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RESULTS OBTAINED IN FICUS ELASTICA CONCERNING ROOTING ON DIFFERENT SUBSTRATA AND DIFFERENT ROOTING ENHANCERS WITH EFFECT ON PLANT ROOTING IN AUTUMN

Anches Codrut Calin[1], Bala Maria[2]

Key words: *ficus, substratum, enhancer, rooting, variance analysis*

ABSTRACT

Ficus is an ornamental plant that roots with difficulty (Militiu A. 1962); this is why we had the idea of using different substrata and different enhancers in a greenhouse with proper climate factors.

Data concerning ficus rooting show that greenhouse climate conditions in Timisoara are favourable to the rooting of the studied plants.

As a result of trials, we could see that bio enhancers and substrata have different effects on rooting and on other aspects. The first plant roots appeared 30 days after planting on the rooting substratum and the rooting period from the first to the last roots was about 35 days.

INTRODUCTION

Ficus is a woody tree-like or creeping ornamental plant that adapts to different conditions (Kiselev G. 1956). All ficus cultivars are elegant and some of them are grown inside for the beauty of their decorative leaves. Their leaves are appreciated for their shape, size and colour. The entire plant contains latex (Bala M., 2012).

The stem is elastic, easy to model, and very branchy. The leaves are long and spear-like, coloured in different shades from dark green to green-yellowish, and shiny. Some ficus cultivars have variegated leaves (Anton D. 2007).

So far, there has been no research on ficus; therefore, the data presented in this paper are the only ones in literature.

In order to get scientific results for the improvement of the cultivation technology of the species ficus in greenhouses, our researches aimed at several goals: determining the effect of the rooting bio enhancers on the plants, obtaining vigorous, healthy plants, obtaining a very high percentage of rooted plants, and observing the behaviour of the three *Ficus* species rooting on different substrata.

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Substratum as main environmental factor, greenhouse climate conditions, and well-applied cultivation technology impact quantity and quality of rooted plants (Lazureanu A 1994).

MATERIAL AND METHODS

Trials were carried out at the Didactic and Research Basis of the Faculty of Horticulture and Forestry, in a greenhouse measuring 1000 m^2 equipped with environmental factors' automatic control systems and equipment.

The trial has a polyfactorial character: the variants were set after the randomised block method with 3 replicas (Ciulca S 2006) specific to trials in forced, protected flower cultivation areas.

The method of interpretation of the results was variance analysis (Ciulca S 2002).

In the trial, we used 3 *Ficus* species from which we harvested cuttings – *Ficus* plants brought from the Mediterranean locality Pistoia (Italy). The 3 *Ficus* species in the trial were *Ficus elastica*, *Ficus Benjamina*, *Ficus Australis*. Cuttings are parts or fragments of plant that are planted to root. The organs sued to get cuttings are shoots, stems, leaves, buds, roots (Cantor M 2009). The rooting substratum we used was sand, perlite, perlite + peat, perlite + peat + sand. These substrata were disinfected before use to remove potential diseases and pests.

The enhancers used to root *Ficus* plants were: atonik, radistim and revital.

RESULTS AND DISCUSSIONS

Substratum had a considerably higher effect (45.54%) on root growing variability compared to the effect of the growth enhancers (14.25%). The combined effect of substratum and of rooting enhancers also had a distinctly significant influence of 33.91% on the variability of this feature.

Table 1

Analysis of variance for the effect of rooting substrate and stimulator on cuttings rooting in *Ficus elastica*

Source of variation	SS	DF	MS	F Test
Total	10511.64			
Replicas	46.82	2	23.41	F=1.14
Substratum	4787.23	3	1595.74	F= 77.83**
Enhancer	1498.37	3	499.46	F= 24.36**
Substratum x Enhancer	3564.16	9	396.02	F= 19.32**
Error	615.06	30	20.50	

Taking into account the unilateral effect of the substratum, the cutting rooting percentage (Table 2) had a span of 25.10% with values ranging between 33.10% when using sand and 58.20% when using perlite, on the background of high variability (24.71%) among results on the four substrata. Using simple perlite was the most efficient substratum

if we take into account that it allowed significant increases of cuttings' rooting (4.17-25.10).

The average values of the cuttings' rooting percentage with different enhancers showed a span of 14.35% ranging within 41.25% in the non-treated variant and 55.60% when treated with Atonik, on the background of medium variability (13.82%) between enhancers.

Table 2

Rooting substratum effect on cuttings rooting in *Ficus elastica*

Rooting substratum	Averages (%)		Relative values (%)	Difference/ Significance
Perlite - Sand	58.20	33.10	175.83	25.10***
(Perlite+Peat) - Sand	54.03	33.10	163.23	20.93***
(Perlite+Peat+Sand) - Sand	41.39	33.10	125.05	8.29***
(Perlite+Peat) - Perlite	54.03	58.20	92.84	-4.17^{0}
(Perlite+Peat+Sand) - Perlite	41.39	58.20	71.12	-16.81^{000}
(Perlite+Peat+Sand) - (Perlite+Peat)	41.39	54.03	76.61	-12.64^{000}

$DL_{5\%}$=3.77 $DL_{1\%}$=5.08 $DL_{0.1\%}$=6.74 $LSD_{5\%}$

Table 3

Rooting enhancers effect on cuttings rooting in *Ficus elastica*

Rooting enhancer	Averages (%)		Relative values (%)	Difference/ Significance
Radistim - Not-treated	47.13	41.25	114.25	5.88**
Revital - Not-treated	42.73	41.25	103.59	1.48
Atonik - Not-treated	55.60	41.25	134.79	14.35***
Revital - Radistim	42.73	47.13	90.66	-4.40^{0}
Atonik - Radistim	55.60	47.13	117.97	8.47***
Atonik - Revital	55.60	42.73	130.12	12.87***

$DL_{5\%}$=3.77 $DL_{1\%}$=5.08 $DL_{0.1\%}$=6.74

We consider significant the differences between the combinations marked as a, b, c – for vertical comparisons and as x, y, z – for horizontal comparisons.

As for the combined effect of substratum and enhancers on cuttings' rooting in *Ficus elastic*, we can see that, on the substratum with sand, this character had a variation span (5.18%) and a low variation (8.48%), associated with insignificant differences between different enhancers. As such, on this substratum, the enhancers used had no significant impact on rooting in Ficus cuttings' rooting.

The rooting percentage of the cuttings on perlite had a very high variability (35.74%) for a span of 48.89%, from 41.67% when treated with Revital to 90.56% when treated with Atonik. On this substratum, growth enhancers had the highest impact

on cuttings' rooting, with differences ensured statistically. Thus, treating with Atonik resulted in a significant increase of the rooting percentage of the cuttings compared to the other variants with increased above 36.00%. Treating with Radistim also had a significant effect on cuttings' rooting.

Table 4

Effect of rooting substratum and enhancer on cuttings rooting in *Ficus elastica*

Enhancer	Substratum					
	Sand	Perlite	Perlite +Peat	Perlite+ Peat+Sand	$\bar{x} \pm s_{\bar{x}}$	S%
Not-treated	y30.00a	x46.11c	x43.33b	x45.56a	41.25±2.16	18.12
Radistim	z33.52a	x54.44b	x57.78a	y42.78ab	47.13±3.33	24.46
Revital	z33.70a	y41.67c	x58.33a	yz37.22b	42.73±2.99	24.27
Atonik	z35.18a	x90.56a	y56.67a	z40.00ab	55.60±6.60	41.13
$\bar{x} \pm s_{\bar{x}}$	33.10±0.81	58.20±6.00	54.03±2.23	41.39±1.29	46.68±2.16	
S%	8.48	35.74	14.32	10.84	32.04	

$DL_{5\%}$=7.55 $DL_{1\%}$=10.17 $DL_{0.1\%}$=13.48

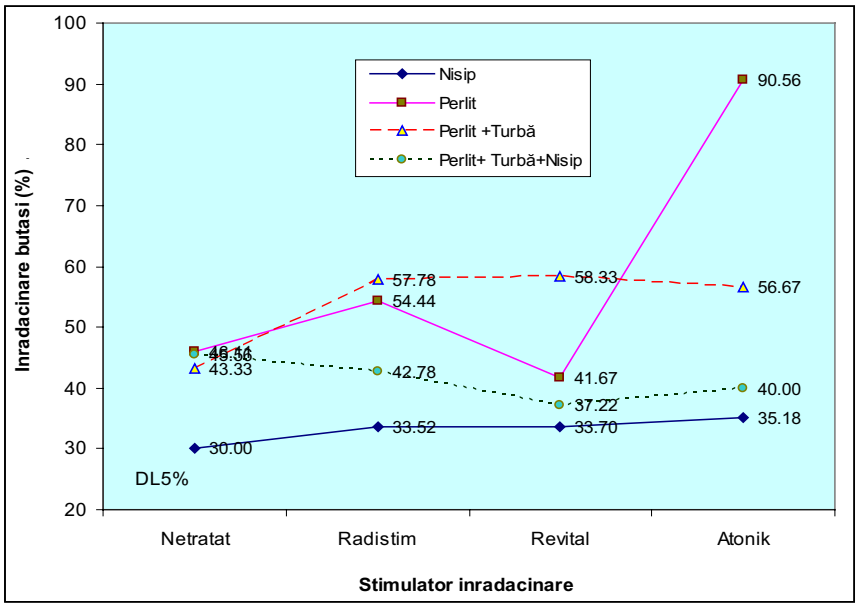

Figure 1. Cuttings' rooting in *Ficus elastica* under the impact of different substrata and enhancers

On a perlite + peat substratum, the cuttings had a span of the rooting percentage of 15.00% associated with an intermediary variability compared to the other two substrata (14.32%), with limits between 43.33% for the not-treated variant and 58.33%

for the variant treated with Revital. Thus, on this substratum, treatments with enhancers determined a significant increase of the cuttings' rooting percentage without statistically ensured differences between the enhancers.

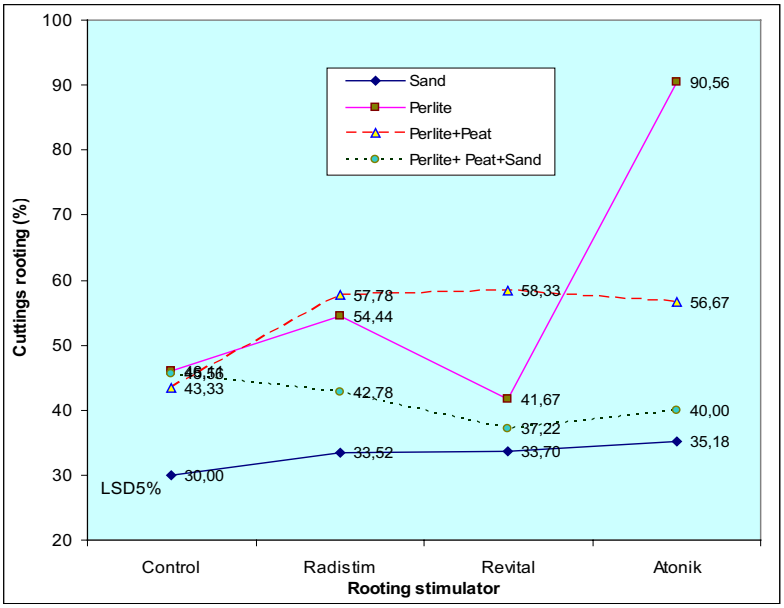

Figure 2. Rooting of *Ficus elastica* cuttings under the influence of different substrates and stimulators

CONCLUSIONS

Based on the variance analysis in Table 1, we can see that substratum and rooting enhancers had a real impact strongly ensured statistically on *Ficus elastica* cuttings' rooting on the ground of homogeneity of environmental conditions at the level of the trial;

Using perlite + peat allowed a rooting level of the cuttings significantly higher than sand or perlite + peat + sand;

On sand, *Ficus* cuttings had a significantly inferior rooting level compared to other substrata studied in the trial;

The enhancer Atonik had a very significant effect on cuttings' rooting compared to the other treatments, with increases between 8.47% compared to Radistim and 14.35% when not treated;

Using Radistim determined a significant increase of cuttings' rooting compared to the variant not-treated or to the variant treated with Revital;

The treatment with Revital had no significant impact on the rooting of the cuttings in *Ficus*. As for the effect of different enhancers on cuttings' rooting on the four substrata, there are significant real differences between the substrata on the ground of the three enhancers. Thus, treating with Radistim allowed the best results on perlite, and on perlite + peat, while sand substratum allowed a significantly lower efficiency.

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STUDIES REGARDING THE SENSORYAL ANALYSES OF YOUNG WINES OBTAINED FROM CHARDONNAY CLONES IN SAMBURESTI VINEYARD

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Key words: *Chardonnay, clone, wines, aroma compounds*

ABSTRACT

The different sensorial appreciation of the wines was burdened by their high quality of alcohol, that emphasized even at the olfactory exam, the alcohol smell being highlighted and contributing up to a great extent to the increase of the smell intensity but at a slight diminishing of its character because it has become a main component of the flavor and, moreover, diminished the flavor fineness so that one was not able to make a real appreciation of the notion of area pattern.

INTRODUCTION

The sensory perception of wine is complex and involves the interaction of both volatile and non-volatile components (Robinson A.L. et.al., 2011). The aroma profile of wine is one of the most important factors in wine evaluation process since it influences both orthonasal and retronasal perception (Petka J. and Farkas P., 2001a). Aromatic compounds from grapes can appear later in must and wines as volatile free forms or as odorless, non-volatile glycosides (Gunata et. al., 1985). As only free forms are volatile and detected by smell sense (Williams et al., 1981, Tambora et al., 2004, Rocha et al., 2005, Sanchez Palomo et al., 2005).

The highest amount of volatile aroma compounds is formed during the alcoholic fermentation of the must of wine. The organoleptically active products of both alcoholic and malolactic fermentation belong to the secondary volatile aroma compounds of wine. By these processes, alcohols, acids, esters, carbonyl compounds, sulphur compounds, nitrogen compounds, lactones and volatile phenols are formed. A detailed assessment of the influence of both processes as well as specific compounds on wine aroma is presented (Petka J. and Farkas P., 2001b).

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According to the quantities of free and glycoside forms of varietal aromatic compounds, grape and wines are classified as neutral or aromatic (Cabrita et al., 2006, Bavčar D. et al., 2011).

MATERIAL AND METHODE

The present paper has been elaborated having as a root the physical, chemical and organoleptical studies of the wines obtained from six clones of Chardonnay, harvested between 2010 and 2011 from Samburesti vineyard, where this wine has recently been introduced in the range. The wines were elaborated in conditions of micro wine-making in the Oenology lab from the Horticulture Faculty of Craiova, in the same location having also been done all the studies regarding the main composition and organoleptical parameters. The six Chardonnay clones that have been studied are: 95/K5BB; 121/K5BB; 548/K5BB; $95/SO_4$; $76/SO_4$ and $548/SO_4$.

The wines belonging to the 2010 harvest have been tasted for several times, but each time was due to the occasion of some technological operations that had as a purpose the assurance of purification and stabilization. The first process of tasting took place in a period of 30 days after the wine production when the first decanting was carried out. The following tasting took place in the beginning of November, when the second decanting was carried out. The third tasting took place in the beginning of December when the wines were almost three months old and the fourth tasting took place in March 2011, so at six months after wine production when the chemical composition analyses were carried out. The wines from the 2011 harvest were tasted at two and six months respectively, after wine production.

RESULTS AND DISCUSSIONS

For the 2010 wines, the study of the tasting results indicates that for all the six wines coming from the studied Chardonnay clones, a likely resemblance was recorded, as they have all been better appreciated until the third tasting carried out at almost three months. In exchange, at the six month tasting all the six wines received lower scores than at the three month tasting. As a result, in the period of time between three and six months, none of the wines improved from the organoleptical perspective but on the contrary all of them were underestimated and evaluated although they were studied by the same tasters and under identical tasting conditions. Therefore, at the first tasting carried out at one month after wine production, the highest praises were targeted towards the wines obtained from clones 548/K5BB which got a score of 80 points and $95/SO_4$ which received 78 points followed at a tiny difference by clone $76/SO_4$ with 77 points and $548/SO_4$ with 75 points. The least appreciated were the wines from clones 121/K5BB (70 points) and especially 95/K5BB which received only 65 points. These two last wines were unappreciated taking into account the fact that the fermentation was either unfinished or barely finished, having also pungent smells of CO_2 and yeast lacking balance and harmony. On the other hand, clones 548/K5BB and $95/SO_4$ were highly appreciated not only for their yellow-greenish color and clarity but also for their intense and pleasant smells. At the first wine, though, the flavor was dominantly vegetal with a firm grassy feature while at the second wine one could have easily noticed an obvious plus of fermentative secondary flavor with prevailing features of warm bread. At all the wines, the presence of the carbonic gas not fully emitted conferred a plus of intensity but at the same time diminished the olfactory pleasure and

from the gustative point of view, the two mentioned clones conferred a plus of volume and pleasure while at the others, it was the gustative equilibrium that spoiled them.

At the second tasting, when the wines were two months old, one can notice that all the six wines have been much better appreciated achieving significantly better scores, which seems to be natural thanks to a favorable evolution which has taken place in the period of time from the previous tasting.

Just like in the previous tasting, the wine which achieved the best appreciation, obtaining the highest score was the one from clone 548/K5BB with 88 points. The tasters noticed at this wine their intense yellow-greenish color, which was also very pleasant and characteristic to a fresh wine with a very good purity. From the olfactory perspective, the wine has remarked through a very pleasant flavor with dominant fruity features (a pleasant smell of sweet apples and ripe pears) and also floral features (the dominant ones being of wax cherry and freesia). When tasting it, the wine proved to be dry, regularly flowing, pleasant with an acceptable acidity and easily sweet.

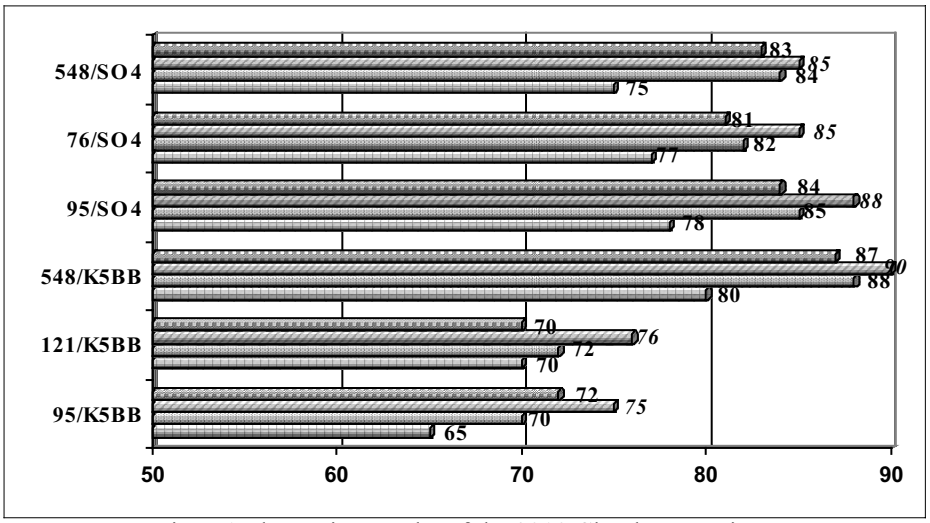

Figure1- the tasting results of the 2010 Chardonnay wines

On the second position, there placed the wine obtained from clone 95/SO_4 just like in the previous tasting, but this time with a score of 85 points (unlike 77 points in the previous tasting). At the visual exam, the wine was highly appreciated for its very good purity and also for its very attractive aspect of color with tents of yellow-greenish characteristic for a very early age. At the olfactory exam, it remarked through both its fruity flavor and the secondary fermentative one with an intense and pleasant smell of baked bread. At the tasting exam, the wine was appreciated as being dry, consistently pleasant, with a high gustative intensity but with a slight lack of acidity, partially compensated by an easy presence of carbonic gas, which had not totally emitted.

On the third position, it was the wine obtained from clone 548/SO_4 with 84 points, the wine that presented the highest increase in score in comparison to the previous tasting(9 points), this wine making a shift of positions with the one coming from clone 76/SO_4 which increased with only 5 points reaching 82.

Both wines did very well at the visual exam with a very good purity and a pleasant, beautiful and clear yellow-greenish color. At the olfactory exam, both wines

displayed intense smells but while the first one displayed features that were dominantly floral (wax cherry) and alimentary (butter, bananas), the second one displayed burnt, fragrant features like those of the matches. When it comes to taste, both wines suffered modifications because of the slight acidity, the most pronounced disequilibrium being of the wine coming from clone 76/SO_4.The slightest scores were obtained, just like in the first tasting, by the wines from clones 121/K5BB(76 points) and especially 95/K5BB. The first one was slightly appreciated because of its flavor which lacked delicacy but which contained grassy and rough features while the second one because of a too powerful smell of yeast, given by the prolonged fermentation(more than 30 days), which affected its taste as well. At the third tasting that took place when the wines were almost three months old, all the six wines displayed a maximum of organoleptical qualities achieving the highest scores.

On the first position, it was maintained the clone 548/K5BB with 90 points, although the score increase was lower in comparison to the other clones unlike the previous tasting. The wine detached itself especially by means of its visual and olfactory aspect, through its intense and fruity flavor of green apples with dominant and strong alimentary features- bread- but of a medium towards low intensity. When it comes to its taste, it was appreciated as being dry, extractive and pleasant. On the second position it was maintained the wine from clone 95/SO_4, with 88 points, with three points more than at the previous tasting. The wine has displayed a yellow-citron pleasant color, with a very pleasant flavor, with dominant alimentary features-bread- but of a medium towards low intensity. When tasting, it was appreciated as being dry, extractive and pleasant.

The positions three and four were occupied just like in the previous tasting by clones 548/SO4 and 76/SO4, this time with an equal score (85 points). Both wines were highly appreciated for their smell, with floral features (clone 548/SO_4) or redundant (clone 76/SO_4).

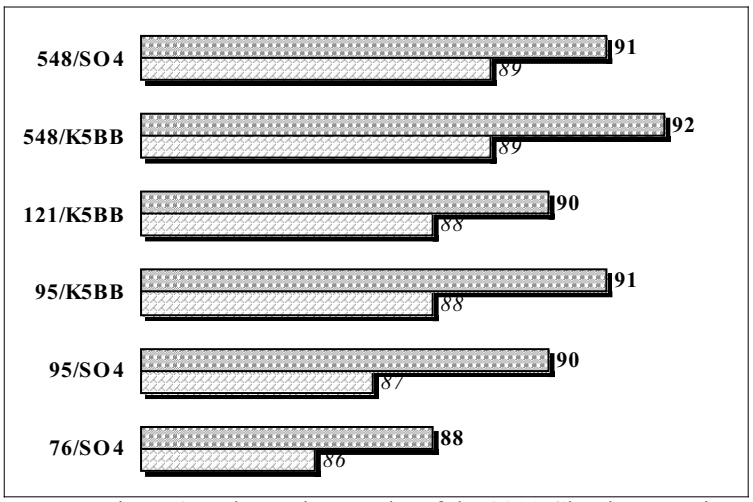

Figure 2 – The tasting results of the 2011 Chardonnay wines

On the last two positions, there were maintained clones 121/K5BB (with 82 points) and 95/K5BB (75 points). The last one was the only of the six wines that did not reach 80 points at none of the tasting processes , while clone 121/K5BB although on the

fifth position, proved to be one of the highest score increase so, the most obvious increase from the previous tasting(from 76 to 82 points). This wine was unappreciated in comparison to the other four because of its rough flavor, with grassy, weedy features and also because of its tough, bitter smell, all of these on a background of acidity lacking. The wine from clone 95/K5BB was one more the most unappreciated of all the wines because of its weak, ordinary flavor, lacking qualities and fineness and also because of its acid, burning taste and features of yeast.

At the last tasting, which took place at six months old, all the wines lost something of their sensorial qualities in opposition to the previous tasting from three months old.

On the first position, just like in the previous tasting processes, it was maintained the wine from clone 548/K5BB with 87 points lowering with three points. Unlike the previous tasting, the wine has maintained its great purity but the color has lost something from the greenish features, directing towards straw-colored with a golden feature. The flavor has also diminished in intensity and the characteristic smells of fermentative secondary flavor have completely disappeared together with the floral and citric smells. The taste was appreciated as being dry, slightly acid but yet pleasant on the background of a greater tannin content, resulted from the pre-fermentative pellicular maceration. Nevertheless, it has lost a lot of its fineness and especially freshness. On the second position, it was maintained the wine from clone $95/SO_4$ with 84 points, lowering from 88 points. Unlike the previous tasting, the wine has maintained almost intact the visual characteristics but lowered from the aromatic point of view, totally lacking the very pleasant fermentative flavor of baked bread, offering a flavor of medium towards low intensity, with fruity and slightly floral features. The taste keeps on being pleasant, flowing even though acidly weak. On the third position, it was maintained the clone $548/SO_4$ with 83 points, lowering with only two points compared to the previous tasting. As a matter of fact, this was the wine that lowered the least compared to the three month tasting. Just like the previous ones, this wine too has almost lost all the floral features of the flavor and at tasting, the lack of acidity is somehow compensated by a certain thickness and a slightly grassy feature achieved through pre-fermentative maceration. On the 4^{th} position, it was maintained clone$76/SO_4$ with 81 points, lowering with 4 points from the tasting that took place three month ago. Throughout this period of time, the wine evolved to straw-colored, the smell achieved stale features, lacking almost completely the flavor and the smell is dry and slightly tougher, unspecific for a refined white wine. On the 5^{th} position, it was maintained the wine from clone 121/K5BB with 80 points, lowering with two points than three months ago, so a slight lowering. The wine has maintained its visual characteristics while the olfactory ones received a feature of redundant, burnt but less vegetal feature. The taste is slightly acid and lacks freshness and fineness. On the last position, it was maintained clone95/K5BB with 72 points. It proved to be mediocre to all the characteristics and it was described to be tough, lacking pleasure, freshness and fineness.

For the 2011 wines, at the first tasting achieved when the wines were two months old, the scores were between 88 and 92 points while at the second tasting achieved when the wines were six months old, the scores were between 86 and 89 points, being thus kept the wine hierarchy. Therefore, at the first tasting, the highest score was the one obtained by clone 548/K5BB (92 points). This one was followed by clone 548/SO4 and 95/K5BB, each one with 91 points. After four months, both wines from clone 548 had the same score (89 points) while clone 95/K5BB had 88 points just like 121/K5BBwhich at the first tasting had 90 points. At both tasting processes, the lowest scores were obtained by clones $95/SO_4$ (90 and 87 points, respectively) and $76/SO_4$ (88 and 86 points, respectively).

On the whole, it is highly important to mention that this is about very small differences which are not highly illustrative. Yet, what made a difference between the varieties of both tasting processes was the manner in which there were perceived the gustative balance of the wines and also the balance between alcohol and acidity. At the second tasting, the main factor that brought its contribution to the scores lowering was the intensity and quality of the flavor, especially the diminishing of fruitfulness and freshness of wines.

Considering the fact that the wines were obtained following schemes of identical wine production, fermented with the same yeasts, they have been very similar from the olfactory perspective, especially in what concerns the secondary flavor of fermentative type, a thing that highly damaged the mission of the tasters to make a real classification of the wines from their sensorial profile point of view.

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RESEARCHES REGARDING THE CHEMICAL AND POLYPHENOLIC COMPOSITION OF THE MERLOT CLONES WINES OBTAINED IN SAMBURESTI VINEYARD IN 2011 YEAR

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Key words: *chemical composition, polyphenolic compounds, Merlot clones wines*

ABSTRACT

In this study the main parameters of chemical and polyphenolic composition of the young red wines obtained in Samburesti vineyard from different Merlot clones were investigated. All the clones can produced high quality red wines, specially the clone 181, which have the best values of analytical parameters and good chromatic structure.

INTRODUCTION

Samburesti vineyard is recognized for this red wines obtained from famous cultivars widely in the world: Merlot (Baduca Campeanu C., 2008). The quality of Samburesti red wines is due to the environmental factors such as climatic conditions and the type of the soils (Popa A., 2008). Environmental variables are considered the most influential factors on grapevine production and berry composition (Montes C. et.al., 2012).

Worldwide, a large number of studies examining the climatic features have provided the description of different *terroirs* and the identification of winemaking regions using different methodologies (Montes C. et.al., 2012). Temperature is widely accepted as being the primary climatic factor affecting the quality of viticultural production (Gladstones 2004). As a consequence, increases in temperature due to an enhanced greenhouse effect will likely have a significant effect on viticultural production. Possible beneficial aspects of climate change include less bud and crop damage from frost events and less extreme winter minimum temperatures that would otherwise damage grapevines (Jones 2005).

The length of the growing season is considered an important determinant of grape quality and consequent wine value (Webb et al., 2007). Therefore, the time at which ripening takes place, whether it be in the heat of midsummer or in cooler autumn months, can determine potential wine quality for a particular vintage. (Hall A.and Jones G.V., 2010).

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There were strong correlations between sugar content, colour and quality perception in grapes and the resulting wines. The best Cabernet-Sauvignon wines were made from grapes rated highly for colour intensity, red berry and black berry with spice aroma. Seasonal differences resulted in larger variance in grape composition than grapes originating from vineyards in different climatic zones. This highlights the difficulties in pinpointing a specific parameter to indicate optimal maturity (Oberholster A. et al., 2010).

MATERIAL AND METHODS

The present paper was achieved as a result of a study on the wines produced in 2011, in conditions of micro wine production, in the Oenology Laboratory belonging to the Faculty of Agriculture and Horticulture, starting from three clones (181, 337 and 343) recently introduced in the range of a traditional type of wine from Samburesti vineyard – more precisely, Merlot.

The first of these clones had been grafted on two stocks (1103 and SO_4), clone 337 is grafted on stock 1103 P and clone 343 is grafted on stock SO_4. The only difference between the variants is given by the single and unique potential of each clone-stock combination because any other factors of variability had been eliminated and that is due to the fact that all the grapes from which wines were obtained had been harvested on the same day and had been produced under identical conditions, following the same wine production scheme. The sulphitation method was identical, the sedimentation being achieved at the same dates and so were the physical-chemical and organoleptical analyses (Gheorghiţă M. et. al., 2006).

The study of the chemical composition was achieved in March 2012 and targeted the main compositional parameters of the wines, being obtained in the Oenology Lab, following official methods of studying wines. (Muntean Camelia et. al., 2001).

The physical studies were obtained by means of spectro-photometric methods by the specialized staff from the Laboratory of Physical and Chemical Studies of Horticulture Products, where there had been determined the index of total polyphenols, the anthocyan content and the chromatic structure(intensity and color shade), the determinations being achieved when the wines were three months old, following the methods described by Ribéreau-Gayon P. et al. (2005). Unlike the compositional studies, the spectro-photometrical ones targeted a greater number of wines, because on the main wines there had been done other combinations between the wines from a clone but grafted on different stocks or between wines coming from different clones but grafted on the same stocks.

RESULTS AND DISCUSSIONS

The results of the composition studies belonging to the four varieties of Merlot wines, displayed in Table 1 show that all of them come from fully developed and ripe grapes having a high degree of maturation, as they offer very high contents of alcohol and glycerol. Thus, the two wines obtained from clone 181 display the highest alcohol contents, clone 343 display an alcohol content of 14,1 %volume, while the lowest content in alcohol being obtained at the wine from clone 337, but, nevertheless, it has to be mentioned that it is a very good content, above many types of Merlot that exist already on the market. Another observation that is essential and that comes from the study of data regarding the alcohol content highlights the fact that the varieties that had as a stock the SO_4 have a higher amount of alcohol than the ones that had as a stock the 1103P. So, in conclusion, the only wine of the four with the alcohol level under 14% is the one obtained from clone 337.

Even though this wine contains less alcohol than the other Merlot wines, it is a good content, enough to produce high quality wines even of D.O.C type.

The following parameter of composition that had been under study, the glycerol, shows that there is a parallelism between the wine contents in alcohol and glycerol. In other words, the wine which is the richest in glycerol (12,8g/l) is also the wine with the highest alcohol strength (181/SO4)while the one with the lowest amount of glycerol (12,0g/l) is at the same time the one with the lowest alcohol strength(337/1109P) and the two intermediate clones share the same alcohol strength (181/1103P and 343/SO_4) and very close glycerol contents, 12,5 and 12, 4g/l respectively.

The third determined compositional parameter has a certain connection with the alcohol fermentation process. This is about the residual sugar, whose values emphasize the fact that all the wines display contents under 4g/l, so they are dry wines. (Băducă Cîmpeanu C., 2008). This is a positive thing that highlights the fact that the fermentation process had been a smooth one, without accidents. This is extremely important for the red quality wines which are, through excellence, dry wines. It is essential that all the wines have been dry and the yeasts have been able to fully metabolise the ferment sugars, under the conditions of some very rich in sugar unfermented wines. This detail has to be kept in mind and also underlined because an accidental fermentation with indigene yeasts might lead to uncertain results. (Popa A., 2008).

Table 1

The chemical composition of wines

Wine	Alcohol, %vol.	Glycerol, g/l	Residual sugar, g/l	Total acidity, g/l H_2SO_4	Volatile acidity, g/l acetic acid	Free SO2, mg/l
181/1103P	14,1	12,5	3,8	4,7	0,38	12
181/SO_4	14,4	12,8	3,6	5,1	0,44	12
337/1103P	13,7	12,0	3,5	4,8	0,40	14
343/SO_4	14,1	12,4	3,2	4,6	0,42	15

When it comes to quantity, there is no doubt that the second compositional parameter of wine, after alcohol is the glycerol, but when it comes to importance, and taking into account the organoleptical equilibrium, then the total acidity is of a highest importance, as well. (Stoian V., 2001). The study regarding the total acidity of the four Merlot wines displays exceptional values of this parameter, between 4,6 and 5,1g/l H_2SO_4. For some wines with the alcohol strength around 14% volume, a total acidity close to 5g/l H_2SO_4 is something rather rare and emphasize the fact that the harvesting of the grapes had not been done at an advanced super maturation but at a maturity that allowed them the maintenance of a good acidity level.

The most significant indicator of the wine health is the volatile acidity, expressed in g/l acetic acid. The data regarding the volatile acidity of the four Merlot wines display values between 0,38 and 0,44 g/l, which is a lot beneath the maximum limit of 1,2 g/l acetic acid, a level which is established by the Romanian wine-making legislation for the red wines beneath two years old, just like these wines. These values, which are almost three times beneath the superior limit, show the fact that all the wines are healthy and there is no danger of short and medium term biological instability.

This aspect is even more assuring as the values of the last studied compositional parameter, the content of free $S0_2$, are tranquillizing, because all the wines have over

10mg/l, the minimum level of preservation. The spectro-photometrical studies for the description of the polyphenolical composition and the chromatic characteristics were performed at a greater number of wines because, besides the four wines resulted as experimental varieties obtained from a single combination clone-stock, there were also achieved other combinations between the wines from the same clone but grafted on different stock, like the case of clone 181. There were also achieved combinations between wines obtained from different clones but grafted on the same stock, but it was also achieved a wine coming from the combination in equal proportion of the four main wines. Under these circumstances, from four initial wines, obtained from three clones, studied from the chemical perspective, at the spectro-photometrical studies, there were studied eight wines. The results of these studies displayed in table 2 show very good values of all the wines, from the point of view of their polyphenolic composition and chromatic features, which suggests that all the wines have a good potential of evolution.

Table 2

The polyphenolic composition and chromatic structure of Merlot wines

Wine	IPT	Anthocyans, mg/l	I	T
181/1103P	42,04	387	1,15	0,58
181/SO4	46,44	403	1,32	0,55
337/1103P	40,31	402	1,27	0,55
343/SO4	43,65	463	1,25	0,51
181	44,07	403	1,23	0,56
181+343/SO4	44,86	431	1,28	0,53
181+337/1103P	41,42	401	1,19	0,56
Merlot	43,35	429	1,22	0,54

As for the index of total polyphenols, its values are between 40,31 and 46,44. the wine with the highest index (46,41) comes from clone 181/SO_4, in other words this is the same wine that had the highest content in alcohol, glycerol and total acidity , a fact which suggests that this is one of the best Merlot clones. The other wine obtained from clone 181 but grafted on stock 1103/P display an index of total polyphenols significantly lower (42,04) and the wine coming from the blending between the two combinations clone-stock has the index of total polyphenols of 44, 07 so close to their arithmetical media. And the wine obtained from the combination of the two clones (181 and 343) grafted on stock SO_4 has a very good value of the index (44,86) which situates it on the second position between the two wines.

The lowest values of the total polyphenolic index belong to the wine 337/1103P and to the one resulted from the blending of the clones grafted on the stock 1103P. Thus, the study of the total polyphenol index obviously and clearly emphasize the fact that the highest values of this index are, from the clone perspective, at the wines obtained from clone 181 and from the stock perspective, at the stock SO4.

When it comes to anthocyans, the greatest content is to be found in the wine obtained from clone 343, grafted on stock SO_4 (463mg/l), followed by the wine obtained through the blending of the clones grafted on stock SO_4 (431mg/l). Thus, it is maintained

the superiority plus for the wines coming from the combination with the stock SO_4 unlike the ones coming from the combination of clones with the stock 1103P, but clone 181 does not have the highest content in anthocyans anymore, like in the case of total polyphenols, but the lowest ones. Taking into consideration all these data, we can draw the conclusion that the wines coming from clone 181 owe their superiority, in what concerns the total polyphenols, not to the anthocyans but to the tannins, the other great class of phenolic elements from wine. Under the conditions of high contents of anthocyans and of some indexes of total polyphenols with high values, it was almost obvious for the wines to display very high values of colorant intensity. The study of the values belonging to the elements of chromatic structure emphasize that all the wines obtained from certain types of combinations containing the stock SO_4, are much more colored than those in which this stock was not included, and as for the clone, even though clone 181 displayed the lowest contents in anthocyans, the wines obtained from it are the most intensely colored.

The values of the color tent comprised between 0,5l and 0,58 are characteristic for some very young wines, even where it is emphasized a weaker contribution of the yellow pigments to the color intensity. This is absolutely natural for the level at which the wines are and it will certainly modify throughout the wine evolution, while the red pigments do not suffer more serious modifications in a greater amount unlike the yellow ones which will have as a result the increase of the tonality value. What is undoubtedly interesting to remark is the fact that the wine which has been highly praised but at the same time extremely criticized by the wine tasters has at its root the same clone, 181. But in the first case, the most praised and appreciated wine was the one obtained from the combination between clone 181 and stock SO_4 while the one obtained from the combination between the clone and the stock 1103P was the least appreciated. As for the wine coming from the blending of the two combinations clone-stock, this one also obtained an intermediate score.

Taking into consideration the tiny differences between the wines at this early phase of their evolution, it is by all means unjustified a comparison between the clones because even the organoleptical features of the wines will evolve until consumption.

CONCLUSIONS

The main conclusions of the study regarding the compositional, chromatic and organoleptical features of the Merlot wines obtained in 2011, are the following:

- The Merlot sort is part of the traditional range of wines belonging to Samburesti vineyard and throughout a more than a century of existence, it has brought its contribution to the vineyard reputation on the wine market from both Romania and Europe. In the present, in the culture, there are more clones of Merlot grafted on different stocks in the structure of the new-founded plantations.
- The chemical composition of the wines obtained in 2011 display exceptional compositional parameters like in the case of top quality wines. That is why it has to be noticed the fact that the best results came from clone 181 and stock SO4, the wines obtained from combinations containing either this clone or this stock detaching themselves from the others.
- From the polyphenolic perspective, all the wines have displayed very good values of the index of total polyphenols, contents in anthocyans and colorant intensity. Even here, clone 181 and stock SO4 proved to be the best.
- The sensorial study of the wines revealed rather small differences between the ones being in an early phase of their evolution, differences that will become even more clear as the wines evolve and outline their sensorial profile, that is why it is relevant a firm

appreciation regarding the organoleptical features even though clone 181 and stock SO4 remarked once more.

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BIOCHEMICAL AND MORPHOLOGICAL INDICES DEPENDING ON ROOTING SUBSTRATE AT THE *Pelargonium peltatum* PLANTS

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Keywords: *rooting substrate, Pelargonium, rooting percentage, biochemical composition*

ABSTRACT

The present researches were focused on the effect of substrate type on the rooting process of the cuttings and on post rooting morphological development and biochemical composition in Pelargonium peltatum plants. Also, some biochemical parameters were analyzed to emphasize if the rooting substrates used influence the biochemical composition of leaves formed on shoots.
The maximum percentage of rooting cuttings and the highest number of leaves on the shoot were registered by using peat + perlite as rooting substrate. Rooting percentage values and morphological indicators were minimal at the cuttings rooted on sand.. The rooting substrate indirectly influenced the content in assimilatory pigments in the leaves and also the accumulation of biochemical compounds depending on the morphological features of the plant roots.

INTRODUCTION

Plant propagation by cuttings is the most commonly encountered method of vegetative propagation. For rooting cuttings there can be used a large variety of substrates, some natural (peat) or obtained by different methods and techniques.

Whatever its origin, a good substrate for rooting must meet certain conditions: to be easy, loose, with a high porosity for air and a good water retention capacity.

Also, the ability propagation by cuttings depends on the species, stock type, and environmental factors (Bhekithemba Mamba, 2010).

Genus Pelargonium includes over 170 species, originating in South Africa. One of the most decorative species is *Pelargonium peltatum*, known as „the flowing geranium", much appreciated thanks to the spectacular flowering in many colors. Very popular, it provides the setting balconies, terraces, windows and gardens throughout the summer (Amariutei A., 2010).

The present researches were focused to compare the effect of various specific rooting substrates on the rooting process on different specific substrates and post rooting morphological development and biochemical composition of the new plants. Therefore, some biochemical parameters were analyzed to emphasize if the rooting substrates used influence the biochemical composition of leaves formed on shoots. Thus, were determined: dry matter content accumulated in the leaves, crude protein content and total lipid and assimilatory pigments content in leaves (chlorophylls and carotenes).

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MATERIALS AND METHODS

In order to estimate the effect of rooting substrates on the rooting process and after rooting of *Pelargonium peltatum* were studied plants from the cuttings cultivated on different specific substrates in the greenhouses USAMV Bucharest, during February-March 2011.

As rooting substrates, which were also the experimental variants were used: perlite (V_1), sand (V_2), a mixture of equal proportions of peat and perlite (V_3) and finnaly, peat (V_4). The biological material consisted of the shoot tip cuttings with standard sizes of 6-7 cm long, harvested from mother plants of *Pelargonium peltatum*.

After trimming, there were selected 10 rooting cuttings by for each type of substrate. Throughout the rooting cuttings were maintained in greenhouse multiplier, where they were provided the same conditions in terms of environmental factors (temperature, humidity, light). After rooting were made observations and measurements on rooted cuttings on: the percentage of rooting, main root length, main shoot length, number of leaves on the vine.

Measurements of the biochemical parameters were made in the *Pelargonium* leaves using proper biochemical methods (Iordachescu D., 1988):

- The investigations of *chlorophyll and carotenoid pigments* were performed spectrophotometrically, after extraction in 80% acetone, at 663 nm, 646 nm and 470 nm wavelength. The results were calculated with Mackiney formula.
- *Dry mass content* was gravimetrically determined with a thermoscales.
- Determination of the content in *crude protein* was made after the digestion of the vegetal material by Kjeldahl method. The content in total nitrogen was measured by volumetrical method and converted in crude protein content.
- Content in *total lipid* was determined using the Soxhlet method: total fat was extracted in petroleum ether; then, the solvent was evaporated at 35ºC using a rotary evaporator; finally, the lipid content of samples was determined gravimetrically.

RESULTS AND DISCUSSION

1. Study of rooting process and of morphological development post rooting

As the percentage of rooting, the experimental results (Table 1) indicate that the best results were obtained by using peat + perlite as rooting substrate (100% rooting percentage). Only 56,66% rooting percentage was registered on sand substrate.

In the variant V_1 and V_2 the results were identical, respectively 83,33%.

Table 1.

Summary of the experimental results

Experimental variants	Rooting percentage (%)	Main root length (cm)	Main shoot length (cm)	Number of leaves/shoot
V_1 (perlite)	83,33	10,5	12,2	7,5
V_2 (sand)	56,66	7,5	9,93	5,6
V_3 (peat+perlite)	100,00	12,3	13,7	12,3
V_4 (peat)	83,33	8,5	13	8,5

Main root length ranged from 7,5 cm in the sand to 12,3 cm in peat + perlite. Close values were registered at the cuttings from variants V_1 and V_2: 10,5 cm in perlite and 8,5 cm in the peat.

Shoot length registered also the highest value (13,7 cm) in peat + perlite, while the lowest value (9,93 cm) was noticed to cuttings rooted in sand. Similar values, meaning 13 cm, respectively 12,2 cm were registered in cuttings harvested from peat and perlite.

The highest number of leaves per shoot was observed on peat + perlite (12,3), while sand cuttings reached the lowest number of leaves per shoot (5,4). The V_1 and V_2 values were very close (7,5 and 8,5) by using perlite and peat as rooting substrates.

2. **Study of biochemical parameters in the *Pelargonium* leaves in all experimental variants.** The content in chlorophyll of the leaves plays an important role in the process of photosynthesis, so that influences the growth and the development of plants. The biosynthesis of the assimilatory pigments depends on the mineral substances, therefore is essential the water and mineral substances supply by the roots (Burzo I, 2005).

Considering these information provided by the literature, the researches performed indicated that the rooting substrate indirectly influenced the content in chlorophyll in the leaves.

Thus, the obtained results were related to the number of the roots and also to the length of the main root. The calculated data (fig. 1) showed that content in chlorophyll *a* registered the highest value in the leaves harvested from the cuttings rooted on peat+perlite (103,57 mg/100 g), which registered also the longest main root.

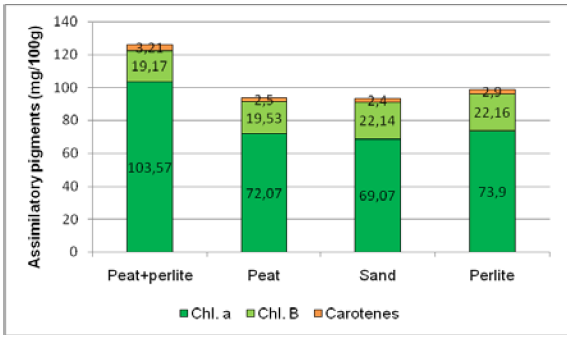

Fig. 1. Content in assimilatory pigments depending on rooting substrates

The plants rooted on perlite and on peat registered similar values of the content in chlorophyll *a*, while the variant which used sand as rooting substrate reached the lowest value of chlorophyll *a* (for 1,5 times lower than the variant on peat+perlite).

In all the studied variants the amount of chlorophyll *a* was higher that chlorophyll *b* in the leaves tissues, the ratio chlorophyll *a*/chlorophyll *b* varied between 5,4 at the plants cultivated on peat+perlite and 3,1at the plants cultivated on sand.

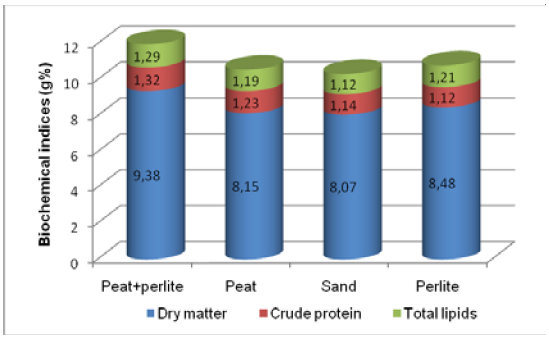

Fig. 2. Biochemical indices depending on rooting substrates

Also the synthesis of the carotenes was influenced by the rooting substrate. The differences between values of carotenes registered at the plants rooted on peat+perlite and at the plants rooted on sand were significant: 3,21 mg/100 g carotenes registered in the first variant compare with 2,4 mg/100 g carotenes in the sand variant.

According to the literature, the plants use water and mineral compounds absorbed by the roots from the soil in order to biosynthesize its own organic compounds (Burzo I., 2005). Therefore, the amount of biochemical compounds in leaves may be influenced by the morphological features of the plant roots, as root lenght (Denny, G.C., 2001).

The accumulation of biochemical compounds was influenced by the rooting substrate (fig. 2), so that the plants cultivated on peat+perlite registered a higher content in dry matter (9,38%) compare with 8,07 % dry matter at the variant cultivated on sand. The plants rooted on perlite also reached a high value (8,48 %), while the variant rooted on peat registered value close to variant on sand.

The results of the researches performed indicated that the use of the mixture perlite+peat as rooting substrate determined also an increased accumulation of proteins and lipids, which registered high values in the analyzed leaves. At the plants rooted on peat + perlite 1,32 g% crude proteins and 1,29 g% total lipids were determined compare with 1,14 g% crude protein and 1,12 g% total lipids at the sand variant. Also at the variants that used peat and perlite as rooting substrate were registered better results than the sand variant (1,23-1,12 g% crude protein and 1,19-1,21 g% total lipids).

CONCLUSIONS

The results obtained in the researches performed indicated that substrates used for the rooting of the *Pelargonium peltatum* plants influenced the rooting process and the morphological and biochemical features of the plants in post rooting period:

After all the observations and measurement better results were registered for rooted cuttings in substrate consists of peat + perlite.

- For cuttings rooted in perlite and peat were found close values of the analyzed indicators. Lower values were observed in cuttings rooted in sand.
- The maximum percentage of rooted cuttings was on peat + perlite.
- Values were identical percentage of rooting for cuttings rooted in perlite and peat, while the minimum value was registered in cuttings from sand.
- Maximum length of main roots was noted on the peat + perlite, while sand registered the lowest value.
- A greater number of leaves per shoot achieved the cuttings rooted in peat + perlite.
- The researches performed indicated that the rooting substrate indirectly influenced the content in assimilatory pigments in the leaves, which registered the highest value in the leaves harvested from the cuttings rooted on peat+perlite.
- The accumulation of biochemical compounds was also influenced by the rooting substrate, so that the plants cultivated on peat+perlite registered the highest content in dry matter, crude protein and total lipids.
- Using sand as rooting substrate determined low value of the analyzed parameters, may be because of the poverty in nutrients of this substrate.

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THE WALNUT CULTURE IN THE REPUBLIC OF MOLDOVA AND ITS PERSPECTIVES FOR DEVELOPMENT

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Key words: *walnut, variety, seedlings, growth, economic efficiency*

ABSTRACT

Since the year 2000 the walnut culture has known a substantial evolution in the Republic of Moldova, greatly sustained by the joint efforts of some active promoters and of the Government financial assistance. The methods of propagation of the walnut varieties have been consistently improved in the last ten years. In the meantime, better terms and conditions of the establishment and care of the walnut orchard have been implemented. Also, by the end of the '90, it came clear which are the best walnut varieties and to what use are they best suited.

The establishment of the walnut orchards has been greatly stimulated by informing the population about the existing demand for the walnut products, but also by helping out the business by granting it with important financial resources.

INTRODUCTION

In the light of the globalization process, the increasing cross-border movement of the goods produced by the national economies and their delivery at an international scale inherently determines these economies to focus their energies and resources on certain industries and on strengthening those industries' potential. For the decision makers in Moldova this phenomenon is to be taken very seriously and it is to play a key role in the further elaboration of the economic strategies for the country and in the identification of the resources needed in that respect.

It is known that the industrial complex of the Republic of Moldova is insufficiently developed to enable the country to get important incomes out of its exploitation. Therefore, taking that into consideration, altogether with the available soil and climate resources, one of the major value creation sectors Moldova has is agriculture. This branch supplies about 60% of the country's GDP. In response, it has been decided to spot the most profitable agricultural fields of activity and provide them the best stimulus.

Therefore, in 1999 the Parliament has enacted the Law of the walnut and the nut cultures. By doing so it created a legal framework designed to give technical and financial support to the sector. So we can come to the conclusion that at a nationwide level it has been acknowledged that walnut represents a revenue source, both for rural populations

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directly involved in the field and for a new generation of entrepreneurs that take the conception of the walnut orchard to higher levels of economic performance.

As a consequence of the above mentioned, in the last ten years the Moldovan Government has identified a number of fields of national economic interest and has, since then, multiplied the actions taken in order to assist the people involved and to launch or to sustain ongoing projects destined to promote these industries. One of those is the walnut business and it has gradually been on the rise, mainly by the establishment of more and more commercial orchards.

It's important to note that the majority of the Moldovan walnut orchards are designed in a classic planting system with high trees and big globular crowns that are hard to prune, difficult for harvest and are of low performance in the light of the standard requirements of integrated production (Ţurcan and Comanici 2004). The upgrading process that underwent and still undergoes the walnut culture is determined by the technological methods and instruments through which the process is carried on, based on the soil as a main production factor and resource.

Other features that help in the affirmation of the yield potential of a variety are related to the precocity and the type of fruit bearing, the pruning and training methods, the resistance to pests and diseases, the planting density and the used rootstock (Balan et al. 2001, Cociu 2003, Gena et al. 2004).

The purpose of this research is to promote a modern technology for the walnut culture, based on valuable local genetic resources and adapted to the soil and climate conditions existing here in Moldova. Therefore, new improvements will be brought upon this field through the promotion of modernized elements in the establishment and care of walnut orchards, no matter what the size, referring to the successful experience obtained in this matter in countries like France, the USA, Italy, etc.

MATERIALS AND METHODS

At this time, most of the walnuts that are being sold in the country or outside come from local biotypes that don't comply with the ever increasing consumer preferences. Those nuts come from trees issued from seed propagation that is known to induce a great genetic variability in the resulted hybrid populations. Therefore, by sowing a nut with very interesting features we can obtain very random results in the descendants. In order to have homogeneous walnut planting material, presenting all the advantages this type of material can provide and namely: early crop on setting, uniform fruit lots, standard trees vigor and vegetative growth it is necessary to multiply this material by vegetative propagation methods, instead of the generative ones (Balan et al. 2001, Cociu 2003, Babuc 2012).

In Moldova, in order to identify walnut genetic resources, a selection process has been carried out over the years, focused on spotting and registering of natural valuable biotypes. In this process, priority was given to those varieties that seemed most fit from an ecological and economical point of view. Thereby, several varieties have been selected and named, among them: Cazacu, Cogâlniceanu, Costiujeni, Călăraşi, Schinoasa, Pescianski, etc. If grafted onto Juglans regia seedlings, a walnut tree starts bearing important amounts of fruits from the 6^{th} – 8^{th} year, whereas if issued from seed they start producing in their 10^{th} – 15^{th} year after planting. Fruits obtained from grafted walnut trees have better quality, higher commercial and nutrition value. It is determined that from generatively propagated

walnut trees, only about 20-25% of the crop can be employed for commercial use (Cociu 2003, Ţurcan and Comanici 2004).

RESULTS AND DISCUSSION

The walnut is being cultivated from ancient times in different regions that have the temperate type of climate. On Moldovan soil, this species is being known and grown for about 2000 years. Walnuts represent a complete and concentrated nutrient; they contain fatty, mineral and protein substances, vitamins, carbohydrates. The walnut is a fruit bearing tree with great potential for extension of the orchard acreage and fruit production volumes. This potential is determined by the ever growing global demand for the walnut orchard produce.

Also, the walnut tree produces raw material for numerous industries, is also used in medicine and is one of the few trees that absorbs heavy metals from the atmosphere. Moreover, despite its slow growing rates, the walnut has a strong root system and is widely used in forestry, especially against soil erosion and landslides and as a primarily used species in wind breaking tree screens (Cociu et al. 2003). Based on a census carried out in the naturally existing trees populations, it has been determined that in 1994 there were about 2,2 mln walnut trees. Out of the 2,2 mln ha of farm land available in Moldova, about 800 thousand are predisposed to erosion. The walnut tree grows and performs well in this type of terrains. Therefore, measures should be taken to make sure this fact is known by the stakeholders so a more reasonable use could be made of the existing farm land.

The walnut orchard acreage has evolved from 3300 ha registered in 2003 to approximately 9400 ha in 2010. During the last three years, the nursery material production has amounted in average 230 000 grafted walnut trees per year. Between 2003-2010, the production of kernels has been about 13,1 thousand tons yearly, gradually increasing. Since 2005, the acreage of the planted orchards is constantly on the rise. According to the National Programe for the development of the nut cultures, in the year 2020, the total area of the walnut orchard will amount to 14-15 000 ha and the total in shell nut production will touch up to 60 000 tons (currently 25-35 000 tons).

Additionally, it is important to note that we are on the way of effectively intensifying the walnut orchard and bringing the adaptive measures required in this process (choosing the adequate training systems, establishing the mineral nutrition and irrigation programmers, deciding on how to harvest the fruits, etc.).

There are a number of nurseries (SRL „Kernel Grup", SRL „Gospodarul Rediu", SRL „Pepeniera Pomicolă Voineşti", etc.) that have specialized in the production of grafted planting material thus generally covering the internal demand. Besides the internal market, some of the most successful Moldovan nurseries are now exporting large quantities of grafted walnut trees in nearby countries, mostly Romania and Ukraine while some others have established partnerships with some European walnut nurserists and supply them with walnut rootstocks.

At the same time, we have to underline an important complementarity that is being currently created between several participants in the process (i.e. the process of the development of the nut cultures as an important economic part of the Moldovan agriculture). Since 2006, the Government provides important finances as a stimulus plan for the establishment of walnut orchards. This is being carried out in the form of subsidies.

More specifically, for one hectare of planted walnut orchard the sum amounts to 800$ (The Yearly Regulation on Farmers Subsidizing). Usually, this money is enough to cover the cost of the planting material. By doing so, the Government stimulates both the walnut orchard establishment rates and the walnut nurseries development rates.

At this level, it is interesting to note the direct relation that has developed between the growth dynamics of the yearly planted acreage and the quantity of the walnut planting material produced yearly (tab.1).

Table 1.

The production rates of the grafted walnut planting material (pieces)

year 2009	year 2010	year 2011	year 2012
171 500	198 900	283 600	250 000

With regard to the production of the walnut planting material, which will be the focus of the second part of this study, the Republic of Moldova has made important progress in the last seven years. That has mainly happened thanks to the sustained demand for the product, which in return had encouraged the producers to work on improving the propagation techniques. There has been an important step up in the general production efficiency, going from production rates of 25-50% grafted trees ready for sale (relative to 100% initially grafted) at the beginning of the years 2000 as high as 40-70% at the current time (Turcan and Comanici, 2004).

Concerning the propagation methods, several have been tested until now, including open field grafting. After a ten years period of trials (but also referring to and taking into consideration conclusions of the essays carried out previously, in the last 60 years), the nurseries have retained only the most efficient and stable ones. For instance, it has been demonstrated that the success rate of the open field grafting or budding (chip or patch) is too scarce and inconsistent to recommend it for further use. So, producers have focused their attention and energy on the most promising techniques: the table grafts, done at the end of the dormant period, from February and through April (Babuc 2012). Among these methods, the one that had generated the best results are the whip graft and the omega graft. Except the type of combining the scion and the rootstock, other elements that refer to the grafting method that have been improved are: the length and characteristics of the stratification period, the preparation and type of the sawdust used in the stratification, ways and methods used to force the rootstock into active vegetation prior to grafting, the acclimation of the grafted trees after the stratification, the further care to be given to the plants in the field (Ţurcan and Comanici 2004).

Another important evolution that happened in the Moldovan walnut orchard relates to the varieties employed in the orchard foundation. It is obvious that the yield efficiency greatly depends not only on the soil type and the amount of available rainfalls, or on the agronomic care that is taken of, but also on the genotype of the varieties chosen for a given orchard (Pîntea 2004, Botu at al. 2001). In the last ten years that have seen the rise of the establishment of commercial walnut plantations, the Moldovan producers have succeeded to identify the most valuable varieties. The criteria that have been considered at ranking the varieties have been the following: early crop on setting, yield efficiency per tree, the young tree establishment in the first years of the orchard, the capacity to yield and

to differentiate flowering buds in the conditions of increasingly hot and dry summers, different varieties resistance to extreme winter frosts and to late returning frosts in April and May months, the ability to synthesize organic substances in conditions of extremely high temperatures and acute lack of atmospheric and soil humidity (Ghena and Branişte 2003, Ţurcan and Borozan 2003).

From another point of view, the evolution and selection process through the existing walnut varieties has also taken into account the consumer preferences. As a result, mostly came out the thin shelled varieties with good tasting and easy extractable white kernel. Therefore, the most popular varieties in the Moldovan selection were: Cazacu, Cogâlniceanu, Pescianski, Costiujeni, Calarasi, Schinoasa (Ţurcan and Comanici 2004). Among others, we have to mention the introduction of foreign varieties, mostly French and American, in the Moldovan walnut culture. Franquette, Lara Pieral, Fernor, Chandler, Hartley, Payne (Germain et al. 1999), otherwise very precious varieties, haven't unfortunately proved well adapted in the more hostile continental climate found in Moldova, compared to the milder one encountered in the area of provenance of the above mentioned varieties.

CONCLUSIONS

Referring to the what has been discussed here above, we can firmly assert that currently, the walnut culture is on an ascending trend and, moreover, can already account for some important progress that has been made in the affirming of the most suited and valuable varieties, in improving the most efficient propagation techniques and in determining the most favorable regions in Moldova that can host walnut orchards. From a commercial point of view, there have been comparatively tested technical and organoleptic qualities of these varieties. This has paved the way for facilitating the sale of the walnuts, by establishing different uses (fresh consumption, industrial use, etc.) that could be given to different varieties. Characterized by distinct features, growers now know for what destination each variety is best suited for.

Nevertheless, there still is a considerable progress to be made in order to continue to develop this interesting branch and recently, in the center of the producers' attention have come certain biotypes of rootstocks, but also a new range of varieties for fruit. At the same time, new and potentially interesting propagation methods are being discussed, new trials for determining even better planting distances are being put in place, some producers are trying to find ways to optimize the soil laboring and to improve the mineral nutrition and irrigation programs in the orchard, etc. Research is also and very much required for determining how to better use the available instruments and leverages in order to insure a more efficient control over the mechanisms by which ecological, biological and technological factors have an impact upon the success rate in walnut propagation methods and in walnut orchard establishment and management.

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PARAMETERS CHERRY TREES IN FUNCTION OF VARIETY AND CUTTING SYSTEM

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Key words: *cherry, variety, cutting the trees, growth*

ABSTRACT

We studied the parameters varieties Valerii Cikalov and Record, grafted on Mahaleb rootstock aged 9-10 years. The distance of planting trees is 6x5 m. The trees are driven by free forms natural crown shape with large.

Was studied the cutting of production trees in the rest and wood vegetation period , the cutting of semiskelet branches in the rest and wood vegetation period from 3-5 years.

The phasing rejuvenation of the semiskelet branches for 3-5 years wood depends on the sorts and circumstances of the trees. The cutting of the branches semiskelet is made only for lateral branches or may bouquet which have vegetarian bugs.

INTRODUCTION

The majority cherry orchards from the Republic of Moldova are projected in the classical system with high size, the wreath with big volume; it is difficult to keep and to harvest because they are not performed by the exigency produce standards (Balan et al. 2012, Donică et al. 2005).

The modernization of the cherry is determined by the technological methods and instruments, using the soil as the principal source. Another elements which influence the biologic potential of the soil production are connected by the fruit-bearing , the type of fruit, the way of living and leading, the resistance against illness and pests, the planting density (Budan and Gradinariu 2000, Mitre et al. 2010; Balan et al. 2012).

The enrichment of cutting technology that is introduced through new modern methods in all the world, it makes the pressure upon the traditional technology, they must enrich for getting the maximum of high biologic potential (Cimpoieş 2000, Ştefanco et al. 2009).

The task of the investigation is to promote the modern technology of cherry planting, adapting to the national clime of the Republic of Moldova. In this way we can raise the capacity of the harvest.

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MATERIALS AND METHODS

The investigation was made in the cherry orchard of "Vindex-Agro" planted in 2003 year in Malaesti village, Orhei department. The biological material was made from Valerii Cikalov and Record sorts, grafting on mahaleb. The distance between plants is 6x5 m.

The trees are characterized by their wreath shape with big volume. The wreath is made by bazal level with 3 frameworks that there are 3-4 frameworks, putting axle spiral with 35-40 cm distance. The axle is over the last framework after 3 years growing.

To realize the investigation there are the following variants.

V1- cutting of produce (keeping and fructification) in the rest period.

V2- cutting of produce (keeping and fructification) in the vegetation period.

V3- cutting of semiselet branches, in the rest period of 3-5 years wood.

V4- cutting of semiskelet branches, in the vegetation period.

Every variant includes 4 repetitions of 8 trees. The measures were made in the field condition and laboratory according to the methods made and checked by Мойсейченко et al 1994.

The parameters of wreath were learnt annually at the end of vegetation period after trunk diameter, high and breadth, the average length of branches through measure method of 32 trees. During of vegetation period in the orchard were made 4 tillage and chemical treatment against illness and pests.

RESULTS AND DISCUSSION

The investigation made to optimize the parameters of wreath opened new diversity between height, thickness and shape and cutting production that can keep a big part of energy, harvest and to raise the use of substance of agro biological processes (Cimpoieş 2000, Balan et al 2001, Karoly et al. 2008, Mitre et al. 2007).

Trees growing in the harvest period were different in view point of sort and cutting system (table 1). If we analyze the drunk diameter of every sort, we can observe that Valerii Cikalov sort had the variety from 14,4 cm till 20,3 cm but Record sort had the same values with Valerii Cikalov sort and the diameter was from 16,5 cm till 19,7 cm. In 2012 we notice that the average growing of trunk diameter was 2 cm for both sorts. Cutting system does not influence the trunk growing of the trees.

To form and to keep a wreath must respect the sort and the type of fructification, the maximum illumination and the suitable tillage. Due to the shape and strength, they must have a lot of fundamental structure elements, to be delimitated as number, space orientation, limits of distance between them using the biggest vegetarian potential of the tree. To cut a tree was taken in consideration the biological characteristics of the sort and its reaction of different type of cutting (Perry 1987, Negrun et al. 2005, Cimpoieş 2000, Balan et al 2001, Magyar and Hrotku 2005).

A rational cutting makes the beginning of the crop and to get high and qualitative harvest. They raise the economical efficiencies of the trees (Sansavini 1984, Simon et al. 2004)

Table 1

Parameters cherry trees by variety and cutting system

Mahaleb rootstock, 6x5 m planting distance, improved natural crown shape with large, Ltd Agro Vindex, the years 2011 – 2012

Cutting system	Variety Valerii Cikalov		Variety Record	
	year 2011	Year 2012	year 2011	year 2012
Trunk diameter, cm				
V1	14,4	16,8	18,8	20,6
V2	20,3	22,1	19,7	22,1
V3	18,8	20,8	17,4	19,0
V4	18,2	20,2	16,5	18,3
Crown diameter, m				
V1	3,7	4,7	4,3	4,7
V2	4,0	4,8	4,2	4,8
V3	3,8	4,6	4,4	4,9
V4	4,2	4,8	4,1	4,5
Crown height, m				
V1	4,2	3,9	3,9	3,7
V2	4,3	4,0	3,7	3,8
V3	4,6	4,2	3,6	3,8
V4	4,6	3,8	3,8	3,9

The tree growing in the harvest period was different depending on the sorts and cutting system. The biggest thickness average of wreath was 4,1-4,5 to Record sort. Valerii Cikalov sort is characterized through less insertion than Record sort and it makes bigger wreath.

The wreath diameter of Valerii Cikalov sort is 3,7 m in the production cutting and 4,2 in the vegetarian cutting of semiskelet in the vegetarian period. The Record diameter of wreath is the biggest and was pointed from 4,1 m till 4,4 m. We can observe that we do not use rationally the territory in the ninth and tenth year of planting.

Height growing of the trees were differentiated in the limits of insurance of the sort and cutting system. The trees to all the cutting system are at the height of 3-5,4 m. We observe the investigated trees from 2011-2012 have the bigger height than optional parameters. The conclusion is that the cherry tree in the growing period goes the slowest in the superior part of the wreath. In 2011 the tree height at Valerii Cikalov sort was 4,2-4,6 m.

Keeping the optional age structure and fructification of the semiskelets are determined by the vegetative process of growing, there are the results of the wreath forming. The physiological situation is considerate when the tree has the fruits and growing process is over 35-40 cm (Ghena et al. 2004, Babuc 2012).

The investigation showed that the annual growing depended on the sort, the clime and cutting system. The annual growing of branches are thanks to phasing cutting of semiskelet branches and keeping the optional parameters of geometrical structure of wreath. These influence to the interior light of the tree and to develop the bugs of the harvest for the next year.

The average length and the total branches are very sensible to ecologic and technologic conditions. Putting the offshoot on the wooden spigot of different age (table 2)

is determined by the biological priorities of the soil and less of the cutting system. Analyzing the results we observe that the soil and the cutting system influence very much upon the branch number and the length of annual branches made from the offshoot formation. If we use cutting wreath without spigot we see on the many-sided branches (3-7 year) formed 2-3 offshoots for Valerii Cikalov sort and 1-3 offshoots for Record sort. They depend on the cutting time. The average length of annual branches formed by spigot was influenced by investigated sorts.

Table 2

Parameters annual branches depending on variety and cutting systems

Mahaleb rootstock, 6x5 m planting distance, improved natural crown shape with large, Ltd Agro Vindex, the years 2011 - 2012

Cutting systems	Reduction cuts on wood aged 3-5 years	Number of annual branches formed of buds, pc	The average length of annual branches formed of buds, cm	Annual aggregate length of branches formed of buds, m
Variety Valerii Cikalov				
V1	-	2	52	1,06
V2	-	3	61	1,80
V3	3 (4) 5	6 (4) 2	48 (60) 67	6,71
V4	3 (4) 5	3 (5) 3	39 (38) 53	4,82
Variety Record				
V1	-	1	47	0,47
V2	-	3	50	1,53
V3	3 (4) 5	3 (2) 0	39 (47) 0	2,21
V4	3 (4) 5	3 (4) 0	31 (36) 0	2,35

In the first and the second variant, when it was used the cutting production, the harvest is situated on the outskirts of the wreath, this makes the trees and fruits to damage. In the third and fourth variants, when it was used the spigot cutting of different ages we observe that the annual branches are more. Valerii Cikalov sort forms more offshoots from anticipate bugs than Record sort. In the variant where it is used cutting in the rest period, the number of the formed offshoot are from 3 till 6 annual branches per 3 years and 4 branches per 4 years spigot.

The number of annual branches formed on the spigot (tab.2) is influenced by the sort and cutting system, in spite of the cutting system, Valerii Cikalov sort ensured more annual development then the Record sort.

The biggest parts of values were ensured by phasing cutting of the semiskelet branches in the vegetarian wooden period of 3-5 years. The conclusion is that the branches quantity formed on the spigot for cherries depend on the tree strength and geometrical structure of the orchards.

On the spigot of 5 years for Record sort did not form the annual branches but for Valerii Cikalov was observed few numbers of annual branches. The average length of annual branches formed on the spigots with different ages was from 48-67 cm for Valerii Cikalov sor and 39-47cm for Record.

In 4 variant where cuts were applied during the growing season with fouling formation, the number of annual caps branches and their length recorded lower values, which ranged from 2 shoots average length of 39-47 cm to 5 shoots whose average length is 38-53cm.Research over the years aggregate length of branches annual caps formed on the variety Valerii Cikalov was higher than the variety Record.

So semischelet branches gradually rejuvenate the wood is 3-5 years depending on type and condition of the trees. Semischelet cutting branches is performed only at a lateral branch or bunch more who vegetative buds.

CONCLUSIONS

Analyzing the values of growing for Valerii Cikalov and Record sort of cherries, it can say that they have different sizes and biological status. It is due to that the buds form from the offshoot that they grow constantly. The different biological beginning is due to the situation of the wreath on the biennale and many-sided wood and thanks to different positions of offshoot. In spite of the annual grow has different length and position.

Cherries tree in the period of growing and crop in the rejuvenation condition, 3-4 years wood semiskelet, they have the possibility to form many diversities of annual branches with different biological level.

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COMPARISON OF LIGHT REFLECTION INDICES OF SEVERAL GENOTYPES OF APPLE TREES IN MĂRGINIMEA SIBIULUI

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Key words: *index, reflection, apple tree, chlorophyll*

ABSTRACT

There has been a tendency in the last years to focus on immediate and accurate assessments of crop cultures. In order to meet such expectations, certain small, portable and easy-to-use devices have been produced. For example, Crop Circle ACS-430 has been used to determine the index of light reflection for certain genotypes of apple trees growing in Mărginimea Sibiului. The percentage of chlorophyll in the leaf has also been determined along with the light reflection index. The purpose of this experiment is to correlate the light reflection index with of chlorophyll in the leaf. Acknowledging the relationship between reflectance index and chlorophyll content for certain genotypes of apple trees in the area known as Mărginimea Sibiului is of major importance in differentiating among genotypes.

INTRODUCTION

The research studies focusing on measurements of cultivated plant using Crop Circle ACS-430 have been carried out in USA on corn. At the University of Missouri a series of research has been made related to the index of light reflection in order to assess the health and degree of fertility of nitrogen-fixing crops. In this way, specialists could make a quick assessment of the nitrogen quantity needed to fertilize the crop. (Kitchen et al., 2010; Solari et al., 2008).

The Crop Circle ACS-430 active crop canopy sensor comprises a polychrome emitting diode (LED) and two separate detectors. Designed for nadir sensing, this equipment has a field of view proportional to the height above the target. The response has been described as relatively constant over the field of view (Holland and others, 2005). Green Seeker sensor, developed based on the initial work by Stone et al., (1996) uses a separate LED for each wavelength and only one detector.

The development of automatic-diagnosis technique as well as other expert systems is now on an early, rudimentary stage, although the first steps in automatic image segmentation of damaged tissues of plants have been made (Moshou et al., 2005). Despite difficulties, monitoring the distribution of less developed crown or yellow foliage

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using NDVI airborne sensors can often make available an efficient indicator of vermin or contamination and disease. This is typical especially for those situations where damages may be identified due to the knowledge developed related to the soil behind a specific cause; for example, the spreading of Phytophthora root mould in case of raspberry plots, or vermin in forests. A good example is teledetection of defoliation using NDVI that has long been used starting with the first studies on gypsy moth in North America in the 1970s, using the Mss Landsat data, and up to the most recent studies like those on the birch moth in Scandinavia (Jepsen et al., 2009).

In such cases the general cartography describing the spreading of contamination over large areas may be an efficient tool to monitor the spreading, also a good aid in soil management, or even in the investigation of climate change impact. In practice, the predominant use of spectral indices on satellitary scale applied on change-detection grounds whether at the level of chlorophyll in the leaf or of LAI according to a spectral vegetation index. Although many of these indices have been tested, successfully in some situations, they only identify chlorophyll or LAI changes, and not so much the specific disease.

The purpose of measurements is to determine the correlation of light reflection index of x foliage system and the percentage of chlorophyll in the leaf. This way we can determine and explain the differences among the apple tree genotypes.

MATHERIAL AND METHODS

The study focused upon several genotypes of apple tree in the region called Mărginimea Sibiului, to which certain measurements have been applied using Crop Circle ACS-430 and Spad 502.

Crop Circle ACS-430 is a light sensor that has been designed to take measurements related to disease and vermin control at day or night. The sensor can be mounted to almost any type of vehicle at a desirable distance of the target crop or plants. The sensor comprises its own polychrome technology, made up of one source of light that detects the green part of plants.

The technology of the light source emits simultaneously both visible light and near-infrared (NIR) from one LED light source only. The greatest advantage this technology offers is that the source of light that can be detected on the surface of plants, under analysis, for both visible and infrared band specters of light. Crop Circle ACS-430 measures reflectance in three bands simultaneously: 670 mm, 730 mm and NIR bands. Reflectance data measured by the Crop Circle ACS-. 430 allows the user to calculate classic vegetation indexes from plant canopies such as the NDVI and SRI indeces. This allows the user to evaluate thousands of detected vegetation indices for airbore measurements, as well as satellite ones, sent with the help og GPS teledetection applications. The serial data generated by the sensor may be easily processed with the help of a laptop, PDA or other data storage devices.

NDVI (The Normalized Difference Vegetation Index) is a simple graphical indicator that can be used to analyze remote sensing measurements and evaluate whether the object under analysis comprises or not green vegetation, and implicitly if the vegetation is dead or alive.

Live green plants absorb solar radiation in the photosynthetically active radiation (PAR) spectral region, which they use as *a* source of energy in the process of photosynthesis. Leaf cells have also evolved to scatter (i.e., reflect and transmit) solar radiation in the near-infrared spectral region (which carries approximately half of the total incoming solar energy), because the energy level per photon in that domain (wavelengths longer than about 700 nanometers) is not sufficient to be useful to synthesize organic molecules. A strong absorption at these wavelengths would only result in overheating the plant and possibly damaging the tissues. Hence, live green plants appear relatively dark in the visible spectral region (VIS), leaves absorbing this radiation, and relatively bright in the near-infrared (NIR) due to the infrared absorption. The pigment in green plants, chlorophyll, strongly absorbs visible light (from 0.4 to 0.7 μm) for use in photosynthesis. The cell structure of the green plants, on the other hand, strongly reflects near-infrared light (from 0.7 to 1.1 μm).

Remember, NIR and VIS stand for the spectral reflectance measurements acquired by the device in near-infrared regions and the visible (red).

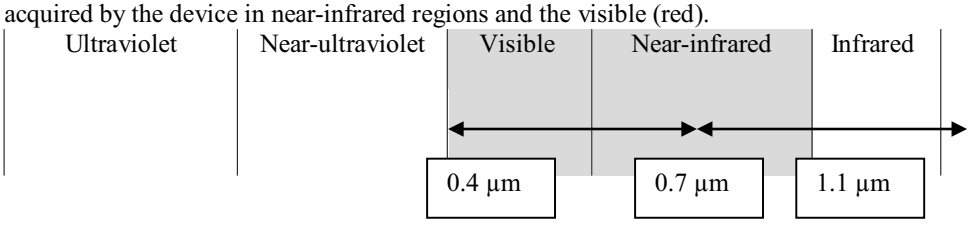

Fig. 1. Regions identified by the device, corresponding to absorption in the visible range and near-infrared reflectance

The NDVI Vegetation Index is calculated using the following formula:

$$\mathrm{NDVI} = \frac{(\mathrm{NIR} - \mathrm{VIS})}{(\mathrm{NIR} + \mathrm{VIS})}$$

SPAD-502 easily performs quick measurements of the chlorophyll content of plant leaves without damaging the leaves exposed to analysis.

The measured spectrum capacity of SPAD-502 varies from 650 nm to 940 nm.

After measuring the chlorophyll content, the device has the option to measure the ideal moment to add the supplementary fertilizers during the growing season.

The chlorophyll content in leaves has been measured separately, on leaves infected by *Venturia inaequalis,* without involving the ones that were not affected by it. The chlorophyll content in the leaves has been measured on the south part of each tree crown in turn, repeating the 30 measurements taken 3 times in a row. In a parallel manner, we have measured the percentage of chlorophyll in the healthy leaves that were not afflicted by black spot.

The measuring has been carried out on the middle part of the leave, both on its length and its width. Light reflectance has been measured on the south part of each tree crown in turn, while measuring the chlorophyll at the same time.

The study consisted of 10 apple tree genotypes, growing on the territory of 8 villages.

RESULTS AND DISCUSSIONS

The measurements on the 10 target genotypes included in the study prove that the medium-green index varies from 0.54 for the Fântânele 44 genotype to 0.76 in case of Cisnădioara 21. In Tabel 1 we can see that the average reflectance index in infrared spectrum range (NIR) bears the lowest value in case of Sibiel 43 genotype, of only 3.23 nm, while the average 9.48 nm value is the highest in Răşinari 18 genotype case.

The average reflectance index in the visible spectrum range (VIS), for the 10 apple tree genotypes studied, varies from 0.50 nm in Sibiel 43 genotype to 0.86 nm in Fântânele 44 genotype. The differences among these genotypes can be explained by the fact that the lower values describe the Sibiel 43 genotype, since it is in decline.

The average content of chlorophyll in the leaves that haven't been attacked by the black spot varies from 45.10% in Apoldul de Sus 2 genotype, while the lowest value is of 35.60% and could be found in Fântânele 30 genotype.

The average content of chlorophyll in the leaves that haven't been attacked by the black spot varies from 41.80% in Cisnădioara genotype, while the lowest value of 29.86% describes the Fântânele 30 genotype. The density of chlorophyll in the leaves differs greatly from the healthy leaves to the ones infected by *Venturia inaequalis.* Thus, in case of Răşinari 18 genotype we have a difference of 9.17% chlorophyll density, while in Fântânele 41 genotype a concentration of 7.44% chlorophyll content.

Small differences in the percentage of chlorophyll in the leaves may be observed, according to table 2, in such genotypes as Topârcea 22—a 1.13% difference—and Cisnădioara 21, bearing a difference of 2%. The relation between the chlorophyll content and the intensity of the black spot attack on the leaves varies from 0.76 in Răşinari 18 genotype to Topârcea 22 genotype, described by 3.61.

Table1

Average vegetation indices generated by carrying out measurements on apple trees using Crop Circle ACS-430

No.	Name of Apple Tree Genotype	NDVI	NIR (nm)	VIS (mn)
1.	Păltiniş 42	0.72	8.29	1.32
2.	Topârcea 22	0.59	7.72	1.79
3.	Fântânele 30	0.57	3.66	0.91
4.	Fântânele 41	0.74	9.33	1.36
5.	Sibiel 43	0.72	3.23	0.50
6.	Tilişca 49	0.67	4.61	0.85
7.	Cisnădioara 21	0.76	6.60	0.88
8.	Răşinari 18	0.68	9.48	1.62
9.	Apoldul de Sus 2	0,70	4,87	0.86
10.	Fântânele 44	0.54	6.56	1.86

NDVI – vegetation index
VIS– visible spectrum (nm)
nm – nanometer

Table 2

Correlation between normalized difference vegetation index and intensity of attack by *Venturia inaequalis* to some apple tree genotypes

No.	Name of Apple Tree Genotype	Average chlorophyll content in healthy (unattacked) leaves (%)	Average chlorophyll content in attacked leaves (%)	Attack intensity *Venturia inaequalis* %	Relation between attacked leave chlorophyll and intensity of attack
1.	Păltiniş 42	41.53	38.5	35	1.10
2.	Topârcea 22	36.16	35.03	10	3.61
3.	Fântânele 30	35.60	29.86	10	2.98
4.	Fântânele 41	43.30	35.86	20	1.79
5.	Sibiel 43	39.10	36.60	15	2.44
6.	Tilişca 49	41.16	37.60	25	1.50
7.	Cisnădioara 21	43.80	41.80	40	1.04
8.	Răşinari 18	43.70	34.53	45	0.76
9.	Apoldul de Sus 2	45.1	38.70	35	1.20
10.	Fântânele 44	38.10	32.46	10	3.24

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COMPARISON OF THE LIGHT REFLECTION INDEX OF SEVERAL GENOTYPES OF PEAR TREES IN MARGINIMEA SIBIULUI

Barbu Ion[1]

Key words: *index, reflection, pear tree, chlorophyll*

ABSTRACT

In agriculture, as in all economic fields, one aims at maximizing the profit through the increase of production with low expenses but with the keeping of the environment in its best possible state. The present study focuses on the correlation between the light reflection indices measured with Crop Circle ACS-430 and the percentage of chlorophyll in the pear leaves measured with SPAD 502.

Both devices are of small size and they are either portable or assembled to different farm equipments. The percentage of chlorophyll in the leaf has also been determined along with the light reflection index. The purpose of this experiment is to correlate the light reflection index with the percentage of chlorophyll in the leaf. Acknowledging the relationship between the reflection index and chlorophyll content for certain genotypes of pear trees in the area known as Mărginimea Sibiului is of major importance in differentiating among genotypes.

INTRODUCTION

The development of automatic-diagnosis technique as well as other expert systems is now on an early, rudimentary stage, although the first steps in automatic image segmentation of damaged tissues of plants have been made (Moshou et al., 2005). Thus, it is necessary to find ways to automate the identification and quantification of certain diseases based on the observed pattern, as long as air sensor scaling and satellite remote sensing is not possible at present.

The research studies focusing on measurements of cultivated plant using Crop Circle ACS-430 have been carried out in USA on corn. At the University of Missouri a series of research has been made related to the index of light reflection in order to assess the health and degree of fertility of nitrogen-fixing crops. In this way, specialists could make a quick assessment of the nitrogen quantity needed to fertilize the crop. (Kitchen et al., 2010; Solari et al., 2008).

The Crop Circle ACS-430 comprises a polychrome emitting diode (LED) and two separate detectors. Designed for nadir sensing, this equipment has a field of view proportional to the height above the target. The response has been described as relatively constant over the field of view (Holland et al., 2005). The Green Seeker sensor, developed

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based on the initial work by Stone et al., (1996) uses a separate LED for each wavelength and only one detector.

Monitoring the distribution in the areas with poorly developed or yellow canopy, using NDVI airborne sensors, may frequently represent a useful indicator of pests or diseases. This aspect occurs especially where damages can be assigned to a specific cause based on the familiarity with the soil (for instance, the spread of Phytophthora root rot to the raspberry plots, or of pests in the forest areas). For instance, the remote sensing of unleaving using NDVI was highly used since the fist studies on the gipsy moth in North America, in the 1970's, using the Mss Landsat data, until several recent studies such as those regarding the birch moth in Scandinavia (Jepsen et al., 2009).

In such cases, the general mapping concerning the infestation spreading on large areas may be a useful instrument for monitoring spread and for supporting soil management, or even for investigating the impact of climatic change. As far as the practical aspect is concerned, the large use of spectral indices on a satellite scale was based either on detecting changes on the level of chlorophyll content of the leaf, or on LAI, through a spectral index of vegetation. Although several such indices were tested, sometimes successfully in a certain context, they rather track down the chlorophyll or LAI changes than the specific disease.

MATERIAL AND METHODS

The study was based on several genotypes of pear trees in Mărginimea Sibiului, which were measured with Crop Circle ACS-430 and Spad 502.

The studied genotypes of pear trees were divided on the territory of 5 localities, namely: Tilişca, Fântânele, Sibiel, Cisnădioara, Topârcea.

The Crop Circle ACS-430 is a light sensor that can perform measurements in the field of diseases control and pests, integrated both on sunny days and at night. The sensor can be installed on almost any type of vehicle at the desired distance from the analyzed crop or from the target plants. The sensor is incorporated with a proper polychrome technology made of a light source in order to illuminate the foliar system and to detect the green part of the plants.

The technology of the source light simultaneously emits both visible light and near infrared (NIR) from only one source of light LED. The major benefit of this new technology is the light source that can be detected on the surface of the plants submitted for analysis, which is identified both for the visible lights specters and for the infrared light beams. The Crop Circle ACS-430 can perform measurements on three bands simultaneously: 670 mm, 730 mm and NIR bands. The data measured by Crop Circle ACS-430 allows its user to calculate the vegetation classic index of plants, such as NDVI and SRI indices. This allows the user to make use of scores of vegetation indices detected for air measurements and transmitted through satellite by GPS and remote sensing applications. The serial data produced by the sensor can easily be processed by using a data processing device.

NDVI (Normalized Difference Vegetation Index) is a simple digitizer that can be used in order to analyze remote sensing measurements and to evaluate if the observed objective contains green vegetation or not.

Alive plants absorb solar radiation in the region of the electromagnetic spectrum called photosynthetically active radiation (PAR) and use it a source of energy for the specific processes of photosynthesis. Leaves cells have the ability to reflect the solar energy

form the spectral region of the near infrared (which transports approximately half of the solar energy) as the energy per photon in this field (wavelengths over 700 nanometers) is not sufficient in order to be useful for synthesizing organic molecules. A high absorption at this wavelength (if leaves would not reflect it) would have as result the overheating of the plant and a possible damage of tissues. As a result, alive green plants appear relatively dark in the visible spectral area (VIS), al leaves absorb this radiation, and rather luminous in the spectral area specific for the near infrared, due to the infrared reflection (NIR).

The pigment of living plants, chlorophyll, intensively absorbs the visible radiation (between 0.4 and 0.7 μm) for its use in the photosynthesis. The cellular structure of living plants highly reflects radiation from the near infrared spectrum (0.7-1.1 μm).

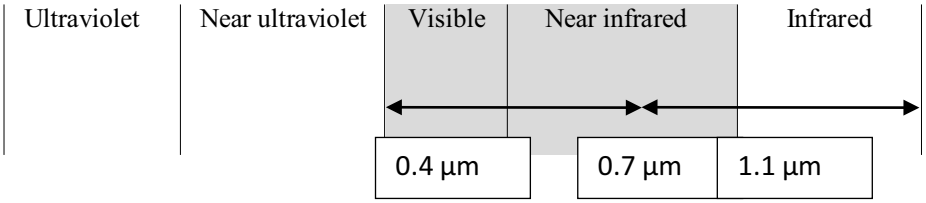

Figure. 1 The detection areas of the device corresponding to the visible absorption and to the near infrared reflection

The calculus formula of NDVI vegetation index is:

$$\mathrm{NDVI} = \frac{(\mathrm{NIR} - \mathrm{VIS})}{(\mathrm{NIR} + \mathrm{VIS})}$$

Where NIR and VIS are the reflectances obtained by the device for near and visible infrared.

SPAD-502 allows the rapid and easy measurement of the chlorophyll content of the plant leaves, without damaging the analyzed leaves. The measurement spectrum of chlorophyll concentration of SPAD-502 varies between 650 nm and 940 nm.

Once establishing the leaf's chlorophyll content, one can determine the optimal moment for applying additional fertilizer during the trees' vegetation. The chlorophyll content was measured in the southern part of each pear tree canopy with 3 repetitions of 30 determinations. The measurement point of chlorophyll was in the middle of the leaf on both its length and its width. Light reflection measurement was performed on the southern part of each canopy parallel to the chlorophyll measurement. The determination of the chlorophyll content for the studied genotypes of pear trees was performed separately on the leaves attacked by *Venturia pirina* and on those uninfected. The determination of the chlorophyll content was performed through 3 repetitions of 30 measurements for both types of leaves, namely: infected leaves and uninfected leaves.

RESULTS AND DISCUSSIONS

According to table 1 we can observe that out of the 10 studied genotypes, Fântânele 6 genotype has the green index with the lowest average value of 0.62, and the highest average value is to be found on Topârcea 13 genotype with 0.75.

The average refection index in infrared spectrum (NIR) has the lowest average value of 1.28 nm for Fântânele 6 genotype, as compared to Fântânele 5 genotype, with an average value of 7.92 nm.

The average reflection index of the visible spectrum (VIS) for the 10 studied genotypes of pear trees varies between 0.52 nm for Topârcea 12 genotype and 1.24 nm for Fântânele 9 genotype.

Table 1

The average vegetation indices determined for pear trees using Crop Circle ACS-430 devices

Running number	Name of genotype	NDVI	NIR (nm)	VIS (nm)
1.	Tilişca 3	0.74	4.13	0.59
2.	Fântânele 5	0.72	7.92	1.23
3.	Fântânele 6	0.62	1.28	0.27
4.	Sibiel 7	0.67	5.13	0.91
5.	Fântânele 8	0.67	4.14	0.76
6.	Fântânele 9	0.67	6.14	1.24
7.	Cisnădioara 10	0.67	3.13	0.57
8.	Topârcea 11	0.74	4.05	0.53
9.	Topârcea 12	0.71	3.20	0.52
10.	Topârcea 13	0.75	4.28	0.60

NDVI – vegetation index
NIR – infrared spectrum (nm)
VIS – visible spectrum (nm)
Nm – nanometer

The average content of chlorophyll in the leaves that were not attacked by *Venturia pirina* varies between 36.6 % at Fântânele 5 genotype and 43.73 mg/dm^3 at Fântânele 9 genotype. The infection with *Venturia pirina* makes the average chlorophyll percentage to decrease. Thus, at Fântânele 5 genotype is 33.96 mg/dm^3 and at Fântânele 9 genotype reaches a value of 42.00 mg/dm^3.

The ratio of the average percentage of chlorophyll of the attacked leaves and the intensity of *Venturia pirina* attack varies between 1.13 at Fântânele 5 genotype up until the value of 8.40 at Fântânele 9 genotype. Table 2 points out the fact that the ratio between the chlorophyll content in the leaves and the intensity of the scab has higher values when a small attack of the disease is registered on the leaves, and it decreases along with the intensification of the attack on the leaves.

