

THE OENOLOGICAL CHARACTERIZATION OF YEAST STRAINS POTENTIALLY PERFORMANCE IN THE ALCOHOLIC FERMENTATION PROCESS, ISOLATED FROM THE INDIGEN FLORA OF THE VINEYARD OF IAȘI

CARACTERIZAREA OENOLOGICĂ A UNOR SUȘE DE LEVURI POTENȚIAL PERFORMANTE ÎN PROCESUL DE FERMENTAȚIE ALCOOLICĂ IZOLATE DIN FLORA INDIGENĂ A PODGORIEI IAȘI

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Abstract: *With the purpose of using the new yeast strains isolated and selected from the vines of the vineyard of Iași – the Copou Wine-Growing Center in the current wine-growing biotechnological practice, the study in standardized conditions of the following oenological characters was necessary: the degree of foam production, the clearness of the wine obtained, the type of yeasty sediment formed after the fermentation, the alcoholigenic power, the capacity of leading the alcoholic fermentation to different temperatures (11 – 13°C, 22°C and 35°C), as well as the resistance to different concentrations of sulphur dioxide (60 mg/L SO₂, 100 mg/L SO₂, 150 mg/L SO₂ ad 200 mg/L SO₂). The data obtained from the application of tests for the identification of the oenological characters allowed the selection of nine yeast strains considered of high performance in the production of high quality white wines.*

Key words: Copou, vineyard, yeast, fermentation, quality

Rezumat: *În vederea utilizării în practica biotehologică vinicolă curentă a noilor sușe de levuri izolate și selecționate din plantațiile viticole ale podgoriei Iași – centrul viticol Copou s-a impus studiul în condiții standardizate a următoarelor caractere oenologice: gradul de producere a spumei, limpiditatea vinului obținut, tipul de sediment levurian format după încetarea fermentației, puterea alcooligenă, capacitatea de a conduce fermentația alcoolică la diferite temperaturi (11 – 13°C, 22°C și 35°C), precum și rezistența la concentrații diferite de dioxid de sulf (60 mg/L SO₂, 100 mg/L SO₂, 150 mg/L SO₂ și 200 mg/L SO₂). Datele obținute în urma aplicării testelor pentru determinarea caracterelor oenologice au permis selectarea unui număr de nouă sușe ce pot fi apreciate ca fiind performante în producerea vinurilor albe de calitate.*

Cuvinte cheie: Copou, podgorie, levuri, fermentare, calitate

INTRODUCTION

Specialists in the field classify the yeasts important from the oenological standpoint based on their physiological and biochemical properties or on general criteria or criteria that correspond to particular wine-production conditions (1 – 4).

Polsinelli and his collaborators consider that the selection of yeasts involved in

the wine-production involve, on the one hand, knowing the fermentative properties (the quick trigger of fermentations, high capacity of fermentation, tolerance to alcohol, osmotolerance, resistance to temperatures, aromatic properties etc.) and, on the other hand, knowing the technological particularities (genetic stability, resistance to SO₂, low formation of foam, sulphites etc.)

Thus, with the purpose of implementing in the wine-producing biotechnological practice new sources of yeasts isolated from the vineyard of Iași – Copou Wine-Growing Center, a study was necessary of the main oenological characters of the new yeast strains isolated in pure culture, in standardized conditions.

MATERIAL AND METHOD

In order to determine the degree of foaming of the yeast strains, calibrated cylinders of 1,000 cm³ were used, in which 800 cm³ of sterile must was put. The sterile must divided to calibrated cylinders was inoculated with the same quantity of active inoculum. Upon inoculation, the density of the cells/mL was approximately the same (1 x 10⁵). The cylinders were maintained at the temperature of 22°C. For 72 hours, the quantity (cm³) of foam produced by each yeast was recorded every 24 hours. The clearness and the characteristics of the yeasty sediment were established by visual examination, after the alcoholic fermentation. From the clearness standpoint, the wine was evaluated as clear, opalescent, turbid, and the appearance of the yeasty sediment was evaluated as powdery or granular. The degree of adherence to the walls of the recipients was also evaluated.

