ABSTRACT

"RESEARCH ON THE OPTIMIZATION OF BIOTECHNOLOGIES, THAT APPLY TO THE ELABORATION OF WHITE WINES, SEMI-AROMATIC WINES, AND AROMATIC WINES FROM DRĂGĂŞANI VINEYARD"

Keywords: vine varieties, grapes, glucides, acidity, polyphenols, flavors, production, yeasts, fermentation, maceration, activators, enzymes, chemical composition, organoleptic characteristics

The research on the optimization possibilities of biotechnologies that apply to the elaboration of white wines, semiaromatic wines as well as aromatic wines in Dragasani vineyard, have been developed during wine - growing years 2006 – 2009. There have been used grapes productions belonging to the following vine varieties: Crâmpoşie Selecţionată, Riesling Italian and Fetească Albă – for "neutral" white wines; Fetească Regală and Sauvignon – for white semiaromatic wines; Muscat Ottonel and Tămâioasă românească – for typical aromatic white wines, located, especially, within the large area that can be found between Pesceana stream and Mamu stream.

In order to achieve this purpose, the following four objectives have been taken into consideration:

- Objective no. 1: Defining the technological potential of already mentioned vine varieties taking into consideration the main climatic elements of the four wine-growing years (2006, 2007, 2008, 2009).
- Objective no. 2: Dimensioning the indigenous wine growing microbiologic background as well as its specific structure; obviously, there are different years from a climatic point of view, as well as with a reference to the phenophases of vine varieties maturity.
- Objective no. 3: Experimental identification of the most adequate biotechnological possibilities regarding the elaboration of "neutral" white wines with compositional parameters and organoleptic parameters at the requested level.

- Objective no. 4: Establishing the possibilities of improving the biotechnologies for obtaining high – quality semiaromatic and aromatic wines.

Obtained results

From ancient times, Drăgăşani vineyard has demonstrated an exceptional degree of favorability in obtaining all wine categories, among which there can be mentioned the white wines, semiaromatic and aromatic wines, that have been considered as privileged varieties.

The high degree of favorability for a high - quality and sustainable viticulture within the large Oltenia vineyard is conferred by: Olt and Oltet rivers, that flow on both sides of the large viticulture area, the same way as their numerous affluents; woody mountains from the vicinity of plantations; the chain hills with numerous and dominant eastern, western, south - eastern, and south - western expositions; the mild climate with a low mediterranean influence in the south - western part and with a low continental influence in the north - eastern part; the soils, with domination of eroded pseudorendzinic clay brown variety, followed by the varieties of levigated chernozem varieties, rendzinic regosol, regosol, vertisol etc.

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Generally, the quality parameters of grapes and the parameters of productivity and efficiency, for the same area and for the same wine - growing year, represent essential characteristics of the genetic structure of every soil, their levels being different, in terms of different phenophases, according to the climate conditions of the production years.

Both quality features of grapes and productivity and efficiency elements, belonging to the white wine variety, as well as semiaromatic and aromatic wines, are highly advantageous during the years with a good water supply belonging from previous year, with a normal pluviometric regime, accompanied by a thermal regime and a superior insolation regime, from the active period of the grapevine, as it took place during the year 2006.

In general, for all vine varieties from superior categories, the composition parameters of grapes reach the appropriate levels of the

varieties in question, during the following 10 - 15 days after the complete maturity has taken place.

- ♦ In the case of technological maturity, with a view to the variety and viticulture years:
- The lowest level of glucides has been registered to the Crâmpoşia selecţionată (174 g/l), and the highest level to Muscat Ottonel (234 g/l). Averages at glucides belonging to the four years reveal Muscat Ottonel variety (with 228,5 g/l), followed in a descending order by: Sauvignon (with 225g/l); Tămâioasă Românească (with 222,6 g/l); Fetească albă (with 216 g/l); Fetească Regală (with 213,8 g/l); Italian Riesling (with 211,5 g/l); Crâmpoșie Selecţionată (with 183,8 g/l);
- Concering grapes acidity, the lowest contained level has been noticed in the case of Muscat Ottonel (2,67 g/l, in H_2SO_4) and the highest level in the case of Sauvignon (4,74 g/l). Registered average levels during the study, have situated on the first place of the variety, the Italian Riesling (with 4,58 g/l), closely followed by: Tămâioasă românească (with 4,54 g/l) and Fetească Regală (4,52 g/l). At a more important distance there can be found the varieties Fetească albă (with 4,44 g/l) and Sauvignon (with 4,40 g/l). On the last place, there can be found the Muscat Ottonel (with only 2,85 g/l);
- The tartaric acid, from the content of the grapes acidity is situated, on an average note, between 60,7 % and 65,9 % to all varieties, except Muscat Ottonel in the case of which, this component reaches 78,7 %, meaning that the high decrease of the acidity in this case takes place on the basis of malic acid;
- Both total polyphenols and tannins, at lowered levels as opposed to complete maturity, present significantly higher levels in the case of semi-aromatic and aromatic varieties (between 2,17 g/kg b in the case of Sauvignon and 2,63 g/kg b in the case of Muscat Ottonel and Tămâioasă românească, respectively 2,00 g/kg b in the case of Sauvignon and 2,50 g/kg b in the case of Tămâioasă românească, as opposed to the grapes belonging to white varieties (among 1,43 g/kg b Crâmpoşie Selecţionată and 1,96 g/kg b Fetească Regală, respectively between 1,33 g/kg b Crâmpoşie selecţionată and 1,77 g/kg b Fetească Regală).