Table 2

Correlation between the vegetation index and the intensity of *Venturia pirina* attack for some genotypes of pear trees

No.	Name of pear tree genotype	Average chlorophyll content in leaves that were not attacked (mg/dm^3)	Average chlorophyll content in attacked leaves (mg/dm^3)	Intensity of *Venturia pirina* attack %	Ratio of chlorophyll for attacked leaves /attack intensity
1.	Tilişca 3	40.10	37.96	30	1.26
2.	Fântânele 5	36.30	33.96	30	1.13
3.	Fântânele 6	39.06	37.8	5	7.56
4.	Sibiel 7	43.30	40.46	5	8.09
5.	Fântânele 8	39.16	37.20	10	3.72
6.	Fântânele 9	43.73	42.00	5	8.40
7.	Cisnădioara 10	40.53	40.03	5	8.00
8.	Topârcea 11	38.80	37.5	10	3.75
9.	Topârcea 12	41.43	36.2	30	1.20
10.	Topârcea 13	42.36	40.43	10	4.04

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ASPECTS OF THE INTERSPECIFIC RELATIONSHIPS BETWEEN KLOECKERA APICULATA AND SACCHAROMYCES CEREVISIAE var. ELLIPSOIDEUS YEASTS

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Victoria Leustean[4]

Key words: *alcoholic fermentation; inhibition period; mixted inoculation.*

ABSTRACT

We have studied during alcoholic fermentation, some aspects of the interspecific relationships between the apiculated yeasts belonging to the Kloeckera apiculata species and the elliptical yeasts belonging to Saccharomyces ellipsoideus species, isolated from Murfatlar vineyard, viticol centre Cernavoda. The two species yeasts were inoculated separately and mixedly, in sterile Pinot Gris must. At the mixted inoculation, an inhibition have been noticed, particulary for Saccharomyces ellipsoideus species. The inhibition was maximum, when the inoculation with Saccharomyces ellipsoideus species, is done when there is a maximum activity period of Kloeckera apiculata species. Mixed inoculation leads to the occuarence of two maximum in the fermentation process: the first maximum occurs due to the activity of the Kloeckera apiculata species and the second maximum occurs due to the activity of the Saccharomyces ellipsoideus species, after the passing of the inhibition period.

INTRODUCTION

In the wine microbiology majority of the researches are orientated upon the relationships between microorganisme species and from inside of the same species. All the time were studied the yeasts attend in the fermentation process (Beleniuc G., 2006; Castelli T., 1973; Gandini –1966), the yeasts influence upon lactique bacterias and viceversa (Ribereau-Gayon and Peynaud, 1960, 1961) the action of the acetic bacterias and moulds upon the yeasts and lactic bacterias (Ribereau-Gayon, 2000). Were studied the relationships between the mains yeasts groups, apiculate and elliptical (Domerq S., 1956). Some authors, have seen the negative role of the apiculate yeasts in alcoholic fermentation process and therefore even recommend their elimination from the must (CoteaD.V., 1985). The others authors, showed the main role of apiculate yeasts in the wine flavours formation. They showed that the famous wines, can not be obtained in exclusivity with elliptical yeasts, only by using the spontaneous microflora from the vineyard.

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MATERIAL AND METHOD

Were studied some aspects of the relationships between apiculate yeasts, belonging to Kloeckera apiculata species and elliptical yeasts, belonging to Saccharomyces ellipsoideus, isolated in the viticol Centre Medgidia, from Murfatlar vineyard, and identified by „Yeasts A taxonomic study, 6-th Revised and Enlarged Edition"[Kurtzman, C.P., şi J.W. Fell, 2006]. The researches were made, using like fermentation medium, Pinot gris sterile must with the following characteristics: 230 g/l sugars and 6,24 g/l H_2SO_4 total acidity. The experiments were made in seven variants, as following:

V_1 – inoculated only with Kloeckera apiculata species;

V_2 – inoculated only with Saccharomyces ellipsoideus species;

V_3 – inoculated simultaneous with Kloeckera apiculata and Saccharomyces ellipsoideus species;

V_4 – inoculated with Kloeckera apiculata species and after one day with Saccharomyces ellipsoideus species;

V_5 – inoculated with Kloeckera apiculata species and after 2 days with Saccharomyces ellipsoideus species;

V_6 – inoculated with Kloeckera apiculata species and after 4 days with Saccharomyces ellipsoideus species;

V_7 – inoculated with Kloeckera apiculata species and after 31 days with Saccharomyces ellipsoideus species.

The samples were keep at 25^0C temperature and was followed the fermentation process by daily yeasts population evolution (daily counting–Thoma mount) and the lost in weight registration, by CO_2 lost (g%). Finally, the wines obtained were analised from the chemical characteristics point of view, using the O.I.V methods and the Romanian standards in force.

RESULTS AND DISCUTIONS

The results are showed in the charts 1-7 and in the table 1.

Following the curves of the cells yeasts evolution and the lost in weight, for the sample inoculated with one yeasts species (V_1 and V_2), we can show (chart 1): Kloeckera apiculata yeasts has a big fermentative intensity in the first four-five days, and Saccharomyces ellipsoideus species in the first eight days.

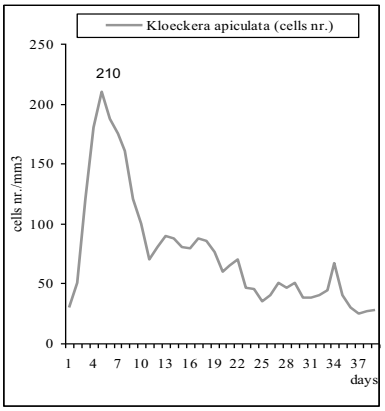

Chart 1. The cells number evolution at V1

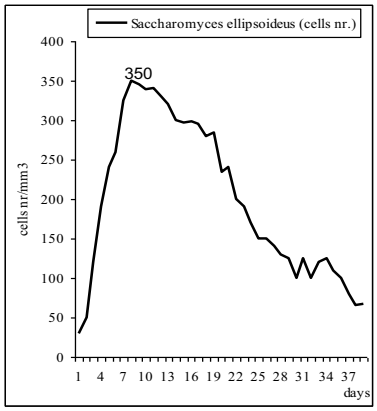

Chart 2. The cells number evolution at V2

After these periods, the fermentation has a low intensity. In the power fermentation period (1-5, 1-8 days) the number of yeasts cells are increase and, after this period, was hardly to establish with accuracy a correlation between total yeasts cells number/mm^3 and the fermentation intensity.When the samples were mixed inoculated (V_3-V_8), we have seen a mutual inhibition of two yeasts species, correlate with their inoculated moment. When the two yeasts species are simultaneous inoculated (V3, chart 3) was showed a mutual inhibition thus that, the Kloeckera apiculata species can not reach a big cells number like in V_1 (inoculated only with Kloeckera apiculata species).

To the other mixed variants inoculated (V_4-V_7) due the lag of inoculation with Saccharomyces ellipsoideus, the yeasts Kloeckera apiculata, achieve a number of cells/mm^3, approached V_1 (inoculated only with Kloeckera apiculata species). Only at the V_7 variant the Kloeckera number of cells/ mm^3, is the same as in V_1 (chart 7).

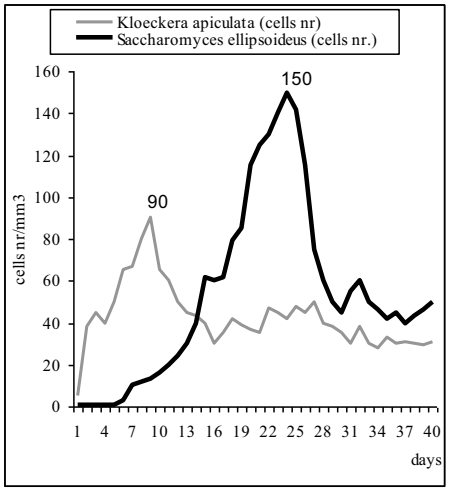

Chart 3 – The cells number evolution at V3

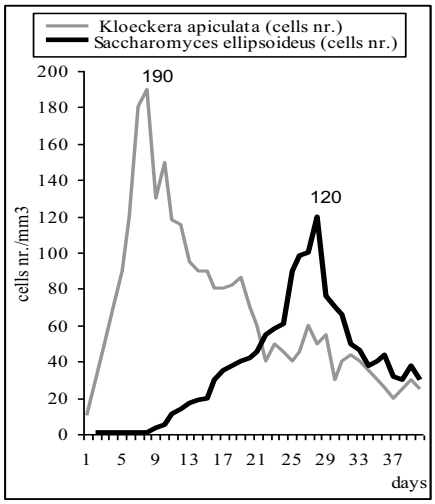

Chart 4 – The cells number evolution at V4

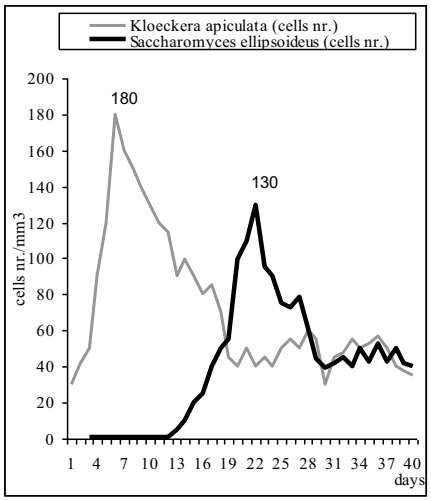

Chart 5 – The cells number evolution at V5

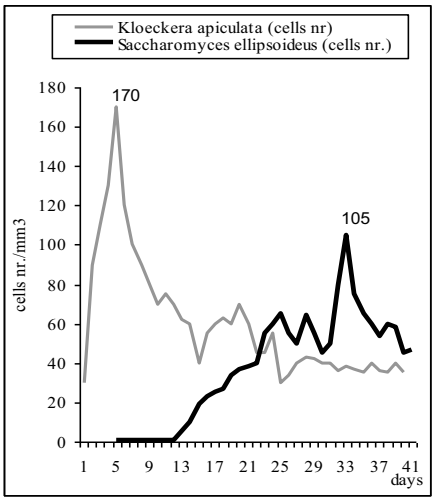

Chart 6 – The cells number evolution at V6

As the strain of Saccharomyces ellipsoideus behavior in mixed inoculated variants (V_3-V_7), there is an inhibition of its multiplication by Kloeckera apiculata, for a number of days, correlated with the lag time of both species yeast inoculation. This inhibition is: - almost 5 days for V_3; - almost 7 days to V_4; - almost 10 days for V_5; -almost 8 days for V_6 and almost 5 days for V_7.

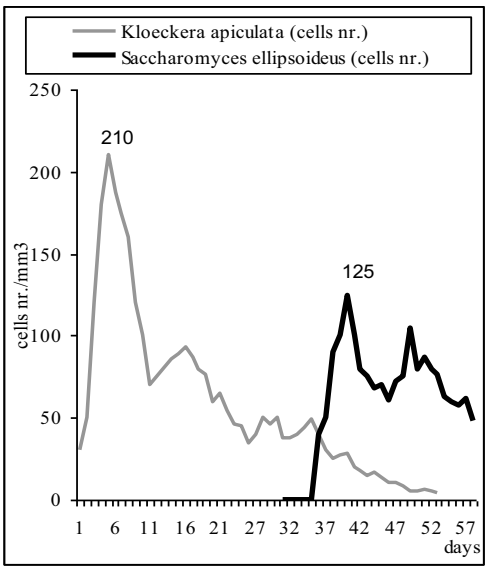

Chart 7 – The cells number evolution at V7

The lost in weight curves are different for variants mixed inoculated (V_3-V_7), comparatively with variants inoculated with one species of yeast (V_1 and V_2). If to V_1 and V_2, fermentation curve (CO_2 % release) recorded a single maximum (chart 1 a and chart 2a), the V_3-V_7 variants, it has two maximum (chart 3a, chart 4a, chart 5a, chart 6a, and chart 7a):

- the I-st maximum, came up in the 6-7 day fermentation and is the most species Kloeckera apiculata fermentation product, due to its intense activity in the first days of fermentation;
- the II-nd maximum was in the 26-27 days (V_3), the 31day (V_4), the 27 day (V_5), the 33 day for V_6 and 46 day (V_7) and is generated by Saccharomyces ellipsoideus, after passed the period of inhibition produced by species Kloeckera apiculata.

Table 1

The physico-chemical composition of the wines obtained

Sample	Sugar g/l	Alcohol % vol.	Total acidity g/l H_2SO_4	Volat. Acidity g/l H_2SO_4
Must	229,0	-	6,74	-
V_1	128,0	4,85	6,20	1,40
V_2	43,0	11,0	4,80	0,43
V_3	67,3	8,82	6,03	1,04
V_4	67,3	8,75	6,00	1,09
V_5	76,3	8,40	5,94	1,03
V_6	73,6	8,42	6,10	1,06
V_7	74,5	8,49	6,01	1,00
V_8	68,0	8,24	5,97	0,93

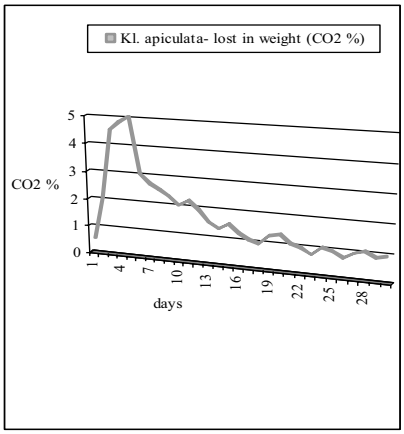

Chart 1a. - The lost in weight at V_1

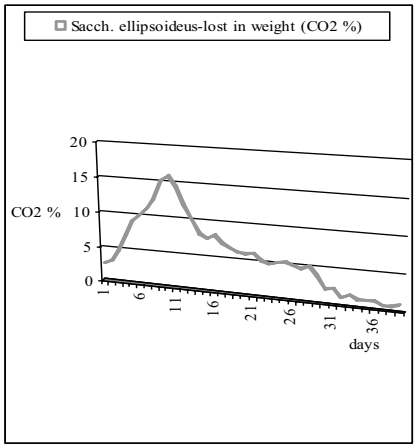

Chart 2a. - The lost in weight at V_2

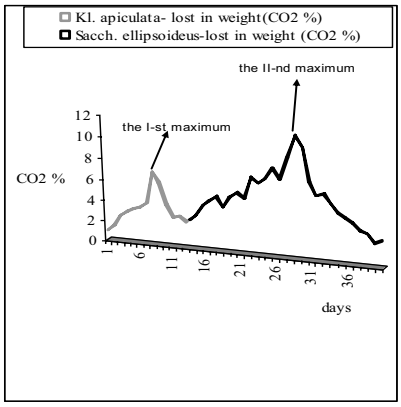

Chart 3a -The lost in weight at V_3

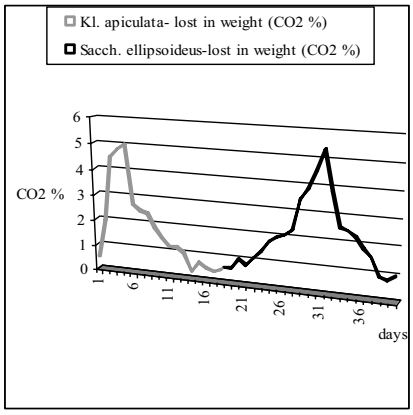

Chart 4a -The lost in weight at V_4

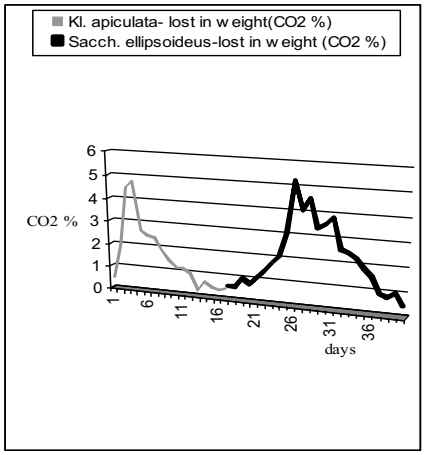

Chart 5a -The lost in weight at V_5

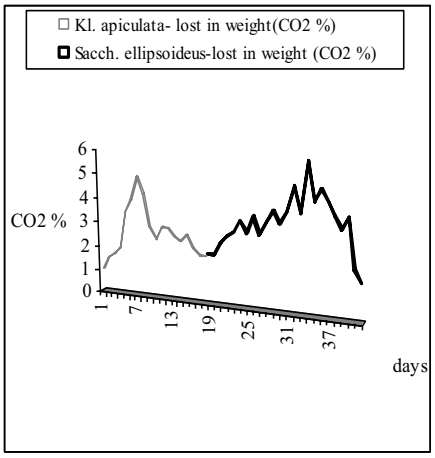

Chart 6a -The lost in weight at V_6

CONCLUSIONS

- The apiculate yeasts has the main role to the formation of wine flavours.
- The famous wines, can not be obtained in exclusivity with elliptical yeasts, but by using the spontaneous microflora from the vineyard.
- Kloeckera apiculata yeasts has a big fermentative intensity in the first four-five days, but Saccharomyces ellipsoideus species in the first eight days.
- In the power fermentation period (1-4, 1-8 days) the number of yeasts cells are increase and after this period was not possible to establish a correlation between total yeasts cells number /mm^3 and the fermentation intensity.
- When the samples were mixed inoculated (V_3-V_7), we have seen an mutual inhibition of two yeasts species, correlate with their inoculated moment.
- The lost in weight curves are different to the samples mixed inoculate (V_3-V_7) comparatively, with the samples inoculated with one yeasts species (V_1 and V_2).
- In conclusion, during alcoholic fermentation process, between the two species of yeasts have established negative relationship, of antagonism, which indicated that the Kloeckera apiculata species, by his activity of the metabolites produced in the fermentation medium, unfavorable to the Saccharomyces ellipsoideus species, inhibiting its development and activity for a specified number of days, correlated with their lag time of inoculation.

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RESEARCH ON PHYSICO-CHEMICAL AND SENSORIAL CHARACTERISTICS OF RED WINES FROM CORCOVA-MEHEDINTI

Bică Mircea[1]

Key-words: *Lubricity, color, intensity, fullness, smoothness*

ABSTRACT

During the period 2009-2011, there have been analyzed the physic-chemical and sensorial characteristics of red wines belonging to Cabernet Sauvignon, Merlot and Syrah, the latter newly introduced in culture.

By setting the values of wines, in the content of alcohol, of the level of total and volatile acidity, of the content in extract and glycerol, but also the presence of glucose and fructose, but especially of the chromatic characteristics of wines, we found that they can be part of the best quality red wines.

INTRODUCTION

The grape has in its constitution all the influences that natural factors and technique cultural factors have on the vine, but only wine is capable of sensorial expressing them and, thus, showing what should be analytically investigated in order to explain the generosity or deficiencies of natural conditions, human merits or faults (Teodorescu Ştefan et al. 1987; Popa Aurel 2008; Cotea D. Valeriu et al. 2009).

It is judiciously known that quality assets are usually realized within the vineyard. Therefore a quality wine could only be realized from a quality grape (Fregoni M., 1998, Morlat R., 2007). In its bacca, the grape does accumulate the compounds which a quality winw could not lack of. Amoung these compounds, sugars are of prime interest, together with organic acids, colouring substances and flavouring ones (Sequin G., 2007). Yet, for a given viticultural area, its vocation for quality could only be expressed by the wine itself only, understood as a complex structure able to define by itself a label of quality.

MATERIAL AND METHODS

Through these researches we have tried to prove that in Craiova-Mehedinti, the main production direction in viticulture must be getting high quality red wines.

For establishing the composition characteristics of the wines, we have used the methods approved and recommended by the O.I.V., and for catching the sensorial

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characteristics we have used the compensation system with 1 to 20 points. The organoleptic assessment was done by commissions of approved tasters.

RESULTS AND DISCUSSION

Wine quality is stated in the number and proportion of the present fixed and volatile components. For a high quality of the wine it is necessary that both the components number and the proportions, in which they are present, be as large as possible (Popa Aurel 1996; Stoian Viorel 2001).

From over 1000 known components that make up the wine, associated in a highly complex and inconsistent manner, some come from unchanged grapes, such as tartaric acid, malic acid, citric acid, carbohydrates, mineral substances, etc.; others are formed during alcoholic fermentation or other fermentation processes such as alcohols, lactic acid, succinic acid, etc.; anyways, other part is formed through non-fermentative processes and also through reactions that occur between incipient substances or between existing substances, as it is the case of the esters and acetals.

When referring to the alcohol concentration of the three wines (*Cabernet Sauvignon, Merlot, Syrah*) we notice (table 1) that it is high in all wines, over 12.9% alcohol volume.

Cabernet Sauvignon wines have an alcoholic potential between 12.5 and 13.8 vol% with a median of 12.9 vol% alcohol. *Merlot* wines have a higher alcoholic concentration, fluctuating between 13.2 and 14 vol% with a median of 13.6 vol% alcohol. *Syrah* wines also have a good alcoholic concentration, between 12.8 and 13.4 vol%.

This (high) level of alcohol content is a consequence of the fermentation of sugars accumulated in grapes, especially during the period of post-ripening. Of course, this is a consequence of favorable natural conditions, of breeds' potential and of the applied agro-technique.

High alcohol wines are powerful, generous. On the other hand, alcohol is the support for other constituents of wine.

Total acidity expressed in tartaric acid, determined in Corcova red wines is between 6g/l (*Merlot*) and 6.4 g/l at *Cabernet Sauvignon* and *Syrah*.

Total acidity is an important parameter for wine quality. The lack of acidity (below 4.5 g/l tartaric acid) makes the wines taste flat and do not resist to retention. The excess of acidity impressed a hard taste (harshness) and a lack of organoleptic harmony of the wine.

Volatile acidity determined at Corcova red wines, is very small; it fluctuates between 0.17 g/l (acetic acid) at *Cabernet Sauvignon* and 0.32 g/l (acetic acid) at *Syrah*. At *Merlot* wines there were determined 0.30 g/l (acetic acid).

Volatile acidity is formed as a barometer for the evolution of wine, its health, and the difficulties that are expected in wine storage.

In appreciating a wine, fixed acidity is of utmost importance and it includes the main fixed acids in wine (tartaric, malic, citric, galacturonic, succinic, lactic) and the inorganic acids present in wine as salts. Fixed acidity represents an index for wine quality and authenticity. The normal range for fixed acidity of wines is between 2.5 and 5.8 g/l (tartaric acid). Corcova wines have a good fixed acidity, are resistant to disease and are better preserved.

We find that real or actual (pH) acidity of Corcova red wines (Table 1) is between 3.32 (Syrah) and 3.45 (*Cabernet Sauvignon*). *Merlot* wines have a pH value of 3.35, so that Corcova red wines are prepared, in this regard, for safely running over a ripening and an ageing period ensuring itself well appreciated sensorial qualities.

Table 1

Physico-chemical characteristics of wines
(limits minimums, maximums and median) Corcova 2009-2011

Type of wine	Alcohol (vol%)	Total acidity (g/l tartaric acid)	pH	Volatile acidity (g/l acetic acid)	Residual sugar g/l	Non-reducing extract g/l	Malic acid (g/l)	Citric acid (g/l)	Glycerol g/l	Glucose (g/l)	*Fructose (g/l)*
Cabernet Sauvignon	12,5-13,8 12,9	6,0-6,8 6,4	3,58-3,69 3,45	0,15-0,24 0,17	1,1-1,2 1,1	29-36 31	0,4-1,7 1,0	2,3-2,9 2,5	7,6-9,2 8,7	2,2-3 2,2	*0,3-1,0* *0,9*
Merlot	13,2-14,0 13,6	5,8-6,7 6,0	3,30-3,57 3,35	0,29-0,30 0,30	1,8-2,8 2,1	28-33 28	0,5-1,2 1,0	2,2-3,6 2,3	8,7-9,0 8.7	1,9-2,3 2	*0,1-0,7* *0,6*
Syrah	*12,8-13,4* *12,9*	*6,0-6,6* *6,4*	*3,30-3,41* *3,32*	*0,26-0,34* *0,32*	*1,2-2,2* *1,5*	*28-33* *32*	*0,7-1,4* *1,1*	*2,2-3,8* *2,9*	*10-11,2* *11*	*0,3-0,8* *0,7*	*0-0,2* *0,1*

Table 2

The amount of phenolic compounds at different times of ripening of grapes (limits, minimums, maximums, median) Corcova 2009-2011

Wine breed	Vinicultural year	Full ripening			Phenolic ripening			*Technological ripening*		
		Antocians mg/Kg grape grains	Total poliphenols (mg/l galic acid)	Technological provision mg/l	Antocians mg/Kg grape grains	Total poliphenols (mg/l galic acid)	Technological provision mg/l	Antocians mg/Kg grape grains	Total poliphenols (mg/l galic acid)	*Technological provision mg/l*
Cabernet Sauvignon	2009-2012	1199-1246 1231	1420-1489 1456	649-668 650	1510-1596 1514	2140-2273 2256	821-830 827	1399-1432 1400	2110-2129 2120	*795-832* *830*
Merlot	2009-212	1199-1212 1204	1401-1459 1430	711-739 729	1514-1573 1562	2110-2124 2119	814-857 849	1392-1401 1399	1996-2028 2000	*919-931* *927*
Syrah	*2009-2012*	*1222-1250* *1242*	*1419-1459* *1444*	*811-840* *829*	*1591-1600* *1599*	*2107-2164* *2147*	*799-830* *825*	*1397-1408* *1400*	*2014-2070* *2045*	*899-951* *942*

It is known the fact that total wine extract is represented by the total fixed substances, wine components, which remain after the removal of volatile substances by evaporation under conventional and demanding conditions.

However, it is important to know the size of the non-reducing extract in wines. Non-reducing extract is deduced by subtracting from the total extract the amount of reducing sugars expressed per liter, minus 1g (representing the content in reducing pentose). Non-reducing extract provides the wines the quality of "fullness" or consistency. Non-reducing extract content of natural wines varies from 16 g/l to over 22 g/l depending on the quality category they belong to, on the breeds, but especially on the climate and soil conditions of the vinicultural area where the grape production was obtained.

Non-reducing extract content of Corcova red wines is extremely generous (Table 1), ranging between 32 g/l in Syrah wines and 31 g/l in Cabernet Sauvignon red wines.

The high content in non-reducing extract of Corcova red wines, justifies the fullness or consistency that these wines reveal through sensorial analysis, and especially their personality and authenticity.

The small content of malic acid of Corcova red wines shows that these wines have also known the malolactic fermentation.

The colour of Corcova dry red wines becomes less brightly red, as a consequence of the acidity decrease. The flavor also changes, new tastes are added and the wine character becomes stronger.

The presence of glycerol in Corcova red wines fluctuates between 7.6 and 11.2 g/l and influences the quality of wine by giving a velvet taste and the feeling of lubricity.

The colour of wines is an essential quality.

Colour characteristics assessment is based on the fact that, optically, the colour resulting from selective absorption of elementary radiations that form the solar spectrum (daylight), so the wine colour characterization is reduced to determining the absorption of light radiation.

For characterizing the colour of wine, the colour intensity (Ci) and the hue or tint of the colour (Tc) are determined.

Determining the colour characteristics of Corcova red wines, i.e. the colour intensity (Ci) and the colour tint (Tc), we notice (Table 2) that *Cabernet Sauvignon* wines have Ci values between 11.99 and 13.58.

The colour tint (Tc) varies from 0.28 to 0.33. Consequently, in sensorial analysis, there have been assessed with the highest grades 19.60 and 19.80 (1-20 points scoring system).

Merlot wines had Ci values ranging from 12.15 to 12.53; in sensorial analysis they got between 19.20 and 19.30 compensation points for olfacto-gustative qualities. The colour tint (Tc) of *Merlot* wines had values between 0.30 and 0.33.

Both the colour intensity and the colour tint are influenced in a great manner by the climatic conditions. Oscillations were observed from simple to double. In organoleptic assessment, *Syrah* wines were, however, compensated with 18.90 to 19 points.

CONCLUSIONS

Cabernet Sauvignon, Merlot, Syrah and Corcova red wines are endowed with a rich composition, have enough colour intensity and beautiful hue;

Of the three types of wine, *Cabernet Sauvignon* wines rank first in terms of quality, being followed by *Merlot* and *Syrah* wines;

Syrah wine has some inconstancy in terms of colour, so that it can be better valued as a sort, especially with *Merlot* wine;

Corcova red wines are temperamental, refined and generous, which is why it is appreciated at the highest level.

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GROWTH AND RIPENING PROCESS OF CORCOVA BLACK GRAPES

Bică Mircea[1]

Key-words: *full ripening, over ripening, absolute sugar*

ABSTRACT

In 2009-2011, we conducted research on how the black grapes for red wines, under the climatic conditions of Corcova-Mehedinti, go through stages of growth and ripening and the consequences. The results of the research highlighted the fact that at Corcova, Cabernet Sauvignon, Merlot and Syrah black wines go through stages of growth and ripening starting from late June until early October. During this period grapes accumulate in their grains enough sugars and colouring substances, so that after the winemaking process we achieve high quality red wines.

INTRODUCTION

Joining the genetical endowment of the grapes' kind, the paedo-climate circumstances from a given viticultural area are the determining events which decide of how the grapes' growth and maturation processes are carried on (Labruyere A., Schirmer R., Spurr M., 2006, Leeuwen C., 2000). Where the grapes are able to accumulate a lot of sugars, of colouring and flavouring substances, this reaching to a convenient level of organic acids, we are entitled to state that we are witnessing for a viticultural area with a vocation for quality (Morlat R., 1998, Oz Clarke, 2008).

MATERIAL AND METHODS

By monitoring the processes of growth and ripening of black grapes for red wines in Corcova-Mehedinti, we wanted to notice the behaviour of black grapes in the pedoclimatic conditions offered by this area and the calling of these types of grapes for obtaining high quality wines.

For capturing the growth and ripening stages, we have used the methods recommended by O.I.V. regarding the evolution of grain weight, the accumulation of sugar in the grain, and also the values of organic acids in the grape grain determining the total acidity expressed through tartaric acid.

Tracking these processes was done for *Cabernet Sauvignon, Merlot* and *Syrah* wines.

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RESULTS AND DISCUSSION

Depending of the vineyard area and on the evolution of the main climatic elements, grape varieties go through stages of growth and ripening differently (Teodorescu Ştefan et al., 1987; Popa Aurel, 2008; Tuţă Veronica, 2011; Ţâştea George, 2012).

If this process has favourable conditions (climate, technology), the types of grapes that have proven to better exploit the offered potential benefit of a normal growth of the grain and accumulates in the grain the most valuable chemical components (sugars, acids, colouring and aromatic substances, etc.) at very high levels (Popa Aurel, 2007; Munteanu Camelia, Nicolăescu, C., 2010).

Following the processes of growth and ripening of Corcova grapes, in 2009-2011, very important aspects are to be noticed (Table 1).

Herbaceous growth period runs from tying up the first flowers and fruits and varies from one type to another.

At Cabernet Sauvignon, 100 grain weight at the end of the herbaceous growth (10^{th} of August) is of 93.2 g and the sugar content reaches the level of 134 g/l whereas organic acids content reaches the maximum level of 14 g/l (tartaric acid).

Al merlot, the same period frequently ends on the 1^{st} of August, when the weight of 100 grains is of 91 g, the sugar content reaches 125 g/l and total acidity reaches 12.2 g/l (tartaric acid).

Under the same conditions, at Syrah grapes, herbaceous growth period ends also on the 1^{st} of August, but the weight of 100 grains is 111 g, the sugar content does not exceed 105 g (commonly 95 g) and total acidity reaches the level of 15 g/l (tartaric acid).

Herbaceous growth behaviour is determined by the genetic heritage of each grape type, the pedoclimatic conditions being the same.

Frequently, after the start of the first fruits period, the acidity content slowly decreases, and the relative content of sugar firstly increases and sometimes insignificantly decreases, then throughout the period of the first fruits, the increase of these constituents (sugars) takes place in a very high rate, especially the types with high capacity of accumulation in the grains.

The end of the first fruits period is considered to be when the acidity content decreases slowly and stays at the same level while the amount of sugar increases quite obvious.

The grains weight is continually growing, but at different rates, determined by the intervention of rainfall and temperature variations.

At Corcova, for *Cabernet Sauvignon*, the first fruits period begins on the 10^{th} of August and lasts until 30^{th} of August, when 100 grain weight varies from 11 to 123 g, the sugar content is between 170 and 180 g/l and total acidity has values between 8 and 10 g/l (tartaric acid).

Merlot grapes, also at Corcova, spend the first fruits period from the 1^{st} of August until 30^{th} of August when the 100 grain weight ranges between 135 and 137 g.

The sugar content varies at the end of the first fruits period between 189 and 193 g/l.

Under the same conditions, the newly introduced type, *Syrah*, spends the first fruits period between the 1^{st} of August and 20^{th} of August, when the grape grain accumulates between 137 and 140 g/l sugars and the acidity content varies from 10.5 to 11.3 g/l (tartaric acid).

Table 1

The process of ripening of Corcova grapes (2009 – 2011); limits minimums, maximums and median

Wine breed	Denomination	*Date*									
		1.08	10.08	20.08	30.08	5.09	10.09	15.09	20.09	25.09	*4.10*
Cabernet Sauvignon	100 grain weight (g)	68,5-87,6 86,4	90,6-95,0 93,2	90,4-117 90,1	110-123 121	119-127 122	121-130 128	129-136 130	130-13 133	136-140 137	*138-140* *135*
	Sugar (g/l)	95-115 110	130-140 134	153-167 157	170-180 176	188-193 190	190-201 200	205-212 209	215-220 216	218-225 217	*229-233* *230*
	Total acidity (g/l tartaric acid)	13-15 14	12-15 14	10-12 11	8-10 9	7-8,9 8	6,8-8 7	6,7-7,8 7	6,3-7,6 6,9	6,3-7,5 6,7	*5,0-5,4* *5,2*
Merlot	100 grain weight (g)	89-99,7 91	115-118 114	127-130 129	135-137 136	140-146 142	149-153 150	150-156 155	151-154 153	150-153 150	*147-150* *148*
	Sugar (g/l)	124-128 125	156-160 157	171-175 170	189-193 191	195-207 201	212-215 212	218-222 220	225-229 227	229-234 232	*230-236* *234*
	Total acidity (g/l tartaric acid)	11,5-13,6 12,2	10,5-11,4 11,0	9,1-9,7 9,5	6,9-7,5 7,1	6,8-7,5 7,1	6,3-7 6,8	6,4-6,6 6,5	5,9-6,1 6,0	5,5-6,1 5,8	*5,7-5,9* *5,7*
Syrah	100 grain weight (g)	113-116 111	115-118 116	129-131 129	141-146 143	150-167 159	171-179 178	187-191 189	190-202 196	180-202 185	*178-200* *183*
	Sugar (g/l)	96-105 95	120-127 124	137-140 138	143-147 144	151-154 153	159-167 160	171-175 172	179-185 182	180-192 190	*208-215* *212*
	Total acidity (g/l tartaric acid)	*14-15,2* *15*	*14,2-14,5* *14,3*	*10,5-11,3* *10,9*	*9,5-10,1* *10,9*	*8-8,5* *8,1*	*6,9-8* *7,7*	*6,5-7,8* *7,0*	*6,2-7,5* *6,0*	*5,9-6,9* *6,0*	*4,6-5,0* *4,7*

Table 2

Summary of results regarding the first fruits period of black grapes from Corcova (2009-2011)

Type	Period moment		Determining			Period duration (days)	*Accumulation rate of sugars g/day*
			Sugar	Acidity g/l (tartaric acid)	Weight of 100 grape grains (g)		
Cabernet Sauvignon	Start	10.08	134	14	93,2	20	*2,1*
	End	30.08	176	9	121		
Merlot	Start	1.08	125	12,2	91	30	*2,2*
	End	30.08	191	7,1	136		
Syrah	Start	1.08	95	15	111	20	*4,3*
	End	*20.08*	*138*	*10,9*	*12*		

Ripening period comes after the first fruits period. In this stage, the sugar content increases considerably, especially if the weather is warm, there is abundant light and the soil moisture is sufficient.

The obvious increase of sugars takes place this time due to a corresponding intensification of photosynthesis.

At Corcova, in 2009-2011, the ripening period of *Cabernet Sauvignon* grapes started from 30th of August until 25th of September, when full ripening is reached (grains weight is maximum, as well as absolute sugar weight).

At this point, 100 grain weight reaches 137 g, the sugar content is 217 g/l and the must acidity varied from 6.3 g/l to 7.5 g/l (tartaric acid).

Merlot grapes go through the ripening period from 30th of August until 15th of September, when the median weight of 100 grains is of 155 g, the relative sugar content ranges from 218 g to 222 g and total acidity from 6.4 to 6.6 g/l (tartaric acid).

Under the same natural and culture conditions, *Syrah* grapes go through the ripening period from 20th of August to 20th of September (full ripening moment) when 100 grains have the weight of 156 g, the sugar content reaches 182 g/l and total acidity of the must varies from 6.2 to 7.5 g/l (tartaric acid).

If grapes are kept on the grape vine after full ripening, they go through the so-called period of over-ripening (post-ripening).

In post-ripening period, there occurs a continuous increase of the relative content of sugar in grapes, but this increase is not a result of an accumulation of carbohydrates from the leaves (by biosynthesis), but the result of water evaporation from the grains.

In other words, there is a gradual concentration of juice from the grains, although they do not receive sugars anymore.

At the same time, absolute content of sugars (g/grains weight) decreases more or less depending on the grapes type, place and climatic conditions, due to the frazzle taking place in grains cells.

In over-ripening, the absolute content, as well as the relative one generally continues to decline, but sometimes it slowly increases, due to climatic factors.

Water evaporation from the grains and determining the acidity are much faster as the temperatures are higher.

In over-ripening stage of *Cabernet Sauvignon* grapes, sugar concentration may reach up to 233 g/l, acidity drops to 5 g/l (tartaric acid) and the weight of 100 grains also decreases to 140 g/l.

Under the same conditions, *Merlot* grapes accumulate in over-ripening period up to 236 g/l sugars, total acidity decreases to 5.7 g/l (tartaric acid) and the weight of 100 grains also decreases to 147 g/l.

Syrah grapes accumulate in over-ripening period up to 215 g/l, the must acidity reaches 4.6 g/l (tartaric acid) and the weight of 100 grains decreases to 178 g.

CONCLUSIONS

Under the pedoclimatic conditions of Corcova-Mehedinti, black grapes for red wines – *Cabernet Sauvignon, Merlot, Syrah* – go through stages of growth and ripening with good results, meaning that the accumulation of the main components in the grapes has values that ensure high quality wines, so popular on worldwide wine markets;

Of the three grape types, *Cabernet Sauvignon* is the one that accumulates enough sugars and colouring substances;

In order to find the optimum harvest time for grapes (technological ripening) there should be noticed the sugars content, the grapes weight, total acidity content, colouring or aromatic substances content.

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AGRICULTURAL BIOTECHNOLOGIES, BALANCE FACTOR FOR THE SUSTAINABLE DEVELOPMENT OF THE SOCIO-ECONOMIC SYSTEM

Bonciu Elena[1]

Key words : *Biotechnologies, agriculture, development, sustainability, safety*

ABSTRACT

This paper addresses the broad concept of agricultural biotechnologies in terms of their role in the sustainable development of the socio-economic system, but also in terms of questions posed mainly due to the lack of information or scientific ignorance. We hope that, in a certain extent, the work clarifies some safety aspects of the new scientific methods and substantiates that, because of their advanced knowledge and extremely careful control, plants and foods that are produced based on modern biotechnologies can be even more reliable than those produced by conventional breeding methods. As well as all biotechnologies, agricultural biotechnology can be effective only as part of an integrated plan for use, seen in terms of systemic ecology, which aims at maximizing material and energetic flows in parallel with environmental care.

INTRODUCTION

Agricultural technology is based on the domestication of wild plants to create the crops that we have come to depend upon. Humans invented agriculture approximately 10,000 years ago when they began to harvest and cultivate specific plants to produce food. The improved plant traits selected by early agriculturalists were transmitted genetically to succeeding generations of plants. For example, domestication of corn by prehistoric agriculturalists has modified the plant to such an extent that it hardly resembles the wild teosinte plants from which it was originally selected (Suslow et al. 2002).

For most of human history, plants and animals have been selectively bred to improve particular traits, such as yield, disease resistance and hardiness. The making of bread, wine and beer by microbial fermentation processes are age-old activities, documented in our historical development even as far back as Egyptian times. Archaeological evidence suggests that the early Romans recovered copper leached by bacteria from natural copper sulphide deposits (www.wikipedia.com).

The current biotechnological revolution is based on scientific knowledge, which is closely related to the current economic development. Most of the current industry is based on biochemical engineering processes, molecular biology, fermentation, and so on, which led to a considerable increase in the use of biotechnologies in various fields of economy (www.biotechnologies.com). The scope of biotechnologies is very high, but in particular, they apply to agriculture, speaking here of the so-called agricultural biotechnologies.

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In the practice of current breeding as well as modern genetic research, haploid generations are obtained by using two biotechnological method categories: one of them based on somatic embryogenesis (androgenesis, microsporogenesis and ginogenesis) and the other based on zygotic embryogenesis (zygotes produced by intergeneric crossing followed by the elimination of pollinic partner genome and "haploid embryo saving" by in vitro culture. By both method categories, one can achieve: fixation of useful genetic variability after the first recombination cycles (F1, F2, F3) by complete homozygosity in one generation; acceleration and increasing of selection efficiency (connection/phenotype); significant reduction (5-6 years) of period to obtain new varieties; varieties with double haploid origin are in accordance with EU diversity, uniformity and genetic stability criteria (Verzea and Raducanu, 2007).

MATERIAL AND METHODE

This is a documentation study. The source of documentation was the various national and international bibliography and different internet sites. The work clarifies some safety aspects of the new scientific methods and substantiates that, because of their advanced knowledge and extremely careful control, plants and foods that are produced based on modern biotechnologies can be even more reliable than those produced by conventional breeding methods. Usually, scientific claims about agricultural biotechnologies benefits for society are not accepted without criticism. Today, it is more realistic to see the development of a new technology as a result of a complex social system of interactions and decisions.

RESULTS AND DISCUSSION

Biotechnology is defined as a set of tools that uses living organisms (or parts of organisms) to make or modify a product, improve plants, trees or animals, or develop microorganisms for specific uses.

Modern biotechnology is a term adopted by international convention to refer to biotechnological techniques for the manipulation of genetic material and the fusion of cells beyond normal breeding barriers. The most obvious example is genetic engineering to create genetically modified/engineered organisms (GMOs/GEOs) through "transgenic technology" involving the insertion or deletion of genes (Berger, 2008).

The ability to manipulate living organisms at the genetic level is one of the principal tools of modern biotechnology. Although the aim of traditional biotechnology, such as selective breeding, was to develop new traits or enhance existing functions, new biotechnology allows sophisticated manipulation of the genes in plants and animals which encode for particular characteristics in a more direct, precise manner. Genetic engineering is capable of providing an organism with a specifically chosen, designed and desirable new ability or property.

Agricultural biotechnology is the term used in crop and livestock improvement through biotechnology tools. Biotechnology encompasses a number of tools and elements of conventional breeding techniques, microbiology, molecular genetics, biochemistry, plant physiology, and molecular biology. Agricultural biotechnologies mainly include directions as:

• The micro-reproduction of plants and animals, through genetic engineering techniques, somatic hybridization, selection, "in vitro" cultures, plant and animal cellular cultures can be obtained through the intensive replication of seeds, plantlets or animal selected lines;

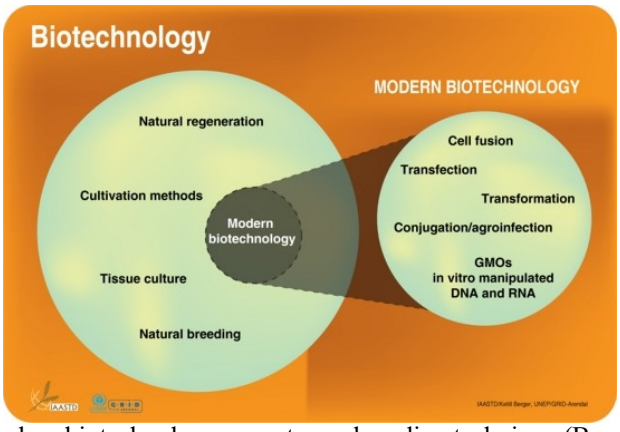

Fig. 1. Modern biotechnology, an extreme breeding technique (Berger, 2008)

• Plant and animal improvement, for obtaining highly productive lines, resistant to pests, diseases and extreme weather conditions. This direction includes the production of plants with increased rates of photosynthesis, plants resistant to cold or high temperatures, drought or increased soil salinity. We can also obtain animals with increased production of meat, milk, eggs, wool, etc. Efforts shall be made in the direction of obtaining nitrogen fixing plants other than legumes and crop plants that produce biocides (Ofiţeru, 2009);

• The transfer of embryos, in vitro fertilization and, most recently, cloning fall within the sphere of biotechnologies applied to animal bodies. The protection of animals through vaccination and treatment with synthetic biotechnological substances also contribute to increased production and economic efficiency.

Plant breeders have not exhausted the genetic resources available for crop improvement through traditional techniques, and recombinant DNA methods have now greatly expanded those potential resources. Transgenic crop varieties already on the market are providing value to farmers and consumers while reducing the use of agrochemicals. The fact that with biotechnology, foodstuffs can be produced faster, may mean for some consumers that they could be less tasty, which means that one would be more less inclined to indulge oneself with such a product. On the other hand, it could mean that the product would become cheaper, which could mean more efficiency in the management of the household purse (Hamstra and Smink, 1996).

Modern biotechnology is developing in this social atmosphere. Scientific claims about benefits for society are not accepted without criticism, that's why it is more realistic to see the development of a new technology as a result of a complex social system of interactions and decisions. However, the public opinion cannot be ignored.

Figure 3 shows that the public clearly distinguish between different applications of biotechnology. In terms of overall support, Europeans are neutral about agricultural biotechnology, and opposed to both GM foods and the cloning of animals. By contrast, and despite the opposition to GM foods, perceptions of medical biotechnologies (genetic testing, and the production of pharmaceuticals) and environmental biotechnologies (bio-remediation) are very positive. So the idea that the European public is anti biotechnology can be discounted (www.agbioforum.org).

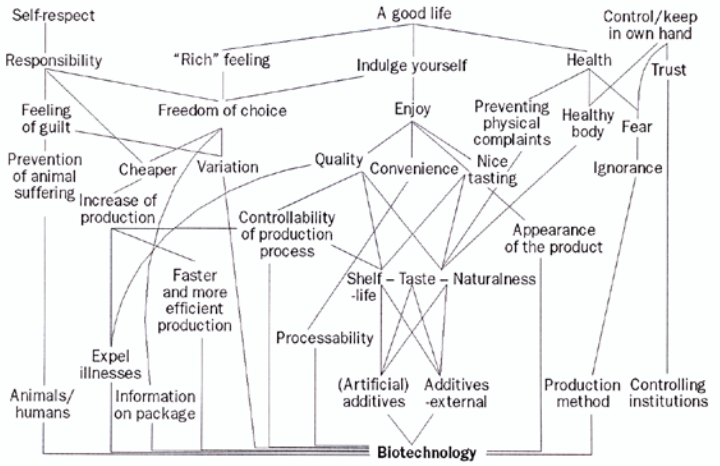

Fig. 2. Image of products, made with modern biotechnology (Hamstra and Smink, 1996)

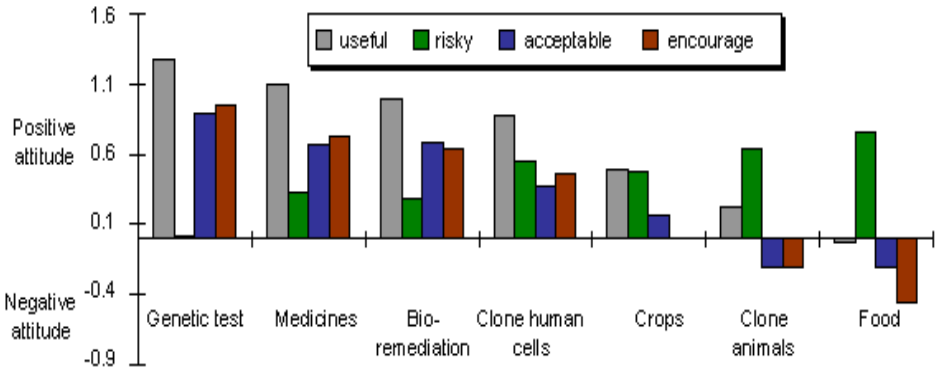

Fig. 3. European attitudes to seven applications of biotechnology (www.agbioforum.org)

In the same vein, here are a few myths about agricultural biotechnology, dismantled by the N. Hristea, in his work "Agricultural biotechnologies, myth and reality" (2009):

Myth: Modern biotechnology is inherently different from conventional plant breeding and involves more risks. Reality: Modern biotechnology represents an extreme breeding technique which has been used for thousands of years for plant breeding. The main difference is that modern biotechnology is more accurate and the range of characters that can be used to improve the performance of plants is much wider than conventional breeding. Many authoritative scientific bodies - including National Academies of Sciences - have reached the same conclusion, namely that crops improved by using modern biotechnologies are as safe as crops improved by conventional breeding methods.

Myth: Foods created by using biotechnologies contain genes, while improved plant-derived foods, traditionally, do not contain them. Reality: All whole foods contain genes that are broken down in the digestive process. Genes provide the instructions

necessary for plant growth and determine the characteristics of the plant and the type of food product. For thousands of years, mankind has changed the plant genetic map - most recently through modern biotechnology - to improve their characteristics. These changes have resulted in more resistant crops with higher yields and superior nutritional qualities.

Myth: Meat, milk and eggs from birds and animals fed with plants improved through biotechnologies are not as safe as similar products from livestock and poultry fed with fodder produced by conventional methods. Reality: Scientific evidence supports the safety of meat products, milk and eggs from animals fed with plants improved through biotechnology. Dr. Jimmy Clark, professor of animal breeding at the University of Illinois at Urbana-Champaign, reported the results of 23 studies on animals fed with plants improved through biotechnologies. These independent studies have found that forage plants improved through biotechnologies are as safe as crops improved through conventional methods.

Myth: Safety tests performed on foods produced through biotechnologies, realized or sponsored by modern biotechnology firms, is uncertain and serves their own purposes. Reality: The results of these tests are reviewed by scientific experts from appropriate regulatory agencies. These agency tests are very rigorous and operate as a parallel analysis. Prestigious independent scientific committees have also reviewed the scientific research on crops and food products based on biotechnologies and have not found any evidence that foods produced by using biotechnologies would be unsafe.

Animal feeding studies have been conducted as part of the toxicological assessments on different cultures improved through biotechnologies and they have not revealed any negative effects. For example, tomatoes produced through biotechnology have been fed to mice in a study conducted at the State Institute for the Quality Control of Agricultural Products in Wageningen, the Netherlands. Although the mice consumed the equivalent of 13 fresh tomatoes daily, no harmful effects were observed; a higher consumption would have been toxic due to the usual nutrients existing in this vegetable, such as potassium (Hristea, 2009).

Myth: Food products based on biotechnologies will introduce new allergenic substances in foods, exposing susceptible people at risk. Reality: All known food allergens are proteins, but a small number of proteins are allergenic. Common sources of food allergens include widely consumed foods, such as milk, eggs, wheat, fish, hazelnuts, peanuts and soybean. Biotechnology is also used by researchers to remove allergens from foods. Experimental rice has been modified through biotechnology to remove allergenic proteins, and other researches are under investigation for removing or neutralizing protein allergens from other foods, such as peanuts. The future development of allergen-free foods can expand the range of healthy foods available for those suffering from allergies.

Myth: Foods developed through biotechnology are not as nutritious as conventional foods which are achieved through breeding classical methods. Reality: An independent research has shown the nutritional composition of biotechnology-based products, being equivalent to that of conventional foods. An ongoing research explores ways to increase the nutrient content of foods using biotechnology methods. For example, researchers are trying to improve the content of antioxidants, vitamins and minerals from food and to improve the absorption and utilization of nutrients consumed from food. Foods with these advantages are still in the research phase, therefore it may take many years until they reach the market, but they present special advantages that are desired by consumers.

Food safety is the assurance that a food will not cause harm when it is prepared or eaten according to its intended use. The Food and Agriculture Organization (FAO) and the World Health Organization (WHO) of the United Nations advocate the concept of

'substantial equivalence' as the most practical approach to address the safety evaluation of foods or food components derived by modern biotechnology. This approach states that if a new food or food component is found to be substantially equivalent to an existing food or food component, it can be treated in the same manner with respect to safety. Researchers must prepare comprehensive data to support the safety and wholesomeness of new crop varieties developed through biotechnology. This process requires years of laboratory and field testing before a product can be brought to the market (www.fao.org).

The development of agricultural biotechnologies is dependent on continuing to break down barriers on a global basis, including addressing the regulatory environment and IP issues, promoting biotechnology acceptance and responding to the need for increased science and technological education and training (Niebur, 2009).

CONCLUSIONS

The benefits of agricultural biotechnology are becoming reality. In the future, biotechnology will lead to more obvious improvements in the nutritional profiles and other qualities of many foods. The technology itself holds enormous potential. Molecular breeding and transgenic crops will continue to play a key role in improving productivity in a sustainable development of the socio-economic system.

Because the features that are transferred by using modern agricultural biotechnologies are less numerous and even more predictable than those using hybridization, scientists better understand the changes that are induced and they are better able to assess the safety degree.

Public opinion has shaped and will continue to shape the social and political environment of modern biotechnology, and as such will have a determining influence on the trajectory of the technology itself.

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THE INFLUENCE ON THE CALLOGENESIS PROCESS OF WARM WATER TREATMENT APPLIED TO ROOTSTOCK CUTTINGS AND SCIONS BEFORE GRAFTING

Cristian Burlacu[1]

Keywords: *warm water, treatment, cuttings*

SUMMARY

The preliminary heating of the rootstock cuttings with warm water determined a more rapid start in vegetation of the grafted cuttings, depending on the duration of the treatment. Average productivity of STAS grafted vine varied from 52.1% to 65.6%. Distinctly significant positive differences (7.76 and 7.72%) were recorded for the variants 2 (rootstock cutting kept 10' in 45° warm water)and 4 (rootstock cutting wetted 6 hours 10' in 30° warm water). In the case of the determinations done on STAS vines regarding the matured length and the young cane width, the number of roots per vine and the way of joining between graft and rootstock, the values are close to the sample variant, no significant differences were recorded.

INTRODUCTION

Researches made so far in our country in the planting material production approached more or less all technological sequences involved in producing seedlings, but we face a low yield of obtained vines and problems about the grafted vines tehnical quality. Research carried out by Ophelia K. and collab. (1990), Moretti G. and F. Anaclerio (2000) and G. Moretti and collab. (2002) highlighted the positive influence of hot water treatment on the processes of callusing, rooting and increase of STAS vines yield.

The study represents the researches results conducted during 2005 - 2008, aiming to establish the extent to which the treatment of the rootstock cuttings and scions with warm water influence the process of callusing, rooting, twigs growing and STAS vine yield.

MATERIAL AND METHOD

Physical treatments applied to rootstock cuttings and scions consisted of the moistening in water heated to different temperatures:

- V_1 - rootstock cuttings wetted 24 hours (grafted eye and 36 hours (rootstock cuttings) in normal water at 12 ° C (sample control);
- V_2 - rootstock cuttings wetted 10 ' in water heated to 50 ° C;
- V_3 - ootstock cuttings wetted 3 hours in water heated to 45 ° C;
- V_4 - rootstock cuttings wetted 6 hours in water heated to 30 ° C.

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There were grafted 1000 cuttings for each variant from Feteasca regală variety using as rootstock the Berlandierii x Riparia sel. Crăciunel 2 variety. After forcing, the planting of grafted cuttings in school vine was done using the latin square scheme with 4 variations and 4 repetitions.

RESULTS AND DISCUSSION

The main results obtained highlight the following issues: preliminary wetting of the rootstock cuttings in warm water caused a faster start in vegetation of grafted cuttings, depending on the duration of the treatment (Table 1). It is found that the lowest number of days required to start growing the grafted eye was in the variant V_4 respectively after wetting rootstock cuttings for 6 hours at 30 ° C.

Table 1

Influence of physical treatments on the dynamics of starting in vegetation of vine shoots when forcing

Variants	Number of days to onset of growth restraint in:		
	1-5 % of cuttings	about 50% of cuttings	> 90% of cuttings
V_1	6	14	21
V_2	4	10	17
V_3	4	9	15
V_4	3	8	14

The situation is similar as regards the occurrence to the point of grafting of callusing (Table 2).

Table 2

Influence of physical treatments on the dynamics of callus occurrence at the point of grafting during forcing

Variants	Number of days until the appearance of callus forcing at:		
	1-5 % of cuttings	about 50% of cuttings	> 90% of cuttings
V_1	14	20	24
V_2	12	18	22
V_3	11	17	22
V_4	10	15	20

An upper callusing of sample (V_1) was achieved in V_2 and V_4 variants (Table 3):

Table 3

Influence of physical treatments on the callus formation of grafted cuttings after forcing in the grafting point

Variants	Cuttings with callus (%)		Cuttings without callus (%)	Callus on cutting weight (mg)
	diffuse	easily revealed		
V_1	71,6	24,4	4,0	615,2
V_2	79,0	20,2	0,8	685,2
V_3	64,0	19,6	16,4	492,0
V_4	82,4	17,2	0,4	646,6
Average	**74,2**	**20,4**	**5,4**	**606,5**

The percentage of cuttings with callus scattered is higher in variant V_4 as compared with other variants, this also having the lowest percentage of cuttings without callus.

Highest callus weight per cutting was recorded for V2 variant followed by V4 variant (Fig. 1).

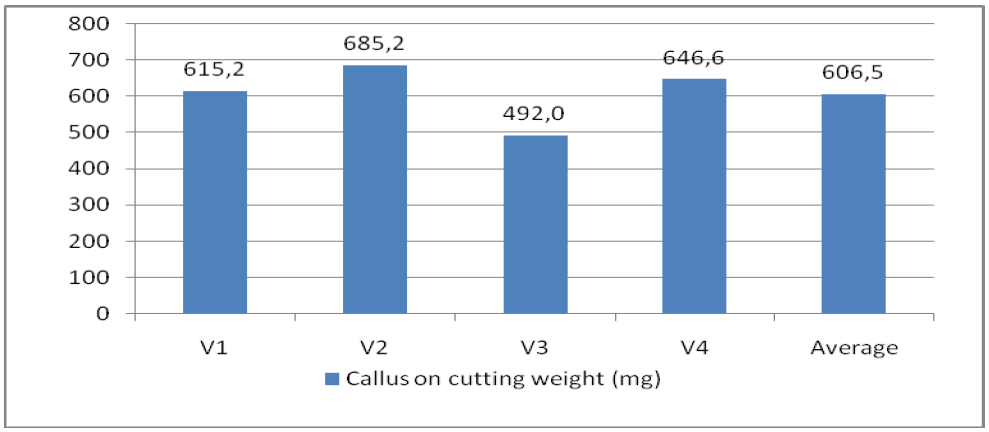

Figure1- Influence of physical treatments on the callus weight of grafted cuttings after forcing

Analyzing the influence of applied physical treatments on the process of callusing at the base of grafted cuttings it is found that V_4 variant had the highest percentage of cuttings with callus scattered (55,4%) and the highest callus weight per cutting, 590 mg (table 4).

Table 4

The influence of physical treatment on the callus formation process at the basis of grafted cuttings

Variants	Cuttings with callus (%)		Cuttings without callus (%)	Callus weight per cutting (mg)
	diffuse	easily revealed		
V_1	46,4	16,0	37,6	443,2
V_2	48,8	17,2	34,0	507,8
V_3	40,2	12,2	47,6	410,4
V_4	55,4	13,0	31,6	590,0
Average	**47,7**	**14,6**	**37,7**	**487,8**

These results confirmed that the treatment with warm water at 30 º C for 6 hours had a positive influence on the process of callusing. V_2 version, in which the rootstock cuttings were kept in water at 50 º C for 10 minutes, achieved a callus weight of 507,8 mg per cutting and had a percentage of cuttings with callus scattered of 48,8%.

The lowest percentage of cuttings without callus was of 31,6 recorded by V_4 version . V_3 variant had the lowest percentage of cuttings with scattered callus (40.2%) and

simultaneously the highest percentage of cuttings without callus (47,6%). Average scattered cuttings with callus was 47,7%, those with slightly pointed callus of 14,6% and 37,7% cuttings without callus. The average weight of callus per cutting was 487,8 mg (Fig. 2).

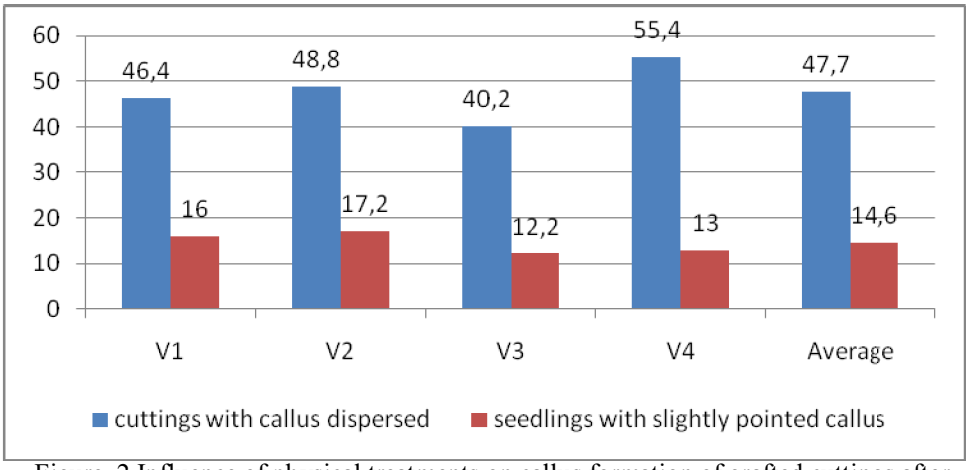

Figure. 2 Influence of physical treatments on callus formation of grafted cuttings after forcing

Analyzing the influence of physical treatments with warm water on the rooting of grafted cuttings (after forcing) shows that the V_4 variant has the highest percentage ofcuttings, with more then three roots (25,6%) and a root average weight of 265,6 mg/cuttings and a small percentage of cuttings without roots, of only 43,8%.

The V_2 variant, when the rootsock cuttings were kept in water at 50 ° C for 10 minutes, has the lowest percentage of cuttings without roots, but having a smaller percentage of cuttings with more then 3 roots (24,6%) and 268,6 mg/ cuttings average weight of roots (Table 5).

Table 5

Influence of physical treatments on the rooting degree of grafted cuttings (after forcing)

Variants	Number of cuttings (%)		Number of cuttings rootless (%)	The average weight of roots from a cutting (mg)
	over 3 roots / cutting	with 1-3 roots / cutting		
V_1	22,8	30,4	46,8	205,6
V_2	24,6	31,8	43,6	268,6
V_3	3,6	18,0	78,4	226,6
V_4	25,6	30,6	43,8	265,6
Average	**19,1**	**27,7**	**53,2**	**242,3**

The influence of physical treatment applied to the rootstock cuttings before grafting and forcing was monitored even in vine school in order to show any influence on STAS grafted vines quality and yield.

The obtained experimental data showed that for the starting in vegetation of vines in the greenhouse forcing, the influence of physical treatments applied to cuttings was observed in V_2 variant, when the rootstock cuttings were kept in 50 ° C water for 10 minutes. In this case, the starting in vegetation rate of grafted vines was of 87,4%, with

0,1% higher than V_4 version (rootstock cuttings kept in water for 6 hours at 30 ° C) and 3,5% more than the sample control (table 6). Regarding the main twig average length of growth was remarked V_2 variant (17,8 cm) with 0,7 cm more than the V_4 variant (17,1 cm) and 2,5 cm more as compared to the sample control (15,3 cm), (table 6).

Table 6

Influence of physical treatments on the starting in vegetation of vines in nursery

Variants	Rooted vegetation started (%)	The average length of main shoot growth (cm)
V_1	83,9	15,3
V_2	87,4	17,8
V_3	74,0	14,9
V_4	87,3	17,1
Average	**83,1**	**16,8**

Analyzing the obtained experimental data concerning the applied physical treatments influence on the vegetative development of vines grafted in vines school, it is found that the V_2 variant, when rootstock cuttings were wetted in 50 ° C water for 10 minutes, has a 83 4% rate of vines in vegetation, which have a main twig total length of 54.8 cm, with a 14.5 cm matured length. V_4 version has a 83.0% percentage of growing vines, with 52.9 cm main twig total length and 14.7 cm matured length (Table 7). V3 variant, in which rootstock cuttings were wetted in 45 ° C water for 3 hours showed the lowest percentage of vines in vegetation of 67.8%, compared with the sample control (V_1), which was of 79.7%. As regarding the main twig growth, the total twig length at V3 variant was 48.3 cm and matured length was 14.7 cm, as compared with the sample control, which recorded 49.7 cm total length and 13.3 cm matured length. The differences between versions highlighted the V_2 version which, compared with the sample control (V_1) presented a percentage of vines in vegetation higher with 4.6%, a main twig length higher with 5.1 cm and a matured length largest with 1.2 cm.

V_4 variant also showed differences from the sample control of 4.1% for vines which had started in vegetation, of 3.2 cm in case of the twig total lenght and of 1.4 cm for main twig matured length (table 7).

Table 7

Influence of physical treatments on growing of grafted vines in nursery

Variants	Rooted vegetation (%)	Main shoots	
		Overall length (cm)	Length matured (cm)
V_1	79,7	49,7	13,3
V_2	83,4	54,8	14,5
V_3	67,8	48,3	14,7
V_4	83,0	52,9	14,7
Average	**78,5**	**50,9**	**14,3**

The physical treatments applied to grafted cuttings had also effect on STAS vines yield. Analysis of variance for this parameter indicates that, compared with control variant

(V_1), V_2 and V_4 variants showed a highly significant positive difference and V_3 variant showed a distinct significant negative difference (Table 8).

Table 8

Influence of applied physical treatments on the yield of STAS vines

Variants	Average /variant	Differences	Significance	%
V_1(Sample)	57,82	-		100,00
V_2	65,58	+7,76	xxx	120,51
V_3	52,14	-5,68	00	84,98
V_4	65,54	+7,72	xxx	120,41

DL 5% = 1,7 DL 1% = 3,9 DL 0,15% = 7,4

CONCLUSIONS

The treatments that had a distinct significantly influence on STAS vines yield were those in which the treatment lasted 10 minutes at 50 ° C temperature (variant V_2) and 6 hours at 30 ° C temperature(variant V_4), at these variants being noticed:

- a positive influence on calogenessis and rootedness processes at cuttings grafted after forcing;
- a twigs better growth and maturation;
- an increased production exceeding 13% STAS grafted vines.

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GRAPES PRODUCTION AND ITS QUALITY DEPENDING ON VITICULTURAL AREAS AND VINE LOCATION ALONG THE HILL SLOPE

Laura Călugăru [1]

Key words: *slope, area, content, fine wines, balanced, features, times*

ABSTRACT

Research made during 2007-2009 concerning the grape production and its quality in two grapevine types (Sauvignon and Tamaioasa Romaneasca), grown in two winegrowing areas (Calina-Dragasani Hill and Bolindetu-Samburesti Hill) highlighted the fact that the biggest grape productions are performed at the hill base and smallest ones - in the upper third of the slope. In turn, the best quality of the grape crop is obtained in the upper third of the slope. In many years, the two types grown in the upper part of the slope take the grapes to the botrytisation process, which means wines will be obtained in the highest quality class (DCO-HGE). Relatively similar results were obtained in the middle third of the slope.

INTRODUCTION

At this time which the mankind are crossing, winegrowing around the world is in its final phase of going on to best quality products (grapes, must, wine, raisins, wine distilled products).

The decisive factor supporting this necessity lied in the delimitation of areas with a predisposition to quality winegrowing.

Scientific research in winegrowing and oenology managed to make available to winegrowers the criteria allowing them to plant grapevine in those areas where grapes – raw matter for wine – should be of best quality (Asselin C., Falcetti M.,1996, Axente D.2009, Dejeu L.,1986, Fregoni M.,1998, Genoiu T., Popa A. 2010, Guillaume Girard, 2010, Macici M.,2008, Morlat R.,2007, Popa A. and collab. 2007).

MATERIALS AND METHOD

In order to capture the quantitative and qualitative elements, we followed the wine production for Tamaioasa Romaneasca and Cabernet Sauvignon along the slope. Laboratory methods as per O.I.V. recommendations were used.

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RESULTS AND DISCUSSIONS

The outcomes obtained during the four years of research about the production of Sauvignon grapes and their main composition features (upon harvesting) in the Calina - Dragasani Hill and Bolindetu-Samburesti Hill are comprised in charts 1 and 2.

The Sauvignon type was brought from Sauternes-France, where it is grown together with the Semillon and Muscadelle types, which gives the famous Sauternes wine. Apparently, from the winegrowing areas in Romania where it was grown, Dragasani provides the opportunity for obtaining fine, balanced and full of character wines, with a high inclination to longevity. Not all winegrowing areas provide the necessary pedoclimatic conditions for the acquirement of high quality wines with a denomination of controlled origin, even when it comes to Dragasani ones.

The outcomes obtained and presented in table 1 show that Sauvignon in the Calina – Dragasani Hill provide constant grapes productions also in areas of exquisite quality. Thus, in the upper third part of the slope on Calina Hill, the grapes production varied between 7,010 kg/ha (in 2010 – a year with heavy rains during the growth and ripening of grapes) and 9,930 kg/ha (in 2007). In good winegrowing years, the content of sugars in grapes harvested from the upper third part of the slope reaches 242 g/l (in 2008), when grapes can be botrytised and fine wines can be obtained, much appreciated for their stoutness, fructuousity and discreet but persistent flavour. The high sugar content is also supported by the high acidity, which frequently exceeds 4.30 g /l (H_2SO_4). In the middle third part of the slope, the grapes' production is somewhat higher, reaching 10,050 kg/ha, but the sugar content is lower than in the upper third part of the slope, but even here it does not, however, drop below 228 g/l during good winegrowing years, and good quality wines can be obtained. During the heavy rain years (like 2010, for instance), the sugar content drops heavily and can reach 186 g/l; as a result, obtained wines can no longer be quality wines.

The biggest grapes productions are provided by the Sauvignon type in the lower third part of the slope, reaching 10,240 kg/ha (in 2007), but the sugar content drops even more, and sometimes drops below 212 g/l even during good winegrowing years, the result being wines lacking strong personality, the acidity content of grapes in the lower third part of the slope is high, frequently exceeding 4.59 g/l; together with the alcohol obtained by fermentation, this acidity no longer provides the inclination to longevity of wines.

Table 1.

Grapes production and their main composition features upon harvesting
Sauvignon type - Calina-Dragasani Hill

Determination	Grapes harvested from the											
	upper 1/3 part of the slope				Middle 1/3 part of the slope				Lower 1/3 part of the slope			
	Harvesting year											
	2007	2008	2009	2010	2007	2008	2009	2010	2007	2008	2009	2010
Grapes production (kg/ha)	9,930	9,820	9,730	7,010	1,0050	9,920	9,934	7,100	10,240	9,820	9,915	7,850
Sugars, g/l	236	242	228	196	228	232	228	186	212	220	218	185
Acidity, g/l	4.56	4.30	4.38	5.35	4.85	4.46	4.49	5.70	4.87	4.59	4.62	5.92

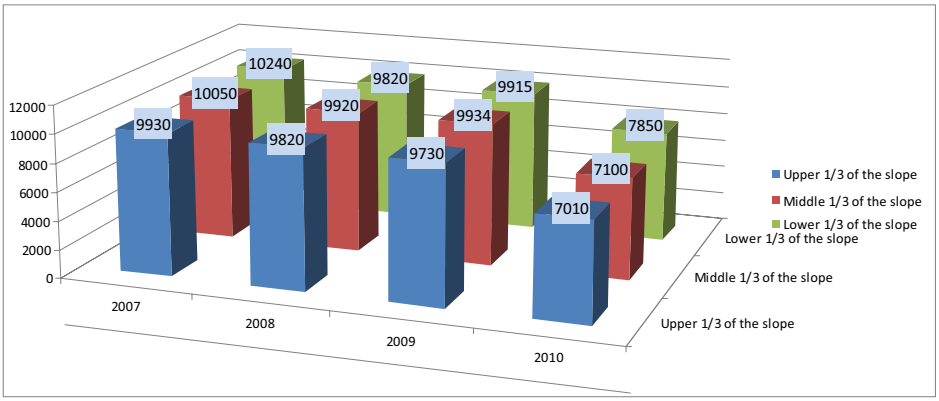

Fig. 1. Grapes production for Sauvignon- Calina-Dragasani Hill by years depending on the location on the slope

Table 2.

Production of grapes and their main composition features upon harvesting Sauvignon Type - Bolindetu-Samburesti Hill

Determination	Grapes harvested from the											
	upper 1/3 part of the slope				middle 1/3 part of the slope				lower 1/3 part of the slope			
	Harvesting year											
	2007	2008	2009	2010	2007	2008	2009	2010	2007	2008	2009	2010
Grapes production (kg/ha)	9,120	9,010	9,300	6,845	9,320	9,320	9,520	6,760	9,914	9,432	9,620	6,880
Sugars, g/l	232	245	229	193	224	232	226	188	216	218	216	180
Acidity, g/l	4.64	4.40	4.52	5.78	4.78	4.62	4.68	5.96	4.81	4.86	4.99	6.02

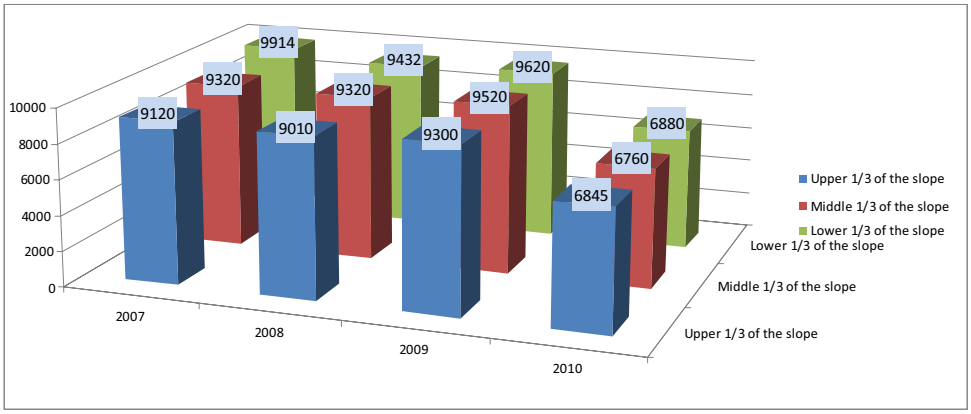

Fig 2. Grapes production for Sauvignon- Bolindetu-Samburesti Hill by years depending on the location on the slope

Hence, in the Calina-Dragasani Hill, best quality grapes are obtained in the upper third part of the slope (sometimes in the middle third part, also). The grapes botrytisation phenomenon may frequently occur in the upper third part, thus providing the opportunity to obtain exceptional sweet or licquorish wines.

In the second venue of the research - Bolindetu-Samburesti Hill, the Sauvignon type grapes production in the upper third of the slope reaches 9,300 kg/ha (2009), while the poorest production was recorded in 2010 (6,845 kg/ha), as a result of heavy rains recorded during July-October. In this area, also, the grapes sugar content reaches 245 g/l, with the specification of high acidity content (over 4.40 g/l H2SO4).

Similar outcomes were obtained in the middle third part of the slope, with slightly higher grapes production (9,520 kg/ha), but the sugar content does not drop below 224 g/l during good winegrowing years. In one year alone (2008) of the four considered during the research did the grapes gather 232 g/l. In this third part, the poorest grapes production with the lowest content in sugars of the grapes was in 2010, with 188 g /1. The highest harvest increments are recorded in the lower third part of the slope, as a result of high content in nutrients of the types of soil at the base of the slope. The sugar content of grapes in this area does not exceed 216 g /1, except in good years.

Considering the obtained outcomes, we can also emphasise that particular quality Sauvignon grapes can be grown in the upper third part of the slope in Bolindetu-Samburesti as well, with high content in sugars, with convenient acidity maintained. In the middle third part of the slope, only in some years, the grapes production is of the best quality. A rich production of grapes can frequently be obtained in the lower third part of the slope, however with low sugar content and higher acidity (6.02 g/l, H2SO4).

The first prizes and medals awarded to the Dragasani winegrowers were in relation to the Tamaioasa romaneasca wines. Even today we relish in remembering that the gold medal was awarded to the Tamaioasa romaneasca wine produced by Iordache N. Ionescu, at the Paris International Exhibit in 1887. Ever since, the Dragasani Tamaioasa romaneasca has represented the most illustrious ambassador of Romanian wines. Whether by accident or not, the Tamaioasa romaneasca type has been grown by winegrowers without considering the locations, which confer several valences to the type – from a pedoclimatic perspective, so that grapes and wines are of best quality.

Starting from this state of affairs, I have attempted, during the four years of research , to highlight the following as part of the two locations: Călina-Drăgăşani Hill and Bolindeţu-Sâmbureşti Hill: the area along the hill where grapes have the richest and the most complex chemical composition, based on which we can obtain wines of certain quality value, allowing them to be part of the great wines category.

In Călina-Drăgăşani Hill, in the upper third part of the slope (table 3), the Tamaioasa romaneasca type grapes production ranges between 6,430 kg/ha (in 2010 – poorest winegrowing year) and 8,480 kg/ha (2008 – the best winegrowing year). At the level of these recorded productions, when harvesting tok place after 15 October, the grapes sugar content in the years with good climate conditions ranges between 226 g/l (in 2009) and 248 g/l (in 2008). In two of the four years of research (2007 and 2008), the grapes dealt with the botrytisation phenomenon. In these sugars contents, acidity is kept within convenient ranges, between 4.65 g/l (in 2009) and 5.02 g/l (in 2007). The poorest wine harvest was recorded in 2010, with less favourable winegrowing conditions (6,430 kg/ha), as well as the lowest content in sugars (196 g/l).

Table 3.

Grapes production and their main composition features upon harvesting
Tamaioasa Romaneasca type - Calina-Dragasani Hill

Determination	Grapes harvested from											
	the upper 1/3 part of the slope				the middle 1/3 part of the slope				the lower 1/3 part of the slope			
	Harvesting year											
	2007	2008	2009	2010	2007	2008	2009	2010	2007	2008	2009	2010
Grapes production (kg/ha)	8,040	8,480	8,100	6,430	8,174	8,520	8,185	6,504	8,203	8,610	8,240	6,702
Sugars, g/l	230	248	226	196	226	236	221	187	218	228	214	180
Acidity, g/l	5.02	4.65	4.72	5.26	4.91	4.76	4.87	5.45	4.98	4.87	4.90	5.68
Index of flavoured substances, ml $Na_2S_2O_3$/100g Grapes	17.50	18.50	17.00	15.45	17.30	18.01	16.90	15.20	17.50	17.20	16.01	14.01

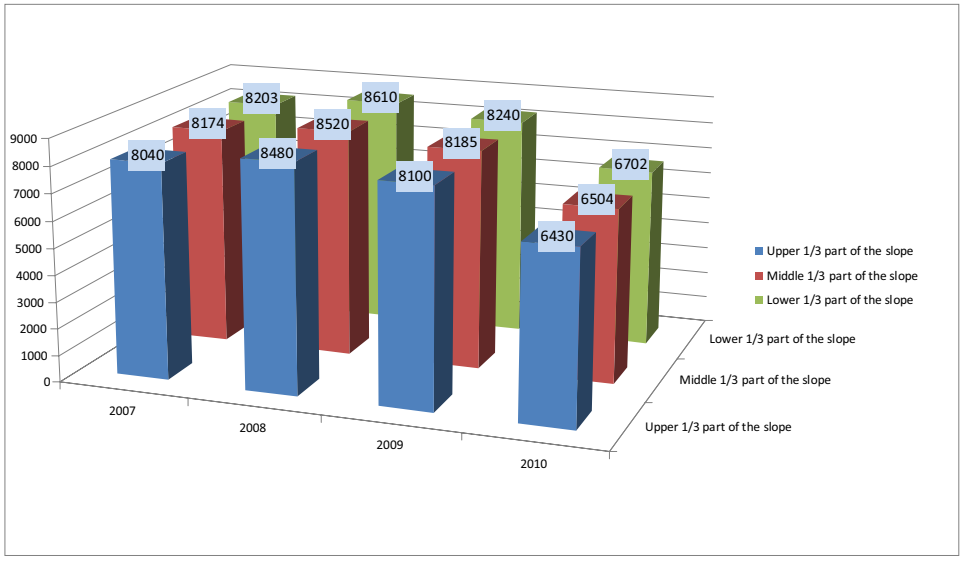

Fig. 3. Production of grapes for the Tamaioasa romaneasca type - Calina-Dragasani Hill by years depending on the location on the slope

Table 4.

Production of grapes and their main composition features upon harvesting
Tamaioasa romaneasca type - Bolindetu-Samburesti Hill

Determination	Grapes harvested from											
	the upper 1/3 part of the slope				the middle 1/3 part of the slope				the lower 1/3 part of the slope			
	Harvesting year											
	2007	2008	2009	2010	2007	2008	2009	2010	2007	2008	2009	2010
Production of grapes (kg/ha)	7,840	7,650	7,545	6,245	7,905	7,704	7,602	6,304	7,984	7,850	7,784	6,350
Sugars, g/l	228	236	219	186	216	226	212	180	209	216	209	179
Acidity, g/l	4.75	4.42	4.64	5.25	4.87	4.65	4.72	5.32	4.89	4.72	4.84	5.46
Index of flavoured substances, ml $Na_2S_2O_3$ /100g grapes	17.00	17.50	17.01	15.02	16.50	17.10	17.00	15.01	16.42	16.95	17.00	14.92

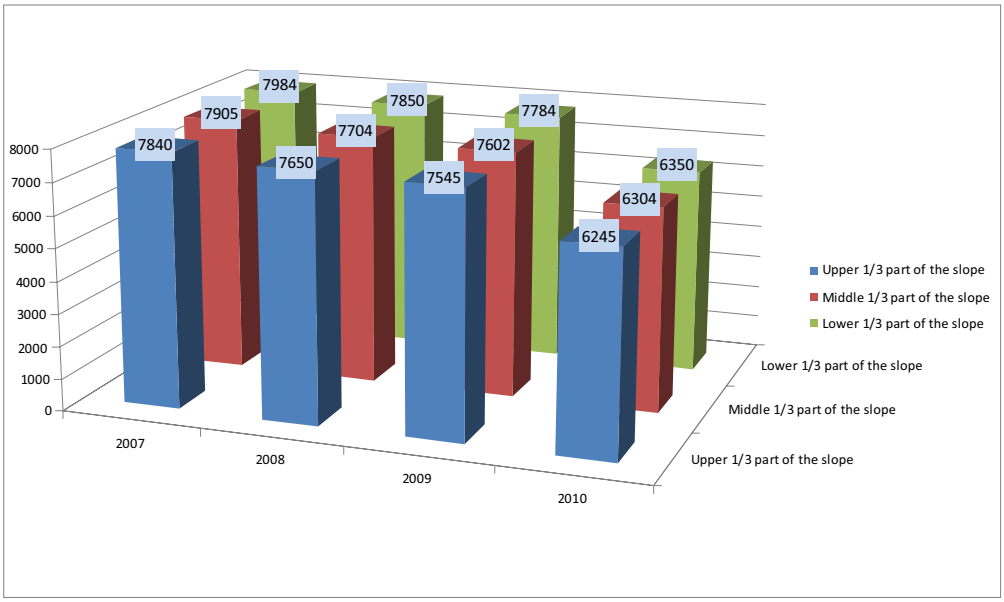

Fig 4. Production of grapes for the Tamaioasa romaneasca type - Bolindetu-Samburesti Hill by years depending on the location on the slope

As it is a flavoured type, one needs to take into account the aspect focusing on the gathering of flavoured substances. The obtained data highlight the fact that most flavours are accumulated in the upper third of the hill side, which shows the benefit the plant has from the richness of sun energy.

During the four years of research, the index of flavoured substances ranges between 17.0 (in 2009) and 18.50 (in 2008), while the lowest value was recorded in 2010 – 15.45.

In the middle third part of the slope, production is slightly richer, ranging between 6,504 kg/ha (in 2010) and 8,520 kg/ha (in 2008). The sugar content of grapes upon harvesting remains high, at values quite similar to those in the higher third part of the slope – in this area of the slope, the botrytisation phenomenon is rarer, and the flavour index is slightly lower than in the upper third part of the slope. The richest grapes production is recorded in the lower third part of the slope, reaching 8,610 kg/ha (in 2009). Even in the good winegrowing year (2008), the sugars content did not exceed 228 g/l, and the same direction is recorded in the flavour index, which ranges between 14.01 (in 2010) and 17.50 (in 2007). The acidity content of grapes upon harvesting stays within convenient ranges.

In the Bolindeţu-Sâmbureşti Hill area, the Tamaioasa romaneasca type has considerable contents in sugar and flavoured substances which clearly range lower than in the Călina-Drăgăşani Hill .

In the upper third part of the slope (table 4), the grapes production tanges between 6,245 kg/ha (in 2010) and 7,840 kg/ha (in 2007). The highest sugars content is 236 g/l (in 2008), and the lowest reaches 186 g/l (in 2010), while acidity does not drop below 4.42 g/l (2008). The highest value of the flavour index is recorded in 2008, and the lowest in 2010.

The grapes production in the middle third part of the slope comes across some enhancements, but the sugars content drops – the highest is 226 g/l (in 2008). In fact, the content in flavours is lower too – it reaches 17.1 in the best winegrowing year, compared to 17.50 in the upper third part of the slope (in 2008).

Wine production in the lower third part of the slope also rises but not considerably, as the top value recorded was 7,984 kg/ha (in 2007). The sugar content drops significantly – it barely reaches 216 g/l in the best winegrowing year (2008). In fact, flavoured substances also come in more modest quantities in this area.

Considering these outcomes, we can conclude that the Tamaioasa romaneasca type in Bolindeţu-Sâmbureşti provides sufficient satisfaction from a qualitative perspective. The botrytisation phenomenon was not recorded in this area.

CONCLUSIONS

The studied winegrowing area at Călina Hill allows for the Sauvignon grapes in the higher and middle third of the cliff to come across the Botrytisation phenomenon in some years, and thus it is possible to obtain wines with denomination of controlled origin harvested upon the grapes' enrichment or harvested late, in the worst case scenario.

At the Bolindeţu Sâmbureşti Hill, we find a similar situation to the one at the Călina Hill in the higher and middle third of the cliff; we can obtain superior quality wines with denomination of controlled origin, however harvested at full maturity.

The Tămâioasă românească specimen, harvested at the Călina-Drăgăşani Hill, in the higher and middle third part of the cliff, encounters optimal conditions for the development of the photosynthesis process, and it is possible to come across a Botrytisation phenomenon in some winegrowing years, and thus it is possible to obtain superior quality wines with denomination of controlled origin - harvested upon the grapes' enrichment or late harvest. The Tămâioasă românească grapes harvested from the lower third part of the cliff, can only generate superior quality wines with denomination of controlled origin-harvested at full maturity.

In the Bolindeţu-Sâmbureşti Hill, the Tămâioasă românească species grown in the higher third of the cliff allows for the production of superior quality wines with denomination of origin - late harvest. From the grapes harvested from the middle third of the cliff, one can obtain superior quality wines with denomination of controlled origin - late harvest or harvested at full maturity. The wines obtained from the grapes harvested from the lower third of the cliff can be qualitatively classified as superior quality with denomination of controlled origin - harvested at full maturity.

During al winegrowing years with abundant precipitations, during the grapes ripening period, no superior quality wines with denomination of controlled origin can be obtained, irrespective of the winegrowing area or of the cliff area.

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PHYSIOLOGIC AND BIOCHEMICAL ASPECTS DETERMINED ON WINE VARIETIES CULTIVATED IN DIFFERENT POSITIONS ON THE SLOPE ON DEALUL CALINA-DRAGASANI AND DEALUL BOLINDETU-SAMBURESTI

Laura Călugăru [1]

Key words: *slope, position, rate, transpiration, photosynthesis, respiration*

ABSTRACT

Research performed on 2 sites (Dragasani and Samburesti) concerning the physiological and biochemical activities present in 2 wine types (Tamaioasa romaneasca and Cabernet Sauvignon) cultivated on different slope positions, capture very interesting and useful phenomena related to the intensity of those processes in vine. Photosynthesis intensity decreases as we move from the upper to the lower third part of the slope. Leaves' respiration process registers a similar direction. Catalase activity is maximal in the Lower third of the slope and minimal in the upper one. Water quantity lost through transpiration is highest in the Lower third of the slope and lowest in the upper one.

INTRODUCTION

Across the viticulture world, including Romania, there are objective criteria set for quality vine areas delimitation (Carbonneau Alain, 2001; Condei Gh and collaborators, 2008; Constantinescu Gherasim, 1964; Falcetti M and collaborators, 1994; Popa A. and collaborators, 2007; Teodorescu St. and collaborators, 1987).

Our research aimed at highlighting the type of quality supply for two wine types (Tamaioasa Romaneasca and Cabernet Sauvignon) on the hill slope of two vine areas (Dealul Calina-Dragasani and Dealul Bolindetu-Samburesti).

MATERIALS AND METHOD

In order to capture the quantitative and qualitative elements, we followed the growth and maturation process of shoots, paying heed to the development of physiological and biochemical processes (photosynthesis, respiration, transpiration, catalase activity) in Tamaioasa Romaneasca and Cabernet Sauvignon grown along the slope. Laboratory methods as per O.I.V. recommendations were used.

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RESULTS AND DISCUSSIONS

The dry substance accumulations in Cabernet Sauvignon - Samburesti wine range between 7.0 mg DS (upper third) and 8.0 mg DS (lower third). The Cabernet Sauvignon - Dragasani photosynthesis intensity is higher than that recorded in the same variety in Samburesti, reaching values between 8.2 mg DS (upper third) and 8.7 mg DS (lower third).

Note that the intensity of photosynthesis decreases from the upper third (receiving more light) to bottom (receiving more moisture and soil fertility). Leaf respiration recorded a similar trend to that made by photosynthesis - values increase from top to bottom third in the studied varieties. Thus, the intensity of respiration showed values between 1.23 mg CO_2/dmp/hour (upper third) and 1.38 mg CO_2/dmp/hour (lower third) in the Tamaioasa romaneasca variety; between 1.20 mg CO_2/dmp/hour (upper third) and 1.30 mg CO_2/dmp/hour (lower third) for Cabernet Sauvignon Samburesti; between 1.26 mg CO_2/dmp/hour (upper third) and 1.39 mg CO_2/dmp/hour (lower third) in Cabernet Sauvignon Dragasani. The quantity of water waste through transpiration exceeds 1,000 mg/dmp/hour in the studied varieties.

Table 1

Main physiological and biochemical processes of vine, 5-10 August (2008-2009)

Physiological and biochemical processes	Slope Position								
	Upper third			Middle Third			Lower third		
Photosynthesis (mg DS/dmp/hour)	4.2	7.0	8.2	4.7	7.6	8.5	5.3	8.0	8.7
Respiration (mg CO_2/dmp/hour)	1.23	1.20	1.26	1.27	1.24	1.29	1.38	1.30	1.39
Transpiration (mg water/dmp/hour)	1147	1111	1128	1217	1224	1276	1272	1298	1317
Economic coeficient of transpiration (g water/ g DS)	273	159	137	258	161	150	240	162	151
Catalase Activity (ml $KmnO_4$)	0.9	1.0	1.2	1.0	1.7	2.0	1.2	2.6	3.0

The amount of water lost is the largest in the lower third and lowest in the upper third of the slope, and the direction is given by the soil water reserve available to grape vines-vine, in the phenophase in which determinations were made (first fruits of grape – 1st decade of August). Transpiration intensity in Cabernet Sauvignon Dragasani (with values between 1,128 mg water/dmp/hour in the upper third of the slope and 1,317 mg water/dmp/hour in the lower third of the slope) is higher than that obtained from Cabernet Sauvignon Samburesti (from 1,111 mg water/dmp/hour in the upper third to 1,298 mg water/dmp/hour in the lower third of the slope). The difference recorded between the two areas studied is neither large nor significant.

