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THE INFLUENCE OF ULTRASOUND TREATMENT ON MUST FERMENTATION PROCESS

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Abstract: *In order to meet consumer requirements for food safety products, the producers need to provide high quality wines. As an alternative to the traditional winemaking practices, new emerging technologies can be used among which is the ultrasound technique. During fermentation, yeasts transform sugars present in the juice into ethanol and carbon dioxide. Controlling the fermentation process is essential to improve the quality and organoleptic properties of the final product. Ultrasound can be used in the must fermentation process to both monitor and influence its progress. In this paper, the effects of ultrasound on the evolution of must fermentation during the winemaking process were investigated. The main objective was to examine the influence of ultrasonic treatment time on alcoholic fermentation.*

Keywords: *ultrasound, winemaking, fermentation, maturation.*

1. Introduction

The compounds contained in the grapes together with the winemaking process influence the final quality of the wine. In order to express and define the overall quality of the wine, the physicochemical parameters like pH, total and volatile acidity, alcohol content, sulphur dioxide and sugars are generally used.

The sensory analysis, including the colour of the wine, total phenolic compounds, total concentration of antocyanins and the level of tannins complete the panel of measurable parameters related to wine quality and stability [5].

For wineries, the fermentation process is a very important technological part of the winemaking process due to the influence in the obtained wine.

Face with consumer demand for high quality wines and increasing pressure to optimize production and costs, wineries are constantly looking for alternatives to apply during processing. One of such emerging technologies is ultrasound that can be used in fermentation to either monitor the progress or to influence it [6].

Ultrasound is an efficient physical method, non-thermal, non-hazardous, environmental friendly, and inexpensive.

Ultrasound is sound waves with frequencies higher than the upper audible

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limit of human hearing (16–20 kHz). In the food industry, the sound ranges employed can be divided into high frequency diagnostic ultrasound and low frequency ultrasound. High frequency diagnostic ultrasound (above 100 kHz) is a non-destructive technique used for quality assurance and, monitoring of food processes and causes no physical or chemical alterations. The low frequency ultrasonic waves (18 – 100 kHz) are capable of altering material properties: physical disruption, acceleration of chemical reactions [4], [7].

In oenology, ultrasound has been studied for its potential to accelerate reactions within the wine, to increase phenolic compounds extraction or antimicrobial effects [5]. In the literature, the latest research showed the potential uses of ultrasound in winemaking including reducing the fermentation time and increasing the extraction of phenolic compounds [1-2], [8-11].

Ultrasound is a simple and rapid extraction method, being an alternative for the analysis of wine flavour components through advantages that include higher reproducibility and possibility of simultaneous extraction of several samples [5].

Some works indicate that application of ultrasound to wine might cause ingredients interactions, leading to chemical and structural changes in wine [5]. Other authors [3] studied the effect of combined ultrasound (40 kHz/20min)/SO_2 treatment on microbial-stability of Italian Riesling low alcohol sweet white wine and observed the improving of micro-stability of raw wines and a better taste, with typical and complex aromas and flavour. Also, no significant influence on titratable acidity, pH, free and total SO_2 was reported.

2. Objectives

In this paper, we examined the influence of ultrasonic treatment time on the alcoholic fermentation of the white wine by applying a laboratory-scale power ultrasound system to white grape must obtained from indigenous Riesling grape variety.

Riesling is a white aromatic grape variety which originated in the Rhine region of Germany. It has pronounced fruit flavours, and flowery, almost perfumed aromas as well as high acidity. Usually, it is used to make dry, semi-sweet, sweet, and sparkling white wines.

In winemaking, the delicate nature of the Riesling grape requires special handling during harvesting to avoid crushing or bruising the skin. Without this care, the broken skins could leak tannin into the juice, giving a markedly coarse taste and throwing off balance the Riesling's range of flavours and aromas.

The literature mentioned that during fermentation, the wine is cooled to between 10 and 18 °C in controlled stainless steel fermentation tanks. Most Riesling does not undergo malolactic fermentation in order to preserve the tartaric acid, an important parameter for Riesling wine quality. Riesling is often put through a process of cold stabilization, where the wine is stored just above its freezing point. After this, the wine is normally filtered again to remove any remaining yeast or impurities.

3. Material and Methods

Grapes were supplied by Pietroasa Development Research Center for Viticulture and Wine-making, located in Dealu Mare vineyard, Romania. The

variety used for this research was Riesling.

The grapes were transported on the same day to the laboratory for processing.