In order to determine the alcoholigenic power, grape must sterilized in the steamer was used, in different concentrations of sugars by adding concentrated must. The must was allotted to recipients and then sown with 1 mL of three-day-old suspension, with the density of 1 x 10⁵ cells/mL and thermostatically subject to 22°C. After 30 days from the sowing, the alcoholic degree (vol. %) and the non-fermented sugars (g/L) were determined. The alcoholic power was determined after the alcoholic fermentation by measuring the ebullition and recording the obtained alcohol degrees.

The fermentative capacity at different temperatures was also tested on a set of glass recipients in which must sterilized in the steamer at 0.8 atm. was put. For each yeast strains, three recipients were prepared that were inoculated and incubated at different temperatures of 13-14°C, 22°C and 35°C. After 30 days, the alcoholic degree and the residual sugar were established.

The fermentation capacity of the sugars of the must in different concentrations of sulphur dioxide was evaluated, for each yeast strains, on a set of glass recipients of 500 mL, in which sterilized grape must was introduced. After the allotment of the must, calculated amounts of SO₂ were added for the achievement of the concentrations of 60 mg/L SO₂, 100 mg/L SO₂, 150 mg/L SO₂ and 200 mg/L SO₂. Such recipients were inoculated with 1 mL of 3-day old yeasty suspension of 1 x 10⁵ cells/mL and thermostatically subject to 22°C. After 30 days from the sowing, the alcoholic degree and the non-fermented sugars for each yeast were measured.

RESULTS AND DISCUSSIONS

With a view to selecting new yeast strains of high performance in the production of high quality white wines, 17 yeast were studied. The results regarding their oenological characters are presented in tables 1-4.

The analysis of the data specified in table 1, regarding the degree of foaming,

the type of sediment and the clearness of the end product, reveal that six yeast sources are average foaming, six are minimally foaming and five are non-foaming, namely MNF6, F1(75), S6(75), MNC2 and C1(100).

Depending on the type of sediment, it was found that 70 % of them formed compact deposits and only five yeast strains formed powdery or floury deposits. According to the clearness character of the obtained wines ten yeast sources led to clear wines, namely four yeast sources isolated from the plantation of Fetească alba, three yeast strains of the plantation Sauvignon blanc and three from the plantation of Chardonnay. The rest of the yeast strains led to the achievement of turbid or opalescent wines.

Table 1

Evaluation of the degree of foaming, of the type of sediment and of the clearness of the end product

No.	Yeast code	Foam volume, (cm ³ /L)			Type of sediment	Clearness
		24 hours	48 hours	72 hours		
1	MNF1	10	5	-	compact	opalescent
2	MNF5	60	10	10	compact	turbid
3	MNF6	-	-	-	compact, non-detachable	clear
4	F1(75)	-	-	-	compact	clear
5	F1(100)	50	-	-	compact	turbid
6	F2(100)	140	5	5	floury, easily detachable	clear
7	F1(200)	10	-	-	compact, difficultly detachable	clear
8	MNS6	6	-	-	compact	clear
9	S4(75)	10	10	5	floury, easily detachable	opalescent
10	S5(75)	15	5	-	powdery, dense	turbid
11	S6(75)	-	-	-	compact	clear
12	S3(150)	9	-	-	compact	clear
13	BC1	50	50	50	powdery, detachable	turbid
14	MNC2	-	-	-	compact	clear
15	MNC4	25	-	-	Powdery, easily detachable	turbid
16	C1(100)	5	-	-	compact	clear
17	C4(100)	-	-	-	compact	clear

Note: for the yeast strains 1 – 12 the must concentration was 210 g/L;
for the yeast strains 13 – 17 the must concentration was 235 g/L;

The results regarding the capacity of the studied yeasts of fermenting the musts with different concentrations of sugars are specified in table 2. The specified data reveal that the new yeast strains isolated lead to the achievement of dry wines when using musts with concentrations of 210 and 235 g/L respectively in sugars.

The presence of musts with concentrations of 260 g/L sugars, the yeasts MNF6, S6(75), MNC2 and C1(100) led to the achievement of dry wines and the yeast strains F1(75), F2(100), MNS6 to semi-dry wines. The rest of 58.8% of the tested yeast strains produced semi-sweet wines. As regards the concentration of 305 g/L sugars in the must, all tested yeast strains achieved sweet wines with non-fermented sugars between 58 and 87 g/L.