♦ The terpenic aromatic fund from the grapes belonging to the Sauvignon semi-aromatic variety and those belonging to aromatic varieties gather in decided proportions until reaching complete maturity. By a comparison with medium contents registered at the moment of the harvest, the proportions to complete maturity are: 90 % - in the case of Sauvignon and 94,6 % - in the case of Tămâioasă românească. In the case of the Muscat Ottonel the situation is opposite: at the time of technological maturity, the flavors represented 96 % as opposed to the level registered during complete maturity.

 \diamond At the time of technological maturity, the total flavors have fulfilled the contents: 1530 µg/kg b – in the case of Sauvignon; 9134 µg/kg b – in the case of Muscat Ottonel; 11820 µg/kg b – in the case of Tămâioasă românească.

In the content of the total aromatic background of grapes, in the case of Sauvignon, the free flavors are dominant (73 %), while in the case of Muscat Ottonel and Tămâioasă românească the forerunner flavors are dominant: 61,5%, respectively 66,1%.

♦ The productive potential presents a considerable variability within the variety, during the entire experimental period of time, and differences according to wine - growing years as well as for one and the same variety.

The average grapes productions place ahead of the variety the Crâmpoşie selecţionată (with 10574 kg/ha), and on the last place, the variety Muscat Ottonel (with 6904 kg/ha). The other varieties produce average productions between 7050 kg/ha (Sauvignon) and 9751 kg/ha (Tămâioasă românească).

On the basis of grapes productions, of their efficiency at primary vinification and of relative contents in glucides, the synthetic technological indicator – the weight of ferementescible glucides from the obtained grapes must, reveal the first place occupied by Tămâioasă românească variety (1521 kg/ha) followed in a descending order by: Crâmpoşie (1385 kg/ha), Fetească Regală, (1276 kg/ha), Italian Riesling (1190 kg/ha); Sauvignon (1097 kg/ha), Muscat Ottonel (1075 kg7ha), Fetească albă (1073 kg/ha).

Within Drăgăşani vineyard, wine - growing area with a multi - centannial background, the microflora specific to the wine - growing, compactly built as a fermenting capacity and structure, has had an essential contribution to the obtaining of high - quality wines, highly appreciated at national and international contests.

Lately, within Drăgăşani vineyard, have frequently appeared, fermentation difficulties, especially in the case of white vinification. Most certainly, "the pressure" of climate parameters, intense chemical treatments, antagonic phenomena and antibiosis are the causes of fermentation problems.

- ♦ The investigations over wine growing microbiological background, during two different years from a climatic point of view, have evidenced the following features:
- For the same variety type and the same phenophase of grapes ripenning, the number of microorganisms of grains and the specific structure of populations differs according to climate conditions of wine growing years;
- Under the same climatic conditions of the wine growing year, the indigenous microbiologic background, in the case of the same phenophase of ripenning differs according to the variety;
- For the same phenophase of grape ripenning, during the best winegrowing year (2006), the number of wine – growing microorganisms, in the case of all varieties, is considerably higher, as opposed to the situation from a highly warm and relatively droughty season (2007);
- Both in the first phase of grape ripenning, as well as at the time of their technological maturity, keeping the differences in the case of populations dimensions, determined by the climatic conditions of the considered years, with the lowest levels the variety called Crâmpoşie selecţionată is included, and the highest values have been registered in the case of Muscat Ottonel, followed by Tămâioasă românească and, at a larger distance by Sauvignon. Thus, it can be noticed the direct relation between the gathering of glucides and the growth of indigenous microbiological populations;