The grapes were destemmed and crushed, and deposited in a stainless steel tank where the temperature was measured and controlled. The resulting must was treated with Bentonite (1 g. per litre) and enzyme (Enozyme Arome white skin maceration, 4g. per 100 litres) were added. After 24 hours, selected yeasts were added (Viniferm Saccharomyces cerevisiae Agrovin, Spain, 20g. per 100 litres). Four different samples were treated with a laboratory-scale power ultrasound system (Sonics & Materials Inc. U.S.A.) for 3 (US3), 5 (US5), 8 (US8) and respectively 10 (US10) minutes. The system operated at 750 W and 20 kHz frequency. A batch of must was not treated (control vinification). 5 litre vessels were filled with the same quantity and proportion to assure the same liquid ration in each vessel and fermented in a temperature-controlled climate chamber at 17-19 °C.

When alcoholic fermentation was finished, the wines were cold stabilised at 9°C and then the SO2 was applied and filtration stage took place. Must and wines were analysed prior to the ultrasound treatment, after the ultrasound treatment, during the fermentation period and at the end of alcoholic fermentation. Figure 1 describes the flow diagram of white winemaking process.

4. Analytical Determinations

4.1. Spectrophotometric Parameters

The colour density (CD) for the white wine was calculated as the absorbance at 420 nm using the optical system of Hanna HI 83742 series colorimeters. The measurement process was carried out in two phases: first the meter was zeroed and then the actual measurement was performed.

Total phenols (TP) were calculated based on the reaction of phenolic substances with Folin-Ciocalceu reagent. The reaction between phenols and the Folin-Ciocalteu reagent involves oxidation of the phenolic groups (R-OH) with a mixture of phosphotungstenic acid (H3PW12O40) and phosphomolybdenic acid (H3PMo12O40) to the quinoid form (R=O). The concomitant reduction of the Folin-Ciocalteu reagent causes a blue colour in the sample that is proportional to the total phenolic content that, in turn, is expressed as g/L of Gallic Acid Equivalents (GAE). TP were calculated by measuring wine absorbance at 610 nm.

4.2. Chemical Analysis

The tartaric acid was determined using a HI83748 photometer that uses a method with two reagents to determine the concentration of tartaric acid less than 5.0 g/L (ppt). When both reagents are added to a sample containing tartaric acid, the sample turns an orange-red hue; the greater the concentration, the deeper the colour. The associated colour change is then colorimetrically analyzed according to the Beer-Lambert Law. This principle states that light is absorbed by a complementary colour, and the emitted radiation is dependent upon concentration. For determination of reducing sugars, a narrow band interference filter at 525 nm (green) allows only green light to be detected by the silicon photodetector and omits all other visible light emitted from the

tungsten lamp. As the change in colour of the reacted sample increases, absorbance of the specific wavelength of light also increases, while transmittance decreases.