In the Tamaioasa romaneasca variety, transpiration recorded values generally higher than in Cabernet Sauvignon, ranging between 1,147 mg water/dmp/hour (upper third) and 1,272 mg water/dmp/hour (lower third).

Transpiration economic coefficient indicates the efficiency with which water is used and the nutrients absorbed from the soil, more specifically, the amount of water lost through transpiration so that, through photosynthesis, one unit DS/dmp/hour is achieved.

The data presented show that the values expressing the best use of water from transpiration are in the case of Cabernet Sauvignon from Dragasani, ranging between 137 (upper third) and 151 (lower third) , followed by those reached in the case of Samburesti Cabernet Sauvignon (with values from 159 – upper third to 162 - lower third). It is found that the water absorbed from the soil in Dragasani is better used by vine.

In the case of Tamaioasa Romaneasca, values range between 240 in the lower third and 273 in the upper third of the slope, this means that the vine had to absorb a larger amount of ground water in order to synthesize a unit of DM, compared with Cabernet Sauvignon.

The fact that photosynthesis in the lower third is greater than in the other slope positions is due to higher soil fertility, as well as to bigger soil moisture, which means that solar heat gain is lower. Similarly, one can understand the progress of transpiration and respiration along the slope.

Catalase activity as final redox enzyme confirms through the found values the respiration intensity progress along the slope. Catalase activity reached maximum values in the lower third and minimum in the upper third of the slope, as in the case of the respiration intensity. As shown in the table, catalase intensity values ranged from 0.9 ml to 1.2 ml $KMnO_4$ $KMnO_4$ for the Tamaioasa romaneasca variety, between 1.0 ml and 2.6 ml $KMnO_4$ for Samburesti Cabernet Sauvignon and between 2 ml and 3.0 ml $KMnO_4$ for Dragasani Cabernet Sauvignon.

Depending on the intensity with which the key physiological processes took place, in conjunction with climate and soil conditions according to the position on the slope, the maturation of shoots also occurs, expressed here by water content and carbohydrates of shoots (annual strings).

Table 2

Annual aging of wood for Tamaioasa romaneasca, Dragasani, (2008-2009)

Specification	Slope Position		
	1/3 upper	1/3 middle	1/3 lower
Aged wood water content (%)			
- shoots base	48.0	48.4	48.7
- shoots middle	48.2	48.5	48.7
- shoots top	48.6	48.9	49.5
Aged wood sugars content (g %)			
- shoots base	9.9	9.7	9.2
- shoots middle	10.4	10.1	9.6
- shoots top	9.6	9.4	8.8
Aged wood starch content (g %)			
- shoots base	8.2	8.1	7.6
- shoots middle	8.5	8.2	7.9
- shoots top	8.0	7.8	7.5
Specification	Slope Position		
	1/3 upper	1/3 middle	1/3 lower
Aged wood hydrates content (g %)			
- shoots base	18.1	17.8	16.8
- shoots middle	18.9	18.3	17.5
- shoots top	17.6	18.2	16.3

The water content of matured shoots in Tamaioasa romaneasca (Table no 2) rises from the base of the shoots to their top, and from the upper third to the lower third of the slope. Thus, the water content of shoots (string) in the shoots top ranged between 48.0% in the upper third and 49.5% in the lower third of the slope. Carbohydrates content is relevant for the assessment of the shoot (string) maturation level. Usually, fewer carbohydrates are associated with higher water content in strings. Carbohydrates content shows good maturation of the shoots on the string portions analyzed. The carbohydrate content of the Tamaioasa romaneasca variety ranged between 16.3% for the shoots top in the lower third of the slope and 18.9% for the mid-shoots in the upper third of the slope.

It is found that the best maturation of the shoots occurs in the middle portion of the shoots for the upper third of the slope.

Given the time of determination of the carbohydrates level (the first decade of November, 2008-2009), we find that the highest rate in the carbohydrates level is that of sugars compared to starch. The two found components (sugars and starch, as backup substances in shoots) progress similar to carbohydrates, however with differentiated values, depending on the string portion, the slope position and varieties reviewed. In Cabernet Sauvignon, the string water content has similar development to that in Tamaioasa romaneasca, however with slightly higher values (Table. 3, table. 4).

Table 3

Annual aging of wood for Cabernet Sauvignon, Dragasani, (2008-2009)

Specification	Slope Position		
	1/3 upper	1/3 middle	1/3 lower
Aged wood water content (%)			
- shoots base	48,.4	48.9	48.8
- shoots middle	48.7	48,7	49.2
- shoots top	48.9	48,9	49.4
Aged wood sugars content (g %)			
- shoots base	10,1	9,7	9.2
- shoots middle	10,6	10,0	9.6
- shoots top	10,0	9,6	8.9
Aged wood starch content (g %)			
- shoots base	7.8	7.7	7.5
- shoots middle	8.0	7.8	7.7
- shoots top	7.6	7.6	7.3
Aged wood carbohydrates content (g %)			
- shoots base	17,.9	17.4	16.1
- shoots middle	18.7	17.8	17.3
- shoots top	17.6	17.2	16.2

It should be noted that in the case of the variety located in Dragasani (table no. V.11), the water content in the string was slightly higher (with values between 48.4% for the shoots base in the upper third of the slope and 49.4% for the shoots-top of the lower third of the slope) than in the variety located in Samburesti (table V.2) (with values from 48.3% on the shoots-base of upper third to 49.3% for the shoots-top of the lower third of the slope).

Table 4

Annual aging of wood for Cabernet Sauvignon, Sâmbureşti (2008-2009)

Specification	Slope Position		
	1/3 upper	1/3 middle	1/3 lower
Aged wood water content (%)			
- shoots base	48.3	48.7	48.5
- shoots middle	48.5	48.6	48.9
- shoots top	48.6	48.7	49.4
Aged wood sugars content (g %)			
- shoots base	10.4	9.9	9.5
- shoots middle	10.8	10.2	9.8
- shoots top	10.0	9.7	9.1
Aged wood starch content (g %)			
- shoots base	8.0	7.8	7.7
- shoots middle	8.1	8.0	8.0
- shoots top	7.8	7.7	7.6
Aged wood hydrates content (g %)			
- shoots base	18.4	17.7	17.2
- shoots middle	18.9	18.2	17.8
- shoots top	17.8	17.4	16.7

Carbohydrates content rises in Samburesti Cabernet Sauvignon from 16.7% (shoots-top of the lower third of the slope) to 18.9% (mid-shoots in the upper third of the slope), and in Dragasani Cabernet Sauvignon from 16.2% (top of shoots in the lower third) to 18.6% (mid-shoots in the upper third of the slope).

CONCLUSIONS

. Viability of shoots was also influenced by the weather conditions recorded during the resting period of the grapevine and by the position of vines along the versant. It was greater in the cliff higher third, and towards the middle or the lower third of the cliff, viability drops gradually.

The total and maturated length of annual shoots increases from the higher third of the cliff towards the lower third of the cliff. However, mention needs to be made of the fact that, through calculation the maturated wood percentage from the total annual wood in the higher third is higher compared to that in the lower third.

The vine's leafy surface is maximal in the case of vines located in the lower third of the cliff and drops towards the middle of the cliff and the higher third of the cliff.

Irrespective of the soil, the vineyard where the species were studied, from a fertility and productivity viewpoint, it became obvious that in the higher third of the cliff, smaller values were identified compared to the other positions on the cliff; the weight of grapes is also smaller – all these in the context of smaller soil nutrient reserve and smaller water reserve available to vines in the maximum stress stages from a nutritional and water viewpoint.

. Intensity of photosynthesis decreases from the higher third (which benefits from more light) to the lower part (benefiting from more soil humidity and fertility).

. The progress of the leaves' breathing process registers a similar direction to that made by photosynthesis, namely, values increase from the higher third to the lower.

Through the established values, the activity of the catalysis confirms the evolution of breathing intensity along the versant. The catalysis activity reached maximum values at the lower third and minimum values at the higher third, similar to the breath intensity.

The quantity of water lost through transpiration is higher in the lower third and smallest at the higher third, which direction is given by the soil water reserve available to the grapevine vines at the phenophase during which findings were made (ripeness of grapes – 1st decade in August).

Depending on the level of intensity with which the main physiological processes took place, corroborated with the climate and soil conditions provided by the cliff position, the maturation of shoots also takes place, herein expressed through the water and carbon hydrates content of shoots (annual canes). A small carbon hydrate content is associated with high content of water in the canes. The carbon hydrates content shows very good maturation of the shoot on the analyzed canes places. The best maturation of the shoot is done in the middle place of the shoot and in the higher third.

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STUDY OF THE INFLUENCE OF TOBACCO LEAVES STORAGE ON THE CHEMICAL QUALITY OF CIGARETTES

Căpruciu Ramona[1]

Key words*: tobacco chemical composition, storage conditions*

ABSTRACT

This study aims to observe if the chemical characteristics of the tobacco leaves used in high quality cigarettes manufacturing are changing under different storage conditions. There is observed mainly the tobacco that is harvested and stored for a period of three months in order to perfect its qualitative characteristics. Two experiments were conducted on the stored tobacco leaves as follows: the first experiment was to determine the chemical elements of Virginia 180 and Djebel cultivar leaves kept under normal storage conditions (ventilation, optimal lighting, etc.) with an optimal level of humidity in the storage room. In the second experiment two bundles (carrots) of tobacco were kept under high humidity conditions (by watering the floor and walls during the month of August when the temperatures exceeded 35°C), and chemical determinations were carried out under these conditions, the obtained values being compared with the values of the first experiment.

INTRODUCTION

The smoking is usually widespread throughout the world and it is practiced equally by men and women, children and adults and affects the lives and health of millions of people (Nicolae Hodişan-2006).

The main chemical components of tobacco are: carbohydrates, proteins, alkaloids, polyphenols and colouring substances, organic acids and lipids, essential oils and resins (Giurgiulescu Liviu - 2002, 2006).

The nicotine content is higher under wet weather conditions, on heavier and colder soils, that are fertilized with nitrogen and it is lower under dry climate conditions and on soils with low nitrogen fertilization (after Constantin Bolcu-2007).

All the varieties of tobacco cultivated in our country are grouped into several types, each type containing qualitatively, biological and even morphological similar

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varieties. The types of tobacco are: oriental (Djebel), semi-oriental, Virginia, of high consumption and Burley (after Aniţia N. et al. -1993, Căpruciu Ramona-2008).

Researchers such as Giovino G.A. et al. 1995, J. Seglera et al. 2000, indicate through studies the harmfulness use of tobacco products (mainly cigarettes) on the human body in various stages of development.

Besides the manufacturing of the cigarettes, from tobacco leaves it is extracted the nicotinic acid (provitamin PP) used in the pharmaceutical industry, as well as the acetic acid.

MATERIAL AND METHOD

The tobacco used in the study is from the 2012 crop, the analyzed varieties were obtained in the natural growing area of Şimnicu de Sus.

The analyses were conducted on the strips belonging to Virginia 180 and Djebel cultivars. The stripping process consists of removing the tobacco ribs from the foliar limb by obtaining the parts of the leaves that were submitted to the analysis.

From the strips there were determined within the Research Base with Multiple Users - University of Craiova, the chemical characteristics of the analysed leaf according to the effective methodology.

The total reducing sugars (TRS) extracted from the tobacco sample, after inverting the sugar solution, are reduced under heating to a copper salt solution (Fehling's solution). The cuprous oxide resulted from the reaction is titrated indirectly with a solution of $KMnO_4$.

The nicotine extraction from the leaves of the analyzed cultivars was done by vapour stripping. A vacuum evaporation device was also used. For dosing there were used two methods for comparing the results: the method in aqueous environment (acidimetry), the sulphuric acid was used as an indicator to those transferred to neutral pH, and the spectrophotometric method: there were used the electromagnetic radiation in the UV region (185-400 nm).

The albumins are soluble in water and in dilute solutions of electrolytes (acids, bases, salts). The precipitation with neutral salts, sodium sulphate, in saturated solution was the used method.

RESULTS AND DISCUSSIONS

The two studied tobacco cultivars are part of the higher quality tobacco class. It is noteworthy to see how in different storage conditions, the chemical composition of the same cultivar changes, thus the storage condition becomes important in choosing the type of tobacco that is used in the recipe of manufacturing the end product (the cigarettes).

Virginia 180 and Djebel cultivars, kept under the two systems (high humidity and normal humidity conditions) were analyzed to determine the chemical characteristics represented by: total reducing sugars (TRS), albumins, nicotine, values expressed in percents % of the dry tobacco substance. Based on these values, the Kovalenko quality index could be calculated. The values obtained in the two experiments are shown in Tables 1 and 2.

Virginia type is the only type of tobacco that has the Superior (S) quality class, the other types of tobacco (also the Oriental type which includes Djebel cultivar) have the first class (I) of quality ranging until Scrap, refuse class (R).

In the two tables there is observed that both Virginia 180 and Djebel cultivars kept under normal humidity conditions have a chemically composition of superior quality compared with the tobacco leaves kept under high humidity conditions.

There is also noted that Virginia 180 cultivar has a lower content of nicotine, total nitrogen and total reducing sugars (TRS) than Djebel cultivar (table 1).

For both analyzed cultivars, the chemical content increases when the quality class decreases, this phenomenon is more clearly observed for the leaves stored under high humidity conditions.

Table 1

The chemical composition of Virginia 180 and Djebel cultivars stored under normal humidity conditions

Variety	Quality class	Total reducing sugars (%)	Total nitrogen (%)	Nicotine (%)
Virginia 180	S	5,49	2,48	1,21
	I	5,61	2,63	1,41
	II	6,00	2,71	1,52
Djebel	I	5,98	2,85	1,55
	II	6,45	2,96	1,62

Quality classes: S= Superior , I = 1st class, II = 2nd class

Table 2

The chemical composition of Virginia 180 and Djebel cultivars stored under high humidity conditions

Variety	Quality class	Total reducing sugars (%)	Total nitrogen (%)	Nicotine (%)
Virginia 180	S	5,68	2,53	1,23
	I	5,79	2,72	1,44
	II	6,05	2,78	1,52
Djebel	I	6,23	2,94	1,54
	II	6,99	3,02	1,67

Quality classes: S= Superior , I = 1st class, II = 2nd class

The heat and water from the foliar (leaf) tissue and those dispersed inside the storage determined an increased humidity above the optimum storage threshold of tobacco.

It can be seen (Table 2) that under these conditions (high humidity), the amount of total reducing sugars (TRS) decreases especially in the case of Virginia 180 cultivar. The reducing sugars have the role of producing an acid reaction to cigarette smoke during burning, leading to a pleasant, soft, slightly sweet, satisfying taste for smokers when there are obtained quantities up to 5-6%. If the percentage increases, the cigarettes will have a choking taste when they are smoked. This is observed especially at Djebel cultivar, kept under high humidity conditions for both 1st class of quality (6.23%), and especially for the second class of quality (6.99%).

The quantity of total nitrogen in the analyzed leaves is influenced by the nitrogen content of the soil. One can notice an increase in total nitrogen both for Virginia 180 and Djebel cultivars, with higher percentages, for storing them under high humidity conditions.

For the first quality class, Virginia 180 cultivar stored under normal conditions contains 2.63%, and under high humidity conditions it reaches 2.72%.

Comparing the two analyzed cultivars in terms of total nitrogen content, Djebel cultivar has a higher total nitrogen content for both quality classes in comparison to Virginia 180 (3.02% for 2nd quality class - Djebel cultivar compared to 2.96% for the 2nd quality class- Virginia 180 cultivar).

To underline the role of the storage conditions on the quality of tobacco there was also determined the albumin content. By comparing the two storage systems used for Virginia 180 and Djebel cultivars, it appears that the presence of humidity determines a higher albumin concentrations compared to the situation when they are kept under normal conditions (figure 1 and figure 2).

In terms of quality classes, the highest values of albumins are recorded for the lower classes of the studied cultivars (2nd class) in both storage systems, variations being still quite low (8.98% for Virginia 180 cultivar and 9.05% for Djebel cultivar).

However there are noted relatively high albumin values for all quality classes, which means that the land on which it was the tobacco crop was enriched with NPK complex that was above the necessary dose required for normal growth and development of the analyzed tobacco cultivars, this thing being also influenced by the drought during the leaves maturation period (in August).

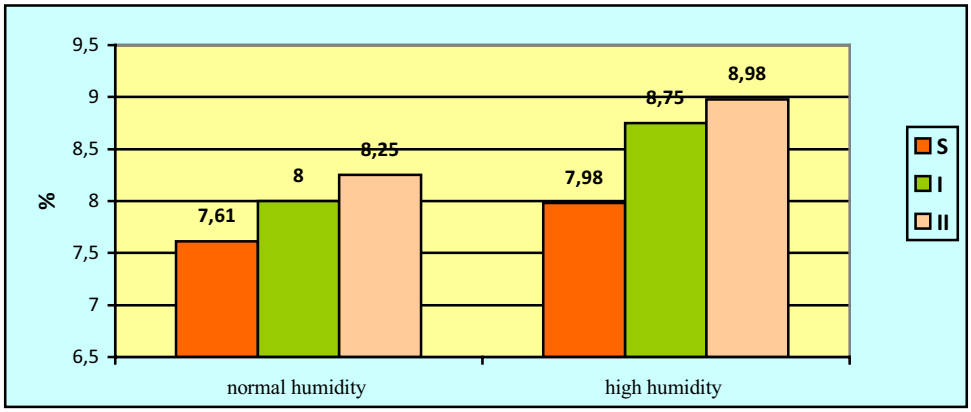

Figure 1 The albumin content of Virginia 180 cultivar depending on storage conditions

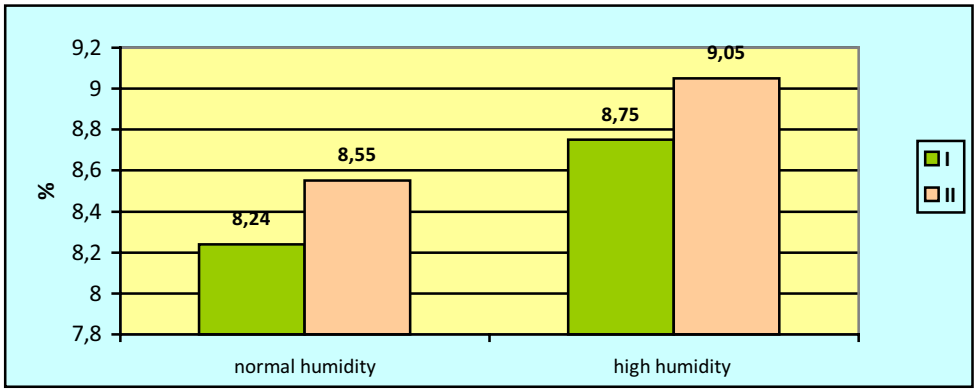

Figure 2 The albumin content of Djebel cultivar depending on storage conditions

For a meaningful assessment of the quality of tobacco kept under these conditions there was determined the Kovalenko quality index, specific for tobacco.

The Kovalenko quality index is the ratio of total reducing sugars and the difference between total nitrogen and nicotinic nitrogen. The higher the values of these indices are, the higher is the quality of the tobacco used in manufacturing (Tables 3 and 4).

Table 3

Kovalenko quality index evaluation for Virginia 180 cultivar

Variety	Storage conditions	The Kovalenko quality index		
		S	I	II
Virginia 180	normal humidity	5,50	4,25	5,04
	high humidity	4,60	4,05	4,00

Quality classes: S= Superior , I = 1st class, II = 2nd class

Table 4

Kovalenko quality index evaluation for Djebel cultivar

Variety	Storage conditions	The Kovalenko quality index	
		I	II
Djebel	normal humidity	4,60	4,51
	high humidity	4,45	4,17

Quality classes: S= Superior , I = 1st class, II = 2nd class

Analyzing Tables 3 and 4, there are observed high values for both analyzed cultivars, with higher values for the tobacco stored under normal conditions. In terms of quality classes the Superior class is determinant for Virginia 180 cultivar and at the 1st quality class, Djebel cultivar obtains a higher quality index (4.60 compared to 4.25 for Virginia 180 cultivar). For the 2nd quality class, the situation is reversed; Virginia 180 cultivar has a higher Kovalenko index than Djebel cultivar.

CONCLUSIONS

Comparing the data recorded for the two tobacco cultivars (Virginia 180 and Djebel) there is observed that the tobacco leaves stored under normal conditions of humidity have a chemical composition much better than the tobacco stored under high humidity conditions.

This implies a higher total reducing sugar for the leaves kept under normal conditions. For the 2nd class of quality, the sugar content decreased by half for both analyzed cultivars that were kept under high humidity conditions, correlated also with other factors.

Virginia 180 cultivar has a content of nicotine, total nitrogen and total reducing sugars (TRS) lower than Djebel cultivar, both kept under normal storage conditions and under high humidity conditions, which means that they are suitable for manufacturing high quality cigarettes.

The humidity from the storage space leads to higher albumin concentrations for the analyzed bundles. In terms of quality classes, the highest values of albumins are recorded at the lower classes for both storage systems, a phenomenon observed for Virginia 180 and for Djebel.

After calculating the Kovalenko quality index, it results that the obtained values emphasize the laboratory results in terms of the chemical parameters, namely: the tobacco kept under normal humidity conditions is chemically speaking superior to the tobacco stored under high humidity conditions.

Knowin the changing degree of the chemical parameters from the analyzed cultivars leaves under different conditions of storage, the following measures can be taken:

If we want a light, bright coloured tobacco (of bright yellow specific to Virginia type), with a specific smell and flavour, a balanced taste (a low degree of bitterness, a medium sweet taste), the humidity in the storage areas must not exceed the 20 -30% threshold;

On the contrary, if we need dark tobacco to produce cigarettes, with an intense bitter taste (a high content of albumins) then we can keep the leaves in the storage space at a higher humidity (50-70%) for a certain period of time.

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CLIMATIC CONDITIONS ARE THOSE THAT DECIDE THE QUALITY OF RED WINES FROM BOLINDEȚU – SÂMBUREȘTI

Chirca Ion, Luţu Florea [1]

Key-words: *vocation, amplitude, zoning, types, ferruginous*

ABSTRACT

Research conducted in 2009-2011 at Bolindeţu – Sâmbureşti has shown the fact that the climatic conditions they offer: amount of rainfall (682 mm per year and 424 mm during the growing season); the average sunshine (1784 hours/year and 1600 during the growing season); monthly average temperature 10,5^{0}C; relative air humidity (66,9%); the sum of temperature grades during the growing season (3226^{0}C); but also through the predominant soil types (Brown argilo iluvial) rich in iron oxides and chalk, outlines this area's vocation for obtaining high quality red wines.

INTRODUCTION

One of the "pearls" of Romania's viticulture is Samburesti vineyard, with its hills Bolindetu and Bolovanu, located on the left side of Olt River, somehow across Dragasani, slightly above it.

The name Samburesti makes one think about the red wines produced here, primarily about Cabernet Sauvignon wines as well as Pinot Noir, Merlot and Feteasca Neagra.

Samburesti Cabernet is distinguished from other wines produced from this bred for its extra force, its strength and firmness, for its great personality given by the wealth of its compounds that participate at forming the color, aroma and taste. There are wines that are associated to heavier dishes and steaks and that transmit long unique sensations.

Samburesti red wines, primarily Cabernet Sauvignon, complete their qualities through barrel maturation and bottle ageing. They are raw, even wild in the first months of life, the round off, they are finished under the biochemical transformations effect, and they are wines that impress all senses: sight, smell, taste and they require to be enjoyed, hard to forget.

The vocation for quality of this area was observed and enhanced by generations of wine-growers, but especially by the scientific research done here (Cimpoacă C., 1998; Condei Gh. et al., 2008; Genoiu T, Popa A., 2008; Popa A., Dicu C., 2010; Măcău I., 1994).

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MATERIAL AND METHODS

During 2009-2011 we conducted research in order to capture the climatic elements that favor high quality red wines at Bolindetu-Samburesti.

In order to capture the climatic conditions at Samburesti, there have been used the meteorological data recorded at Dragasani Station during 50 years and the last 3 years (2009-2011), as well as those recorded during the research by the local Expert weather station.

RESULTS AND DISCUSSION

Besides other factors, the climate can improve grapes' quality and thus wines' quality to the highest levels of appreciation, just as it could collapse it.

Referring to the chemical constituents of grapes, we should mention the special case of some of them of highly oenological importance (malic acid, aromatic substances, anthocyanin pigments) which through their relatively law proportion, though certain properties (volatility, oxidisability, respiratory combustion) through their peripheral position in the grain rind, are extremely vulnerable and exposed to the most significant risks of depreciation caused by certain manifestations of climate during grapes' ripening. (Popa A., 2008; 2010).

In tables 1-4 there are presented the main climatic elements (their manifestation) recorded at Bolindetu-Samburesti Hill.

Bolindetu-Samburesti viticultural area is located near the Sub Carpathians at latitude of $44^0$32' and an altitude of 285 m (Table 4).

The average annual temperature is $10,5^0$C. The amount of annual rainfall is about 682 mm. The amount of rainfall recorded during the active growing period does not exceed 395 mm. This marks a good water supply of the wine.

Monthly average rainfall distribution during the active growing period (Table 1) also preserves the same aspect of oenoclimatic favorability as in all the centers belonging to the A3 oenoclimatic area – hilly vineyard centers, represented by the continuous decrease of the rainfall proportions during the months of August, September and October, which creates more reliable conditions for ensuring a better health of the crop.

Monthly average duration of sunshine at Bolindetu-Samburesti has high values, thus providing the vine abundant heliothermal resources.

Consequently, the monthly average temperature is one of the most favorable for the vine. The hottest month, with the highest temperatures is July (21.5^0C).

In August, which is also hot, the monthly average temperature is around 21^0C. Further on, the average temperature decreases, reaching values of 16.6^0C in September and of 11^0C in October.

These climatic availabilities that Bolindetu-Samburesti viticultural area benefits from, allow the vine to achieve in grapes, at a large scale, the synthesis of any of the constituents, no matter how complex their chemical structure is. It is also noted that the abundance of these availabilities is not accompanied by climatic hardness causing damages to their constituent harmony, through loss and degradation. Thus, the average of high and low air temperatures in August and September (Table 2) indicate moderate values, even in July, the hottest month, the average of maximum temperatures does not exceed 27.4^0C and in August is not higher than 27.3^0C. Similarly, the absolute maximum temperature in

August is 36.4^0C. In September, both the maximum average and the absolute maximum temperature are even more moderate than in August.

The difference between the absolute maximum temperature and the absolute minimum temperature in August keeps quite low values.

The theoretical balance of soil water, during the growing period, calculated on the basis of the potential evapotranspiration shows a slight deficit. The values of the hydrothermal and hygrohydrothermal are, naturally, smaller.

The data concerning air relative humidity (Table 1) indicate values that prove the existence of a satisfying level.

Also, the deficit of air steam saturation (Table 3) marks moderate values, around 17.9 mb/cm^2.

Similarly, air relative humidity in August, at 1 p.m. slightly bears away from the values of 55%.

A general observation that can be drawn from the entire set of data is that in Bolindesti-Samburesti viticultural area there is distinguished a high proportion of insolation hours which can sustain a high level of the synthesis processes and other physiological processes.

The location of this area in a hilly region makes this heliothermal abundance not to have excessive character, so that in July, the hottest month of the year, the average air temperature is 21.3^0C and in August is 21^0C. Also, average air relative humidity in August is 65% and in August at 1 p.m., 55% is achieved.

The fact that the wealth of sunlight and heat resources is not excessive is undoubtedly confirmed by the generosity of Samburesti red wines obtained here and that have brought a well-deserved fame to this place.

So, through the climatic offer of this viticultural area, we have the chance of obtaining red wines that, through their composition and their olfactogustative characteristics, can occupy the leading places on the highest step of appreciation.

CONCLUSIONS

1. At Samburesti, Bolintetu area, there are the most favorable climatic conditions for the physiological-biochemical processes of vine to take place efficiently.

2. The climatic conditions firstly make Samburesti red wines to be distinguished through their extra force, their strength and firmness, through their great personality given by the wealth of the compounds that participate at forming the color, aroma and taste.

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Table 1

The main climatic elements and their size in Bolindeţu – Sâmbureşti viticultural area - average on 50 years (1950-2000) and during 2009-2011

Climatic element	Period (years)	Month							Annual
		April	May	June	July	August	September	October	
Average monthly rainfall (mm)	1950-2000	58	72	82	84	57	40	31	682
	2009-2011	31,8	66	67,4	76	62	56	42	636
Average duration of sunshine (hours)	1950-2000	194	242	267	304	287	232	188	1684
	2009-2011	170,8	223,9	249,9	285,9	270,6	216,1	133,9	1551
Average monthly air temperature (^{0}C)	1950-2000	11,4	16,2	19,4	27,3	27,0	16,6	11,0	10,5
	2009-2011	11,4	16,3	20,0	27,8	27,2	17,1	11,0	10,7
Relative air humidity (%)	1950-2000	66	6,5	68	64	65	68	69	73
	2009-2011								

Table 2

Average air temperatures – maximum and minimum – and of the absolute maximum and minimum in August and September recorded at Bolindeţu – Sâmbureşti viticultural area (1950-2000 – average)

Average temperatures									August The difference (Tx-Tn)
Maximum			Minimum						
July	August	September	August	September	Absolute maximum		Absolute minimum		
					Tx		Tn		
					August	September	August	September	
27,4	27,3	23,7	15,6	11,9	36,4	33,5	8,6	1,7	28

Table 3

Climatic characteristics of Bolinteţu – Sâmbureşti viticultural area during 50 years (1950-2000)

August 1 p.m.		Hydrothermal index	Hygro-hydrothermal index	Oenoclimatic aptitude
Relative air humidity %	Deficit – saturation with air steam (mb/cm^2)			
55	17,6	1,19	6,7	4627

Table 4

Geographical and climatic elements of Bolindeţu – Sâmbureşti viticultural area (1950-2000)

Latitude	Altitude	Annual average temperature (^{0}C)	Annual rainfall amount (mm)	Sum 1.IV – 30.X			Oenoclimatic aptitude A=T+I-(P-250)
				T	I	P	
				Temperature(^{0}C)	Insolation hours (hours)	Rainfall (mm)	
44,32	285	10,5	682	3226	1536	395	4627

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CHARACTERISTICS OF SOIL TYPES AT BOLINTEŢU – SÂMBUREŞTI, RESPONSIBLE FOR THE HIGH QUALITY OF RED WINES

Chirca Ion, Luţu Florea [1]

Key-words*: type, horizons, formations, clay, gravel*

ABSTRACT

In Samburesti vineyard there are present 6 types of soils: Brown clay pseudogleizate, Brown argilo-iluvial slightly pseudogleizate, Brown argilo-iluvial pseudogleizate with frame; Brown rodic pseudogleizate with frame, Brown red luvic; Brown argilo-iluvial rodic.

In our research there were found connections between the physico-chemical properties and the morphological properties on one hand and wines' quality on the other hand.

In the present paper there are presented the results obtained concerning the soil types and their characteristics.

INTRODUCTION

Besides the climatic factors, the edaphic factors influence the processes of growth and fruiting of the vine, the quantity as well as the production quality, plantation life, resistance to diseases and to weathering. The certain and complex influence of the soil is unfortunately still difficult to express oenologically, under analytical aspect.

To the greatest extent, the soil characteristics that influence the quality of wine are also closely related to or conditioned by the climate. Of great importance are the chemical properties, but mostly the physical properties of the soil and subsoil which adjust the water, air and temperature regime, the permeability and water retention power (Popa A., 2008; Condei Gh. et al., 2008; Teodorescu St. et al., 1987; Călugăru Laura, 2012; Dejeu Liviu, 2010; Urucu Iustin, 2011; Morlat, R., 2001).

In cold viticultural areas, gravel soils provide a good drainage of excess water and gently heat allowing through the heat retention a better ripening of grapes. Also, heavy rains that occur after the first fruits are less favourable if the soil is permeable and allows a rapid infiltration of water.

Well drained soils are a first major factor of quality, especially in cooler wine regions. Good drainage arises from two factors: the nature of soil (sand, gravel, etc.) and the slope (the bigger it is the easier water flows).

It contributes to rapid soil warming during the day. Drainage is very important not only in cooler regions, but also in others (certainly not in warm areas) in autumn and during

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ripening, preventing the installation of grey mold and favouring the noble mold, protecting plantations, with breeds for red wines or favoured wines.

It is sufficiently clear the soils' participation in defining the composition and quality of grapes and wine function that reaches the level of an obvious vocation to carry out certain valuable characteristics of quality.

\MATERIAL AND METHODS

Research was done on the vineyards of Bolindetu-Samburesti Hill, belonging to S.C. „Vitipomicola" Samburesti. Soil profiles were conducted at the ground of the slope, in the middle and on the plateau. There were studied pysico-chemical properties and morphological characteristics. In conducting the research we have been supported by OSPA Laboratory Slatina – Olt County, who used the methodology recommended by the National Soil Science from Romania.

RESULTS AND DISCUSSION

In Samburesti vineyard there are present 6 types of soils: Brown clay pseudogleizate, Brown argilo-iluvial slightly pseudogleizate, Brown argilo-iluvial pseudogleizate with frame; Brown rodic pseudogleizate with frame, Brown red luvic; Brown argilo-iluvial rodic. At Samburesti, the lithological substrate of soils is represented by the clayish Pleistocene terrace deposits and by terraces' gravel in the left of Olt River (Cotmeana Platform-Getic Piedmont).

Generally, at Samburesti, the profiles of quality oriented soils have the following sequence of genetic horizons:: Ao – Bt – Cca; Ao – Bt – C

Fig. 1. Argilo iluvial pseudorenzinic soil

Dominant soils at Samburesti vineyard (Table 1) are characterized by the fact that:

- they have a high content of clay, especially in AB and Bt1 horizons, so the permeability is very low, also appearing features of pseudogleying by rainwater stagnation due to poor internal drainage;
- they are heavy soils, compact, difficult to work with, having clayish texture and prismatic structure;
- they are medium supplied with humus, nitrogen, phosphorus and well supplied with potassium;

Table 1

Physico-chemical properties of soils at Samburesti vineyard

Soil type	Depth (cm)	Genetic horizon	Physico-chemical properties									
			N total %	P_2O_5 Total %	P_2O_5 mobile mg/100 g soil	K_2O access mg/100g soil	Humus %	pH	$CaCO_3$ %	SH ml %	SB ml %	V ml %
Brown luvic pseudogleizate	0-15	A0	0,08	0,10	6,4	9,2	2,029	4,55	-	8,25	10,9	60
	15-26	B1	0,07	0,07	2,2	5,3	1,168	4,83	-	8,25	16,05	66
	26-30	BB	0,05	0,06	2,0	5,1	1,230	4,82	-	10,7	19,36	66
	30-62	Btw1	0,03	0,05	1,8	4,2	1,230	5,76	-	8,25	18,99	70
	62-120	Btyw2	-	-	-	-	2,337	7,91	0,21	0,91	30,40	97
Brown argiloiluvial weakly pseudogleizate	0-20	A0	0,11	0,14	10,6	10,8	2,715	4,44	-	13,7	15,31	63
	20-45	Bt1	0,10	0,09	6,8	20,4	2,010	6,11	-	4,58	23,04	85
	45-90	Bt2	0,08	0,07	6,2	19,0	1,476	5,85	-	4,56	22,30	88
	90-120	Cca	-	-	-	-	0,615	8,21	6,94	0,91	-	-
Brown argiloiluvial pseudogleizate with frame	0-13	A0	0,07	0,12	18,2	12,8	0,988	6,12	-	4,58	12,74	73
	13-42	AB	0,05	0,14	10,5	7,2	0,615	6,98	-	0,91	14,57	94
	42-75	Btyw	0,04	0,06	0,8	16,2	0,123	8,33	1,05	0,91	31,13	97
	75-110	Cca	-	0,03	0,4	15,0	0,369	7,20	-	1,82	23,04	93
Brown rodic pseudogleizate with frame	0-18	A0	0,08	0,11	20,6	13,5	2,090	4,58	-	11,7	12,74	70
	18-52	AB	0,07	0,12	8,8	9,5	1,270	5,05	-	7,33	17,15	70
	52-92	At1	0,05	0,09	1,1	16,7	0,615	4,52	-	9,16	21,57	86
	92-105	At2	0,04	0,05	0,7	14,2	0,738	6,24	-	3,67	22,30	96
	105-130	Cc2	-	0,03	0,2	10,5	0,492	7,68	-	0,91	23,19	92
Brown red luvic	0-40	(A+B)d	0,06	0,10	9,3	14,0	1,845	4,92	-	4,58	12,74	73
	40-50	AB	0,05	0,12	10,1	12,8	2,465	5,29	-	5,49	12,74	70
	50-82	Bt1	0,03	0,09	6,5	9,2	0,868	5,51	-	5,49	17,15	76
	82-117	Bt2	0,02	0,07	4,7	8,3	0,492	5,26	0.42	5,49	17,52	76
Brown argilo iluvial rodic	0-20	A0	0,13	0,12	12,2	21,6	2,583	4,81	-	10,07	13,47	57
	20-40	A/B	0,10	0,09	8,8	9,5	1,942	5,37	-	4,58	16,41	78
	40-100	Bt	0,08	0,06	1,2	13,2	0,700	4,71	-	8,25	27,45	77
	100-125	Cca1	0,06	0,04	0,8	7,4	0,615	7,97	13,04	0,91	-	-
	125-150	Cca2	0,04	0,02	0,2	5,8	0,615	7,74	33,66	0,91	-	-

Table 2

Iron content, available potassium and humus of the soils from Samburesti vineyard

Soil type	Depth (cm)	Mobile Fe (ppm)		Available K_2O_4 - 100g soil	Humus
		HCl 0,1 n	Olson		%
Brown luvic pseudogleizate	0-15	1,2	0,2	9,2	2,029
	16-25	9,4	4,6	5,3	1,168
	26-30	3,2	0,35	5,1	1,230
	31-80	4,1	1,6	4,2	1,230
	-	-	-	-	-
Brown argilo iluvial weakly pseudogleizate	0-20	14,0	4,4	10,8	2,715
	20-45	21,8	7,0	20,4	2,010
	46-90	3,7	0,35	19,0	1,476
	91-120	1,5	0,22	-	0,615
	-	-	-	-	-
Brown argilo iluvial pseudogleizate with frame	0-10	10,4	3,1	12,8	0,988
	11-42	8,5	3,0	7,2	0,615
	41-75	0,7	0,2	16,2	0,123
	-	-	-	-	-
Brown rodic pseudogleizate with frame	0-10	16,2	5,6	13,5	2,090
	11-52	24,5	7,5	9,5	1,270
	53-90	6,7	2,6	16,7	0,615
	91-105	4,2	1,7	14,2	0,738
	106-130	0,3	0,18	10,5	0,492
Brown red luvic	0-20	9,2	3,0	14,0	1,845
	21-40	10,8	3,2	12,8	2,465
	41-70	7,7	2,8	9,2	0,868
	71-100	3,3	0,4	8,3	0,492
Brown argilo iluvial rodic	0-20	11,4	4,0	21,6	2,583
	21-40	9,4	3,1	9,5	1,942
	41-100	22,6	4,8	13,2	0,700
	101-125	10,4	3,2	7,4	0,615

- they are ferruginous soils with high content of Fe_2O_3 ranging from 7 to 30 ppm in Bt3;

- they are acid or moderately acid soils with pH ranging from 6.20 to 6.50 decreasing in depth;

- on Regosol soils situated on the top of the hills (Bolindetu and Bolovanu) there was found the existence of $CaCO_3$ (at 90 – 100 cm) quantifying between 13.40 and 33.66%, at Bolindeţu 22%.

Acid and moderately acid reaction of the soils from Samburesti vineyard is benefic for quality red wines, as they are wines with a high colour durability. A high content of iron stimulates the forming of chlorophyll and thereby it helps vine growing phenomenon and anthocyanin forming in black grapes' rind.

Our preliminary research has showed the fact that hydro physical indexes of the soils we studied, have very high values for the bleakness coefficient, field capacity has medium to high values and useful water capacity has low and very low values. Humus content in soil types decreases from surface to depth except for the Brown red luvic soil and Brown luvic pseudogleizate where there is recorded a slight increase in the profile (Table 1). The phosphorus content (P_2O_5) has normal values.

Potassium supply (K_2O_5) available on the first 20 cm presents a weak increasing scale for the following types of soil: Brown argilo iluvial pseudogleizate, Brown argilo iluvial rodic, Brown rodic pseudogleizate with frame. Soil reaction varies from strongly acid to acid or slightly acid.

The degree of base saturation is moderate for the following types of soil: Brown argilo iluvial rodic, Brown luvic podzolit, and the Brown red luvic soil the saturation degree is strong. The iron content is between 10.4 and 27.88 ppm (Table2), for the Brown argilo iluvial litic soil, the Brown argilo iluvial weakly pseudogleizate and between 22.6 and 24.5 ppm for the Brown argilo iluvial rodic soil and the Brown rodic pseudogleizate litic.

CONCLUSIONS

In Samburesti vineyard there are 6 (six) types of Brown soils: Brown clay pseudogleizate, Brown argilo-iluvial slightly pseudogleizate, Brown argilo-iluvial pseudogleizate with frame; Brown rodic pseudogleizate with frame, Brown red luvic; Brown argilo-iluvial rodic;

All soils types encountered at Samburesti are suitable for quality viticulture, especially for red wines of the highest quality;

Brown rodic pseudogleizate soil with frame is normally supplied with nutritive elements, disposing of a good drainage at the depth of 105 – 130 cm where $CaCO_3$ is present. As a consequence, grapes accumulate great sugars amounts (225 – 230 g/l), great quantities of coloured substances. Wines are balanced, fine, well coloured, with characteristic flavour, they are generous.

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RESEARCH ON ROOT SYSTEM DEVELOPMENT FOR THE APRICOT SPECIES ON SANDS, IN THE OLTENIA SOUTH

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Key words: *sand, radicular system, rootstock, variety, apricot.*

ABSTRACT

By studying the features of root system of trees, grafted on vegetative rootstock, we aimed to increase knowledge on development features of root systems, under the sands of the south and the ratio between the aerial and root system. A very important objective is adaptability of rootstocks on different soil types and their compatibility with all varieties. Sands may give to the grafted trees, the adaptability to climate and soil conditions, making the fruit earlier and obtain a quality high yield, year after year.

INTRODUCTION

A fruit tree species of great interest for the fruit growers' specialists is apricot. Primarily because the fruits have great food value, the positive effect they have in terms of health but also because of the products derived from apricot (nectar, jam, distilled, compotes etc.).

Of course, the productivity and the early growing of the species in relation with favorable natural conditions for apricot are necessary to be studied in different fruit growing centers in Romania. In this respect Baciu A., (2000) argues that the general trend is to diversify rootstocks in order to recovery soil and to control the vigor of plant-rootstocks association.

A more economical use of sands can be achieved by planning and cultivation methods, all these valuing the favorable characteristics of sands. Due to the influence of dunes and in between dunes on growth and fruitfulness of the trees, must be established the most suitable rootstock for the area but also for the main apricot zone varieties.

Popescu M., (1992) points out in his work *Fruit growth (general and specific)* that the engrafted apricot is the most suitable apricot rootstock regarding the compatibility, the productivity, and the root depth. Studying the behavior of several varieties on the engrafted apricot rootstocks, Cichi M., (2006) in his work *Root system development on different cultivars rootstocks combination to apricot,* mentions the very good behavior of the

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engrafted apricot on apricot varieties, air and root system growth. Also Sergiu Ancu and collaborators, (2009) highlights the Apricor vegetative rootstock that is compatible with apricot varieties.

MATERIALS AND METHODS

Apricots can be grown on irrigated sands but also on non irrigated sands in southern Oltenia. This can be done if you consider the planting method, the assortment, the protection against late frost in spring, before emergence of the premature death phenomenon on apricots.

The experience on the behavior of apricot on different rootstocks was located on Tâmburesti research station, on the sands. The layer of sand is over 10 m and the groundwater is located at depths varying from 7-9 m. The percentage of sand is 90%. Apricot tree was grafted on three rootstocks namely: engrafted apricot, peach and almond and the used variety was Dacia.

The plantation land was maintained black field and it was not irrigated. The crown shaped was laid flat dish, the experience being placed in 1998 at a planting distance of 4,5 / 4 m.

The view mode of horizontal root was achieved by applying the profile method. The increasing air and root system are observed in the seventh year of planting.

Generally in the process of the experiment, through the observations and measurements we followed:

- the main growth and fructification phenophases;
- the increasing of the air system and the ratio of grafted elements;
- the development of the root system.

Basically we try to observe the influence of rootstocks on growth and fructification of the apricot variety, of the behavior of the three rootstocks with Dacia variety, on sand. Botu I. and collaborators (2003), notes that each apricot variety requires a particular type of rootstock.

RESULTS AND DISCUSSIONS

On growth and fruiting phenophases we find that the start of unburgeoning occurs early on the Dacia variety grafted on almond and engrafted apricot (21-22. III), and on the peach rootstock occurs later (25. III), (Table 1).

Table 1

Completion the main growth and fruiting phenophases

No. crt.	Variant variety/rootstock	Start of unburgeoning	Start of blooming	Ripening fruit	Fall leaves
1.	Dacia/engrafted apricot	22.III	3.IV	5.VII	1.XI
2.	Dacia/peach	25.III	6.IV	7.VII	1.XI
3.	Dacia/almond	21.III	2.IV	5.VII	27.X

We note that even in terms of flowering, flowering occurs earlier in the case of grafting on almond and engrafted apricot rootstocks (2-3.IV) and flowering occurs on peach rootstock after 3-4 days (6.IV).

Fruit ripening is earlier on almond and engrafted apricot rootstocks (5.VII) and two days later (7.VII) on peach rootstock.

On the first day of November fall leaves in the Dacia variety grafted on peach and engrafted apricot rootstocks and earlier (27.X) in the same variety grafted on almond.

Table 2

The increase of air system and the ratio of grafted elements

No. crt.	Variant variety/rootstock	The height of the tree (m)	The crown diameter (m)	The amount of vegetative growth (cm)	The increase in thickness ratio: rootstock/graft
1.	Dacia/engrafted apricot	5.5	4.5	180	1.00
2.	Dacia/peach	4.0	3.0	120	1.03
3.	Dacia/almond	5.0	4.2	160	0.94

Analyzing air system increases (table 2), made in the first seven years after planting, we see that a greater height (5.0-5.5 m) showed in the Dacia variety grafted on engrafted apricot and almond rootstocks. Crown diameter is greater for grafting on the engrafted apricot and almond rootstocks (4.2-4.5 m). A greater influence on vegetative growth shows Dacia variety grafted on engrafted apricot and almond rootstocks.

Basically, the engrafted apricot rootstock imprints a greater increase in the Dacia variety; almond rootstock imprints a medium growth and a low growth on peach rootstock.

After studying the increase in thickness ratio, in the grafting zone we note that this ratio is very favorable (i.e. closer to the value of one or even one) for Dacia variety grafted on engrafted apricot and peach rootstock. It is less close to the value of 1.00 or less favorable for

Table 3

The development of the root system of the three rootstocks

No. crt.	Variant	The depth distribution of horizontal roots (cm)	The extension diameter of horizontal root (m)
1.	Dacia/engrafted apricot	20-50	5.20
2.	Dacia/peach	40-70	5.50
3.	Dacia/almond	25-60	4.80

Dacia variety grafted on almond rootstock, there is a danger of division for both partners.

Analyzing the root system architecture to the three rootstocks, we find that an important aspect is the rooting depth, (Table 3). Thus, very good rooting has the peach and engrafted apricot rootstocks, a strong attachment to roots in soil.

A shallower rooting was observed, (Table 3) to the almond rootstock (25-60 cm) and also a lower horizontal roots expansion in the soil (4.80 m).

CONCLUSIONS

In conclusion the apricot grafted on engrafted apricot and peach rootstocks shows a good growth both in terms of increased air system but the root system.

A great affinity between the two partners rootstock-graft has apricot/engrafted apricot and apricot/peach.

We do not consider an unfavorable association apricot/almond, but there may be adverse ratio growth in thickness of the two partners in the grafting zone and also a more superficial in depth root system formation on sand.

It is worth noting that, on the peach rootstock, the apricot shows a moderate growth, lower vigor, which is a favorable aspect for cutting trees, applying the treatments, harvesting.

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EFFECT OF VARYING DIETARY RATIOS OF AFALFA SILAGE TO CORN SILAGE ON MILK PRODUCTION AND CHEESE YIELD POTENTIAL IN LACTATING HOLSTEIN COWS

Ciobanu Radu[1]

Key words*; milk production, dairy cows, cheese yield potential, alfalfa silage, corn silage*

ABSTRACT

In this study was investigated the effect of different dietary ratios of alfalfa silage (AS) to corn silage (CS) on milk production and cheese yield potential. The 4 diets contained (dry matter basis:A) 50% AS, 50% concentrates, B) 40% AS, 10 % CS and 50% concentrates, C) 25% AS, 25% CS and 50% concentrates, and D) 10% AS, 40% CS and 50% concentrates). Intake of dry matter, yield of milk, milk fat all decreased when CS replaced AS. Milk protein yield was maximized in diet B and Cheese yield potential in diet C. Corn silage is a complementay with AS to maintin high milk protein yield and cheese yield potential.

INTRODUCTION

The quality of Telemea cheese begins with quality milk. One measure of quality milk is the chesee making potential of milk definded as the yield and quality of cheese produced from a unit of milk.

The most significant factors affecting the cheese making potential of milk include bovine genes, environment and physiolgy. These factors are behind the differences in chimical and pysical-properties of milk that influence the yield and quality of cheese.

Relationship between milk composition and milk processability in terms of cheese yield have been documented. (Amenu et al.2006).

In Oltenia area (S.W of Romania) the decline in milk protein during summer period may be a result of reduced nutrient intake owing to declines in the soluble carbohydrates metabolisme energy (ME) and protein content of forage avaible to the dairy cows.

In summer time reduced forage quality may be related to lower levels of milk protein (Beever et al, 2001). There is also the direct effect of heat an the cow, wich causes reduction in milk protein content (West, 1999).

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In south-west of Romania, enviromental modifications of the dairy-production system ase needed to enable Holstein-Friesian cows to express their genetic potential in milk production and to improve the cheese yield efficiency.

The study reported in this paper is part of a research program examining various strategies in intensive dairy production at Agricultural Research development Station (ARDS) Simnic Romania, with the objectives of increasing profitabilty.

Of the variation in milk composition, 55 percent is due to heredity and 45 percent is due to environmental factors, such as feeding management.

The nutritional factors receving the most attention the last 25 years for their influence on milk protein content were forage to concentrate ratio, the amount and source of dietary protein, and the amount and source of dietary fat.

A low transfer efficeiency (25-30%) of dietary protein to milk is a major factor accounting for the inability of diet to markedly alter milk protein content (Jenkins and McGuire, 2006).

Inefficeinet Nitrogen (N) utilization necessitates feeding large amounts of supplemental protein, increasing milk production costs and contributing to environmental N pollution (Broderick, 2006).

Feeding programs that maximize milk yield while maintaining normal component percentage will be the most profitable way in dairy farming. Feeding programs balanced for protein, energy and fiber, along with good bunk management, will increase milk yield and component procentaje.

Cheese makers need two main milk components: casein and fat. The amount of fat that can be used in making cheese is limited by the amount of casein present to hold the fat in a stable system.

The aim of this study was to evaluate the effects of the dietary ratio of alfalfa silage and corn silage on milk yield, milk composition, and cheese yield potential.

MATERIAL AND METHODS

Animals:

Twelve multiparous Holstein Friesian cows, averaging 104 (±42) kg of body Weight (BW) at the beginning of the experimment, were blocked by days in milk (DIM) and randomly assigned within blocks to treatment sequences in 3 replicated 4x4 Latin squares. Each experimental period lasted 28 days and consisted of 14 days for adaptation and 14 days for sample and data collection. Cows were housed in tie stalls and had free access to water.

Diets:

Table 1 shows the chemical composition of the silages and concentrates used in this experiment. Treatmants were fed as Total Mixed Ration (TMR) and contained the following proportions of alfalfa silage (AS): corn silage (CS) as perccentage of Dry matter (DM): 51:0 (diet A), 40:10 (diet B), 25:25 (diet C) and 10:40 (diet D). Concentrates comprised the rest of 100% DM of the diets. Diets were formulated to contain equal crude protein (CP) based on the initial composition of feed ingredients. Diets wese offered once daily at 10:00. Orts were collected daily at 09:00 and the amount of feed offered to the cows adjusted daily to yield refusals equal to aproximately 4-7% of intake.

Sampling and Laboratory analyses

Daily samples of 0.5 kg of AS, CS, concentrates, TMR and orts were collected, stored at -20 ^{o}C and used to make weekly composites.

Dry matter (DM) contents of weekly composites of all feeds, TMR and orts were determined by dryng al 60 ^{o}C for 48 hour and used to adjust as-fed compozitions of TMR. Ash content byashing the sample at 550 ^{o}C for 2 hours. Neutral detergent fibre and acid detergent fibre content was determinet by Fibretherm FT12 Cows were milked twice daily and milk yield was recored at each milking. Milk samples from a.m. and p.m. milkings were collected on day 19 and 26 of each period and analyzed for fat, solids non fat, total protein by ultrasonic milk analyzers with Ecomilk (EON Trading). Casein content was determined by gravimetric method. This method consits in the casein precipitation with 2 NCH_3COOH at pH=4.7. The precipitate is filtred, dried and weighed.

Cheese yield potential of milk was determined using theoretical cheese yield:

$$\text{Teoretical yield} = \left[(0.93 \times \text{fat} + (\text{casein} - 0.1)\right] \times 1.01 / \left(1 - \frac{\text{Cheese moisture}}{100}\right)$$

Were: 93% fat recovery in cheese;

0,1 theoretical lass at casein and 1,01 is a factor in theoretical yield formula (retention factor for otter milk solids).

Statistical analysis was performed. All data for the investigated parameters are shown as mean values.

RESULTS AND DISCUSSION

Foraje and diet composition

Chemical composition a feeds are shown in table 1.

Table 1

Chemical compositive of feed ingredients

Item	Alfalfa silage	Corn silage	Corn grain	Soybean seeds	Pea seeds
Dry matter (DM)	40.3	42.0	87.5	90.0	87.2
Cnide protein % of of DM	21.5	6.9	9.0	42.1	26.0
Ash % of DM	10.6	5.1	4.6	7.0	3.7
Neutral detergent fibre % of DM	38.1	42.0	9.0	11.4	20.3
Acid detergent fibre % of DM	29.0	20.9	4.4	5.4	10.8
pH	4.72	3.70	-	-	-

Crude protein content of AS and CS averaged 21,5 and 6,9%. Neutral detergent fibre (NDF), acid detergent fibre (ADF) contents wese tipical of high-quality forages.

Dietary soybean seeds was increased from 2.9 (diet A) to 16.0% of DM (diet D) to maintin CP levels as CS replaced AS and corn grains. Crude protein contents declined from diet A to diet D, and ash contents of the diet also decreased from 8.0 (diet A) to 6.4 % of DM (diet D) reflecting the higher ADF levels in AS (table 1).

Acid detergent fibre contents decreased from 16.8 (dietA) to 14.7 % of DM (diet D) refletcting the higher ADF levels in AS (table 1).

Neutral detergent fibre contents increased slightly from diet A to diet D (table 2) as a rezults of the higher NDF content of CS (table 1).

Composition of diets are shown in table 2.

Table 2

Composition of diets

Item	Diet AS:CS			
	A (50:0) % of DM	B (40:10) % of DM	C (25:25) % of DM	D (10:40) % of DM
Ingredient:				
AS	50	39.5	25.0	10.0
CS	-	10.5	25.0	40.0
Corn grains	40.4	37.4	32.3	27.3
Soybean seeds	2.9	5.2	11.0	16.0
Pea seeds	1.5	1.5	1.5	1.5
Brewers grain wet	4.0	4.0	4.0	4.0
Dicalcrum fosfate	0.2	0.2	0.2	0.2
Sodium biocarbonate	0.7	0.7	0.7	0.7
Salt	0.2	0.2	0.2	0.2
Vitamin premix	0.1	0.1	0.1	0.1
Chemical composition:				
DM%	50.2	51.0	52.0	52.8
CP % of DM	17.2	16.9	16.7	16.3
Ash % of Dm	8.0	7.8	7.0	6.4
NDF % of DM	24.3	24.5	24.9	25.0
ADF % of DM	16.8	17.0	15.0	14.7

Animal production

Tratament effects on dry mater intake (DMI), milk yield, milk composition and cheese yield potential are ashwn in table 3.

Table 3

Effects of varyng dietary rations of alfalfa silage (AS) to corn silage on the production of lactating dairy cows and cheese yield potential

Item		Diet (AS:CS)			
		A (51:0)	B (40:10)	C (25:25)	D (10:40)
DMI	kg/day	20.80	20.60	19.80	18.80
Milk yield	kg/day	29.10	29.80	28.60	27.60
3.5 % Fat concreted milk	kg/day	30.59	30.61	28.62	27.19
Milk fat	%	3.72	3.68	3.61	3.42
Milk fat	kg/day	1.082	1.078	1.003	0.941
milk protein	%	3.25	3.30	3.38	3.40
Milk protein	kg/day	0.945	0.983	0.96	0.938
Casein	%	0.47	2.58	2.67	2.64
Casein	kg/day	0.718	0.768	0.743	0.278
Chesse yield potential kg/100 kg of milk		11.756	11.800	11.840	11.440

The 9.6 % reduction in DMI from diet A to diet D was probably related to depressed ruminal fiber digestibility resulting from the effect of greater strach intake, lower ruminal pH and inhibition of cellulolytic bacteria.

Milk yield decreased as CS plus soybean seeds replaced AS plus corn grins. Cows fed diet D, with highest proportion of CS yielded 2.2 kg/day less milk than those fed diets with lower CS. Yield of 3.5 % fat content milk (FCM) also declined; difference of 3.4 kg/day of 3.5 % FCM was observed between cows fed diet A and those fed diet D.

The reduction in milk yield paralleled the decrease in DMI from diet A to diet D, suggesting that lower DMI accounted for depressed milk production. Several other authors (Onetti et al 2002, Krause Combs 2003, Ruppert et al 2003) reported similar milk yield when different dietary retios of AS:CS were fed.

Milk dfat content and yield decreased when CS replaced AS in the diets (table 3). For both variables diet A was highest (3.72%; 1.082 kg/day) and diet D lowest (3.42 %; 0.941 kg/day). Milk fat depression has been associated with increassed ruminal concentration of trans 18:1 fatty acid (mainly trans-10, 18:1 and trans-10, cis-12 conjugated linoleic acid). The full fat soybeen seeds increased from 2.9% of DM in diet A to 16 % of DM in diet D. A higher rumen avilability of C 18:2 faty acid from soybeean seeds is expossed to biohydrogenation and trans isomers C 18:1 result.

Milk protein content increased when CS was added to diets (table 3). The milk protein concentration of cows fed diet D was 4.6% higher than that of cows fed diet A. increassed milk protein content in the present study probably resulted from linear decrease in milk volume.

Milk protein yield was maximized at dietary AS:CS ratio of 40:10 possibbly because of the higher avilability of fermentable energy for ruminal microbes. This further supports the complementary characteristics of these forages.

Milk casein content increassed when CS replaced AS in the diet. Milk casein yield was the highest in diet B (0.768 kg/day) and the louwest diet A (0.718 kg/day), due probably by higher essential aminoacids supplied by diet B in ratio to the metabolisable protein relative to the ratio in milk.

The cheese yield potential of milk depends of the two main components: casein and fat. The amount of fat than can be used in making cheese is limited by the amount of casein present to hold the fat in curd.

The cheese yield potential of milk of cows fed diet C was the highest (11.840 kg of cheese per 100 kg of milk) due probably of optimum casein: fat ratio of cheese milk in diet C (0.74) compared with casein: fat ratio of cheese milk in diet A (0.66), B (0.70) and D (0.77). It is necessary to study the casein: fat ratio of cheese milk and its implications for cheese composition, cheese yield and the procentage recoveries of milk faat, milk casein and water to cheese.

CONCLUSIONS

- Under the conditions of this study, decreasing dietary AS and corn grains and increasing dietary CS and soybean seeds reduced DM intake.
- This may have contributed to reduced milk fat content and yield and depressed milk yield when the diet contained an AS:CS ratio of 10:40.
- Milk protein content increased when CS was added to diets.
- Milk protein yield was maximized at dietary AS:CS ratipo of 40:10. This further support the complementary caracteristics of these forages.
- Milk casein content was the highest in diet B wihen an AS:CS ratio was 40:10; higher essential amino acid supplied.
- The cheese yield potential of milk of cows fed diet (25:25) was the highest (11.840 kg/100kg of milk) when an casein: fat ratio of cheese milk was 0.74.
- The promotion of a protein feed (soybean and pea seeds) production directly on the farms is important in order to reduce the dependence of farms on the market.

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TRANSFER OF COW MILK COMPONENTS TO TELEMEA CHEESE

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Key words: *cow milk, milk constituents, telemea cheese, whey*

ABSTRACT

The aim of this study was to determine some of the milk components retention and losses in Telemea cheese manufacture and to calculate cheese yield efficiency as a percent of theoretical yield. The study was carried aut on partially skimmed vat milk in 6 chesse-making trials. The percentage of component lasses and cheese yield were calculated as described. The results cleary demonstrate that milk components (fat and casein) of cheese milk have implication for cheese yield and in the percentage ricoveries of milk fat and protein to cheese. Cheese yield as a percent of theoretical yield was 93,33 %.

INTRODUCTION

The principal constituents of milk of any species are water, fat, protein (caseins and whey proteins), lactose, minerals and trace quantities of vitamines and enzymes. Milk is a complex system, being an emulsion (of milk fat in globules protected by the milk fat globule membrane), a colloidal suspension (75-80% of the protein is casein, wich is found in aggregates called casein micelles) and a solution containing many dissolved components (Banu and Vizireanu, 1998).

The most important constituents in cheese making are:

-casein (formes the rennet gel which is primary structural element of cheese).

-lactose (fermentation substrate for starter lactic acid bacteria).

-calcium (essential for formation of a rennet gel in the initial stages of cheese making).

The levels of fat in milk ase much more variable than those of other constituents. Milk composition can vary acconding to diet of the cows, stage of lactation, mastitis and seasonality.

In cheese manufacture the objectives are:

-collect protein and fat and remove whey;

-ferment lactose into lactic acid;

-establish proper,,ripening environment".

Telemea cheese belongs to the group of white brined cheese. Common features of this group are that the cheeses are white in color and are ripended and kept in brine.

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Brine stage has a determinable effect on the biochemical, textural and structural changes that occur in the cheeses and leads to the developemnet of their characteristic flavor.

Telemea cheese is soft, rindless cheese salted, matured and kept in brine; it is acid and sligtly salty. It originates in Romania and named ,,Branza de Braila".

Telemea cheese is a concentrated gelled product that structurally consists of a para-casein matrix, enclosing the rest of milk components.

The retention of milk dry metter components in cheese is a rezult of interactions of numerous complementary factors, such as the chimical composition of milk, conditions of

milk collection and storge, type of coagulating enzyme and starter, production technology and technical equipment used at a chese dairy (Bohdziewicz, 2006).

The aim of this study was to determine some of milk components retention and losses in Telemea cheese manufacture.

MATERIAL AND METHODS

This study was carried ant at Agriculture Research and Developement Station (ARDS) Siminic-Fomania- dairy plant, on parttially skimmed vat milk. The milk was obtined by mixing skimmed milk of the evening milking and the whole milk of the morning milking.

The cheeses were made in april 2011 in 6 cheese-making trials. A total of 6 samples of cheese milk, each one representative of 100 kg of milk and the coresponding 6 samples of whey, were evaluated for the following parameteters:

-total solids, solids non fat, fat and protein content using ECOMILK M ULTRASONIC MILK ANALYSER and Gerber method;

-total casein content by gravimetric method (Popescu, 1988);

-curd fines as dried sediment expressed as mg in 1000g of whey;

-total wey protein by determining the N content of the fimes-free supernatant using Kjeldahl method.

The amount of milk in the vat was determined by weighing of the milk.

The percentage of component losses and cheese yield were calculated as de described by Guinee, 2006.

Clotting time indicated as time from rennet addition to the formation of vizible floccules was measured visually, using a spatula dipped in the cheese milk.

Titrable acidity was recorded as Thörner digree (^{o}T) by acid base titration with a 0.1 N Na OH solution and fenolftalein as indicator. Cheese weights were determined after pressing (day 1).

$$\text{Teoretical yield} = \frac{\left[\,0.93 \times \text{fat} + (\text{casein} - 0{,}1) \times 1.09\,\right]}{} \quad \frac{\text{desired cheese moisture}}{100}$$

Statistical analysis was performed. All data for the investingated parameters are shown as mean, ± standard deviation values and variability (%).

RESULTS AND DISCUSSION

Results of investigation are shown in table 1.

Wat milk:

The partially skimmed vat milk had an average dry matter content of 11.92%, fat content of 2.52, protein content of 3.24% and casein content of 2.50%. Fat content variability (3.1%) was higher than protein content variability (18%). Casein content variability (3.2%) was higher than protein content variability (18%).

Average titrable acidity of vat milk before starter culture adding was 17.67 ^{o}T and rennet clotting time (visually observed) veried from 16.1 to 22.1 minutes (mean 19.49 minutes).

The Holstein –Friesian breed is well known for high milk production low protein content, and poor coagulation properties of milk (de Marchi, 2007).

The dray metter of the whey resulted was 7.6 g/100g and varied from 7.1 to 8.0 g/100g. The fat content 0.36 g/100g, was characterised by 8.3% variability (table 1). The average of cnede protein was 0.56 g/100g with 8.9 % variability. The average of crude fines was 204 mg/kg with minimum 190 mg/kg and maximum 225 mg/kg.

Mean cheese yield was 9.8 kg/100kg of milk and varied from 9.0 to 10.15 kg/kg.

Average theoretical cheese yield was 10.5 kg/100kg of milk. Failure to achieve theoretical yield means that some casein and fat were lost during the manufacturing process. Yield losses occurred as dispersed fat globules, soluble breackdown products of casein and filtrable curd fines expelled into the whey (table 1).

Table 1.

Vat milk, whey characteristics and cheese making lasses

		Mean	SD ±	Variability %	Minimum	Maximum
Vat milk (Dray matter g/100g		11.2	0.29	2.5	11.60	12.40
Fat	g/100g	2.52	0.08	3.1	2.40	2.62
Crude protein	g/100g	3.24	0.06	1.8	3.18	3.34
Casein	g/100g	2.50	0.08	3.2	2.40	2.65
Titratable acidity	^{o}T	17.67	1.21	6.8	16	19
Clotting time minutes		19.49	2.17	11.1	16.10	22.10
Whey: Dray matter	g/100g	7.60	0.29	3.8	7.1	8.0
Fat	g/100g	0.36	0.03	8.3	0.32	0.41
Crude protein	g/100g	0.56	0.05	8.9	0.50	0.62
Curd fines	mg/kg	204	12.00	5.8	190	225
Cheese and whey yield: Theoretical cheese yield	kg/100kg	10.50	0.23	2.1	10.18	10.80
Cheese yield	kg/100kg	9.80	0.42	4.2	9.0	10.15
Wey yield	kg/100kg	89.71	0.42	0.4	89.40	90.50
Losses: Dray matter %		63.75	1.37	2.1	61.20	65.00
Fat loss %		14.16	1.26	8.9	12.26	15.64
Protein %		17.24	1.10	6.3	15.62	18.56

Average of whey fat of 0.36% seems like small content, but quantitatively represents 14.16% fat loss from fat of vat milk (table 1).

The Van Slyke equation assumes that a cheese plant should be able to recoveer 93% of the original milk fat in the finished cheese. In this study mean fat recovery was 85.84%.

Manufacturing performance is based upon how efficiently is converting solids from milk supply into actual cheese yield.

As a percent of theoretical yield, cheese yield efficiency was 93,33%. The Van Slyke equation did well predicting magnitude and direction of changes of cheese yield.

CONCLUSIONS

- The results clearly demonstrate that milk components (fat and casein) of cheese milk have implication for cheese yield and in the percentage recoveries of milk fat and protein to cheese.
- A major reson for low actual cheese yields was excessive lass of fat during cheese making.
- It is necessary to find the optimum ratio of casein to fat in standardization of vat milk.
- Accurate casein determinations are very important, but analytical methods for casein have not been refined to the same extent as methods for milk fat.
- The Van Slyke cheese yield formula predicts higher cheese yields than actual cheese yields, but is useful for improving in-plant cheese yield.
- Cheese yield efficiency as a present of theoretical yield was 93,33%.

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ASSESSMENT OF THE HEREDITARY VARIABILITY OF F1 INTRASPECIFIC HYBRID VINE PLANT DESCENDENTS USING AMPELOGRAPHICS AND AMPELOMETRICS METHODS

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Key-words: *hybrid, hereditary, intraspecific combinations, parental forms.*

ABSTRACT

To create new varieties of the table grapes is important to understand the genetic basis of the varieties involved in hybrid combinations and mode of transmission to offspring of different characters and value combination thereof.

So determinations were made on descriptors ampelometric and ampelographic and adult leaves from five elite ((BpxV21, BpxV52, BpxV63, BpxV74, BpxV135), in combination Victoria X Black Pearl, and the leaves belonging to varieties used as maternal and paternal genitors and then calculated the average of these measurements genotypes compared with parental varieties.

*Ampelometric determinations included the following measurements: leaf area, length of main veins in order to determine their relationships ((N2/N1=A=*21, N3/N1=B=*31, N4/N1=C=*41, the sum of the angles α, β,γ of the ribs, and the length ratio of the distances sinus nerves (d1/N2, d2/N3).*

INTRODUCTION

Romanian viticulture orientation provides assertion by reconsidering share the sorts of valuable native varieties and promoting quality culture variety of existing and valuable to be obtained.

An important of the work of improvement is to create new varieties adapted vine current climate and soil conditions, the complex biological resistant varieties.

The numerous varieties in cultivation, vine polymorphism, and technological and agro biological complexity characters used in vineyards led to confusion on the recognition of varieties.

Therefore the description of varieties should follow standards developed by international organization accredited (OIV; UPOV; IBPGP) as common code list descriptors varieties and Vitis genus (OIV, UPOV, IBPGP).

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To create new varieties of table grapes is important to understand the genetic basis of the varieties involved in hybrid combinations and the mode of transmission to descendants of various characters and their combination value.

In the Laboratory of improvement were obtained by controlled hybridization (sexual) 102 elite hybrids from five combinations, which were planted in the greenhouse. Hybridization were made in 2007 and in 2012 entered the fruit plants.

MATERIAL AND METHODS

To know the main characters ampelographic of elites, but also an understanding of how the characters have been transmitted from parent to progeny varieties, ampelographic determinations were made on descriptors: chord, wheel, young shoot, young leaf, mature leaf, frames, flower, grape and grain.

Because the leaf has very important character recognition vine varieties were determined 29 important descriptors in mature leaf to 52 new genotypes, from four hybrid combinations.

This paper aims to present some results on the transmission to F1 hybrid progeny of characters such as: shape, size, colour and leaf appearance; number, depth and shape of the lateral sinus; petiole sinus form; shape and size of teeth; pubescent blade; colour veins; stem length and colour.

At crossings were used 5 vinifera varieties, different in terms of morphological and physiological traits: Victoria, Canner, Black Pearl, Muscat Iantarnâi şi Muscat de Hamburg.

Data interpretation was done by comparing the elite hybrid leaf descriptors obtained from genitor varieties, the hybrid combinations.

Were analyzed and determined ampelographic 52 hybrid genotypes from four combinations: Mt. Iantarnâi x Canner, Muscat Hamburg x Canner, Victoria x Black pearl, Victoria x Canner, coded as follows:

- IXC-19 elite hybrid, HXC-11 elite hybrid, VXBP-11 elite hybrid, VXC-11 elite hybrid

Ampelometric and ampelographic determinations were performed at ten leaves mature from elite hybrids and varieties parents.

According to *The methodology described descriptors ampelographic* each character and digit coded terms, represents a level of expression, as the smallest units in presenting a character. To avoid confusion and facilitate determination for each expression level were established reference varieties for comparison.

To achieve ampelometric determinations were made following measurements: leaf area, length of main veins, in order to determine their relationship (N2/N1=A=*21, N3/N1=B=*31, N4/N1=C=*41), sum of the angles α, β,γ of veins, ratio of distances sinus and length of veins (d1/N2, d2/N3).

RESULTS AND DISCUSSION

Hybrid descendants have intermediate characters and traits between the two parental forms in generation I (F1), but there are also cases overall dominance of one of genitors, or only a character and trait. A series of descriptors for mature leaf underlying identification and description of vine varieties.

To define the morphological characteristics of elite hybrid derived were made ampelographic and ampelometric determinations to mature leaf, comparative with varieties used in hybrid combinations. So can be seen the mode of transmission of characters from genitors to the descendants but and combinative value of several varieties for table grapes.

Size of blade estimated after lenght and width of leaf ,structure of surface, shape of blade, number of lobes, size of teeth in relation to blade size, opening/overlapping of petiole sinus. After ratio between lenght of blade and internode of support, leaves are: *small*, when the ratio of the two elements is less than 1; *large*, when the ratio is 1; and *very large,* when the ratio between them is 1,5. Leaves from an elite hybrid, from the combination HXC was classified as very large, transmitted character from Canner variety, 11 elite from Muscat Iantarnâi x Canner combination had small leaf, similar maternal genitor variety (Mt. Iantarnâi). Black Pearl variety has sent large character from 3 elite hybrid, and Victoria and Mt. Hamburg varieties propagated medium size leaf to 5, respectively 3 elite. Variety Canner (used as male) shows large leaf to 7 hybrid descendants, and the rest elites showed intermediate characters from the varieties used in directed hybridization.

Shape of blade. Five types of leaves are different: wedge-shaped (elites from all hybrid combinations-10), cordate (7 hybrid elite from combinations VXC), pentagonal (24 genotypes from the four combinations), round (3 combinations) and kidney-shaped (only combination IXC by 2 individuals). 64% of individuals obtained from Victoria x Canner combination showed cordate leaf, similar maternal variety.

Number of lobes. Leaves with two upper sinuses and one petiole are *trilobite* (6 elite, prevailing HxC combination with 4 elite) and the two with upper sinuses, two lower and one petiole are *five lobate,* character typical for most elites analyzed but to the varieties used as genitors. *Seven lobed* leaves meet to one elite from each combination, except combination VXC with two individuals.

After form, are distinguished: the angular *teeth* with straight edges and sharp point; teeth with convex edges, or concave-convex edges. Most elites have leaves with straight teeth (45) and convex (20).

Ampelometric method represents the expression of leaf morphological characters by numerical values. Values obtained by ampelometric measurements were coded and interpreted by the method proposed by P GALET. With the aid the results related to the leaf surface were able to establish correlations between the blade leaf size of the hybrids studied and the varieties genitors.

The method consists in taking the 10 leaves from each elite hybrid studied and measuring length main veins (N1, N2, N3, N4), angles formed between the main veins α, β,γ, the sinus depth (d1/N2 si d2/N3) wich determines the degree of sectional blade. Tabel nr.1

Measurements were made at five elites (BpxV21, BpxV52, BpxV63, BpxV74, BpxV135), from the combination Victoria x Black Pearl, and the leaves belonging to varieties used as genitors maternal and paternal and then calculated the average of these measurements genotype compared with the parental varieties.

Analyzing data on length of veins it was found that elite hybrid ranged between 8,9-15,7cm median vein length (N1), 8,4-12cm upper side main veins length (N2), 5-8,6 cm lower side main vein length (N3), 3,2-4,9 cm lower secondary vein length (N4), compared with genitor varieties Victoria (9,3-10,8cm N1; 8-8,4cm N2; 4,8-5,6cm N3; 2,9-3,5cm N4) and Black Pearl (9,0-10,0 cm N1; 8,4-9,7cm N2; 6,4-7,3cm N3; 3,5-7,6 cm N4)

which presented the length veins with values generally lower exceptions doing only two parameters (N3 şi N4) from the male variety.

To present the more conclusively multitude of data obtained we used the distribution on frequency class of mean values of parameters determined for elite hybrid and parents.

For the values representing sum of the angles α+ β+ γ it indicates a distribution very close to normal.-Fig1. The value ratio in histogram is almost symmetrical about the mean 3,9. Of 35 determinations on the sum of the angles resulting that most elites are below average, significant excesses were Victoria variety and 63 elite.

A completely different distribution by index vaulting meets for the ABC ratio. The empirical distribution rises above the normal to the mean values. Here we meet an excess of values placed above average and far from it, the distribution is the limit of normality rejection-Fig 2

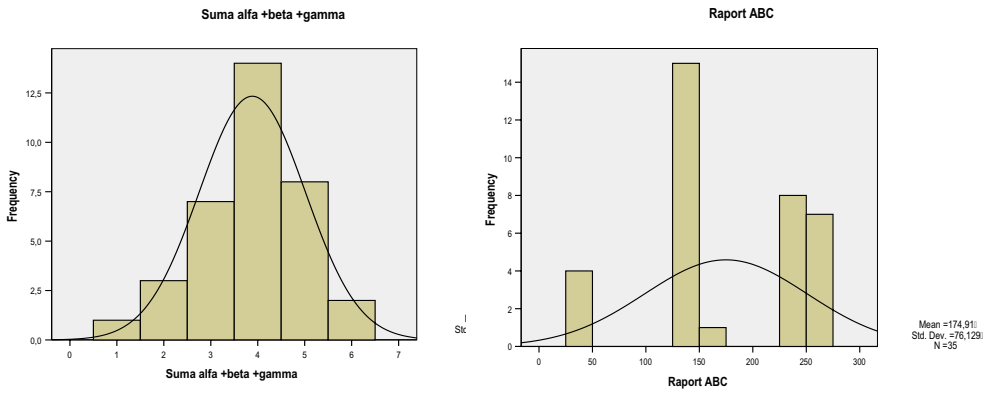

Figure1-2. Distribution on frequency class of average values of the sum of the angles α + β + γ and ratio ABC

Table nr.1

The variation of the leaf shape to the parental varieties and hybrid elite – codes

Nr.crt	Variety/Hybrid	Sum α+β	Sum α+ β+ γ	Ratio A BC	Shape of blade
1	*Victoria*	3	5	246	*wedge-shaped*
2.	*Black Pearl*	3	5	035	*circular-kidney-shaped*
3.	Victoria x Black P21	3	3	126	*wedge-shaped*
4.	Victoria x Black P52	2	3	246	*wedge-shaped*
4	Victoria x Black P 63	3	4	146	*wedge-shaped*
5.	Victoria x Black P 74	3	4	257	Cuneo-cordiformă
6.	Victoria x Blck P 135	3	4	145	*wedge-shaped*

The size of leaf was determined by the method Kişkin, applying the formula $\pi d^2/4$ in which is the distance between the top mucron and the farthest point according to OIV 065. For combination VxBp the values obtained put the leaf in the level of expression medium. Values are within the limits 142,28cm^2 (small) and 214,4 cm^2 (medium). The values obtained for genitors put the mature leaf: Black Pearl: 124,618 cm^2 (small) - 162,77cm^2 (medium) and Victoria 120,70cm^2 (small)- 167,33 cm^2 (medium).

Overall study of performed measurements and codification of relations veins and the sum of angles presented in Table 1, show the shape of leaf of elite hybrid and parental varieties.

The analysis of d1/N2 d2/N3 ratio allowed the evaluation of the sectional leaves, represented parameters by Duncan test, thus highlighting the differences between elites hybrid and genitors leaves.

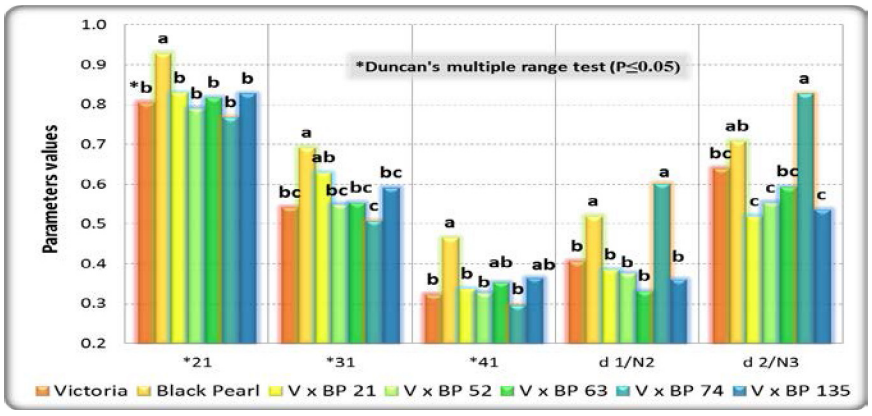

Figure3. Differences recorded to the characters *Sinus depth* to the elite hybrid

As regards the character *21(A) was noted that all hybrid descendants they borrowed this trait just from the variety Victoria. Also elite VXBP 21 borrow character * 31 (B) from both genitors all the other progenies having character *Lower side main vein length/Median vein length* (*31) just like Victoria and completely different from Black Pearl.

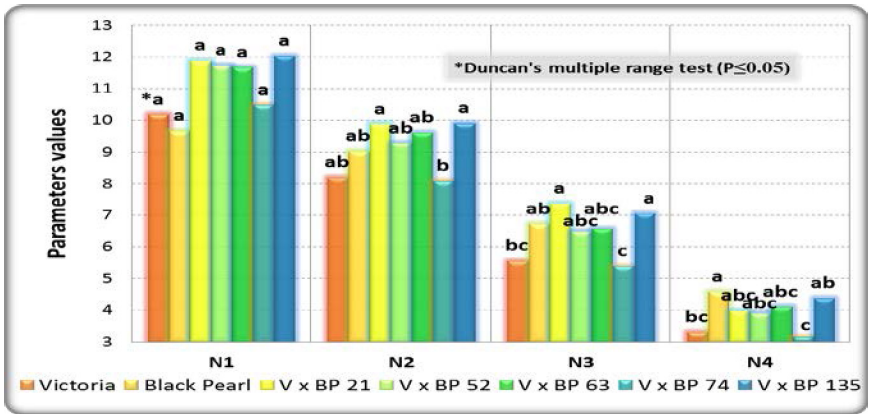

Figure 4. Differences recorded to the characters *Length veins* to the elite hybrid

Ratio *41 is taken from both genitors only by VxBP 63 and VxBP135, remaining descendants borrow only character from Victoria. Both ratios d1/N2 and d2/N3 reach high values genitor Black Pearl, only for VxBP hybrid elite 74, other progenies remaining to close values by Victoria variety. Fig.4

Regarding the length veins the differences between genitors are statistically assured only for N4, in that case only hybrid VxBP 74 showing different values of Black Pearl, the others progenies having intermediate characters.

CONCLUSIONS

- By studying a rich collection hybrids on separate groups of interbreeding between different species of the genus Vitis, were followed in lineage hereditary particularities characteristic leaf varieties Victoria and Black Pearl.
- Knowing the main phenological characteristics and quality of these varieties help to orientation towards the desired characters to be fitted to new varieties created by hybridization. All these varieties are potential genitors for obtaining new varieties.
- With the help of results related to leaf surface (method Galet) were able to establish correlations between foliar size of hybrids studied, but also genitors varieties.
- Ampelographic and ampelometric analysis results of the 7 varieties of Vitis sp. (5 elite hybrid and 2 varieties used in intraspecific hybridization) revealed genetic particularities for each genotype.
- Elite hybrid obtained have characters inherited from the parents, but also many intermediate characters. Victoria is a good variety genitor for obtaining valuable varieties for table grapes.

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EVALUATION CONCERNING THE PHYTOSANITARITY QUALITY OF SOME PLANT SPECIES OF THE GENUS *Prunus*

Ciomaga Fraguta[1]

Key words: *Plum pox virus, plum, apricot, resistance, detection*

ABSTRACT

Plant pathogen interaction remains one of the most important problem addressed by the entire Europa.PPV - (Plum pox virus) that is responsible for Sharka disease, is one of the most important quarantine organisms. This paper aims to present an evaluation of plant quality assessment of plum and apricot genotypes that can be considerate the perspective one after their infection with PPV and testing them using serological and molecular techniques. For the few plum and apricot local genotypes that showed no symptoms of PPV infection under field conditions, were grafted in greenhouse conditions on GF 305 rootstock which were previously inoculated with PPV (strain D) chip budding method. After leaving the vegetation were tested both, the rootstocks and grafted genotypes by ELISA and PCR methods.

INTRODUCTION

Sharka disease, caused by a *virus* (PPV) is one of the most serious viral diseases of stone-fruit crops, including peach (*Prunus persica* L.), apricot (*P. armeniaca* L.), plums (*P. domestica* L. and *P. salicina* Lindl.) as well as sweet and sour cherries (*P. avium* L. and *P. cerasus* L.) that may be systemically infected by a few unique PPV strains (. 1999), (Audergon *et al*. 1994, Damsteegt *et al* 2011,Egea *et al*. 1999, Karayiannis *et al* 1999, Polák 1994).

In apricot (Prunus armeniaca), as a result of the intensive search for a source of resistance within available apricot germplasm, some North American cultivars ['Stark Early Orange' (SEO), 'Goldrich', 'Harlayne','Stella', and others] were found to have natural resistance to PPV (Martinez-Gomez *et al.* 2000). These cultivars were used as donors for a resistance trait in conventional breeding programs based on crosses between resistant and the best local cultivars susceptible to virus. The different sources of resistance have been described (Dosba *et al.* 1994), whereas the resistance was shown to be quantitative and partial (Ion-Nagy *et al.* 2006).

In peach (Prunus persica L. Batsch), all cultivars are susceptible, and no source of resistance has been identified (Escalettes et al. 1998) up to now. However, in Prunus davidiana, a wild species a relative of peach, the clone P1908 was shown to bear resistance to PPV movement and incidence (Decroocq *et al.* 2005).

For the plum, a transgenic individuals developed by Scorza *et al.(*1994) has a durable PPV coat protein-mediated resistance (Hily *et al.* 2004). Unfortunately, the

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inability to reliably regenerate transgenic plants with other Prunus species hinders the application of transgene-mediated resistance.

Breeding programs for PPV resistance have met with limited success due to the multigenic nature of the resistance, long juvenile period of seedlings (4–8 years for Prunus species) and the strain specific nature of resistance (Kegler & Hartman, 1998).

The introduction of resistant cultivars of stone fruits into the orchards is the best long-term solution to virus control. The strategy for obtaining resistant cultivars is to identify natural resistance present in Prunus germplasm and introduce this resistance into commercial cultivars by standard breeding practices

The goal of the work presented in this communication is the identification of natural resistance in breeding programs screening the individual trees for the resistance phenotype.

Prunus genetic map (Joobeur *et al.* 1998). This Prunus genetic reference map is the comparative map standard for the Prunus community and contains anchor markers for integration of genetic information from all Prunus species (Aranzana *et al*. 2003, Dirlewanger *et al*. 2004). In addition, Lalli *et al*. (2005) published the first resistance gene map for Prunus identifying 42 resistance gene containing loci on the general Prunus map. The combined reference or resistance map serves as a resource for identification of candidate resistance genes in Prunus breeding materials and for utilization of MAS in resistance breeding.

MATERIALS AND METHODS

Plant materials

The Romanian plum genotypes('Myrobolan 4Kr' - rootstock, 'Andreea', 'Carpatin', 'Centenar' – plum varieties,'Pixy' and 'Scoldus', 'Otesani 11', 'Calugaresti'), the local adapted apricot varieties ('Dacia', 'Traian', 'Nicusor', 'Carmela', 'Amiral', 'Sirena', 'Olimp', 'Litoral) and 46 Romanian old varieties of peach germplasm were tested for their ability to support the artificial PPV infection.All this material was grafted onto inoculated (by chip budding) peach GF305 considerate indicator for PPV. (Fig. 2 and 3)

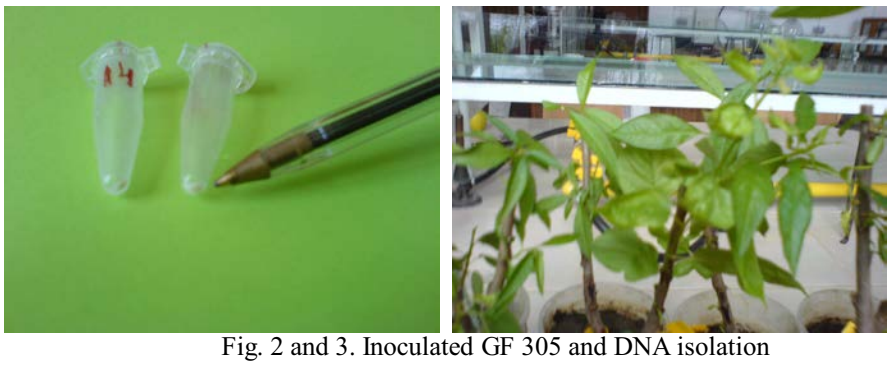

Fig. 2 and 3. Inoculated GF 305 and DNA isolation

PPV isolate

For this work was used one strain of PPV; D (Dideron).

The PPV isolate assay was RB3.30, a Dideron type from the collection of the SCDP Bistrita Romania isolated from the plum. Strain D is less aggressive.

Serological method – ELISA of detections

Presence of PPV in both the scion and the rootstock of infected plants were examined by serological ELISA tests, three times during each vegetative cycle. The mashed leaves (samples) in extraction buffer (AFT 0,2 % + Dieca 2% + PVP – 10) were placed in holes in the plate previously tapisated with polyclonal immunoglobulins conjugated (anti-PPV) and incubated at 4 ^{0}C for 16h. After 3 washes (with AFT- Tween) were added 200 µI specific monoclonal antibodies for PPV and incubated at 37 ^{0}C for 2 h. The last step was the implementation of immunoglobulins conjugated with alkaline phosphatase 1:1000 (200µI) and incubated for 2h at 37 ^{0}C. Readings were performed at 405 nm considering the positive values exceeding twice the value of negative test reading (T-x 2).

Molecular detection was performed by RT-PCR (Reverse Transcription-Polymerase Chain Reaction) using a primer pair (Pl/P2) that amplifies a 243 bp fragment located at the C-terminus of the PPV CP gene. PPV was trapped with PPV-polyclonal antibodies adsorbed on an Eppendorf micro tube. Enhanced Avian kit provided by Sigma was used for RT-PCR., according to Olmos *et al*. (1997). The thermal cycling scheme used was the following: RT- 30 min at *50°C,* denaturation / RT inactivation - 2 min at 94°C followed by *35* cycles: template denaturation - 30 s at 94°C, primer annealing - 45 s at 61°C and DNA elongation- 60 s at 72°C. Following to the last cycle, amplified DNA was elongated for 10 min at72°C. An aliquot of the amplified products (10 µI) was fractionated onto *1.5%* agarose gel electrophoresis in 1x TBE buffer. Bands were visualized by ethidium-bromide staining under UV light. (Fig. 1)

Total RNA was isolated *from* leaf tissue located at three different locations (tip,middle and bottom).

RESULTS AND DISCUSSIONS

Local plum genotypes identified in different parts of the country were selected (Otesani 8, Alina, Tita, Mirovice, Rival, Oltval, Tuleu dulce) and grafted on GF305 peach rootstock considered indicator of PPV, it was previously infected with strain D (Dideron) by chip-budding.

M 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16

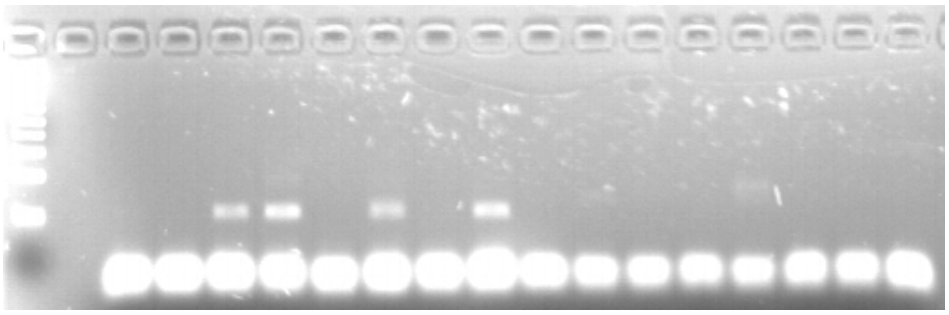

Fig. 1 Agarose gel electrophoresis to detect infection with PPV in apricot genotypes

Table 1

Monitoring by molecular and serological test in terms of PPV infection at some local plum genotypes.