- During extremely favourable wine growing years from a climatic point of view, within the variety, the yeasts represent proportions situated between 12,8 % and 18,6 %. During the last favorable years, the yeasts are comprised between 16,6 % and 23,6 %, registering growths between 3,8 % and 5,0 %, corresponding the diminish of yeasts populations;
- During a very good wine growing year, the sporogenous yeasts represent proportions comprised between 78,8 % and 87,4 %, and the asporogenous ones between 12,6 % and 21,2 %. In the same conditions, the acetic bacteria proportions are comprised between 23,5 % and 27,9 %, and the lactic ones between 72,1 % and 76,5 %;
- During a highly warm and relatively droughty season, the sporogenous yeasts register decreases with limits between 73,2 % and 84,1 %. Under the same conditions, the populations of lactic bacteria rise, with approximately 3 4%;
- On the sporogenous yeasts background, the two species Saccharomyces ellipsoideus and Saccharomyces oviformis occupy 84,6% during the favorable years and 82,8 % during climatic difficult years;
- In the case of asporogenous yeasts background, the species Candida vini and Kloeckera apiculata, represent all together 59,5 % during good wine growing years and 63,4 % during excessive warm years.

Therefore, during extremely warm and droughty years, as well as during extremely rainy years, the spontaneous fermentations may be uncertain, thus, the use of selected yeasts is necessary.

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 \diamond Once the wine decantantion and the clearance of wine take place, no matter which is the applied technological process, the contents in glucides and tannin are unsignificantly diminished. When the low temperature operates, it is diminished with approximately 4,2 %, and total nitrogen contents are drastically diminished (with 27 – 30 %) during singular bentonite interventions or in association with SO₂.

The highest clearance degree of the grapes must is obtained when applying pectolitic enzymes and the bentonite in association with SO_2 . The bentonite determines also a massive release of microorganisms, reaching 91 %.

♦ In the case of cleared and decantated musts, yeasts popultions – both the indigenous as well as the selected ones evolve differently according to the technological process that has been applied. The highest rate of multiplication is registered in the case of enzymes cleared approach and slower when using the betonite.

According to the evolution of yeasts population, the matabolization of glucides also takes place, the fermentation periods being longer when the grape musts have been cleared with betonite together with SO_2 .

- \diamond For the two types of yeasts (indigenous yeasts and selected yeasts) with an activity in musts that present the same origin, as the fermentation temperature becomes higher, there are different processes that take place : diminished fermentation periods, growths of volatile acidity contents, acetic aldehyde, and glycerol, diminished residual sugar. For the same stage of the fermentation temperature, the content parameters present a degree of convenience in favor of the induced yeasts (with selected yeasts). When analyzed from an organoleptic point of view, the temperatures of $19-20^{\circ}$ C are highlightened, by the obtained notes, as presenting something more when applying the selected yeasts.
- \diamond Both spontaneous fermentations, as well as induced fermentations in the presence of a fermentation activator end up with the growth of alcohol and glycerol contents as well as the diminished contents in volatile acidity, of the fermentation efficiency, of the residual sugar, as well as of the fermentation periods of time, all in favor of the selected yeast. As a result of acetaldehyde lower contents, the contents of free SO₂, for the same total dose, are high.
- ♦ Fermentation activators as well as added oxygen in nonproliferated stages of yeasts stimulate the fermentation process and in highly rich glucides musts, in the case of activities of selected yeasts, the alcoholic degree reaches the value of 14 %vol.
- ♦ Pellicular maceration represents a possibility of improving the organoleptic composition and quality of Drăgăşani white wines, in the case of grape harvests with a perfect phytosanitary state:
- When obtaining high quality white wines (Italian Riesling, Fetească albă), the compositional and organoleptic effects of pellicular maceration are

produced at temperatures comprised between $14 - 15^{\circ}$ C and $19 - 20^{\circ}$ C and contact durations between must phases between 18 - 24 hours;