Table 2

Capacity of accomplishing alcoholic fermentation in different concentrations of sugars

No.	Yeast code	Must with 210 g/L și /235 g/L sugars		Must cu 260 g/L zaharuri		Must cu 305 g/L zaharuri	
		Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L
1	MNF1	12,3	-	13,8	25,0	13,2	80
2	MNF5	12,3	-	15,3	-	14,2	63
3	MNF6	12,3	-	14,4	15,0	12,8	87
4	F1(75)	12,2	2,0	14,9	6,0	12,8	87
5	F1(100)	12,0	6,0	14,3	16,0	12,9	85
6	F2(100)	12,3	-	14,9	6,0	13,2	80
7	F1(200)	12,3	-	14,3	16,8	12,9	85
8	MNS6	12,3	-	14,9	6,0	14,8	53
9	S4(75)	12,3	-	15,0	5,6	11,8	104
10	S5(75)	12,3	-	14,4	15,0	12,8	87
11	S6(75)	12,3	-	14,2	18,0	11,8	104
12	S3(150)	12,3	-	15,1	3,0	14,5	58
13	BC1	13,8	-	13,0	39,0	12,8	87
14	MNC2	13,8	-	15,2	-	12,8	87
15	MNC4	13,8	-	13,2	35,0	10,7	123
16	C1(100)	13,8	-	15,0	4,8	14,5	58
17	C4(100)	13,8	-	14,7	10,0	14,5	58

Note: for the yeast strains 1 – 12 the must concentration was 210 g/L;
for the yeast strains 13 – 17 the must concentration was 235 g/L;

The experimental data regarding the checking of the fermentative capacity at different temperatures are specified in table 3.

Table 3

Capacity of accomplishing alcoholic fermentation at different temperatures

No	Yeast code	Temperature at which the alcoholic fermentation process takes place					
		13 - 14°C		22°C		35°C	
		Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L
1	MNF1	12,3	-	12,3	-	10,8	26
2	MNF5	12,2	-	12,3	-	9,9	41
3	MNF6	12,1	-	12,3	-	9,7	45
4	F1(75)	11,6	12,7	12,2	2,6	9,2	53
5	F1(100)	10,9	2,4	12,0	6,0	9,7	45
6	F2(100)	12,2	2,0	12,3	-	9,2	53
7	F1(200)	12,3	-	12,4	-	10,2	36
8	MNS6	12,2	2,6	12,3	-	12,2	2,6
9	S4(75)	12,3	-	12,3	-	9,3	51
10	S5(75)	12,3	-	12,3	-	8,6	63
11	S6(75)	12,3	-	12,3	-	12,0	6
12	S3(150)	12,3	-	12,3	-	12,3	-
13	BC1	13,0	11,0	13,2	10	9,2	141
14	MNC2	13,8	-	13,8	-	13,8	-
15	MNC4	13,0	14,0	13,5	5	10,1	63
16	C1(100)	13,7	2,0	13,8	-	13,8	-
17	C4(100)	13,8	-	13,8	-	13,7	2,0

Note: for the yeast strains 1 – 12 the must concentration was 210 g/L;
for the yeast strains 13 – 17 the must concentration was 235 g/L;

At temperatures of 13-14°C, 82 % of the yeast strains tested led to the achievement of dry wines, the rest achieved sweet wines. Nevertheless, at 22°C, all yeast tested led to obtaining dry wines, except for two of them, namely F1(100) and BC1.

Along with the increase in the temperature to 35°C, the data obtained reveal different aspects. Four yeast strains, namely S3(150), MNC2, C1(100) and C4(100) led to the achievement of dry wines, the rest of 64% of the tested yeast led to achieving sweet and semi-sweet wines, except for one yeast, BC1, which was not able to lead the alcoholic fermentation process to 35°C and thus approximately 60% of the sugars of the must remained non-fermented.

The last aspect approached was the testing of the capacity of the 17 yeast strains of accomplishing alcoholic fermentation in different SO₂ concentrations. The data obtained are specified in table 4.

