidant activity was determined on a PU-1 polarograph by taking a voltammogram of the current of the test substance, and NaClO₄ dissolved in dimethylformamide was used as the background electrolyte.

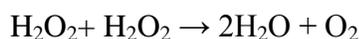
All groups of wine substances are involved in redox reactions - carbohydrates, phenolic and nitrogenous substances, organic acids. The intensity of the passage of oxidative enzymatic processes depends on technological methods, creating conditions for the passage of secondary redox processes. The state of the components of antioxidant protection determines the resistance to environmental influences.

Studying the antioxidant protection of red wines will allow you to properly build the technology of wine making. Crushing red grapes does not change the oxygen concentration and remains in the amount of 14 mg / dm³. The activity of the catalase enzyme in red wort increased from 3.93 μmol / min / dm³ to 4.35 μmol / min / dm³. Peroxidase activity, on the contrary, decreased from 40.6 μmol / min / dm³ to 22.3 μmol / min / dm³. Superoxide dismutase activity increased from 2.93 conventional units to 4.60 conventional units.

In general, crushing red grapes activates the superoxide dismutase (SOD) enzyme and catalase in the wort. SOD and catalase reduce the level of primary reactive oxygen species (ROS) and contain iron ions as catalysts.

An increase in SOD activity determines the presence of a superoxide oxygen radical, which intensifies the oxidation process and SOD protects against excessive oxidation (6).

Catalase, by oxidizing one hydrogen peroxide molecule with another hydrogen peroxide molecule, forms two water molecules and an oxygen molecule:



When red wort drains, oxygen saturation is observed, at which the oxygen concentration increased from 12 to 20 mg / dm³. That is, the intense absorption of oxygen led to an increase in its concentration by 8 mg / dm³. Peroxidase activity increases, but the enzymes SOD and catalase somewhat lose their activity.

Peroxidases are oxidizing agents of peroxides, that is, peroxidation is carried out according to the scheme: Molecular oxygen is gradually reduced to 2H₂O₂ during the oxidation process. Free superoxide (O₂ • -) and peroxide

radicals (O₂²⁻) are formed, which are directly reduced by stronger than O₂ phenolic oxidizing agents (7).

Pressing the red pulp reduces the accumulation of oxygen and the enzymatic activity of peroxidase, but the activity of the enzymes SOD and catalase increased. These data determine the presence of the superoxide oxygen radical during the pressing period, while the superoxide dismutase protects against excessive oxidation. Pressing activates this enzyme from 2.81 conventional units to 5.74 conventional units, which indicates the predisposition of red wort to oxidation. Here, an increase in catalase activity is observed from 3.95 μmol / min / dm³ to 4.77 μmol / min / dm³, which oxidizes one molecule of hydrogen peroxide. Apparently, pressing, as a technological method, enhances peroxidation, thereby providing antioxidant protection for red wort.

When red wort settles, peroxidase activation is observed and oxygen accumulation is intensified. The activity of SOD and catalase decreases from 4.76 μmol / min / dm³ to 1.9 μmol / min / dm³. The infusion increases the oxygen concentration in the red wort from 10.3 mg / dm³ to 24 mg / dm³. Peroxidase activity intensifies peroxidation reactions. The chemical composition of the red wort makes it possible to prevent the ionization of molecular oxygen.

Fermentation of red must is not the same effect on the enzymatic activity of the antioxidant defense system. High oxygen saturation is observed during fermentation, when the oxygen concentration increased to 13 mg / dm³.

If we judge the oxidation of wort by the number of active enzymes that make up the antioxidant defense system, then when crushed, red grapes are most susceptible to

oxidation. Processing of red grape varieties is characterized by the maximum activity of SOD during runoff, then during fermentation, crushing, pressing and is absent only with the infusion of red wort. That is, all operations in the processing of grapes, except for the wort infusion, tend to oxidize red wort, and the danger of oxidation of one hydrogen peroxide molecule by another hydrogen peroxide molecule with the formation of two water molecules and an oxygen molecule is significant (8).

But peroxidase activity in the processing of red grapes appeared in only two cases: during runoff and infusion. It can be concluded that when processing red grapes, the wort is less susceptible to peroxidation.

Red wines are characterized by a rich complex phenolic complex and are characterized by high antioxidant ability, predetermined by a high content of phenolic and coloring substances.

From the results of the analysis it follows that the redox processes occurring in red wines during the ripening period are due to the absorption of oxygen contained in the air and entering the wine during technological operations.

Pasting with bentonite saturates red wine with oxygen. And the maximum (8.5 mg / dm³) increase in the concentration of molecular oxygen was noted after treatment, and the smallest increase (only 0.62 mg / dm³) was observed during cold treatment.

Hydrogen peroxide is a relatively weak oxidizing agent with respect to the components of wine, but again, in the presence of iron, it is reduced to the hydroxyl radical, a very strong oxidizing agent. The highest catalase activity was determined before cold treatment. It is the low-

temperature treatment that inactivates it. An increase in its activity is provided by pasting with bentonite, and during this technological treatment, the increase in catalase activity is about ten times higher than during heat treatment.

Peroxidase is active at the beginning and end of wine ripening. Glutathione peroxidase activity in all samples increases, and it is maximal after pasting the wine and minimal (0.059 mmol / min / dm³.) With heat treatment, which confirms the existing hypothesis that heat treatment accelerates the maturation of wines. And this technique is recommended to accelerate the maturation and typing of wines.