- By applying the pellicular maceration, in the mentioned conditions, the additions as oppposed to classic wine-growing, are: with 15 20 % at total polyphenols; with 14 21 % at tannin; with approximately 5% at non-reducing extract; with 9 10 % at ash. As a result of the organoleptic examination, the varied aromatic profile stands up, the marks received being higher with approximately. 3 %;
- By applying enzymes to the must, for the same durations and temperatures of pelicular maceration, there has been noticed a growth of the efficiency in must, extract, ash, polyphenols, and in the case of organoleptic appreciation. The residual sugar presents diminished values.
- \diamond Current market wines from Crâmpoşie selecţionată can be organoleptically and compositionally improved by applying pelicular maceration (at a temperature of $18-19^{\circ}$ C and contact duration between phases within 18-24 hours) or of the pelicular criomaceration (at $6-8^{\circ}$ C and durations comprised between 18-24 hours). Under these circumstances, there have been observed additions to: the yield in must, non-reducing extract, ash, tannin, and at the organoleptic appreciation.
- ♦ Pectolitic enzymes addition in the must brings technological organoleptic, and compositional improvements.
- \diamond Cryoextraction, by cooling grapes at $-6 8^{\circ}$ C would be another option of concentrating glucides, acidity and tannin in the case of Crâmpoşie, but with relevant diminishings in the case of the efficiency and the must volume resulted from the processing of frozen grapes. The procedure is not convenient from an economic point of view.

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When obtaining aromatic and semi-aromatic wines, for a correct interpretation of maceration effects regarding the levels of the terpenic flavors and those of the polyphenols from finite products, expressed in litres, must be take into account the fact that these constituents are extracted from existing backgrounds in the grapes quantities situated between 1445 g (in the case of Tămâioasă Românească) and 1486 g (in the case of Sauvignon).

- When obtaining Sauvignon wines:
- For equal durations of maceration, the contents in extract, ash, total nitrogen, and tannin continuously grow as the maceration temperature grows, the sulfation doses and pectolitic enzymes doses grow as well. The convenient results are obtained at temperatures of $19 20^{\circ}$ C, sulfitation doses of 50 75 mg/l and enzymation 2,0 g/hl. The same levels have been proved to be efficient in the case of terpenic aromas extraction as well;
- In equal conditions regarding the maceration temperature, the sulfation dose and the enzymes dose, the contact durations between the must phases comprised between 18 and 24 hours, when obtaining Sauvignon wines, have proved to be very favourable from an organoleptic and compositional point of view;
- In the case of Sauvignon grapes vinification, there will be avoided the conjugated action of maceration temperature of over 20° C, the dose of SO_2 over 75 mg/l and the maceration duration of over 24 hours, in order to limit the extraction of nitrogen and tannin in high proportions;
- The fermentation parameters of selected specialized yeasts, in the case of Sauvignon musts obtained after a 24 hours maceration, are highly superior to those obtained in the same conditions from indigenous yeasts;
- The pectolitic enzymes influence the extraction of polyphenolic compounds, both in the case of spontaneous yeast as well as in the case of induced one, when the Sauvignon grapes vinification takes place. For the maceration duration of 24 hours the growths are: with 78,5 % higher in the case of indigenous yeasts and with 90,9 % higher in the case of selected yeasts, for total polyphenols.
 - When obtaining Muscat Ottonel wines:
- Within the same biotechnological phase, the contents in alcohol, glycerol, non reducing extract, ash grow as the maceration period becomes longer. In the sense of the growth of the contact duration between phases, the fermentation ratio, the residual sugar, and the fermentation durations evolve in a descending manner;
- Under all fermentation and composition aspects, the use of singular selected yeasts or in association with pectolytic enzymes presents the best results, even in the situations in which glucides contents are very high: the

alcohol contents reach the level over 13,5 % vol, the glycerol over 10 g/l, the extract just below 24 g/l, residual sugar under 10 g/l, in the conditions of fermentation proportion lower than 17;

- From the point of view of total terpenic aromatic fund of wines, favorable results are obtained both at the level of spontaneous yeast as well as in the case of selected yeast, but in both cases in association with pectolitic enzymes, without maceration periods that exceed 30 hours from the spontanoues fermentation, and 24 hours from the induced fermentation;
- But, for the same maceration duration, the level of total aromatic fund and of the flavors connected in forerunners are obviously superior in the case of the fermentation with selected yeasts + pectolytic enzymes, as opposed to the fermentation with indigenous yeasts + enzymes.
 - ♦ When obtaining Tămâioasă românească wines:
- For the same raw material, the organoleptic and compositional results are different according to the yeast type, as a singular factor or in association with additional components;
- For the same yeast type (indigenous and selected yeasts) as a singular factor and in association, the results differ in accordance to the maceration duration;
- When using singular induced yeast, or in association, the advantages refer to the diminish of: the fermentation ratio, of volatile acidity, of maceration duration, of residual sugar and to growths of contents in: alcohol, glycerol, extract, polyphenols, ash, and total nitrogen, terpenic aromas;
- When adopting the procedure that comprises : selected yeasts $+SO_2+$ pectolitic enzymes, the maceration duration will not exceed 36 hours form the elaboration of Tămâioasă românească wine.