Table 4

Capacity of accomplishing alcoholic fermentation in different concentrations of the sulphur dioxide

No	Yeast code	Sulphur dioxide concentration in the must							
		60 mg/L SO ₂		100 mg/L SO ₂		150 mg/L SO ₂		200 mg/L SO ₂	
		Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L	Alcohol, vol %	Non-fermented sugars, g/L
1	MNF1	12,3	-	12,3	-	12,3	-	12,3	-
2	MNF5	12,3	-	12,3	-	12,3	-	<i>Does not trigger fermentation</i>	
3	MNF6	12,2	2,6	12,3	-	12,4	-	12,3	-
4	F1(75)	12,2	2,6	12,3	-	12,3	-	12,3	-
5	F1(100)	12,2	2,0	12,2	2,0	7,3	84	7,3	84
6	F2(100)	12,2	2,1	12,3	-	12,3	-	12,3	-
7	F1(200)	12,1	-	12,3	-	12,4	-	12,3	-
8	MNS6	12,3	-	12,3	-	12,3	-	12,3	-
9	S4(75)	12,3	-	11,4	16,0	9,0	5,7	7,5	82
10	S5(75)	12,3	-	11,7	11,0	11,1	21	9,8	43
11	S6(75)	12,3	-	12,3	-	12,3	-	12,3	-
12	S3(150)	12,3	-	12,3	-	12,3	-	12,3	-
13	BC1	13,8	-	13,2	10,0	12,0	31	12,3	37
14	MNC2	13,8	-	13,7	-	13,8	-	13,7	-
15	MNC4	13,8	-	13,8	-	11,6	36	10,2	62
16	C1(100)	13,8	-	13,8	-	13,8	-	13,8	-
17	C4(100)	13,8	-	13,8	-	13,8	-	13,8	-

*Note: for the yeast strains 1 – 12 the must concentration was 210 g/L;
for the yeast strains 13 – 17 the must concentration was 235 g/L;*

The analysis of the data reveal the fact that in doses of 60 and 100 mg/L SO₂, the newly isolated yeasts achieve alcoholic fermentations, resulting in dry wines, except for the sources S4(75), S5(75) and BC1.

As for the use of the concentration of 150 mg/L SO₂, the yeasts BC1 and MNC4 resulted in alcoholic fermentation, with remaining sugar, between 21 and 36 g/L. The yeast F1(100) proved to be sensitive to this dose of SO₂, as in the

achieved wine, non-fermented sugars were found of the concentration 86 g/L.

As regards the variant with addition of 200 mg/L SO₂, the behaviour of the tested yeast strains mark out their good resistance to this dose, leading 64.7 % of the fermentations to dry wines and 29.4 % to semi-sweet and sweet wines. The yeast MNF5 did not trigger the alcoholic fermentation in 200 mg/L SO₂.

CONCLUSIONS

All in all, the data obtained as a result of the tests for the identification of the oenological characters of the potentially high performance, 17 yeast strains reveal the following aspects:

- from the point of view of the foaming degree, six yeasts are average foaming, six are minimally foaming (3 – 5 cm³/L foam) and five are non-foaming;

- according to the type of sediment, five yeasts formed powdery and floury deposits and twelve formed compact deposits;

- as for the clearness characteristic, five yeasts completed the alcoholic fermentation process and produced turbid wines, two gave opalescent wines and ten led to clear wines;

- all the yeasts strains tested are alcoholigenic and have the capacity of triggering and ending fermentation at temperatures of 13 – 14°C;

- of the 17 yeast tested, in the process of alcoholic fermentation of grape must treated with doses of SO₂ between 60 and 200 mg/L, the sources BC1, F1(100), MNC4, S4(75) and S5(75) proved to be sensitive. One yeast (MNF5) did not trigger the alcoholic fermentation process of the grape must with 200 mg/L SO₂.

After the evaluation of the oenological characters, nine yeast were selected, being considered capable of performances in the production of high quality white wines, namely three yeasts from the plantation of Fetească albă (MNF6, F1(75), F1(200)), three yeasts from the plantation of Sauvignon blanc (MNS6, S6(75), S3(150)) and three yeasts from the plantation of Chardonnay (MNC2, C1(100), C4(100)).

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