Nr..	Genotypes	2011 (frst scoring)		2012 (second scoring)	
		DAS Elisa	RT - PCR	DAS Elisa	RT- PCR
1.	Otesani 8	+	+	+	+
2.	Alina	+	+	+	+
3.	Tita	+	+	+	+
4.	Rival	-	-	-	-
5.	Miroval	-	-	-	-
6.	Oltval	+	+	+	+
7.	Tuleu dulce (control po	+	+	+	+
8.	C+ control pozitiv	+	+	+	+
9.	C- control negativ	-	-	-	-

After serological and molecular tests, during two growing seasons showed that genotypes "Rival" and "Miroval" proved to be resistant to artificial infection with PPV. In the table 1 are presented the results at serological and molecular tests for some plum genotypes after 2 years scoring.

Table 2

Serological and molecular tests of apricot genotypes after artificially infection with PPV

Nr. crt.	Genotipuri	2011 Das Elisa	2011- RT -PCR	2012 DAS Elisa	2012 RT-PCR
1.	Traian	-	-	-	-
2.	Auras	-	-	-	-
3.	Sirena	+	+	+	+
4.	Olimp	+	+	+	+
5.	Orizont	-	-	-	-
6.	Sulmona	+	+	+	+
7.	Ovidiu	-	-	-	-
8.	Amiral	-	+	+	+
9.	Euxin	-	-	-	-
10.	Tudor	-	-	-	-
11.	Augustin	-	-	-	-
12.	Ceres	-	-	-	+
13.	Danubiu	-	-	-	-
14.	Histria	-	-	-	-
15.	Harcot	-	-	-	-
16.	SEO	-	-	-	-

Genotypes:

‘Traian’,‘Auras’,‘Orizont’,‘Ovidiu’,‘Ceres’,’Euxin’,’Tudor’,’Augustin’,’Danubiu’,’Hristia” and varieties 'SEO' and 'Harcot' considered to be resistant to PPV were tested by serological, DAS ELISA l and molecular RT-PCR (Fig. 1). The results showed that Romanian varieties 'Trajan', 'Auras', 'Ceres', 'Euxin', 'Tudor', 'Augustin' are resistant to

artificial infection with PPV in greenhouse conditions, the results are presented in the table 2.

CONCLUSIONS

Evaluation of local varieties and genotypes of the genus Prunus in terms of resistance to PPV (Plum pox virus) allows identifying new sources of resistance to this quarantine disease "Sharka". Regarding knowing already given plum variety Andreea and local genotypes "Mirobolan 4Kr" and "Local Dragasani 'has already been declared resistant to PPV, research in this paper come with other additions with two genotypes of plum" Rival "and" Miroval" which proved resistant to PPV by serological and molecular tests after artificial infection in greenhouse conditions.

Regarding the Romanian the apricot varieties:

Traian', 'Auras', 'Orizont', 'Ovidiu', 'Ceres', 'Euxin','Tudor','Augustin' Danubiu', 'Hristia' proved to be rezistentae the PPV. Information is coming to bring extra value to Romanian varieties of apricot.

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COMPOUNDS CAPTURED IN CARBON DIOXIDE EFFLUENT DURING AROMAT DE IASI FERMENTATION

Lucia-Cintia Colibaba[1], Valeriu V. Cotea[2], Marius Niculaua[3], Bogdan Nechita[2], Maria Codreanu[2]

Keywords: *Aromat de Iasi, aroma compounds, exhaust CO_2*

ABSTRACT

The volatile aromas that are lost during the must's fermentation into wine represent a department that is not very much analysed. The capturing and analysing of the volatile compounds that are trapped in the CO_2 flow during gas exhaustion of the fermentation stage are the main objectives of the present study. The Aromat de Iasi grapes, harvest of 2011, were processed according to the aromatic wine technology During fermentation, the volatile aromatic compounds were captured using SPE cartridges attached to the airlocks of the fermentation vessels. After the fermentation ended, the extracts were obtained. Gas-chromatography coupled with mass-spectrometry was used to identify the captured compounds. Esters and alcohols, as well as terpenes are found in the exhaust air of the fermentation process. The identified compounds are found in trace quantities.

INTRODUCTION

The sensoric character of wine is influenced by the grape variety, the maturity degree of the grapes at harvest, the yeast activity, pre-fermentative technologies and aging techniques.

The aroma compounds are the ones that give wine its "genius", specificity and individuality. More or less pronounced aromas are the base of each wine's personality. The aromas evolve during the grape maturation, becoming, from an organoleptic (qualitative) point of view, „personal" for each wine and ecosystem.

In specific literature, more than 800 aroma compounds are attributed to wine, among which are alcohols, aldehides, ketones, esters, acids and terpenes.

The most important stage of the wine production is the fermentation, the "birth" of the wine. At this time, the second class of aroma compounds is formed, after the varietal aromas and before the aging specific sensorics.

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During fermentation, a large volume of CO_2 gas is produced, about 260 mL/g glucose. This equates to over 50 times the volume of the juice fermented. Carried off with CO_2 are various volatile compounds (Jackson, 2000).

This study aims at analysing the volatile aroma compounds that are lost during the fermentation stage, by flowing out together with the exhaust of carbon dioxide (Nasrawi *et al.*, 1990; Muller *et.al*., 1993), from Aromat de Iasi, a new grape creation, obtained in 1980 by Danulescu D. et al. through free fecundation of Tamaioasa romaneasca grape seeds. These will be harvested by use of SPE (solid phase extraction) cartridges with an ability to retain volatile compounds.

MATERIAL AND METHOD

Aromat de Iasi grapes were used, harvested from Iasi - Copou vineyard in 2011. The grapes were processed according to the specific literature (Cotea *et al.*, 1986) for neutral and aromatic wine technologies. After desteming and crushing, the marc was macerated for 12 hours; for a better extraction of the aroma compounds commercial pectolytical enzymes (Zymoclaire M®, Sodinal) were added. Pressing of the must was done with a hydraulic press. Selected yeasts (Fermactive Muscat®, Sodinal) were added. The must was transferred into glass vessels for fermentation.

The capturing of the volatile compounds (fig.1) that are lost during fermentation was done as follows: SPE (solid phase extraction) cartridges were conditioned (6 mL diclormethane, 6 mL ethanol and 6 mL ethanol solution (14%) were passed through the C18 bed) and fixed to the fermentation airlocks so that the exhaust CO2 flow, together with the volatile compounds passed through them. The volatile substances were „trapped" in the SPE device for further analysis.

Figure 1 Capturing of exhaust CO_2 and compounds in Aromat de Iasi fermentation process

After the fermentation process ended, the attached SPEs were disconnected and analysed. The volatile compounds were obtained by washing the SPE bed with 2 mL diclormethane.

The obtained extract was injected into a Shimadzu GC coupled with a QP2010 Plus mass-spectrometer.

1 μL extract are injected into a Supelco SLB 5 ms GC column, 15 m length, column oven temperature 30 ^{o}C, injection temperature 250 ^{o}C, in splitless mode, initial temperature 30 ^{o}C for 1 minute, then if grows at a rate of 8 ^{o}C until 240 ^{o}C where it stays for 2.75 minutes. The carrier gas was Helium, column flow 0.75 mL/min, ion source temperature 250 ^{o}C, interface temperature 250 ^{o}C, detector voltage 0.9 kV.

The aroma compounds were determined by means of the NIST 08, Wiley 08 and SZTERP spectrum library. The program lasts for 30 minutes.

RESULTS AND DISCUSSIONS

Part of obtained chromatogram after analysing the volatile compounds found in the exhaust CO2 flow of fermenting Aromat de Iasi grapes is reproduced here (fig. 2)

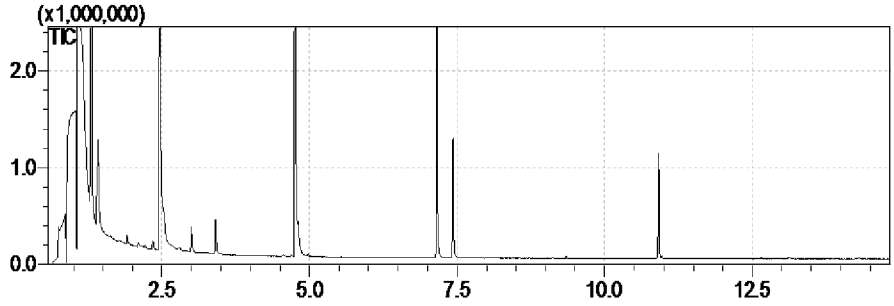

Figure 2. Chromatogram (part) of exhaust CO_2 flow of Aromat de Iasi wine

The compounds indentified in the CO_2 exhaust flow are in very small quantities and very volatile, with small masses, being able to get "lost" during the fermentation process.

The identified compounds are from major chemical classes specific to wine: terpenes, alcohols, esters (Table 1). The compounds are characterised by their area.

Table 1

Compounds identified in exhaust CO_2 flow

No.	Retention time	Area	Identified compound
1	1.930	3808487	1-Propanol, 2-methyl-
2	3.009	43962167	1-Butanol, 3-methyl-
3	16.858	68814	Linalool
4	3.373	96978	Ethyl isobutyrate
5	4.641	366693	Ethyl butirate
6	7.995	492024	Isoamyl acetate
7	12.368	2288426	Ethyl hexanoate
8	17.464	4799274	Ethyl caprylate
9	20.628	2121992	Ethyl decanoate

Among the terpenes, linalool was identified in the exhaust CO_2 of the fermenting Aromat de Iasi must.

Linalool is a component of many essential oils, including orange, lavender, rose, rosewood, and coriander. It is a naturally occurring terpene alcohol chemical found in many flowers and spice plants with many commercial applications, the majority of which are based on its pleasant scent (floral, with a touch of spiciness).

From the volatile alcohols class, isobutyl alcohol and 1-butanol, 3-methyl- were identified. Isobutanol has a strong solvent smell, bitter while 3-methyl-1-butanol is a main ingredient in the production of banana oil, an ester found in nature and also produced as a flavouring substance in industry.

The identified esters are ethyl isobutyrate, ethyl butyrate, isoamyl acetate, ethyl hexanoate, ethyl caprylate, ethyl decanoate.

The esters of fatty acids (ethyl hexanoate, ethyl octanoate, ethyl decanoate) have specific aromas, those of fruit, respectively apple and grape.

Isoamyl acetate smells nice, of bananas and melon, is characteristic of cool-fermented whites.

Ethyl isobutyrate has a sweet odour, while ethyl butirate has an aroma associated with fresh orange juice (Ján Farkaš, 1988).

CONCLUSIONS

During fermentation of grape must, an important number of aroma compounds, from almost all chemical classes specific to wine aroma, are lost in the exhaust CO_2 flow.

A SPE trap has succeeded in capturing aroma compounds from the exhaust CO_2 flow of fermenting must.

Further research will analyse the possibility of separating the identified compounds and their reuse as aroma additives in certain food and cosmetic industries.

ACKNOWLEGMENTS

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THE INFLUENCE OF CLIMATIC CHANGES OVER THE AGROBIOLOGICAL AND TECHNOLOGICAL CHARACTERISTICS OF THE TABLE GRAPE VARIETY – ARGESSIS

Costescu Adriana[1], Popa Camelia[2], Dejeu Liviu[3]

Key words: *table grapes, climatic change, agrobiological descriptors*

ABSTRACT

The studied variety in this paper – Argessis, had been homologated at INCDBH Stefănești-Argeș in 2002. Under bad weather conditions, due to excessive drought but also to high temperatures recorded during the last years, the results of qualitative and quantitative production can be considered satisfactory. Since this variety ensures the largest quantitative productions for the moment, it must be extended in culture. The study achieved in this paper, refers to its behaviour before homologation (1999-2001), as compared to the last years (2009-2011) when climatic changes had been significant, as compared to the homologation year. This paper presents the results obtained in the Ștefănești-Argeș vineyard regarding the fertility, the productivity, the quantity and the quality of grapes for the variety Argessis. During the studied period, the variety Argessis, distinguished through a weight of the cluster of 564 g, a production of 24 t/ht and a very pleasant aspect of the grapes.

INTRODUCTION

Among all the edible fruit, grapes are extremely appreciated. Their pleasant aspect, the nectarious taste but mainly their special nutritional value ensure them a preferential place in consumption, being considered in the category of relish (Băicoianu Floarea, 2003). They have a complex structure with a direct implication in the human body, bringing vitamins and minerals and with with energetic, refreshing, dietary and therapeutic qualities (Dejeu L., 2011). Grapes are fruit and at the same time a medicine, since they have special therapeutic virtues while also the grapevine and the wine have the properties of natural medicines (Messegue M., 1998; Petrescu E., 2002).

2-3 decennies ago, the surface cultivated in our country with table grape varieties was about 40 000 ht. 60 000 t. of grapes were exported yearly. Presently, this surface decreased at about 10 000 ht with an average production of 90 000 t., and the export is practically non-existent (Dejeu L.,2010). Nowadays, through the import from other countries, especialy from the southern hemisphere (Brazil, Chile, South Africa), grape

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consumption can be planned through the whole year (Dejeu L., 2011). However, the grape consumption per capita is about 5 kg/year (Dejeu L., 2010).

The newly created varieties, homologated during the latest years, but which are only known in the units where they had been created, must be necessarily introduced in culture in order to replace older varieties which do not meet the changing requirements of the consumers (Popa Camelia and coll., 2003). The necessity of extending the period of consuming table grapes and the diversification of the choice constitute the objectives of study for the scientific research (Damian Doina and coll., 2006).

MATERIAL AND METHOD

As geographical location, the Ştefăneşti vineyard lies between 44° 42′ and 44° 55′ northern latitude, at the southern limit of the Cândeşti platform, in the contact zone of the Romanian Field. The viticultural plantations are located at altitudes ranging between 200 and 415 m (maximal altitudes are Izvorani Hill = 415 m and Pietroasa Hill= 325 m).

In the experimental plot located in the representative viticultural area of Muntenia (Ştefăneşti Argeş), observations and determinations have been made as regards the quality of the table grapes of the variety Argessis in the pedoclimatic conditions of the Argeş country. The grapevine plantation has a distance of 2,5 m/ interval and 1,0 m/row (4000 vines/ht), the applied cutting is the Guyot type on semistock, supported by 5 wires espaliers. The parent stock used for grafting the variety Argessis was Kobber 5BB.

The studied variety had been planted in a brown, colluvial, clayey soil, with a medium input of phosphorus and potassium, weakly carbonated, with a low acid Ph (6,2-6,4).

In order to emphasize the agrobiological and technological characteristics of the variety, comparative observations have been made between years 1999-2001 before homologation and between 2009-2011, as regards the phenology, fertility, the productivity, the quantity and the quality of the crop, in a direct relation to the continuously changing climatic factors.

The studied variety is representative for the Muntenia country, where Ştefăneşti vineyard is located. Efforts have been made on the national and international level, in order to obtain early, superior, very productive table grape varieties, resistent to handling and transport, with large cluster and berries, seedless and with an increased biological resistence to harmful environment factors, to diseases and pests (Oprea Şt and coll., 2007; Sestraş R., 2004 cited by Cichi Daniela, 2010).

The description of the studied variety:

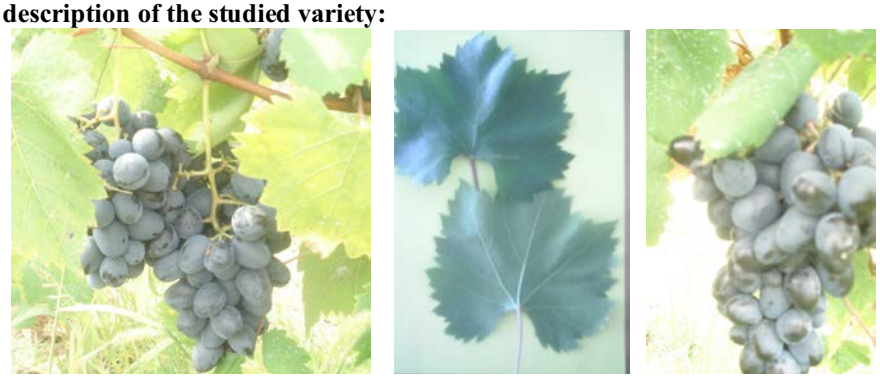

Photo 1. Aspect of the grape and leaf of the variety Argessis

ARGESSIS (photo 1.) – variety homologated in 2002 at SCDVV Ştefăneşti. The first variety of tablegrapes obtained in the pedoclimatic conditions specific to the Ştefăneşti-Argeş vineyard. Pleasant commercial aspect, large berry (7,5-8,0 g), globular, bluish-black. Large cluster (450-480 g), uniaxial. Good tolerance to cryptogamic diseases (mildew, blight, rot) . High vigour vines, suitable for arbor culture (Popa Camelia and coll., 2009). Average production reaches over 15 t/ht .

RESULTS AND DISCUSSIONS

The climatic factors during the studied periods corresponded to years with different weather conditions. The period of the years 1999-2001, before the homologation of this variety, shows seasons with normal temperatures and rainfall, while beginning with the year 2009 the water condition is deficitary during the growing period of the grapes and registered great differences of temperatures in summer and winter. Temperature is an ecological factor which influences the growing and development of plants, the quantity and the quality of the production of grapes (Dobrei A., 2003).

The values of the minimal absolute temperatures in the air were situated within tolerable limits being of -14,5ºC in 2010 and -7,0ºC in 2000. The lowest temperatures have been recorded in 2010 in air (-14,5 ºC) but there wasn't any recorded loss of buds (figure no. 2).

The mean monthly temperature from the vegetation period ranged between 11,0 ºC (2001) and 23,5 ºC (2010). The sum of the global temperatures oscillated between 3321 and 3847. The quantity of rainfall was different from one period to another, affluent in 2001 (623 mm) and deficitary in 2010 (248 mm). The number of insolation hours was an average of 2300, with low values (1596) in 1999.

Thus, taking into account the evolution of climatic changes, the studied variety behaved normally, regardless the recorded temperatures, the recorded quantity of rainfall, and achieved normal sugar accumulations both in the years with rainfall and during those with excess of humidity.

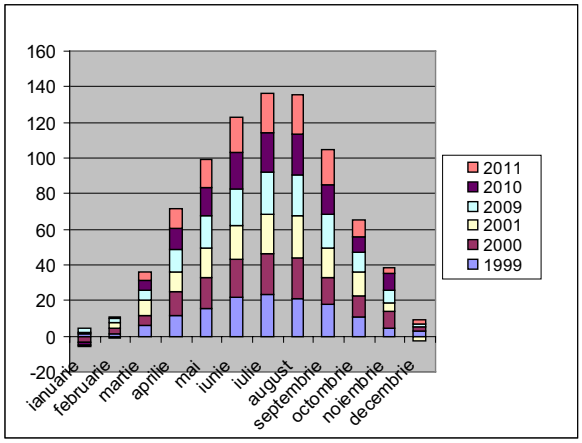

Figure 2. The values of the mean temperatures in air (1999-2001; 2009-2011)

All the data recorded after the observations and determinations concerning the agrobiological and technological characteristics effectuated are influenced by the weather conditions of the mentioned viticultural years.

Determining the fertility of the soil (average over 82%) one can affirm that the soil has a high fertility and the losses of buds were reduced during the winter periods during the years under observation.

The coefficients of fertility (absolute and relative) have been of superior value, ranging between the values 0,46-1,50. This coefficients are useful for the quantization of the qualitative indicators of the production of grapes (Dumitriu I.C., 2008).

Table . 1

The main agrobiological characteristics of the variety Argessis
(1999-2001; 2009-2011)

Investigated element name	**Year research**					
	1999	**2000**	**2001**	**2009**	**2010**	**2011**
Opening buds	21.04	17.04	13.04	18.04	15.04	14.04
Flourished	12.06	29.05	31.05	28.05	01.06	06.06
Early ripening grapes	21.07	20.07	26.07	22.07	21.07	21.07
Consumer maturity	15.09	13.09	18.09	14.09	11.09	12.09
The fertile shoots %	55	77	69	68	79	70
The coefficient of absolute fertility (cfa)	1,50	1,20	1,52	1,20	1,50	1,50
The coefficient of relative fertility (cfr)	0,82	0,46	1,08	1,02	0,79	0,94
The absolute productivity index (ipa)	685	580	553	588	840	846
The relative productivity index (ipr)	375	222	393	499	442	530
The force growth	high	high	high	high	high	High
The frost resistance, % viable buds	90	86	90	82	86	86

The production of grapes per vine and per hectare was superior, with a recorded average of 5,9 kg/vine and 23,6 t/ht calculated production.

The parameters of productivity had mean values of 410 (relative) and 682 (absolute). Technological elements influenced the high productivity of this variety: the weight of the cluster (564 g) and of the berry (8,26 g). The variety distinguished through the size of the cluster and especially through the weight of the berry. The variety has been also studied under the pedoclimatic conditions from Oltenia, where the productivity parameters ranged between 252-548 (ipr) and 500-676 (ipa) (Cichi Daniela and coll., 2011).

The early maturation and the quality of the grapes of this variety has been comparatively studied in two different locations (Banu Marăcine/Ştefăneşti), thus the

maturation epoch was ahead of time with two weeks at Craiova, as compared to Stefăneşti (Vintilescu Monica and coll., 2010).

Table 2

The main technological characteristics of the variety Argessis
(1999-2001; 2009-2011)

Investigated element name	Year research					
	1999	**2000**	**2001**	**2009**	**2010**	**2011**
Average weight of a grape (g)	457	483	364	490	560	564
Weight of hundred grains (g)	775	617	669	784	814	826
Sugars in wine (g/l)	155	169	165	185	180	177
Acidity in wine (g/H_2SO_4)	4,23	4,36	3,42	3,5	3,2	2,3
Production of grapes vine (kg)	4,2	4,9	5,6	5,5	5,9	5,8
Production calculated/ha (t)	16,8	19,6	22,4	22,0	23,6	23,2

The qualitative parameters of the variety have been appreciated through the sugar content and the total acidity of the must. Thus the values had been almost similar in all years (155-185g/l), the corresponding quantity for table grapes; the total acidity in must ranged between 2,30-4,36.

CONCLUSIONS

Under the conditions of the viticultural ecosystem from the Ştefăneşti-Argeş vineyard, variety Argessis recorded a normal cycle of vegetation in both periods of study, thence proving to be resistent to the present climatic factors.

The qualitative and quantitative production of the variety Argessis recommend it as a qualitative and productive variety; it is free from the major viral diseases recorded in the country.

Variety Argessis completes successfully the varied range of the area, as an early variety of table grapes.

The extension in culture of the variety Argessis, is highly recommended in the Muntenia region, but also in other viticultural areas in the country, being a variety with a large cluster and a pleasant commercial aspect.

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THE BEHAVIOUR OF CERTAIN SEEDLESS GRAPEVINE VARIETIES UNDER THE CONDITIONS OF THE STEFANESTI-ARGES VINEYARD

Costescu Adriana[1], Dejeu Liviu[2], Popa Camelia[3]

Key words: *fertility, productivity, seedless varieties, commercial maturity*

ABSTRACT

The dehydrated grapes –the raisins- are energetic, having additional sweetening qualities, being recommended in the treatment of the pulmonary (asthma, bronchitis), kidney and hepatic diseases. The table grapes are named by the International Office of Vine and Wine (IOVW) the fruit of the grapevine, having the special destination to be consumed fresh and is obtained from varieties cultivated to this purpose.

This paper presents the results obtained in the Stefanesti vineyard Argeş as regards the resistance to cold, the fertility and the productivity, the quality and the quantity of the annual grape production for the varieties: Canner, Otilia, Călina, as compared to the witness Perlette. The study has been completed during the period 2009-2011.

INTRODUCTION

The grape is the most complete nutriment from the vegetal world, being compared to mother's milk (Messegue M., 1998). The raisins represent a product which can be easily transported and preserved, obtained from the dehydration of the seedless grapes. Raisins represent a high energetic value nutrient: 3.340 kilocalories/kg (Dejeu L., 2011).

Romanian viticulture includes very few varieties destined to the production of raisins; thus, it is desired that new varieties of this group should be extended in culture and new clones should be selected. Paradoxically, the multiple achievements in the creation of new varieties situate our country on top level in the world (Dejeu L., 2010). Unfortunately, these varieties are not known in the country and abroad, only few of them managed to be known farther that their original areas.

In Romania, the table grapes varieties are less and less cultivated; the year 2008 recorded a production of grapes for fresh consumption of 87,2 thousand tones, far below the mark of the demand for grapes on the Romanian market (Cichi Daniela and coll., 2010), and this production is on the decrease.

A comparative study has been achieved on the experimental plots of INCDBH Stefanesti on the main seedless varieties from the culture, over three years. The varieties

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studied were: Canner, Otilia and Călina as compared to the witness Perlette, a known variety, extended in culture. Mostly, the variety which came into notice was Canner, an seedless variety with middle maturation. The study has been performed over a period of 3 years (2009-2011) and consisted in the ampelographic, agrobiological and technological determination of the varieties, as compared to the witness.

MATERIAL AND METHOD

During the period 2009-2011 research has been made on certain varieties of table grapes and raisins, varieties having valuable agrobiological qualities which can complete the demands of the Romanian market. It is true that these varieties have been obtained long time ago, but they are appreciated also nowadays, except that Romanian producers cannot afford to set seedless varieties surfaces due to the high cost (planting material, labour, support system, etc.) The varieties are in fruition, in an ampelographic collection at I.N.C.D.B.H. Stefănesti. The studied varieties are grafted on the parent stock Kober 5 BB and planted at the distance 2,5 m between rows and 1,2 m on the row. The applied cutting is the Guyot type on high semistocks. The parent stock on which seedless varieties are grafted, gives them vigour in culture, thus the variety Crimson Seedless grafted on 1103 Paulsen has great vigour in culture but also high productivity (Feldberg N.P. and coll., 2007). Under the conditions of the Stefănești vineyard, the parent stock used at grafting gave the vines great culture vigour and also productivity.

Geographical coordinates of the viticultural centre, longitude and latitude.

As regards the geographical location, the Stefănesti vineyard is located between 44°42' and 44°55' northern latitude, at the southern limit of the platform Cândesti, in the contact zone with Câmpia Română. The viticultural plantations are located at altitudes between 200 and 415m, the maximal altitudes being the Izvorani Hill (415m) and the Pietroasa Hill (325m).

The meteorological data have been extracted from the database of the Stefanesti Institute, collected during the interval 2009 - 2011 (3 years).

The average, climatic conditions over three years (2009-2011): thermal global balance, active and effective: thermal sum (ºC) effective: 1.742; thermal sum (ºC) active: 3.712. The absolute minimal temperature was -21,4 ºC (24.01.2009); real solar heat gain: 1764 hours; rainfall: total viticultural year 566 l; total during the vegetation period 423 l. Number of days of bioactive period 197. Observations and determinations have been made on the culture vigour of vines, on the development of the main phenophases, the duration of the vegetation period, the absolute and relative fertility coefficients have been calculated, the productivity parameters, the quality and quantity of the production, the resistance to cold.

RESULTS AND DISCUSSION

Variety description:

CANNER (figure 1)**:** is a variety obtained by OLMO in 1969, by cross-breeding the varieties Hunisa x Sultanina. It is a variety with white, large, oval, seedless grapes which can be used for raisins, conserves and stew.

The grape is large, branchy, lax. *The berry* is medium-sized, elliptic has yellow-greenish colour, golden on the sunny side. Fleshy pulp, crisp with rudiments of seeds. The variety falls in the group of the varieties having a very high vigour of culture. It has a

medium tolerance to low temperatures during winter, to mildew and rot. Maturation in phase IV.


Figure 1 - Variety Canner

OTILIA (figure 2)**:** variety obtained at SCDVV Pietroasa through the sexuate hybridation of the varieties Alphonse Lavallee x Perlette, homologated in 1987; apirene variety for tablegrapes and for industrialization (sraisins, conserves, stew); medium-sized grapes (297 g average), conical, biwinged or triwinged, semicompacted, very pleasant aspect. Medium, globular, slightly oval, with black-violaceous rind, elastic, covered with a thick layer of bloom. Crisp, seedless pulp or with rudiments of green seeds. Otilia gives good results in the viticultural regions from the southern part of the country, mainly for the production of the early table grapes. It can also ensure availabilities for the export of grapes, production: 16.7 t/ht (Ţârdea C., Rotaru Liliana, 2003).


Figure 2- Variety Otilia Figure 3 -Variety Calina Figure 4 - Variety Perlette

CĂLINA (figure 3)**:** variety obtained at SCDVV Dragasani, through the controlled sexuate hybridation of the varieties Braghină x Sultanină (MĂRCULESCU M.). The homologation of the variety has been made in 1985. It is the first seedless variety created in our country and introduced in culture. It corresponds to the apireny of the "Sultanina" type. The grapes are long (30 – 38 cm), large (250 g average), branchy, semicompacted, with a lignified stem. The berry is small (1,7 g), oval, having a pink colour in different shades, according to the maturation degree and to the exposure to heat. The rind is thin and covered with a fine layer of bloom. The pulp is juicy, seedless or with undeveloped rudiments of seeds. Călina has been introduced in culture in the viticultural regions in the southern part of the country (Ţârdea C., Rotaru Liliana, 2003).

PERLETTE (figure 4)**:** has been obtained at Davis University in California, through the hibridation of the varieties Queen of vineyards x White Sultanină (H. P.OLMO, 1936). It belongs to the same group with the varieties Delight and Beauty Seedless. It can

be noticed through early maturation and the commercial value of the grapes. The grapes are uniaxial, taper, medium-sized (240 – 380 g, average), compacted. The berry is medium-sized (2,65 – 5,39 g), globular, white-greenish due to the thick layer of bloom; the pulp is fleshy, crisp, not flavoured. The stem and the cob are herbaceous (nonlignified). Biological resistances: sensitive to cold (-16°C . . . -18°C): very sensitive to mildew; splits the matured berries very easily, even at reduced rainfall; is attacked by wasps (Dobrei A. and coll., 2008).

The development of the phenophases is synchronized multiply by the influence of the climatic factors. The table nr.1 shows the data of the main phenophases during the three years

Table no. 1

Dynamics of the vegetation phenophases (2009-2011)

Nr. crt.	Name of the variety	Debudding	Blooming	Ripening	Full maturity
1.	Canner	13.04-23.04	26.05-30.05	20.07-28.07	05.09-09.09
2.	Otilia	13.04-17.04	20.05-27.05	28.07-30.07	09.09-12.09
3.	Călina	11.04-17.04	19.05-25.05	29.07-31.07	18.09-21.09
4.	Perlette (witness)	09.04-18.04	24.05-30.05	15.07-25.07	19.08-28.08

Theis table shows clearly that at all the studied varieties, the main phenophases had a similar period, namely the debudding at all the 4 varieties took place in April; blooming in May; maturation between 15-31.07 and harvest between 19.08-21.09, specific to the seedless table grape varieties.

The varieties Călina and Otilia have been studied at the Experimental Vitivinicultural Station Iasi as compared to the variety Perlette, where the vegetative growths had been very extensive; the maturation epochs had been recorded at mid-August for the variety Otilia and end of September for the variety Călina; the average weight of 100 berries is 204 g. at the variety Otilia and 222 g at the variety Călina (Damian Doina and coll., 1998).

Table no. 2

Quantity and quality of production (average years 2009-2011)

Nr. crt.	Name of variety	Mass of 100 grapes (g)	Production kg/vine.	Weight of a grape (g)	Quality of the production	
					Sugar g/l	Acidity g/l H_2SO_4
1.	Canner	430	5,1	462	189	4,1
2.	Otilia	220	4,8	285	166	3,4
3.	Călina	218	4,5	333	203	3,9
4.	Perlette (witness)	209	4,0	327	135	4,7

The most appreciable production had been recorded at the variety Canner, both on the vine and at hectare (15,3t/ht); mass of 100 berries had the highest value at the variety

Canner (430g), and the lowest at the variety Perlette (209 g); the wight of a grape ranged between 285 g at the variety Otilia and 462 g t the variety Canner; as regards quality the variety Canner recorded a content in sugars of 189 g/l, and the variety Perlette only 135 g/l. All these data contributed to the qualitative characteristics of the varieties. The variety Canner, as compared to other seedless varieties, according to the research, proved the best behaviour, recording productions of 5,5 kg/but., sugars of over 220 g/l (Costescu Adriana and coll.2012).

The temperatures registered during the winters of the years 2009-2011 did not exceed the frost limit for the grapevine, thus all the studied varieties had a normal viability (over 75%), normal cuttings of production have been applied and the quantity/quality had been those planned for the variety. Generally, the seedless table grapes are less resistant to low temperatures.

Fertility at the grapevine must be understood as a fundamental characteristic by which fructification organs are formed yearly, as an initial basis in obtaining the grape crop. Although it has an unitary character, fertility can be appreciated and interpreted in terms of potential fertility and real fertility (Dumitriu IC., 2008). Real fertility depend on potential fertility and is expressed through the percentage of fertile shoots and the absolute and relative fertility coefficient (Pop N., 2003).

The fertility of the studied varieties is shown in fig. 5, thus the variety Perlette had the lowest absolute and relative fertility coefficient during the period of the study and the varieties Canner and Otilia recorded the highest fertility coefficient.

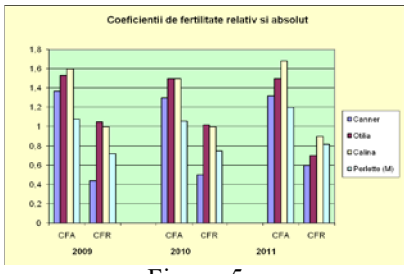

Figure. 5
Fertility coefficients

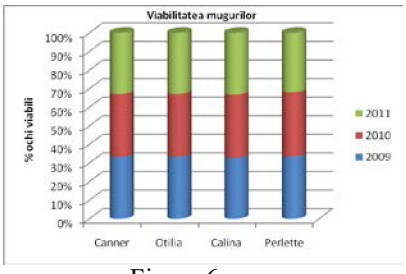

Figure 6
The viability of the buds

The value of the viability percentage of the buds demonstrated the variety Canner has a medium resistance to cold, which is specific to seedless varieties (Necula Cezarina and coll., 2010). The four studied varieties presented over 75% viable eyeholes (fig. 6).

The production of grapes constitutes the basic indicator in setting agrotechnical measures applied in a grapevine plantation.

The quantity of the crop depends on he size of the grapes and of the number of inflorescences, thus the varieties which had the same number of fertile shoots or the same fertility coefficient (Canner, Otilia), gave different crops in production (Călina - 4,5 kg/stock., Canner - 5,1 kg/stock.) .

As regards the weight of 100 berries, the differences are significant, varying between 218 g at the variety Călina and 430 g at the variety Canner as compared to the witness Perlette with an average of 209 g. The variety Canner had the most appreciable weight of the grape. In kindly years for viticulture, the berry of this variety exceeds 7 g, a value rarely met at seedless varieties.

The content of the must in sugars id specific to the group of the grapes for conserves, with medium maturation. Appreciable accumulation of sugars had been recorded to the variety Călina - 203 g/l and the lowest 135 g/l at the witness variety Perlette.

The acidity of the must had been higher on the average at the variety Perlette (4,7 g/l H_2SO_4), and the lowest to the variety Călina (3,4 g/l H_2SO_4).

CONCLUSIONS

Under the climatic conditions of the viticultural centre Stefănesti-Arges , the seedless variety Canner had the best behaviour, being distinguished through the size of the grapes and of the production. Consequently, it has to be studied thenceforth, in order to obtain a clone.

It is recommended that seedless varieties should be used in hybrid combinations with a view to create new varieties.

All the 4 studied varieties had remarkable qualities in order to be cultivated for fresh consumption and for the production of raisins, conserves and stews.

The private producers should be oriented towards the growing of the Romanian quality table grape varieties.

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THE INFLUENCE OF FERTILIZATION WITH THE LATEST GENERATION OF MANURE TO THE PRODUCTION AND ITS QUALITY IN FIELD GROWN PEPPERS

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Key words: *pepper, the latest generation fertilizers.*

ABSTRACT

Pepper crop fertilization is a sure way to increase production because this species reacts strongly to fertilization of the land culture through the use of organic and chemical fertilizers (Andronicescu D., Anghelescu H. , 1968).

In the current situation, in the south part of Romania, manure is available in quantities increasingly smaller and with prices increasingly higher.

Starting from this reality, the scientific work that we are presenting, deals with the basic fertilization and the first phase of peppers in the field using last generation complex fertilizers in order to achieve productions over 7kg/m2 in cultivars which are commonly used in the vegetable basin in Braila.

INTRODUCTION

In recent years, the Romanian chemical fertilizers market, besides the conventional, such as the 15 N-15 P2O5-15 K2O, the offer includes controlled release granular fertilizer complex (Agroblen - used for basis fertilization) and complex soluble fertilizers with microelements applied by fertigation (range Universol, Scotts products), (Davidescu, D., Davidescu Velicica, 1992).

Such fertilizers were used to develop experimental variants, which are presented in the following.

MATERIALS AND METHODS

The experimental variants on which we worked are presented in Table 1.

To achieve experimental factor graduations "fertilization" these fertilizers were used:

Complex III - the fertilizer complex III produced in our country, with the following percentages of active substance 15% N, 15% P2O5, 15% K2O.

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Table 1

The experimental variants
Field pepper fertilization, in Braila 2011

Var. no.	Fertilization		Cultivars
	Basic	1st phase	
V1(mt)	Complex III – 250 kg/ha 15N-15P-15K	Complex III 15N-15P-15K	Yellow Superior
V2			Bianca F1
V3			Atris F1
V4	Agroblen total(5 months) 20N-10P-10K-4MgO 0,04 kg/m^2	Blue Universol 18N-11P-18K-2MgO-Microelements 8g/m^2/week Purple Universol 9N-9P-9K-3MgO-Microelements 15g/m^2/week	Yellow Superior
V5			Bianca F1
V6			Atris F1
V7	Yellow Universol 12N-30P-12K-2MgO-microelements 15g/m^2/week	Purple Universol 9N-9P-9K-3MgO-Microelements 15g/m^2/week	Yellow Superior
V8			Bianca F1
V9			Atris F1

Agroblen total: basic fertilizer for vegetable crops that are harvested mainly fruits (tomatoes, peppers, cucumbers and so on). The main macro-elements NPK, are partially film-controlled formula that extends the period of their release to the root, providing the necessary development throughout the plant, phosphorus, much needed during flowering, or potassium, necessary in the period of fructification. Longevity fertilization is 5-6 months, the formula 20 +10 10 4 MgO (N 95%, P2O5 50%, K2O 100% A% MgO).

Universol: permeability and excellent solubility make use of high performance on all types of crops can be applied to both foliar and soil, by all methods of irrigation (Buzescu, D., 1994).

Universol contains all the necessary elements that a plant needs: nitrogen, phosphorus, potassium, magnesium and trace elements binders of. To ensure rapid solubility and total absorption, all the formulas Universol have in their composition citric acid and trace elements are chelated EDTA.

Universol blue: 18 +11 +18 - 8g/m2 / week
Universol yellow: 12+30+12 - 15g/m2 / week
Universol violet: 9+9+27 - 15g / m 2 / week.

Blue Universol balanced formula - universal. Use during the period of vegetation provides balanced nutrition, leading to a harmonious development until the formation of the first fruits on plants, and beyond. Universol purple, with higher potassium content very well support fructification, ensuring a high level of production and quality of fruits.

The Yellow Universol with a high content of phosphorus, used after crop establishment or flowering. It favors the root growth and development. It stimulates flowering process.

It was made an experience bi-factorial type 3x3 with 9 variants, nonrandomized blocks mounted with 4 repetitions. Witness was set V1.

In the experience were made following cultivars:

- Yellow Superior: Variety native, created the ICLF Vidra, large fruit, bright yellow, elongated cone. In medium conditions production is about 35-40 t / ha with average fruit weight of 100-110 g.

- Bianca: A pepper successfully cultivated in Romania widely in greenhouses, solariums and field. Very early with good fruiting capacity, large fruit with 3-4 lobes white to yellow. Average fruit weight of 150-200 g has great power growth, short internodes and strong root system giving a good balance between growth and fruiting. Protected crops, 40% of the total production is obtained from the first harvest. High production potential. Respond well to technology applied correctly.

- Atris: pimiento is a hybrid type, early and highly productive fruit conical, elongated and rapid ripening from green to red. Fruits long (15-20 cm), straight, thick and extremely uniform pulp. The plant is strong cord does not need to field crops. Foliage is rich, balanced fruit, with a length of 19 to 22 cm and the shoulder diameter 5-6 cm. Is a suitable material for paprika processing but also for fresh consumption (The official catalog of varieties (hybrids) of crops 2005).

TERMS OF EXPERIMENTATION. In 2011, the first year of research, the experience has been made on chernozem soil type in the county of Braila (SC Agroleg Silistraru), in an area dedicated for growing vegetables. Soils are classified in class of mollic soils, represented by typical chernozem, chernozem cambic characters vermouth, carbonated poorly formed on loess and loess. Soils are natural chemical and biological attributes gives them high fertility, so are part of the best soils.

Specific elements of culture technology applied in the experience:

The autumn furrow to a depth of 28-30 cm , with incorporation of crop residues.

In spring, Harrow shredding and loosen the soil surface.

Seedlings were produced on a bed of peat, (Biolan) with a density of 400 yarns / m2 (Florescu, E., Popescu, V., Ciofu, R., Atanasiu, N., 1998).

The experimental crop has been planted in late May (approx. 18 to 20 May) in equidistant rows with 70 cm between rows and 20 cm between plants in the row, distances providing a density of 7.14 plants/m2(Atanasiu N., 2005).

Besides basic fertilization, which was performed according to the variants shown in the Table 1, in culture experiments were conducted following works:

- Filling gaps;
- Hoeing to control weeds and loosen soil;
- Phytosanitary treatments for preventing and combating attacks from diseases (mildew - Leveillula Taurus) and pests (thrips - Thrips ssp). Fighting powdery mildew was made with substance Rubigan 12 EC 0.04% and thrips were fought Admiral 10 EC 0.05% (Costache, M., Roman, T., 1998).

Harvesting, differentials plot was made nominal gradually, with counting and weighing all fruit valuable for consumption in fresh condition, technological maturity for cultivars Superior Yellow and Bianca and physiological maturity for Atrix F1.

In the experience were made observations and measurements to determine the dynamics of vegetative increase, growth and harvest production, total production and its quality (Ciofu, R. et al, 2004).

RESULTS AND DISCUSSION

In this first year of research (2011) we have given a great importance to production results, which are presented and discussed briefly in the following.

Results on production components are shown in Table 2.

Table 2

Production per plant and per unit area
Field pepper fertilization, in Braila 2011

Var. no.	Fertilization	Cultivars	Density Plants/m^2	Production Kg/plant	Production Kg/m^2	Production, no. of fruits STAS/plant	Average fruit weight (g /fruit)
	Conventional Complex3	Yellow Superior	7,14	0,618	4,415	5,80	106,8
		Bianca	7,14	0,766	5,471	6,81	112,5
		Atris	7,14	0,930	6,640	4,43	210,1
	Agroblen +Universol	Galben Superior	7,14	0,675	4,820	6,11	110,4
		Bianca	7,14	0,835	5,964	7,02	119,0
		Atris	7,14	0,968	6,916	4,20	230,5
	Universol	Yellow superior	7,14	0,786	5,618	6,66	118,1
		Bianca	7,14	0,890	6,340	7,04	126,4
		Atris	7,14	1,100	7,860	4,65	236,7

The quantities of peppers harvested Dynamics were combined and reported as total production in kg/m2 ranging between 4,415 kg/m^2 to Yellow Superior and 7,860 kg/m^2 to Atris F1.

Subsequently, depending on the crop density (7.14 pl/m^2) and number of fruit harvested on variants were calculated average weight of fruit, which varies between 106.8g/m^2 in Superior Yellow and 236.7 g/m^2 Atris F1 ..

And depending on the average weight of fruit, the 3 cultivars can rank, in accordance with current standards of quality.

It is also found differences in production created by fertilization differentiated.

Using total production calculated on variations and repetitions, was conducted a statistical calculation of two-way analysis of specific variation experiments. Between the synthesis made along the multiple comparison , we have chosen for presentation in this paper those within which are examines the influence of cultivar and fertilization influence on production.

Summary data presented in Tables 3 and 4 highlight the following aspects:

Equipment and fertilization regime influence significantly distinct production realized in variant 3 (Table 3). On the average variable, average production of the three cultivars is 1.098 kg/m2 higher than similar variant fertilized with Complex III. Distinct positive difference above is significant.

The influence of assortment (of cultivar) is marked by good and very good productivity of the 2 hybrids-Bianca F1 and Atris F1. Thus, Bianca F1 the production of

5925 kg/m2 is with 0.974 kg/m2 higher than production similar Yellow Superior variety. This is distinct significant positive difference.

Table 3

Summary of experimental results
Influence of fertilization on the production technology,
Pepper, Braila 2011

Nr. crt.	Fertilization technology	The average production of the 3 cultivars Kg/m2	Production %	Output gap		The meaning
				Kg/m^2	%	
	Complex 3	5,508	100,00	-	-	-
	Agroblen+Universol	5,900	107,2	+0,392	+7,2	-
	Universol	6,606	119,34	+1,098	+19,34	xx

DL – 5%- 0,415 kg/m^2
DL – 1% - 0,824 kg/m^2
DL – 0.1% - 1,261 kg/m^2

Table 4

Summary of experimental results
Influence of the assortment over the production
Pepper, Braila 2011

No. crt.	Assortment (pepper cultivars)	Average production of the three fertilization technologies kg/m^2	Production %	Output gap		The meaning
				Kg/m^2	%	
1	Yellow Superior	4,951	4,951	100,00	-	-
2	Bianca	5,925	5,925	119,67	0,974	+19,67
3	Atris	7,138	7,138	144,17	7,187	+44,17

DL-5%-0,415 kg/m^2
DL-1%-0,824 kg/m^2
DL-0,1%- 1,261 kg/m^2

If Atris F1 hybrid, the production of 7.138 kg/m^2 exceeds with 2.187 kg/m2 the similar production of Yellow Superior. This positive difference is very significant.

CONCLUSIONS

The assortment of peppers recently introduced in culture in vegetable Basin Braila is certainly more productive than some local varieties grown in the area. Thus, Bianca Atris

F1 and at F1 Of production differences from controls (0.974 kg/m^2 and 2.187 kg/m^2) are distinct significant, very significant respectively.

Of production the differences mentioned are due first of all to average fruit weight (111.7 g Yellow Superior 119.3 g and 225.7 g Bianca Atris F1 F1).

The number of fruits per plant ranges from 4.43 in relatively low 4.65 V3 and in V9 for Atris F1 cultivar is offset by their average weight (210.1 g / fruit, or 236.7 g / fruit).

Fertilization with advanced manure of recent generation contributes to the increase significantly distinct in production compared with that achieved in the control.

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RESEARCH REGARDING THE ECOLOGICAL CULTIVATION OF PEPPER IN FIELD

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Key words: *pepper, organic culture in the field.*

ABSTRACT

On global horticulture production, European and national, was found in the last few years an increased interest of the producers and the consumers for organic fresh food, of which, pepper is well appreciated due to its nutrients. Although Romania is part of this trend, organic pepper is currently one of the species that local producers have relatively less taken care of in the last years.

Full paper shows the results obtained from organic farming in the south of our country.

INTRODUCTION

The specialty literature from countries with advanced agricultural technology, treats, in the last few years the specific problems of farming. (Stoian 2009, SILGUY 1999).

From the vegetable species grown organic, field pepper has been neglected in the last years comparison with other species from vegetable native assortment.

Because of this, in the last 2-3 years have seen a regression of acreage in this species, compared with tomatoes, brassicaceae's family species, root vegetables and some aromatic species, although there is clear legislation that regulates this issue and country Our. (REGULATION (EC) NO. 889/2008 of 5 September 2008 laying down detailed rules for implementing Regulation (EC) no. 834/2007 on organic production and labeling of organic products with regard to organic production, labeling and control.

Emergency Ordinance no. 34 of the 17 April 2000 on organic foods.

The objectives of the research presented in the paper were:

Study the behavior of pepper cultivars that are suitable for organic cultivation;

Determination of organic crop production realized in field pepper.

I approached this issue to determine how to implement the technology applied to this culture in France. (Otto Schmid, Henggeler, 2000).

Objectives and organizing research presented consistent in the literature.

The experimental variants are presented in Table 1

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Table 1

The experimental variants
Peppers grown in the field, the ecological culture 2011

Var. no.	Cultivar (a)	Crop systems
1	Yellow Superior (a1)	Conventional crop
2		Organic crop
3	California Wonder (a2)	Conventional crop
4		Organic crop
5	Atris F1 (a3)	Conventional crop
6		Organic crop

The 6 variants were performed using 3 cultivars: Yellow Superior, California Wonder, Atris F1 culture two separate technologies: conventional and organic.

The experience two-way, such as 3x2with 6 variants has been fitted in an area dedicated to vegetable production, as follows:

- On the organic parcel, versions 2.4 and 6.
- On the conventional parcel (separated from organic) versions 1.3 and 5.

The 4 repetitions were not randomized.

MATERIALS AND METHODS

In our experience these cultivars were used:

Yellow superior: semi-early variety of sweet pepper, weighing 110-120 g per fruit. Eaten at the maturity the color is light yellow, and at physiological maturity the color is red. Vigorous shrub with a height of 55-60 cm. Fruits with 3-4 lobes are pointed, conical, 8-10 cm long and 6.5-7 cm in diameter. Pulp thickness is 7.5-8 cm. Production potential is 35-40 t / ha.

Atris F1: is a hybrid of pimiento type, early and highly productive fruit conical, elongated and rapid ripening from green to red. Fruits long (15-20 cm), straight, thick and extremely uniform pulp. The plant is vigorous, with no need of trellising to field crops. Culture in greenhouses: trellising and weeding are required. The foliage is rich, balanced fruit, with a length of 19 to 22 cm and a maximum diameter of 5-6 cm. Is a suitable material for paprika processing but also for fresh consumption.

California Wonder: pepper, late growing determined.

Plant height of 50-80 cm, is very vigorous and fast growing.

The fruit is with four lobes, blocky type, with sweet and firm flesh. Growing season: 75 - 80 days.

Fruit weight: 130-150 g fruit color: dark green at maturity of consumption red at the physiological maturity. Way of culture: through seedling. Culture type: in field, protected.

For plant health control, were used intercropping aromatic plants (Sage - Salvia officinalis, Lamiaceae's family. Good protection against insects and fungicide.).

Intercalation with herbs such crops, was done in three ways:

- Balanced rows of peppers. 70 cm between rows and 20 cm between plants in the row. On every second interval between rows of peppers, sage was planted.

- 70/20 cm along the row, 2 sage plants after 5 pepper plants.

As witnesses we used 2 ways: superior yellow in organic culture with sage stripes and conventional culture with the same kind located on a separate parcel, at a vegetable grower from the neighbor.

Besides cultivars presented, to achieve the experience, decoctions and infusions were used for foliar treatments. The materials used for this purpose are:

- Nettle (Urtica dioica). In organic agriculture, nettle is recommended as bio-stimulant, treatment against aphids, acaricide and as an adjunct in compost fermentation. Aggressively macerate of nettle: macerate 1 kg of fresh plant in 10 liters of water. Plastic vessel is exposed to sunlight for 4-5 days. A quart of nettle macerate is diluted 50 times and sprinkle seedlings and leaves before blooming the flowers for the combating aphids and mites.

Fermented nettle soak: Use the same proportions, just let it soak for 10-15 days until the water is no more spewing, mixing it every day. Diluted 10%, can be used as plant growth stimulant by watering their roots.

- Wormwood (Artemisia absinthium). Wormwood contains, besides an alkaloid, absinthin responsible for insecticide effect, and resins, tannin substances, organic acids and nitrates. Maceration of wormwood is obtained from 3 kg fresh plant floriferous stems in 100 l water. After 2-3 weeks is filtered and stored in tightly closed plastic containers up to 2 years. Maceration thus obtained can be used in combating aphids and ants in horticultural crops. (Bertrand, Collaret, Petiot 2004).

In the experience were used observations and measurements regarding the dynamics production, the total productivity and its quality in the culture and the availability of certain populations of pests (thrips, Thrips ssp). Thrips are polyphagous insects. insect measuring 1-2 mm long, yellow-brown or dark. usually they have three generations over a summer. Thrips maggots are light colored. Adults and maggots attacks on the underside of the leaves (Candea, 1986).

The specific elements of culture technology applied in experience:

For the versions with conventional technology we respected the current recommendations concerning pepper crop in the field.

For the organic technology, located on a certificate lot the main differences from conventional are:

- Strict adherence to crop rotation;
- Basic fertilization with manure came from breeders and producers of beef meat and milk;
- Remove in all stages of the crop the production of pollutant materials that are not allowed in organic crops.

In the experience were made observations and measurements concerning growth and fructification of pepper from conventional crops and organic crops.

RESULTS AND DISCUSSION

To achieve the experience we paid special attention to production results, which are presented in Table 2.

Table 2

The total production kg / plant
Organic and conventional crops of peppers in the field – 2011

Var. no.	Cultivar	Tehnologie	Total Production		The output gap kg/m2	
			Kg/plant	Kg/m^2	From mt1	From mt 2
1(mt1)	Yellow Superior	Conventional	0,642	4,583	-	+1,228
2(mt2)		Organic	0,470	3,355	-1,228	-
3	California Wonder	Conventional	0,716	5,112	+0,529	+1,757
4		Organic	0,508	3,627	-0,956	+0,272
5	Atris F1	Conventional	0,894	6,385	+1,802	+3,030
6		Organic	0,581	4,147	-0,436	+0,792

Table 3

Average weight and number of marketable fruits per plant
Organic and conventional crops of peppers in the field – 2011

Var. no.	Cultivar	Technology	Kg/plant	Number of fruits / plant	Average weight (g / fruit)
1	Yellow Superior	Conventional	0,642	5,96	107,7
2		Organic	0,470	5,14	91,4
3	California Wonder	Conventional	0,716	4,16	172,1
4		Organic	0,508	3,49	145,6
5	Atris F1	Conventional	0,894	4,82	185,4
6		Organic	0,581	3,83	151,4

The data from table 4, the synthesis of the experimental results, highlight the production differences in between cultivations technology. Thus, it is noted that the cultivars with conventional production technology is significantly higher than cultivars with green technology. The differences are observed and at cultivars California Wonder and Atris F1. Thus the plant production in kg is significantly higher than the witness Yellow Superior making a difference size and weight of fruit (table 3).

Table 4

Synthesis of the experimental results
The total production kg / plant

Var. no.	Cultivar	Technology	Kg/plant	Difference from MT1 (kg / plant)	The meaning	Difference from MT2 (kg /plant)	The meaning
V1 (mt1)	Yellow Superior	Conventional	0,642	-	-	+0,172	xxx
V2 (mt2)		Organic	0,470	-0,172	ooo	-	-
V3	California Wonder	Conventional	0,716	+0,074	x	+0,246	xxx
V4		Organic	0,508	-0,134	oo	-0,038	-
V5	Atris F1	Conventional	0,894	+0,252	xxx	+0,424	xxx
V6		Organic	0,581	-0,061	-	+0,111	x

DL – 5% - 0,072
DL – 1% - 0,121
DL – 0, 1% - 0,156

CONCLUSIONS

New range introduced in culture is more productive than native range.

Although the production difference is significant in cultivars with conventional technology, high demand toward consumption of organic vegetables, including peppers, allow us to further cultivate this plant. The production differences can be recovered as costs easily obviously higher market price for organic products.

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✔*Ingineria mediului*

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EFFECT OF CARROT SEED WETTING WITH BIOSTIMULATING SUBSTANCES ON CROP PRODUCTION IN EARLY FIELD

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Keywords: *Daucus carota L., wetting, humic acids, Vitis vinifera seed extract, production*

ABSTRACT

This study was carried out in the field educational at the Faculty of Agriculture and Horticulture in Craiova. The purpose of this paper is the cumulative effect of plant growth substances with humic acids and antioxidants from the seeds of Vitis vinifera on the production of thick roots of carrot that came from early culture (with moistening seeds for 6 and 12 hours).

It was found that the process of wetting of seeds caused substantial production increases, the wetting time is critical in the production of seeds produced. It also shows that the seed's biostimulator extracted from Vitis vinifera determine differences in production of 1.6 t / ha after 6 hours of wetting of seeds and 6.2 t / ha after 12 hours of wetting of seeds.

Production increases, particularly noteworthy were obtained and variants wetted with humic acids (3.4 to 5.0 t / ha) due to the combined effect of the two factors.

INTRODUCTION

Carrot is one of the most important vegetable species from which we consume thickened root, fresh, and we use also as raw material for canning and pharmaceutical industries, because of dietetic and therapeutic qualities.

It is well suited to culture in the open field. Originated in Mediterranean Sea area and Black Sea region spreading out then in time of the Romans, in North Africa, Asia and then later in America (Ciofu, 2004).

In our country, in 2011 the culture occupied an area of 16,000 ha and there has been a production of 245,000 t (INS press release nr.70/2012).

Application of growth regulators for both the seed germination as well as for supplementary fertilization is a key objective in organic farming, both in our country and around the world (Kähkönen et al., 1999; Vijayakumar et al., 2009; Dinu et al., 2009.2010 ,).

Humic substances are found in all types of soil or water, and are the result of decomposition of plant products.

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Seed treatment with a dilute solution humic acids stimulates the cell membrane and the metabolic activity leading to the a higher percentage of germination.

The important early period of growth, humic acids promote root system development, support and consolidate extraction of nutrients and the healthy development of the plant (Albayrak 2005, Popov 2008). Shall enlarge the absorption capacity of roots, leading to increased crop by 30%.

The use of humic acids enlarge the effectiveness of fertilizer and nutrient uptake by the root system. (Pena-Mendez et al. 2005) This can reduce the required amount of fertilizer by up to 30%, by default reduction of production costs. Lowering the amount of fertilizer is beneficial effect also on the environment.

The humic acids can be an excellent foliar fertilizer with action on the development of leaves, roots and fruits (Albayrac 2005). He contributes to plant growth by acting on the carbohydrate content of the leaves and stem. These carbohydrates are transported to the roots through the stem once they arrived are released into the soil for use as nutrients for organisms (Kulikova et al. 2005);

Foliar application of the humic acid based products promotes photosynthesis and improves plant growth and development. Also increases tolerance to stress factors (biotic and abiotic), including infection with phytopathogenic agents. In addition, the humic acids act as natural chelating agent or complexing agent for those substances that contribute to plant nutrition.

Using the Vitis vinifera seed extract (Bita, 2008) in the carrot seed germination and vegetation in the plant is due to its rich content of polyphenols (5-8%) (Thorngate et al., 1994; Prieur et al., 2000, Shiraz et al., 2000). Antioxidant activity is 20 times greater than vitamin C and 50 times stronger than vitamin E (Shi et al., 2003). The polyphenols from Vitis vinifera extract contribute to the protection of the seed's compounds during germination and plant development from oxidative reactions that occur over time, the reaction may be due to lack of moisture, temperature, nutrients, etc..

By using polyphenolic extract is increased the concentration in the plant of polyphenolic compounds this concentration has beneficial effects in the development of the plant:

- Protects plants from UV that affects DNA, proteins and cell membranes, leading to altered metabolism by generating reactive oxygen species (ROS) acting as a screen inside the cell layer (Vinkel, 2002; Carletti et al. , 2003);

- Are involved in plant defense mechanism against pathogens, insects, herbivores, etc.. (Groger and Kokubun, 2001; Lattanzi et al., 1994; Huitema et al., 2003, Mysore and Ryn, 2004; Nurnberger and Lipka, 2005);

- Have a positive effect on the process of pollination of flowers by shades of pigment given by the content in anthocyanin (Harborne and Grayer, 1993).

- Contribute to the increase of the auxins who have an important role in plant growth (Stafford, 1991, Mahesius, 2001).

- Manage the dynamics of organic matter in the soil, causing its accumulation in the soil, and ion exchange capacity increases with reducing the loss of the nutrient's essential cations.

By increasing the production, improving the quality and reducing the fertilizer costs, the profit resulted from the use of products based on humic acids and polyphenols from seeds of *Vitis vinifera* is obvious. These benefits, economically and environmentally, arising only through long-term use of these products. All these are the guarantee of successful crops, whatever soil type, location or method of culture.

MATERIAL AND METHODS

The experience was bifactorial the factor A was represented time of wetting of the seed and had two graduations, a1-wetted seeds for 6 hours, a2 - wetted seeds for 12 hours and factor B was the Biostimulator used to the seed germination and had three graduations, b1 - water, b2 - humic acids (HA) and b3 - Vitis vinifera seed extract (ESVv). We study the hybrid F1 Danvers.

Carrot seeds were moistened in water, humic acid solution and the Vitis vinifera seed extract for a period of time of 6 and 12 hours. The seeds were placed in gauze bags in equal amounts in each bag (10 g of seeds) to provide approximately the same number of plants per unit area. After every two hours with seed bags were removed from the solution for half an hour for ventilation seeds.

After moistening the seed before planting, they were left to be dry while they were prepared the furrows of seeding, (this is performed on 26.02.2010).

In the vegetation were carried out the specific maintenance works of carrot culture established in the field.

Harvesting was carried out in the second decade of June.

RESULTS AND DISCUSSIONS

After harvesting of carrot roots thick observations and measurements were made about the quality and quantity of the production achieved. Regarding the amount of carrots in Tables 1-3 refer to the products obtained under the influence of factor A (wetting time seed), of the factor B (Biostimulator used for moistening seeds) but also because of the combined effect of the two factors .

Regarding the total production of carrot the seed obtained as a result of wetting is observed that after 6 hours after the momment when seed has been wetted we obtain a production of 15.04 t / ha while after the wetting for 12 hours we obtain a production increase with 3.36 t / ha compared to the control (table 1).

To the production achieved as a result of stimulation of the seed with water, humic acids and the Vitis vinifera seed extract we found that the humic acids determines a production increase with 2.80 t / ha, while seed polyphenol extract of Vitis vinifera, we obtain a difference of production of 1.60 t / ha relative to referential stimulated only with water where the production obtained was 18.40 t / ha. (Table 2).

The combined effect of these two factors (A x B) resulted in yield increases ranging from 2.0 t / ha and 6.1 t / ha (Table 3). Highest yield was obtained in variants where seed has been wetted for 12 hours with the Vitis vinifera seed extract and humic acids.

It is noted that biostimulator used affects the seed germination phenomenon due to absorption of humic substances, which, as you get inside the cells increases respiration rate and cell division is accelerated. Same respiratory processes causes the root meristem formation. An important role in the germination of the seeds of this species had their time of wetting.

Doing an analysis of factor B graduations in relation with graduations of factor A we note that the crop obtained at 12 hours of wetting recorded the increases of production ranging from 2.5 to 3.4 t / ha compared to yields obtained after 6 hours of wetting seeds.

Table 1

The significance of differences in production of factor A

Fact. A	Significance	Total production (t/ha)	Relative production (%)	± Relative to control (t/ha)	Statistical assurance
a_1	Carrot seeds moistened 6 ore	15,04	100,0	Control	-
a_2	Carrot seeds moistened 12 ore	18,40	122,34	3,36	x

DL 5% = 2,92 t/ha; DL 1% = 9,00 t/ha; DL 0,1% = 37,62 t/ha

Table 2

The significance of differences in production of factor B

Fact. B	Significance	Total production (t/ha)	Relative production (%)	± Relative to control (t/ha)	Statistical assurance
b_1	Carrot seeds moistened with water	18,40	100,0	Control	-
b_2	Carrot seeds moistened with humic acids	21,20	114,67	+ 2,80	
b_3	Carrot seeds moistened with Vitis Vinifera extract	20,00	108,69	+1,60	

DL 5% = 2,90 t/ha; DL 1% = 6,41 t/ha; DL 0,1% = 15,76 t/ha

Table 3

Significance of AxB interaction

Fact. A	Fact. B	Total production, t/ha)	Relative production (%)	± Relative to control (t/ha)	Statistical assurance
a_1	b_1	23,4	100,0	Control	-
	b_2	26,2	111,9	+ 2,8	
	b_3	25,0	106,8	+ 1,6	
a_2	b_1	26,8	114,5	+ 3,4	
	b_2	29,6	126,4	+6,2	
	b_3	28,4	121,3	+ 5,0	

DL 5% = 11,45 t/ha; DL 1% = 21,71 t/ha; DL 0,1% = 41,30 t/ha

CONCLUSIONS

In this study it was found that to the species *Daucus carota (L.)* seeds wetting process resulted in obtained of the substantial production increases; knowing that these seeds germinate very difficult.

Also, time wetting of the seeds influenced long enough thick root production achieved per unit area and not least, the biostimulator used to the wet of the seeds. In this sense the best results were observed in variants where seed has been moistened with Vitis vinifera seed extract. In addition, the humic acids led to the substantial increases production after 6 hours of seed wetting and especially after 12 hours of wetting.

This study will continues because these stimulators were caused and an improvement in product quality and better plant behavior in culture regarding specific diseases, and pests.

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RESEARCH ON CHEMICAL COMPOSITION AND SENSORY FEATURES OF THE WINES OBTAINED IN GALICEA MARE VITICULTURAL AREA IN THE CLIMATIC CONDITIONS OF THE YEAR 2011

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Key words: *red wines, chemical composition, maceration*

ABSTRACT

Any discussion on the quality characteristics of these wines must be made subject due to the fact that for wine year 2011 was a special year when weather conditions allowed all varieties to biosynthesised in proportions well above the annual average, a number of basic compounds of plant metabolism and therefore the composition of red wines. Thus, the grapes have accumulated very large proportions of sugars and polyphenols, which are found in unusually high content of wine in alcohol, glycerol, in anthocyanin and in the incredible value of coloring intensity .

INTRODUCTION

Wine is the first alcoholic beverage produced and consumed by humans. Over time many testimonies have been reported on the health benefits of moderate wine consumption. The most important beneficial effect attributed to moderate wine consumption is related to the protection which he provides to cardiovascular disease, the leading cause of mortality in developed countries (Băducă Cîmpeanu C., 2008).

The studies of Leighton F. (2001) and Mansvelt E.P.G. (2001)have shown the importance of a moderate daily consumption of wine as part of a diet or a diet complementare already rich in polyphenols or to compensate for the negative effects of a high fat diet.

With red wine, both the alcoholic and the polyphenolic components seem to contribute to its beneficial effects (Guilford J.M. et al., 2011). The alcoholic component is known to increase high-density lipoprotein cholesterol and to decrease fibrinogen concentrations (Hansen A.S. et al., 2005). When the effects of 3 alcoholic beverages, red wine, beer, and vodka, were compared in a recent study (Krnic M. et. al., 2011), only red wine provided protection against vascular oxidative stress. Indeed, red wine polyphenols seem to shield the vasculature by reducing reactive oxygen species and by inhibiting

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endothelin-1 expression (Schini-Kerth V.B. et al., 2011, Jiménez R. et. al., 2005, Nicholson S.K. et. al., 2010).

MATERIAL AND METHODS

This paper was prepared based on chemical and chromatic analyzes of a 6 red wines obtained from wine center Galicea Mare from Dolj county, in 2011. The wines were developed in the Laboratory of Oenology of Faculty of Agriculture and Horticulture, University of Craiova, where they were analyzed.

Chemical analyzes focused on the main parameters of the wine composition : alcoholic strength, total acidity and volatile acidity, residual sugar and glycerol (Cotea D.V., 2009),using the official methods of analysis . The 6 wines were obtained from three varieties: Fetească neagră, Merlot and Cabernet Sauvignon vinified as pure or mixed. Thus, Cabernet Sauvignon is found in 4 of the 6 wines analyzed, of which 3 variants as pure variety but with different maceration times: 3 days, 6 days and 9 days. A fourth version is present by a Cabernet Sauvignon and Merlot combination, in equal proportions. Merlot and Fetească neagră varieties are two other versions of the experimental scheme.

Wine tasting was conducted twice, on the wines of 3 and 6 months, were made by a panel of tasters and authorized wine-makers that tasted the wines by a common O.I.V.-U.I.O.E. sheet (Stoian V., 2001).

RESULTS AND DISCUSSIONS

The results on the chemical composition of wines presented in Table 1, carries clearly the mark of a specific character of the year 2011, characterized as a year in which the grapes have accumulated a large amounts of sugars, well above the annual average, in all growing areas of Oltenia, including Galicea Mare.

The first analyzed parameter was the alcohol content of wines and it shows that some wines have incredibly high values, especially those derived from Feteasca neagră and Merlot. Thus, Fetească neagră wine presented an alcoholic strength of 16.4% vol, while Merlot wine presented an alcoholic strength of 17.0 vol, unnatural for a wine. An unusually high alcoholic strength also presented the Merlot and Cabernet Sauvignon combination (16.2% vol), particularly due to alcohol intake of the variety Merlot while the Cabernet Sauvignon alcoholic strength not passed 15% vol.

Fetească neagră and Merlot wines reached to such alcoholic strengths because of the sugars accumulation in grapes from over 280 to 300 g/l, while the harvesting was not made very late in calendar view and the production levels were average to good really. This is a clear indication that 2011 was a year of special wine, which can hardly be met.

The alcohol content of the 3 Cabernet Sauvignon wines are very interesting because it's about three different alcoholic strengths, while the wort was divided into 3 variants, with different maceration time. Because the wine with the shortest duration of maceration (3 days) had the lowest alcohol content (13.9% vol) and the wine with the longest duration (9 days) had the highest alcohol content (15.1% vol) seems inexplicable at first, because it does not exist a direct correlation between alcohol content and the maceration time, especially differences of more than 1% vol. A closer look to the values of this parameter leads to the explain of situation that since this is a very rich in sugar wort, a longer time of contact of the grape pomace and the wort allowed the yeast to lead the fermentation to higher alcohol content, allowing them to resist in high concentrations of

alcohol and to ferment a higher proportions of sugars due to a better resistance in the decline phase of population.

In conditions of high alcohol content due to the accumulation of excessive proportions of sugars, the glycerol content in wines was well above normal, ranging between 13.2 g / l and 15.0 g / l, values which, normally not found in the wines themselves. This has led to a greasy perception in tasting of all the wines. The lowest glycerol content was for Cabernet Sauvignon wine 1 (3 days maceration time), being 13.2 g/l. Highest glycerol content was Merlot wine (15 g/l), the wine with highest alcoholic strength.

The third composition parameter analyzed, the residual sugar content, showed very interesting situations, meaning that only two wines (Merlot and Cabernet Sauvignon 3, with 9 days of maceration) were dry wines, so below 4 g/l residual sugar. Other 3 wines (Fetească neagră, 5.8 g/l, Merlot + Cabernet Sauvignon, with 8.2 g/l and Cabernet Sauvignon 2, 10.2 g/l) were semi-dry . The sixth wine, Cabernet Sauvignon with 3 days of maceration, was sweet, with 16.4 g/l residual sugar, but of the 3 wines Cabernet Sauvignon is the wine with the shorter maceration time. So the yeasts faced harder with sugar content and the alcoholic fermentation stopped before exhausting its fermentable sugar content.

Table 1

The chemical composition of wines

Wine	Alcohol %vol.	Glycerol g/l	Residual sugar g/l	Total acidity g/l H_2SO_4	Volatile acidity g/l acetic acid
Fetească neagră	16,4	14,2	5,8	4,4	0,52
Merlot	17,0	15,0	3,6	3,2	0,36
Merlot+Cabernet Sauvignon	16,2	14,0	8,2	3,9	0,36
Cabernet Sauvignon 1 (3 days)	13,9	13,2	16,4	4,0	0,44
Cabernet Sauvignon 2 (6 days)	14,3	13,6	10,2	3,8	0,40
Cabernet Sauvignon 3 (9 days)	15,1	13,8	3,2	3,9	0,32

Total acidity of wine, expressed in H2SO4, has contents between 3.2 and 4.4 g/l, low but perfectly explicable, given the fact that are red wines produced from overripe grapes.The lowest amount of the total acidity of 3.2 g/l, was Merlot wine. The 4 wines obtained from Cabernet Sauvignon (3 vinified as pure and one mixed with Merlot variety) had contents between 3.8 to 4.0 g/l. Fetească neagră had the highest value of total acidity 4.4 g/l but if this case volatile acidity was also the highest of all 6 wines analyzed, which means that it has also contributed to total acidity of the wine.

The volatile acidity, which is linked to the health and biological stability of wines, presents values between 0.32 and 0.52 g/l acetic acid. It is very important to note that these values are very good, not because of the low level compared to the legal limits allowed for this parameter of 1.2 g/l as the fact that wines were produced from overripe crops, which made the fermentation times longer, 10-15 days. It is well known that at high content of sugar and long period of fermentation are formed higher proportions of volatile acidity, which did not happen to these wines because the grapes were very healthy and winemaking was done in proper conditions of hygiene wine.

The results of the organoleptic characteristics of wine are shown in Figure 5. How they were perceived by tasters in wine tasting sessions, was strongly influenced by the particular conditions in which the wines were produced , taking into account all aspects of the quality of the grapes mainly the very advanced degree of ripeness. A first observation that must be made in analyzing the results of the two tasting sessions is that 5 of the 6 wines were assessed with higher scores tasting at 6 months, so there is an upward trend in terms of organoleptic characterization and only one wine was considered weaker at the second tasting. The only wine that was seen better at the first tasting to the second tasting was the wine with the shortest maceration time, Cabernet Sauvignon or with three days of maceration, which scored excellent at the 3 months tasting (93) greatest of all 6 wines analyzed, and at the 6 months tasting, though it received one point less, still remained the wine with the highest score of all. The reason for this is that wine produced by a short maceration time is slimer, with impressive color, especially in terms of shade and clarity, characteristics that were appreciated at its best in both tasting, receving excellent rating. The wine was also better appreciated against the other in terms of quality and flavor, especially fruitiness. In fact, this was a very important element to assess, since it was the only wine that was given excellent rating for the quality of flavor, this was primarily influenced by the fact that under a shorter period of maceration and the extraction of a smaller proportion of polyphenols, the fruit and fermentative flavors were better perceived . At the taste examination this wine has dominated the others because it was lighter, less tannic, astringent, agressive.

Unlike him, the wine with 6 days of maceration received 5 points less than this one at the first tasting and one point less at the second tasting. This is explained by the fact that Cabernet Sauvignon wine with maceration for 6 days was intensely colored, but with more pronounced violet hues, less shiny, almost opaque. The smell was more intense and greater of typical variety but less appreciated in terms of flavor quality that was dominated to a greater character of astringent berries, with notes of blueberries and blackberries, however less enjoyable than the first wine. At the taste examination the wine was full bodied, was much more structured but with a more pronounced touch of astringent, especially at the first tasting. At the next tasting, the astringent character wasn't so pronounced, the wine appreciation getting better, what made the difference in scores from the wine with the short maceration time to be smaller. So, in terms of sensory perception, wine increased from the first to the second tasting.

Regarding Cabernet Sauvignon wine obtained by the longest macerating, 9 days, it was considered an even lower score at first tasting (86) because of the color that was completely lacking in transparency, with a pronounced purple tinged, but also because of the biting taste, very heavy drinking. Over the next three months, these characters were slightly attenuated, being helped to a large extent by the high content in alcohol and glycerol, which gave the wine a sweet character, soft, silky and so the astringency wasn't so aggressive. A very appreciated wine was that obtained by combination of Merlot and Cabernet Sauvignon, which received 88 points from the first tasting and 90 points to the next tasting. Both scores are very good for a young red wine and opens interesting perspectives in the production of wines from Galicea Mare assortment type, the formation of which to attend two or more varieties. Also of interest is the obtain of wines of the same variety but different proportions to see which combination would give the best results in relation to what is expected of them, namely the desired sensory profile, taking into account and categories of consumers whom it is addressed. The lowest obtained scores were the Merlot and Fetească neagră wines but this does not mean that there are not good wines. Contrary, even if the points they obtain are just below the points of other wines, it is

important to note that there are still very high for a few months wines and that are growing. At these two wines, the element that made them to receive fewer points at the tasting was the alcohol content, unusually high . This meant that their flavor was dominated, obviously, by the smell of alcohol, slightly fiery chacter, which fell both typicity and quality of flavor, which contributed to a decrease in the score, even if the other sensory characteristics examined were appreciated at its best at Feteasca neagră and Merlot wines .

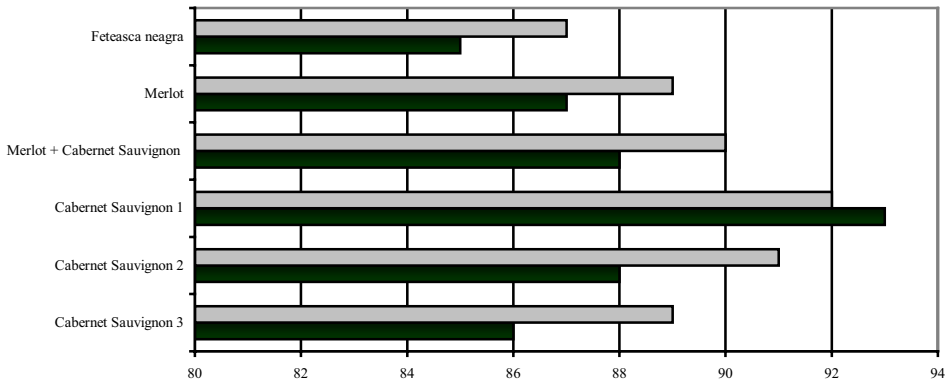

Figure 1 - Results of sensory analysis of wine at 3 and 6 months

CONCLUSIONS

The study undertaken on the 6 red wines produced in 2011 from grapes from Galicea Mare wine center in Dolj, highlighted a number of conclusions on the chemical characteristics, color and organoleptic properties of wine.

A technological factor of the primary winemaking which normally is not related to the main chemical constituents of the wine, as the length of maceration, in the particular conditions of an overripe crops had influence on alcohol and glycerol content and balance of taste perception. In other words, it is an unusual situation that the maceration time influence the alcohol content, but this should be blamed on the quality characteristics of raw grapes.

Under the same wort, uniform in terms of chemical composition, biochemical and microbiological, divided into 3 types, which have different periods of maceration was found that the wines showed higher alcohol content as maceration time was higher. The only plausible explanation in this case is that, given the advanced state of maturation of the grapes, maceration extension allowed the extraction of the solid parts, especially from the skin, of a higher proportions of substances used by yeasts in mineral nutrition, which helped them to a better withstand in front of the difficult conditions of fermentation, especially towards the end of fermentation,when the yeast population is in decline and thus to increase the rate of fermentation of sugars.

From the sensory point of view, all the wines showed a balance olfactory-taste marked by alcohol and glycerol excess. If alcohol is excess, it has disadvantaged in terms of smell, the Fetească neagră and Merlot wines, which covered the smoothness of flavor

variety, the glycerol excess had the gift to alleviate hardness, roughness and astringency of tannins.

The different maceration time allowed a very good appreciation of wine with shorter duration of maceration, which presented an obvious plus of freshness and suppleness, which was very important in the first months of wine but if you judge wines by their suitability for maturation and aging, a longer maceration was beneficial.

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CHEMICAL METHODS TO PREVENT THE PHYLLOXERA GALICOLA

Ficiu Lidia, Dejeu L.[1]

Key words: *grapevine, phylloxera, vinifera variety, insecticide, rootstock*

ABSTRACT

. To prevent the attack of phylloxera galicola present on the leaves of Vitis vinifera varieties in the experimental device of Valea Calugareasca were made chemical treatments with insecticides and acaricides with curative potential in combating this pest. Experimenting was done using Cabernet Sauvignon and Feteasca alba varieties grafted on Kober 5BB and SO4 rootstocks.

After each treatment applied to 3 days, were made observations and determinations at the leaves of varieties mentioned the number of present gale on the leaves in order to register the frequency, the intensity of the attack and to follow the damage caused plant and harvest grapes by this pest.

From the determinations and observations it was found that in the case of use abamectin based product, the frequency attack with phylloxera galicola was greatly diminished.

INTRODUCTION

Phylloxera (*Phylloxera vastatrix*) is considered as the most important pest of the vineyard in all world. It was discovered in 1868 year, its attack causing the biggest disaster in the viticultural world (Bazille M., Planchon J.E., Sahut F., 1868; Foott J.H., 1989).

In Romania phylloxera was identified in 1884 in Dealu Mare vineyard, from where was spread in all vineyards.

This required changing the technology of culture by passing to the system based on the use of grafted vines (Tomoiagă Liliana, 2006). This species has several generations per year and have a full life cycle on American vines and direct producer hybrids and an incomplete life cycle in European vines.

Phylloxera is presented in four forms which are distinguished by distinct morphological characters galicola, radicicola, sexupara and sexual forms.

Phylloxera is an insect of small dimensions that bites vine roots and leaves for to feed the sap of them, and finally grapevine dried. (Grecu V., 2000).

The adults and larva form of galicole parasites the leaves producing damage depending on the population density. The stings caused by the insect determined the appearance of the characteristic gala.

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Climate change (warm winters, high temperatures, low rainfall), cultural techniques of non-compliant (planting too deeply rooted grafts) will facilities appearance of some strains with a high degree of pest and enhancing the area of spread of the pest (Ray M.K., 2006, Sabbour M.M., Abbass M.H., 2006). From observations and determinations made to found an increase in aggressiveness and virulence, an increase in the number of generations and a worrying attack of galicole form a growing number of vinifera varieties (Jeffrey Granett, M. Andrew Walker, 2001).

Until now not found the effective methods to fight, but to limit the damages produced by this pest.

This study was developed during the process of doctoral studies programe, financed through project POSDRU/107/1.5/S/76888.

MATERIAL AND METHODS

The experimental plots were located in two viticultural parcels within Valea Calugareasca viticultural center, where the biological reserve of phylloxera was very high.

Have been selected Cabernet Sauvignon and Feteasca alba varieties grafted on rootstock Kober 5 BB and SO4.

In the experience were taken into study the following factors:

The factor A – vinifera variety, with graduations: a_1 - Cabernet Sauvignon;
a_2 - Feteasca alba;

The factor B – rootstock variety, with graduations: b_1 - Kober 5BB;
b_2 - SO_4;

The factor C – active substances, with graduations:
c_1 - untreated (control);
c_2 - Tiacloprid, in concentraţion of 480g/l (Ti);
c_3 - Abamectin, in concentraţion of 18g/l (Ab);
c_4 - Lambda cihalotrin, in concentraţion of 50g/l (La);
c_5 - Oxamil, in concentraţion of 240g/l (Ox).

Of their combinations resulted 20 experimental variants.

For each variant were chosen each 4 control vine which were applied operations of vegetative uniformity.

The treatments were began and applied in view of pest biology, so that the first generation were applied at bud burst phenological stage BBCH 15 (five leaves displayed), the treatments is then repeated every 10 days.

The treatments were done manually with spraying STYLE apparatus.

The observations and measurements performed every 3 days after each treatment were made in order to record the frequency, intensity and attack damage from this pest plant and harvest the grapes.

For each variant were analyzed 4 control vine which were made observations to 100 leaves.

Lambda-cyhalotrin is a pyrethroid insecticide, highly effective with a strong contact and ingestion action on a wide range of pests, apply by spraying the warning during the growing season.

Oxamyl is used as an insecticide to kill and control a broad spectrum of insects. Its action is both systemic and contact. Abamectin works on the nervous system of insects causing few hours, their irreversible paralysis. When insects coming into contact with the product they die without the presence of any movement or spasm. Acts both by contact and ingestion and especially high insecticidal activity is observed starting from the third day after application. Thiacloprid is a systemic insecticide with spectrum wide counter.

RESULTS AND DISCUSSION

The observations in experimental plot showed that the leaves phylloxera attack was manifested itself early, because high biological reserve, since the stage of vines presented 4-5 leaves displayed, the attack showed by the appearance on the surface of the leaves of gale (figure 1and 2).

The experimental data obtained show that in the two varieties grafted on Kober 5BB and SO_4 rootstock, the best results in combating phylloxera galicola attack were obtained by using abamectin active substance in concentration of 18g/l (figue 3, 4, 5 and 6).

Fig.1-Phylloxera galicola to Cabernet Sauvignon variety / SO4

Fig.2- Phylloxera galicola to Feteasca alba variety/SO4

So while to the untreated variant the intensity attack registered an increasing trend from values of about 75% to values of 84%, when using of this substances.

The intensity of the attack decreased to value of 70% on 14.05.2012 at levels of 40% on 24.06.2012 at Cabernet Sauvignon variety and value of 70% to 48% at Feteasca alba variety for the same date.

The lowest results were obtained when using of lambda cyhalotrin as an active substance in concentration of 50 g/l. Cabernet Sauvignon showed values from 75% to 53% on 14.05.2012 on 24.06.2012 and Feteasca alba variety of values of from 75% to 58% for the same dates.

The rootstock partner used in combination not influenced by an evident manner efficacy for control of phylloxera galicola.

Also considering the evolution in time of phylloxera galicola attack is found that the influence of treatment made it decreased in intensity control of all substances tested, especially when used as abamectin active substances, while the untreated control the intensity of attack showed increasing trend.

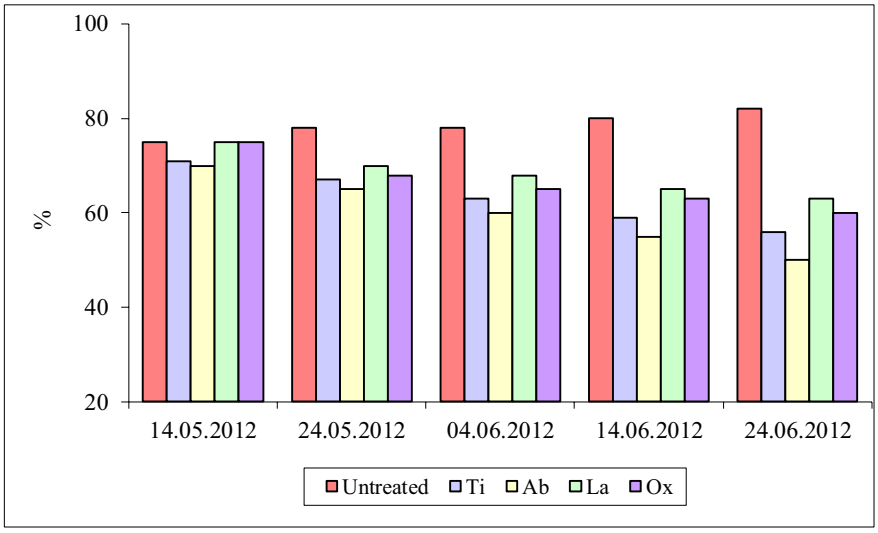

Fig.3 Results the intensity attack of phylloxera galicola (%)
to Cabernet Sauvignon variety/Kober 5BB

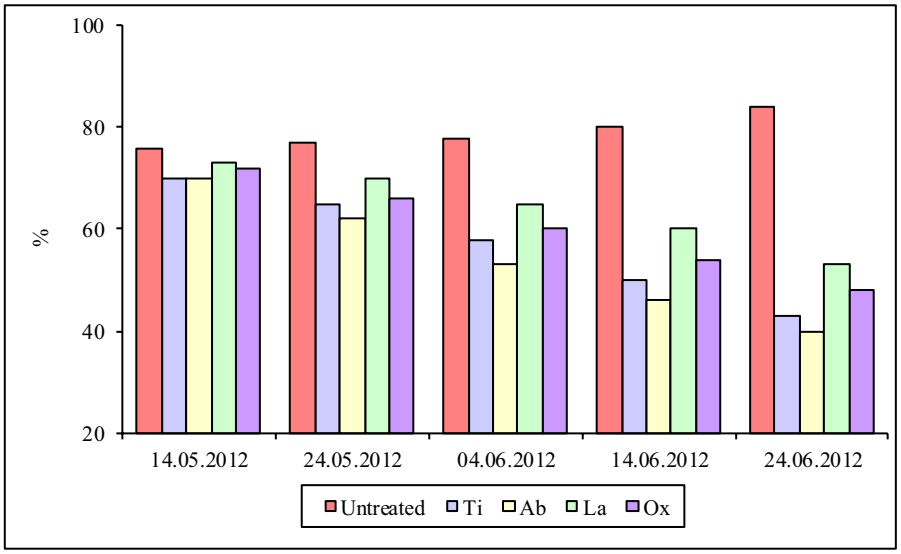

Fig.4 Results the intensity attack of phylloxera galicola (%)
to Cabernet Sauvignon variety/SO_4

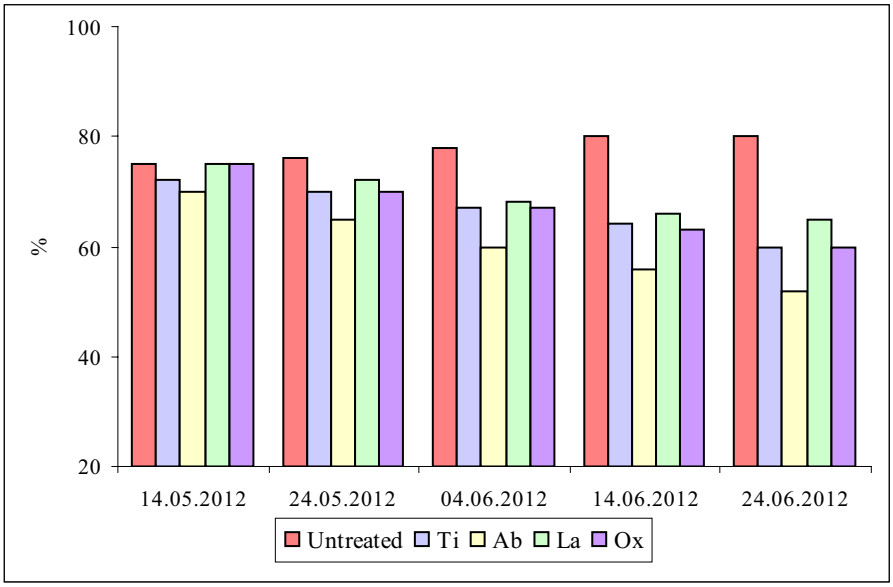

Fig.5 Results the intensity attack of phylloxera galicola (%)
to Feteasca alba variety / Kober 5BB

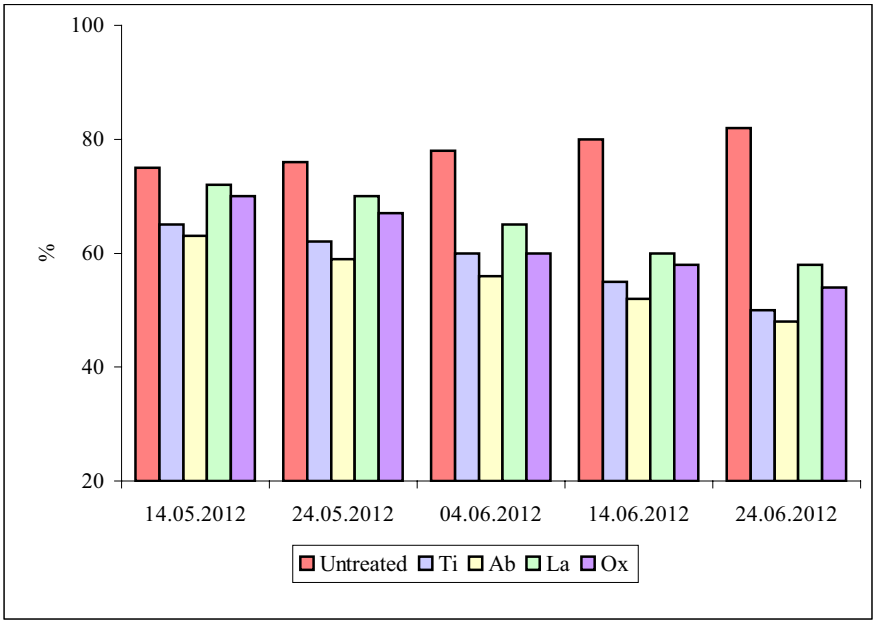

Fig.6 Results the intensity attack of phylloxera galicola (%)
to Feteasca alba variety /SO_4

CONCLUSIONS

The experimental data obtained showed that the active substances tested the treatment with abamectin in concentration of 18 g/l had the best efficacy in reducing the attack of galicola phylloxera both varieties studied.

