CHARACTERIZATION OF SACCHAROMYCES CEREVISIAE STRAINS FROM PUHOI VINEYARD AT THE LABORATORY SCALE.

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Abstract: This paper presents the provisional results of the studies on the selection of most suitable yeast strains in a lot of 57 strains from Puhoi vineyard-Bulboaca wine centre, in order to be used for dry red wines. For achieving this objective, we used at first an initial test for checking the fermentation features of yeast strains, with the following targets: foaming ability, evolution in time (triggering, finish) of fermentation process stages. We selected 17 from 57 yeast strains. In the second test for checking fermentation features, conducted in the laboratory and using fermentation tanks of 2 litters, we checked the selected strains ability to stick or not to the walls of the fermentation tanks, the formation of granular or compact yeast deposits, the ability to completely ferment sugars from must, the property not to produce hydrogen sulphide and the influence on organoleptic properties. For this test we selected 10 strains from the 17 yeast strains.

Key words: strain, yeasts, fermentation, red dry wine.

Introduction

The use of selected yeasts for winemaking has clear advantages over traditional spontaneous fermentation. Selection of wine yeasts is usually carried out within the Saccharomyces cerevisiae species. Yeast strains produce different amount of secondary compounds that impart specific characteristics to the wines. This suggests that it is necessary to isolate naturally occuring autochthone strains, which exhibit a metabolic profile that corresponds to each wine.

The selection process of yeast strains depends on their oenological characteristics, such as fermentative rate, tolerance to ethanol, flocculent characteristics, the presence of killer factors, acetic acid production, H_2S , higher alcohol production, alcohol yield, glycerol production, etc. [1]. These and other technological developments have contributed to an improvement in the quality of wine, and have enhanced the ability of winemakers to control the fermentation process and achieve specific outcomes. Commercially available dried yeast strain starter culture of *S.cerevisaee* could be inoculated into the grape juice in order to establish a high population and accomplish well-controlled must fermentation. However, the use of local, indigenous, selected strains of *S.cerevisaeas* a starter culture are preferable [2], since these yeasts are better acclimated to micro area conditions of the wine producing region [3, 4, 5, 6, 7] and can dominate the natural flora easier. Moreover, such treatment, among others, could assure maintenance of the typical sensory properties and a characteristic profile of local wines.

The aim of our research was to select *Saccharomyces cerevisiae* strains that are starters for the production of dry red wines. We examined oenological differences,

affinities and suitability of yeasts for wine production at a laboratory scale for possible future starter production.

Materials and Methods

In the preliminary test, for the selection of yeast strains we used minifermentation tanks of 0,5 L, into which was poured 0.35 L of grape must from the variety of which the yeast strains were isolated. An inoculum was prepared from each activated yeast strain and its number of cells/mL was measured to determine the volume of inoculum introduced so that the density of cells/mL should be of $5 \cdot 10^6$ cells/mL in the grape must. For the test using fermentation tanks of 2 L we followed the same procedure. After introducing the inoculum and the fitting of boiling tanks, the alcoholic fermentation process was monitored each day, recording, for the preliminary test, the level of foaming and the quantification of the fermentation process' stages (hours /days), and in the test performed with 2 L fermentation tanks we tracked the adherence or non-adherence to the walls of fermentation tanks, the type of yeast lees and the reproducibility of the fermentation process stages. At the end of the alcoholic fermentation, the wines were characterized from physical-chemical and organoleptic point of view, according to OIV methods.

Results and Discussion

In 2013-2014 study period, we isolated 57 yeast strains from Merlot vineyard all pertaining to Bulboaca wine centre. In the preliminary test for detecting the fermentation characteristics of yeast strains, according to the foaming ability and the evolution in time (triggering, ending) of the fermentation process' stages, we selected 17 yeast strains.

The remaining strains checked, representing 70% of the total number, were removed, because they either triggered large amounts of foam, some of them even brimming over the fermentation recipients, or triggering the fermentation process after 2-3 days or, once activated it had a slow evolution, never finishing.

The 17 yeasts were selected because they produced very few foam in the first 24-48 hours, five strains being even included in the non-foaming yeasts category, namely PVM1, PVM9, PVM17, PVM20 and PVM24.