The superoxide dismutase enzyme is most active before cold treatment and it is this technological technique that sharply reduces its activity (by 5.75 conventional units), and when glued and treated with heat, its growth is the same (1.74 conventional units). Superoxide dismutase catalyzes the dismutation reactions of the superoxide radical to produce hydrogen peroxide and oxygen. An increase in the activity of superoxide dismutase, catalase, peroxidase, and molecular oxygen is observed during gluing, as well as during heat treatment, which is a vivid demonstration of the presence of antioxidant protection of red wines (9).

Compounds of antioxidant protection behave somewhat differently in low-temperature processing. Thus, cold treatment gives a small increase in the concentration of molecular oxygen and peroxidase, but the activity of superoxide dismutase and catalase is reduced. In the process as cold treatment, a sharp drop in the activity of superoxide dismutase by 5.75 conv. units, loss of catalase activity by 1.45 conventional units

Technological method-pasting gives the greatest increase in the activity of catalase and

peroxidase with an increase in all the studied parameters of antioxidant protection.

Heat treatment enhances the activity of the antioxidant defense system. However, the increase in molecular oxygen concentration during heat treatment is approximately two times less than during gluing.

Consequently, cold treatment enhances the tendency of wine components to oxidize, while technological methods like gluing and heat treatment protect wine components from oxidation.

Antioxidant activity accepts free radicals. No direct correlation between antioxidant activity and molecular oxygen concentration was found in this case. The absence of such a correlation indicates the presence of antioxidants of various nature.

The results of studies to determine the antioxidant activity of red dry wine material during technological processing are shown in the diagram (Fig. 3). Explicit antioxidant activity is typical for heat treatment (64 nmolO₂ / min / ml), antioxidant activity is slightly lower during cold processing (28 nmolO₂ / min / ml) and minimal antioxidant activity is characteristic (10 nmol O₂/ min / ml) for gluing the technological method used to clarify wines and wine materials. In this case, the quantitative component remains unchanged throughout the duration of the treatment. A sharp increase in antioxidant activity gives a temporary factor in the heat treatment of red wine. Slightly lower than antioxidant activity during cold processing

It should be noted that the studied enzymatic systems of antioxidant protection are quite complex and therefore the technological methods adopted in winemaking inadequately affect their activity. So, the maximum concentration of molecular oxygen (10.64 mg

/ cm³) was noted after heat treatment of dry red wine. The greatest change in molecular oxygen concentration was observed in red wine after treatment with cold. The oxygen concentration decreased by 5.57 mg / cm³, and the largest increase in oxygen concentration (4.63 mg / cm³) was noted in red wine after heat treatment.

Catalase activity is maximal (8.12 μmol / min / dm³) in red dry wine after pasting it with bentonite. The increase in the activity of the catalase enzyme was the largest (2.2 μmol / min / dm³) in red dry wine treated with gluing agents.

Peroxidase is an enzyme related to catalase and inactivates H₂O₂ and other peroxide compounds (10). Peroxidase had the highest activity among other samples in red dry wine after processing with cold. Pasting red dry wine gave the greatest increase in the activity of the enzyme peroxidase (0.438 mmol / min / dm³).

AOZ from reactive oxygen species is essential when crushing grapes, and slightly lower when wort drains

When processing red grapes, the wort is less susceptible to peroxidation.

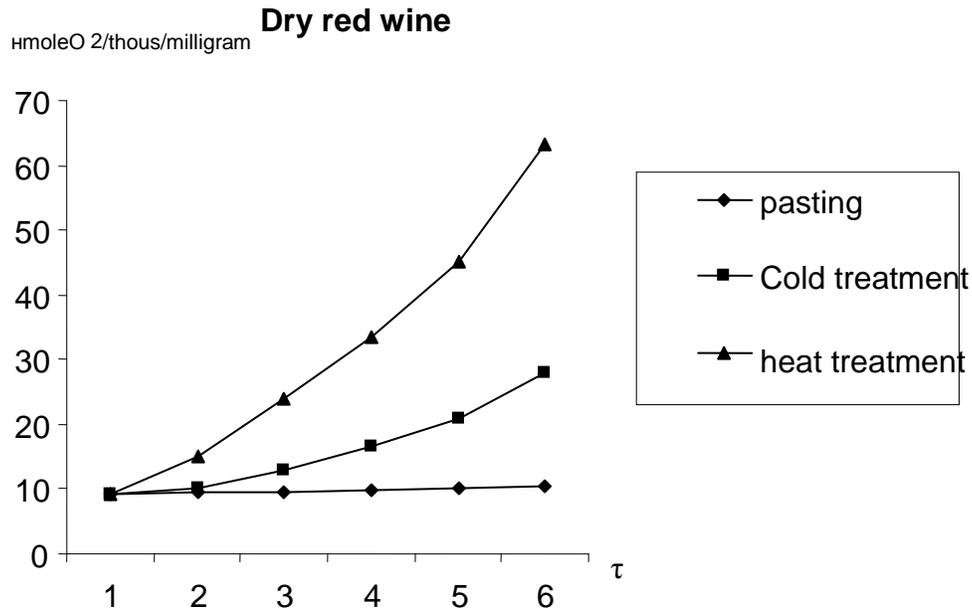
Processing of red grapes is clearly not sufficiently provided with antioxidant protection

Pasting of red wines is characterized by high AOD;

Cold processing inactivates all the studied enzymes of the AOZ system, that is, this technological technique gives the wine a steady state against oxygen stresses;

Since cold treatment of dry red wines leads to an increase in oxygen concentration, which

Fig.3 The antioxidant activity of red wine during technological operations



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How to cite this article:

Gulmira R. Irgasheva. 2019. Antioxidant Protection during Processing of Red Grapes and Processing of Red Wine. *Int.J.Curr.Microbiol.App.Sci.* 8(11): 2026-2033.
doi: <https://doi.org/10.20546/ijemas.2019.811.235>