The grafting rootstock of the combination partner not influenced in a obvious manner the resistance of experimental varieties to the attack of phylloxera and not effectiveness of the tested substances.

Mode of action of the active substances used is also essential.

To reduce pests population below the threshold there are necessary treatments repeated.

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NITRATE CONTENT OF RAW COW MILK

Gingioveanu Oana Andreea[1]

Key words: *nitrate, raw cow milk, metallic cadminm column.*

SUMMARY

The purpose of this study was to find out content of nitrates in raw cow milk. The levels of nitrate were determined in 40 sourples of cow raw milk produced at Agricultural and Development Statistics Şimnic Romania from April to July 2011. Following the analyse the result reveal a variation of nitrate content in fresh cow milk. The average of nitrate level was 1,17mg NO_3^-/l of milk in April 1,28mg/l in May, 1,71mg/l in June, 1,64mg/l in July. The highest level of nitrate was in July 2011 (2,5mg NO_3^-/l of milk). The nitrate content of examined sourples could be attributed external sources such as green forages consumed by dairy cows. The data revealed that nitrate devels found in all cow raw milk sourples were inside the legal limits.

INTRODUCTION

Nitrate and nitrite are naturally accurring ions thet are part of the nitrogen cycle and are abiquitons in the environment. Both are products of the oxidation of nitrogen by microorganism in plants, soil or water. Studies have suggested that nitrates, nitrites and nitrosamines have an etiologic rol in adverse reproductive autcomes (Ward et al. 2005) and other health conditions: neural tube defects (Brender et al. 2004), type 1 and type 2 diabetes mellitus (Longueeker and Daniels, 2001), effect in thyroid function (Bloonefield et al. 1961), or increase in blood pressure in school children (Pomeranz et al. 2000).

Humans are exposed to nitrates primarily through diet and drinking water, with vegetables contributing the largest amount of dietary nitrates per serving (Griesenbeek et al. 2009).

The occurrence of nitrates in food may be considered hazardous because nitrates can be reduced to nitrates before ingestion, in saliva and in the gastro-intestinal duct (Anonymous, 1981). Nitrites may react in the stomach with secondary or tertiary amines and amides present in foods to form n-nitrosococeepocends which are potentially corcingeus (Anonymous, 1981).

The nitrates may naturally present in milk and the level of it depends are the quality of feedstuffs and water for lactating cows. Following the personal application of KNO_3 to dairy cows, a worked increase in nitrate content in milk appeared in dependence on applied KNO_3 (Baranova et al. 1993). Average value of residual nitrate in milk two

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hours after administration of 150g of KNO_3 was 34,60mg of NO_3^-/l milk (Baranova et al.1993).

The natural levels of nitrite in fresh plant material, with certain exception, are generally very low and few data have been published (EFSA, 2008).

As in the case of vegetables grown few human consumption, several plant species are known to accumulate nitrate up to very high levels, but majority do not represent major feeds for livestock. The accumulation of nitrate in plants is influenced by a range of factors: stage of maturity, fertilizer application, growing conditions, plant species. Levels of nitrate tend to be highest in the lower stems and leaves of plants, while very little is found in the seeds. Forage crops represent the greatest risk of diets exceeding the maximum permitted levels of nitrite (10mg/kg nitrite ion) as specified in Council Directive 2002/32/EC.

The purpose of this study was to find out content of nitrates in raw cow milk produced in April, May, June and July 2011 period in wich green forages are consumed by lactating dairy cows.

MATERIALS AND METHODS

A total of 15 samples of raw cow milk produced at Agricultural Research and Development Statistics Șimnic (dairy center) were analyzed from April to July 2011.

Nitrates and nitrites were determined according to International Dairy Federation standard method (1 DF 1984) and a method described by Popescu 1988.

The method involves dilution of 10 ml milk saceeple in warm distilled water, precipitation of fat and proteins with protein free substances and filtrates to obtain a filtrate volume of 200ml, volume that will be considered in the calculation.

Nitrate was reduced to nitrite in a position of the filtrate by means of metallic cadmic in a glass column.

A red color was developed in a position of both reduced solution and unreduced filtrate by addition of sulfanilamide and Nj-naphtyl-ethylenedianide. Measurement of the color intensity was made by spectrophotometer.

Calculation of the nitrate content was made from difference between these two analytical results. Concentration of NO_3=Concentration of NO_2 (after reduction), minus concentration of NO_2 (without reduction). Standards of nitrite were measured in the same way. Nitrite calibration curve was determined using series of concentration of nitrite of 0; 2; 4; 6; 8; 10; 12; 16 and 20 ml from the standard nitrite solution with 0,1 μg/ml.

All analysis were repeated twice for each sample.

RESULTS AND DISCUSSION

Nitrate contents of milk samples are presented in table 1.

The analysis of data for april 2011 show that nitrate ions (NO_3-) levels were between 0,07 to 2,10 mg/l of milk with an average of 1,17 mg/l. In may 2011 the limits were between 0,1 and 2,2 mg/l with an average of 1,28 mg of NO_3-/l of milk.

Table 1

Levels of nitrates in raw cow milk (mg/l) from april to july 2011

Months	Number of samples	Nitrates (NO_3-)			Legal limit
		Average	SD ±	Limits	
April 2011	10	1,17	0,88	0,07-2,10	max 10 mg/l
May 2011	10	1,28	0,73	0,10-2,20	max 10 mg/l
June 2011	10	1,71	0,52	1,00-2,30	max 10 mg/l
July 2011	10	1,64	0,50	1,10-2,50	max 10 mg/l
Total/average/limits	40	1,45	0,27	0,07-2,50	max 10 mg/l

In june 2011 the limits were between 1,0 and 2,3 mg with an average of 1,71 mg NO_3-/l of milk and in july 2011 the average was 1,64 mg NO_3-/l of milk and limits between 1,1 and 2,5 mg NO_3-/l of milk. For all months, the level of nitrates in raw cow milk was an average of 1,45 mg NO_3-/l of milk (table..).

The nitrate content increased by 9,4% in may, 46,1% in june and 40,1% in july compared with april 2011. The nitrate content of examined milk samples could be attributed to external sources such as green forage consumed by dairy cows. This data revealed that nitrate levels found in all cow raw milk samples were inside the legal limits.

Several authors have reported higher nitrate levels compared to the results of this study. For instance, Osvat and Bara 2010, reported in 2007 an average value of 2,66 mg NO_3-/l, in 2008 2,39 mg NO_3-/l and in 2009 3,08 mg NO_3-/l of fresh collected cow milk.

Nitrates are considered harmful to health and for this reason their levels must be controlled in milk destinated for human consumption.

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MAXIMUM RETENTION TIME OF ANTIBIOTIC RESIDUES IN RAW MILK FOLLOWING TREATMENT OF BOVINE INFECTIONS

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Keywords: *antibiotic ,residue ,retentions, microbial, inhibitor.*

SUMMARY

Mean retention time was 51,80 hours, 45,00 hours and 9,25 hours, for *intiamammary infusion, intramuscular injection and respectively intrauterine infusion of the antibiotics. Recommended withdrawal time was72 hours for intramammary infusion and intramuscular injection and 48 hours for intrauterine infusion of antibiotics. It was observed a cow with antibiotic residues at the end of recommended withdrawal time as result of therapy administered in excess of label directions. I recommend to check each treated cow at the end of recommended withdrawal time until two consecutive samples assayed negative for antibiotic residues, or to use drugs with less potential for residues in raw milk.*

INTRODUCTION

Treating lactating animals with antibiotics is a veterinary practice to cure the disease.

The antibiotic used may persist in the milk of cows for a period of time depending on drug, dosage applied, route administered, body weight of the animal treated.

The presence of antibiotics in milk is a major concern for public health and dairy industry. This is due to the generation of multi-resistant bacteria and allergic processes caused to the consumer, as well as by the interruption of milk fermentation in milk processing.

Antibiotic residues are remnants of antibiotic drugs or their active metabolites that are present in the milk of treated animals. Levels of the drugs and their metabolites may persist at unacceptable levels and consumers can be exposed to then.

Milk may be contaminated with compounds of one of the antimicrobial drug classes: beta-lactames sulphonamides tetracyclines, marolides, quinolones and aminoglycosides.

Penicillins, along with apholosporius are the main constituents of the class of antimicrobials known as β-lactam antibiotics. All the drugs in the group have very similar modes of action, pharmacokinetics and side effects.

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The most commonly used penicillins in live stock production are procaine penicillin G, potassium penicillin and potassium in international units. Procaine penicillin G has o potency of 900-1050 1 Un per mg.

International organizations such as codex alimentrius commission (FAO/OMS), European Union, Food and Drugs Administration (FDA) and The United States Department of Agriculture (USAD) have established maximum residue levels (MRL) for veterinary drugs in food intended for human consumption.

Veterinarians in the EU are allowed to prescribe drugs in an off-label manner but are then obligated to assume that residues do not enter the food chain.

Accurate detection of low levels of antibiotic residues in milk is of great importance for farmeys to ensure that contaminated milk from individual cows do not enter in the bulk milk tank.

In order to keep antibiotic residues out of the food chain of humans, regulatory authorities have established the withdrawal period for antibiotics that must be observed by producers before the milk from heated cows can be sent to market. Never the less, drug residues in marketed milk continue to occur (Grădinaru 2010, Grădinaru şi colaboratorii, 2011).

It is necessary to monitor a number of milk samples for the presence of the important antimicrobiel drugs residues by using inexpensive, rapid and simple screening methods.

Of the methods used, microbial inhibitor method suited best these requirements (Navratilova, 2008).

In general, antimicrobial agent control system can be divided in two steps. The first step is general monitoring of these agents during witch tests are used making it possible to establish rapidly the presence of inhibitory agents. The second step is a specific analysis which makes use of the methods which enable the identification and quantification of inhibitory agents.

This study was designed to investigate persistence of antibiotic residues in row milk of the intramammary, intramuscular and intrauterine treatment of lactating cows.

MATERIALS AND METHODS

Retention times of some antibiotics for cows treated for infection was observed at Agricultural Research and Development station Şimnic. Diary herd consists of approximately 250 Holsteins. Cows diagnosed with clinical mastitis were treated by one of two procedures: intramammar infusion with an antibiotic preparation containing procaine penicillin ant sodium novobiocin and intramuscular injection with an antibiotic preparation of procaine penicillin and dihydrostreptomicin sulfate.

Milk sample were collected each milking in sterile bottles. Teats were pre-dipped in an iodine-based product and died with individual paper towels. Three streams of fore milk from each quarter were discarded before sample collection, and 100 ml of quarter composite milk was collected from each cow.

Cows diagnosed with clinical metritis were treated by uterine infusion with an antibiotic preparation containing procaine penicillin and dihydrostreptomicin sulfate milk samples were collected as described.

For each of the treatments, milk samples were collected and tested for antibiotic residues until two consecutive samples assayed negative.

The antibiotic detection test was the Ecotest (EON TRADING LLC USA). The test sensibility is sufficient to guarantee finding any substances in suppressing concentration in milk raw material. The method involves incubating milk sample with active substance at 44^0C for 10 minutes. The quantity of acid developed was titrated in the presence of ph indicator.

The samples with color white are negative and the samples with color pink are positive.

RESULTS AND DISCUSSION

Maximum retention times of antibiotic residues in milk after final intramammary infusion was 78 hours, 60 hours after final intramuscular injection and 12 hours for intrauterine infusion of antibiotics (table 1).

Table 1

Retention times of antibiotic residues in milk after the final intramammary infusion, intramuscular injection and intrauterine infusion of antibiotics

Specifications	Animals					X	SD	Variab%	Recommended withdrawal time
	1	2	3	4	5				
1. Intramammary infusion: Retention time-hours	60	78	25	48	48	51,8	±8,7	16,8	72
2. Intramuscular injection: Retention time-hours	36	48	60	36	-	45	5,7	12,66	72
3. Intrauterine infusion: Retention time-hours	6	12	8	11	-	9,25	2,75	29,7	48

Mean retention time was 51,8 hours, 45,0 hours and 9,25 hours for intramammary infusion, intramuscular injection and respectively intrauterine infusion (table 1).

Table 1 shows that 60% cows with intramammary infusion produced milk clear of antibiotics residues by 24 hours before the end of recommended withholding time.

Also table, shows that antibiotic residues may be present in milk at the end of recommended withdrawal time (cow nr. 2 with intramammary infusion of antibiotics).

This is due when therapy is administrated in excess of label directions.

Drug elimination from a cow typically follows a characteristic pattern, in that the amount of drug eliminated per unit of time is proportional to the amount of drug present (1^{st} order of time linear kinetics).

Because of this logarithmic nature of drug depletion, doubling a drug dose only adds one elimination half life to the withdrawal time.

Giving label doses to a sick or geriatric cow significantly increases the risk for having a residue drug in milk even if the label withdraw all time is followed (Chicoine, 2007).

Pathology such as renal or hepatic dysfunction dehydration, or hypoproteinemia can alter drug distribution and clearance. These changes may prolong the elimination half life. Variation in the half life can have profound effects on the withdrawal time.

Improper injection techniques can cause residues in milk even with label dosages. Most drugs will be absorbed more quickly and completely if given intramuscularly in the cow's neck rather than the hindquarter muscles. Injecting excessive drug volumes per site can also result in a decreased absorption rate and elongated withdrawal time (Chicoin, 2007).

CONCLUSIONS

- In general monitoring of antibiotic residues in milk of cows treated for bovine infections, microbial growth inhibition method is easy to perform and enable the detection of residues.
- This results suggest that a 72 hours withdrawal period might be insufficient to secure the clearance of antibiotics used, because treatment dosage contribuie to the antibiotic clearance.
- To reduce the potential for significant antibiotic residues an ideal methods are to test milk samples from individual treated cows and then discard the milk with positive results, or to use other drugs to that bovine infections that are less potential for residues in raw milk.
- A large scale of samples is needed for further investigations.

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SELECTED PERSPECTIVE ELITE FROM 'TĂMÂIOASĂ ROMÂNEASCĂ' IN DRĂGĂŞANI VINEYARD

Gorjan Sergiu Ştefan[1]

Keywords: *elite, productivity, clonal selection, vineyard, germplasm, conservation*

ABSTRACT

To identify varieties and vine clones and their preservation from saving the viticultural germplasm from Dragasani Vineyard, was made a study in the Drăgăşani vineyard plantations in 2010 - 2011. This study was carried out at 'Tămâioasă românească' grape variety in a very old plantation observing that some grape vines have distinct agroproductive and technological characters. One of these hubs was particularly significant and was taken into study. This elite will be approved in the next future in order to be placed in culture.

INTRODUCTION

The vine is along with wheat, one of the oldest cultivated plants which arose about 4000 years i.H (Dejeu, 2010).

From vine genetic resources have resulted many varieties, either by empirical selection or by modern breeding. Much of the genetic variability is still insufficiently used, many old varieties, local, have been abandoned or neglected.

Actions are being taken in Europe to make landraces more competitive with modern varieties. Interventions to increase competitiveness have included better characterization of local materials, improvement through breeding and processing, greater access to materials and information, increasing consumer demand, and more supportive policies and incentives (Devra Jarvis, 2009).

This study was conducted in private plantations from Drăgăşani Vineyard, one of the oldest and most reputed vineyards in Romania at 'Tămâioasă românească' old grape variety that is part of the traditional assortment of Drăgăşani Vineyard.

Over time this variety has been very beneficial for viticultural Romanian research, obtaining new cultivars and clones.

Reminded of varieties as 'Istriţa', table variety obtained at SCDVV Pietroasa through sexual hybridization of varieties 'Tămâioasă românească' x 'Maria Pirovano' and the 'Centenar de Pietroasa', apiren variety obtained through sexual hybridization of

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'Tămâioasă românească and 'Perlette' varieties. Also at S.C.D.V.V. Pietroasa was obtained the variety for quality white wine 'Alb Aromat' a natural hybrid from 'Tămâioasă românească'. Another variety from 'Tămâioasă românească' is 'Aromat de Iaşi' obtained at SCDVV Iaşi , through the free annealing (Varga et al., 2005). Beside these varieties we recall values clones obtained by clonal selection namely 'Tămâioasă românească clone 104' obtained at SCDVV Drăgăşani and 'Tămâioasă românească clone 36 and 45' obtained at SCDVV Pietroasa (Oprea and Moldovan, 2007).

MATERIAL AND METHOD

After identifying and locating 'Tămâioasă românească' grape variety, located in a private plantation in Drăgăşani Vineyard in the wine Drăgăşani Suteşti-Pietroasa (44º40'52 N,24º12'06 E, 220 m. Alt.), we proceeded to analyze the plantation and the hubs. Plantation 'Tămâioasă românească' is very old, over 30 years, with a population of 15 hubs, planting distance 2.0 m between rows and 1.1 m between the hubs. From this study we observed that 5 hubs presents identical characters but different from the others in terms of agroproductive and technological point of view. One of these hubs was particularly significant and was taken into study. A code number was given to these elite, namely 1-2-3, and was made a comparative study between elite and witness variety ('Tămâioasă românească'). Were studied phenology and physiological traits - resistance to diseases comparing the elite with the genitor. The agroproductive characters analyzed were fertile shoots, hub vigor, vine grapes production, grape weight, grape quality and technological elements studied were: weight of 100 berries, sugar content in must, must acidity. Analyses were made on must using Carl Zeiss hand refractometer method and titrimetric method for acidity (H_2SO_4).

RESULTS AND DISCUSSIONS

Variety 'Tămâioasă românească' is part of the old traditional assortment of Drăgăşani Vineyard, being well known and cultivated in Europe, in Romania having as synonyms as 'Tămâioasă de Moldova', 'Tămâioasă de Drăgăşani', 'Busuioacă', 'Muscat', 'Tămâiată' , in Italy it is known as 'Moscato bianco', 'Moscato comune', 'Moscatello bianco', in Spain as 'Muscadel menudo bianco', 'Muscadel Morisco', 'Zoruna', in Germany as 'Muskateller', 'Weisser Muskateller', 'Muskateller gelber', 'Muscat blanc', in Austria as 'Schmeckende Weyrer' and in England the 'White Frontignan' (Constantinescu et al., 1960).

Studying the phenology of these genotypes, we note that bud break takes place in the same period (april – 11th), blooming is the same time at the elite and the witness, (may 27th – june 5th). Regarding the beginning of berry ripening, full maturity of the berry and beginning of wood maturity, these are early at the elite compared to witness (Table 1).

Physiological characteristics shows that the elite has a high resistance to drought, frost, *plasmopara*, *oidum*, but has average resistance to *botrytis* and the witness is resistant to drought, frost, *oidium*, with an average resistance to *plasmopara* and *botrytis*. We note that the elite is superior to the witness regarding *plasmopara* resistance.

The agroproductive characters of the 1-2-3 elite are superior compared to the witness regarding the fertile shoots, the hub vigour, production grapes/hub, the average weight of grapes and quality of grapes. The technological elements are superior, sugar content in must, acidity (H_2SO_4), weight of 100 berries being greater at the elite compared to the witness (Table 2).

Table 1

The phenological data average of clone 'Tămâioasă românească cl 1 BB' and variety witness 'Tămâioasă românească' in the period 2010-2011

No.	Code variety (elite)	Variety (elite)	Bud break	Blooming	Beginning of berry ripening	Full maturity of the berry	Beginning of wood maturity
1	1-2-3	'Tămâioasă românească cl 1 BB'	11-04	27.05 - 05.06	05.08	14.09	10.08
2	Mt.	'Tămâioasă românească'	11-04	27.05 - 05.06	08.08	16.09	12.08

Table 2

The main agroproductive and technological characteristics of clone 'Tămâioasă românească cl 1 BB' and variety witness 'Tămâioasă românească' in the period 2010-2011

Specification	U.M.	(Clone) 'Tămâioasă românească cl. 1 BB' elite 1-2-3	Variety 'Tămâioasă românească' (witness)
Fertile shoots	%	90	85
Vigor hub	-	large	middle
Production grapes/ hub	kg	3,100	2,920
The average weight of grapes	g	225	217
Quality of grapes	%	90	80
Weight of 100 berries	g	200	180
Sugar content in must	g/l	227	220
Acidity (H_2SO_4)	g/l	4,8	4,4

This elite 1-2-3 was grafted on 'Berlandieri x Riparia Kobber 5 BB, Selection Crăciunel 2' rootstock, for more advance study in the test plot and in pots at the University of Craiova - SCDP Vâlcea.

In the photos bellow we present images with 1-2-3 elite, the grape at full maturity, the young shoot and the adulte leave, the test plot image.

Photo 1 **Elite 1-2-3** the grape (originally)

Photo 2 **Elite 1-2-3** the adulte leave (originally)

Photo 3 **Elite 1-2-3** young shoot (originally)

Photo 4 **Elite 1-2-3** test plot (originally)

CONCLUSIONS

The 1-2-3 elite presents differents agroproductive and technological characteristics and it can be introduced on future in culture.

The elite has potential to be used for obtainig DOC (Controlled Origin Denomination) wines.

In the future this study will continue to select more valorous elites from the varieties wich forms the Drăgăşani Vineyard traditional assortment or from grape cultivars, to be homologated, the aim being the salvation and the use of the local viticole germplasm.

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FERTILIZATION MANAGMENT BASED ON SOIL SALINITY LEVEL ON FOUR (4) SUCCESSIVE GREENHOUSE CUCUMBER CROPS

Nikolaos Gougoulias, Alexandros Papachatzis, Adamantia Chouliara[1], Nikolaos Chouliaras[2]

Key words: *Soil Electrical conductivity, Soil salinity, Soil Fertility, Greenhouse crop*.

ABSTRACT

An assessment of a fertilization management practice for exploitation of soil salts was made in a four successive greenhouse cucumber crops. Only an organic commercial fertilizer in the form of black peat moss slightly enriched with inorganic elements was embedded basically, while during crop, a fertigation program was planned based on soil salinity level. This work confirmed that the plant need for nutrients might be covered largely by soil salts. The basic application of the organic fertilizer in relation to fertigation program applied during of crops period led to a reduced soil electrical conductivity from 0.66 to 0.24, 0.58 to 0.24, 0.80 to 0.49 and 0.55 to 0.39 dS/m respectively (soil extract of 1soil: 5 H_2O). Significant increase in soil salinity was observed after soil solarization, in the first and second crops (EC 1,23 dS/m and EC 0.48 dS/m), however, not observed increase in soil salinity(EC 0.44 dS/m and EC 0.41 dS/m) after soil solarization for the third and the fourth crop. Then, excessive salinity levels in soil should be taken into account when fertilization management of cultural practices in successive crops is applied.

INTRODUCTION

Soil salinity assessment is based on measurement of soil electrical conductivity, a quick, reliable and easy method which could be used during cultivation period for indication of soil fertility in a greenhouse crop (Rhoades *et al*., 1999).

The application of high fertilizer rates in intensive cultures, like greenhouse crops, affects electrical conductivity. In soils from several greenhouses in Thessaly, which have been irrigated with low salinity water, the high electrical conductivity (0.4 dS/m) in soil extracts (soil:H_2O ratio 1:5) is related with the increase concentration of soluble N and K. In these soils the base application of N and K mineral fertilizers in the following culture is

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not recommended (Chouliaras *et al*., 1991). In addition, the concentration of phosphorus in soil is not significantly affect salinity (Chouliaras *et al*., 1991). The mobility of P in soil is very limited and therefore could remains in soil for many years, in contrast with N and K. Thus, the knowledge of P history fertilizer applications in formers cultures, consists a valuable guide to efficient plant P-nutrition management.

The aim of this work was to develop a fertilization method for a greenhouse cucumber culture, based on soil salinity and fertilizer inputs of the previous crops.

MATERIALS AND METHODS

Soil samples (to a depth of 15 cm) were taken from a greenhouse located at the Technological Education Institute (TEI) of Larissa, Greece, for physical and chemical soil properties estimation. Soil organic matter, ammonium and nitrate nitrogen, available P and exchangeable K were measured following the Page *et al.* (1982) methods. Organic matter content was calculated by chemical oxidation of soil with 1 mol/l $K_2Cr_2O_7$ and titration of the remaining reagent with 0.5 mol/l $FeSO_4$. Soil organic matter was estimated by multiplying soil organic carbon content by the factor 1.724 as reported by Hesse (1972). Both ammonium and nitrate nitrogen were extracted with 0.5 mol/l $CaCl_2$ and estimated by distillation in the presence of MgO and Devarda's alloy, respectively. Available P (P-Olsen) was extracted with 0.5 mol/l $NaHCO_3$ and measured by spectroscopy. Finally, exchangeable K was extracted with 1 mol/l CH_3COONH_4 and measured by Flame Photometry (Essex, UK).

According to analysis, the greenhouse soil was loamy sand, slightly calcareous, with alkaline pH, low organic matter content and high Cation exchange capacity (Table 1). The electrical conductivity of soil extracts (water soil extract in ratio1:5) was 0.66 dS/m, indicating marginally increased soil salinity (Chouliaras *et al*., 1996).

Table 1.

Chemical soil properties at the beginning of crops

Soil properties	A Cultivation	B Cultivation	C Cultivation	D Cultivation
pH	8.13	7,95	7,50	7,40
$CaCO_3$ (%)	5.80	5,60	5,70	5,28
Organic matter (%)	0.74	1,47	2,10	1,93
Cation exchange capacity (cmol/kg)	23,0	26,5	27,3	23,8
Electrical conductivity in soil extract (1 soil : 5 H_2O, dS/m)	0.66	0,58	0,80	0,55

A. Crop:. Only an organic commercial fertilizer 25 tons/ha (that contains 1,5 kg N, 3,8 kg P_2O_5 and 2,5 kg K_2O per 100 kg fertilizer), in the form of black peat moss slightly enriched with inorganic elements was incorporated in soil before plant transplantation.

Cucumber plants (var. Gador, 10^4 roots per hectare were used) was transplanting at early April 2008 in greenhouse, and in an area of 100 m^2. During cultivation period (April-July 2008) plants were watering with good quality water (EC = 0.5 dS/m) while the fertigation programme organized according to plant growth, blooming and fruit load, and soil salinity changes. Forty days after transplanting, leaf samples were taken for plant nutritive elements assessment. As the concentration of N and K in plants was found to be slightly low, fertigation programme modified according to CTIFL guide (1989). Totally, during the whole growing period 110 kg N, 80 kg P (P_2O_5) and 130 kg K (K_2O) per hectare were used for plant fertigation. At the end of growing season, soil covered by transparent polyethylene plastic for soil solarization and the soil electrical conductivity was measured three months later.

B. Crop: Later settled the second crop (October-April 2009), following the same procedure, and during the whole growing period 200 kg N, 200 kg P (P_2O_5) and 200 kg K (K_2O) per hectare were used for plant fertigation. At the end of growing season, soil was covered by transparent polyethylene plastic for soil solarization, the soil electrical conductivity was measured three months later, and, the same organic commercial fertilizer with 45 tons/ha was applied .

C. Crop: The third cultivation lasted from October-April 2010 and during the whole growing period 80 kg N, 80 kg P (P_2O_5) and 80 kg K (K_2O) per hectare were used for plant fertigation. At the end of growing season, soil was also covered by transparent polyethylene plastic for soil solarization, the soil electrical conductivity was measured three months later, and, the same organic commercial fertilizer of 45 tons/ha was applied .

D. Crop: The fourth cultivation lasted from October-April 2011 and during the whole growing period 80 kg N, 80 kg P (P_2O_5) and 80 kg K (K_2O) per hectare were used for plant fertigation. At the end of growing season, soil was equally covered by transparent polyethylene plastic for soil solarization and the soil electrical conductivity was measured three months later.

The experimental design was completely randomized with four replications for each sample analysis. Data was analyzed using Microsoft Excel analysis Tool-Pak.

RESULTS AND DISCUSSION

The fertigation program decreased soil salinity as indicated by EC reduction from (0.66 to 0.24, 0.58 to 0.24, 0.80 to 0.49 and 0.55 to 0.39 dS/m respectively) during the cultivation period (Fig 1). However, soil solarization after plant removal in the first and second crop increased soil salinity (from 0.24 to 1.23 and 0.24 to 0.48 respectively) due to soluble salts mobility to soil surface caused by intensive evaporation (table 1). After soil solarization the third and the fourth crop the electrical conductivity has remained constant (from 0.49 to 0.44 and 0.39 to 0.40 respectively) which confirms the improvements of the soil salinity.

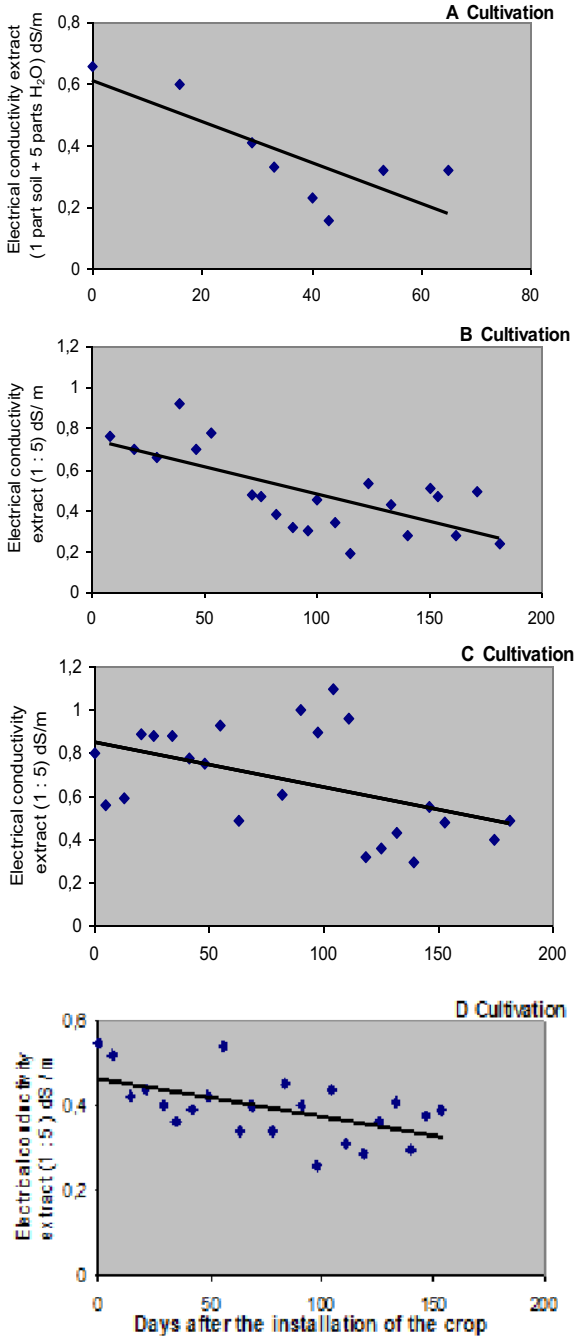

Figure 1. Changes in soil salinity during successive cultivation periods.

Table: 2 shows the balance of available inorganic elements in the soil during growing period. The availability of minerals is due to fertilizer residues from previous crops and nutrients applied via irrigation in the current crop. These data confirm the use of nutritive elements of soil salts from plants. In this case, the salinity level has to be considered for the following crops.

Finally, the fertigation programme did not affect plants yield. The total production of greenhouse cucumber was 6.4 kg/ m^2 (A-Cultivation), 9.0 kg / m^2 (B-Cultivation), 9.0 kg/ m^2 (C-Cultivation) and 9.1 kg / m^2 (D-Cultivation),

Table 2.

Inorganic elements availability in greenhouse soil

	N (kg/ha) Inorganic	P_2O_5 (kg/ha) P-Olsen	K_2O (kg/ha) Exchangeable	Electrical conductivity dS/m
A -Cultivation				
Soil Content at the Starting of growing season	490	70	1040	0.64
Base +Surface fertilizer application	110	80	130	0.24 (end crop period)
Soil Content after soil solarization	730	80	1180	1.23
B- Cultivation				
Soil Content at the Starting of growing	290	72	1020	0.58
Base + Surface	215	238	223	0.24(end crop
Soil Content after soil solarization	270	90	866	0.48
C- Cultivation				
Soil Content at the Starting of growing	556	62	1070	0.8
Base + Surface Fertilizer	106	149	121	0.49(end crop period)
Soil Content after soil solarization	330	140	1030	0.44
D- Cultivation				
Soil Content at the Starting of growing	556	114.9	885.8	0.55
Base + Surface	106	149	121	0.39(end crop
Soil Content after soil solarization	265	115	873.9	0.40

CONCLUSION

This study shows that greenhouse soil salinity due to accumulation of fertilizers (residual effect), could be taken into account for basic mineral fertilizer omission. In this way, plant needs for nutrients might partially be covered by soil salts, leading to limited fertilizer application and soil salinity reduction by the end of the growing season. The salinity level in soil should be considered for the management of the cultural practices in successive crops.

In addition, the high content of organic matter in soil reduces the negative effects of salinity (Chouliaras, 1996).

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THE INFLUENCE OF THE WINEMAKING TECHNOLOGY ON THE SENSORIAL CHARACTERISTICS OF THE VARIETY SAUVIGNON BLANC FROM THE GOLUL DRÂNCEI – MEHEDINTI WINE CENTER

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Key words: *Oprisor, Sauvignon Blanc, yeast, sensorial characteristic,*

ABSTRACT

For the white wines, especially for the authentic semiaromatic and aromatic varieties, the intensity and complexity of the wine flavor represents a factor with a decisive influence over the next steps of tasting and/or consumption. The purpose of this work is to evaluate the influence of the various winemaking technologies and oenological products on the quality, diversity and complexity of the sensorial characteristics of the wines obtained from Sauvignon Blanc grapes from Golul Drâncei – Mehedinți wine center using sensorial analysis. The Sauvignon Blanc wines obtained usind different winemaking technologies have been sensorially analyzed by authorised winetasters using specialised and adapted winetasting sheets for different and specific sensorial descriptors. The results have proven that some winemaking techniques and some oenological materials like selected yeasts significantly influence the wine flavour, increasing even its tipicity.

INTRODUCTION

Ever since the oldest times, the hill areas from south Mehedinti have proven excellent conditions for the vine grow and especially for the red wine producing grapes. In „ Agricultura Romana din judetul Mehedinti", the great savant Ion Ionescu de la Brad (1868) mentionned that „... the greatest benefits from the vineyards come from Blahnita and Campul areas, where are located the famous vineyards of Orevita, Rogova, Drancia, Oprisorul." (Gheorghită M. et al., 2006).

The production of semiaromatic wines with controlled denomination of origin is made in Romania in strictly delimitated areas, where the favourable pedoclimatic conditions are capitalized through the means of grape varieties with a high capability of flavour and sugars accumulation and concentration in the berries (Begea Mihaela et al., 2005). In this context, it is very well known the potential of the wine center Golul Drancei – Mehedinti where there can be produced out of the Sauvignon Blanc variety, DOC wines as

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well as DOC-CT (late harvest) and DOC-CMD (full maturity harvest) (Grigorică L., 2002)– Table 1.

The production conditions of these wines were established as a result of the researches made during the time at the Research Station Banu Maracine and the observations from the production activity (Antoce O.A. et al., 2003).

In this work we will try to evaluate, by means of the sensorial analysis, the influence of the various technologies used in winemaking and of some oenological materials (Bertrand A. and Beloqui A., 1996, Castino M. and Scandolo A., 1996) over the quality, diversity and complexity of the wines made out of the Sauvignon Blanc variety from the Golul Drancei - Mehedinti wine center.

In this work we have studied the influence of the clarification technology and of yeast strains used at the alcoholic fermentation over the organoleptic qualities and tipicity of the wines.

MATERIAL AND METHODS

In order to conduct this study, there were used as raw material, grapes out of the Sauvignon Blanc variety, manually harvested from Oprisor – Golul Drancei – Mehedinti area. There were used grapes from the 2009 vintage campaign, from three different parcels, the grapes from one parcel representing a reherseal for each experimental variant. The experiments took place at the Oprisor winecellar of Carl Reh Winery.

In table 2 there can be found the experimental variants and the winemaking technology used in order to obtain them. All the variants have in common the addition of potassium metabisulphite 55 mgSO_2/l applied on the mash during the maceration, the gravitational clarification and the addition of sulphur dioxide 60 mg SO_2/l after the alcoholic fermentation has ended. The difference between the variants consisted in the way the clarification was made (gravitational or enzyme-gravitational) and the use or not of various selected yeast strains at the alcoholic fermentation.după încheierea fermentației alcoolice. For each variant three repetitions were made, coded with 1, 2, 3 before the variants number. Each repetition was made using homogenous mash obtained from a single parcel grapes. For the enzymatic maceration (Pinsuin M., 1996**)** it was used an enzymatic pectolytic concentrate with secondary glycosidic activities, Lallzyme Cuvee Blanc, product which gave very good results in practice and in previous testing.

As selected yeasts, three strains were used, out of the most worlwide used for the Sauvignon Blanc variety: Anchor Vin 7, Lalvin QA 23 and Lalvin D47. All of them are characterised in the technical sheets as yeasts that generate fermentation metabolites with pleasant floral ot exotic fruit aroma (Sicheri G., 2008).

RESULTS AND DISCUTIONS

The grapes had at harvesting a sugar concentration between 192 g/l and 228 g/l, all of their, being capable to produce dry Sauvignon Blanc wines, with controlled denomination of origin Mehedinti.

The alcoholic fermentation was conducted at a temperature of around 17-18ºC for all the variants, except for variant 4 which fermented at around 12 ºC, this being the recommended temperature of the producer, in order to obtain a higher aromatic intensity. The physical and chemical parameters of the wines are presented in table 3.

The sensorial analysis of the wines was made after the wine was obtained and after the first racking, in order to compare the experimental variants.

The statistic processing (variation analysis) of the results of the physical-chemical analysis has shown that there are no significant differences between the samples from the point of view of the final alcoholic concentration, sugars, total acidity, volatile acidity, free and total sulphur dioxide, the little differences resulting of the inherent variability between the samples. It is observed, however, that in the case of selected yeast use the residual sugar content is lower than in the spontaneous fermentation, probably due to the existence of less performant yeast in the indigenous microflora. The only variant that presented a difference of the extract (of 5,26% compared to the lowest extract variant) is the one that used the selected Lalvin D47 yeast, fact explained by the capacity of this yeast to produce a rather large quantity of manoproteins in the new wine.

Table 1

Sauvignon Blanc types of wine with controlled denomination of origin Mehedinti

TYPE OF WINE – Sauvignon Blanc	Sugar content of grapes at harvest, g/l, min.	Alcohol %vol. Min.	Sugars g/l	Dry extract, g/l
DOC-CT Half sweet	204,0	11,0	min. 15	21
DOC-CMD Dry	187,0	11,0	max. 4	21
DOC-CMD Medium dry	196	11,0	4 - 12	23

Table 2

Technological variants made in order to study the influence of oenological products use in winemaking the Sauvignon grapes variety - Golul Drâncei, 2009

Variant	CODE	TECHNOLOGICAL VARIANTS
Variant 1 repetition 1 Variant 1 repetition 2 Variant 1 repetition 3	SB 1.1. SB 2.1. SB 3.1	Winemaking with 6 hours maceration, gravitational clarification, spontaneous fermentation
Variant 4 repetition 1 Variant 4 repetition 2 Variant 4 repetition 3	SB 1.4. SB 2.4 SB 3.4.	Winemaking with 6 hours enzymatic maceration (Lallzyme Cuve Blanc, 2 g/hl), gravitational clarification, spontaneous fermentation
Variant 4 repetition 1 Variant 4 repetition 2 Variant 4 repetition 3	SB 1.5. SB 2.5 SB 3.5.	Winemaking with 6 hours enzymatic maceration (Lallzyme Cuve Blanc, 2 g/hl), gravitational clarification, fermentation with Anchor Vin 7 selected yeast (20 g/hl)
Variant 6 repetition 1 Variant 6 repetition 2 Variant 6 repetition 3	SB 1.6. SB 2.6. SB 3.6.	Winemaking with 6 hours enzymatic maceration (Lallzyme Cuve Blanc, 2 g/hl), gravitational clarification, fermentation with Lalvin QA 23 selected yeast (20 g/hl)
Variant 7 repetition 1 Variant 7 repetition 2 Variant 7 repetition 3	SB 1.7. SB 2.7. SB 3.7.	Winemaking with 6 hours enzymatic maceration (Lallzyme Cuve Blanc, 2 g/hl), gravitational clarification, fermentation with Lalvin D 47 selected yeast (20 g/hl)

Table 3

Physical and chemical parameters of Sauvignon Blanc wines (Golul Drâncei, 2009)

Variant	Variant code	Alcohol, %vol.	Sugars, g/l	Dry extract, g/l	Total acidity, g/l	Volatile acidity, g/l	Total SO_2, mg/l	Free SO_2 mg/l
V1	SB 1.1.	13,00	4,40	23,89	5,70	0,56	134,0	31,00
	SB 2.1.	13,60	3,80	22,90	5,30	0,47	128,0	21,00
	SB 3.1.	13,50	4,10	21,23	5,20	0,51	121,0	24,00
	average	**13,37**	**4,10**	**22,67**	**5,40**	**0,51**	**127, 7**	**25,33**
	Standard deviation	**0,32**	**0,30**	**1,34**	**0,26**	**0,05**	**6,51**	**5,13**
V4	SB 1.4.	13,20	4,60	22,87	5,68	0,52	124,0	26,00
	SB 2.4.	13,50	4,15	21,90	5,43	0,56	117,0	23,00
	SB 3.4.	13,40	3,96	21,12	5,34	0,59	134,0	32,00
	average	**13,37**	**4,24**	**21,96**	**5,48**	**0,56**	**125,0**	**27,00**
	Standard deviation	**0,15**	**0,33**	**0,88**	**0,18**	**0,04**	**8,54**	**4,58**
V5	SB 1.5.	13,20	2,24	23,32	5,76	0,57	112,0	21,00
	SB 2.5.	13,50	2,45	22,34	5,65	0,53	117,0	26,00
	SB 3.5.	13,70	1,98	20,98	5,23	0,62	121,0	24,00
	average	**13,47**	**2,22**	**22,21**	**5,55**	**0,57**	**116,7**	**23,67**
	Standard deviation	**0,25**	**0,24**	**1,18**	**0,28**	**0,05**	**4,51**	**2,52**
V6	SB 1.6.	13,10	2,89	23,65	5,75	0,34	123,0	25,00
	SB 2.6.	13,60	3,04	23,12	5,54	0,41	117,0	28,00
	SB 3.6.	13,60	2,54	21,14	5,30	0,38	122,0	24,00
	average	**13,43**	**2,82**	**22,64**	**5,53**	**0,38**	**120,7**	**25,67**
	Standard deviation	**0,29**	**0,26**	**1,32**	**0,23**	**0,04**	**3,21**	**2,08**
V7	SB 1.7.	13,20	3,40	24,32	5,65	0,41	116,0	23,00
	SB 2.7.	13,50	3,25	23,24	5,41	0,34	128,0	36,00
	SB 3.7.	13,60	2,98	21,98	5,34	0,32	125,0	26,00
	average	**13,43**	**3,21**	**23,18**	**5,47**	**0,36**	**123,0**	**28,33**
	Standard deviation	**0,21**	**0,21**	**1,17**	**0,16**	**0,05**	**6,24**	**6,81**

CONCLUSIONS

After the sensorial analysis of the technological variants and the use of different yeast strains in the winemaking process of Sauvignon Blanc variety, the following conclusions have been drawn: - the enzymatic clarification does not substantially influence the quality of wine if it is followed by an uncontrolled fermentation, generated by an indigenous microflora.;

- when using a single tupe of must and different selected yeast, the evaluation shows that from the total scoring point of view, the wines are similar in aspect and colour, but different in aroma, body and harmony;

- the use of some selected yeasts rise the quality level perceived by the winetasters because of the more intense floral aroma obtained;
- all the technological variants that have used enzymatic clarification and selected yeasts during the alcoholic fermentation obtained higher scores for nose and taste;
- there are statistically significant differences between the aroma and the taste of Sauvignon Blanc wines fermented with different selected yeasts;
- the aromas of the technological variants that included gravitational clarification and gravitational-enzymatic clarification of the must, fermentation with indigenous microflora, are not significantly different; the highest influence is due to the yeasts used in fermentation;
- in the aromatic panel used we were able to see that the aromas of elder, acacia, exotic fruit, fruit, vegetal aromas, considered to be typical for Sauvignon Blanc have been influenced by the yeast strain used.
- very interesting results are obtained using the selected yeast Anchor Vin 7 with an enhanced aromatic intensity, in what concerns the floral, exotic and elder aroma. In the same time the vegetal aromas increase, aromas that can be considered less pleasant by some consumers;
- the most balanced aroma was obtained with the selected yeast Lalvin QA23, the resulting wine having intense floral and fruity aroma as well as discrete notes of vegetal or green walnuts;
- the selected yeast Lalvin D47 didn't generate a very intense aroma, but its structure (exotic aroma, fruit, acacia) as well as the lack of intense vegetal nuances was very well appreciated by the winetasters;
- if we select the proper yeast, according to the variety and the growth area of the grapes, we can obtain results corresponding to the tipicity of the area (without major differences regarding the aroma and the structure of the wine) but with a plus concerning the finesse of taste and smell; this fact is very important because the winemaker can use requently these selected yeasts regardless of the climate conditions in order to obtain a constant quality of the wines;
- for the musts from the same area and same variety (Sauvignon Blanc from Golul Drancei), regardless the parcel of vineyard from which they come, there is no major influence from the point of view of fermentation cynetics according to the yeasts used; these behave constantly in what concerns the fermentation speed.
- regardless the winemaking technology (traditional or modern) we have the possibility to influence the aromatic tipicity of the variety, its taste and body, by using a selected yeast;
- there is the possibility to enhance the aroma considered typical for the Sauvignon Blanc variety (elder, exotic, acacia) using a selected yeast that generates a large quantity of fermentation aroma, but the winemaker must be careful not to modify the aromatic tipicity and not to enhance some flavours (vegetal aroma) which can be considered unpleasant by the consumers.
- if the consumers demands move towards more aromatic wines, this thing can be achieved using an enzymatic clarification of the must and using selected yeasts capable of producing a higher quantity of aromatic fermentation metabolites.

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RESEARCH REGARDING THE INTENSITY OF PHOTOSYNTHESIS AND RESPIRATION AT PEACH TREES DEPENDING ON VARIETY

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Key words: *photosynthesis, respiration, photosynthetic active radiation*

SUMMARY

The physiological processes conducted at leaf level are dependent on a series of climatic factors- temperature, water, light, wind, etc, but also on biological factors-species, variety, age, the presence or absence of fruit. In the present experience, with 7 peach tree varieties used, the photosynthesis was strongly influenced by the variety, but also by the temperature and light intensity. The intensity of photosynthesis was negatively influenced by strong light. The respiration was negatively influenced by temperature and positively influenced by stomata conductance.

INTRODUCTION

The intensity of physiological processes which develop on leaf level is influenced by many factors such as: the age of the leaf and its position on the shoot (Kappel F., Flore J.A., 1983, Sams C.E., Flore J.A., 1982), the illumination grade, (Marini R.P.,Marini M.C., 1983), the species and the variety, the presence or the absence of fruit (De Jong T.M., 1986, Flore J.A., Gucii R., 1988), the health of leaves, water and mineral elements supply level (De Jong T.M., 1986, Flore J.A., Gucii R., 1988).

For a technologist, the knowledge of these factors is very important in order to assure the fundament of an appropriate supply of organic substances for the plant and the fruit, by assuring proper conditions in which the physiological processes are developed. A defective management and maintenance of peach trees leads to insufficient light supply and to a decrease of photosynthesis rate, with direct influence on the quantity and the quality of the production. On fruit bearing trees, due to consumption centers represented by fruit, the intensity of photosynthesis increases compared to the ones which lack fruit (Hoza, 1997).

In order to acknowledge the way in which the variety can influence the process of photosynthesis and of respiration, the experiment was conducted with 7 peach tree varieties less cultivated in the area, for which the data obtained was combined in different patterns in order to observe possible dependences among studied parameters.

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MATERIAL AND METHODS

The experience was conducted in the Experimental field of Horticulture Faculty, Bucharest, in the year of 2011, with 7 peach tree varieties: Cardinal, Inka, Reliance, Royal estate, Early Rich, October star and Late Luka. The trees were 5 years old, planted at 5/1 m distance and managed with transverse epsilon crown. The soil was worked on the row and maintained with grass on the spacing area. In the orchard, the specific technological measures applied were appropriate for fruit bearing trees.

The measurements regarding the intensity of photosynthesis and of respiration, the photosynthetic active radiation on leaf level (P.A.R), the stomata conductance and the temperature at leaf level were made with the portable device LCi.

RESULTS AND DISCUSSIONS

The studied varieties greatly influenced the intensity of analyzed physiological processes. Therefore, the intensity of photosynthesis had different values among varieties, oscillating from 5.57 mmoles/m^2/s at Reliace and 12.70 mmoles/m^2/s at Cardinal. High values were registered at varieties such as Late Luka (11.81 mmoles/m^2/s), October star (9.61 mmoles/m^2/s) and Inka (9.41 mmoles/m^2/s). Values lower than 8 mmoles/m^2/s were recorded for the varieties Early Rich and Royal estate (fig. 1). Considering the fact that the measurements were conducted in similar climatic conditions and on leaves with similar positions in the crown, the differences can be explained only through the characteristics of each variety. In comparison with values noted in the specific literature (12.7-23.2 mg CO_2/dm_2/hour, Marini and Marini, 1983), the intensity of photosynthesis was lower, probably due to very high temperature registered on the leaf level at that time, over 35 degrees Celsius.

The intensity of respiration was influenced by variety, but variations were lower in comparison with photosynthesis, remaining in the interval of 2.19 mmoles/m^2/s, for Inka variety, and 6.48 mmoles/m^2/s, at Cardinal variety. For this process, the varieties which presented similar values were put together in groups of 2-3 varieties (fig.2).

Both photosynthesis and respiration were influenced by temperature and stomata conductance, but the influence was different for each variety.

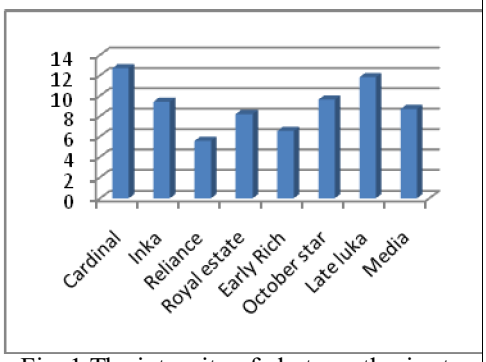

Fig. 1 The intensity of photosynthesis at some peach tree varieties

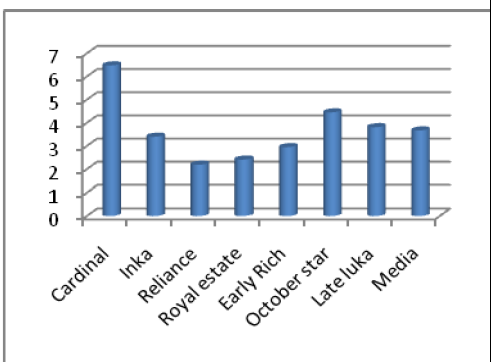

Fig. 2 The intensity of respiration at some peach tree varieties

The dependence between the features studied was proved throughout the correlations made. Therefore, the observations highlighted that photosynthesis was negatively influenced by strong light intensity, registered values being with over 55% from photosynthetic active radiation (3000 mmoles/m²/s). The values of the correlation coefficient, r^2=0,14, show a weak correlation, but it is important to know that the maximum amount of light the leaves can be exposed to is not always favorable for an appropriate synthesis (fig.3).

The calculation of the correlation coefficient between stomata conductance and photosynthesis (fig.4) proved that photosynthesis was not influenced by this feature. The gas exchange from substomata chamber to the exterior was normal, therefore, the decrease of photosynthesis was not charged to this parameter.

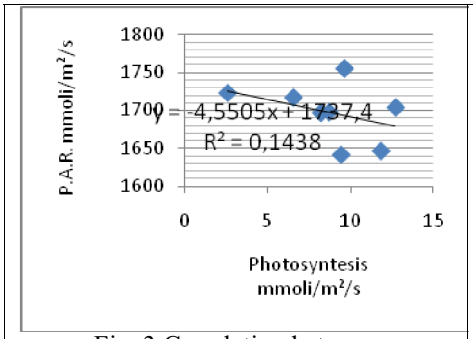

Fig. 3 Correlation between photosynthesis and P.A.R.

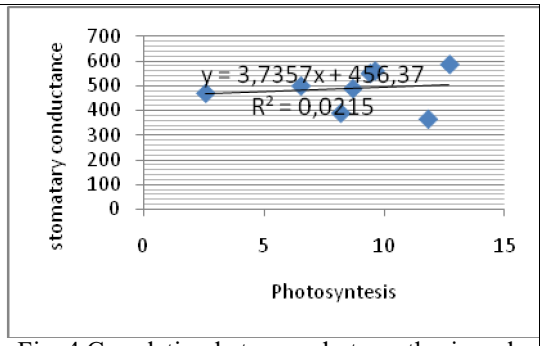

Fig. 4 Correlation between photosynthesis and stomata conductance

The opening grade of stoma and the gas circulation through stoma substantially influenced the respiration. The correlation coefficient and orientation of the regression line prove a direct connection between stomata conductance and respiration (fig.5).

Photosynthesis was negatively influenced by temperature. Figure 6 proves the indirect correlation between photosynthesis and temperature, the first one decreasing as the second one increased.

Respiration was also negatively influenced by temperature. The value of the correlation coefficient was in this case higher in comparison with photosynthesis (fig.7).

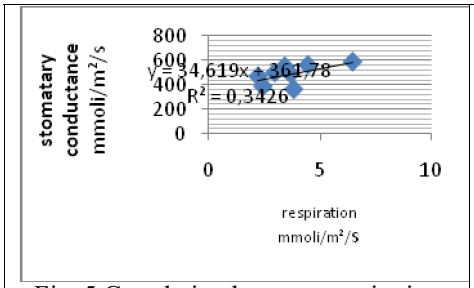

Fig. 5 Correlation between respiration and stomata conductance

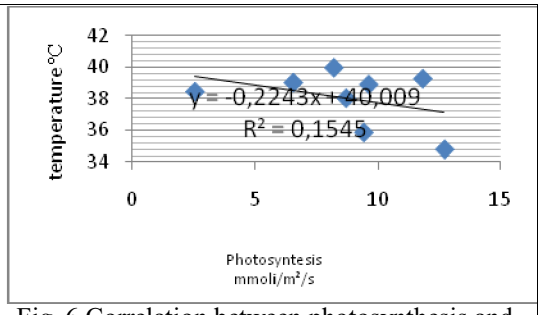

Fig. 6 Correlation between photosynthesis and temperature

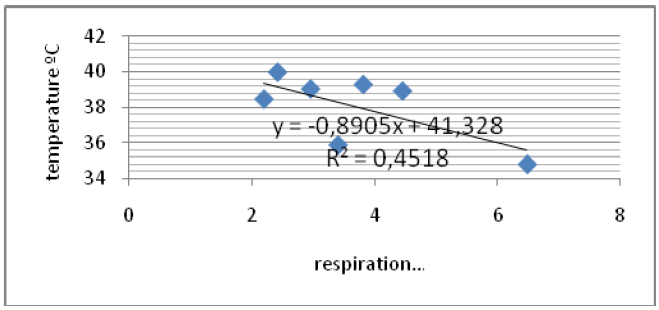

Fig. 7 Correlation between respiration and temperature

CONCLUSIONS

From the present study, the following conclusions can be drawn:

The physiological processes developed on leaf level at peach trees are extremely complex and greatly influenced by biological and environmental factors;

The intensity of photosynthesis was negatively influenced by strong light and high temperature;

The intensity of respiration was negatively influenced by temperature and positively influenced by stomata conductance.

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RESEARCH REGARDING THE INFLUENCE OF TOMATO PLANT MANAGEMENT FOR CULTURES GROWN IN SOLARIUM, IN EXTENDED PRODUCTION CYCLE

Hoza Gheorghița[1], Stanciu Laura Georgiana[1]

Key words: *solarium, tomatoes, two stems*

SUMMARY

The research has been conducted on the experimental production field of Horticulture Faculty, Bucharest. The tomatoes have been cultivated in solarium, in extended production cycle. The plants have been conducted with two stems, in three different variants. The first variant of stems has been represented by the first spring formed at the base of the plant and the stem itself. The second variant has been represented by the stems formed from the springs emerged in the armpit of cotyledonary leaves, resulted after pinching the plants in the second phase of leaf growth. The third variant has been represented by a control singular stem. The planting has been made at 80cm/30 cm for control and 110cm/60cm for variants with two stems, reducing therefore the number of seedlings for 1 hectare with 63% for the last ones. The observations from the vegetative growth point of view have proven that plants managed both with one and two stems reveal similar values, reaching a height of approximately 170cm. From the fruit point of view, the number and the production per plant registered at variants with two stems have been much higher, the percentage calculated being of 203% and 242% in comparison with the control variant. In what regards the production per unit of area, the values have been lower with 12% and 27% for two stems, due to the 63% decrease of seedlings used.

INTRODUCTION

The tomato culture represents one of the most appealing vegetable cultures due to its indubitable importance for human nourishment. Considering its major role, the present paper is aimed to test the growth and fructification capacity of tomatoes managed differently from the classical method, with less density of the plants. The possibility of conducting tomatoes on two stems is not a new idea in other countries, but in ours this method is very rarely used.

The two stems can be obtained as follows: by preserving the first spring formed at the base of the stem and the stem itself (Ceausescu 1984, Arthur şi Henry 1993, Diana Relf, Ronald Morse 2009); by pinching the plant in the first phases of growth, when springs emerge from the buds located at the armpit of cotyledonary leaves. Depending on the planting scheme, managing plants with two stems result in a decrease of number for plants per hectare with 20% (Hoza Gheorghița, 2011).

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MATERIAL AND METHODS

The culture has been established in the teaching field of Horticulture Faculty in Bucharest.

The experimental three variants managed in solarium have been the following:

- V1- the control plant, managed with one stem, the planting scheme: 80 cm between rows/30 cm between plants/row, resulting 41 668 plants/ ha;
- V2- the plant with two stems, the first one being the stem itself and the second one being the first spring located at the base of the initially stem, the planting scheme: 110 cm between rows/60 cm between plants/row, resulting 15151 plants/ha;
- V3- the plant with two stems, formed from springs resulted after pinching the top of the plant in its seedling phase of growth, the planting scheme: 110 cm between rows/60 cm between plants/row, resulting 15151 plants/ha.

Due to the planting scheme presented, the density for one hectare decreased with 26515 pl/ha, reaching a rate of 63% of decrease.

Forming the two stems

The stems have been conducted with two different methods:

- **Stems formed from springs located at the base of cotyledonary leaves.** Immediately after transplanting, when plants regained their natural growth and started producing the second pair of true leaves, the small stem located at the base of cotyledonary leaves has been removed. From buds located at the armpit of cotyledonay leaves two springs have been formed, which have become the two stems needed.
- **Stems formed from the first spring emerged at the base of the plant and the initial stem.** In this case, the first spring from the base of the initial stem has been preserved and sustained individually on the string, becoming the second stem needed. The rest of the springs have been removed.

The observations and measurements conducted in this experiment refer to: the height of seedling and the number of leaves, the height of the plants, average distance between inflorescences, average number of flowers in the inflorescence, average number of fruits in the inflorescence, the percentage of fructification, the production obtained, average weight of fruit and categorizing fruit in weight classes.

RESULTS AND DISCUSSIONS

In what regards the growth dynamics of tomato seedling (table 1) the following results have been registered. For plants with one stem, the height increased gradually from 11.0 cm to 17.3 cm and the number of leaves oscillated between 4 and 7. For plants with two stems, the height has had much lower values, as a consequence of removing the upper part of the plants, preserving only the cotyledonary leaves.

Therefore, the height of the plants, dated when the last measurements have been made, has been of 10.2 cm and respectively of 9 cm for the second stem

Table 1

The growth dynamics of tomato seedlings

Date	24.03				31.03				7.04			
Seedling type	Height		No. lvs		Height		No. lvs.		Height		No. lvs	
Seedling with one stem	11.0		4		15.1		5		17.3		7	
Seedling with two stems	2.4	2.1	3	2	5.1	4.7	4	3	9	10.2	5	4

. In what regards the number of leaves/plant, the seedling with two stems prove different values in comparison with one stem conducted plants and the values constantly modify as the plants grow in height. It can be observed that in the first period of time, plants have had 2-3 leaves on each stem and at the end of the experimental period of time each stem has had 4-5 leaves.

From biometrical point of view, the plants conducted with two stems have behaved extremely well (table 2). The values regarding the height of plants have been proximate, oscillating between 168 and 178 cm. All plants have been conducted in extended cycle of production.

The distance from collet to the first inflorescence has been different depending on the method used to manage plants. For the variant in which one stem is formed from the spring, the distance from collet to the first inflorescence has been of 23.3 cm for the main stem and 21.3 cm for the second stem. For the variant with both stems formed from springs after pinching plants, the distance from the collet to the first inflorescence has had equal values for both stems, but lower values in comparison with the other variant presented with two stems. The average distance between inflorescences located on one plant has had proximate values.

Table 2

Biometrical indicators

Variant	Average height of plants (cm)	Distance from collet to the first inflorescence from first stem (cm)	Distance from collet to the first inflorescence from second stem/spring (cm)	Distance between inflorescen-ces Stem 1 (cm)	Distance between inflorescences Stem 2 (cm)
V1- control	168	28	-	25.4	-
V2- stem+spring	178	23.3	21.3	23.4	24.4
V3- two stems	175	17.1	17.7	23.1	21.6

In what regards the average number of flowers in one inflorescence, values presented in table 3, variants 2 and 3 have presented proximate values, reaching approximate 5 flowers each. Taken into consideration the total number of flowers per plant, variants 2 and 3 have presented higher values in comparison with variant 1, due to the presence of the two stems.

For variants with two stems, the average number of fruit per plant has reached 2-2,3 times higher values than the variant with one stem (table 4). The numbers registered have been of 47. 8 and respectively 53.5.

Table 3

Average number of flowers per plant

Variant	Average number of flowers in one inflorescence		Total number of flowers per plant	
Stem	1	2	1	2
V1-control	4.7	-	28.3	-
V2-stem+spring	5.3	4.6	31.8	27.8
	9.9		59.6	
V3-two stems	5	4.8	29.9	28.9
	9.8		58.8	

Table 4

Average number of fruits per plant

Variant	Average number of fruits in one inflorescence		Average number of fruits per plant	
Stem	1	2	1	2
V1-control	4.2	-	24.9	-
V2-stem+spring	4.2	3.8	25.1	22.7
	8		47.8	
V3-two stems	4.4	4.5	26.5	27
	8.9		53.5	

Analyzing the capacity of fructification (table 5), variant 2 has registered a higher percentage than the other two variants, due to the improvement of environmental conditions in the exact time when the flowers from the spring stem have been opening and forming fruits.

The fruit formation percentage has reached great values at inflorescence 2, at both 2 and 3 variants, being of 83.4 % on stem 1 and 86.8 % on stem 2 and respectively, 96.7% on stem 1 and 98.7% on stem 2. Variant 1 has had a good value, of 93.3 %, but the maximum percentage registered at control has been on the first inflorescence, 95.2%.

Table 5

Fruits formation percentage, %

Variant	Inflorescence											
	1		2		3		4		5		6	
Stem	1	2	1	2	1	2	1	2	1	2	1	2
V1-control	95.2	-	93.3	-	93	-	78	-	78	-	92	-
V2-stem+spring	78.6	85.8	83.4	86.8	81.8	78	76.8	74.9	77	87.6	76.1	85.7
V3-Two stems	87.1	93.3	96.7	98.7	83.4	95.7	90.3	88.1	86.4	87	86	96.2

In what regards the production obtained (table 6) measured in kg/pl, variants 2 and 3 have registered substantially higher values than the control. But at measurements made in kg/sqm, variant 2 (stem+ spring) has registered substantially lower values than control and variant 3 (with two stems) has reached insignificant values in comparison with the control variant.

Considering the production per plant, the higher value has been registered at variant 3, with 6.3 kg/pl, exceeding the control variant with 3.7 kg.

The second place has been taken by the second variant, with 1.06 kg/pl less than variant 3. Even so, variant 2 produced 2.64 kg/pl more than control.

The relative production has been of 242% for the variant with the two stems obtained by pinching the plants and of 203% for the variant with the stems obtained from one spring and the main stem, proving a higher rate of production in comparison with the control variant.

Analyzing the production measured in kg/sqm, the absolute production registered has been of 10.9 kg for control, 8 kg for variant 2 and 9.6 kg for variant 3. Although at first sight the values seem lower for the variants with two stems, overall, the advantages in managing tomatoes with these methods are highly effective due to the 63% decrease of seedling use per hectare.

Table 6

Production of fruit

Variant	Average weight of fruit (g)	Production					
		Per plant			Per m^2		
		kg	%	Signifi-cance	kg	(%)	Signifi-cance
V1	105	2.6	100	Mt	10.9	100	Mt
V2	110	2.8 | 2.5			4.2 | 3.8		
		5.3	203	xxx	8	73	00
V3	118	3.1 | 3.2			4.7 | 4.8		
		6.3	242	xxx	9.6	88	N

DL 5% -0.3 kg
DL 1 % -0.5 kg
DL 0,1 % -0.93 kg

DL 5% -1.61 kg
DL 1 % -2.66 kg
DL 0,1 % -4.98 kg

In what regards the size of fruit measured throughout weigh , the best values have been registered in 100-150 g category, valid for all variants as it follows: 35.4 % for V1, 38.6% for V2, 40.5 % for V3. For the 50-100 g category, the results reached a rate of over 30 % for all variants (table 7).

Table 7

Fruit percentage classified in different classes of weight %

Variant	Under 50 g	50-100 g	100-150 g	Over 150 g
V1-control	27.3	30.3	35.4	7
V2	21.5	35.7	38.6	4.2
V3	15.4	30,7	40.5	13.4

CONCLUSIONS

The following conclusions can be drawn from data obtained while analyzing the tomato culture, cultivated in solarium and managed with two stems:

- Managing plants with two stems results in a decrease of seedling use in order to establish the culture. The number of plants used to start a tomato culture with one stem is 41666 pl/ha and the number of plants used for cultures with two stems is of 15151 pl/ha, which proves a decrease of 63% , depending on the plantation scheme;
- The average number of fruit per plant registered has had values of 2-2,3 times higher for variants with two stems in comparison with control variant, managed only with one stem;
- For variants with two stems, the production per plant has been significantly positive in comparison with control, but the production per unit of area has been lower than the one registered at control due to the lower number of plants used and managed with two stems;
- The relative production rate for the variant in which stems have been obtained after pinching the plants has been of 242% and 203% for the variant with main stem and spring stem, which proves a higher rate of production for two stems in comparison with the control variant;
- The average weight of fruits has been of 120g, the majority being classified in the 100-150g category: 35,4 % for variant 1, 38,6% for variant 2, 40,5 % for variant 3, followed by 50-100 g category which registered over 30% of fruits for all three variants.

RECOMMENDATIONS

- Cultivating tomato plants in solarium, in extended production cycle and managing plants with two stems for a substantially higher production rate;
- Reducing the distance between plants per row for a higher production rate.

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FOOD SAFETY AND HYGIENE ENQUIRY IN HONEY PROCESSING UNIT

Iancu Ramona Maria[1], Oprean Letitia[1], Tita Mihaela Adriana[2], Lengyel Ecaterina[2]

Key words: *food, processing, safety, hygiene, health, ATP bioluminescence.*

ABSTRACT

The central purpose of this study was to evaluate the hygiene conditions in an honey processing unit, to minimize the risk of product contamination, to fulfil their partners requests. The current work was performed during three month in 2011 and the level of contamination was done from walls, equipment surfaces and personal hands using Fast Sanitation Tests with the possibility of reading the results on the spot using Ultrasnap vials. These were analysed by Syste Sure II. In the framework of the audit visit, we observed an increase of positive tests, which exceed the limit of 30 URL but within acceptable limits of 30-100 URL. The basis of excellent hygiene is in well prepared personnel, much attention is paid to food processing equipment, which defined critical control point. Documents sanitation procedures prevents the contamination of product and improve the safety of human health.

INTRODUCTION

Honey production industry is rapidly gaining in importance throughout the world, even in our country. According to Agriculture Minster, in 2010, the main producer of honey in the world is China with 398.000 tones, followed by Turkey with 81.115 tones. Romania ranks only 22.222 tones, but for our healthy is very important to ensure a strong link between conservation and continuous improvement of the genetic local beekeeping and implement practices based upon food storage, sanitation, personal hygiene.

The most important factors contributing to the changes in quality and safety of food are from these specific categories: environment, demographic, economic, technology and infrastructure, social and political. The main routes of contamination are from agricultural sector in general and food chain personal, in particular (Fares, 2010 Nielsen, 2004).

In addition, easier access to safe food increase our productivity and our status. (Fraser, 2009).

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MATHERIAL AND METHODS

The central purpose of this study was to evaluate the hygiene conditions in an honey processing unit, to minimize the risk of product contamination, to fulfil their partners requests.

The current work was performed during three month in 2011 and the level of contamination was done from walls, equipment surfaces and personal hands using Fast Sanitation Tests with the possibility of reading the results on the spot using Ultrasnap vials. These was analysed by Syste Sure II.

The method principle consists in the reading on the bioluminescence of ATP (adenosine triphosphate), the chemical compound in which is stored energy in all living cells. In the case of surface contamination, a pen-like device containing reagents is simply swabbed over an area, then inserted into a handheld reader that displays results within seconds in relative light units (RLU), indicating clean/unclean and, in some cases, marginal results.

The most common method for detecting ATP based on bioluminescence measurement is using the luciferin substrate/luciferase enzyme system (luciferin 4-monooxygenase), the reaction being presented below:

D-luciferin + ATP = luciferase $\xrightarrow{Mg^{2+}}$ luciferase adenylate complex + free phosphate (PPi)

In presence of molecular O_2, the luceferin adenylate complex is oxidized to oxyluciferin. During this metabolic process, photons are omitted. (Ceresa, 2004)

Luciferil adenylate complex + O_2 oxyluciferin-luciferase + light (bioluminescence) + AMP + CO_2

ATP bioluminescence is being used in the food industry as a rapid, simple and reliable test to monitor surface contamination during food processing. (Horiuchi, 2003)

RESULTS AND DISCUSSIONS

Interpretation of the results was made on the basis of the average value obtained in the course of all three visits. According to the hygiene survey results obtained from direct and indirect contact surfaces the amount of tests over limit of 30 RLU varied between visits.

Results obtained from tests Ultrasnap, where the minimum limit underside is 0 RLU (relative light units) and the maximum is 30 superior RLU, carried out in the processing covered by the study, are presented in Table 1.

Table 1

RLU test in direct product contact surfaces

Microbial load (RLU test)	Amount of sample, %								
	Visit 1			Visit 2			Visit 3		
	PL	PE	PH	PL	PE	PH	PL	PE	PH
≤ 30	25	15	30	47	43	67	60	59	72
31-100	8	9	22	32	38	12	36	46	28
101 – 500	53	47	24	12	10	20	4	5	-
501-1000	8	9	24	9	9	1	-	-	-
≥ 1000	6	20	-	-	-	-	-	-	-

PL – production line, PE – production equipment, PH – personal hands

In working conditions it is impossible for the tests to frame within the limits imposed, but notice an evolution, in a positive sense, the number of tests that go beyond the domain of values for older units. Thus, in the framework of the audit visit, we can observe an increase of tests, which exceed the limit of 30 URL but within acceptable limits of 30-100 URL.

In accordance with Smith et all, (2002) and Raspor et all. (2008), the basis of excellent hygiene is in well prepared personnel, much attention is paid to food processing equipment, which defined critical control point.

Documents sanitation procedures prevents the contamination of product and improve the safety of human health.

CONCLUSIONS

The rapid microbiology method has evolved in the recent years in the context of Food Safety and Quality Assurance, that is why it is an important and necessary investment for all stakeholders, conclusion express also by Finardi (2012) in his work „Food safety issues: From Enlightened Elitism towards Deliberative Democracy?".

The relationship between healh and organization, managemnet and coordination of the food chain process is going to be in the focus of public interest and, because of that, Food Industry have to prepare an essential Quality Assurance Programme for every marketed product.

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SEASONAL VARIATION OF NITRATES AND NITRITES IN MILK AS RAW MATERIAL

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Key words: *nitrates residues, Griess method, colorimetric method with m-xilenol.*

ABSTRACT

Nitrates compounds have insignificant toxicity. They are considered virtual toxic substances due to their transformation into nitrites which have well known toxic potential. The milk contaminated with nitrates must be severely restricted. Seizure and destruction of the milk is not necessary in case of nitrates contamination.

The main objective of this study was to determine the level of nitrates and nitrites in 60 samples of raw milk collected from different seasons (15 samples/each season).

INTRODUCTION

The main sources of milk contamination are: water, intentional adulteration of milk with ammonium nitrate and nitric acid used to remove the "milk stone" from plate pasteurizers. Milk nitrites are extremely toxic especially for children.

The polluted water with nitrites and nitrates can reach milk especially through sanitation of equipments and containers. The main source of water contamination is the ammonium nitrate used for basic chemical fertilizer in agricultural practice (Şindilar, 2000).

Usually the quantity of nitrates which can be eliminated through milk is very low – below 4-5mg/l. The concentration of milk nitrites depends also on the milk processing method. Through pasteurization their concentration can increase twice and through thermal treatments like sterilization, it can increase over 4 times. In the case of cheese manufacturing the most quantity of nitrites and nitrates from milk are eliminated through whey. The residual quantity is gradually decreasing through ripening process (Guş Camelia, 2005).

The main source of water contamination is through ammonium nitrate used as basic chemical fertilizer in agricultural practice. The second contamination source with nitrites and nitrates of milk is through nitric acid careless used to remove the milk stone from plate pasteurizers. The most dangerous contamination is performed in case of intentional adulteration with ammonium nitrate (Mirela Jimborean, 2011).

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Nitrites and nitrates end up in the body of animals through feed and water. Although animals can feed with high quantities of nitrates and nitrites, the organic tissues contain relatively low amounts due to fst metabolisation and excretion through urinary tract. In addition, a high amount of nitrites and nitrates is transformed in ruminants by ruminal microflora (Costin, G., 2008).

MATERIALS AND METHODS

SR 10314/84 standard establishes the methods for determination of nitrites and nitrates from raw and pasteurized milk from different animal species. The reference method used for nitrites from milk is Griess method. The reference method for nitrates is colorimetric method with meta-xilenol.

Through this method total nitrogen is determined. The nitrate content was calculated through difference between total nitrogen and quantity of nitrite determined by Griess method and expressed as nitrate equivalent.

The method of reducing nitrate to nitrite in Cadmium- Copper environment is used in case of litigation. It is recommended to use reagents of p.a. purity and distilled water without nitrates or nitrites.

Sampling is performed according to product regulations, following SR 9535/1-74. Samples preparation is performed according to SR 6343/81.

RESULTS AND DISCUSSIONS

Limits for NO_3^- and NO_2^- from milk and recommended measures to be taken are:

For NO_3^- :

- No NO_3^- , but correlated with NO_2^- is admitted for free consumption;
- Up to 10 mg/l is admitted to human consumption without restrictions;
- From 10 mg/l until 100 mg/l is excluded from baby foods under 1 year old and it is admitted for adult foods;
- Over 100 mg/l is excluded from consumption and is used for manufacturing fermented cheese products;

For NO_2^- :

- No NO_2^- present but correlated with NO_3^- is admitted free for consumption;
- Under 10 mg/l is admitted without restrictions in consumption;
- Over 10 mg/l, regardless the quantity of NO_3^- is excluded from per se consumption and it is being used in fermented cheese manufacturing (Bondoc, 2007).

In some countries is admitted the addition of nitrates in milk used for cheese manufacturing in order to prevent cheese bloating due to *Coliform* sp, but especially to

prevent delayed cheese bloating due to *Clostridia sp*. A part of nitrstes are eliminated through whey and some are transformed in maturation process in inoffensive compounds (Costin, 2003, Mirela Jimborean, 2006).

Values of nitrates and nitrites levels determined in milk as raw material are presented in Table 1 and Figure 1. Statistical analysis of nitrates and nitrites values is presented in Tables 2 and 3.

Table 1

Variation of nitrates and nitrites content in raw milk

Crt. No	Season	Samples/ season	Nitrates NO_3^-, mg/l			Nitrites NO_2^-, mg/l		
			0-0,05	0,051-0,1	> 0,1	0-0,05	0,051-0,1	> 0,1
1.	A	15	3	12	-	10	5	-
2.	W	15	-	14	1	8	7	-
3.	Sp	15	4	11	-	13	2	-
4.	Su	15	1	14	-	13	2	-
TOTAL		60	8	51	1	44	16	-
%		100	13,3	85	1,7	73,3	26,7	-

legend: A/ autumn; W/ winter; Sp/ spring; Su/ summer.

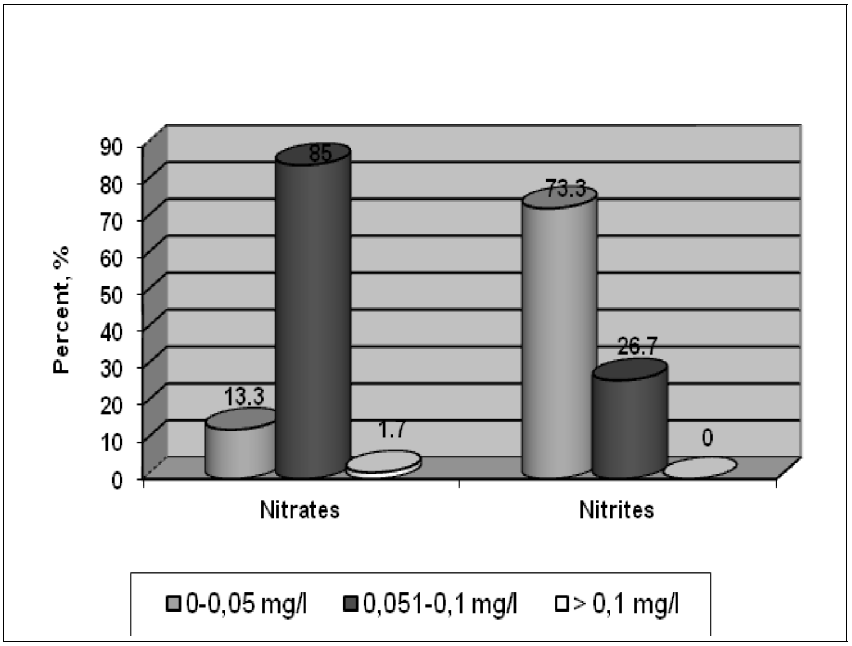

Fig. 1. Variation limits of nitrates and nitrites content in milk

Table 2

Statistical analysis of **nitrates** (NO_3^-) content detected in raw milk per seasons

Season	Minimum value, mg/l	Maximum value, mg/l	Mean, $\overline{\mathbf{X}}$	Standard deviation of the mean	Standard error of the mean, $\mathbf{s}_{\overline{\mathbf{X}}}$	Coefficient of variation, %
Winter	0,0570	0,3000	0,08867	0,05992	0,01547	67.57
Spring	0,0010	0,0920	0,0552	0,01844	0,004762	33.41
Summer	0,0500	0,0610	0,05247	0,002900	0,0007488	5.53
Autumn	0,0010	0,0920	0,05287	0,02442	0,006306	46.19

Table 3

Statistical analysis of **nitrites** (NO_3^-) content detected in raw milk per seasons

Season	Minimum value, mg/l	Maximum value, mg/l	Mean, $\overline{\mathbf{X}}$	Standard deviation of the mean	Standard error of the mean, $\mathbf{s}_{\overline{\mathbf{X}}}$	Coefficient of variation, %
Winter	0,0200	0,0800	0,0500	0,01852	0,004781	37.03
Spring	0,0100	0,0700	0,02667	0,01952	0,005040	73.19
Summer	0,0	0,0600	0,0220	0,01935	0,004995	87.94
Autumn	0,0100	0,0800	0,0360	0,02720	0,007024	75.56

From data presented in table 2 and 3 and Figure 1 it is noticed that from all 60 samples taken into study no sample exceeded the maximum limit admitted for nitrites and nitrates which is 10mg/l milk.

In the dry matter of preserved feed the level of nitrates may reach 4% of dry matter. A part of these nitrates can be eliminated through milk.

Animals can ingest high quantities of nitrates from feed and water and use them in biosynthesis processes of intestinal flora, therefore they are accumulating in small amounts in meat, milk, eggs. About 30-50mg nitrates/l and under 0,3 mg nitrites/l are secreted through milk. In dairy products there is a wide distribution of nitrites but it is appreciated that their share in a normal diet is under 20-45mg/day. Concentrations of nitrites grow 5-10 times during severe thermal treatments, therefore new techniques of semipermeable membrane separation were developed for newlyborn and children feeding.

In Romania there is forbidden to use nitrites and nitrates in milk and milk products. Nitrites and nitrates are not toxic in normal concentrations in food. But exceeding these limits have severe consequences on human organism (Costin, 2008).

A large study performed over 210 milk samples (Şindilar E. V., 2005) included 95 collected milk samples which presented limits between 0-7.5mg NO_3^- /l, and a mean of 2.7 mg NO_3^- /l.

CONCLUSIONS

✓ **Nitrates** had a mean ($\overline{X} \pm s_{\overline{X}}$) of 0,0623 ± 0,004678 mg/l and a variability coefficient of 58.16%. Determined values were situated between 0,001 and 0,3 mg/l. Statistical analysis of seasonal variation of nitrates leads to following conclusions:

- In the autumn nitrates presented a mean value ($\overline{X} \pm s_{\overline{X}}$) of 0,05287 ± 0,006306 mg/l and a variability coefficient of 46.19%. Determined values were situated between 0,001 and 0,092 mg/l.
- In the winter nitrates presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,08867 ± 0,01547 mg/l and a variability coefficient of 67.57%. Determined values were situated between 0,057 and 0,3 mg/l.
- In the spring nitrates presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,0552 ± 0,004762 mg/l and a variability coefficient of 33.41%. Determined values were situated between 0,001 and 0,092 mg/l.
- In the summer nitrates presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,05247 ± 0,0007488 mg/l and a variability coefficient of 5.53%. Determined values were situated between 0,05and 0,061 mg/l.

✓ **Nitrites** presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,03367 ± 0,003034 mg/l and a variability coefficient of 69.81%. Determined values were situated between 0,0 and 0,08 mg/l. Statistical analysis of seasonal variation of nitrites leads to following conclusions:

- In the autumn nitrites presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,036 ± 0,007024 mg/l and a variability coefficient of 75.56%. Determined values were situated between 0,01 and 0,08 mg/l.
- In the winter nitrites presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,05 ± 0,004781 mg/l and a variability coefficient of 37.03%. Determined values were situated between 0,02 and 0,08 mg/l.
- In the spring nitrites presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,02667 ± 0,00504 mg/l and a variability coefficient of 73.19%. Determined values were situated between 0,01 and 0,07 mg/l.
- In the summer nitrites presented a mean value ($\overline{X} \pm s_{\overline{X}}$) de 0,022 ± 0,004995 mg/l and a variability coefficient of 87.94%. Determined values were situated between 0,0 and 0,06 mg/l.

From the total of 60 samples studied no one exceeded the maximum limit admitted for **nitrates and nitrites** which is **max. 10 mg/l milk.**

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RESEARCH REGARDING EVOLUTION OF SOME PHISICO-CHEMICAL PARAMETERS DURING PARMESAN CHEESE RIPENING

Mirela Jimborean[1], D. Ţibulcă[1], Laura Stan[1]

Key words: *ripening, proteolisis, lipolisis, proteins, pysico-chemical indicators*

ABSTRACT

The aim of this research was to determine the physico-chemical properties during manufacturing until the end of ripening of Parmesan hard cheese.

Cheese ripening took place in conditioned warehouses, where air temperature and humidity were monitored. Mean values of temperature were between 8.44 and 9,2°C, and air relative humidity were between 65.57 and 66.67%.

INTRODUCTION

Nutritional importance of cheese derives first of all from their content in proteins with high biological value. The content in these proteins varies widely according to cheese type. In each cheese type the content in proteins varies inversely to fat content.

The porcelain-white, crumbly and bland taste cream cheese from the beginning of the ripening process becomes white to yellowish, elastic, unctuous, taste and aroma specific range. Modifications which take place during ripening develop in a certain order and are based on the milk components: lactose, proteins and fats (Mirela Jimborean, 2006).

In a series of research performed by Stănescu V. şi Laslo C, 1982, it has been shown that the correlation between product freshness and dynamics of total nitrogen, amines and ammonia. In this sense there were established indexes of proteic scindation represented by reports values of:

$$\frac{N-NH_2 \times 100}{N_{total}} \text{ and } \frac{N-NH_3 \times 100}{N_{total}}$$

MATERIALS AND METHODS

Air parameters (temperature and humidity) were monitorized during cheese ripening and their values are presented in Table 1.

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Table 1

Variation of air temperature and relative humidity from ripening space

No.	Sample	Recordings	Mean value	
			Temperature °C	Relative Humidity %
1.	P_1	6	9,17	66,67
2.	P_2	10	9,2	65,9
3.	P_3	14	8,57	65,57
4.	P_4	16	8,44	66,56

Samples were analyzed at certain time periods from manufacturing date and were coded as follows:

P_1 – sample harvested at 48 h from manufacturing;
P_2 – sample harvested at 10 days from manufacturing;
P_3 – sample harvested at 25 days from manufacturing;
P_4 – sample harvested at 50 days from manufacturing;
P_5 – sample harvested at the end of ripening period (after 6 months).

Collection of samples was performed according to STAS 6343/81.
The following analyses were realized on the cheese taken into study:

- Determination of dry matter and humidity (SR EN ISO 5534/2004);
- Determination of sodium chloride (STAS 6354/84);
- Determination of fats (STAS 6344/88) and proteins (STAS 6355/89, Kjeldahl method);
- Evaluation of protein cleavage indexes
- Determination of acidity (SR ISO 1740/2004) and pH.

There is no general method for pH determination in cheese. AOAC recommend the potentiometric evaluation of pH using a suspension of 10 g cheese within 10 ml distilled water. After opinions of many specialists, it is more accurate and representative the pH measured with probe electrodes in the cheese mass. It is therefore avoided to alter the balance between colloidal calcium phosphate and active calcium from solution (Costin at all, 2003).

RESULTS AND DISCUSSIONS

The results obtained regarding the evolution of cheese content in dry matter, humidity, salt, fats reported to dry matter and proteins are presented in Table 2.

The content in dry matter increased during development of experiment (cheese manufacturing – end of ripening) with 8.09%.

The content in humidity recorded a constant decrease during experimental design: it dropped from 41.6% right after obtaining to 33.51% at the end of ripening.

Table 2

Dynamic of physic-chemical parameters analyzed for Parmesan cheese

Sample	**Dry matter, %**	**Humiditaty %**	**NaCl, %**	**Fat, % reported to dry matter**	**Proteins, %**
P_1	58,4	41,6	0,4	40,40	32,71
P_2	58,6	41,4	1	39,82	31,44
P_3	59,6	40,4	1,8	38,08	28,69
P_4	60,2	39,8	2	37,56	28,45
P_5	66,49	33,51	2,30	34,71	26,01
Company standard for end product	min **65**	**33 - 35**	**max 3**	min **36**	min. 21,5

During experimental development it was noticed an increase in salt content with 1.9%, up to 2.3% at the end of ripening.

The content in fats reported to dry matter recorded a continuous decrease during experimental design, namely with 5.69% for the whole period of ripening.

Content in proteins of cheese recorded a continuous decrease with 6.7% during the whole ripening period.

In order to establish indexes for protein cleavage it was determined the amine nitrogen and ammonia. Determination of amine nitrogen based on blocking the amine groups using formic aldehyde, this leading to formation of methylen derived compounds from acidic reaction which can be titrimetric determined. For determination of easily hydrolysable nitrogen the direct titration method with hydrochloric acid was used (Mirela Jimborean, 2009). The results obtained are presented in Table 3.

Table 3

Evolution dynamics of protein cleavage indexes analyzed for hard and semi-hard cheeses

Sample	**Total Nitrogen %**	**Amine nitrogen mg%**	**Ammonia nitrogen mg%**	**Indexes for protein cleavage, %**	
				$\frac{N-NH_2 \times 100}{N_{total}}$	$\frac{N-NH_3 \times 100}{N_{total}}$
P_1	5,13	140	14,96	2,73	0,29
P_2	4,93	168	15,67	3,4	0,32
P_3	4,5	220	17,4	4,89	0,39
P_4	4,46	320	20,88	7,17	0,47
P_5	4,08	140	59,82	3,43	1,47

Among main processes recorded during cheese ripening, proteolysis was the most complex and without doubt the most important.

The content in aminic nitrogen increased in the first step of maturation, and decreased in the following 6 months. Index of proteolytic cleavage ($N\text{-}NH_2/N_{total}$) known similar evolution: from 2.73% at manufacturing time increased in the first 50 days of ripening up to 7.17% and after 6 months of ripening decreased to 3.43%.

The content in ammonium nitrogen increased during ripening, and index of proteolytic cleavage ($N\text{-}NH_3/N_{total}$) had a similar evolution: it increased in the first 50 days of ripening from 0.29% up to 0.47% and in 6 months it went up to 1.47%.

There are taking place a lot of physical, chemical and microbiological processes during cheese processing and ripening which has consequences over the main compounds of cheese. There is enough to mention the 3 fundamental biochemical processes: glicolysis, proteolysis and lipolysis which along with other simultaneous transformations determine the specificity and finesse of aroma and cheese texture (Fox et al., 1996).

Cheese acidity is correlated with pH during technological process as well as in the end product. Cheese with high titrable acidity has a more acidic pH. The value of pH cannot decrease under 3 due to regular organic acids from cheese are too weak to create ionic concentrations $[H^+]$ bigger than 10^{-3} mol/l. In the same time it must be stressed out that the cheese acidity is strongly buffered by aminoacids, peptides and hydrosoluble macropeptides (Feeney et al., 2002). The pH and acidity results obtained for analyzed Parmesan cheese are presented in Figure 1 and 2.

Acidity increased during ripening with 80°T, and pH increased in the ripening step, while at the end of the process it decreased: in the first 25 days of ripening it increased with 0.23 units and then decreased steadily with 0.27 until de the end of ripening.

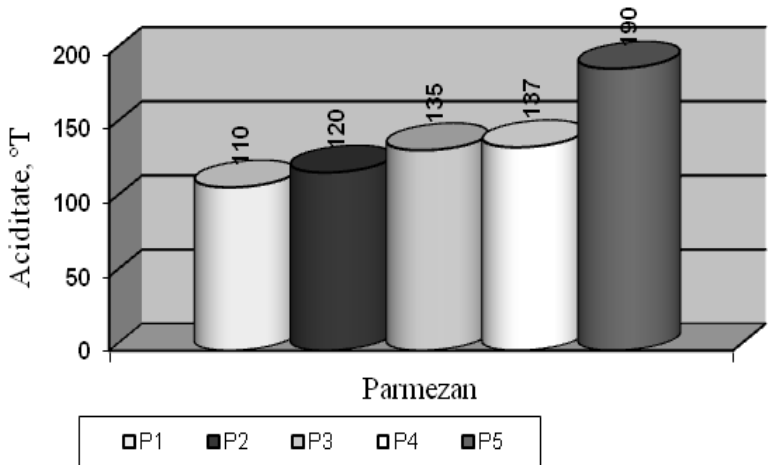

Fig. 1. Dynamics of acidity evolution at analyzed cheese sorts.

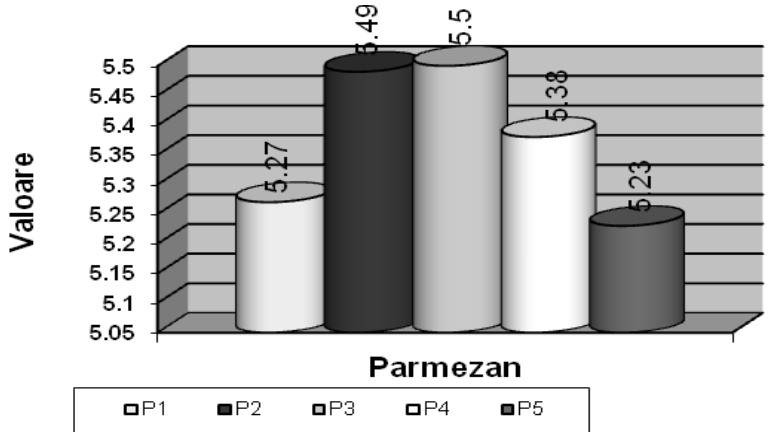

Fig. 2. Dynamics of pH evolution at analyzed cheese sorts.