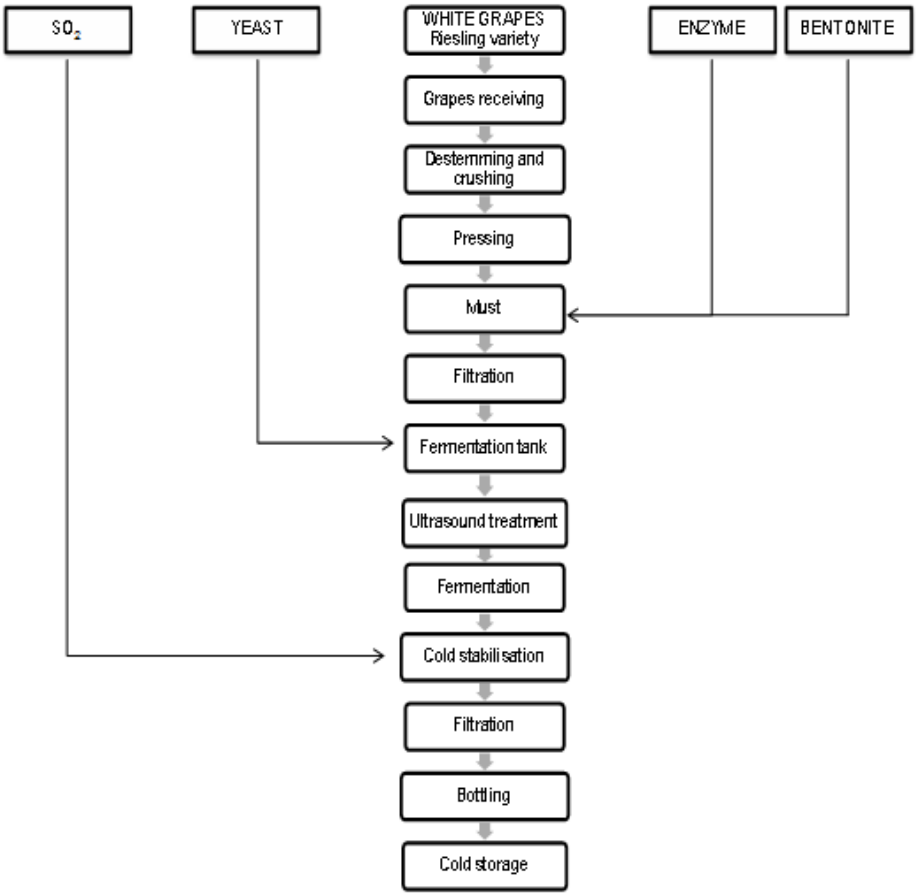

Fig. 1. *The flow diagram of white winemaking process*

The reducing sugars in wine were determined using a HI83746 photometer that uses the Fehling method to determine the concentration of reducing sugars less than 50.00 g/L (ppt). When Fehling's A and Fehling's B Solutions react with a sample containing reducing sugars, the sample undergoes a colour change; the greater the concentration, the deeper the colour. The associated colour change is then colorimetrically analyzed according to the Beer-Lambert Law. This principle states that light is absorbed by a complementary colour, and the emitted radiation is dependent upon concentration. For determination of reducing sugars, a narrow band interference filter at 610 nm (orange) allows only orange light to be detected by the silicon photodetector and omits all other visible light emitted from the tungsten lamp. As the change in colour of the reacted sample increases, absorbance of the specific wavelength of light also increases, while transmittance decreases.

5. Results and Discussions

The results of the chromatic parameters and chemical analysis are shown in Table 1. The initial must already had large differences in total phenols and colour density. The total phenolic compounds showed the highest value in control must, while the sonicated samples had lower values. Comparing the chromatic characteristics of the different samples at the initial moment, 8 min. sonicated samples (US8) showed the highest value of the colour density while the lowest was observed in US3.

The tartaric acid had the lowest value in the control must and the same value for all the sonicated samples. Also, the reducing sugars showed the same value for all the samples.

Comparing the total phenolic compounds, we observed that after fermentation, all the samples have higher values than initial must.

At the end of alcoholic fermentation, we found a slight decrease in terms of colour density except in US5 and US10 that showed higher values than the initial ones.

The highest colour density was found in 10 min. sonicated samples US10, and the lowest was observed in US3.

The values for the tartaric acid were higher at the end of alcoholic fermentation. The highest value was found in US3 while for the control wine, US8 and US10 had the same value.

Concerning reducing sugars, the highest value was observed for 5 min. sonicated samples US5 and the lowest for control wine and US3.

Chromatic characteristics and chemical analysis of the control and sonicated must and wines Table 1

Sample		**TP [g/L GAE]**	**CD**	**Tartaric acid [g/L]**	**Reducing sugars [g/L]**
Initial must	Control must	0,700		3,7	231
	US 3	0,609	0,251	4	231
	US 5	0,582	0,256	4	231
	US 8	0,599	0,263	4	231
	US 10	0,563	0,261	4	231
End of alcoholic fermentation	Control wine	0,951	0,237	4,5	1,25
	US 3	0,917	0,234	4,6	1,25
	US 5	0,917	0,287	4,1	2
	US 8	1,594	0,246	4,5	1,5
	US 10	1,361	0,291	4,5	1,5

TP – Total phenols; GAE - Gallic Acid Equivalents; CD - colour density.

5. Conclusions

The results obtained for total phenols showed that ultrasonic treatment had a large influence on these compounds, the values after fermentation being higher than the values from initial must. Also, an increasing of the colour density was observed for two samples at the end of alcoholic fermentation. The values obtained for tartaric acid showed an increasing for all the samples. All the samples, except US5 were in accordance with Romanian legislation that indicates the value 4.5 g/L as the minimum level of the tartaric acid [12]. The values of reducing sugars obtained at the end of the alcoholic fermentation showed that

sonication had influenced the fermentation time in terms of reducing it, but not for all the samples.

Most research, including this work was performed at a laboratory-scale level. The lack of standardisation relating to the sonication parameters indicates that further studies and assays are necessary due to the large variety of grape and winemaking techniques that might lead to different reactions to the ultrasound treatment.

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