The evolution in time of the fermentation process' stages of the 17 yeast strains highlighted the fact that these ones triggered alcoholic fermentation under the best conditions for making high quality wines.

The next step in the selection of potentially productive yeast strains was the laboratory test in 2 L fermentation tanks. The selection criteria in this test were: the strains' ability to stick or not to the walls of the fermentation tanks, the formation of granular or compact yeast deposits, the ability to completely ferment must sugars and the property not to produce hydrogen sulphide.

The data collected in this test are showed in Table 1. In Table 1 we illustrate the results achieved in the assay conducted on the 17 yeast strains, selected from Merlot vineyard.

Yeast strain code	Stages of alcoholic fermentation process				+		Physical-chemical analyses of wine				
	Prefermentation stage. hours	Tumultuous fermentation, days	Slow Fermentation,	End of fermentation, davs	Adherence – Non-adherence	Yeast less	H ₂ S (quality)	Alcohol, vol. %	Fotal acidity, g/L	Residual sugars, ^{g/L}	Organoleptic analysis
PVM1	18	6	4	10	-	Compact	-	12,9	6,4	<3	Fresh, fruity odour, full
PVM2	18	6	4	10	-	Compact	-	12,9	6,5	<3	Fresh, harmonious odour, full
PVM5	20	7	5	11	-	Powdery	H_2S	12,8	6,4	<3	H ₂ S flavor and odour
PVM7	18	6	4	10	+	Powdery	-	12,7	6,4	<3	Fresh, fruity odour, harmonious, full
PVM8	22	8	5	12	+	Granular	H ₂ S	12,4	6,3	9,8	H ₂ S flavor and odour
PVM9	20	7	5	11	-	Compact	-	12,7	6,5	<3	Fresh, fruity odour, full
PVM13	20	7	5	11	-	Compact	-	12,8	6,5	<3	Fresh, fruity odour, full
PVM17	21	7	5	11	-	Powdery	H ₂ S	12,1	6,4	10,7	H ₂ S flavor and odour
PVM19	20	7	5	11	-	Compact	-	12,7	6,3	<3	Astringent, fruity odour, full
PVM20	18	6	4	10	-	Compact	-	12,8	6,3	<3	Fresh, fruity odour, full
PVM24	19	6	4	10	+	Compact	-	12,8	6,2	<3	Astringent, fruity odour, full
PVM27	18	6	4	10	-	Granular	-	12,7	6,4	<3	Fresh, fruity odour, full
PVM33	18	6	4	10	-	Granular	-	12,7	6,2	<3	Astringent, fruity odour, full
PVM37	20	7	5	11	-	Compact	-	12,9	6,3	<3	Astringent, fruity odour, full
PVM42	21	7	5	11	-	Compact	H_2S	12,4	6,2	8,5	H ₂ S flavor and odour
PVM45	22	8	5	12	-	Compact	H ₂ S	11,9	6,5	13,4	H ₂ S flavor and odour
PVM52	18	6	4	10	-	Compact	-	12,7	6,5	<3	Fresh, fruity odour, full

 Table 1. Characteristics of alcoholic fermentations of yeast strains selected in the preliminary test.

 Stages of alcoholic

From this yeast list we selected PVM1, PVM2, PVM9, PVM13, PVM19, PVM20, PVM27, PVM33, PVM37, PVM52 strains for making the red dry wine, because of the high alcohol content which determines the alcoholic fermentation process and the production of a dry wine, very appreciated from organoleptic point of view. The yeast lees are compact or granular, stable, and easily removable. Moreover, these strains do not stick to the walls of the fermenting tanks and produce dry wines, with no hydrogen sulphide odour, also having very good organoleptic features.

Conclusions

Following the research activity on the selection of yeast strains

suitable for dry red we isolated 57 new yeast strains from Puhoi – Bulboaca wine centre. In the preliminary selection test, 70% of the strains checked in the alcoholic fermentation process were removed. The 17 yeast strains selected were assessed in the process of alcoholic fermentation in tanks of 2 L, analyzing the physical-chemical and organoleptic features of the wines obtained. For this test we selected 10 yeast strains, which can be considered as high-performances ones due to the stable features in the fermentation processes and the achievement of quality wines.

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