Numerous biochemical modifications are caused by microbial enzymes present together with their metabolites and dynamics of the transformations depend on water activity, pH, redox potential, mineral content and other few factors like level of saltiness and method used for it, temperature and duration of ripening, nature of secondary microflora etc (Law, 1984).

CONCLUSIONS

The requirements of Company Standard were fulfilled regarding the minimal content in dry matter. The percent reduction of humidity was compensatory to percent increase in dry matter. The humidity decrease of products is influenced by ripening period, climatic conditions from ripening spaces (temperature and relative humidity of air).

The content in salt increased as a result of humidity reduction from the product and concentration in dry matter. There is a correlation between salt content and humidity of products in the sense that humidity reduction leads to saltiness increase in cheese.

The content in fats reported to dry matter dropped under company's standard limits. The fat content reduces during ripening as a result of degradation events (lipolysis) which are the basis of specific taste and aroma. These phenomena are favored by ripening conditions, time period (these transformations are more intense in hard cheese due to longer ripening period).

The protein content reduces during ripening due to their hydrolysis in the presence of proteolitic enzymes into peptones, aminoacids, ammonia which play important role in aroma, taste and smell. Proteolysis speed is faster at the beginning of cheese ripening. The wetter the cheese is, the faster the proteolysis.

At the beginning of ripening there takes place an acidification of cheese cream through lactose transformation into lactic acid. In the final part of ripening the action of lactic microflora continues at low intensity, and specific microflora interferes also. In this phase the taste and aroma of the end product is defined.

pH value increases in the first ripening step, afterwards it decreases as a consequence of lactic acid accumulation in cheese.

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THE IMPORTANCE OF PLANT ASSOCIATION USING SPECIES OF HORTICULTURAL INTEREST AND MAINTAINING BIODIVERSITY IN ORGANIC AGRICULTURE

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Key words: *Organic agriculture, association, biodiversity*

ABSTRACT

The study shows that a number of species cultivated associated (mixed or nearby) positively influence their growth and development processes.

Using existing biodiversity and rational association using species of horticultural interest creates the appropriate environment for obtaining superior products in terms of quality and quantity, also an important link in a sustainable exploitation of existing resources.

INTRODUCTION

In the current era of expansion policies diet, scientists reported that organic farming is the answer for keeping and preserving the balance in nature (Aubert C, 1970).

Not only that organic farming offers a viable alternative to industrial agriculture, but is also an effective antidote against corporate food helping to revive small family farms and their production.

The ideology of the "organic movement" is derived from social campaigns focused on alternative production technologies, "crusades" taken in support of pure and wholesome food and against anti-culture started in the 1960s.

Organic farming as we know it today emerged in the early twentieth century, initiated by Austrian Rudolf Steiner. It was then diversified and enriched by numerous researchers. Now the principles and rules that they are based, are imposed by the International Federation of Organic Agriculture (IFOAM), founded in 1972.

Organic farming practices were formulated based on the initiators beliefs on the way of perceiving nature. Today those views and ideas are no longer applied.

Currently, in order to understand the modern farming methods and principles such as: excluding water soluble inorganic fertilizers, we must consider the original ideas and arguments of the founders, who shared common principles and methods relying on natural processes that does not affect in any way, the health of human and the soils (Sattler F., and colab.1994).

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Although early principles are not applied or have been modified in modern organic agriculture the principles of founders about excluding synthetic compounds, are the most appropriate methods of control the pests in culture.

The possibility of developing integrated plant protection system is determined using vegetable biocenotic relations between components, agrocoenoses and their operating mechanism, harmless biological methods to control pests and diseases by using the economic damage thresholds (Bălăşcuţă N., 1993).

Biological struggle is primarily aimed at reducing the number density of harmful populations below the economic damage threshold. By changing the ratio of all pests and the biological factors that contribute to reducing or limiting pest populations, contribute to establishing a new equilibrium level biocenotic (M. Ferrari M, 1992).

Practice so far has shown that there are a number of pests better adapted to climatic conditions in Romania, which can not be eradicated, so that all pest control measures will seek to maintain them below the economic damage threshold using the method of "dirty field ". Thus, crop pests exist in small numbers, but do not cause economic damage, which is why the treatments against pest are not recommended (Calin Maria, 2005).

Ability to prepare plants to cope with aggression or combat the effect caused by pathogen attack is certainly the main factor that brings or not , the success in organic farming .

This paper aims to demonstrate the productivity and the benefits of organic agriculture when it comes to inter and intraspecific relations between plants and the environment, relating to the results achieved in plant associations from cultures and the direct action on how plants interacts and influences physiological processes.

MATERIAL AND METHODS

Experience was held in the village of Poieni, Buzau County, on an area of 1.92 ha and in Zamfira village, Prahova County, on an area of 0.30 ha was established the sample field.

The lands on that was done the experience did not receive any chemical treatment, organic or synthetic fertilizer product, and they are under the supervision of Ecoinspect, making them suitable for the type of study proposed.

The semicer material used is certified as organic and is provided by The resort of Research and Development in Vegetables from Bacau and DeBolstner, a Dutch society, certified organic and biodynamic, with a tradition of over 20 years in production and marketing of certified seed .

Seedlings used on the farm were obtained in a, partially buried greenhouse, heated by biofuel, plant were produced using certified seeds by an authorized producer.

Treatments, amendments and fertilizers applied on seedlings, were purchased from a company that importing products that are certified internationally and locally.

The experience focused on monitoring the behavior and response of plants to disease and pest species used in the context of association. Of great importance for the culture is that interspecific associations creates diversity in vegetable farm.

For having a higher relevance, were chosen some representative plant associations made in the crops from Poieni village, Buzau county (PvBc) and compared with results obtained in plantations from Zamfira village, Prahova county (ZvPc), where the applied technology on the organic crops doesn't include associations. The land was divided into

equal sole, each field being cultivated with a botanical family.

RESULTS AND DISCUSSIONS

The attack and attack intensity of pests and diseases at main crops.

In case of plant associations, that contain tomatoes, bean and celery, we observe that in beans and celery crops was not reported any damaging attack, and the atack intensity in case of Phytophthora, is lowest in Roma cultivar 18% and 48% in Unibac cultivar.

In crops where the monoculture was practice, the atack of diseases and pests was much stronger, signaling many pathogens including: Phytophthora infestans, Tetranychus urticae, Thrips spp, most damage was caused by Phytophthora infestans in culture some varieties was affected on a rate of 35-75% (table 1).

Table 1

Tomatoes+Beans+Celery

Crop type	The place where the research was conducted	Variety	Surface (m^2)	Average weight fruit/ Root (g)	Diseases and pests reported in culture	Attack intensity %
Tomatoes	PvBc	Marmande	200	153	Phytophthora infestans	25
	ZvPc	Marmande	100	121	Phytophthora infestans Tetranychus urticae Thrips spp.	38
	PvBc	Roma	400	56	Phytophthora infestans	18
	ZvPc	Roma	200	48	Phytophthora infestans Tetranychus urticae Thrips spp.	35
	PvBc	Unibac	300	68	Phytophthora infestans	48
	ZvPc	Unibac	200	63,5	Phytophthora infestans Tetranychus urticae Thrips spp.	75
	PvBc	Zuckertr.	300	44	Phytophthora infestans	20
	ZvPc	Zuckertr.	300	42	Phytophthora infestans	23
Beans	PvBc	Amelia	1000	10,5	There were no reported	
	ZvPc	Amelia	500	11	There were no reported	
Celery	PvBc	Bistrița	800	325	There were no reported	
	ZvPc	Bistrița	500	346	There were no reported	

In case of plant associations, that contain peppers, carrots and basil any pests or diseases wasn't notified in culture, one of the factors that contributed to the lack of crop pests is the fact that the land was first cultivated with bell pepper and red ruffle pepper.

In crops where the monoculture was practice, bell pepper and ruffle pepper was affected in 78% and 69%, the main pests emerged in culture being: Thrips spp and Myzus persicae (tabel 2).

Table 2

Red Bell Pepper+Red Ruffle Pepper+Carrot+Basil

Crop type	The place where the research was conducted	Variety	Surface (m^2)	Average weight fruit/ Root (g)	Diseases and pests reported in culture	Attack intensity %
Red Bell Pepper	PvBc	Jubilanska	500	114	There were no reported	
	ZvPc	Jubilanska	300	102	Thrips spp. Myzus persicae	78%
Red Ruffle Pepper	PvBc	Creola	500	113	There were no reported	
	ZvPc	Creola	300	97,5	Thrips spp. Myzus persicae	69%
Carrot	PvBc	Nantes	300	105	There were no reported	
	ZvPc	Nantes	150	110	There were no reported	
Basil (var.purpurescens)	PvBc	Red ruby	180	-	There were no reported	
	ZvPc	Red ruby	30	-	There were no reported	

In case of plant associations, that contain cucumbers, zucchini and sweet corn no disease or pest that could jeopardize culture wasn't signaled.

In crops where the monoculture was practiced, cucumbers were affected in 80% the main diseases reported being Pseudoperonospora cubensis and Pseudomonas lachrimans (table 3).

In case of plant associations, that contain letuce, radish, onion and parsley, no patogens were reported.

In monoculture situations, the radish and onion were noticed to have had a low intensity attack caused by the Phyllotreta sp., Delia brassicae, Peronospora destructor and Delia antiqua (table 4).

Table 3

Cucumber+Zucchini+Sweet corn+Thyme

Crop type	The place where the research was conducted	Variety	Surface (mp)	Average weight fruit (g))	Diseases and pests reported in culture	Attack intensity%
Cucumber	PvBc	Cornibac	600	86	There were no reported	
	ZvPc	Cornibac	200	75	Pseudoperonospora cubensis Pseudomonas lacrimans	80%
Zucchini	PvBc	Dana	150	350	There were no reported	
	ZvPc	Dana	50	320	There were no reported	
Sweet corn	PvBc	Dulce de Bacău	300	-	There were no reported	
	ZvPc	Dulce de Bacău	100	-	There were no reported	
Thyme	PvBc	Daria	50	-	There were no reported	
	ZvPc	-	-	-	-	

Table4

Letuce+Radish+Onion+Parsley (root)

Crop type	The place where the research was conducted	Variety	Surface (mp)	Average weight fruit/root (g)	Diseases and pests reported in culture	Attack intensity %
Letuce	PvBc	Blonde de Paris	350	235	There were no reported	
	ZvPc	Blonde de Paris	400	295	There were no reported	
Radish	PvBc	Cherry Belle	100	16	Phyllotreta sp. Delia brassicae	5%
	ZvPc	Cherry Belle	50	14	Phyllotreta sp. Delia brassicae	20%
Onion	PvBc	De Stuttgart	350	76	There were no reported	
	ZvPc	De Stuttgart	5000	70	Peronospora destructor Delia antiqua	15%
Parsley (root)	PvBc	Zaharat	200	146	There were no reported	
	ZvPc	Zaharat	200	135	There were no reported	

CONCLUSIONS

Among the advantages of associations and the usage of existing biodiversity in crops we mention:

The association of species with different radicular sistems has allowed the optimum usage of all the soil layers. The association of beans with tomatoes has controled the attacks of some pests and diseases.

The coupling of letuce with radish has controled the fleas attack on cabbage and radish. The association of onion with radish helps protection against the onion's fly.

Sage and thyme keep at a distance the Pseudoperonospora cubensis and Pseudomonas lachrimans pests.

The usage of corn protection screens has enhanced the cucumber and pepper production. The conservation, protection and improvement of existing biodiversity have significantly redused any form of attack on crops, regardless the patogens form.

Plant association and the usage of existing biodiversity have created the neccesary conditions, so that all cultivated legumicol species could develope harmonious.

Considering the facts mentioned above, over the entire duration of supervision, it has been concluded that the mode of cultivation and association of legumicol species have reduced almost completely the attacks of specific pests and diseases.

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LANDSCAPING THE ENVIRONMENTAL RESPONSIBILITY – VILLAGE PARKS AND EU MONEY

Lazăr-Bâra Alexandru Paul[1]

Key words: *ecologic restoration; landscape architecture; landscape concept; rural heritage; swans*

ABSTRACT

Romanian rural area preserves a great deal of the historic steps that European culture is built upon. In this context, the EU far-sighted policy created mechanisms aimed at encouraging this less competitive rural system to approach the once iconic western wealth while minimizing natural and cultural heritage destruction. The National Program for Environmental Quality Enhancement through Urban Green Space Setting Up used to provide funding for these, appealing the landscape architect's qualifications. Among the hundreds of parks built in this context, this study analyses a case in Plosca village – Teleorman. The bureaucratic mechanisms, the financing criteria, the project theme and real-life processes are analyzed based on a project that has been granted the EU money. Conclusions reach domains ranging from public administration, European and national law system, environmental protection and rural development.

INTRODUCTION

Romania's EU entrance in 2007 unleashed important reforms within a country that seemed to have lost its sustainability mechanisms to Balkan *carpe diem* blase. Yet, occident-supported policy determined sometimes paradoxical impacts in local environment.

Landscape synthesizes the cultural achievements of a society on the environmental space-time scale. The natural capital valuation efficiency, as well as the socio-economic systems' sustainability are reflected in landscape character. Also, landscape is the interaction *locus* of the various fields of socio-economic development.

Some of the structural reforms induced by the EU have targeted landscape quality: connecting people and site in an integrative, ecologically sensitive effort of creating sustainable, resilient and feasible environments for present and future society, with the untold objective of preserving in a geographically and biome-related country a reservoir of biodiversity resources that EU core-nations have depleted in order to fuel their socio-economic development. This biodiversity could eventually spread back to the restored habitats that western countries have started to invest in.

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Based on the 6th Environment Action Programme of the European Union Commission (E.U. 2001), covering the 2002-2010 period, the National Program for Environmental Quality Enhancement through Urban Green Space Setting Up (Administration of the Environmental Fund, 2007) was a tool of directing western financial support to Romanian landscape enhancement programs. According to the program's regulations (The Environment Ministry 2009), local administrations were entitled to financial support for green-space developments, including new parks set-up, territorial expansion of the existing green-space assigned urban lots or enhancements of existing parks.

The program targeted human habitats – essential factor of life quality (Ward Thompson et al. 2007, Council of Europe 2000), one of migration's key reasons and one of the structural elements considered by foreign investors – as well as environmental culture (Plumwood 2002, EIONET 2011), as an essential aspect of socio-ecologic systems' sustainability.

Considering the fact that the financing guide of the program only allowed one call per year for any local council, the result was that most of the engaged resources were invested in rural parks development, for the essential reason that in this country there are far more local administrations in rural areas than in urban environment. Therefore, these environmental funds could have been a necessary instrument of an integrated national programme, meant for the rural heritage recovery, conservation and valuation. Without such a programme, this study analyzes the potential contribution to sustainability of a rural landscape architecture design.

MATERIALS AND METHODS

Fig. 1. The territorial context of the park site (yellow)

This paper approaches the case of a financing demand in 2008 regarding a park setup in Plosca village – Teleorman county.

The project site, as proposed by the local council, covers a modest 0.4 ha, some 700 m away from the most isolated building of the village – the local mill; the national road DN6 is also 1000 m away (fig. 1).

The settlements specific included general deep poverty, frequent floods and an isolated water body with swans:

- Population – 6558 on june 1-st 2007 (before the project design), then 6418 on january 1-st 2009; thus, the demographic decline is 1,073 % per year;

- Terrain – The Romanian Plain, o the nordic limit of the Burnaz Plain, not far from the Vedea river everglade – a Danube afluent; the relative altitude – 50 m;

- The site lyes on the shore of a lagoon of Vedea river, half dried up at the moment of the research, by its inferior segment (fig. 1), this being owed to the clogging of its bottom sources; between the ancient channel and the limit of the set-up site there is an earth dike, 2m high by 6m wide on the bottom, in a state of degradation and probably untrusty;

- Geology – the Moesic plateau (Paleozoic and Mezozoic rocks); large deposits of Neozoic and Quaternary loess (Pârvan *et al.,* 2011); shallow underground water resource (3-6m), exploited for the Alexandria municipality use; the area is exposed to floods (caused mainly by Vedea river), the most recent being recorded in 2005);

- Environment – the continental bioregion; the Romanian Plain sylvosteppe ecoregion; excessive temperate-continental climate, with 550-600 l/m^2/year rainfalls, thermal amplitude between 42,9 ^oC (july 5th 1916) and -34,8 ^oC (january 24^{th}-25^{th} 1942), with a multianual medium temperature of 11 ^oC, strong wind exposure, mainly on the east-west direction (Alexandria Local Development Plan, 2007), the *Polygonum aviculare, Lolium perenne, Scherochloa dura* and *Plantago major* anthropic community habitat, with no conservative value, was identified in the national classified list (Doniță *et al.* 2005) based on observations on vegetation, soil and site placement, together with site climate specific; sylvo-steppe pasture vegetation – local species analysis shown the *Poëtum annuae* vegetal association (Sanda *et al.* 2008), indicating a land recovering after a vegetation destruction process;

- Wild-life – the nearby river lagoon biotope hosts a mute swan population (*Cygnus olor*): thus, in may-june 2008 there were identified 12 mature birds;

- Protection regime – in the Romanian Plain there are no biogeographical importance protected areas (Geacu and Dumitrașcu, 2009); the presence of the swans on the river lagoon near the park site has never been recorded before this research, therefore there are no adequate conservation strategies; the park-assigned area has no remarkable environmental features.

Fig. 2: Park site landscape – plane pasture, earth dike and everglde forest front

The original site was part of the common grazing field, scarecely populated with sheep and cattle. During the field research period there were not noticed recent pasture management interventions. This determined the conclusion that the site is poorely integrated in Plosca village socio-economic system.

Local landscape is a degraded vegetation plain, lacking diversity and dynamics, with a low aesthetic value of the natural elements. The most striking front is a forest in the north (fig. 2). The swans and the remains of their biotope were not noticeable from the park site.

The planned development aimed the spending of 150.000 RON from the Environmental Fund (AFM, 2007) on a power line extension, fencing, indigenous species plantations, pavements, park infrastructure and furniture (fig. 3).

The time space allowed for field studies and project drawing up was not enough to consider also a social investigation, this being a frequent romanian landscaping lack, but unexpected in the context of a european supported development programme. Yet, the project theme was designed following a village hall personnel inquiery.

Fig. 3. The park proposal relies on the personnality of Liviu Vasilica

The work's beneficiary required the setup of a leisure site, neat, well organized, close to the former "pond" (the river lagoon) which they intended to restore for fishing and leisure.

RESULTS AND DISCUSSIONS

The setup plan relied on a simple landscape concept: the memory of a son of the village, Liviu Vasilică (b.1959 – d.2005) – physician, ethnologist and folk singer. There was argued that the image of a local personality could support park's social success and should form a good basis for the adequate cultural integration of the new landscape. Luckily, the personality of Liviu Vasilică was a rare opportunity for the landscape architect, because the short time meant for the landscape concept did not allow the option of a professional local culture analysis.

The park development strategy aimed the setup of a socialization friendly environment. A Sunday *hora* and village feast territory was proposed – with perimeter seats, garbage cans, general lighting system, a wide versatile lawn and public toilets. Some of the elements were dimensioned following the minimum requirements to meet the budget restrictions, hoping that the local authority would invest in the further development of the park.

The environmental strategy required woody species adapted to occasional floodings, that would make a habitat for the singing birds. An irrigation system was necessary especially for the perrenials water requirements during the dry summer months, when wind exposure encreases the heat impact on plants. The lawn maintenance also requires irrigation nomatter the planting solution in order to compensate traffic exposure vulnerability. A wind-proof planted curtain included most woody plants. Livestock proofing, with fencing and acces gates, was an essential condition for the park biotope protection.

The proposals (fig. 3) were for a unitary and functional composition, dominated by beneficiary familiar elements, fundamentaly aiming the creation of a square as the core of a future extended leisure area:

- The main access is oriented toward the village mill – it was designed for occasional maintenance machinery access, and it offers the village the image of bald cypress, gate flanking groups;
- Provisions were made for gates and sight corridors toward the most important outside landscape resources – the forest and the swan river-side lagoon; the possible ecological restoration of the water body was also considered;
- The permeable alleys, as minimum resource designs in the most traffic exposed areas, ensure ground-water and soil protection without imposing through their profile park crossing paths;
- A safe, modern playground, was designed for a capacity of 20 children of various ages;
- The park furniture and equipments are vandal proof, ensuring the minimum confort for the park's functioning and for the site's social appropriation;
- The plant selection involved the correlation of their ecological quality with the plantation's symbols, the setup and maintenance cost, as well as their availability on the national suppliers; the dominant trees – *Fraxinus excelsior*, *Quercus robur*, *Tilia cordata*; accent species – *Abies concolor* and *Taxodium distichum*; dominant shrubs (local forest skirt species) – *Spiraea sp, Crataegus monogyna, Viburnum opulus, Syringa vulgaris*; the budget limitations has forbiden perennials, but the park plan allows their further introduction on site;
- A small wayside wooden crucifix, in a discrete setting, is meant to remind the visitors of Liviu Vasilică; its financing was meant exclusively for the local budget.

The swan population protection was considered through measures taken within the park site, through its territorial integration and through recommendations exceeding the contracted landscape study.

As a key element of life quality in the local community (Ward Thompson *et al.*, 2007) the park plays its part in the environmental protection mainly by means of culture.

CONCLUSIONS

The local ground-water resource exploitation could be the cause of the riverside lagoon sources warping, near the park site. Considering the swan habitat relies on them, a further study of the biotope sustainability is recommended that would eventually determine measures for its restoration and protection programme. Forthseeing the lagoon's ecological restoration, this would form the core of a leisure and ecological education periurban area of regional importance, that Plosca village would drive economical advantages from.

The park setup would determine the good site appropriation by local village people, thus contributing to environmental protection (sanity, vandalisation and theft prevention) and to the site social functionality (permanent visitor resource).

Hopefully, the park would enhance social cohesion and environmental culture (EIONET, 2011). This would form the sustainable development basis of the socio-ecologic system.

Park design involves complex environmental studies, which require data collected over years. The design in respect with local culture also require a time reserve for analysis. Without these, most landscape investments are unsustainable.

The National Program for Environmental Quality Enhancement through Urban Green Space Setting Up (AFM, 2007) didn't require the adequate environmental and cultural studies for the financed projects, thus failing to achieve the E.U. Comission objectives (E.U. Comission, 2001), as well as the European Landscape Convention's reccomendations (European Council, 2000). The Plosca park project approaches a biodiversity element accidentally discovered outside the contracted site, but there is the possibility that more, less charismatic species, or subtle associations between the ecosystem's elements might have skipped the preliminary analysis.

This work proves the necessity of national landscape regulations according to the 2002 signed European Landscape Convention (European Council, 2000) and also in harmony with the E.U. environmental policy and programms (E.U. Comission, 2001).

The European regulations on further landscape financing programmes should also impose more rigid standards on the planned development impact on local site – regarding either culture, society or environment. These restrictions should finally disciplinate the firms and the landscape specialists, forcing them to appeal to all the necessary expertise for site understanding.

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AROMATIC TERPENIC POTENTIAL OF SAUVIGNON AND CHARDONNAY GRAPES UNDER NATURAL CONDITIONS FROM BOLINDEŢU – SÂMBUREŞTI HILL

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Key words: *potential, ripening, moment, conditions, accumulation*

ABSTRACT

Research conducted during 2009-2011, at Samburesti regarding the aromatic terpenic potential of Sauvignon and Chardonnay grapes, has shown that, under the natural conditions offered by Samburesti vineyard, the two breeds accumulate enough aromatic substances for being able to offer a pleasant and persistent flavor to wines which makes them unique among other white wines.

Aromatic terpenic potential is conditioned by the climatic offer of the viticultural year and by the breed's genetic dot. Grapes' terpens content at full ripening is not enough for offering grapes that pleasant flavor.

This is realized at 5 to 10 days from full ripening of grapes when total terpens are 20-30% more and the balance of free terpens reaches 78-80%, this being the optimal moment for harvest.

Chardonnay grapes have an aromatic terpenic potential than Sauvignon grapes.

INTRODUCTION

Samburesti vineyard became famous for the unique quality of Cabernet Sauvignon, Merlot, Pinot noir and Fetească neagră red wines.

The observations made over the years show that in this area there could be also obtained semi-aromatic and aromatic white wines of a great quality, especially from Sauvignon and Tămâioasă Românească grapes. There haven't been made systematic studies in this direction, the research concerning the obtaining of this wine category being a priority in Dragasani vineyard, Olt Hill (Butănescu Gh., 1969; Cimpoacă C., 1998; Condei Ghe. Et al., 2008; Cotea D.V. et al., 2000; Genoiu T., Popa A., 2010; Macici M., 2008; Călugăru Larisa, 2012; Popa A., 2008; Popa A., Dicu C., 2010), who took into account the research completed on an international scale in this field (Carbonneau Alain, 2001; Fregoni M., 1997; Morlat R., 2007; Seguin B., 2007; Vila P., 2010). In the last 10 years, together with Sauvignon breed which occupied small areas at Samburesti, there appeared Chardonnay breed hoping to obtain semi-aromatic wines of certain quality.

In 2009-2012 we studied the oenoclimatic offer of Samburesti vineyard for obtaining Sauvignon and Chardonnay wines.

In this paper we refer to the obtained results concerning the aromatic terpenic potential of the two breeds.

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MATERIAL AND METHODS

Research has been conducted on the vine plantations of the Commercial Society "Vitipomicola" Samburesti, realized by planting 6 years ago over 50 ha of vines belonging to Sauvignon and Chardonnay breeds.

Every year it has been followed the evolution of the growth and ripening processes of the grapes and at the full and technological ripening there has been determined through the gas chromatographic method the content in tarpens (free and bound).

RESULTS AND DISCUSSIONS

At full and technological ripening of the grapes there was established the aromatic terpenic potential of Sauvignon and Chardonnay grapes. The results are presented in Table 1 and Table 2.

It is known the fact that flavors start to form in grapes from herbaceous period, when the grapes are green. Upon entering the first fruits, grapes already contain large amounts of flavors as glycosides predecessors (250-500 mg/kg of grapes). Some terpenol such as Citronellol and α terpineol occur in grapes only after the yellow ripening (Popa A., 2008). Flavor (aromatic) content significantly increases together with the grapes' yellow ripening; some flavors appear only during the grapes' ripening, while others, such as linalool, begin to decrease before the grapes reach full ripening. Flavor accumulation rhythm is closely linked to the evolution of sugars accumulation.

For unflavored, but potential flavored breeds (Sauvignon, Chardonnay, Traminer) it has been noticed that free flavors continually increase in grapes until the end of ripening and the ratio of gycosidic predecessors and free flavors remains in favor of volatile terpenol. The quantities of flavors in Sauvignon and Chardonnay grapes are a lot smaller compared to flavored breeds (Tămâioasă, Muscat) (Târdea C. et al., 2010 – quoted by Popa A., Dicu Cornel – 2010).

The results presented in Table 1 express the terpenic potential of grapes at full ripening during 2009-2011 at Samburesti.

Table 1

The general aromatic terpenic potential of Sauvignon and Chardonnay grapes, at full ripening, during 2009 – 2011 – at Samburesti

Breed	Viticultural year	Total terpens µg/Kg grapes	Free terpens FTV		Bound Terpens	
			µg/Kg grapes	%	µg/Kg grapes	%
Sauvignon	2009	1390	1100	79.1	290	20.8
	2010	1410	1145	81.2	265	18.8
	2011	1435	1200	83.6	235	16.4
Chardonnay	2009	1401	1175	83.8	226	16.1
	2010	1420	1201	84.5	219	15.4
	2011	1455	1234	84.8	221	15.2

It can be noticed that this aromatic terpenic potential is conditioned by the climatic offer of the viticultural year and of the soil's genetic dot.

At full ripening of Sauvignon grapes in 2009, the total terpens have a content of 1390 µg/Kg grapes, actually the lowest terpinic content during 2009-2011. Free terpens are

present in quantities of 1100 μg/Kg grapes, representing 79.1% from the total content of terpens. The free ones represent 20.8%, a quantity of grapes of 290 μg/Kg. In 2010, the total terpens content already reaches 1410 μg/Kg grapes, out of which 1145 μg/Kg grapes, free terpens, 81.2% and 265 μg/Kg grapes, bound terpens, meaning 18.8%.

Year 2011 was the best viticultural year from a climatic point of view. As a consequence, the total terpens content of Sauvignon grapes is 1435 μg/Kg grapes, out of which 1200 μg/Kg grapes, meaning 83.6%, are free terpens and 235 μg/Kg grapes (16.4%) are bound terpens.

Chardonnay grapes were having in 2009 at full ripening 1401 μg/Kg grapes, total terpens, out of which 1175 μg/Kg grapes (16.1%) bound terpens. In 2010, with a bigger climatic offer, Chardonnay grapes have at full ripening 1420 μg/Kg grapes, total terpens, out of which 1201 μg/Kg grapes (84.5%) free terpens and 219 μg/Kg grapes (15.2%) bound terpens.

Free terpens balance unlike bound terpens balance, is also dependent on the climatic conditions of the viticultural year and on the genetic dot of the grapes' breed for this biochemical activity.

Table 2

General aromatic, terpenic potential of Sauvignon and Chardonnay grapes at technological ripening in Samburesti vineyard, during 2009 – 2011.

Breed	Viticultural year	Total terpens μg/Kg grapes	Free terpens FTV		Bound terpens	
			μg/Kg grapes	%	μg/Kg grapes	%
Sauvignon	2009	1580	1201	76	379	24.0
	2010	1601	1236	77.2	365	22.8
	2011	1626	1280	78.7	346	27.3
Chardonnay	2009	1600	1242	77.6	358	22.4
	2010	1645	1269	77.1	376	22.9
	2011	1690	1301	77.0	389	23.0

Until the technological ripening the terpens content is amplified. The rankings by viticultural years and grapes' breeds are kept and the ratio between free and bound terpens is changed, remaining most of the free ones. In 2009 Sauvignon grapes accumulate until harvest 1580 μg/Kg grapes, total terpens, out of which free terpens represent 75% (1201 μg/Kg grapes) and 24% bound terpens (379 μg/Kg grapes).

Grapes of the same breed, in 2010, accumulate until harvest 1601 μg/Kg grapes, total terpens, out of which 1236 μg/Kg grapes (77.2%) free terpens and 365 μg/Kg grapes (22.3%) bound terpens.

The greatest contents of terpens are realized by Sauvignon grapes in 2011, meaning 1626 μg/Kg grapes, total terpens, out of which free terpens 1280 μg/Kg grapes (78,7%) and 346 μg/Kg grapes (21.3%) bound terpens.

At technological ripening Chardonnay grapes, in 2009, accumulate 1600 μg/Kg grapes, total terpens, out of which 77.6% (1242 μg/Kg grapes) free terpens and 22.4% (358 μg/Kg grapes) bound terpens. In 2010 there have been recorded at technological ripening of Chardonnay grapes 1645 μg/Kg grapes, total terpens, out of which 77.1% (1269 μg/Kg grapes) free terpens and 22.9% (376 μg/Kg grapes) bound terpens.

Chardonnay grapes accumulate the most terpens in 2011; total terpens content of Chardonnay grapes at technological ripening is at the level of 1690 μg/Kg grapes (the

greatest terpens content during research) out of which 77% (1301 µg/Kg grapes) free terpens and 23% (389 µg/Kg grapes), bound terpens.

CONCLUSIONS

It could be noticed that under the climatic conditions of Samburesti, Sauvignon and Chardonnay grapes accumulate enough flavored substances, the two breeds offering a high aromatic terpenic potential;

Aromatic terpenic potential, at full ripening, depends on the breed's genetic dot and on the climatic conditions of the soil;

The greatest terpens content is realized by grapes at technological ripening (harvest);

Chardonnay grapes have a bigger aromatic terpenic potential than Sauvignon grapes;

Sauvignon and Chardonnay grapes from Bolinteţu – Sâmbureşti Hill, accumulate enough sugars and aromatic substances, but also maintain a high content of organic acids. All these represent certain premises for obtaining semi-aromatic white wines of high quality.

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TECHNOLOGICAL LINKS THROUGH WHICH SAUVIGNON AND CHARDONNAY WINES CAN INCREASE THEIR SENSORIAL PROPERTIES

Luţu Florea, Chirca Ion[1]

Key words: *yeasts, enzymatic preparation, chemical composition, sensorial*

ABSTRACT

The use of selected yeasts for the process of fermentation and of the enzymatic preparation for the maceration facilitate the increase of the pleasant flavor and taste of wine, but we have to be careful not to influence the aromatic typicity of breed and its taste.

Sauvignon and Chardonnay best quality wines, characterized by a great delicacy conferred by a pleasant flavor, discreet and suave, but persistent, were obtained when for the maceration there has been used the pectin-glucosidase enzymatic preparation Lallzyme cuve blanc and for the alcoholic fermentation of the must there have been used selected strain yeasts Lalvin QA23 and Lalvin D47.

INTRODUCTION

By applying the classic technology of vinification in white of Sauvignon and Chardonnay grapes, there were achieved good wines, but not sufficiently balanced and with a less intense flavor, which is absolutely necessary for this category of wines.

Given the information in the literature (Bartowsky E.J., et al., 2004; Bertrand A., Beloqui M.A.A.; 1996; Cordero Otero R., 2003; Dumont Ann, 2005; Grigorică L.G., Nămoloşanu I., 2008; Grigorică L.G. et al., 2010) we developed a research program through which we could increase sensorial properties of Sauvignon and Chardonnay wines from Bolindetu-Samburesti by intervening in some links of the white vinification technology by using selected yeasts in some enzymatic preparation.

MATERIAL AND METHODS

The experiments were done on Sauvignon and Chardonnay must within Samburesti complex of oenology belonging to S.C. Viti-pomicolă – Samburesti.

Experimental varieties are shown in Table 1. Physico-chemical analyses of wines were made using the methods adopted by the O.I.V., for the sensorial analysis there was used the compensation system with points from 1 to 100 of qualitative sensorial aspects.

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Sauvignon and Chardonnay grapes used for vinification in the three years (2009-2011) had, at harvest, about 230g/l sugars and a good total acidity (6.6-7.1g/l tartaric acid). All varieties had in common the addition of potassium metabisulfite (55 mgSO2/l) applied on the must at maceration. After the alcoholic fermentation, there were provided 60 mmSO2/l of wines. The three selected yeast sources (Lalvin QA23; Anchor vin 7 and Lalvin D47), as well as the enzymatic preparation, were purchased from S.R.L. Bevitech Bucharest.

RESULTS AND DISCUSSIONS

Table 2 summarizes the levels of the main wine components and the sensorial impression they have shown.

The analysis of these results shows that in all cases the fermentation of sugars was almost complete, except for the first variant, followed by the second one, when the alcoholic fermentation of the must was made the second one, when the alcoholic fermentation of the must was made with yeast present in the spontaneous flora.

In the cases where selected yeasts were used for fermentation and pectin-glucosidase enzyme preparation for maceration, volatile acid content of wines is smaller than in the cases where must fermentation was carried out with yeasts present in the spontaneous flora. We also record a slight increase of the unreducible extract (with 1-2g/l)

In organoleptic assessment, Sauvignon and Chardonnay wines have received many compensation points (88-92 points) when selected yeasts and enzyme preparation were used and especially when Lallzyme cuve blanc (2g/hl) was used for enzymatic maceration and Lalvin D47 yeast strain (90-93 points) was used for fermentation.

Detailed results of the sensorial analysis are presented in Table 3 and Table 4.

Sensorial evaluation of the main organoleptic characteristics (aspect, clarity, smell, taste, harmony) of Sauvignon wines, shows the fact that wine quality level firstly depends on the characteristics of grapes – raw material and then vinification technology.

As a result of all three years of study, grapes have different composition characteristics and that is why wines had a different quality level, regardless of the vinification technology. This can be noticed especially in variants 1 and 2, where must fermentation was left to the spontaneous flora. This aspect is less obvious for other variants. However, the best wine years was 2011, followed by 2010 and then 2009.

What should be stressed is that by using selected yeasts and enzymatic preparation, Sauvignon wines impress with a quality smell and especially with harmony. The average score for tasting puts on the leading places the wines obtained by applying vinification variants 5 and 6 when maceration is realized by using selected yeast strains Lalvin QA23 and Lalvin D47.

Wines obtained through the 2 variants are characterized by a great delicacy conferred by a very pleasant flavor, discreet and suave, but persistent, reminding of wild flowers and vine pollen or elderflower flavor, they have a perfectly balanced taste and a perfect harmony.

The grapes influence in the same way the quality of Chardonnay wines. Selected yeasts Lalvin QA23 and Lalvin D47 and the enzymatic preparation Lallzyme cuve blanc provide Chardonnay wines a great delicacy, a harmonious composition and a special softness. They are suave, gentle, but at the same time rich with a persistent flavor

reminding of that of mown hay. Wines delight through their perfectly balanced quality having a happy harmony.

CONCLUSIONS

If selected yeasts with different qualities are used for must fermentation, wines are not very different from a compositional point of view; they are similar in terms of aspect and color, but different in terms of flavor and harmony;

Sauvignon and Chardonnay wines obtained by using the selected yeasts Lalvin QA23 and Lalvin D47 and by maceration in the presence of pectin-glucosidase enzymatic preparation Lallzyme cuve blanc, facilitate the multiplication of the pleasant flavor and taste of wine. Sauvignon wines have a great delicacy conferred by a very pleasant flavor, discreet and suave, but persistent. Chardonnay wines have a harmonious composition, they are fine and soft. They are also suave, gentle as well as rich with a persistent flavor;

The use of selected yeasts, as well as the enzymatic preparation bring a considerable quality increase (flavor, taste), but we have to be careful not to influence the aromatic typicity of breed and its taste.

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Table 1

Technological variants used for Sauvignon and Chardonnay grapes vinification at Sâmbureşti (2009-2011)

Variant number	Technological operations of experimented variants
1.	White vinification without maceration, without settling, spontaneous fermentation at fermentation temperature of 17-18^{0}C
2.	White vinification with maceration 6 hours, gravitational settling , spontaneous fermentation, at the temperature of 17-18^{0}C
3.	White vinification with enzymatic maceration (Lallzime cuve blanc, 2g/hl) 6 hours, gravitational settling, spontaneous fermentation at 17-18^{0}C
4.	White vinification with enzymatic maceration (Lallzyme cuve blanc, 2g/hl) 6 hours, gravitational settling, fermentation with selected yeast Auchor Vin 7 (20g/hl), at the temperature of 17-18^{0}C
5.	White vinification with enzymatic maceration (Lallzyme cuve blanc, 2g/hl) 6 hours, gravitational settling, fermentation with selected yeast Lalvin QA23 (20g/hl), at the temperature of 17-18^{0}C
6.	White vinification with enzymatic maceration (Lallzyme cuve blanc, 2g/hl) 6 hours, gravitational settling, fermentation with selected yeast Lalvin D47 (20g/hl), at the temperature of 17-18^{0}C

Note: All variants had in common the addition of potassium metabisulfite (55mg, SO_2/l) applied on the must at maceration. After the alcoholic fermentation, there were provided 60mg SO_2/l.

Table 2

Physico-chemical parameters at Sauvignon and Chardonnay wines obtained after applying the technological variants (average data 2010-2011)

Variant	Breed	Determination					Organoleptic assessment
		Alcohol (Vol%)	Sugars g/l	Total acidity g/l (Tartaric acid)	Volatile acidity g/l	Unreducible extract g/l	1-100 compensation points
1	Sauvignon	13,4	6	6,4	0,49	22	80
	Chardonnay	13,3	5	6,5	0,48	23	79
2	Sauvignon	13,4	4	6,5	0,39	22,5	84
	Chardonnay	13,3	5	6,6	0,40	23,2	83
3	Sauvignon	13,5	3	6,4	0,34	23	88
	Chardonnay	13,6	4	6,4	0,33	24	88
4	Sauvignon	13,6	2	6,4	0,30	24	89
	Chardonnay	13,5	3	6,6	0,29	23	89
5	Sauvignon	13,5	3	6,7	0,29	24	90
	Chardonnay	13,5	4	6,6	0,30	23	91
6	Sauvignon	13,6	4	6,5	0,26	23	92
	Chardonnay	13,5	4	6,9	0,27	24	93

Table 3

Sensorial evaluation of the main organoleptic characteristics of Sauvignon wines obtained by applying different technological variants (2009-2011)

Variant	Breed	Organoleptic parameters					Total points
		Aspect	Colour - clarity	Smell	Taste	Harmony	
1	2009	8	5	22	34	9	78
	2010	9	5	22	35	9	80
	2011	10	4	23	36	9	82
	Average	9	4,7	22,3	35	9	80
2	2009	9	4	23	36	8	80
	2010	9	5	2,6	37	9	86
	2011	8	5	27	37	9	86
	Average	8,7	4,7	25,3	3,7	8,7	84
3	2009	10	5	26	33	10	84
	2010	10	5	27	37	10	89
	2011	9	5	30	37	10	91
	Average	9,7	5	27,7	35,7	10	88
4	2009	8	5	28	36	9	86
	2010	9	5	29	37	10	90
	2011	10	5	30	37	10	92
	Average	9	5	29	36,7	9,7	89
5	2009	9	4	28	38	10	87
	2010	8	5	30	37	10	90
	2011	9	5	33	38	8	93
	Average	8,7	4,7	30,3	37,7	9,3	90
6	2009	9	5	29	37	10	92
	2010	10	5	30	37	10	92
	2011	10	5	32	37	10	94
	Average	9,7	5	30,3	37	10	92

Table 4

Sensorial evaluation of the main organoleptic characteristics of Chardonnay wines obtained by applying different technological variants

Variant	Breed	Organoleptic parameters					Total points
		Aspect	Color - clarity	Smell	Taste	Harmony	
1	2009	9	4	22	34	9	78
	2010	9	4	23	34	9	79
	2011	10	4	22	35	9	80
	Average	9,3	4	21,6	35,3	9	79
2	2009	9	3	24	34	9	79
	2010	10	4	25	35	10	84
	2011	10	4	25	37	10	86
	Average	9,6	3,6	24,6	35,3	9,7	83
3	2009	9	5	26	36	10	86
	2010	9	5	28	36	10	88
	2011	9	5	29	37	10	90
	Average	9	5	27,7	36,3	10	88
4	2009	9	4	28	37	9	87
	2010	9	5	29	36	10	89
	2011	9	5	30	37	10	91
	Average	9	4,7	29	37	10	89
5	2009	8	5	30	37	10	90
	2010	9	5	31	37	8	90
	2011	9	5	33	38	8	93
	Average	8,7	5	31,3	37,3	9,3	91
6	2009	9	5	29	37	10	90
	2010	9	5	31	39	11	95
	2011	9	5	32	39	11	96
	Average	9	5	30,7	37,7	10,7	93

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EVALUATION OF SOME APPLE PROGENIES REGARDING GENETIC RESISTANCE TO *Venturia inaequalis*

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Key words*:* *cultivar, rootstock, apple, resistance, race*

ABSTRACT

Improving genetic resistance is one of the most effective protection against scab on apple. Crosby et al. (1992) describes two types of apple resistance to scab, one qualitative (monogenic) typical of the genus Malus species and other quantitative (polygenic).While improving genetic resistance to Venturia inaequalis the new races were discovered Parisi et al. , 1993. In march 2011 was started a breeding program in University of Agricultural Sciences and Veterinary Medicine Bucharest , that aims to induce a complete or partial resistance to Venturia inaequalis using new sources of resistance like old local varieties.This paper illustrates the identify the first local varieties, potential resistant's and their involvement in the pollination of apple varieties to obtained an apple with high organoleptic and market, getting F1 progenies, artificial infection of young plants, their phenotyping and testing by molecular markers for highlighting the Vf gene (gene involved in the resistance to scab).

INTRODUCTION

Apple scab (*Venturia inaqualis*) is one of the most dangerous diseases in apple orchards. Under favorable conditions the unkempt orchards, crop losses can exceed 30%. Fruits attacked by this fungus are refused export - which is a good reason why the apple orchards are numerous treatments each year.

Without applying a set of measures to protect against damaging agents, quantitative and qualitative production losses can vary between 28.8 to 50% to total compromise of fruit or seedlings (Amzăr, 1999, Davidescu, 1999, Thomas , 2003).

Phytoprotection in fruit growing importance of technology derives from the amount of direct expenses to be made to protect trees against damaging agents.

Phytoprotections in orchards are an important component of fruit tree production systems sustainable, environmentally and economically integrated.

In recent decades positive changes have occurred regarding phytoprotection on fruit crops due to the emergence of new active ingredients - synthetic or natural - more efficient, less toxic, biotechnical means of easy to use, and technological progress made in

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the means of forecasting and warning, and machinery for the application of phytosanitary treatments.

In recent years is increased the general interest in modern technology, enabling the production of quality fruit beneficial for the consumer, and reduced environmental impact (Achbani et all., 2006; Brisset et al., 2005, Cohen, 2002; Gansca et all., 2007; Maon et all, 2002; Ugalote, 2004, Teodorescu and colab.1993, 1995, 2007).

In the Integrated phytoprotection that work to optimize the number of treatments against agents damage, prevention and reduction of residues to various plant protection products, reduction of waste in integrated fruit production, maintaining and enhancing biodiversity in agro fruit.

In the case of modern technology of phytoprotection for orchards is necessary to be harmoniously combined assortment, technology of culture and the control methods physical, mechanical, chemical, biological and biotechnical, and will apply especially preventively to maintain a lower level of diseases, pests and weeds than that at which economic damage occurs (Amzăr., 2002, Beers et al., 1993; Şerboiu et al., 2001).

The priority objectives are to reduce the number of treatments with chemicals and replace them with bioinsecticides, biofungicide of "RCI" insect growth regulators, etc.. These products have the advantage of very low toxicity to humans and the environment, some are even safe.

Use of plant extracts for disease control is part of European trends sustainable agriculture, thus achieving industrial synthesis of the complementary products which, although high efficacy in combat, the risk of side effects by accumulating in plants and there by transmitting their toxicity to animals and therefore humans (Murariu others, 2005).

One way to control plant diseases is to induce and increase their defense mechanisms of plants (fitoalexine and elicitive), which would avoid the use of toxic components of plants (Brisset et all., 2005, Kenji Matsui, 2006; Owens, 2005; Shelz et all, 2006).

One of the objectives of the research of the new compounds is to found the new structures to looked natural plant sources. Many researchers, particularly in countries with greater biodiversity contributed to the detection of new compounds derived from medicinal plants for phytosanitarity plant use (Alexandru et al., 1988; Brisset et al., 2005, Peacock et al., 1988; Shelz et all., 2006, Stephen A., et al., 2005).

In phytosanitarity health programs that include vegetable active substances, knowledge of plant resistance to pathogen attack is absolutely necessary to obtain it contributes both passive and active mechanisms (Murariu, 2005, Gouramanis, 1999).

Chemically passive resistance is ensured by the presence in plant extracts an important and various antimicrobial substances that inhibit the growth of bacteria and fungi parasites or destroyed it. In this category are: phenols, flavones, terpenes, saponins, etc.. These preparations do not exclude pesticides, but limits their role and place in conventional technology to combat disease.

Integrated control of apple pests and diseases aims to reduce the number of chemical treatments, use mildly toxic pesticides and selective use of biological methods (use of byproducts, launches zoofagi) and biotechnical (capture of males), the warning treatments and only if is exceeds the economic damage limits.

An integrated control indicative system of pests and diseases of apple orchard involves a combination of:

- use of healthy plant material to plantations;- grow the resistant varieties to diseases, pests, drought, frost;

- applying appropriate agrotechnical;
- application of 1-2 treatments winter before flowering and 4-5 after flowering treatments;
- use of pesticides during the growing season and selective systemic useful for protecting wildlife (natural and released);
- use of different biological methods;
- use biotechnical methods;
- use of specific control treatments (forecasting and warning, the economic damage limited
- the specificity of crop and pest specific (nurseries, young plantations);
- choice of appropriate machinery for carrying out phytosanitary treatments;
- integrating the control of diseases, pests and weeds in crop technology.

The use of healthy materials in plantations of apple is an essential element in ensuring a well concluded and healthy plantings. So, check rigorously seed to be free of ponte and pathogens (viruses, mycoplasmas and bacteria).

Creating resistant varieties (to disease, pests, drought and frost) and placing them into production is one of the important links of the concept of integrated control.

MATERIALS AND METHODS

Plant material evaluated were included, progenies of six experimental crosses that were the old apple varieties like 'Prescurate, 'Gurguiate, 'Poinic' resulted fall in spring 2011. The resistance varieties 'Generos',' Florina', 'Bistritean' were donors of dominant allele *Vf* while 'Idared', 'Poinic','Romanesti 1', 'Romanesti 2' were donors of quantitatively based resistance. (Table 1)

Infection tests in greenhouse conditions according to Chevalier *et al.* (1991) were used for selection of resistant plants. Mixtures of pathogen isolates were used for plantlet inoculation. Seedlings were sprayed with a conidial suspension of *Venturia inaequalis* CKE. Seedlings were incubated for 48 hours at 18°C and 100% relative humidity. Disease symptoms were evaluated macroscopically after 21 days of cultivation in a greenhouse. Seedlings were divided into 5 classes. Plants in class 0 were without symptoms of infection. Plants of class 4 had lesions with full sporulation. For PCR analyses only pre-selected seedlings without symptoms of apple scab on the leaves – class 0 to 3 were used .(fig. 1 and 2).

DNA was isolated from leaves in all evaluated genotypes, parents and progenies. The genotypes were grown in greenhouse conditions. Leaves were immediately fixed in liquid nitrogen and used for extraction. The CTAB method according to Saghai-Maroof (1984) was used.

The primers according to Tartarini *et al.* (1999) were used for detection of dominant allele *Vf*. For multi PCR two pairs of primers in single reaction were used. Primers A (5'TGAAAGAGAGATCCAGAAAGTG3') and

B (5'CATCCCTCCACAAATGCC3')

amplified a co-dominant marker. 466 bp fragment characterized dominant allele *Vf* and 724 bp fragment characterized recessive allele *vf*.

The pair of primers C (5'CGTAGAACGGAATTTGACAGTG3') and

D (5'GACAAAGGGCTTAAG TGCTCC 3')

amplified a marker of dominant allele *Vf* (526 bp fragment) in the same reaction.


Fig 1 and 2. Symptoms of apple scab on the leaves - class 0 to 3 were used.

The composition of 25 µl multi-PCR was: 25 ng DNA/25 µl, 0.2 µM primer A, 0.2 µM primer B, 0.1 µM primer C, 0.1 µM primer D, 1.5 mM MgCl2, 0.2 mM dNTP, 0.8 U Taq/25 µl. The program of amplification was: 1× (94°C – 150 s, 60°C – 60 s, 72°C – 120 s), 35× (94°C – 30 s, 60°C – 60 s, 72°C – 120 s) and 1× (72°C – 600 s). All samples were also evaluated by PCR test using the primers of pair E (5'GTAAAGCAAGCACTTCAACG') and F (5'GTAAAATAGATGTGTGGGTAGC') according to Gianfranceschi *et al.* (1996). This pair was able to amplify the 400 bp marker of dominant *Vf* allele. The composition of 25 µl reaction was: 10 ng DNA/25 µl, 0.3 µM primer E, 0.3 µM primer F, 2.5 mM MgCl2, 0.1 mM dNTP, 0.7 U Taq/25 µl. Touchdown amplification steps according to Hemmat *et al.* (1998) were applied to amplify the PCR marker. The program of amplification was: 1× (94°C – 120 s, 69°C – 120 s, 72°C – 120 s), 5× (94°C – 60 s, 68°C – 120 s, 72°C – 120 s), 5× (94°C – 60 s, 67°C – 120 s, 72°C – 120 s), 5× (94°C – 60 s, 66°C – 120 s, 72°C – 120 s),5× (94°C – 60 s, 65°C – 120 s, 72°C – 120 s), 5× (94°C – 60 s, 64°C – 120 s, 72°C – 120 s), 5× (94°C – 60 s, 63°C – 120 s, 72°C – 120 s), 5× (94°C – 60 s, 62°C – 120 s, 72°C – 120 s) and 1× (72°C – 480 s).

Amplified DNA fragments were visualized by ethidium bromide (Sambrook *et al.* 1989).

RESULTS AND DISCUSSIONS

After pollination (fig. 3 and 4) the highest percentage of binding of the fruit is combination II C2 ('Idared' x 'Romanesti 2') 49% followed by combinations of A1 ('Idared' x 'Bistrita') 41.45% and B1 ('Romanian1'x 'Florina ') 41.90% (table 1).

Genotypes ♀ x ♂

'Idared' x 'Bistritean' (A1);'Florina' x 'Prescurate (B1); 'Generos' x 'Prescurate (B2)
'Idared' x 'Prescurate' (B3); 'Idared' x' Gurguiate'(C1) ;'Generos' x 'Gurguiate' (C2).

The highest ratio was recorded in combination 'Florina' x 'Prescurate', followed by 'Idared' x 'Gurguiate'.

Table 1

The breeding progenies characterization

Parents ♀ x ♂	**Donor of Vf allele**	**No. of analysed seedlings**	**Frequency of resist seedlings classes 0-3 (%)**
Idared x Bistritean (A1)	'Bistritean'	234	41,45
Florina x Prescurate (B1)	'Florina'	370	41,9
Generos x Prescurate (B2)	'Generos'	220	31,3
Idared x Prescurate (B3)	'Prescurate'	162	32,7
Idared x Gurguiate (C1)	'Gurguiate'	165	49
Generos x Gurguiate (C2)	'Generos'	213	28,6

Fig. 4. Aspects of pollinations.

The resistant progenitor ('Generos'and 'Florina') was able to transmit the resistance to descendants, in agreement with previous results observed by other authors (among the 213 descendants evaluated, 82 (38.5%) were susceptible to apple scab and 131 (61.5%) were resistant. Within families, the resistant: susceptible ratios were 83:17 in controlled pollination of 'Generos', 28:72 in 'Prescurate ' x 'Idared' and 62:38 in 'Idared' x 'Gurguiate'. A priori, these proportions do not fit to any of the different hypotheses described to date regarding the genetic control of scab resistance in apple. (Durel *et al.* 2003).

Results of macroscopic evaluation of resistant and susceptible plants are presented in Table For highlighting *Vf* allele the PCR were performed using primers sites (5'TGAAAGAGAGATCCAGAAAGTG3 ') and (5'CATCCCTCCACAAATGCC3'), which amplify a fragment of 466 bp allele Vf characteristic for resistant varieties Florina, Bistrita and a fragment 724 bp peak characterizing recessive allele for varieties Prescurate and Gurguiate.

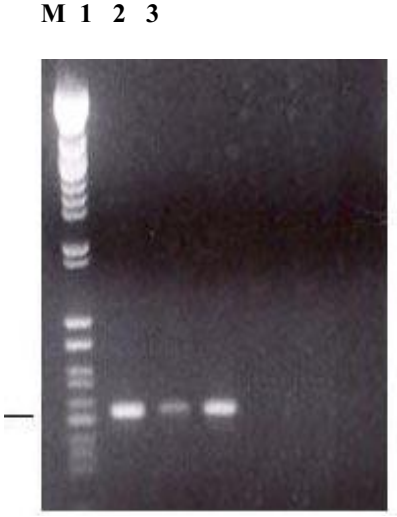

Fig 1. Dominant PCR marker (400pb) in detection of dominant allele *Vf*.

Marker DNA/*Eco 471(Ava II)*:

M= Marker

1= 'Florina'

2= 'Bistritean'

3= 'Generos'

Fig.1- Agarose gel electrophoresis to detect gene *Vf*

CONCLUSIONS

Using the improvement of old Romanian varieties of apple, well adapted to the climate of Romania can be an interesting premise on the induction of natural genetic resistance to *Venturia inaequalis.*

Artificial infection of these varieties and hybrids derived from descendants of these can provide some information about the plant's defense mechanism against pathogen attack (Jha *et al.* 2009). The crosses 'Idared'x 'Romanesti 1' and 'Idared' x' Romanesti 2' are the products of crossing of two heterozygotes from the genetic aspect. In both cases dominant homozygotes were also detected by PCR test.The success of PCR detection of *Vf* was depended on the quality and quantity of isolated DNA. Gardiner *et al.* (1995), Yang and Korban (1996) and Guilford *et al.* (1997) applied a similar method of isolation in CTAB buffer. They considered this method as suitable for PCR analyses in the genus *Malus*. Gianfranceschi et al. (1996) used the method according to Dellaporta's protocol for DNA isolation in apple varieties (Dellaporta *et al.* 1983). Gianfranceschi *et al.* (1996) considered this DNA isolation method as suitable for PCR detection of *Vf* gene in apples.

The frequency of recessive homozygotes in both crosses was 25 and 35%, respectively. This fact confirms that single mass infection tests under certain circumstances are not able to disclose all undesirable genotypes. It could be caused by an inexpressive reaction of the host to the presence of pathogen, failure of inoculation.

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COMPARATIVE STUDY ON THE EFFECT OF SOME HEIGHT CONTROL METHODS FOR *PLECTRANTHUS COLEOIDES* BENTH. PLANTS

Manda Manuela[1], Nicu Carmen[1]

Key words: *Plectranthus coleoides; plant growth regulator; pot size; vegetative characters*

ABSTRACT

The plants reaction to the growth retardants application or to cultivation methods for height reduction has been studied for many species, and the often contradictory results highlight the plant reaction differences between species and even between the varieties within a species.

This study aims to control the height and to stimulate the ramification of the Plectranthus coleoides Benth. 'Marginatus' plants by 2 methods: the chemical method - application of growth retardants (Cycocel); cultivation method (non-chemical) – keeping the plants in small pots in order to diversify the possibilities of use.

The application of Cycocel 0.3% treatments for the Plectranthus coleoides stimulated the vegetative growth, obtaining compact plants by increasing the average number of shoots/plant and improving the decorative appearance of plants by a more intense coloration of the leaves. By reducing the substrate volume, after 7 months of the experiments placement, the plants maintained their height as it was initially and in this case Plectranthus coleoides can be recommended to be used in container gardens, mini gardens or even in vertical gardens, in different contrasting combinations.

INTRODUCTION

For obtaining plants in pots, the height control is often necessary to obtain the desired plant height and shape (Chen & Meister 2006; Milandri et al 2008). The rapid rate of growth, loss of its compact appearance and the lack/delay of flowering prevent sometimes the commercialization of ornamental plants (Banko & Landon 2005).

Although the growth retardands are widely used to control the plants height, there are cultivation methods (non-chemical) increasingly used in floriculture, such as: water stress, nutritional stress, keeping plants in small pots, the temperature difference between day and night (DIF), the quality and intensity of light (Nicu et al. 2009, Manda et al. 2008, NeSmith et al. 1998, Bailey 1991, Larcher et al. 2011, North et al. 2010, Latimer 1998).

Plectranthus L. Her. is a large genus of the *Lamiaceae* family widely distributed in tropical regions of Africa, Asia and Australia (Codd 1985, Ascensao et al. 1999, Abdel-Mogib et al. 2002, Anton 2006). Sixty-two species of *Plectranthus* are reported to be of economic and medicinal interest and some are grown as ornamental plants (Ascensao et al.

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1999, Lukhoba et al. 2006, Khorshid et al. 2010). As an indoor ornamental plant, it is generally cultivated in hanging pots, having long stems up to 1 m.

This study aims to control the height and to stimulate the ramification of the *P. coleoides* Benth. 'Marginatus' plants by 2 methods: the chemical method - application of growth retardants (Cycocel); cultivation method (non-chemical) - keeping the plants in small pots in order to diversify the possibilities of use.

MATERIAL AND METHODS

The initial biological material consisted of *P. coleoides* Benth. 'Marginatus' cuttings, obtained from the Floriculture discipline collection of the Faculty of Agriculture and Horticulture in Craiova. It is a crowling herbaceous plant with purple succulent stems, which are right at the begining, and then start bending and hanging, with persistent leaves. The inflorescences have large, rounded bracts and insignificant lilac tubular flowers. The '*Marginatus*' variety - has cordiforme, pubescent, 5-7 cm long, light green leaves with a wide white margin and it is the variety most commonly grown in apartments in suspended pots, having long stems up to 1 m.

The research was conducted between 2011 and 2012 in the greenhouse of the Floriculture discipline of the Faculty of Agriculture and Horticulture, a temperate greenhouse, with average temperatures ranging between 18 and 20°C.

There were used shoots cuttings harvested in April 2011 from mature plants that were treated with Radistim 1, a powdery biostimulator specific to herbaceous plants, and they were planted in a mixture of: peat and perlite in a ratio of 1:2, and the substrate temperature ranged between 19 and 20°C. For V1, V2, V3 the rooted cuttings were planted in a 2:1 mixture of peat + perlite in pots of different sizes (6, 8, 10 cm diameter). At V4 the rooted cuttings were planted in pots of 10 cm (a size recommended in the literature) and 3 treatments at the soil level with Cycocel 0.3% (3000 ppm) were applied, starting on 26.05.2011, at an interval of two weeks. The following experimental variants resulted: V1 - diameter 6cm/water, V2 - diameter 8 cm/water; V3 Mt - diameter 10cm/ water; V4 – diameter 10cm/Cycocel. For simplicity variants were noted as: V1 - 6cm/w, V2 – 8 cm/w; V3 Mt - 10cm/w; V4 – 10cm/Cycocel.

For 7 months after the experiment placement there was observed the rate of vegetative growth of *P. coleoides* plants, in terms of the cultivation conditions.

The observations and measurements were underlining: the average plant height, the average number of shoots/plant, the average length of shoots, the average leaf size.

RESULTS AND DISCUSSIONS

By reducing the substrate volume, there is found that after 7 months from the experiments placement, the plants maintained their height at values close to the original height, in comparison with the variant treated with Cycocel (V4-18.4 cm), where after 2 months from the first treatment the plant height tripled, and after four months it reached 18.4 cm, in comparison to the original height of 4.3 cm (graph.1).

The plants grown in a small substrate volume (V1 - 6cm/w and V2- 8 cm/w) had a slow growth since their movement in pots until the last measurement, the values range between 4.5 - 5.8 cm (V1) and 4.35 – 7.2 (V2).

At V1-6 cm/w and V2-8 cm/w, the plants kept a dwarf stature (5.8 cm, respectivelly 7.2 cm) compared with V3-10 cm/w, where they recorded higher values, of 14.3 cm at the end of the experimental period.

Noteworthy is the fact that by reducing the substrate volume, after 7 months from the experiments placement, the plants maintained their height at values closed to the original ones, and in this respect *P. coleoides* Benth. may be included in the list of "mini plants".

In graph. 2 there is observed that the treatment with Cycocel 0.3% stimulated the shooting (V4-28 shoots), obtaining plants with compact appearance in comparison with the untreated variants, where the average number of shoots/plant ranged between 3 and 18 shoots at the end of the experimental period.

The lowest values of the average number of shoots/plant were recorded at V1-6cm/water- three shoots, followed closely by V2-8cm/w - 10 shoots. At V1-6m/w there is observed even a decrease of values from the first to the last measurement (from 7 to 4 shoots). At V3 Mt -10 cm/water the values increased significantly compared with the other untreated variants, up to 18 shoots after 7 months of the experiment placement.

The treatments with Cycocel 0.3% (V4), caused a dramatic increase in the average number of shoots, giving the plant a compact appearance, this parameter being negatively influenced by the reduction of the substrate volume (V1, V2).

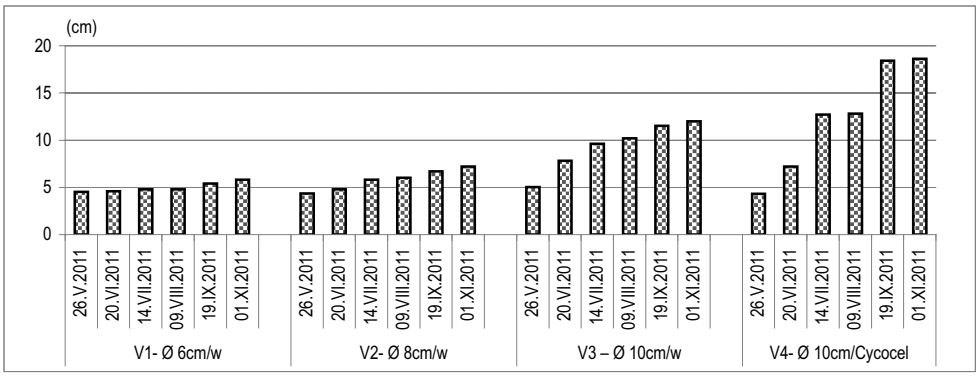
Regarding the average length of shoots, there were obtained the biggest differences between the variant treated with Cycocel (V4-15.4 cm) and the untreated ones (V1, V2, V3) with values ranging from 1.8 to 3.1 cm, from which it appears that the effect of Cycocel treatments was to stimulate the plant growth and not to reduce or maintain the height (graph. 3).

As in the case of the average plant height, the values of this parameter were close to the variants grown in a reduced substrate volume (V1-6cm/w and V2-8cm/w), ranging between 0.4 and 1.8 cm (V1) and between 1 and 2.1 cm (V2) compared to V3 Mt -10 cm/water where there were recorded higher values (4.1 cm) after 4 months from the experiments placement.

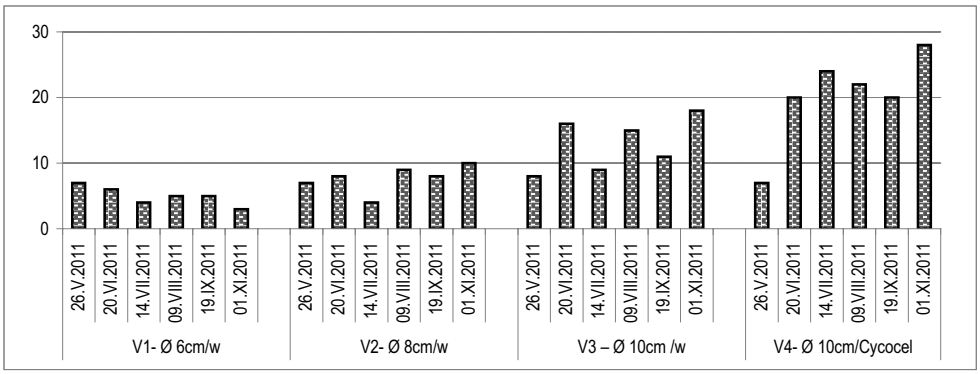
The mode of action of Cycocel in its inhibition of gibberellins is not totally resolved. While the majority of the studies emphasise the ability of Cycocel to retard plant growth only a few experimental works have shown that small doses of Cycocel solution can significantly stimulate or increase growth (Reid & Crozier 1970; Ojeda & Trione 1994 apud Al-Maskari 1998).

The average size of leaves had maximum values also at the plants treated with Cycocel (V4 - 2.5 cm long and 2.3 cm wide) and the lowest values at V1-6cm/water (1,5 cm long, respectiv 1,2 cm wide) followed by V2-8cm/w and V3-10cm/w with very small differences (graph. 4).

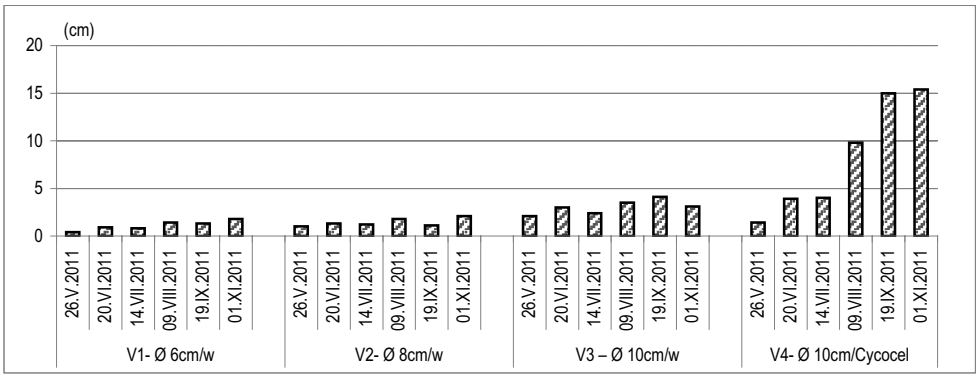
In addition to the measurable parameters, there is noted that by the application of Cycocel treatments the decorative appearance of plants was improved due to a sharp leaf colour, due to the contrast white - deep green, the leaves are healthy compared to the untreated variants, characterized by a softer colour and even the appearance of leaves with green-sickness at the end of the experimentation period.



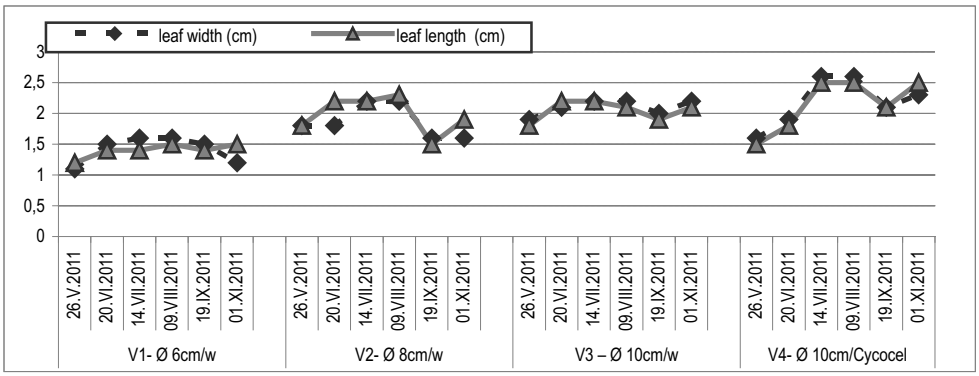
Graph 1. The average height of *Plectranthus coleoides* ‚Marginata' plants



Graph 2. The average number of shoots/plant



Graph 3. The average lenght of shoots



Graph 4. The average size of leaves

CONCLUSIONS

The presented data show that the treatments with Cycocel 0.3% at *Plectranthus coleoides* determined a vegetative growth, plants with compact appearance by increasing the average number of shoots /plant, especially in the first four months of experimentation, then the character of pendent plant became very obvious because of the exaggerated growth in length of the shoots.

The decorative appearance of plants was improved due to the sharp leaf colour, the contrast white-deep green, the healthy leaves, compared to the untreated variants, characterized by a softer colour and even the appearance of leaves with green-sickness at the end of the experiment. The reaction of plants to the treatments with Cycocel could be considered an advantage for the situation when *P. coleoides* is grown in floral suspensions.

We recommend testing the cultivation in pots of 6-8 cm combined with the application of up to 3 treatments with Cycocel 0.1-0.2% (1000-2000 ppm), every 2 weeks for height control and for obtaining compact bush plants of *Plectranthus coleoides* 'Marginatus'.

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RESEARCH REGARDING THE AUTENTIFICATION OF BIOACTIVE COMPOUNDS IN CHERY BEER

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Key words: *beer, cherry, anthocyanin, authentification, UV-Vis spectroscopy*

ABSTRACT

Special beers, especially cherries beers are drinks by consumers for their sensorial properties and due to intake of bioactive substances, especially anthocyanins compounds. Fermentation maceration technology of beer with cheery fruits is the process that brings valuable compounds in beer. Because there is a category of drinks that can be obtained by using food additives, it is necessary to identify biochemical biomarkers to authentification the originality of the product. Using HPLC-MS techniques anthocyanin compounds, 3.6 cianidin cumaroil glucoside was identified as markers of genuineness of cherries in beer.

INTRODUCTION

Cherries beer is based on fermentation process over several months in the presence of cherries often added during secondary fermentation, followed by a maturation period of several months. During this time there is a secondary fermentation in the presence of sugars from cherries and continued even after removing them, so that beer has a sweet flavor. In the last time, breweries add cherries, or replace them with cherry syrup towards the end of the production process to make it less intense and more accessible to the consumer. Beer color is pink to red-purple, sour dry taste, slightly bitter and strong carbonated.

The total antocyanins content was determined with different methods described by Boyles and Wrolstad, 1993; Gusti and Wrolstad, 2001; Shin et al., 2008, Tsantili et al.,2010. Novel technic have been develop for identification and cuantification of this compunds. Pappas et.al., 2011, used diffuse reflectance infrared Fourier transform spectroscopy for quantitative determination of antocianins in sweet cherry varieties.

MATERIALS AND METHODS

The identification of bioactive compounds from cherries and beer it was done for establish the traceability of anthocyanins from fruits to final products. For beer production it was use the traditional technology. The malt it was mashing with water and

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saccarification it was done by infusion regime. The mash it was filtered in lauther tun and the sweet wort it was boiled for 90 minute.

For hopping a rate of 8 mg isoalphaacid/hl wort it was used from bitter hops varieties, Hallertau Magnum. The fermentation process was conducted in two stages, primary and maturation steps. Primary fermentation was performed with lager yeast at 10 ^{o}C, for 7 days. The maturation period it was a maceration fermentation of beer with sour cherries fruits (*Prunus cerasus*) for two month. The finished beer was filtered and packed in bottle.

For traceability of antocyanins from fruit to beer it were identificate the antonyanins in cherries fruits and then in beer.

Identification of antocyanins from cherries fruits

1 gram of sour cherry is mixed with 10 ml of methanol (HPLC grade) and 1 ml hydrochloric acid (36,5%) and the sample is shaken for 15 minute for antocyanins extraction. The extract is diluted with distillated water in 1:10 ratio. The total antocyan content is determinate by UV –Vis spectroscopy method. A spectrophotometer Shimadzu was used to measure the absorbance of extract between 220-700 nm.

Identification of antocyanins from beer

The cherries beer is degassed for elimination of CO_2 content and filtered for UV-Vis analysis. The sample absorbance is determinated by UV-Vis spectroscopy at 220-700 nm.

The confirmation of presence of antocyanins compunds in cherries and beer have been done by HPLC-MS technique.

RESULTS AND DISCUSSIONS

In the UV-Vis spectrum of sour cherries (fig. 1) and cherries beer (fig.2) it was identificat a maximum absorption at 500 nm for both sample. Analyzing the UV-VIS spectra of both samples is observed that the maximum absorbance, both for cherry methanol extact and cherry beer stands where around 500 nm, the wavelength corresponding anthocyanin compounds.

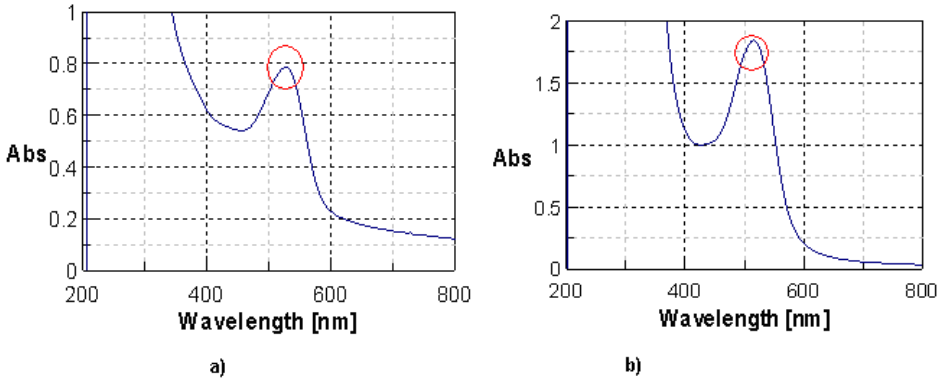

Figure 1. UV-VIS spectrum of methanolic extract of cherries (a) and cherries beer (b)

Presence of anthocyanins compounds in beer with cherries show that they do not suffer oxidation or degradation processes during the maturation of beer.

For accurate identification of anthocyanin compounds detected by UV-VIS spectrophotometry a HPLC-MS analysis was performed to determine if the same compounds were found in cherries and cherries beer. The results are presented in the chromatograms in figure 2 and figure 3.

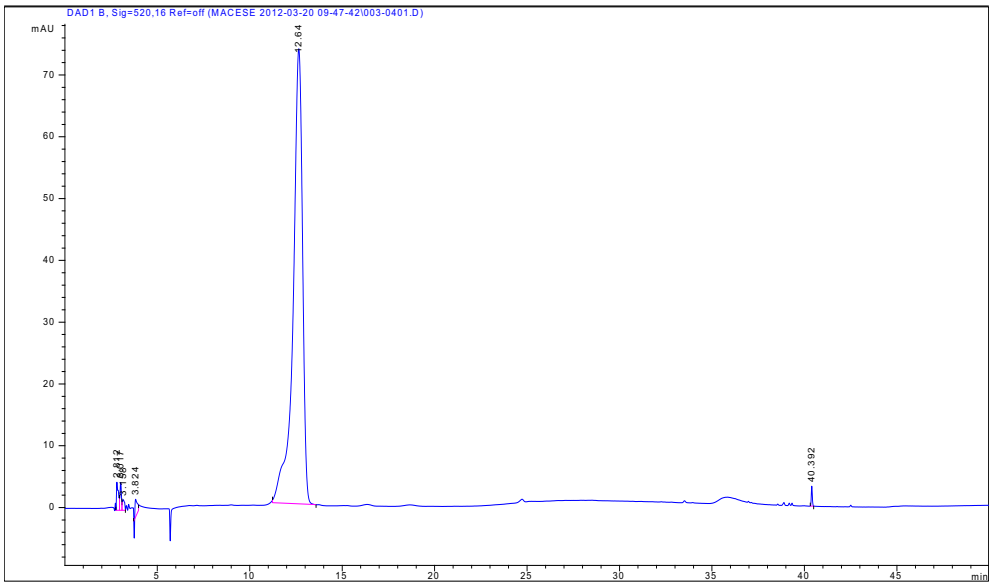

Figure 2. HPLC chromatogram of methanolic extract of cherries at 520 nm

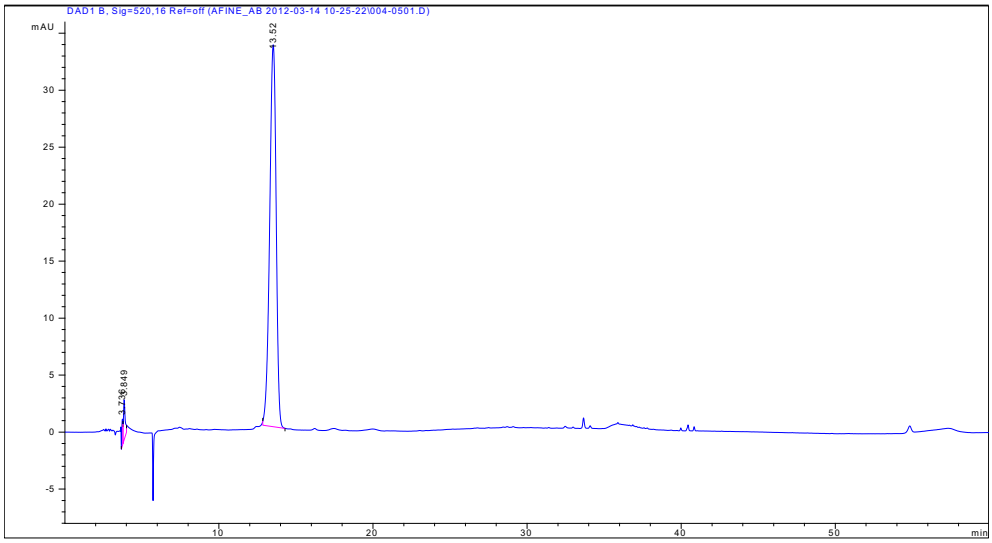

Figure 3. HPLC chromatogram of cherries beer at 520 nm

Chromatograms reveal the existence of the same compound in cherry extract and cherry beer. In order to determine the class of compounds in which the compound shown in chromatogram at a wavelength of 520 nm at retention time of 12.627 minutes from methanolic extract of cherry and at the retention time of 13.670 minutes for beer with cherries, the results are subjected to spectral analysis.

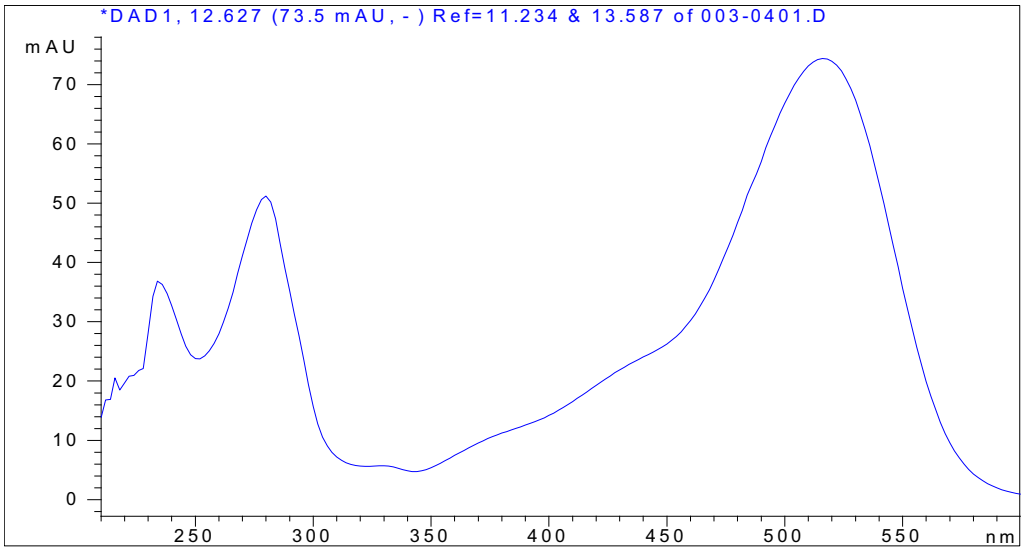

Figure 4. HPLC-MS spectrum of methanolic extract of cherries

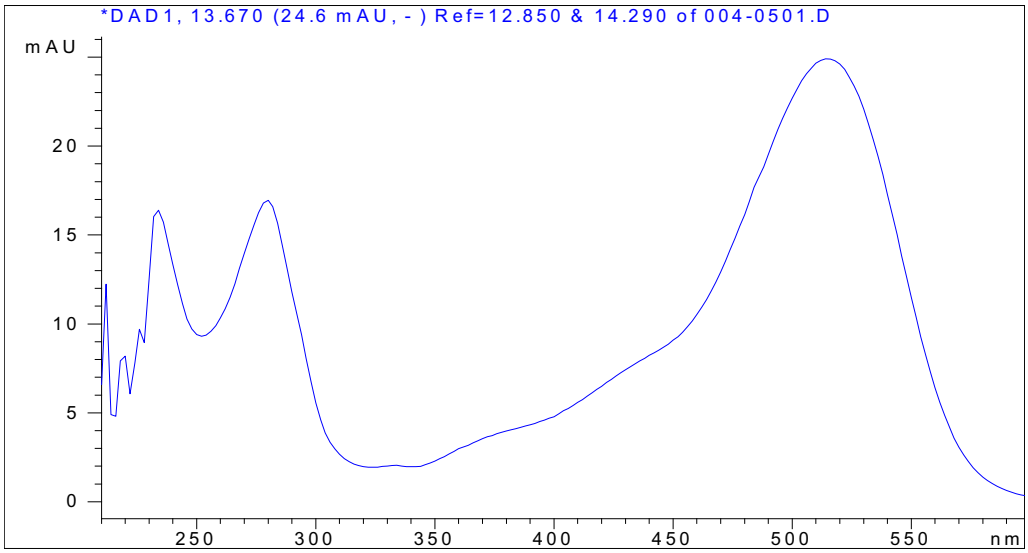

Figure 5. HPLC-MS spectrum of cherries beer

Spectrum analysis of both samples revealed the presence of two absorption bands of the compound at wavelengths 530 nm and 280 nm.These wavelengths are specific for anthocyanin compounds (maximum absorption for anthocyanin compounds are at 270 .. 280 nm, 330 nm and 315 ... 500 ... 550 nm).

HPLC-MS enables precise identification of the compound. Based on the molecular weight can accurately identify compounds present in the samples.

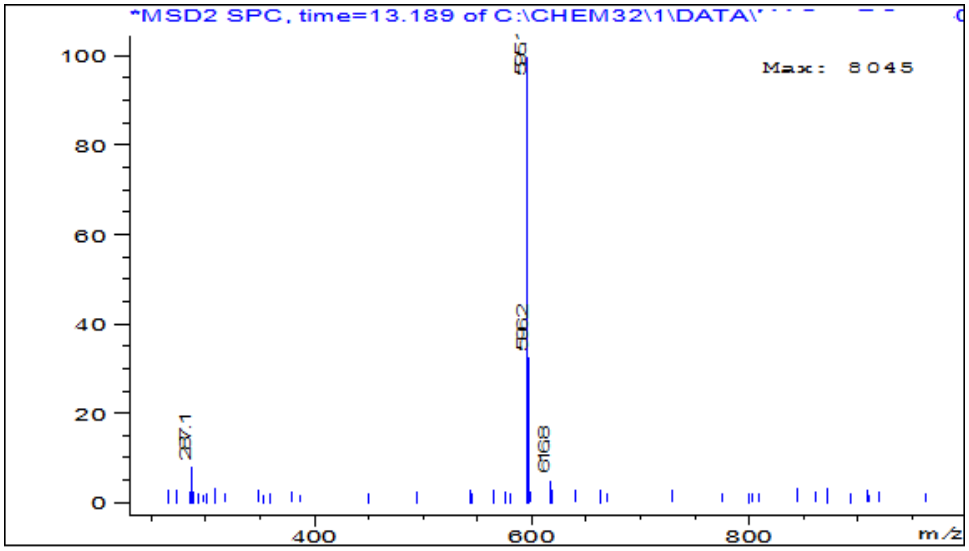

Figure 6. Mass spectrum of methanolic extract of cherries

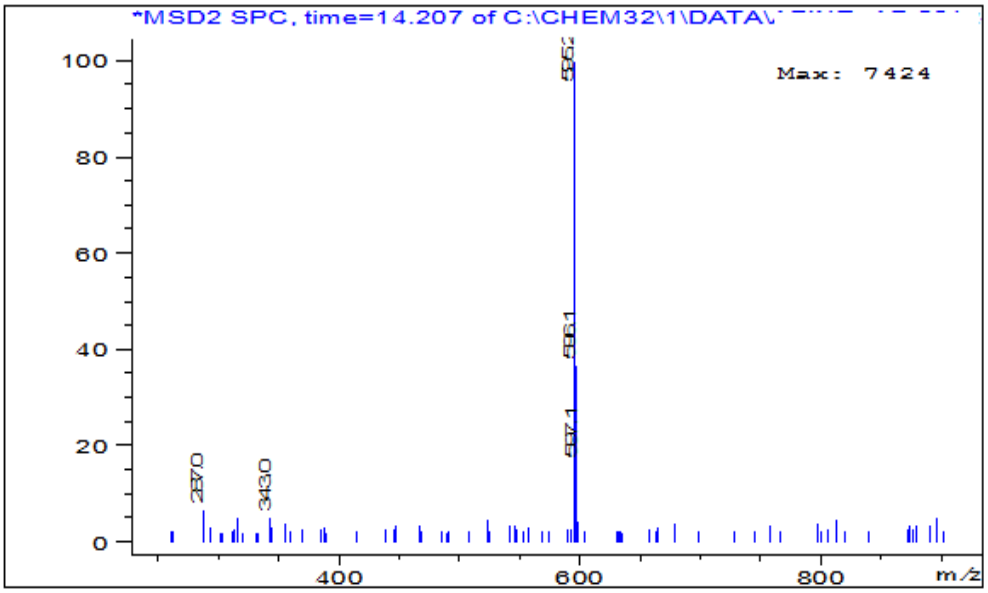

Figure 7. Mass spectrum of cherries beer

Mass fragments detected by mass spectrometry present the same molecular weight (595.2 and 287.1) for both compounds identified at retention time 12.670 minutes and 13.679 minutes. Anthocyanin compound identified by molecular weight is 3.6 cianidin cumaroil glucoside.

CONCLUSIONS

Traceability of anthocyanin compounds in beer production has been proved by spectrophotometric and chromatographic techniques. It was identified a marker compound, 3.6 cianidin cumaroil glucoside from cherries beer. UV-Vis spectrum analysis of beer can be a simple and rapid method for product authentication.

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RESEARCHES REGARDING THE QUALITY OF THE WINES OBTAINED FROM DIFFERENT CABERNET SAUVIGNON CLONES IN THE SAMBURERSTI VINEYARD IN 2011 YEAR

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Key words: *Samburesti vineyard, quality, clones*

ABSTRACT

Cabernet Sauvignon is the main grapevine variety in Samburesti vineyard. The wines obtained in 2011 year by young vines have a great evolution potential, good values of alcohol, glycerol and total acidity content. Concerning the amount in polyphenolic compounds, all the wines have higher values of index of polyphenols totals, tannins and anthocyans contents and intensity of colour.

INTRODUCTION

Samburesti vineyard is recognized for this red wines obtained from famous cultivars widely in the world: Cabernet Sauvignon (Baduca Campeanu C., 2008). Environmental variables are considered the most influential factors on grapevine production and berry composition (Montes C. e.a., 2012). The influence of climate on wine quality is well known, through the effect of both regional and local-scale climatic conditions during the growing season, and by its interannual variability, which generates variations in grapevine growth and then in berry composition (Soar et al. 2008).

Worldwide, a large number of studies examining the climatic features have provided the description of different *terroirs* and the identification of winemaking regions using different methodologies (Montes C. e.a., 2012). Temperature is widely accepted as being the primary climatic factor affecting the quality of viticultural production (Gladstones 2004). As a consequence, increases in temperature due to an enhanced greenhouse effect will likely have a significant effect on viticultural production. Possible beneficial aspects of climate change include less bud and crop damage from frost events and less extreme winter minimum temperatures that would otherwise damage grapevines (Jones 2005).

There were strong correlations between sugar content, colour and quality perception in grapes and the resulting wines. The best Cabernet-Sauvignon wines were made from grapes rated highly for colour intensity, red berry and black berry with spice aroma. Seasonal differences resulted in larger variance in grape composition than grapes

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originating from vineyards in different climatic zones. This highlights the difficulties in pinpointing a specific parameter to indicate optimal maturity (Oberholster A. et al., 2010). Phenolic compounds are extracted from skin and seeds, and their extraction is influenced by winemaking procedures (Moreno-Perez A. et al., 2010).
The Samburesti vineyard is well-known in our country for the quality of the red wines which are produced from acknowledged sorts worldwide, Cabernet Sauvignon and Merlot **being probably the** most important ones. Lately, the Samburesti vineyard has been part of an extensive process of reconversion, through the clearing of the old plantations and the replacement of them with new, modern plantations with sorts that belonged to the previous sorts but with new ones as well.

MATERIAL AND METHOD

The present paper was achieved taking into account the studies regarding the main chemical composition and polyphenolic parameters of the wines obtained in 2011 from different clones of Cabernet Sauvignon from the Samburesti vineyard. The wines were obtained by means of micro wine production at the Agriculture and Horticulture Faculty of the University of Craiova. For all the clones, the grapes have been harvested on the same day and there have been applied the same technological operations of wine production, so that the differences between the wines to be given only by the potential of each clone, avoiding thus the influence of some technological factors.
In the study, there were taken three clones: 169 (grafted on stocks SO_4 Gravesac and R110), 337 (grafted on stock Gravesac) and 685 (grafted on stock SO_4). Therefore, there are five experimental variants of wines, at which it is added the sixth one, CSR that comes from an over 20 year old Cabernet sauvignon plantation.

The study of the chemical composition was achieved in March 2012, aiming at the main compositional parameters of the wines being achieved in the Oenology lab following the official analyses methods of the wines.

The physical studies were achieved following spectro-photometrical methods by the specialized staff from the lab off physical0chemical studies of the horticulture products, where there were determined the index of totals polyphenols, the content in anthocyans and the chromatic structure(intensity and color tent), the determinations being achieved when the wines were three months old following the methods described by Ribberau. Unlike the composition studies, the spectro-photometrical ones aimed at a greater number of wines, because on the main wines there have been accomplished combinations between the wines obtained from a clone but grafted on different stocks or wines coming from different clones but grafted on the same stock.

RESULTS AND DISCUSSIONS

The study of the main compositional parameters of studies Cabernet Sauvignon wines, presented in table 1, indicated a significant difference between the wine obtained from an old plantation and the ones obtained from young plantations that were in the first year of production. From the alcohol strength perspective, the wine coming from the old plantation presents a great value(14,8% volume), due to the fact that the wine came from very rich in sugar grapes. The wines obtained from new clones display values of alcoholic strength with 1-2% lower. The greatest alcohol strength of the wines obtained from new

clones is of 13,7% volume at the wine obtained from clone 685/SO_4. The latest is followed by the three wines obtained from clone 169 of which the one grafted on stock Gravesac has 13,5%volume, the one grafted on stock SO_4 has 13,3% volume and the one grafted on stock R110 has 13,1 % volume. The only wine with the alcohol strength beneath 13,0% volume is the one obtained from clone 337 grafted on stock Gravesac, with 12,8% volume.

Even though the five wines obtained from the newly-introduced clones have the alcohol strength lower than the one belonging to the wine obtained from a mature plantation, all of them have exceptional alcohol strength especially under the conditions of some grapes productions which were higher than the old plantation. The fact that all the wines, with one exception, had over 13% alcohol volumes is remarkable and acknowledges an outstanding qualitative potential of these clones.

When it comes to the glycerol content, it was again clone 685/SO_4 that remarked with a content of 12,8 g/l. On the last position, it was situated once more clone 337/Gravesac with 11,5g/l the only one with content lower than 12,0g/l. Of the three varieties of clone 169, on the first position, it was the one grafted on stock Gravesac, followed by the one grafted on stock SO_4 and the one grafted on stock R110. So, at the glycerol content, we have the same order like in the alcohol content, taking into account that both parameters are tied by the sugar content of grapes and by the fermentative capacities of the yeasts used at fermentation.

Table 1

The chemical composition of the Cabernet Sauvignon wines

The wine	Alcohol, % vol.	Glycerol, g/l	Residual sugar, g/l	Total acidity, g/l H_2SO_4	Volatile acidity, g/l achetic acid	SO_2 free, mg/l
C. S. R	14,8	13,2	3,0	4,5	0,38	12
169/SO_4	13,3	12,1	3,2	5,0	0,42	16
169/R110	13,1	12,0	3,1	4,7	0,36	20
169/Gravesac	13,5	12,4	3,6	5,2	0,40	15
337/Gravesac	12,8	11,5	3,4	5,3	0,33	22
685/SO_4	13,7	12,8	3,5	4,9	0,42	14

The results of the studies regarding the total acidity of the wines indicate the fact that the wine with the lowest acidity is the one coming from the old plantation from the ripest grapes. The other wines present contents between 4,7 and 5,3 g/l. Obviously, these values of total acidity are more seldom met at red wines, especially when it comes to those that contain over 13,0% alcohol volume. It is, undoubtedly, a proof of the qualitative potential of the clones and show that the harvesting of the grapes could have been delayed if it had been desired the accumulation of some great proportions of sugars (although it was useless) because the total acidity of the grapes would have allowed this thing.

Regarding the values of the volatile acidity in the case of the six Cabernet Sauvignon wines, regardless of the grapes they are coming from, all have values with a lot

beneath the legal limit, of 1,2 g/l acetic acid and even beneath the perception limit, of 0,6-0,7 g/l acetic acid. Values of volatile acidity of 0,33-0,42g/l acetic acid are excellent and prove that the wines are healthy and stable from the biological point of view, due to a successful wine making

The spectro-photometrical studies that targeted at the characterization of the polyphenolic composition and of chromatic features have been done on a number of nine wines compared to the chemical composition where six wines were studied. The results of these studies are presented in table 2 and suggest that all the wines have a good evolution potential.

The first element of polyphenolic composition that has been studied in the index of total polyphenols, determined at 280 nm under the form of optical density. At this indicator, on the first position it situated the wine from clone 169/R110. On the following positions, there were the wines from clones 337/Gravesac and 685/SO_4 (46,64 respectively 44,18). On the 5th position, it was the wine obtained from the combination of the variants of clone 169 (41,35) and on the 6th position, the wine obtained from the combination of clones grafted on stock SO_4 (40,50). The last three wines had the value of the IPT index below 40. Thus, on the 7th position, it was the wine obtained from the combination of the two clones grafted on stock Gravesac(39,22) on the 8th position the wine from clone 169/SO_4 (38,139) and on the last position the wine from clone 169/Gravesac.

At the Cabernet Sauvignon wines, the determination of anthocyan contents emphasized the fact that all the wines are very rich, displaying values between 573 and 863 mg/l. But this thing is not a surprise, the issue in discussion being the sort with the greatest capacity of anthocyans accumulation and also the vineyard with a high degree of favorability for obtaining red wines. It was also the fact that the wine-producing year was an exceptional one from the climatic point of view which meant that there were met all the conditions for easing the accumulation of some important proportions of anthocyasns in grapes. The wine production conditions, the manner of managing the processes of maceration-fermentation, even though it was about micro wine- production, allowed the extraction of some important proportions of anthocyans from grapes that is why all the natural and technological factors allowed the obtaining of wines rich in anthocyans and intensely colored.

Table 2

The chromatic structure of Cabernet Sauvignon wines

The wine	IPT, D.O. 280 nm	Anthocyans mg/l	I	T
C. S. R	45,98	731	1,36	0,48
169/SO_4	38,13	662	1,11	0,52
169/R110	48,37	863	1,45	0,51
169/Gravesac	36,69	573	1,16	0,53
337/Gravesac	44,64	734	1,50	0,48
685/SO_4	44,18	779	1,16	0,56
169	41,35	629	1,30	0,52
169+685/SO_4	40,50	667	1,10	0,54
169+337/Gravesac	39,22	596	1,40	0,53

The highest content in anthocyans was met at the clone that also displayed the highest index of total polyphenols, 169/R110, followed at a distance of over 80mg/l by clone 685/SO4.

It was followed by clone 337/Gravesac and very close to her, the wine obtained from the old sort (731 mg/l). The lowest contents in anthocyans were displayed by two wines obtained from clone 169 and those obtained from the combination of the two clones grafted on stock Gravesac: 169 and 337. Therefore, all that was grafted on stock Gravesac displayed contents in anthocyans that were with a lot beneath other clone-stock combinations. At least in the case of clone 169, the differences are huge, with 89 mg/l lower than the combination with stock SO4 and with 190mg/l lower than the combination with stock R110.

All the nine wines displayed values of the colorant intensity comprised between 1,10 and 1,50 which signifies a great richness of color. Taking into account that these are very young even raw wines, which do not meet all the conditions that would allow them to be consumed their color richness is understandable.

From the study of the presented values belonging to the nine wines, it has to be noticed that on the first position, it was situated the wine obtained from clone 337/Gravesac. On the second position, at the colorant intensity, it was the wine obtained from the combination of the two clones (169 and 337) grafted on stock Gravesac (1,40). These three wines significantly got ahead of the one from the old plantation (Cabernet Sauvignon R). On the fifth position, it was the wine from the combination of the variants of clone 169, grafted on both stocks. On the 6^{th} and 7^{th} position, there were the two wines obtained from clones 169/Gravesac and $685/SO_4$ and on the last two positions, two wines obtained from clone 169 grafted on stock SO_4 (the simple variant and the one combined with clone 685).

As for the tonality values, it can be observed that they are quite low, between 0,48 and 0,56, which means two or three times lower as intensity, this being the natural consequence of the fact that the main contribution to the colorant intensity of the wines was due to the red pigments in other words the optical density at 520 nm.

CONCLUSIONS

The researches regarding the wines obtained from new clones of Cabernet Sauvignon, the 2011 harvest, allowed the accumulation of the following series of conclusions:

- Considering the aspect of chemical composition, all the wines obtained from young plantations from the first year of production, displayed lower contents in alcohol and glycerol compared to the wine obtained from a mature plantation but considering the aspect of polyphenolic composition, they proved to be superior.
- The fact that we are talking about wines coming from a young plantation from the first year of production, it has to be regarded as a promise that on the future, the wines will be displayed at a superior qualitative level, while the plantation will strengthen and will better resist to the climate factors with a disturbing influence upon the process of raising and maturation of grapes.
- Clone 685 seems to be the one that best adapted itself to the specific conditions of the Samburesti vineyard and offers the most interesting perspectives of obtaining high quality wines.

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THE INFLUENCE OF WATER STRESS AND SUBSTRATE VOLUME ON GROWTH AND DEVELOPMENT *BELOPERONE GUTTATA* Brandeg. PLANTS

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Key words: *water stress, nutrition space, growth control, morphological characters*

ABSTRACT

In this paper there are presented the results of the substrate volume and water stress influence on the Beloperone guttata Brandeg. plants` growth and development in order to obtain reduced size plants.

The main morphological characters analyzed were influenced obviously by the reduction of the substrate volume (the size of flower pot) and the amount of water given to plants. The minimum values of the analyzed parameters were recorded for the plants that were grown in a reduced substrate volume (flower pots with a diameter of 10 cm) and for the plants that have received a smaller amount of water (150 ml/pot).

These cultural methods can be considered ecological variants of the plants height control in the future in order to reduce the application of chemicals (retardants), which indirectly pollute the environment.

INTRODUCTION

Beloperone guttata Brandeg. known as the "shrimp plant" is a small shrub with thin and flexible stems and oval-elliptic leaves with pointed tip, of light green colour. The main decorative element is represented by inflorescences similarly spikes, formed of pink or reddish-brown bracts, overlapped and persistent, with scaly shape very similar with a shrimp. It is a plant with a rich flowering, cultivated in pots, being used in decorating the apartments, terraces and balconies or in the garden during summer.

The reduced size plants (miniplants) have different origins: specific, genetic and cultural (Vidalie 2004, Anton et al. 2008), being very popular in recent years due to various possibilities of use.

Different techniques can be applied to obtain small size plants with compact shrub, such as: reduction of substrate volume by growing plants in small pots (Nicu et al. 2009, Vita & Lauro 2002, Nesmith & Duval 1998); applying water stress by reducing the amount of water given to plants (Cameron et al. 1999, Manda et al. 2008); plants height control by applying DIF - difference between day and night temperature (Blanchard & Runkle 2011,

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Berghage 1998); plants height control through mechanical methods (Latimer 1998), reducing fertilization, application of retardants etc.

This paper presents the results concerning the reaction of *Beloperone guttata* Brandeg. plants to the application of some cultural methods (water stress and reduction substrate volume).

MATERIAL AND METHODS

The biological material consisted in cuttings of shoots rooted in perlite, from the *Beloperone guttata* Brandeg. plants belonging to the Floriculture discipline collection.

Two experiments were conducted to study the influence of substrate volume and water stress on plant growth and development. In the first experiment the rooted cuttings were planted in May, in pots of different sizes (V1 control -14 cm, V2-12 cm and V3-10 cm diameter) in a mixture of peat and perlite (1:1). In the second experiment the cuttings were planted in pots of 14 cm diameter, in a mixture of peat and perlite (1:1). There were given different amounts of water weekly (V1 control - 250 ml/pot, V2 - 200 ml/pot, V3 - 150 ml/pot).

Observations and measurements were effectuated on the morphological characters (plant height, number of shoots, number of leaves, leaf length and width, number and length of inflorescences) during May-September 2011 period and there was observed the rhythm of vegetative growths (plant evolution) depending on the amount of water given and the volume substrate (the space of nutrition).

RESULTS AND DISCUSSIONS

In the first experiment, the results obtained show that after five months after planting, the highest values of height were recorded by the cuttings planted in pots with a diameter of 14 cm (30.2 cm) and the lowest values by the cuttings planted in pots of 10 cm (17.2 cm).

As the rapid increase of plant height is not an advantage for the reduced size plants, the 13 cm difference recorded between V1 (control) and V3, confirms that by reducing the space of nutrition, the growth rate of the *Beloperone guttata* plants is slower (graph 1).

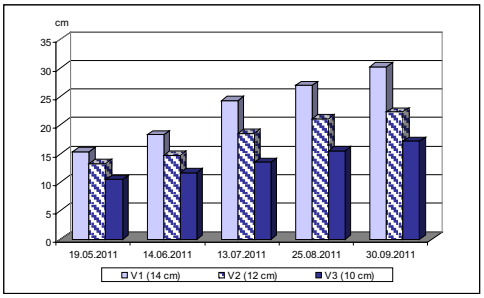
Depending on the substrate volume (pots size), the average number of shoots per plant, showed similar values in the first two months after planting in pots. At the end of September, when the last observations and measurements were done, there was found that the average number of shoots per plant tripled for the plants that have benefited from a larger space of nutrition (12.0 shoots at V1) in comparison with the plants grown in a reduced substrate volume (7.0 shoots at V3) (graph 2).

The average number of leaves per plant was influenced by the reduction of substrate volume. The lowest number of leaves was recorded for the plants grown in pots with a diameter of 10 cm.

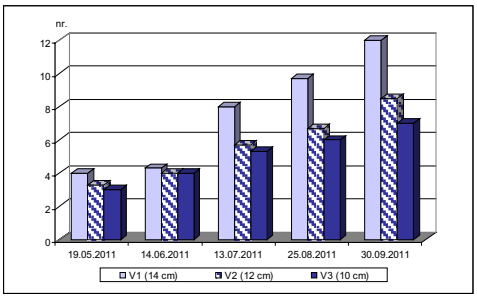
Reducing the substrate volume influenced also the leaves dimensions (length and width) of the three studied variants. We can notice in graph 3 that the lower value of leaf length (4.5 cm) was recorded by the plants which were grown in pots with a diameter of 10 cm (V3) and the highest value (8.4 cm) at V1 (control plants). Regarding the average width

of the leaves, the lowest values were recorded at V3 (3.0 cm) in comparison with the control plants (4.3 cm) (graph 4).

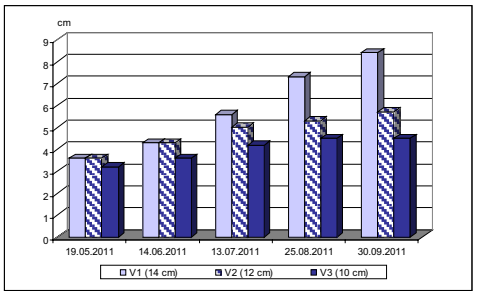
The beginning of flowering was observed for all the plants, regardless of pots size, at a month after planting. Regarding the dynamics of the average number of inflorescences per plant and the inflorescences length there were recorded values below the control at the plants that were grown in pots with smaller diameter (graphs 5 and 6).



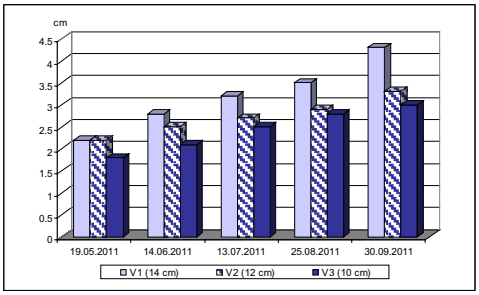
Graph 1. The average height of plants



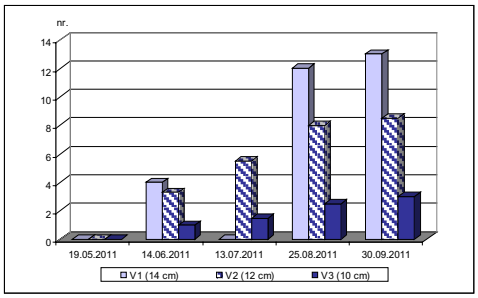
Graph 2. The average number of shoots/plant



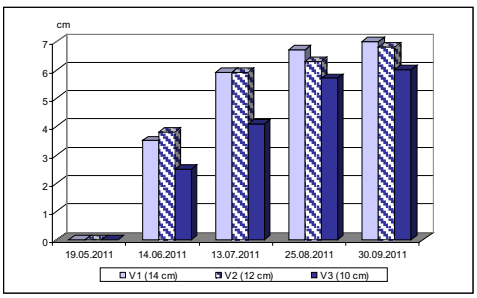
Graph 3. The average length of leaves



Graph 4. The average width of leaves



Graph 5. The average number of inflorescences/plant



Graph 6. The average length of inflorescences

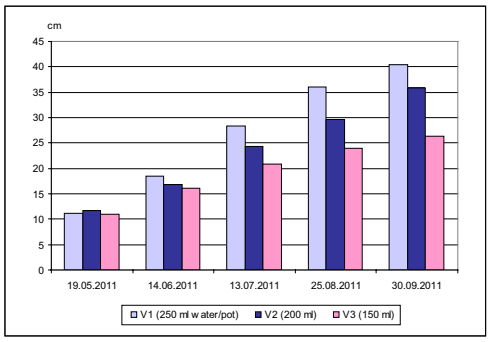
In the second experiment there was observed the influence of water stress on plants growth and development. From the analysis of the results obtained after 5 months of planting in pots, there is observed a reduction in plant height by 35% for the plants that received a smaller amount of water/pot (V3 - 150 ml) and by 12% for the plants that were given 200 ml water/pot (V2), in comparison to the control (250 ml) (graph 7).

The average number of shoots per plant, depending on the amount of water given, had similar values in all variants, throughout the period of observations and measurements. The lowest number of shoots was recorded at the plants that received the smallest amount of water (150 ml), namely 5.0 shoots/plant (graph 8).

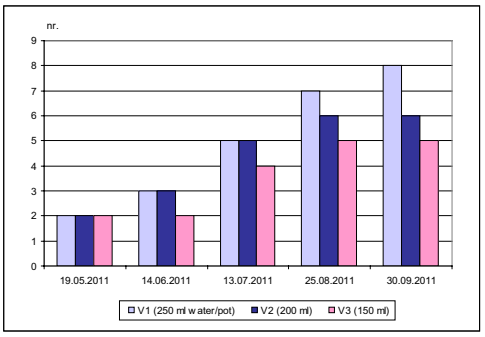
Regarding the average number of leaves per plant, respectively the leaves sizes (length and width), the lowest values were recorded at the variant which received 150 ml of water/pot (graphs 9 and 10).

The water stress applied to *Beloperone guttata* plants influenced the number of inflorescences per plant and the inflorescences length.

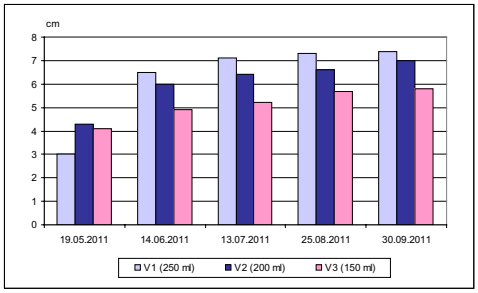
Thus observing the dynamics of the average number of inflorescences per plant, the higher values were recorded, compared with the control, by the plants that received a smaller amount of water (graph 11). Regarding the length of inflorescences, the results obtained show that the water stress had a positive effect on this analyzed morphological character, achieving higher average values at the other variants in comparison with the control (8.6 cm at V2 and 8.0 cm at V3) (graph 12).



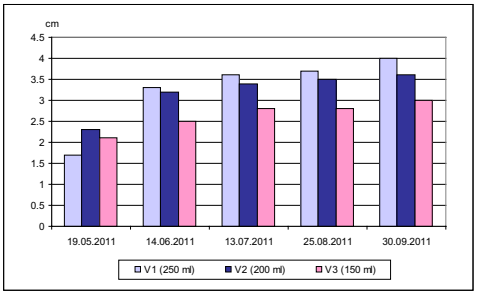
Graph 7. The average height of plants



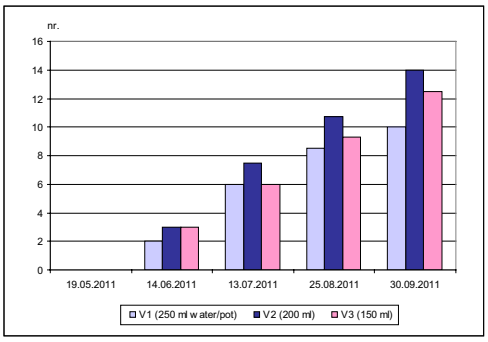
Graph 8. The average number of shoots/plant



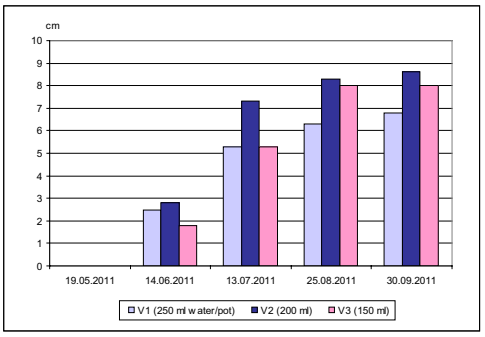
Graph 9. The average length of leaves



Graph 10. The average width of leaves



Graph 11. The average number of inflorescences/plant



Graph 12. The average length of inflorescences

CONCLUSIONS

The used substrate volume influenced the main analyzed morphological characters, the highest reduction occurred at the plants grown in a reduced substrate volume, respectively in pots with a diameter of 10 cm.

The results obtained show that the reduction of substrate volume (nutrition space) can be recommended as a cultural method for plants height control at *Beloperone guttata* Brandeg. species.

After 5 months from planting in pots, the application of water stress determined a reduction of plant height by 35% at the variant which received 150 ml water/pot and by 12% at the variant which received 200 ml water/pot, in comparison with the control.

The main analyzed morphological characters (plant height, number of leaves per plant, leaf length and width), recorded the lowest values for the plants that received a minimum amount of water (V3 - 150 ml/pot).

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THE INFLUENCE OF ORGANIC FERTILIZERS ON *SAINTPAULIA IONANTHA* SPECIES

Osiceanu Silvia-Marcela[1]

Key words: *Saintpaulia ionantha, culture background, organic fertilizer, cuttings, dividing mature plants.*

ABSTRACT

The paper presents the influence of Siforga organic fertilizer on the blooming capacity of Saintpaulia ionantha species. The Saintpaulia ionantha plants have been obtained by dividing mature plants and planting on two types of background. The trial has been set up in December 2005 and kept 3 years, till 2008 with three replications. The optimal doses of Siforga fertilizer have ensured the maximal number of flowers (in average, 26; 18-28; 20) have been 99-113N, 60-69P, 158-182K, which means average experimental doses and the best background proved to be black peat + leaves earth + sand in ratio of 2:1:1.

INTRODUCTION

The African violet *Saintpaulia ionantha* H. Wendland is a pot plant that is extremelly popular all over the world and its trade reaches millions of dollars annually (Ambrozevicius 2002). The demand for this ornamental plant are growing though the low level of income make many romanians to buy less flower than would want, often only for certain occasions.

The *Saintpaulia* genus are among the most interesting plants that can be grown indoor due to the fact they produce flowers all year round and the flowers are special, colorful; the plant is dwarf and can easily find a place to make beautiful, it can be used in floral arrangements, baskets or larger pots among other flowers (Pavel 1977).

Another advantage over other flowers is that it can be easily reproduced vegetativelly. It represents a useful researching material for genetics due its capacity to form sexuated hybrids (Şelaru 2004).

Saintapulia ionantha H. Wendl. belongs to *Gesneriaceae* family and it is the base species for obtaining all cultivated forms (Burtt 1958).

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MATERIAL AND METHOD

Within experiments we have used plants and cuttings of Uzambar violet, group *Inova Spectra* that belong to *Yelda* and *Figaro* cultivars. There have been used mature plants resulted from both classic cutting and shrub dividing (Răuță & Chiriac 1980).

The backgrounds researched for the influence of organic fertilizer on the blooming capacity of *Saintpaulia ionantha* H. Wendl. have been:

- a1 – black peat + leaves earth + sand 2:1:1 and
- a2 – red peat + leaves earth + sand 2:1:1(Fauchier 1990).

The organic fertilizer Siforga is a solid fertilizer as pellets with slow decaying and that is the reason why it was incorporated into the background.

The calculus for the fertilizer quantity per surface unit was made using the following formula

$$\frac{a_{g\ fertilizer}}{\pi R^2 cm^2} = \frac{100000\,x\,a}{\pi R^2} \quad \frac{kg\ fertilizer}{ha}$$ (Răuță & Chiriac 1980)

Whether the fertilizer contains C% active ingredients (N, P_2O_5, K_2O etc.), then the fertilizer dose D a.i. (dose of active ingredient) will be calculated, for Siforga fertilizer, after the following formula:

$$D_{s.a.} = \frac{1000\,x\,a\,x\,C}{\pi R^2} \frac{kg s.a.}{ha} \text{ N, } P_2O_5\text{, } K_2O \text{ etc.}$$

C = 5% N, 3% P_2O_5, 8%K_2O

V = 1; 1,5; 2; 2,5; 3 g/pot

b_1 = Ctrl – not fertilized

$b_2 = N_{71}P_{42}K_{113}$ corresponding to 1 g/pot

$b_3 = N_{85}P_{51}K_{136}$ corresponding to 1,5 g/ pot

$b_4 = N_{99}P_{60}K_{159}$ corresponding to 2 g/ pot

$b_5 = N_{113}P_{69}K_{182}$ corresponding to 2,5 g/ pot

$b_6 = N_{127}P_{78}K_{205}$ corresponding to 3 g/ pot

In order to perform the experiments there were used ceramic pots with the following dimensions: h = 15 cm, φ = 15 cm (r = 7,5 cm).

As working method there was set up an experiment with two factors and three replications:

- The A factor – the crop background with a1 and a2 graduations;
- The B factor – the Siforga fertilizer doses, with 6 graduations: b_1, b_2, b_3, b_4, b_5, b_6.

The experiment started in 2005 and lasted till 2008.

Apart from food crops where the quantity is appreciated and the quality means a certain taste, a chemical composition, with flowers the quality is given by the appearance and express a ratio between certain morphological features (e.g. number of flowers, leaves, etc.) (Ceapoiu 1976).

In the interpretation of the results of the experiment the analysis of variance is very important and it has to be completed with biometrical measurements (Săulescu & Săulescu 1964).

Along with the analysis of variance there was used graphics as well as the calculus of multiple comparisons (Iancu 2002).

RESULTS AND DISCUSSIONS

The blooming capacity (the number of flowers) of *Saintpaulia ionantha* species was very much influenced by the two experimented factors (the culture background and the dose of Siforga fertilizer) as well as of their graduations.

In order to determine the effect of these two factors on the number of flowers within 2006-2008 period it is useful to analyze, first, the separate influence of each factor.

The culture background, as a separate influence has modified the number of flowers in the following way: the plants cultivated on black peat + leaves earth + sand have had 25.33 flowers and the ones cultivated on red peat + leaves earth + sand have formed 22.55 flowers that represents a negative significant difference (Fig. 1)

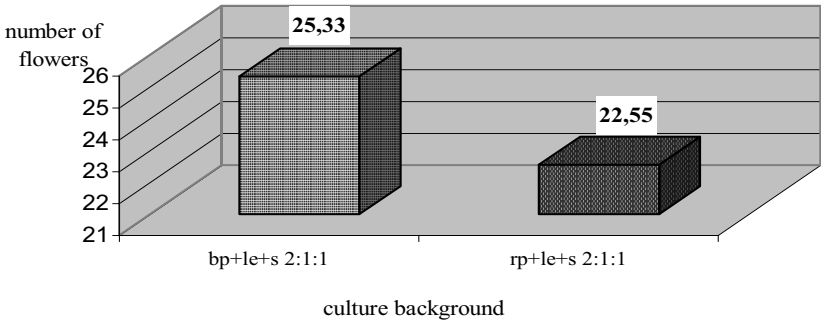

Figure 1 The separate influence of the culture background on the number of flowers with Saintpaulia ionantha (average 2006-2008)

The result is that the black peat used for nutritive background is more indicate because it determine more flowers.

Whether we reffer to the NPK dose as Siforga fertilizer there can be demonstrate that it has had a higher effcet on the blooming capacity of Uzambar violet (Fig.2).

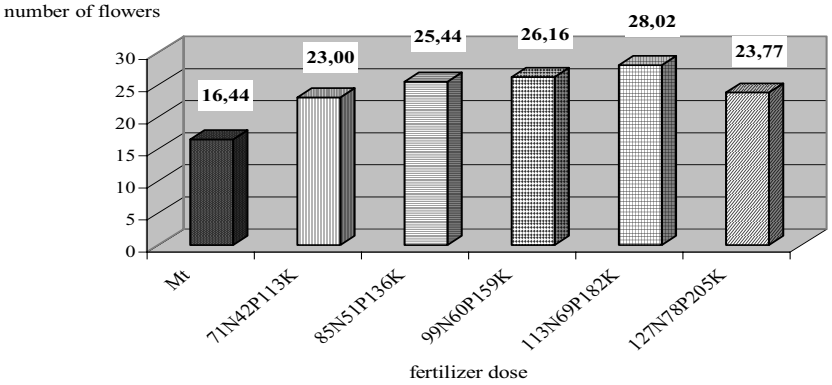

Figure 2 The separate influence of Siforga organic fertilizer doses on *Saintpaulia ionantha* number of flowers (average 2006-2008)

When no fertilizer was used, on average for three years, on each plant there were counted 16.44 flowers. When several fertilizer doses were used, the number of flowers has increased from 23.00 (71N, 42P, 113K) to 28.02 (113N, 69P, 182K). The differences between the five treatments have been of 6.56 – 11.58 flowers from significant to very significant positive. The best results were given by the 113N, 69P, 182K variant which gave the maximum number of flowers (28.20) and a very significant positive difference over the control variant.

The combined influence of the two factors emphasizes the following aspects (Fig. 3, Table 1):

- with the 12 treatments the blooming capacity of *Saintpaulia ionantha* widely ranged from 15.88 flowers/plant (red peat without fertilizers) to 30.17 flowers/plant (black peat, Siforga fertilizer -113N, 69P, 182K)
- on black peat + leaves earth + sand culture background the number of flowers with all 5 treatments of Siforga fertilizer was between 24.33 and 30.17 with 127N, 78P, 205K variant and, respectively, 113N, 69P, 182K variant;
- on red peat + leaves earth + sand culture background the Siforga doses have determined 19.66 flowers (71N, 42P, 113K) to 25.88 flowers/plant (113N, 69P, 182K).

The result is the Uzambar violet can be stimulated to form more flowers by cultivating on a culture background formed of two parts black peat + one part leaves earth + one part sand and fertilization with Siforga organic fertilizer in 113N, 69P, 182K dose (Osiceanu unpubl.).

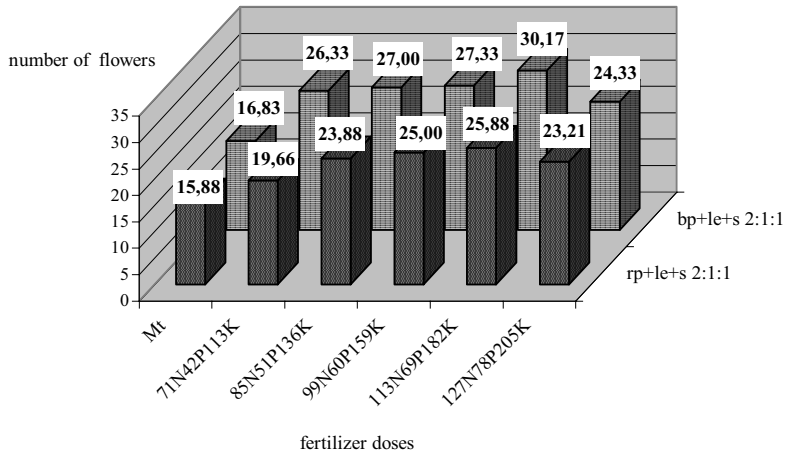

Figure 3 The combined influence of the culture background and the Siforga fertilizer doses on the number of flowers with *Saintpaulia ionantha* (average 2006-2008)

Table 1

The calculus of multiple comparisons on the combined influence of the culture background and Siforga fertilizer doses on the number of flowers of *Saintpaulia ionantha* (average 2006-2008)

Nr crt	Nr. flowers	Combination	a_2b_1	a_1b_1	a_2b_2	a_2b_6	a_2b_3	a_1b_6	a_2b_4	a_2b_5	a_1b_2	a_1b_3	a_1b_4
1	30,17	a_1b_5	14,29***	13,34***	10,51**	6,96*	6,29*	5,84*	5,17*	4,29$^-$	3,84$^-$	3,17$^-$	2,84$^-$
2	27,33	a_1b_4	11,45***	10,50**	7,67*	4,12$^-$	3,45$^-$	3,00$^-$	2,33$^-$	1,45$^-$	1,00$^-$	0,33$^-$	-
3	27,00	a_1b_3	11,12***	10,17**	7,34*	3,79$^-$	3,12$^-$	2,67$^-$	2,00$^-$	1,12$^-$	0,67$^-$	-	-
4	26,33	a_1b_2	10,45**	9,50**	6,67*	3,12$^-$	2,45$^-$	2,00$^-$	1,33$^-$	0,45$^-$	-	-	-
5	25,88	a_2b_5	10,00**	9,05**	6,22*	2,67$^-$	2,00$^-$	1,55$^-$	0,88$^-$	-	-	-	-
6	25,00	a_2b_4	9,12**	8,17*	5,34*	1,79$^-$	1,12$^-$	0,67$^-$	-	-	-	-	-
7	24,33	a_1b_6	8,45*	7,50*	4,67*	1,12$^-$	0,45$^-$	-	-	-	-	-	-
8	23,88	a_2b_3	8,00*	7,05*	4,22$^-$	0,67$^-$	-	-	-	-	-	-	-
9	23,21	a_2b_6	7,33*	6,38*	3,55$^-$	-	-	-	-	-	-	-	-
10	19,66	a_2b_2	3,78$^-$	2,83$^-$	-	-	-	-	-	-	-	-	-
11	16,83	a_1b_1	0,95$^-$	-	-	-	-	-	-	-	-	-	-
12	15,88	a_2b_1	-	-	-	-	-	-	-	-	-	-	-

DL 5% = 4,63 flowers; DL 1% = 8,67 flowers; DL 0,1% = 11,06 flowers;

CONCLUSIONS

The researches on the influence of Siforga organic fertilizer on the blooming capacity of *Saintpaulia ionantha* H. Wendl. have demonstrated that the black peat + leaves earth + sand 2:1:1 culture background give good results in comparison with the one based on red peat. These results refer to the number of flowers per plant.

The optimal doses of Siforga that ensured the maximal number of flowers (26.16 – 28.20 on average) have been 99-113N, 60-69P, 158-182K. With this treatments the differences over the not fertilized variant were very significant.

An overall analysis of the effect of Siforga fertilizer on the blooming capacity of *Saintpaulia ionantha* H. Wendl. species show that the best results are given by average fertilizer doses, the higher or lower doses have given superior results yet not significant.

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TWO VARIANTS OF THE USE OF STARTER YOGURT CULTURES TO OBTAIN OF CEREAL-BASED FERMENTED BEVERAGES

Pacala Mariana-Liliana[1], Brudiu Lucica[2], Lengyel Ecaterina[1], Begea Mihaela[3]

Key words: *cereal mashes, lactic acid bacteria, yeast, fermentation*

ABSTRACT

The main objective of this research was to study and compare the effect of using of the two starter yogurt cultures (2, 3(+/-0.01%w/w dry wheat beer yeast) and 4%w/w) on the main physical-chemical characteristics (pH, total and volatile acidity, alcohol concentration and dynamic viscosity) of the complex fermented cereal mash. The mash was produced from debranned white millet, maize, wheat and barley malt grists and wheat bran and water (initial ratio cereal ingredients:water=1:12) by boiling, filtering and adding of 10%w/w white crystalline sugar. Coarse filtered sweet mash was fermented at 42°C for Lactoferm® Natur Yogurt (NY) culture and at 40°C for Lactoferm® Bifidus Yogurt (BY) culture for 15h and for these both variants followed by a cold maturation at 6°C for 75 h. It was found that addition of dry wheat beer yeast determine obtaining a high alcohol concentration and volatile acidity without negatively influence on the dynamic viscosity. Inoculum of 2%w/w is recommended for both variants of fermentation. If using yeast then it is recommended BY cultures.

INTRODUCTION

Production of cereal-based fermented beverages (*C-BFB*) represents a successful valorization of nutritional and functional potential of cereals and a research topic of interest (Blandino A. et al., 2003, Mollendorff von. J.W., 2006); researchers are increasingly concerned over scientific explanation of beneficial health properties of these beverages (Stancu C.S. et al., 2012), considered as traditional for a series of geographical areas in Europe, Africa and Asia (Todorov S.D. et al., 2008; Das A. et al., 2011). By fermenting of cereals mashes using different microorganisms (lactic acid bacteria, probiotic bacteria, yeasts) in single or mixed culture, or complex cultures can diversify the probiotic food category, with special sensory properties, but also with functional role in case of using of cereals (Botes A. et al., 2007; Rathore S. et al., 2011). In the most cases were study simple cereal mashes, composed of one or two types of grain, spontaneously fermented or using a culture from a single strain of lactic acid bacteria (Rathore S. et al., 2011). The main aim of this work is to made a preliminare investigation of the effect of using a starter natur and

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bifidus yogurt culture at different amounts of inoculum for the fermentation of one mash produced from debranned white millet, maize, wheat, barley malt grists and wheat bran and water.

One series of fermentations is made with addition of dry wheat beer yeast. Because, the consumers do not approve of sour or too acidic products (Rathore S. et al, 2011) to improve the sensory characteristics of cereal-based fermented beverages in the research was used *Pilsner* malt barley. Furthermore, due to consumer concern for a healthy diet and consumption of products with a high content of dietary fiber, it is justified the option to add in the manufacturing recipe, the wheat bran (Arici M. et al., 2002; Charalampopoulos D. et al., 2002; Das A. et al., 2011).

MATERIALS AND METHODS

To achieve the experimental matrix were chosen two variants of the fermentation of cereal-based mashes obtained using the technological chart described by Pacala et al., 2012, but with several modifications (final dilution is 1:9 w/w, adding of 10% w/w white crystalline sugar, total time of fermentation is 90 h):

-with starter natur yogurt culture (*Lactoferm*® - dry natur yogurt culture from *Brouwland*, Belgium; containes strains *Streptococcus thermophilus* and *Lactobacillus bulgaricus*; variant denoted by *NY*) for 15 h at 42°C and

- with starter bifidus yogurt culture (*Lactoferm*® - dry bifidus yogurt culture from *Brouwland*, Belgium; containes strains *Streptococcus thermophilus*, *Lactobacillus acidophilus* and *Bifidobacterium infantis;* variant denoted by *BY*) for 15 at 40°C. Starter yogurt cultures are activated using whole sterilised milk according to the instructions of use. Quantities of inoculum used for each variants of starter yogurt culture were: 2, 3 and 4%w/w. For the inoculum of 3% w/w was selected, also, for a comparative series the addition of 0.01% w/w dry wheat beer yeast (*Safale wb-06* from *Fermentis*, Division of S.I. *Lesaffre*, France; manufactured in Belgium). Cereals grinding was done with *Bühler*'s Universal Laboratory Disc Mill, type *DLFU* set to 0.1 mm disc gap for debranned millet and set to 0.4 mm disc gap for barley malt and wheat. The ratio between of used grist quantities to obtain the mash is debranned white millet (*Panicum miliaceum,* S.C. *Sano Vita* SRL, Rm. Valcea/Romania, Ukraine) : degermed extra corn flour (*Zea mays* var. *amylacea*, S.C. SamMills Distribution SRL) : whole *Spelta* wheat (*Triticum spelta*, S.C. *Solaris Plant* SRL, Romania) : *Pilsner* barley malt (*Weyermann* Specialty Malting, Germany) : fine wheat bran (S.C. *Sano Vita* SRL, Rm. Valcea, Romania) = 3:1:1:3:2 w/w/w/w/w. Hardness of water is less than 5°dH. Cereal-based fermented mashes was evaluated according to the analitical methods described in the specialty literature and in the previous researches (Ergun K. et al., 2003; Hayta M. et al., 2001; Kedia G. et al., 2007; Pacala M.-L. et al., 2012): - pH: at 20 °C using a pH-meter Orion 2 STAR (*Thermo Electron Corporation*, Ltd.) with calibration in 2 points; - total acidity (% w/w, g lactic acid per 100 g of mash): by titrating of mashes with 0.1 N NaOH in the presence of bromothymol blue indicator, alkaline solution 4‰; - volatile acidity (% w/w, g acetic acid per 100 g of mash): by titrating of distillates of mashes with 0.1N NaOH in the presence of bromothymol blue indicator, alkaline solution 4‰; - alcoholic concentration (% w/w): analyzing of corresponded distillates (obtained with *Raypa*'s *Alcotest-1* (*R. Espinar*, S.L.), the semiautomatic distiller with steam-drive) using direct method with the *Alkotest Analyzer* alcoholtest (*Bulteh* 2000, Ltd.). Volatile acidity of distillate must be less than of 0.1 g acetic acid/100 mL of distillate; - dynamic viscosity (cP): at 20°C using a Yield Stress Rheometer *Brookfield* YR-1 (*Brookfield Eng.Labs.*, Inc.) on mode for calibration, equipped with a

spindle S72 at 100 RPM and connected at a refrigerated circulating water bath *Brookfield* TC-502 (*Brookfield Eng. Labs.*, Inc.)). Statistical analysis: data are shown as mean (n=3) ± SD (Standard Deviation) and represent the experimental results of three independent fermentations with three replicates each.

RESULTS AND DISCUSSIONS

The evolution of total and volatile acidity, alcohol concentration and dynamic viscosity for studied variants are comparatively shown in Figure 1, 2, 3 and 4. The values of pH during fermentation of cereal mashes are shown in Table 1.

Table 1

The evolution of pH* of mashes for the experimental matrices (2 variats x 4 series)

Duration of the fermentation		Type and quantity of innoculum of Starter Yogurt Cultures (*SYC*)							
		2% w/w of *SYC*		3% w/w of *SYC*		3% w/w of *SYC* and 0.01% w/w of yeasts		4% w/w of *SYC*	
Code	U.M.	Series no. 1		Series no. 2		Series no. 3		Series no. 4	
		1-NY	*1-BY*	*2-NY*	*2-BY*	*3-NY*	*3-BY*	*4-NY*	*4-BY*
		pH*							
0	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
t_1	0	5.88	5.68	5.88	5.68	5.88	5.68	5.88	5.68
t_2	15h30'	3.57	3.45	3.47	3.49	3.35	3.37	3.44	3.42
t_3	38h30'	3.87	3.72	3.79	3.70	3.77	3.64	3.76	3.67
t_4	63h30'	3.75	3.64	3.60	3.61	3.50	3.53	3.66	3.51
t_5	90h	3.63	3.48	3.61	3.61	3.62	3.45	3.52	3.49

* - mean values (n=3); - for all determination *SD* (Standard Deviation) was under 2%.

Analyzing the graphs from Figure 1 it is found that the total acidity shows higher values for all determinations made at 15h and 30 min., 38 h and 30 min., 63 h and 30 min. and 90 h from the beginning of fermentation for samples *1-BY* si *2-BY*. In the case of *3-BY* sample after 50 h of fermentation and of *4-BY* sample after 40 h of fermentation the evolution of total acidity is reversed in favor of series denoted with *NY*. Total acidities correspond to the data obtained by the de Arici M. et al., 2002 and Kose E. et al., 2003. In the case of series no. 4 the total acidity increases more slowly for *4-NY* and get to corresponding values only after 40 h of total fermentation duration. For series no. 4, also, the gradients of other physical-chemical analyzed parameters are smaller, but *4-BY* samples evolve better. The graphs from figure 2 reveals that for samples of all series *NY* volatile acidity is higher for, approximately, entire period of fermentation. Thus, considering the volatile acidity is recommended to use the starter bifidus yogurt culture inoculum for the fermentation of studied cereal mashes and for this variant of the fermentation the samples with *1-NY* and *3-NY* are optimal, adding to the interpretation of results also the values obtained for alcoholic concentration and dynamic viscosity. By the interpretation of graphs from Figures 3 and 4 it can by said that for quantities of ethyl alcohol accumulated in samples, the dynamic viscosity is not significantly influenced, dynamic viscosity values being similar to those from scientific literature, for these four series of samples (Hayta M. et al., 2001). For small amounts of inoculum (2 and 3% w/w) is a more rapid increasing of dynamic viscosity, if starter culture is *NY*. At an inoculum of 4% w/w representation of those two curves of dynamic viscosity, corresponds to evolution of volatile acidity of samples. Considering, also, values of total acidity for this series and that it is a mainly lactic acid, can say that at higher concentrations of inoculum the cultures *NY* is recommended for a specific volatile acidity of these drinks.

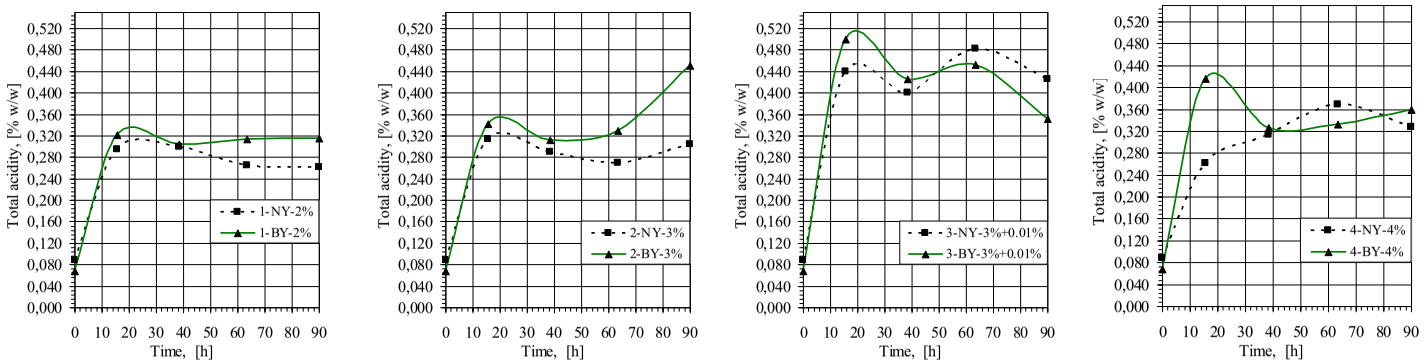

Figure 1 The dynamic of total acidity of those four series of cereal mashes during of the total period of fermentation (90 hours)

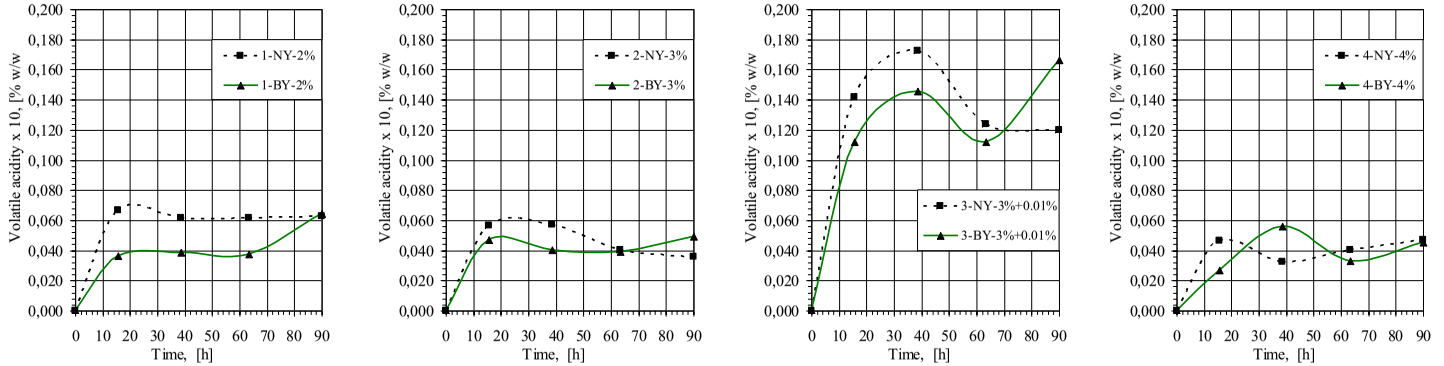

Figure 2 The dynamic of volatile acidity of those four series of cereal mashes during of the total period of fermentation (90 hours)

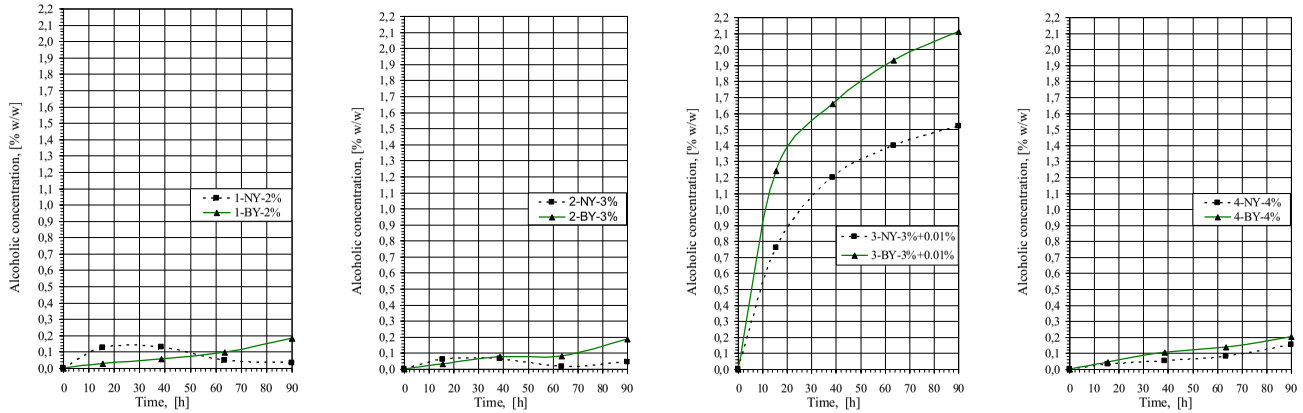

Figure 3 The dynamic of alcoholic concentration of those four series of cereal mashes during of the total period of fermentation (90 hours)

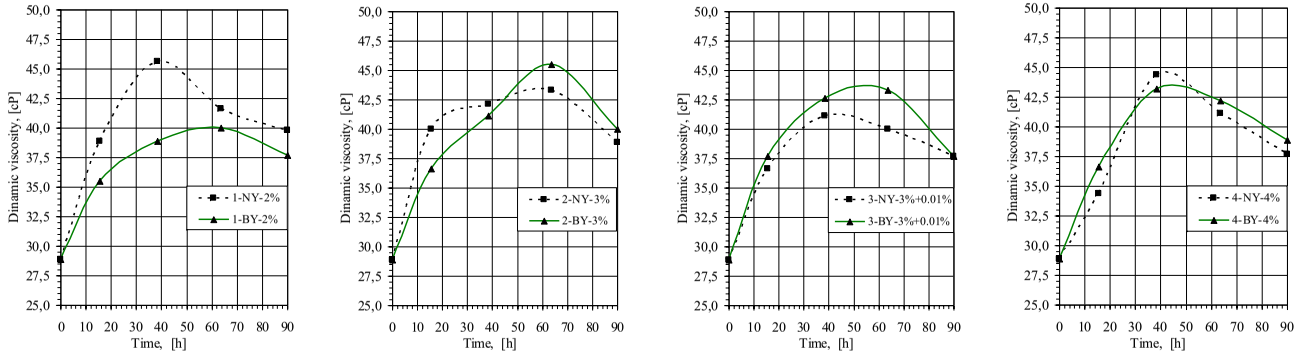

Figure 4 The dynamic of dynamic viscosity of those four series of cereal mashes during of the total period of fermentation (90 hours)

CONCLUSIONS

Using the addition of yeast (*3-BY* sample is considered optimal) allows obtaining of *C-BFB* with a higher content of alcohol and volatile substances, without negatively impacting on dynamic viscosity. May be considered sufficient a total duration of fermentation around 40 h for series no. 1, no. 2 and no. 3 and around 25 h for series no. 4 of both variants of fermentation for the condition of *shelf-life*. If using only yogurt starter culture, is evaluated as optimal series with the inoculum of 2%w/w, for both variants of fermentation.

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THE GROWTH AND DEVELOPEMENT OF APPLE TREES ON DIFERENT ROOTSTOCKS IN THE FRUIT NURSERY

Ananie Pesteanu[1]

Keywords: *Apple, Varieties, Rootstock, Two-year trees, Bench-graft. Fruit Nursery, Planting material*

ABSTRACT

Investigations were conducted during the period of 2008-2009 years in Fruit Nursery of Company „Codru-ST" Ltd., which is located in the centre of Moldova. As objects of the investigation served three apple varieties: Gala Must, Golden Reinders and Idared, witch were bench-grafted on five rootstocks such as: M 9, 62-396, M 26, M 7 and MM 106. Planting distance was 90x35 cm.

It was established, that in the first and second fields of the fruit nursery, the main indicators of apple tree growth manifest significant increases depending on the increase of rootstocks' vigor of growth used in the process of grafting and the evidence obtained corresponds to the current standards.

INTRODUCTION

One of the main apple crop increased, including the Republic of Moldova, is the establishment and efficacious exploitation of intensive and superintensive orchards, used the grafted trees by small and medium vigor rootstocks led by well-structured system of crown, which can ensure the early economical fructification once two-three year after planting in the orchards (Babuc and Rapcea, 2002, Pesteanu, et al. 2010).

From the experience of the countries with a developed fruit growing – Italy, Holland, Poland, etc., the superintensive apple trees are established with crowned apple trees produced during the period of two years (Gudumac, et al. 2007, Peşteanu, 2007, Petre, et al. 2006).

The crowned apple trees in the fruit nursery, being planted in the orchard, have an early fruit production and increase more rapidly the fruit production in comparison with the planted trees without crown (Bielicki and Czynczyk 1994, Mika, et al.2003, Sadowski, et al. 2005).

Of recent research (Gudumac, et al. 2007, Pesteanu, et al. 2010), on crown

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structure in second field of the nursery (of apple trees grafted by M 9 rootstock) was elaborated a mixed method which consists in the formation of base of crown by four normal (grown from last year buds), well-developed longer than 60 cm branches, and to the shoot axis extension two-three early branches obtained of early buds.

It is very important number, distribution uniformity and length of the shoots on the central axis bazitonic strict compliance with the principle, which are large determined by biological features of varieties (Ghena, et al. 2004).

Taking into the consideration that the determinative factor of having an early fruit production in the orchard constitute the type and quality of the planting material, the investigations had the aim to determine the development of apple trees, grafted on different types of rootstock for the apple tree plantations (Babuc, et al. 2009, Petre, et al. 2006).

For these requirements it is necessary to develop and practice the formation methods of the crown, suitable with features of variety-rootstock associations, beginning in the fruit nursery, the formation of trees crown base.

MATERIAL AND METHODS

The research was carried out during 2008-2009 in the fruit nursery of company „Codru-ST" Ltd., which is located in the central area of Moldova, research items were used for apple varieties: Gala Must, Golden Reinders and Idared bench-grafted on M 9, 62-396, M 26, M 7 and MM 106 rootstocks.

The bench-grafting was performed in March, using the perfected copulation method with detached branch. Grafting site was tied with porous polyethylene tape designed specifically for graft and graft was paraffined. The obtained graftings were stratified by placing them upright in containers, so that the basal layers (20-25 cm) to be placed in a layer of wet sand. The stratification temperature in the refrigerator was +2...+4^{0}C. To produce grafted trees were used well-developed layers of 10 mm diameter and graft branches with higher biological values.

The first field of tree nursery was established in the second half of April, with bench grafts. Distance of planting grafted plants was 90x35 cm. The aerial part was palisated on a stick of bamboo.

In the second field of nursery, early spring annual stems have been shortened to a height of 75-80 cm above the grafting site. During the vegetation was carried trunk release, being left only 4-5 shoots to form the crown base. To obtain sylleptic shoots on the central axle, when they reached the length of 15-20 cm, it was made the remove of apical leaves without hurting the point of growth. This operation is repeated every 5-7 days for 5-6 times. To stimulate the strong development of shoots are made more frequent irrigation and fertilization based on macro-and micronutrients.

The usual black soil, the content of humus is 2,6%, that is maintained as cultivated field, irrigation is made by sprinkling keeping the soil wet at 75-80% from the capacity of field.

The number of repetition in each variant is 4. The number of trees in each repetition is 20. The researches were made in field and laboratory conditions according to the required methods for doing experiments with fruit growing plants. The main results obtained were statistically processed.

RESULTS AND DISCUSSIONS

On the basis of the results obtained it was demonstrated that the type of the roostock has an influence on apple tree's growth in the fruit nursery.

The degree of striking the bench graftings in the first field of the fruit nursery during the period of investigation (fig. 1), for all the rootstocks taked into the study, is between the limits of 95,6-99,2 %.

In 2008 the highest degree of striking was registered by the Gala Must variety that was grafted on MM 106 rootstock, being of 99,1%, and the lowest striking degree – by the Idared variety , being grafted on M 9 rootstock (95,6 %).

In 2009, the appropriateness exposed previously remains. The varieties Gala Must and Golden Reinders, have the identical the degree of striking, reducing most only on M9 rootstock. The variety Idared, in 2009, the degree of striking is higher as in 2008, but lower than the varieties Gala Must and Golden Reinders.

Having a more detailed study of this indicator, it may be observed that a lower value was registered at the investigated varieties which were grafted on rootstocks M 9 and M 26. This is due to the fact that this rootstock have already formed a smaller quantity of roots in comparisson with the rootstocks 62-396, M 7 and MM 106, that to their hereditary characters form a bigger quantity of roots (Adăscăliţei, et al., 2004, Babuc, et al. 2009, Ghena, et al. 2004).

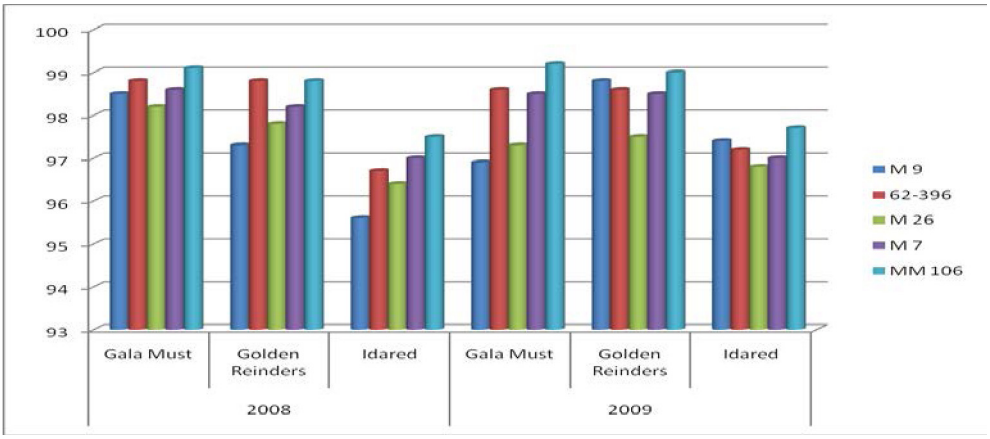

Fig. 1. The degree of striking bench graftings planted in the first field of the fruit nursery, %.

At the end of the first period of vegetation in the first field of the fruit nursery it was established that the height of trees (tab. 1) for all the varieties and types of rootstocks the investigations were corresponding with the limits of 111-135 cm in 2008 year and 109-125 cm in 2009.

Table 1

Growth main indicators of apple trees in the first field of the fruit nursery depending on the rootstocks type

Rootstock	Variety					
	Gala Must		Golden Reinders		Idared	
	graft height, *cm*	graft diameter, * *mm*	graft height, *cm*	graft diameter, * *mm*	graft height, *cm*	graft diameter, * *mm*
2008 year						
M 9	124	8,9	113	8,3	111	8,6
62-396	126	9,0	120	8,5	115	8,7
M 26	128	9,6	123	8,9	117	8,9
M 7	131	10,0	124	8,9	118	9,0
MM 106	135	10,2	129	9,4	124	10,0
$Dl_{0.05}$	*1,98*	-	*3,03*	-	*2,86*	-
2009 year						
M 9	114	9,4	111	8,9	109	8,7
62-396	116	9,8	112	9,4	114	8,8
M 26	117	9,8	117	9,5	114	9,2
M 7	120	10,0	117	9,6	115	9,5
MM 106	125	10,1	120	10,0	116	9,7
$Dl_{0.05}$	*2,75*	-	*2,40*	-	*3,01*	-

*- at 10 cm above the graft's place

The highest values of graft height were recorded in both years by Gala Must variety grafted on MM 106 rootstock, which has a medium force of growth and is 125-135 cm. The Golden Reinders variety, grafted on the same rootstock, registered intermediary values of this index, being 120-129 cm. At the Idared variety, the highest value of this index was recorded at the same event, being 116-124 cm. With decreasing growth vigor of rootstocks studied, there is a decrease in the value of this index and is 109-111 cm at the Idared variety grafted on the rootstock M 9, and 111-113 cm at the Golden Reinders variety, 114-124 cm at the Gala Must variety grafted on the same rootstock.

The graft diameter at 10 cm above the graft's place in the years 2008-2009, which is affected by increasing the vigor of the rootstocks and varieties investigated was within the limits of 8,3 mm and 10,2 mm. As the graft's height, the diameter increases at the same time with the increase of vigour of growth of the investigated rootstocks.

The leaf surface (fig. 2) grows at the same time with the increase of growth vigor of rootstock from 0,20-0,27 m^2/tree to 0,26-0,35 m^2/tree.

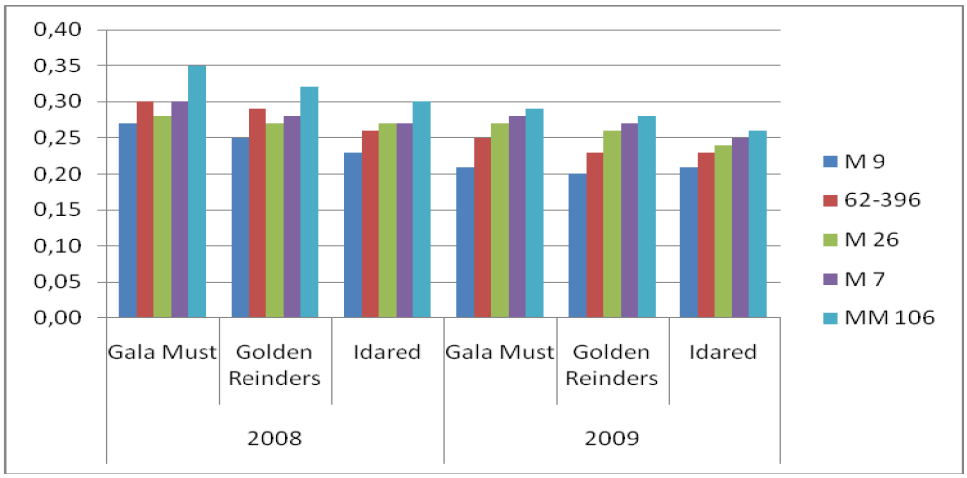

Fig. 2. Leaf surface of apple trees in the first field of the fruit nursery depending on rootstock type, m^2/tree.

At the end of the second period of vegetation in the first field of the fruit nursery it was established that the height of the fruit trees (tab. 2) grows consequently with the vigour of the investigated rootstocks to the limit of 190,25-194,75 cm or with 3-13 %.

The trunk diameter above with 10 cm from the graft's place as an integral indicator, in 2009 had higher values at the grafted trees on rootstock M 7 (16,3-17,0 mm) and respectively the grafted trees on the rootstock M 9, the lowest diameter was of 15,0-15,5 mm due to the fact that this rootstock give the lowest vigour of growth to the fruit trees.

The number of normal leaves formed at the base of the crown at the varieties taken into the study in the second field of the fruit nursery is between the limits of 3,5-4,5 pcs/tree.

The average length depends on the biological peculiarities of the varieties and rootstocks taken into the study and, also their number, so as in the second field of the fruit nursery the values of this indicator is between the limits of 67,24-92,88 cm.

The longest lenght of normal shoots was registered at all the investigated varieties, grafted on rootstock M 7. The average length of normal shoots was registered at the apple trees grafted on the rootstocks MM 106, M 26 and 62-396.

The number of sylleptic shoots formed from early buds on the extension shoot of the axle, at the varieties taken into the study, depends greatly on the variety's capacity to emit sylleptic shoots, and the vigor of growth of the rootstock researched.

The most pronounced hereditary capacity to form sylleptic shoots on the central axle was registered at the Gala Must variety – 3,50-7,50 pcs/tree with an average length of 23,00-33,39cm, followed by the Golden Reinders variety with 3,50-6,25 pcs/tree with a length of 26,5-43,88 cm and, respectively, by Idared variety with the lowest hereditary capacity to emit sylleptic shoots that had formed 1,75-5,25 pcs/tree with their average length of 31,49-37,96 cm.

If to compare the above mentioned indicators according to the vigour rootstock type, it may be observed that the rootstocks M 7 and MM 106 have formed the greatest number of sylleptic shoots in comparison with the rootstocks M 9 and 62-396 with a low vigour of growth.

Table 2

Growth main indicators of apple trees in the second field of the fruit nursery depending on rootstock type, 2009

Rootstock	Tree height, *cm*	Trunk diameter,* *mm*	Crown dimensions			
			normal branches		sylleptic shoots	
			number, *pcs/tree*	average lenght, *cm*	number, *pcs/tree*	average lenght, *cm*
Gala Must variety						
M 9	170,00	15,00	4,50	80,00	3,50	23,00
62-396	176,50	16,33	4,50	73,13	4,00	30,75
M 26	191,25	15,65	4,25	78,78	4,75	33,39
M 7	186,25	16,33	4,25	86,50	7,25	28,00
MM 106	193,75	16,25	4,50	82,94	7,50	31,98
LSD $_{0,05}$	*4,32*	-	-	*2,72*	-	*3,76*
Golden Reinders variety						
M 9	184,50	15,33	4,00	75,63	3,50	35,75
62-396	185,00	17,00	3,50	72,85	3,50	34,50
M 26	191,50	16,50	4,00	81,88	3,75	43,88
M 7	185,00	17,00	4,00	92,88	6,25	26,85
MM 106	194,75	16,13	4,25	74,25	6,00	44,47
LSD $_{0,05}$	*7,71*	-	-	*6,74*	-	*2,79*
Idared variety						
M 9	186,25	15,50	4,00	67,24	4,25	34,38
62-396	182,50	15,68	3,75	72,50	1,75	37,16
M 26	190,00	16,33	4,00	80,30	3,00	32,50
M 7	192,00	16,30	4,00	83,44	4,00	37,96
MM 106	190,25	16,25	4,00	75,94	5,25	31,49
LSD $_{0,05}$	*4,77*	-	-	*5,09*	-	*1,68*

*- at 10 cm above the graft's place

The leaf surface of apple trees in the second field of the fruit nursery (fig. 3) is majored concomitantly with the increase of rootstock vigor of growth, from 0,71-0,82 m^2/tree in the case when the varieties taken into the study were grafted on M 9 to 0,89 m^2/tree at the trees of Idared variety grafted on rootstock M 26, 0,95 m^2/tree at the variety Golden Reinders and 1,00 m^2/tree at the Gala Must variety, both grafted on rootstock MM 106, or with 18-30%.

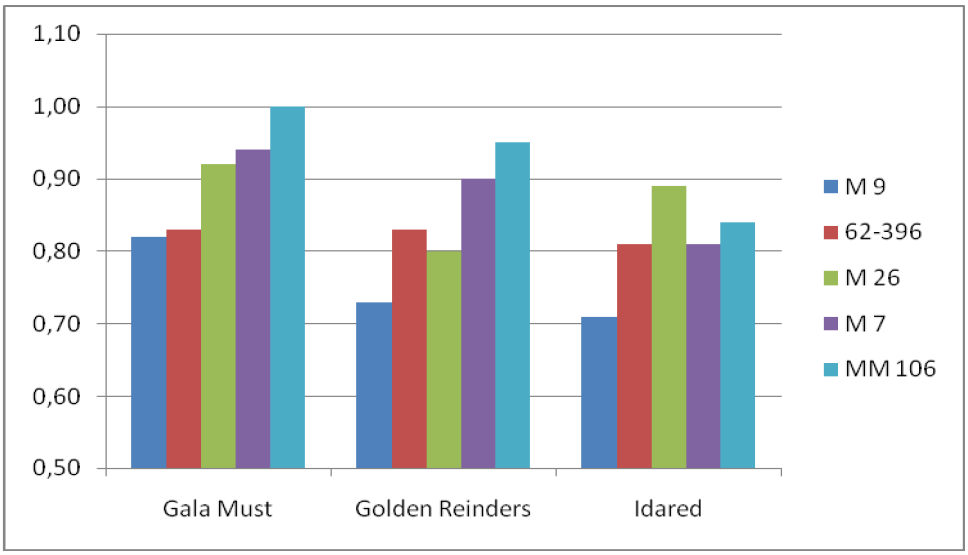

Fig. 3. Leaf surface of apple trees in the second field of the fruit nursery depending on rootstock type, m^2/tree, 2009.

On the basis of the researches made and according to present standards of the Republic of Moldova (SM 155:2003), it was found that the apple trees obtained on different variety-rootstock combinations in the second field of the fruit nursery correspond to first category quality.

CONCLUSIONS

1. The degree of striking and starting to grow of the bench-graftings is very high, being between the limits of 95,6-99,2%.

2. The principal indicators of apple tree growth in the first and second fields of the fruit nursery demonstrate significant increases depending on the increase of rootstocks vigor of growth that were used in the process of grafting;

3. The parameters of apple trees in the second field of the fruit nursery had registered values that correspond to 1st category of quality according to the present standards on: height, trunk diameter, number of branches, as well as their average length;

4. The biological peculiarities of the types of investigated rootstocks influence apple trees growth and development in the nursery. It is recommended to use for the apple tree superintensive system grafted apple trees on rootstocks M 9, 62-396 and M 26, and for the intensive system – more suitable are considered to be the rootstocks M 7 and MM 106;

5. Superior values of the main indicators of apple tree's vigor of growth in the first and second field of the fruit nursery were registered according to the varieties under the investigation, at the varieties Gala Must and Golden Reinders – varieties of perspective, being followed by Idared – a homologated variety in the Republic of Moldova.

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STUDIES ON THE MULTIPLICATION OF TWO NEW FRUIT-GROWING SPECIES, *ACTINIDIA DELICIOSA* AND *ACTINIDIA ARGUTA*

Peticila A.G.[1], Stanica F.[1], Oana Venat-Dumitriu[1], Roxana Madjar[2]

Key words: *Actinidia arguta, Actinidia deliciosa, kiwi plant, micropropagation, European funds*

ABSTRACT

The research on the multiplication of kiwi varieties and hybrids is conducted by the Micropropagation Laboratory within the Horticulture Bioengineering Resources Department of the University of Agronomical Sciences and Veterinary Medicine of Bucharest. Biological material used during observations was represented by 2 (two) varieties: Hayward, Tomuri from Actinidia deliciosa and other 2 (two) hybrids from Actinidia arguta which are direct descent from cultivars and hybrids from Faculty of Horticulture kiwi plants collection. The in vitro culture was ensured in a hormone-free MS environment in three variants: classic MS, MS with 2x NH4NO3 and MS with 3x NH4NO3. Ethyl alcohol, 70% was used for disinfection purposes for 5 seconds, as well as sodium hypochlorite, 10%for 2 minutes, followed by 3 flushing cycles with sterile distilled water. The culture initiation had a 95%success rate and is currently in the multiplication stage.

INTRODUCTION

Due to its many benefits, *Actinidia* succeeded in getting a 'competitive edge' over other exotic fruits and in conquering new lands, regions and fans. Fruits can be eaten fresh or stored up to 6 months or used by the food industry for the preparation of marmalades, sauces, jams and syrups. All these beneficial features of the fruit call for the extension of *Actinidia* –growing areas in our country. Due to the quite large area occupied by this plant type and to the various geographical and weather conditions in the areas of origin, these plants have a good capacity to adapt. *Actinidia* has often been compared to the peach in terms of growing area and soil requirements since it is more demanding than the grape vine. Its growing area is limited to a land strip with the latitude coordinates 34° to 46° north and 30° to 42° south. Romania is located on the northern boundary of the *Actinidia* growing area and offers the proper climate for the cultivation of this variety.

There are regions in Romania which favour the cultivation of *Actinidia*, for instance the mild micro-climates where the peach, the apricot and the almond trees grow, e.g. the first terraces of the Danube Meadow ('Lunca Dunării'). *Actinidia arguta*, which

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proved to be more resistant at low temperatures, may be cultivated by farmers in almost all the regions of Romania, with outstanding crop figures in the first years due to the minimum protection required by the plant during winter. These studies focus on the introduction of *Actinidia arguta* and *Actinidia deliciosa* on the list of species cultivated in Romania and rely on complex scientific methods and in-depth study of the plant's reaction to environmental conditions, of propagation techniques and improvement of technologies to obtain young and healthy plants.

The first information on *Actinidia* were published in 1821 by the Danish researcher Nathaniel Wallich who initiated his studies on the *Actinidia callosa* sample bought from Nepal. This new cultivar was included by former authors in the *Dilleniaceae* family. In 1899, Van Tieghem suggest the creation of the *ACTINIDIA*CEAE family (Zuang H., Musard M., 1984)

Actinidia is a dioecious unisexual plant, i.e. has separate pistiliferous flowers (female) and staminiferous flower (male) plants. Fruits resulting from entomophilic pollination are berries of various shapes according to variety. They may be brown and covered or not in stiff fine hairs. The perfumed and sweet sour kiwi flesh may have different colours from bright green to red. Fruits weight may vary from 60-70 g to 90-200 g. The production rate/plant may reach 65-80 kg.

Only 4 species of *Actinidia* are of interest for the production of fruits: *chinensis*, *deliciosa*, *kolomikta* and *arguta.* These species may be cultivated on the fruit-growing lands of Romania.

Actinidia deliciosa (also known as 'kiwi') is pest and disease resistant and requires little pesticide treatment. This is the main benefit prompting the development of integrated kiwi cropping system. The long oval-shaped fruits of *Actinidia deliciosa* are covered with brownish yellow dense and long-lasting hairs. The flesh may vary in colour from yellowish green to emerald green.

*Actinidia arguta (*also known as 'baby kiwi'*)* is considered to be quite heteromorphic. Various cultivars different by shape, colour of the seed vessel and of the mesocarp and flavour are found in the growing areas and classified into 5 botanical varieties: *arguta*, *nervosa*, *purpurea*, *giraldi*, *cordifolia* (Sieb., Zuccherelli 1994). In terms of phenology, *Actinidia arguta* is a woody wine similar to *Actinidia deliciosa,* however less vigorous. It has smaller and brighter finely serrate leaves than the kiwi plant. The size and shape of leaves is common to almost all varieties. It produces bright hairless fruits which are eaten without peeling it off. The fruits usually vary in colour from green to dark green and may bear different shapes: long, round or oval. The fruit flesh is green and of variable intensity much similar to that of the kiwi fruit. Its sour but sweet taste bears the kiwi-like flavour.(Geoof, Rodd, Segall, Turner jr., Wasson, 2001)

Due to its many benefits, *Actinidia* succeeded in getting a competitive edge over other exotic fruits on the Romanian marketplace and in conquering new lands, regions and fans.

Fruits can be eaten fresh and may be stored up to 6 months. Kiwi fruits are called 'Natural Health Capsules' due to the vitamin C content equalling that of 5 lemon and 2 orange fruits. The vitamin E content equals the avocado fruit with 60% of the calories. The carbohydrates content is similar to banana, however with 40% of the calories. The water content is 81.8% and the sugar content is 11.7%. The energetic value of a fruit is 48 kcal. (Ferguson, 2011). Moreover, the kiwi fruit is rich in iron (3.7 mg./180 g) and is one of the richest sources of Calcium (30 mg./100 g). The kiwi fruit also contains other alkaline elements (K, Mg) and organic acids (apple, citric, tartaric acids) that help regulate the acid-base balance of the body. The nutrients per 100 g. edible part of the kiwi fruit are listed in

Tabel 1.The presence of regions providing proper climatic conditions is another major factor favouring kiwi cultivation in Romania. Due to the quite large area occupied by this plant varieties and to the various geographical and weather conditions in the areas of origin, these plants may easily adapt to other climatic conditions (Davidescu et al, 2010). *Actinidia* has been often associated to the peach in terms of growing area and soil requirements since it is more demanding than the grape vine. Its growing area is limited to a land strip with the latitude coordinates 34° to 46° north and 30° to 42° south. For as far as the warm temperature s concerned, the long cycle of vegetative growth (7 to 8 months) requires clearly-defined geographic boundaries. Our country, as already shown above, is located on the northern border of the *Actinidia* growing area (Zuccherelli 1994). Within the kiwi-growing areas of Romania, the rainfall depth is of about 200-250 mm which is not enough for Actinidia. The water deficit may be counterbalanced by irrigations. Micro-irrigation is the best method recommended also because it increases air humidity.

As far as the soil is concerned, *Actinidia* needs to be cultivated on clay and sandy soils built on drifty and permeable in-depth layers rich in organic substances (The Agricultural Atlas of Romania and so on). Water availability must be constant and not excessive and must not go below 70% of the field capacity. The soil pH value must vary between 5.5 and 7. Higher pH values may lead to issues in connection to the ferrous chloride with the same impact as the high content in active lime

In Romania, a commercial plantation of about 2 hectares was created in the Danube Meadow at Ostrov, as well as a collection of kiwi cultivars and hybrids for teaching and research purposes at the University of Agronomical Sciences and Veterinary Medicine (U.S.A.M.V.) of Bucharest, Horticulture and Vine Growing Department, in the period between 1993 and 1996.

The studies aim to analyse the kiwi breeding techniques and to improve existing technologies (Debersaques, Mekers, 2011) based on plants acclimated in growing areas in order to obtain hardy and healthy shoots in relatively short time while maintaining their genetic identity.

MATERIALS AND METHODS

In order to preserve the identity of clones, I used asexual production methods for *Actinidia* as for other fruit-bearing species, i.e. cutting propagation, grafting and micro propagation.

Since the industrial propagation method most used so far is the cutting propagation followed by micro propagation (Zuccherelli, 1994), these methods have been applied for the study on kiwi offsets obtained from the collection of cultivars and hybrids owned by the Faculty of Horticulture.

The rooting of herbaceous cuttings is carried out in a humid environment protected against slight tissue dewatering, fungi and bacteria. The results are largely conditional upon the nutritive condition of the mother plant, the time of material sampling, the method and type of hormones used to stimulate rooting, temperature, brightness and effectiveness of the moisture system, etc**.** (Zuccherelli, 1994)

The biologic material taken from *Actinidia deliciosa* cultivars Kramer, Hayward, Katiuscia, *Tomuri* and *AD20* hybrid and from *Actinidia arguta* cultivars Jumbo and Francesca was used for cutting propagation. The experimental study was conducted at U.S.A.M.V's Greenhouse in Bucharest. The plant rooting tab was used to root the cuttings

in perlite + sand substrate of 25 cm thickness. (Conte, Bevilacqua, Di Cintio, Terlizzi, Sartori, 2011)

Cuttings have been collected between June and July and the rough conversion was performed after harvesting on a section of 15-20 cm which corresponds to 3-4 internodes (Davidescu et al, 2001).. The upper part of cuttings was covered in paraffin in order to prevent dehydration. Hormone testing consisted in the flushing of various rooting stimulators at the foot, each of which being a different alternative:

- V1- Treatment with Radistim
- V2- Treatment with α naphtylacetic acid (ANA) of 2000 ppm.
- V3- Treatment with α indolyacetic acid (IBA) of 2000 ppm.
- V4- Treatment with ANA 1000 ppm and IBA 1000 ppm solutions.

Treatment was applied for 10 seconds. Cuttings supervision consisted in the definition of rooting rate per species, cultivars, hormone treatment and cutting propagation period, as well as of the characteristics of the resulting root system (Stănică, Peticilă, Davidescu, Dumitrascu, Majdar, 2003)

Apart from cuttings propagation, micro propagation is another kiwi fast-growing method. In-vitro propagation studies aims at adapting the above technologies to the conditions provided by the micro propagation laboratory of the Faculty of Horticulture within the USAMV University of Bucharest where experiments are conducted according to the research plan below: two cuttings were selected from the two species of interest, namely *Actinidia deliciosa* and *Actinidia arguta*; one was used as female genitor and the other as male genitor. The vegetal material needed to initiate growing consists in seedlings developed from vegetative buds.

The vegetal material used was represented by the plant R10P14 belonging to *Actinidia arguta* (male), R10P2 representing *Actinidia arguta* (female), R1P16 – representing *Actinidia delicioasa* (male) and R1P1 representing *Actinidia deliciosa* (female).

Three types of growing media standing for three experimental variants were prepared according to the reference bibliography. The first culture medium variant is the classic MS with unchanged components, the second is the modified MS medium with a double quantity of ammonium nitrate (2N) and the third variant is the modified MS medium with a triple quantity of ammonium nitrate (3N) in the composition of macroelement salt solutions. These changes result from the study of reference literature where the ammonium nitrate was modified in order to boost the vegetative growth of transferred plants.

We thus have for *Actinidia arguta* the male V 1- classic MS medium for seeding, V2- MS 2N for seeding, V3 MS 3N for seeding. After 2-3 follow-up months of the seeding phase, we pass to V4-MS classic propagation medium, V5-MS 2N for propagation, V6-MS3N for propagation, for a period of 6 months of follow-up and measurements. The variant V7 MS classic for rooting, V8 MS 2N for rooting and V9 MS 3N for rooting follow for a period of 2 months, and after that the acclimation of in-vitro plants. This experimental scheme will also apply for the female genitor of *Actinidia arguta* and for the two plants of *Actinidia deliciosa.*

The experiment started in January 2012 by the imposed planting of rows of seedlings according to the aforementioned study plan.

RESULTS AND DISCUSSION

The results obtained in the rooting of *Actinidia arguta* and *Actinidia deliciosa* after the green cuttings propagation are given in Table 2 and Table 3.

The analysis of the variant shows major differences according to the cultivar or hybrid to the same rooting stimulation treatment applied. The highest rooting rate over all other cultivars and hybrids used for rooting on sand + perlite substrate observed for the cultivar AA5 (Jumbo) of *Actinidia arguta*. No differences given by the type of cultivar/hybrid were observed during rooting stimulation with the ANA 2000 ppm product for *Actinidia deliciosa* and for cultivars Kramer and Tomuri for which the rooting rate obtained is of 37.50%.

Various differences are witnessed in the study of the same cultivar/hybrid tested during rooting with various mixes of rooting stimulators. The best rooting results are obtained with the mix of ANA+IBA 1000 ppm solutions, followed by ANA 2000ppm and IBA 2000ppm. The rooting stimulation with Radistim demonstrated that *Actinidia* is the last ranked compared to all other treatment variants using ANA and IBA solutions, as well as the ANA + IBA mix. The untreated variant, irrespectively of the type of cultivar or hybrid tested, shows the lowest rooting rate.

The output for the initiation of in-vitro cultures of *Actinidia arguta* and *Actinidia deliciosa*.

The review of Table 4 shows that the highest rate of success in initiating the culture of *Actinidia arguta* for the male plant (86.6%) was obtained by V2 (V2- MS 2N for seeding). while for the female plant (66.6%) by variant V1 media (V 1- MS classic) and V2 (V2- MS 2N for seeding).

The review of Table 5 shows that the highest rate of success in initiating the culture of *Actinidia deliciosa* for the male plant (60 %) was obtained by V3 (V3 MS 3N for seeding). while for the female plant (60%) by the V2 (V2- MS 2N for seeding) again.

The rate is calculated by taking into account that the disinfection protocol used was the same for all transferred plants and the most effective.

Studies undertaken to establish the micropropagation reproduction protocol are part of the European project and continue with the reproduction and rooting phases according to the initial research plan.

Table 1

The nutrients per 100 g. edible part of the kiwi fruit

Kcal.	61
KJ	254
Carbohydrates	18g
Fibres	1.1g
Proteins	10g
Water	83g

Table 2

Bi-factor experiment layout, a = variety/hybrid, b = treatment applied

a factor = variety/hybrid	b Factor = treatment applied
a1 = AA2 (Francesca) a2 = AA5 (Jumbo) a3 = AD20 a4= Hayward a5= Katiuscia a6= Kramer a7= Tomuri	b1 = untreated (control) b2= Radistim b3= ANA 2000 ppm b4 =IBA 2000 ppm b5= ANA + IBA 1000 ppm

Table 3

Influence of the variety/hybrid (a factor) and treatment applied (b factor) on the rooting percentage (%) at Actinidia sp. on substrate of sand and perlite

Species	a\b	% rooting				
		Experimental variants				
		V1 Untreated witness	V2 Radistim	V3 ANA 2000ppm	V4 IBA 2000ppm	V5 ANA +IBA 1000ppm
Actinidia arguta	AA2 (Francesca)	b53.10d	b59.30c	b62.10b	b61.80b	b65.30a
	AA5 (Jumbo)	a61.20e	a63.10d	a67.50b	a64.50c	a75.90a
Actinidia deliciosa	AD20	g15.10d	g29.50b	f30.10b	g27.10c	g32.10a
	Hayward	c30.00e	c41.90d	c48.80b	c47.50c	c53.30a
	Katiuscia	f18.10c	f31.20b	e33.10a	f30.10b	f34.10a
	Kramer	d27.60e	d36.10d	d37.50c	d39.10b	d42.10a
	Tomuri	e21.10e	e34.10d	d37.50b	e36.10c	e39.50a
B constant A variable: Dl 5%=1.19*% ; Dl 1%=1.60%; Dl 0.1%=2.12 % A constant B variable: Dl 5%=1.26*%; Dl 1%=1.68 %; Dl 0.1%=2.19%						

There were made interpretations by Dl 5% indicated in the table by *

Table 4

Actinidia arguta

	CULTURE MEDIUM	21.03	26.03		3.04		17.04		1.05	
		INITIAL NUMBER	No.	%	No.	%	No.	%	No.	%
R10P14	V1	15	15	100	12	80	9	60	9	60
	V2	15	14	93.3	13	86.6	13	86.6	13	86.6
	V3	15	10	66.6	9	60	7	46.6	7	46.6
R10P2	V1	12	10	83.3	8	66.6	8	66.6	8	66.6
	V2	12	10	83.3	8	66.6	8	66.6	8	66.6
	V3	12	10	83.3	10	83.3	4	33.6	4	33.6

Table 5

Actinidia delicioasa

	Culture medium	24.04	1.05		6.05		11.05	
		Initial number	No.	%	No.	%	No.	%
R1P16	V1	23	15	65.2	12	52.1	9	39.1
	V2	29	20	68.9	16	55.1	15	51.7
	V3	20	15	75	12	60	12	60
R1P1	V1	19	15	78.9	10	52.6	10	52.6
	V2	15	11	73.3	10	66.6	9	60
	V3	12	8	66.6	5	41.6	1	8.3

CONCLUSIONS

The use of rooting treatments is recommended to obtain higher rooting rates. The best experimental variant was to stimulate the rooting with a mix of ANA + IBA 1000 ppm solution.

To initiate the in-vitro culture of *Actinidia arguta*, recommended the culture media corresponding to V2 variant, as well as to the V1 must be used for the male and the female plant respectively. To initiate the culture of *Actinidia deliciosa,* the culture medium corresponding to variant V3 and to variant V2 must be used for the male plant and the female plant respectively.

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THE PEPPER (*Capsicum annuum* L.) –ALIMENT AND REMEDY

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Key words*: long pepper, ascorbic acid, chlorophyll, carotene , technological maturity*

ABSTRACT

This study refers to the results of research in the didactic field of legumiculture teaching area to "Staţiunea Didactică Banu Mărăcine Craiova", on the 6 variants of long pepper fruit to highlight the content of ascorbic acid, chlorophyll and carotenoids.High content of ascorbic acid have been noted for varieties "Cosmin" (160 mg/100), "Lung de Isalnita" (156 mg/100 gsp) and "Bogdan" (140 mg/100 gsp) compared to other variants On carotenoid content were observed following values for varieties "Bogdan" (1.30 mg/100 gsp), "Siret" (1.12 mg/100 gsp) and "Lung de Isalnita" (0.98 mg/100 gsp).

INTRODUCTION

The pepper (*Capsicum annuum* L) originates from Central America and South America. Christopher Columbus found the pepper in Haiti, where it was brought from, to Europe. (Ileana Beresiu & collaborators, 1977)

The pepper is grown in Europe for the first time mid way through the 16th Century in Spain and Portugal, followed by Germany, England and Hungary. The pepper reached Romania much later, being brought here by Bulgarian gardeners in the 18th Century. It was fist grown in the south of the country, and was later taken to other more favourable regions. (D. Andronicescu & collaborators, 1968)

The fruit of the pepper present a high value as an aliment, due to its elevated content of natural sugars and vitamins and the fact that it is habitually consumed as raw, state in which these components are processed directly, by the human body.

Ascorbic acid is found in large quantities in the fruit of the pepper, this varies according to species, variety or the maturity of the fruit, reaching levels of 139-160mg/100g of raw substance for fruits arriving at technological maturity and 211-300mg/100g of raw substance for fruits arriving at physiological maturity. (Bodea, 1972); some species of the Caspicum variety may reach higher levels up to 400mg/100g of raw substance – for the Caspicum frutescens species. (V.K. Andryaschenco & collaborators, 1983)

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On top of ascorbic acid, the fruits of the pepper contain other vitamins, such as B_1 şi B_2, PP and E.

Capsaicin, active component in peppers, give it the hot/chili taste, characteristic to many varieties. The levels of Capsaicin defers according to species and variety, between 0.27-1.12mg/100g of raw substance for hot/chili fruits, and minute quantities for sweet varieties. (A. Somos, 1966)

The high levels of ascorbic acid and capsaicin in the Capsicum variety give it the status of medicinal plant. The human body requires a minimum of 100mg of ascorbic acid daily, which can be easily ensured by a daily intake of pepper in our diet, consumed raw or in salads, and by no means heat-processed.

Elevated levels of capsaicin are characteristic to hot/chili pepper varieties. While in the 70s the consumption of hot/chili peppers was linked to a series of illnesses of the digestive tract and of the blood vessels, recent studies taken place in Australia, Hungary and the USA (countries where the pepper is highly consumed) have reveled that, in truth, the chili pepper prevents cardio-vascular diseases, cures some illnesses of the digestive tract, prevents prostate cancer and type II diabetes. Recently, the chili pepper has started being used in cosmetic products targeting cellulite and has proven to be a great success.

This paper aims to recommend some Romanian varieties of long pepper for raw consumption, according to their levels of ascorbic acid and carotene.

MATERIAL AND METHOD

Were studied 6 genotypes of long pepper, grown in a tunnel-type solarium in SDE Banu Maracine which had the following features found listed in table 1.

Table 1

Long pepper genotypes analized

No.	Genotypes	The color of the fruit when reaching maturity		Source
		Technological	Physiological	
1	Lung de Isalnita	Dark green	Dark red	SCDL Isalnita
2	Bogdan	Green-yellow	Red	SCDL Isalnita
3	Siret	Yellow-green	Red	SCDL Bacău
4	L – 54	Yellow-green	Red	SCDL Isalnita
5	L – TP	Yellow-green	Orange	SCDL Isalnita
6	Cosmin	Dark green	Dark red	ICDLF Vidra

The content of ascorbic acid, carotene and chlorophyll was determined for the state of technological maturity.

Determination of ascorbic acid

A sample of 5-10 g of pepper, previously ground with quartz sand has been put into a 100ml balloon by using a solution of 2% hydrochloric acid. It has been stirred and after sedimentation it has been filtered into a dry glass. A 10 ml aliquot has been passed into a Berzelius glass, to which 30 ml of distilled water, 5 ml of 1% potassium iodate and 1 ml solution of starch have been added. It has been then titrated with potassium iodate N/250 stirred until becoming bluish (Biţa *et al.*, 2009, Dumitru *et al.*, 2010, Samuella *et al.*, 2004, Roberts *et al.*, 1987).

The calculation of ascorbic acid concentration is made by using the equation

Vitamin C mg % = 352. n.f / G

Where:

n - ml used for titration;
f – the factor of the potassium iodate N/250;
G – the sample weight in grams.

The data recorded were statistically processed by using the variant method and by establishing the limit differences (DL).

Determination of total carotenoids

The weighed samples, having been put separately in 95% in acetone (50 ml for each gram), were homogenized with Braun MR 404 Plus for one minute. The homogenate was filtered and was centrifuged using the Hettich Universal 320/320R centrifuge at 2500 rpm for ten minutes. The supernatant was separated and the absorbances were read at 400-700nm on Cary 50 spectrophotometer. It was recorded that Chlorophyll a showed the maximum absorbance at 662 nm, chlorophyll b at 646 nm and total caroten at 470 nm and the amount of these pigments was calculated according to the formulas (Lichtentaler et al., 1985,Grung et al., 1992, Wellburn et al., 1994).

Ca = 11.75 A662 - 2.350 A645
Cb = 18.61 A645 - 3.960 A662
Cx+c = 1000 A470 - 2.270 Ca - 81.4 Cb/227
Ca = Chlorophyll a, Cb = Chlorophyll b, Cx+c = Total carotene

RESULTS AND DISCUSSIONS

The determinations were made on samples of fruits harvested at physiological maturity when their colour was red and they represent average values of the repetitions within the studied variants.

Green, yellow, red or orange peppers are remarkable for their capacity of strengthening the immunity system and their fight against free radicals, they represent one of the most important sources of vitamin C (red peppers contain three times more vitamin C than oranges), as well as of β - carotene (important compound in the fight against cancer), arguments which have determined several scientists to thoroughly study these miraculous species.

Table 2

Principal chemical components of pepper genotypes

No.	Genotypes	Ascorbic acid mg/100g of raw substance	Chlorophyll content			Carotene mg/100g of raw substance
			Total	Types		
				"a" Chlorophyll	"b" Chlorophyll	
1.	Lung de Isalnita	156	136,5	91,5	45,0	0,98
2.	Bogdan	140	131,6	87,5	44,1	1,30
3.	Siret	104	125,5	83,5	42,0	1,12
4.	L – 54	75	117,0	79,6	37,4	0,81
5.	L – TP	96	103,5	67,4	33,2	0,65
6.	Cosmin	169	159,8	104,0	55,8	0,30

Ascorbic acid, the principal component in peppers, varies, indicating its value as an aliment and the therapeutic value of each genotype. Lung de Işalniţa (156 mg/100gsp) and Cosmin (169 mg/100gsp) varieties present a significant level; the fruit for these varieties can be recognized by its dark green color, determined by accounting for the total chlorophyll levels. Even without a thorough analysis of the link between the content of

ascorbic acid and chlorophyll, the data in table 2 indicate that genotypes with a high level of ascorbic acid also have high levels of chlorophyll.

Bogdan variety also presents an increased level of ascorbic acid (140mg/100g of raw substance) and is a highly productive variety; its fruits ooze of quality and have a green-yellow color upon reaching technological maturity – these features are preferred by consumers and make the Bogdan variety highly recommended and sought for raw consumption.

The carotene levels in the technological maturity stage of the pepper fruits for Cosmin and Bogdan varieties is surprisingly low, 0,30 mg/100gsp (for Cosmin) and 1,30 mg/100gsp (for Bogdan).

CONCLUSIONS

Results highlight the qualities we have these long pepper genotypes grown under the unheated greenhouse condition and recommend them to be taken into cultivation on large areas due to:

High content of ascorbic acid showed the genotypes "Cosmin", "Lung de Isalnita" and "Bogdan";

Carotenoids present in greater quantities in genotypes "Bogdan", "Siret" and "Lung de Isalnita".

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✓*Tehnologia prelucrării produselor agricole*
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Vol. XVII (LIII) - 2012

ELABORATION OF THE PROTOCOL FOR *IN VITRO* PROPAGATION OF SOME BIOLOGICAL INDICATORS USED IN VIRAL TEST TO FRUIT TREES SPECIES

Catita Plopa[1], Maria Isac[1]

Key words: *protocol, meristem, multiplication, fitohormon, genotype*

ABSTRACT

For shortening of the time to produce the biological material and elimination of the viral infection danger, was tested the pretability to in vitro propagation of the Malus platycarpa, Golden delicious, Virginia Crab, Lord Lambourne, SPY 227, Tuleu dulce, Vânăt de Italia and GF 305 indicators. The differentiation of the stone genotipes was higher on MS culture media with hormonal balances by 0,05 mg/l GA_3 and 0,1 mg/l IBA. Pome genotypes recorded the best results on QL culture media with Walkey vitamins and GA_3 quantity reduced to 0.01 mg/l. The hormonal combination BAP and NAA on MS and QL culture media give a different multiplication potentialy, one of the influence factor being genotype. The rooting capacity: % rooted plants and roots number/plant has a evolution in accordance to nutritional formulas and genotypes.

INTRODUCTION

The biological indicators have a great utility in detection of the viral diseases to plants, identification of the viruses bybiological method is a diagnosis procedure very important and necessary according to OEPP standards.

The sortening of production time of the biological material and elimination of the viral infection danger in propagation, the conservation through storage in small space of a great number palnts, allowing use in any season (Gaspar, 1981; Holdgate, 1982) represent the advantages that determined new search for optimal solutions for *in vitro* propagation of many plants species.

For fruit trees plants the reserches regarding to *in vitro* propagation are successful for many species and within species for many cultivars. This thing cannot generalize because exist many factors.

In the studying of the influencing factors on the evolution of explants *in vitro* culture has been taken to ensure that there are information that citochinines stimulate bud growth, but the results differ depending on the variety (Davies, 1988).

The culture media used in micropropagation should to offer the nutrient and organic substances for celular metabolism and the growth factors with responsability in shoot diferentiation and rooting (Schuch & Peters, 1993; Lombardi et al, 2007).

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Requirements for nutritional formulas and composition hormonal balance may differ even within the same genotype across phases of culture (Zuccherelli, 1979, Hussain, 1983 etc., Isac, 1983, Sharma, 2000).

Starting to the utility in detection of the viral diseases to plants of the biological method due to lack of data in the literature regarding the behavior of these genotypes to propagation process, Virology-Tissue Culture Laboratory of the Research Institute for Fruit Growing Pitesti-Măracineni, aimed to realise some efficient propagation protocols for establishing of *in vitro* differentiation, multiplication and rooting of a series of biological indicators used to viral test of the pome species: *Malus platycarpa:* ***ACLSV,*** Golden delicious: *APP, ApMV, Apple dapple, Apple scar skin viroid,* Virginia Crab: *ASGV, ASPV,* Lord Lambourne: *ApMV,* SPY 227: *ASPV and stone species* GF 305: *PPV, ACLSV, ApMV, MLRSV, PDV, PNRSV, TBRV, SLRV, CLRV, ArMV, PLMVd, Peach asteroid spot, European stone fruit yellows phytoplasma,* Tuleu dulce*: PPV, PDV, PNRSV,* Vanat de Italia*: PDV*

MATERIAL AND METHOD

Explant sources. The biological material used was represented by apically buds from annual branches of Malus platycarpa, Golden delicious, Virginia Crab, Lord Lambourne, SPY 227, Tuleu dulce, Vânăt de Italia și GF 305 genotypes . The explants obtained were meristems of 0.5-1 mm size.

Table 1

Experimental variants

Variants	Basal medium	Culture phases			
		Initiation			
		Vitamins	***Growth regulators (mg/l)***		
			GA_3	**IBA**	
V1	MS	MS	0,01	**0,1**	
V2	MS	MS	0,05	**0,1**	
V3	MS	MS	0,1	**0,1**	
V4	QL	Walkey	0,01	**0,1**	
V5	QL	Walkey	0,05	**0,1**	
V6	QL	Walkey	0,1	**0,1**	
		Multiplication			
		Vitamins	***Growth regulators (mg/l)***		
			BAP	**NAA**	
V1	MS	MS	0,1	**0,2**	
V2	MS	MS	0,5	**0,2**	
V3	MS	MS	1	**0,2**	
V5	QL	Walkey	0,1	**0,2**	
V6	QL	Walkey	0,5	**0,2**	
V7	QL	Walkey	1	**0,2**	
		Rooting			
		Vitamins	***Growth regulators (mg/l)***		
			GA_3	IBA	**IAA**
V1	MS	MS	0,01	1,5	**-**
V2	MS	MS	0,01	-	**1,5**
V3	QL	Walkey	0,01	1,5	**-**
V4	QL	Walkey	0,01	-	**1,5**
V5	MS	MS	0,01	1,0	-
V6	QL	Walkey	0,01	1,0	-

Desinfections of biological material consist of: washising with water and liquid detergent Tween 80 for 5 min; immersion in 6 % (w/v) $Ca(OCl)_2$ for 20 min; immersion in 90 % ethanol for 10 min; rinsed three times in sterile distilled water.

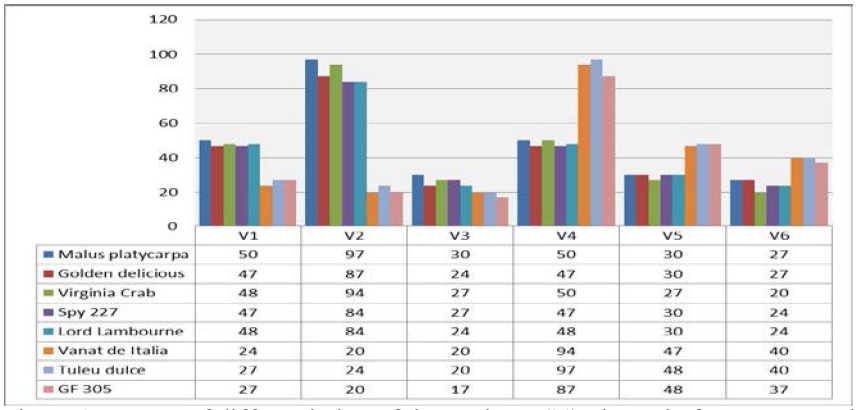
Culture media were represented of Murashige & Skoog (MS,1962), and Quoirin & Lepoivre (QL,1977) with vitamins Walkey, 1972. All media contained 40g/l dextrose, 8 g/l agar and 32 mg/l Na Fe EDTA. Resulted variants are shown in the table 1.

Culture conditions After dissection and inoculation the cultures were maintained at 20-22^0 C, 16 light / 8 dark hours photoperiod.

RESULTS AND DISCUSSION

After 30 days from the culture start were observed the efects produced by culture media compounds on studied genotypes (fig. 1).

In vitro stability give by the degree of differentiation of explants, was according to the culture media through macro and micronutrients composition and hormonal balance used. Pome genotypes recorded higher results on MS medium with hormonal balance represented of 0.05 mg/l GA3 and 0.1 mg/l IBA instead the genotypes of stone fruit species preferred the poorest backgrounds in macro and micro (macro and microlemente QL and vitamins Walkey) and a lower hormonal balance in terms of concentration GA3 (0.01mg/ l).



	V1	V2	V3	V4	V5	V6
Malus platycarpa	50	97	30	50	30	27
Golden delicious	47	87	24	47	30	27
Virginia Crab	48	94	27	50	27	20
Spy 227	47	84	27	47	30	24
Lord Lambourne	48	84	24	48	30	24
Vanat de Italia	24	20	20	94	47	40
Tuleu dulce	27	24	20	97	48	40
GF 305	27	20	17	87	48	37

Figure 1. Degree of differentiation of the explants (%) depend of genotype and components of the culture media

Multiplication phase

Expression of the replicative capacity of studied genotypes was performed according to the culture media and hormonal balance (Table 2). In this case also the pome genotypes have a better evolution on MS medium compared with QL culture media, where better results were obtained for stone genotypes.

In the same basic medium MS, the differences were recorded depending on the concentration of BAP. The pome genotypes showed a multiplication rate at a concentration of 0.5 mg/l BAP in combination with 0.2 mg/l NAA, throughout the multiplication phase, phase consisting of primary culture, subcultura1, subculture 2, subculture, subculture 3 and subculture 4. Although the data about the genotypes that covered this study were not common in the literature, however, can be made comparations with other fruit genotypes studied for this purpose. So, on multiplication during after subculture 4 was observed a decline in training capacity of shoots in all genotypes as a result of the accumulation of

inhibitors, this situation was reported and by other authors in different genotypes of *Prunus* (Vujovići et al, 2012).

Both *Prunus* species as well as for others studies have reported better action in terms of the type citochinines used: zeatin and 2iP were commonly used for *in vitro* multiplication. At some peach rootstocks (Eccher & Noé, 1989, Popowich & Filipenya, 1997; Gonzales et al, 2000; Debnath, 2004), zeatin induced a higher number of shoots compared to 2iP. On the other hand, other authors noted no significant differences Ribas et al., 2005, in zeatin action and BAP on *in vitro* multiplication.

Table 2

The effect of the hormonal combination BAP and NAA on the multiplication rate (average shoot/explant Cp + S1 + S2 + S3 + S4)

Genotip	V1	V2	V3	V4	V5	V6
Malus platycarpa	1:3	1:7	1:2	1:3	1:3	1:2
Golden delicious	1:3	1:6	1:2	1:2	1:3	1:2
Virginia Crab	1:2	1:6	-	1:2	1:2	1:2
SPY 227	1:2	1:7	1:2	1:3	1:2	1:2
Lord Lambourne	1:2	1:5	-	1:3	1:2	1:2
Vanat de Italia	-	1:3	1:2	1:4	1:6	1:7
Tuleu dulce	-	1:2	-	1:5	1:7	1:8
GF 305	-	1:2	-	1:5	1:6	1:8

Rooting phase

The rooting ability was studied in two aspects: the number of rooted plants and number of roots/plant.

The number of the rooting plants was influenced by the concentration and the auxin type. Exposure to higherconcentration by 1.5 mg/l IBA in combination with 0.01 mg/l GA_3 and salts MS was efficienty only for rooting inductions to *Malus platycarpa*, Golden delicious, Virginia Crab, Lord Lambourne, SPY 227 genotypes (fig. 2).

The combination 0.01 mg/l GA_3 and 1.0 mg/l IBA (V6), was efficienty for Tuleu dulce, Vânăt de Italia and GF 305 genotypes. The effect of IBA concentration on the percentage of rooted plants was signaled by Sharma et al., 2007, to apple rootstock, which reached a maximum of 89.63% rooting at a conc. by 2.5 - 3 mg/l IBA, but with abundant callus formation at the basal part of the stem.

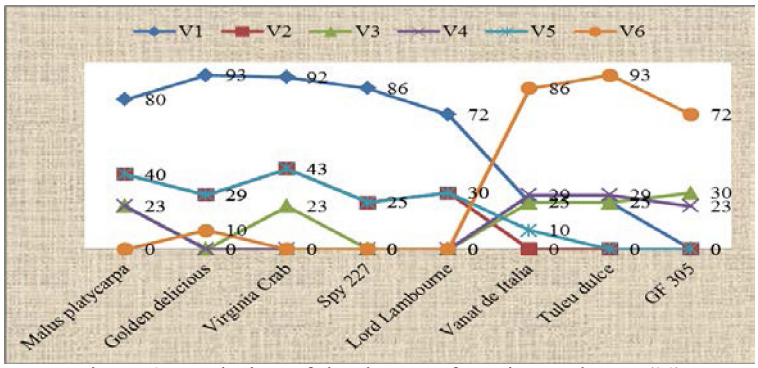


Figure 2. Evolution of the degree of rooting cultures (%) according to nutritional factors - hormonal balance and culture medium

Results abtained after using IAA auxin for rootedness process, does not recommend using this auxine under culture conditions for the studied genotypes.

Maximum number of roots/plant was observed at about 5 weeks of culture for pome genotypes (5-7 roots) for the culture media V1, and approximately to 4 weeks on culture media variant V3 (4-6 roots), for stone species (fig. 3).

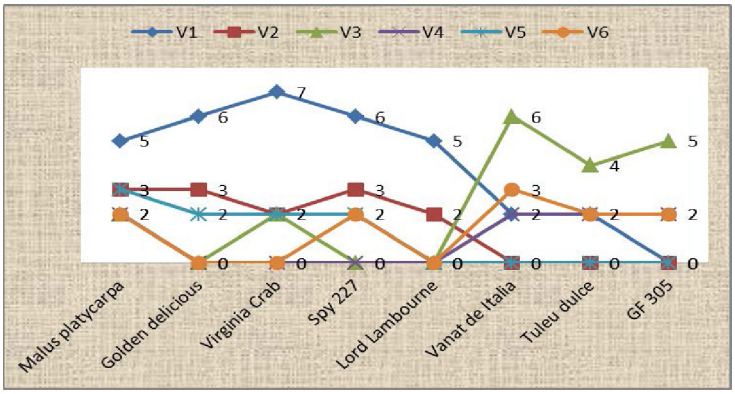


Figure 3-The evolution of the cultures regarding to number of roots / plant according to nutritional factors - hormonal balance and culture medium

The environmental factors provided, represented by culture media, temperature and light regime, produced plants with normal appearance, without being reported during the growing phase vitrification phenomena, yellowing, excessive callus, etc, (photo 1 , photo 2).

This paper is carried out as a result of the Plan sectorial ADER 2020 - Proiect ADER 2.2.7.

Foto 1-Tuleu dulce, aspect from multiplication phase

Foto 2-Spy 227,aspect from rooting phase

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SOME ASPECTS ON CONFORMITY EVALUATION IN THE FIELD OF FOOD SAFETY

Poenaru Maria-Magdalena[1], Iacobescu Fanel[2]

Keywords: *method, validation, conformity evaluation, accreditation, RENAR*

ABSTRACT

This paper presents aspects of conformity assessment infrastructure, the role of the main actors that are part of the conformity assessment infrastructure, RENAR, ASRO and BRML, but also the other bodies, organizations, institutions and associations involved and that contribute to the technical-scientific foundation of numerical values reported about the quality of a food or a food product. Also, in the context of meeting the accreditation requirements by the laboratories for food analysis are presented performance characteristics which are base of the validation studies of methods of analysis.

INTRODUCTION

On the basis of measurements results many important decisions are made in support of legislation or in industrial processes or social aspects. In the field of food safety, the measurement quality is important to enable an equivalent implementation of the European Union regulations and directives across an enlarged EU.

The European Union has developed technical instruments to remove the barriers to free circulation of products, services and person. Among these, the New Approach to product regulation and the Global Approach to conformity assessment take place. New Approach directives apply to products which are intended to be placed on the Community market for the first time. Consequently, the directives apply to new products in the Member States. The concept of product varies between New Approach directives, and it is the responsibility of the producer to verify whether or not the product is within the scope of one or more directives.

For an easier understanding I will explain some definition of terms [EN ISO/IEC 17000:2004], as follow:

Conformity assessment: The conformity assessment body has to demonstrate that specified requirements relating to a product, process, system, person or body are fulfilled. Activities which is the subject in the field of conformity assessment include *testing*, *inspection* and *certification*, as well as the accreditation of conformity assessment bodies.

Conformity assessment body: Performs conformity assessment.

Accreditation: attestation related to a conformity assessment body conveying formal demonstration of its competence to carry out specific conformity assessment tasks.

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Accreditation is part of an overall system, including conformity assessment and market surveillance, designed to assess and ensure conformity with the applicable requirements.

Accreditation body: body that performs accreditation.

Romanian Accreditation Association – RENAR, a non-governmental, non-profit organization, is formally recognized as unique Romanian national accreditation body by Government Ordinance 23/2009 and based on the provisions of (CE) Regulation no. 765/2008. RENAR operates under the coordination and surveillance of the Ministry of Economy, Trade and Business.

Recognition of conformity assessment results: Acknowledgement of the validity of a conformity assessment result provided by another person or body.

THE CONFORMITY EVALUATION INFRASTRUCTURE

The EU Commission has approved a new Regulation No 765/2008 setting out the requirements for accreditation and market surveillance relating to the marketing of products, which was applied from January 2010. It has been developed against the background of a growing recognition of the importance of accreditation to the EU's economic infrastructure. The Regulation, which establish a legal framework for the provision of accreditation services across Europe, covers the operation of accreditation in support of voluntary conformity assessment as well as conformity assessment required by legislation. Under the Regulation, accreditation, when carried out against the recognised harmonised standards, is regarded as a public authority activity.

"A system of accreditation which functions by reference to binding rules helps to strengthen mutual confidence between Member States as regards the competence of conformity assessment bodies and consequently the certificates and test reports issued by them. It thereby enhances the principle of mutual recognition and therefore the provisions of this Regulation on accreditation should apply in relation to bodies carrying out conformity assessments in both the regulated and the non-regulated areas. The issue at stake is the quality of certificates and test reports irrespective of whether they fall within the regulated or the non-regulated area, and no distinction should therefore be made between those areas." [Regulation No 765/2008]

Conformity assessment is divided into several operations (modules) and the principles underlying the conformity assessment are[Iacobescu F et al 2008]:

- a coherent approach of the conformity assessment;
- widespread use of standards from EN ISO 9000 and EN 45000 series;
- promoting the development of accreditation systems and the techniques intercomparison;
- promoting mutual recognition agreements of testing and certification in the mandatory field, but also in the voluntary field;
- reducing disparities between Member States and between industrial sectors in terms of existing quality infrastructure through developing of some programs;
- promoting commercial trade between members of the European Economic Community and third countries.

Main actors of the evaluation infrastructure for conformity assessment in Romania are presented in Fig. 1.

RENAR, the national accreditation body certifies competence to perform specific tasks for laboratories, certification bodies and inspection bodies.

BRML, regulatory authority in the field of metrology, has as main tasks the ensurance of consistency and accuracy of measurements.

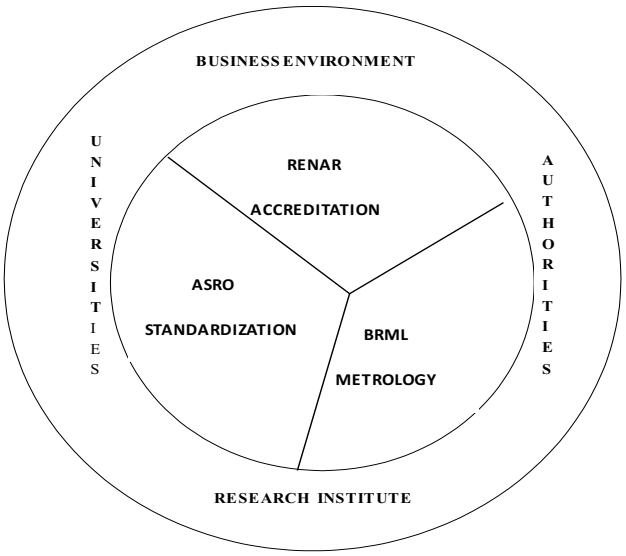

Fig. 1 Infrastructure actors for conformity assessment

Among its duties, ASRO, the national body of standardization, elaborates and approves, reconfirms, modifies and/or cancels the national standards and also adopts international standards, regional and European standards as national standards.

THE RECOGNITION SYSTEM

"In order to avoid multiple accreditation, to enhance acceptance and recognition of accreditation certificates and to carry out effective monitoring of accredited conformity assessment bodies, conformity assessment bodies should request accreditation by the national accreditation body of the Member State in which they are established." [Regulation No. 765/2008]

The default value of accreditation is that it provides an authoritative statement on the technical competence of the bodies whose task is to ensure that products comply with the requirements applicable to them, namely RENAR.

Only one accreditation requirements of the food analysis laboratories is that the laboratory must demonstrate that applies standardized test procedures (published in national and/or international standards); in case there are no standardized procedures, then they should be validated.

The validation of a method is made by the laboratory that applies the method or by a group of laboratories that agree to study a method with wide applicability which has potential to be adopted as *standard*.

Validation of a method is a process of [Poenaru M.M. et al 2008]:

- establishing performance characteristics and the limitations of a method;
- identifying the influences that may change these characteristics and to what extent;

- establising the analyte to be determined, in what matrix and in the presence of what interference;
- verifying that a method is appropriate for the purpose that is going to be used.

Validation studies of the method are based on determining the overall method performance parameters. They are made either during the method development, during interlaboratory study or respectig validation protocols inside the unit. The individual sources of uncertainty are investigated only when they are significant compared with the indicators of accuracy in use, the ultimate goal being to identify them and remove their significant effects, except to correct them [Iacobescu F et al 2008]. This leads to the situation where most factors with significant potential influence compared to the overall accuracy have been identified and shown to be negligible.

The Romanian Bureau of Legal Metrology – National Institute of Metrology (BRML-INM) actively supports the participation of Romanian authorized and field laboratories in interlaboratory comparisons, the instrument of validation method. The results from Romanian laboratories participating in IMEP-12 (water), IMEP-16 (wine), IMEP-17 (human serum) and IMEP-20 (tuna fish) are presented by S. Duta et al.

CONCLUSIONS

It is easily seen that the effective participation of various bodies, organizations, institutions and involved associations confirms the interdisciplinary character of the quality concept, as a way of meeting the requirements specified in the conformity assessment procedures and the need for objective approach of all relevant aspects in the food domain.

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ŞARBA 2 Şt – A NEW CLONAL SELECTION FOR AROMATIC WINES

Popa Camelia[1], Onache Petronela- Anca[1]

Key words*: grape wine, clonal selection, variety, agrobiological and technological value*

ABSTRACT

Beginning with the 1993 year, a clonal selection was applied at grape wine Şarba, at INCDBH Stefanesti-Arges. Valuable clonal selection of the Şarba variety were selected as follows: during the first stage (1993-2011) the elite vines were studied in mother plantation and the best ones chosen; in the subsequent stegea comparative plantation was created and comparative study of the clonal progenies carried out. Through the repetate verification of elites in contest parcels,they have remarked by quality which is manifesting constantly clonal selection Şarba 2Şt. It was certified in year 2012.

INTRODUCTION

Clonal Selection represents a more advanced stage of the work for the selection, because it contributes to improving radical redesign of existing varieties of the vine. Consists in the choice and multiplying separated in the best fouling themselves, coming from the unequalled hubs. Careful tracking of each lines fouling originating from a single hub allows biological and economic determination of the value of each clone. Many varieties are widespread in cultivation are devoid of economic value, and others reverts once by multiplying their heterogene populations, by making the value vineyard, established without measures for the application of selection, to fall gradually.

In the period 1993-2011 years to INCDBH- Ştefăneşti have been selected and entered in the contest plantations of the 12 elite clonal variety representing Şarba. The selection was obtained as a result of the application of clonal selection scheme drawn up by I.C.D.V.V. Valea Calugareasca in the year 1972 for the production of clones upper population from which they come, both quantitatively and qualitatively, in such a way as to be a substitute variety gradually to population which he represents.

According to this methodology, the selection has been carried out in three distinct phases:

Fixing the selection of plantation in full capacity of fertility. Within the framework of this stage, clonal elites have been studied 1-3 years and it has been determined: the vigor of growth, plant health, the production of grapes and their quality. It is valuable elite harvested for shoots and have vegetative multiplied.

[1] National Research and Development Institute for Biotechnology in Horticulture Stefanesti-Arges

Setting up the comparative field and study the elite clonal. The comments and determinations carried out in the comparison field have contents: completing blindspots occurring; strenuousness determination of growth, the phenology spectrum of elites in the relationship with climatic factors which influenced the develop of the main phenophases; fertility and productivity elites; actual production of grapes for the hub and calculated per hectare; quality of the harvest appreciated by the content in sugar and acidity must; average mass of grape and the graines; technological indices of the grape; the percentage of degenerated grains; behavior to frost and specific disease vines. Were chosen and reproduced the most valuable elite, even in the early years of study.

In the agrotechnics improved conditions after testing viral analysis of biological material resulting from selection, the clonal elites have been entered in testing field. Studies and observations were similar to those of the preceding stage, as they are completed with data on surface of the foliar hubs; tests relating to the absence or the presence major viruses and determining quality of the wine.

Submission for the year 2012 of the elite clonal in the official catalogue of the elite of clonal plant species which growing in Romania.

MATERIAL AND METHODS

The original material for performing the works of clonal selection was the variety of production Şarba and plots occupied by variety. Identification, selection and marking of valuable elites from the variety above mentioned was carried out in groves between 20-25 years after planting with a status plant health. Previously this work was carried out a rigorous positive selection.

The choice of the elite was based on recommended criteria: plant health, the vigor of growth, production of grapes and its quality.

Number of clonal elite multiplied and introduced in the compared field was much lower than that of the initial plantation, chosen from the plantation due particularly demanding, in order to ensure the promotion of the most valuable biotype.

Şarba 2Şt elite selection proposed for approval has been made in the comparative field by the laboratory improvement, over an area representative for the vineyard Ştefăneşti, on the middle third of a slope betwen 10 -15 %, in southern exhibition. The soil is coluvial brown, clayey-sandy, medium stocked with phosphorus and potassium, low carbonatated, weak acidic pH (6.2-6.4). The hubs were grafted on the rootstock Kober 5 BB, they have been led to the half-high form (DrawString bilateral) with support on stakes with three double wires. Planting distances is 2.2 m between rows and 1,0 m between hubs (4500 hubs/ha).

Clonal elite evolution was accompanied by keeping valuable characters and biological particularities of hereditary inherited from the mother plantation elites.

Observations and determinations were made on clonal selection Şarba 2Şt. and Şarba variety, on frost resistance during the winter, the deployment of the main phenophases, the duration of the period of vegetation, the coefficients were been calculated absolutely and relatively fertility, indices of productivity, the quantity and quality of rapes.

RESULTS AND DISCUSSION

Data recorded from the observations and calculations of agrobiological and technological conditions, bears the climatic conditions of wine years 2010-2011.

Climatic conditions of these wine years research is characterised in particular by water regime lacking, especially in critical periods of growth and maturation of the grapes, by large differences in temperature between summer and winter. Excessive Heat (34-34,7^{0}C July-Sep.) and precipitation reduced from the period of maturation of the grapes (14.5 - 8,6l/m^2 July and August) since the summer 2010-1011 years, have contributed to knowledge of how to adapt the elite clonal to these conditions.

Technological and agrobiological characteristics of Şarba 2 Şt elite and comparison of clonal variety witness Şarba, are shown in figures 1, 2, 3, 4. and is represents the average over two years (2010-2011).

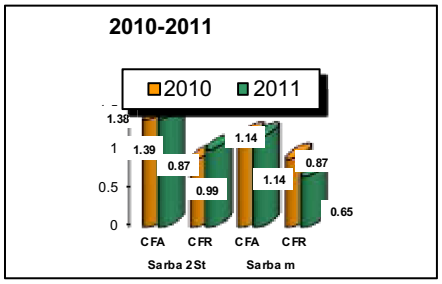

Fig.1 The fertility quotas

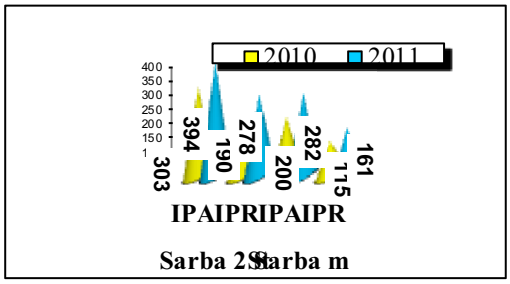

Fig.2 The productivity indexes

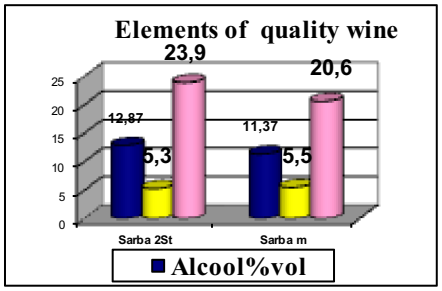

Fig. 3. Qualitative characteristics of clonal elite Şarba 2Şt

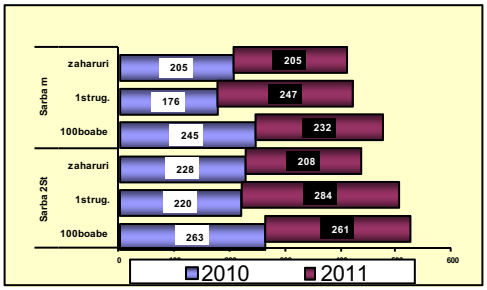

Fig. 4. Physico-chemical characteristics of the win

Phenophases like budding - total maturity have been proceeded on the same dates for both clone as well as for the blank variety - population. (18.04-22.09 2010 and 26.04-29.09 2011). Elite selected has proved resistant to frost, which recorded losses of only 6% of the preceding two years, compared to analyze the variety Şarba in 2011 that has lost 13% of buds. Higher fertility shoots the clone had submitted (65-68%) compared with blank variety (57-58%) actually proved both percentage calculated on the block fertili shoots, but also from determining coefficients of fertility absolutely and relatively (Fig. 1).

From the point of view of production of grapes on the hub, the clone Şarba 2 Şt. is much higher population of that has been selected. Thus, the production weighed/hub has been with 2 kg higher in 2010 for clone (4.3 kg /hub compared with 2.3 kg /hub for the blank), and in the year 2011 Şarba 2 Şt. expressed a yield of 8.2kg /hub, far exceeding 4.5 kg variety blank. (Fig. 2).

High production recorded by clonal selection Şarba 2 Şt, was not to the detriment of the quality of the grapes. Grapes and grains have weighed more than the blank variety

and the content of the must was richer in sugars (Figure 3), corresponding to acidity of less 3,4-3,6 g/l H_2SO_4.

After it has been made wine elite clonal and blank variety, they result in the following physico-chemical characteristics of wines (fig 4): clonal selection Şarba 2 Şt recorded total alcohol 12,87% vol., total acidity 5,3 g/l TA , dry extract of 23.9 g/l, and the Şarba variety presented total alcohol 11,3% vol., total acidity 5,5 g/l TA and dry extract 21.4 g/l. Again you can see the superiority of clonal elite and in terms of the quality of the wine.

Fig. 5. Clonal selection 2 Şt.

CONCLUSIONS

From quality and quantity point of view, the selected clone Şarba 2Şt. proved to be superior to the variety that was selected from.

The sugar accumulation potential corresponds to the quality varieties.

The selected clones can replace successfully the Şarba variety.

The obtained wine after the vinification of the selected clone Şarba 2 Şt. meets all the characteristics of a quality wine, fact that can lead to the vinification of this one in big quantities.

By component type-approval clonal elite Şarba Şt., try inserting the culture of most valuable selections from indigenous varieties which in time can lose a part of qualitative traits.

Correlating factors of production with the quality in the field of comparison, it is found that the selection has maintained the characteristics of the variety, but mostly on the size of the grain and its consistency and accentuated muscat flavor .

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✓*Tehnologia prelucrării produselor agricole*
✓*Ingineria mediului*

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THE INFLUENCE OF HYDRIC STRESS ON THE BEHAVIOR OF SOME VINE VARIETIES RECOMENDED FOR THE MURFATLAR VINEYARD

Ranca Aurora[1], Artem Victoria[1], Petrescu Anamaria[1], Stanciu Iulia-Cristina[1], Dina Ionica[1]

Key words: *production, vulnerability, physiological processes, climate changes, harvest year*

ABSTRACT

The data presented in this study was recorded in the SCDVV Murfatlar area from representative plantations for the viticultural center and refers to 2009-2012. There had been taken into consideration six vine varieties, monitoring their evolution depending by their specific vulnerability to climate changes. The viticultural years 2009 and 2010 had a poor quantitative and qualitative production, because of the great influence of the frost (2009) and excessive moisture during the period of ripening and full maturation (2010). By contrast, the viticultural year 2011 records higher production compared to other years under investigation. In terms of quality, the production of 2011 recorded lower concentrations of sugars in grape must compared to the year 2012, but kept a good quality. The highest sugar contents were recorded in 2012, but the high temperatures and the lack of rainfall have favored the instalation of hydric stress, the production obtained under these conditions being low due to the disturbance of the physiological processes. From the quantitative perspective in 2012 it is observed that the average decrease of production is 25%

INTRODUCTION

Detailed knowledge of the annual evolution of the main environmental factors is an essential condition for ensuring high grapes productivity and quality, in terms of increased economic efficiency. Heat, light and moisture are climatic factors that the vinegrower must constantly take into account (Dejeu 2004, Book 2004). Hydric stress occurs as a result of high temperatures and poor precipitation regime and is a physiological reaction of the the vine to insufficient water supply. The action on the plant consists in exaggerated intensification of transpiration process and can lead to the disturbance of the balance between water absorption and transpiration intensity. For this reason the plant begins to wilt (Berbecel & Neacsa 1966, Book 1966). Among the physiological responses are reduced cell division, reduced cell size, stomatal closing at the level of the leaf, reduction of the photosynthesis process and in the worst case desiccation and cell death.

[1] Research Centre for Viticulture and Enology Murfatlar

Most of these processes are dynamic, largely following water stress level. For example, regarding the stomatal response (which have a strong influence on the process of photosynthesis and therefore on the potential for accumulating sugars), they do not close completely at the first signs of hydric stress, but gradually close as the phenomenon accentuates.

Hydric stress can also have less obvious effects on the harvest productivity and quality. For example, grain size reduction leads to a increased ratio between skin and must, which can increase the concentration of anthocyanins and polyphenols in grape must and red wine (Bahar et al.2011). Water stress may also affect the processes of formation and breakdown of acids and important flavoring substances.

Classically water stress level must be measured by monitoring one or more physiological responses of the vine such as the leaf water content, stomatal conductance and cell expansion level. But these techniques are difficult, expensive and time consuming. By means of the current technology it is easier measuring the soil moisture (Department of Primary Industries, 2002).

MATERIALS AND METODS

The study was conducted in the Murfatlar vineyard at the Murfatlar Research Center for Viticulture and Enology between 2009 and 2012. The investigated varieties are Columna, Pinot Gris, Feteasca Regala, Mamaia, Feteasca Neagra and Cabernet Sauvignon.

Registration of climatic data was performed using the meteorological station located in the research area.

In order to study the hydric regime soil samples were collected at depths of 0-20 cm, 20-40 cm, 40-60 cm, 60-80 cm and 80-100 cm for each of the 6 parcels in the study. Sampling was performed using a agrochemical probe, and samples were processed using the gravimetric method.

In order to determine the influence of water stress on grape production for studied varieties under the local climatic conditions samples were collected at the time of the grape technological maturity during the four years of the experiment. It was determined the sugar content of the must expressed in g/l, the acidity in g/l H_2SO_4 and the weight for 100 grains in grams and the must yield. Sugar content was determined refractometrically and the acidity by titration with NaOH in the presence of phenolphthalein.

The results were analyzed, monitoring the sugars accumulation and acidity decrease of the must in order to establish the influence that climatic factors have on grapes quality.

RESULTS AND DISCUSSIONS

During the years under study we observed that the weather is characterized by dry periods alternating with rainy periods, disturbing the life cycle of the vine.

The climate of the region has an excessive continental character given by pronounced contrasts between summer and winter. The annual average sum of temperatures recorded in the area is 4200°C while the sum of temperatures recorded only during the vine growing period is 3500ºC. The aridity of the Murfatlar viticultural center is characterized by 450mm of average annual precipitation of wich only half falls during the vegetation period, the phenomenon being accentuated by frequent winds - almost daily.

The characteristic soil for the experimental perimeter is calcareous chernozem with loamy structure, rich in carbonates. It has a medium texture, the percentage of humus

ranging from 1.8% to 3%. It's a dark soil with a high natural fertility, consisting of herbaceous vegetation in temperate conditions (Ranca 2007, Book 2007).

Research results include data on production quality, influenced by the ecopedoclimatic conditions of the region.

In terms of soil humidity, the initial moisture content for 2009 is low, the deficit was moderate but low enough (Table no.1) to affect crops. The moisture deficit will accentuate strongly during the summer months, especially in July. It decreases slightly in the next months, but still remains at a very low level compared to the plants' needs. The year 2009 can be characterized in terms of soil moisture as very dry.

The year 2010 starts with a moderately negative deficit, which indicates that the soil moisture level is slightly above the field capacity of the soil, very suitable for the begining of vegetation of vines. Moisture deficit is kept at a moderate level in the next months to return to negative values during July and August. During the warm summer months excessive soil moisture is not desirable, because it facilitates pest attacks on plant and slows the accumulation of sugars in the grapes.

In 2011 the initial water reserve value is situated well above the average, favoring the entry into vegetation, but since June, when the plant needs water is still high, there is an aggravation of water deficit, with repercussions on the development of shoots and grapes, having though a beneficial effect on sugar accumulation.

In 2012 the initial water reserve recorded a moderate deficit, approaching the normal in May and then registeres a strong deficit, with highly negative effects on vines and grape.

Table no.1

Avearge soil water deficit during 2009-2012

Month	**2009**	**2010**	**2011**	**2012**
IV	224.3	-81.1	-307.1	296.9
V	164.4	104.7	-19.5	1.7
VI	633.4	168.7	403.6	396.4
VII	673.8	-209.6	537.1	621.2
VIII	456.5	-112.6	533.1	563,2
IX	425.6	-56.6	394.7	595,3
X	211.8	-63.8	481.1	-

Table no. 2

Annual average temperature values, real heliothermic index, hydrothermic index and grapevine bioclimatic index during 2009-2012

Viticultural year	The normal annual average temperature (ºC)	The annual average temperature (ºC)	Real heliothermic index	Hydrothermic index	Grapevine bioclimatic index
2009	11.4	13.3	3.9	1	8.8
2010		14.5	3.8	1	8.0
2011		13.5	3.7	0.6	15
nov 2011 sep 2012		13.2	4.1	0.6	17

The climatic factors registered permited the determination of several indexes and coefficients used in viticultural climatology for the appreciation of the viticultural biotope characteristics, such as: real heliothermic index, hydrothermic index and grapevine bioclimatic index (Popescu, 2011).

For our country vineyards the values of the real heliothermic index (Huglin, 1978) ranges between 1-5 and is considered optimum for vine when its value is greater than 2.6. Normal values for the hydrothermic coefficient (Selleaninov, 1936) situated between 1 and 1.9 allow obtaining qualitative productions, values greater than 3 indicate an excess of moisture and a lack of heat and if the value reaches 0.6 – 0.7 vines should be cultivated in a irrigated regime. The grapevine bioclimatic index (Constantinescu et al., 1964) ranges between 5-15. Values lower than 5.7 indicate rich water resources, and values above 15 indicate vineyards with rich heliothermal resources and periods with deficient precipitations (Oslobeanu 1980, Book 1980).

Analysing climatic conditions for the year 2009 the annual average temperature has a value of 13.3 °C, the average temperature of the warmest month being 26.2ºC and the average temperature of the coldest month 1.7ºC. The sum of insolation hours has a value of 2181.3 and the total amount of precipitation is 565.0 mm of which 131.2 mm only in July (Figure 1). The beginning of the vegetation period is marked by a climatic accident, negative temperatures of -1.5ºC being registered on April 24, thus affecting the studied varieties. The Feteasca Neagra variety was the most affected. Variation in sugar content and acidity influenced positive the quality of the harvest, quantitative affected (Figure No. 2), with a yield in must of 72%. Regarding the values of the climatic synthetic indicators used to assess vine biotope we can say that the viticultural year 2009 was within normal limits (Table 2).

In 2010 the annual average temperature had a value of 14.5°C, with the average temperature of the warmest month of 29.6 ºC and the average temperature of the coldest month 0.5 ºC. The insolation registered a value of 1902.9 sunshine hours and the total amount of precipitation was 710.3 mm of which 211.5 mm only in July and 1.2 mm in August (Figure no.1). The climatic conditions during the vegetation period favorized disease and pests attacks, especially in July when heavy precipitations were registered, with a value of 211.5 mm compared to the average amount of 35.6 mm for this month.

Compared to the viticultural year 2009, this year recorded similar values concerning the quality given by the sugar content and acidity of the must (Table no.3), followed by a yield in must of 72% for all six varieties. Most sensitive variety was Cabernet Sauvignon.

Climate synthetic indicator recorded normal values for Murfatlar vineyard (Table no. 2).

The climatic conditions for 2011 were caracterized by a annual average temperature of 13.5°C, the average temperature of the warmest month being of 26.6°C and 10.5°C for the coldest month. The insolation recorded 2092,3 sunshine hours and the total amount of precipitation was 326,8 mm, slight below the normal annual average (Figure no.1).

Regarding the quantity and quality ratio (Figure no.2), this year was superior to the other years studied, sugar accumulation (Table no.3) have pleased oenological requirements, the yield given by the must recorded satisfactory values (75%). Also this year Cabernet Sauvignon recorded a lower value of the yield in must than the other six varieties. Synthetic climatic indicators recorded normal values only for the real heliothermic index, the other indicators rated the viticultural year 2011 as being arid, in which the water stress had a negative impact of plant physiological processes (Table no.2).

During November 2011 - September 2012 the average temperatures recorded the value of 13.2°C, with the average temperature of the warmest month being 28.0°C and the average temperature of the coldest month -2.3°C. The insolation recorded 2241.9 sunshine hours and the total amount of precipitation had a value of 361.7 mm of which mostly had fallen in January (95 mm) and May (145 mm) (Figure no.1). For this period the synthetic indicator registered values ranging the viticultural year 2012 as being very dificient in terms of water, thus the vine varieties were subjected to both water stress and heat stress (table 2).

The hydric stress affected the development of the berries and the weight of 100 berries. In consequence the average weight of the grapes (Figure no.2) registered values below the normal average weight for each variety, which led to lower yields in must (67%), the variety Feteasca Neagra being the most affected of all six wine varieties (Table no.3)

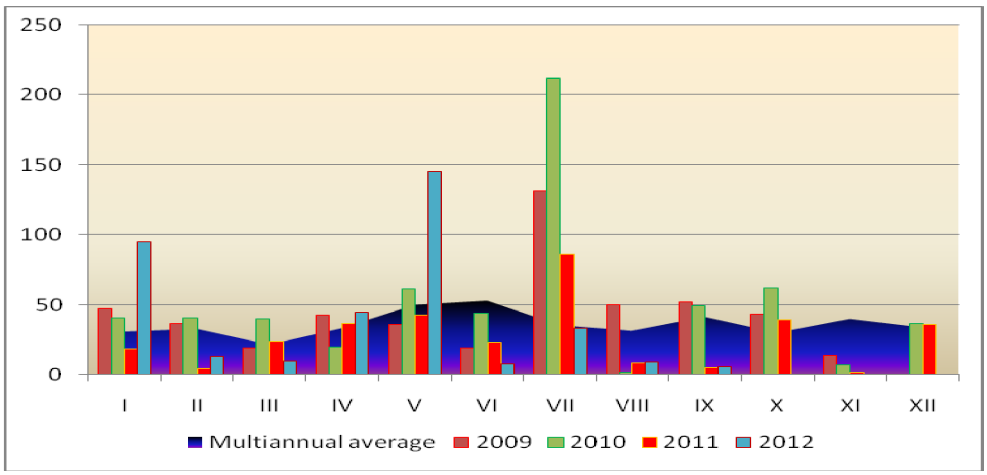

Figure no. 1 Amount of precipitation (mm) during 2009-2012

Table no. 3

The compared content of sugars, total acidity, weight of 100 berries and the must yield for the viticutural years 2009-2012

Variety	Viticultural year	Sugar content (g/l)	Total acidity (g/l H_2SO_4)	Weight for 100 berries (g)	Must yield (%)
Columna	2009	192	5.3	207	74.4
	2010	189	5.4	204	75.5
	2011	192	7.8	222	77.8
	2012	196	4.0	198	66.5
Pinot Gris	2009	226	4.7	136	72.7
	2010	217	3.8	146	67.5
	2011	225	4.2	162	73.1
	2012	238	3.0	111	62.4
Feteasca Regala	2009	210	3.5	142	71.7
	2010	206	3.6	163	75.0
	2011	211	3.4	156	75.4
	2012	215	3.3	131	69.6
Mamaia	2009	218	3.4	208	74.0
	2010	210	4.5	212	72.3
	2011	216	3.8	216	75.2
	2012	229	3.7	198	72.2
Feteasca Neagra	2009	223	3.6	104	70.3
	2010	212	7.2	116	70.8
	2011	226	3.6	127	73.2
	2012	232	4.5	102	70.8
Cabernet Sauvignon	2009	216	7.5	126	70.9
	2010	209	7.0	132	72.0
	2011	213	4.4	136	72.3
	2012	226	3.6	108	62.0

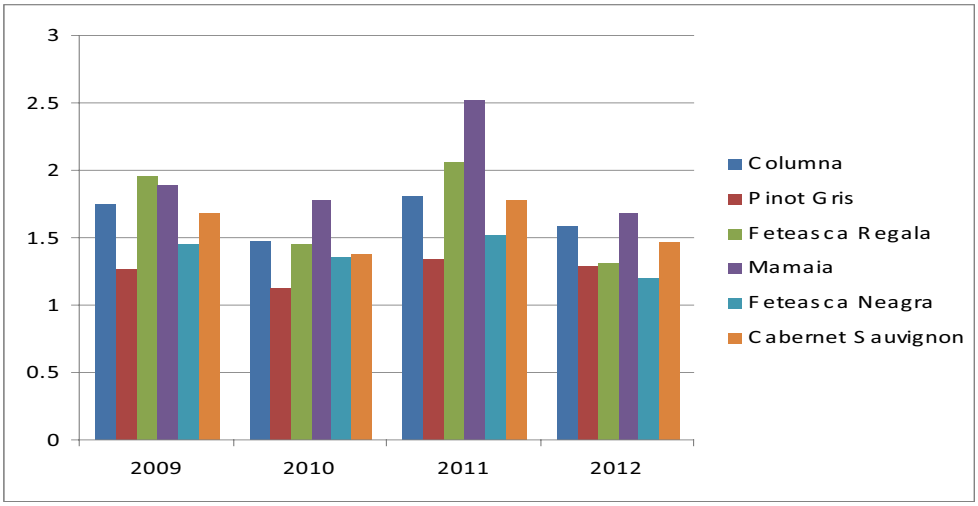

Figure no. 2 The average production (kg/vine stock) during 2009-2012

CONCLUSIONS

The viticultural years 2009 and 2010 had a poor quantitative and qualitative production, because of the great influence of the frost (2009) and excessive moisture during the period of ripening and full maturation (2010). By contrast, the viticultural year 2011 records higher production compared to other years under investigation. In terms of quality, the production of 2011 recorded lower concentrations of sugars in grape must compared to the year 2012, but kept a good quality. The highest sugar contents were recorded in 2012, but the high temperatures and the lack of rainfall have favored the instalation of hydric stress, the production obtained under these conditions being low due to the disturbance of the physiological processes. Linking climatic data with the results obtained for each variety we can say that the best viticultural year concerning quantity / quality was recorded in 2011 for all six varieties recommended for Murfatlar vineyard.

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THERMOVISION – AN IMPORTANT QUALITY ASSURANCE METHOD IN FOOD INDUSTRY

Roşca Adrian[1], Roşca Daniela[2]

Key words: *thermovision, machines elements, food quality assurance*

ABSTRACT

Thermovision has a wide range of application in food industry to control the temperature of perishable goods throughout production, transportation, storage, and sales.

The most important benefit of Thermovision over the other predictive quality assurance methods is easy infrared camera operation for process monitoring, and fast interprets of the results..

This paper presents examples of Thermovision applied in quality assurance in food industry.

INTRODUCTION

In the food industry, it's essential to carefully control the temperature of perishable goods throughout production, transportation, storage and sales.

Accurate, reliable, portable and easy to use, the infrared cameras provide the solution for monitoring temperatures at every stage.

Thermal imaging can also be used to detect problems in electrical and mechanical systems in the factory, warehouse, retail stores, and refrigerator trailers.

Excessive heating or cooling detected by using Thermovision can signal a problem in devices such as cooking vessels, ovens, heat exchangers, freezers, compressors and all the mechanical and electrical components of the food processing chains (motors and motor control centers, electrical connections, breaker panels, disconnect switches, substations, switchgear and circuit breakers) without disassembling equipment or disturbing operations (http://www.flir.com/thermography/, 2011).

Thermovision infrared cameras are capable of detecting temperature variations as small as $0,1^0$ C accuracy, that allows personnel to quickly detect out-of-tolerance temperature conditions and take action to prevent spoilage and loss of product stock.

Thermovision can often assist in identifying the actual cause of temperature problems, such as a faulty component, and suggest a course of action (Roşca, 2010).

Thermovision has a wide application in various fields such as industrial maintenance, engineering (mechanical, civil, electric), aerospace, fire and explosion hazards prevention, medicine, pharmacy, veterinary, agriculture and food industry (Vadivambal &. Jayas, 2011).

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MATERIAL AND METHOD

Infrared cameras are the ideal solution for assuring that temperature tolerances are maintained throughout all of the equipments operations, and in addition, can instantly reveal the operation condition of electrical components and machines' elements in the factory, warehouse, retail stores and refrigerator trailers (Roşca &. Roşca, 2011).

In order to perform the interdisciplinary studies in food process concerning *thermovision applications in food industry*, *thermovision predictive maintenance for large capacity milling process in food industry*, and *thermovision as a reliable method to prevent fire hazard in flour milling industry*, in the Unconventional Technologies and Equipment for Agro-Food Industry Laboratory within Faculty of Agriculture and Horticulture in University of Craiova, a FLIR Infrared Thermovision Camera (0,1^{0}C accuracy) was used (Roşca &. Roşca, 2011).

RESULTS AND DISCUTIONS

Flour milling is a mechanical manufacturing process which produces flour from wheat through comprehensive stages of grinding and separation. Gluten is the natural protein material which gives wheaten flour ability to make leavened bread and baked products, but during milling process must prevent water absorption in gluten. Therefore during the grinding, the operation temperature in all the milling process must not exceed 45^{o}C.

The quality of the roller mill is of decisive importance to the efficiency of the mill and must create the optimal conditions for excellent product quality and yield in the field of grain milling. Poor roller mills distance alignment requires static and dynamic balance measurements using a specialist shaft alignment system.

During grinding, as a result of breaking grains between the rollers mill, the milling process develops heat which generally adversely affect the process and the final produce quality.

To prevent the overheating during the grinding process there are made rollers mill equipped with internal water cooling system consisting in a cold water that enters through a pipe, then passes through the rolling / antifriction bearing cases, and passing through several nozzles, finally sprays the inner wall of the roller mill (http://www.buhlergroup.com., 2011).

In figure 1 is presented a roller mill with low quality maintenance of the internal water cooling system that cause fast increasing of the temperature more than 45^{o}C during operation. In this figure is observed the fast increase of milling temperature with maximum temperature during the operation up to 73,1^{o}C (Roşca &. Roşca, 2011; Roşca, Roşca &.Vlăduţ, 2012).

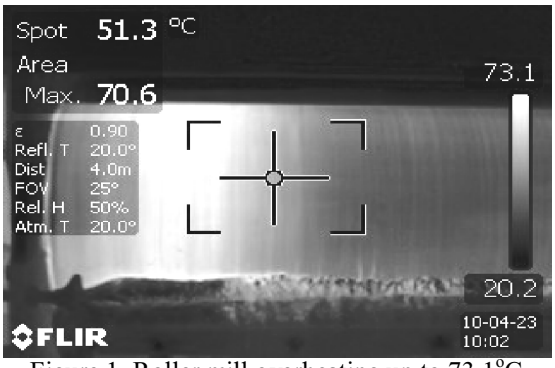

Figure 1. Roller mill overheating up to 73,1^{o}C

During the maintenance activity it was observed that water supply system filter and the manual valve were severe clogged, and the water flow in the pipe of cooling system was blocked; therefore, with no water flow in the inner of the roller mill, the milling process temperature was very fast increased more than 45°C. In figure 1, the left side is hotter than the right side with more than 30°C. It was proposed roller mills distance alignment measurements, and after static balance it was observed more than 0,5mm parallelism misalignment in the roller mill system (Roşca &. Roşca, 2011).

In figure 2 and figure 3 are presented IR thermal scanning in a small enterprise bakery. In figure 2 thermogram are observed heat losses more than 126°C when the oven is closed. After this Thermovision evaluation, the thermostat was metrological verified, and the mechanical system of oven gates was sated for a lower gap, with heat losses up to 80°C.

In figure 3 thermogram is observed the fresh bread cooling process during a winter season; the fast and intensive cooling process will determine a low bread quality when selling in the shop.

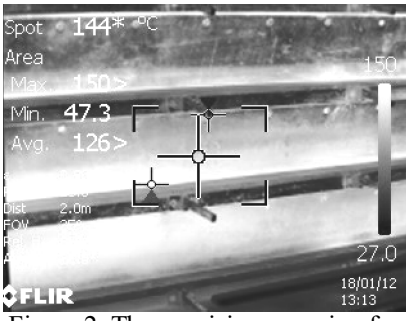

Figure 2. Thermovision scanning for closed oven heat losses up to 126°C

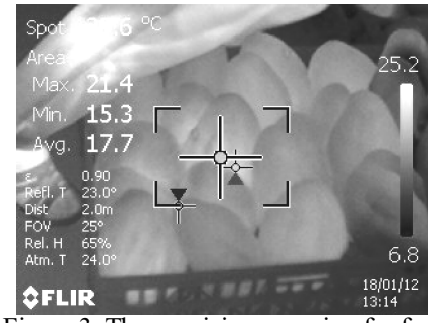

Figure 3. Thermovision scanning for fast and intensive fresh bread cooling process

In figure 4 is presented the temperature of cooked meat for a shaorma produce that must be stored / preserved at least 70°C. It is observed the produce temperature is only 35,6°C that will determine the microbiological problem and produce quality losses.

Figure 5 presents the wine conditioning temperature in a winery. The contact temperature gauge position on the vessel wall was above the liquid level and indicated only the inlet air temperature, but not the wine temperature that was 17,8°C. The wine storage temperature must to be maximum 12°C, and all the wine was low qualitative affected.

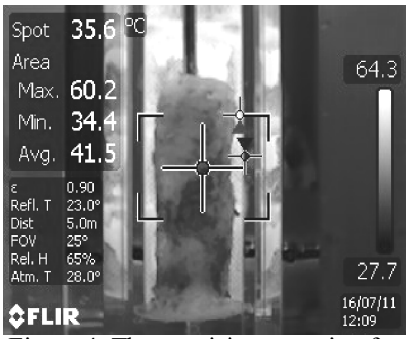

Figure 4. Thermovision scanning for shaorma stored/preserved at 35,6°C

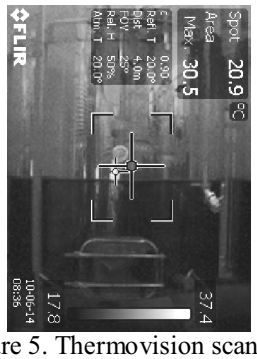

Figure 5. Thermovision scanning for wine storage temperature at 17,8°C

CONCLUSIONS

Thermovision is widely applied in all fields where temperature differences could be used to assist in evaluation, diagnosis, or analysis a process or product.

Thermovision can be applied in food processing, in predictive maintenance of food industry equipments and food products safety such as detection of foreign bodies in food material, temperature distribution during cooking and freezing process or in transportation and storing activities.

Thermovision has a large scale of utilizations in agriculture including planning irrigation scheduling, predicting water stress in crops, disease and pathogen detection in plants, predicting fruit yield, evaluating the maturing of fruits, bruise detection in fruits and vegetables.

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✓*Ingineria mediului*

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IDENTIFICATION OF MICROORGANISMS INVOLVED IN DISTILLED SPIRITS CONTAMINATION AND THEIR EVOLUTION DURING FINAL PRODUCT STORAGE

Ancuţa M. Rotar[1], Cristina Anamaria Semeniuc[1], Mihaela Duma[2], Nadejda Costru[1], Marinela Curliuc[1]

Key words: *Distilled spirits, contamination, moulds, storage, glycerol*

ABSTRACT

The aim of this study was to identify the microorganisms in distilled spirits and their evolution during storage of the final product using Standard microscopy and microbiology techniques. Because moulds were found in distilled spirits, selective media were used for TYMC determination. Strains of Penicillium, Oidium, Alternaria and Geotrichum were isolated from vodka and spirit drink samples. The initial presence of moulds in distilled spirits is a consequence of microaeroflora contamination. Glycerol used as ingredient offers protection against the effect of alcohol from distilled spirits. During the final product storage moulds consume glycerol, and no longer protected, are destroyed by the alcohol.

INTRODUCTION

Distilled alcoholic beverages contain ethanol between 20 and 50% alcohol by volume, derived from the distillation of agricultural raw materials subjected to alcoholic fermentation. Due to the wide range of fermented vegetal raw materials and manufacturing technologies, a variety of distilled spirits with specific sensory characteristics are available, which are consumed either as an appetizer or as a dessert drinks (Banu et al., 1999; Cioltean, 2008, 2009). According to Standards, distilled spirits must be sterile, free of microorganisms and without risk to consumer health. The processing units are completely responsible to this aspect (Cioltean, 2008, 2009). In the manufacturing of natural distilled spirit, the extractive substances used for raw material processing can influence the taste and aroma of the product. Ethanol, the main ingredient of natural alcoholic beverages, can be obtained by distillation or by synthetic pathway (Iovu, 2005).Vodka represents a spirit drink which is produced from ethanol of agricultural origin. It has a minimum alcohol content of 37.5 % alcohol by volume. The most important raw materials used are potatoes, different kind of cereals or molasses. The flavorings added in spirit drinks can be allowed only if the type of spirit permitted it (is not allowed in the case of vodka) and must be specified on the label.

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The main aim of this investigation was to identify the species of mould involved in the contamination of distilled spirits from a local distillery, the cause of the appearance and their evolution during the final product storage.

Table 1 shows the maximum levels of ethanol residues present in the two spirit drinks analyzed in our study (according to REG 110/2008).

Table 1

The maximum levels of ethanol residues present in spirit drinks

Maximum level of ethanol residues	Vodka	Spirit drink (not flavored)
Methanol, mg/100 ml pure ethanol	10	200
Esters, mg ethyl acetate/100 ml pure ethanol	1.3	1.3
Aldehydes, mg acetaldehyde/100 ml pure ethanol	0.5	0.5
Higher alcohols, mg 2-methyl-1-propanol/100 ml pure ethanol	0.5	0.5

MATERIALS AND METHODS

Sampling and experimental design

Samples of vodka and spirit drinks, with different alcohol content, were aseptically collected from a local distillery (Table 1) and tested for TYMC at 30 and 60 days of storage. Sealed bottles from the warehouse were used for distilled spirits sampling. Each type of colony was confirmed by microscopy.

TYMC test. The reference method (SR ISO 21527-1:2009) was used for enumeration of yeasts and moulds in distilled spirits samples. Media used were Dichloran Rose Bengal Chloramphenicol Agar, DRBC (Oxoid, Basingstoke, Hampshire, UK) and Dichloran 18% Glycerol Agar, DG-18 (Oxoid, Basingstoke, Hampshire, UK). Chloramphenicol (Oxoid Ltd., Basingstoke, Hampshire, UK) was added (100 mg/L) to the media as a selective agent to inhibit the growth of bacteria.

Preparation and dilutions of distilled spirits and drinking water samples was in accordance with ISO 7218:2007, SR EN ISO 6887-1:2002 standards. For each sample was prepared two successive dilutions. Tree Petri dishes was used for each dilution. An aliquot of 0.1 mL of the diluted sample was inoculated using a sterile pipette into each Petri dish containing DRBC and DG-18 media. Then, the Petri dishes were sealed in plastic bags (to avoid contamination) and incubated at 25 ± 1°C for 5 days.

After five days of incubation, the colonies are counted. The total number of yeasts and moulds is calculated using the formula specified in the standard:

$$N = \frac{\sum c}{(V(n_1 + 0.1 \times n_2) \times d} \quad \text{[cfu/mL]}$$

where:

Σc-colonies sum;

V-inoculum volume (mL);

n_1-Petri dishes number for first dilution;

n_2-Petri dishes number for second dilution;

d-dilution.

Macro- and microscopic characterization of colonies.

Macroscopic characterization

The following criteria were used: shape, edge aspect, size, colony profile, colour and colony type.

Microscopic characterization

Each type of colony was sampled using a microbiological loop and loaded on a glass slide, covered with sterile saline and a cover slip. The preparations were observed under a microscope (Motic AE31 microscope).

RESULTS AND DISCUSSIONS

In distilled spirit samples were identified different mould species: *Penicillium, Oidium, Alternaria* and *Geotrichum*. The predominant colonies belonged to *Penicillium* genus, subgenus *Furcatum*, with approximately 1 cm colony diameter, rough surface, green colour with a white border, circular shape and raised elevation (Fig. 1a). Figure 1b shows the microscopic image of *Penicillium* subgenus *Furcatum* with phialides that are ampulliform and shorter than their supporting metulae (Pitt and Hocking, 2009).

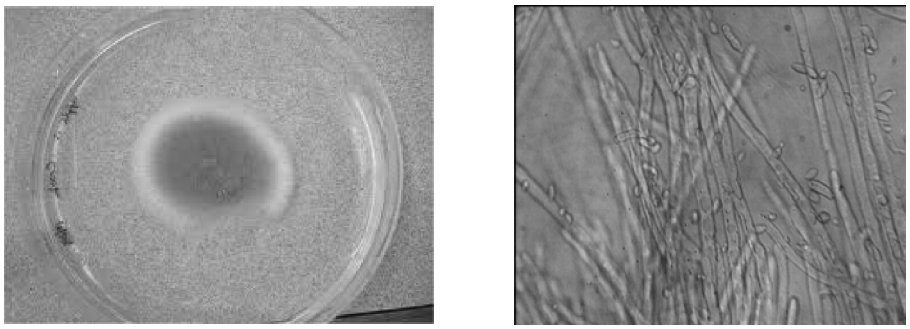

Figure 1a, b. *Penicillium* genus, subgenus *Furcatum* (macro- and microscopic images)

Colonies of *Alternaria* genus were also identified. Different sized colonies with brown color, rhizoidal form and edges with convex profile and type R rough surface (Fig. 2a). Under the microscope, ovoid conidia with a short conical beak and pale brown color (Fig. 2b) were observed. This saprophyte mold can be present in the freshly harvested grains, being an indicator for their freshness (Dan et al., 2009).

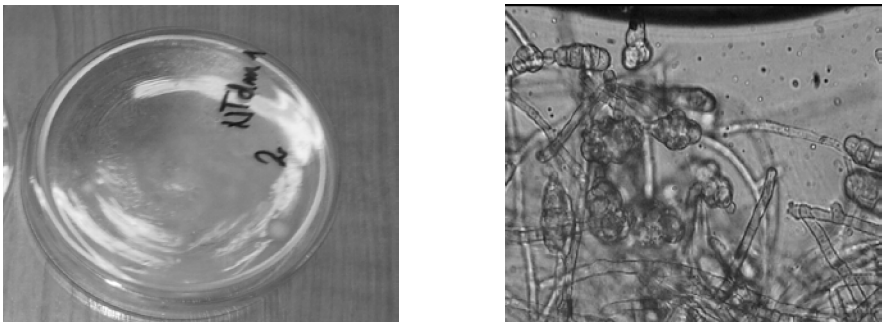

Fig. 2a, b. *Alternaria* genus (macro- and microscopic images)

Also white flat colonies with rhizoidal edges, hilly profile, and type R rough surface were identified (Fig. 3a). The colonies belong to *Oidium* genus. Under the microscope, *Oidium* looks like a string of beads (with conidiophores arising from a hyphal cell) (Fig. 3b).

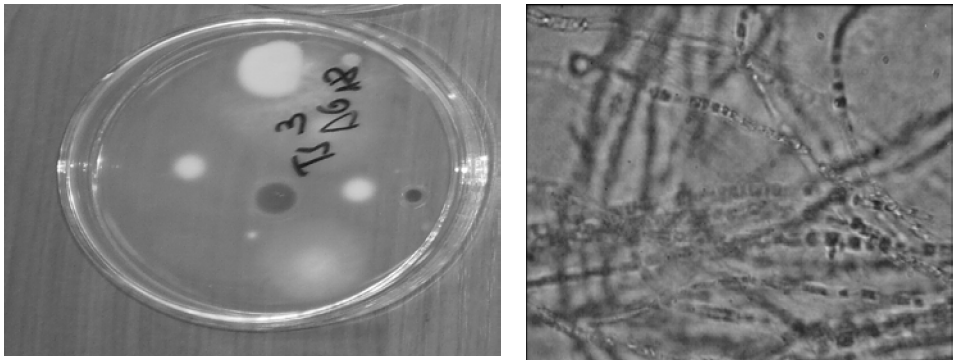

Fig. 3a, b. *Oidium* genus (macro- and microscopic images)

The last type of colonies identified belonged to *Geotrichum* genus and were characterized by white color, rhizoidal and filamentous edges, bulging profile and type R rough surface (Fig. 4a). Under the microscope was noticed the presence of hyphae with reproductive structures completely fragmented at maturity (Fig. 4b).

Geotrichum is a cosmopolitan fungus with a world wide distribution that can be isolated from soil and plants. (http://www.forensica.com/f/lab-services/microbiology/library/geotrichum.asp). It is a common contaminant of equipment and for this reason is called "mold machines". *Geotrichum* genus was identified in all distilled spirits samples.

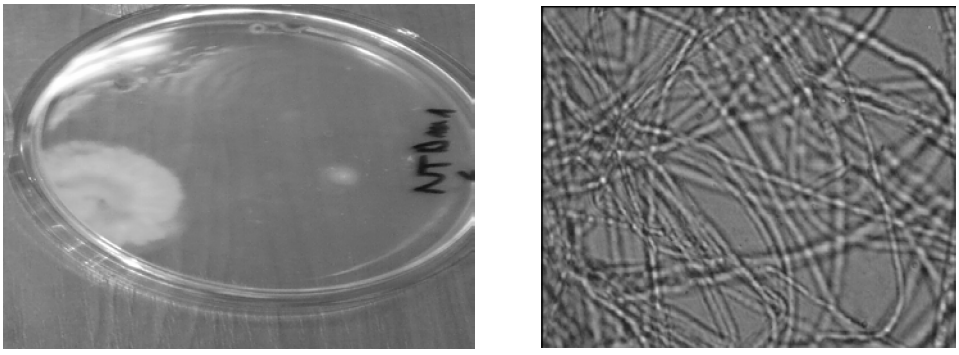

Fig. 4a, b. *Geotrichum* genus (macro- and microscopic images)

In our previous study (Rotar et al., 2012), microaeroflora from processing areas was identified as source of microbial contamination of distilled spirits. Table 1 shows TYMC in distilled spirits after obtaining (Rotar et al., 2012), at 30 and 60 days of storage.

Table 1

TYMC (cfu/mL) in distilled spirits

Sample	TYMC		
	After obtaining	30 d of storage	60 d of storage
Vodka 1, 40% v/v alcohol	7 cfu/mL	1.5 cfu/mL	absent
Vodka 2, 40% v/v alcohol	6.6 cfu/mL	1.3 cfu/mL	absent
Vodka 3, 37.5 % v/v alcohol	4.0 ufc/mL	2.5 ufc/mL	absent
Vodka 4, 37.5 % v/v alcohol	3.5 ufc/mL	2.1 ufc/mL	absent
Spirit drink, 26 % v/v alcohol	4.1 ufc/mL	0.5 ufc/mL	absent
Spirit drink, 26 % v/v alcohol	4.5 ufc/mL	0.5 ufc/mL	absent

A significant decrease of TYMC can be observed during distilled spirits storage. Studying the chemical composition of distilled spirits we noticed that glycerol, used as ingredient in distilled spirits, protects the moulds against the destroying effect of alcohol. Subsequently, glycerol (which is the nutritive substrate for moulds) is consumed during the final product storage; moulds are no longer protected, therefore are absent after 60 days of storage.

CONCLUSIONS

The presence of moulds in distilled spirits is a consequence of microaeroflora contamination. In distilled spirits *Penicillium*, *Oidium*, *Alternaria* and *Geotrichum* genus were identified macro- and microscopic. Good hygiene of production areas is imposed.

ACKNOWLEDGMENTS

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ISO 7218:2007-Microbiology of food and animal feeding stuffs-General requirements and guidance for microbiological examination

Regulation (EC) No 110/2008 of the European Parliament and of the CounciL of 15 January 2008 on the definition, description, presentation, labelling and the protection of geographical indications of spirit drinks and repealing Council Regulation (EEC) No 1576/89

http://www.forensica.com/f/lab-services/microbiology/library/geotrichum.asp).

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NATURAL OCCURENCE OF DEOXYNIVALENOL AND OCHRATOXIN A IN CONVENTIONAL MAIZE HYBRIDS AND THEIR BIOSAFETY COMPARED WITH GM EQUIVALENTS

Muhammad Sajjad [1]

Key words: *conventional maize, deoxynivalenol, ochratoxin A, food safety*

ABSTRACT

Grain samples from the harvest lot of 10 maize hybrids in the year 2011 were collected arbitrarily. Well ground and homogenized samples were analysed for the mycotoxins deoxynivalenol and ochratoxin A. Contamination rates and levels of DON and OTA were low and did not exceed the maximum levels indicating their possible safe use as food and feed under the EC regulation 1881/2006.The samples were further analysed for the possible effect of concentrations of mycotoxins upon that of starch and proteins. The study reveals the absence of any negative impact of presence of mycotoxins upon these biomolecules as their concentrations lie in the normal range. A comparative review of data for the mycotoxins in conventional maize grains invalidate the argument from the producer of GM maize hybrids that conventional hybrids fare inferior for food biosafety with respect to mycotoxins.

INTRODUCTION

Conventional maize hybrids are grown and consumed on a much larger scale than GM hybrids and are regarded as safe for human/animal consumption as well as for their environmental interactions due to their long history of persistence and utilization. They provide a baseline data for the limits of chemical composition,degradation time period for the biomass and its impact upon the environment. Under the familiarity based acceptance of a GM event, the comparison is made not only between the GM and its non transformed counterpart; but also to any published data of that plant species i.e. any cultivar of maize including publications predating World War II (Dolezel et al. 2009).

Diseases of maize plants caused by Fusarium species occur worldwide, causing lodging, significant yield losses and qualitative loss of the harvested produce through contamination with mycotoxins. Fusarium a general class of fungi produces deoxynivalenol (DON) in maize grains mainly at the ear stage. The occurrence of DON is primarily associated with *Fusarium graminearum* (*Gibberellazeae*) and its concentration is significantly affected by the susceptibility of cultivars towards Fusarium species, weather

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conditions, previous crop culture, cultural practices and use of fungicides (Thirtle et al., 2003).

Ochratoxins are a group of mycotoxins produced as secondary metabolites by several fungi of the Aspergillus or Penicillium families. The ochratoxin A production on cereals and derivates is the consequence of the interaction of different factors such as fungal species, composition of the substrate and the environment including oxygen, temperature, humidity, carbon dioxide, pH and incubation time . Ochratoxin is considered to be nephrotoxic, immuno toxic and carcinogen.

The maize grains have the capacity to accumulate large quantities of carbohydrates and proteins (upto 13%) by weight and the GM maize events are under development to enhance their accumulation by using seed specific promoters. A higher starch content in newly developed GM hybrids results in a slow decomposition of the maize crop left over and the availability of soil nutrients to the following crop plants may be compromised. Today from the GM producer companies, the introduction and utilization of new GM IR/HR food grain events is pretexed on grounds that it is more biosafe than the conventional food grains. The view point that GM grains contain less concentration of mycotoxins as compared with conventional hybrids necessitates the analysis of level of mycotoxins in conventional hybrids to establish the degree of biosafety of the existing cultivars.

The following graph gives the comparative concentrations of mycotoxins in conventional and YieldGard hybrids commercialized in Romania (Monsanto 2009). The concentrations are expressed in µg/kg.

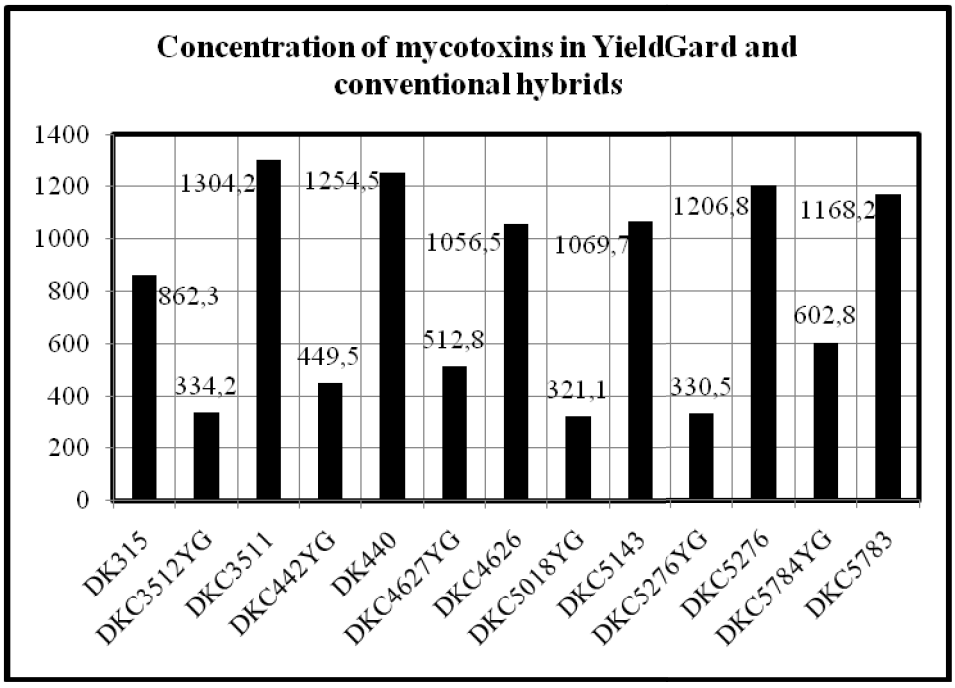

Fig.1. The comparative difference of cumulative concentration of mycotoxins in YieldGard and conventional maize hybrids. Source: Monsanto (2009)

The results indicate the following observation:

"The YieldGard hybrids always exhibit a lower concentration of mycotoxins as compared to conventional hybrids -both iso/ non-isogenic hybrids".

However, the data does not indicate the type of mycotoxins analysed as there is a multitude of toxin-causing fungi which damage the maize plant and grains.

Moreover, the concentrations of mycotoxins may not be reduced in resistant maize under certain environmental conditions or if ear injury has been caused by agents other than insects. The following experiment was designed to evaluate the safety of conventional maize hybrids for the levels of specific mycotoxins and concentration effect of these toxins on major cellular biomolecules.In this experiment, the comparative biosafety of the conventional maize hybrids was analysed for concentration of DON and OTA and their effect on amounts of starch and proteins. The purpose was to review an overall natural resistance of conventional hybrids to resist contamination with mycotoxins and to compare the biosafety grade of GM and non GM maize hybrids from the view point of fungal pathogenicity.

MATERIALS AND METHODS

Samples of conventional maize grains were obtained at the harvest time at the Fundulea Research Institute, Fundulea in September 2011.For DON, 1 kg sample was sieved by applying ¼ method till a laboratory sample of 100g was obtained.The sample was ground. A mixture of 12.5 g/25 ml distilled water was prepared and agitated at 200rmp/30 min./25 ^{0}C.The filtered elute was centrifuged.After centrifugation and filteration,the elute were extracted through acetonitrile+methanol+water (5:5:90). A 100ml elute was injected into HPLC apparatus for quantification of the mycotoxin. The limit of detection (LOD) for DON was 0.02 µg/ml.

For OTA, the samples were prepared according to standard procedure. Dilution was carried out with 44 ml tampon phosphate 20mM pH 7,0 followed by washing through Ochraprep immunoaffinity columns. After the segregation of molecules,100 µl was injected for chromatography. LOD was kept at 0.005 µg /ml. The data for DON and OTA was obtained and worked upon using the software Empower.

The amount of proteins was measured through the standard Kjheldal method while the concentration of starch was measured by Lippich polarimeter by using the expression: Starch %=$\alpha \cdot Vt \cdot 100 / [\alpha]^{20} D.l.p$ where α =measured angle, Vt=sample volume, $[\alpha]$ = standard angle (183.7 for maize), l = tube length (20 cm), p = sample mass (g).

RESULTS AND DISCUSSION

Deoxynivalenol detection and quantification for the 10 non transgenic varietal samples revealed only one positive sample. With respect to EU regulations for fungal toxins in food stuffs (EC No.1881/2006), the samples deem suitable for use as food and feed (table 1).

The results indicate that the concentration of Deoxynivalenol in nine varieties is below the detection limit of 0.02 µg/ml. Only the hybrid Mostistea contains identifiable levels of DON being at 143µg/kg. However, no influence of these concentrations is found on amounts of starch and proteins (figure 2, 3).

Eight of the 10 selected varieties were analysed for the concentration of Ochratoxin A. As the samples were obtained on the harvest day, the detected concentrations correspond to accumulation of OTA in the field conditions. Certain mycotoxins e.g. hepatotoxins are known to disrupt the cellular functions via oxidation of key proteins. However in this study, no influence is established for DON and OTA upon the levels of starch and proteins. The obtained data values show no significant changes in concentration of starch and protein quantities in any of the varieties as all values lie within the normal concentration range of these biomolecules in maize grains. Only the variety F-475 showed a higher value for OTA than the value permitted for infants.

Table1

The data for concentrations of OTA, DON, starch and proteins

Variety	DON (µg/kg)	OTA (ng/g)
Crisana	< LOD	N/A
Milcov	< LOD	< LOD
Mostistea	143	0.06
Olt	< LOD	0.24
Rapsodia	< LOD	< LOD
F-322	< LOD	<LOD
F-125-06	< LOD	< LOD
F-254-08	< LOD	0.12
F-475	< LOD	0.03
F-225-06	< LOD	N/A

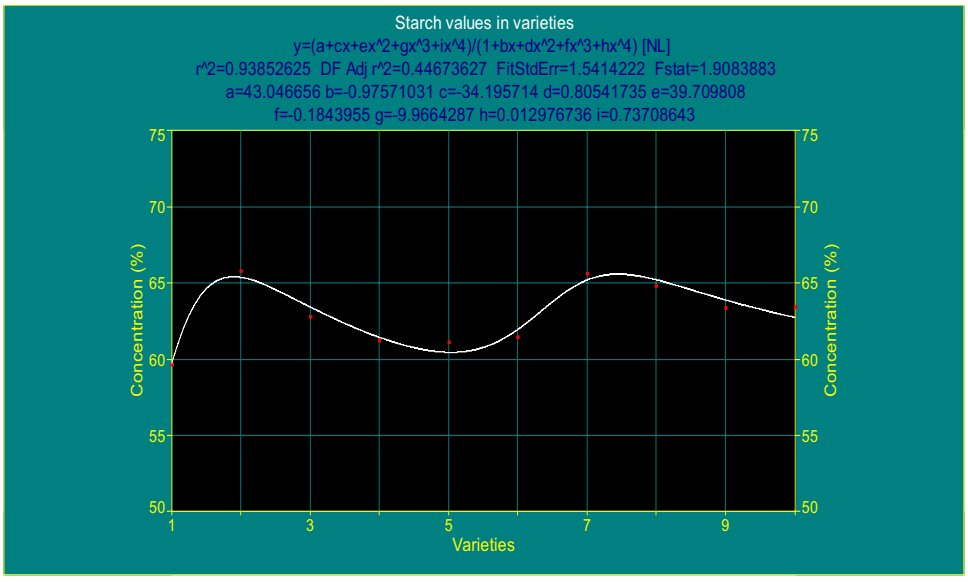

Fig.2. The variation of concentration of starch content in varieties
Mean=62.952 std.dev.=2.072

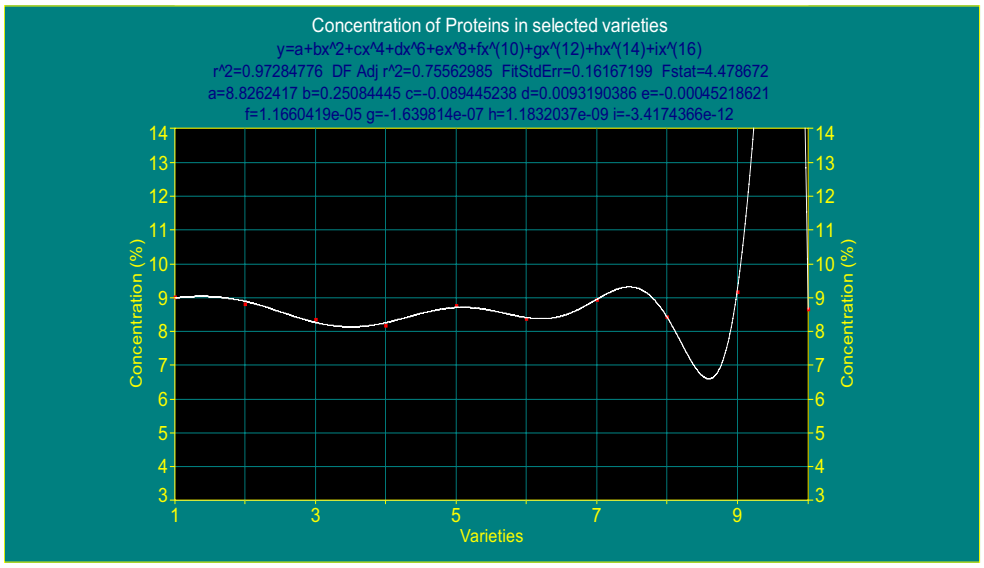

Fig.3. The concentration of proteins in samples
Mean=8.675 std.dev.=0.326

The data is in conformation with previous results obtained in 2007 when 43 samples were collected and analysed for the presence of OTA.The results indicated that samples were positive in a significantly big proportion i.e. 24 from 30 of samples (80%). Only two positive samples had values for Ochratoxin A higher than the admissible value of 5 ng/g (EC 1881/2006),the levels being at 19.92 and 11.72 ng/g. Rest of the positive samples were below than 2 ng/g (Muhammad and Cristina,2011).

A review of the peer studies upon the resistance potential of genetically modified maize hybrids reveal varying opinions. The results of the above experiment support the view that conventional crops are no inferior to GM counterparts with respect to permitted levels for fungal toxins. The studies by Florentina et al.,2009 revealed that out of 125 samples of conventional maize grains (samples obtained from five wide apart counties viz: Galati, Bacau, Oltenia, Timisoara, Bihor),only 25 % samples were positive for DON and only 23% samples were positive for OTA. Moreover, the majority of the samples were found fit for human and animal consumption. Similarly in the studies by Tabuc et al., (2009) conventional maize hybrids (no of samples=54) did not contain neither fumonisins nor ochratoxin A. For an overwhelming majority of the samples, the measured values were either below the limit of detection or under permitted concentration for consumption.

CONCLUSIONS

As the interaction of fungi upon maize crop is a complex phenomenon and yet not understood well, the role of transgenes for conferring the resistance against fungal pathogens is not yet established. If on one hand the data from the producer company has shown the reduced level of mycotoxins in Bt maize as compared with conventional hybrids, then on the other hand conventional varieties also exhibit fairly low levels for them. The concentration of mycotoxins depends upon a number of agronomic and environmental factors such as insect attacks, abundance of weeds, humidity, temperature & moisture

content in grains at the time of harvest etc. The results provide crucial evidence that the newly developed conventionally bred hybrids exhibit fairly low levels of mycotoxins in grains. The reduction in mycotoxin concentration needs to be further analysed in a variety of experiments involving ecological, agronomic and germplasm relationships for controlling or reducing the extent of fungal contamination. Such data is helpful in making decision about the biosafety status of submitted GM maize events in new regions and countries such as Pakistan where VT Double Pro GM maize event (*cry1A.105+cry2Ab2*) is under trials for the 3rd consecutive year.

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MANAGERIAL PROBLEMS RELATED WITH THE INTRODUCTION AND UTILIZATION OF GENETICALLY MODIFIED CROPS IN PAKISTAN

Muhammad Sajjad Ali[1]

Key words: *GMCs, adoption, evaluation, regulatory guidelines*

ABSTRACT

Pakistan is an agricultural country and a new member of the countries which have commercialized genetically modified crops. Crop biotechnology is not a solution itself but a useful tool in solving the agricultural problems. The adoption of GMCs, if not accompanied with proper regulatory protocols, may result in not only monetary loss but environmental ill effects as well. In this review, an analysis of the regulatory issues related with the introduction of products of modern biotechnology in the environmental conditions of Pakistan is carried out with the aim of postulating guidelines for their management.

INTRODUCTION

Agriculture is the largest economic sector of Pakistan and contributes 25% to GDP. Out of a reported area of 59.32 million hectares, only 21.92 million hectares is cultivated while 24.62 million hectares is not available due to one reason or the others. Since the arrival of multinational pesticide companies in early 1980s, not only a culture of heavy and indiscriminate spraying on crops was promoted but the degradation of ecological resources also took place. Excessive use of fertilizers and pesticide sprays has led to serious environmental hazards and health problems for the rural community and wildlife (figure 1). Moreover, chemical insecticides are generally not very effective as compared with the control achieved by Bt-crops (PARC, 2008). Yield losses are lower in Bt-crops than in pesticide treated crops.

As shown in other adopting countries, crop biotechnology if adopted with proper guidelines and safety measures regarding their impacts on the environment, then there will be real socio economic benefits (RAO, 2007).In order to enhance the productivity of food and cash crops, biotechnological approaches can play an essential role especially for the adaptation of crops in problem soils, enhanced nutrition (e.g. sugar content in sugarcane and sugarbeet) and reduction in use of pesticides etc. The present study focuses on the nature and developments in agricultural biotechnology and their management in the domestic agricultural environment.

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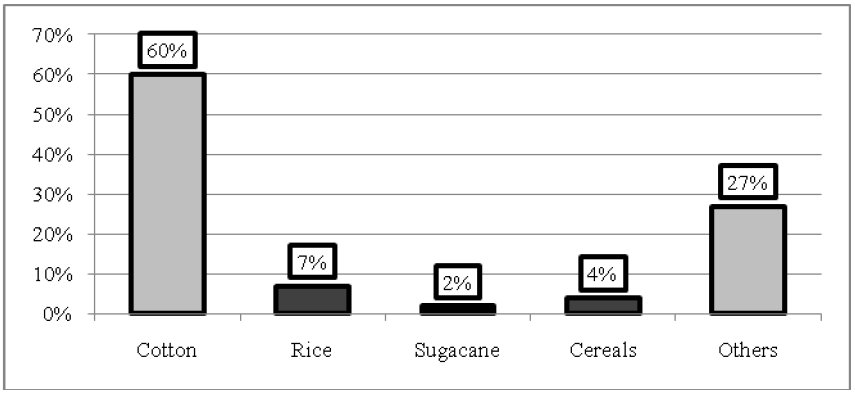

Figure 1: Consumption of pesticides on various crops in Pakistan

MATERIALS AND METHODS

The methodology for review and data collection consisted of investigation of relevant literature at the local and global level where the GM crops are grown and considered as the main source of farmer's income. Information was also collected from the available progressive growers, researchers, extension workers and policy makers through personal communication and interactive on-farm dialogue.

RESULTS AND DISCUSSIONS

CURRENT SITUATION OF ADOPTION

The official adoption of the first GM cotton crop has been delayed due to legal complexities, but the institutions are engaged in high tech biotech research in novel areas of breeding, mutation and insertion of desirable traits in locally developed field crops e.g. cotton, maize, rice and sugarcane. Because Pakistan occasionally imports large amounts of agricultural commodities, the potential benefits of adopting crop biotechnology are greater. It is because the countries with low adoption rate and high imports of commodities benefit from the resulting reduction in the prices of these commodities at the world level (figure 2).

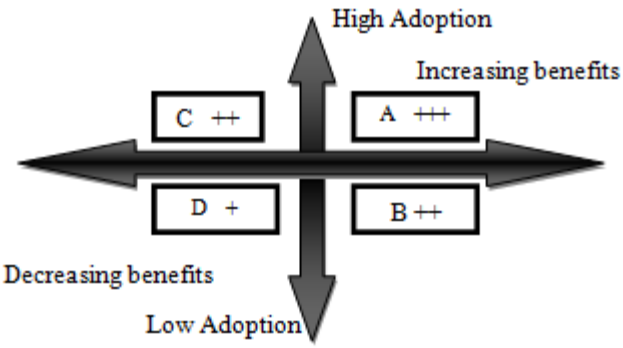

Figure 2: The adoption quadrants for socioeconomic effects of GM crops
source: adopted and modified from Abare, 2001.

The sign + represents the level of adoption of GM technology. The positive signs indicate the status of adoption, three signs being the highest adoption status. The quadrant A represents a situation in which there is a high adoption of GM crops in a country. The agricultural commodities receive a reduction in prices and become more competitive in domestic and international markets. Currently the situation in Pakistan regarding the adoption of GM technology lies in quadrant B moving towards A. Both agriculturists and govt. institutions are probiotech and actively participate in the developmental programmes for the GM crops. At present, about 29 institutions are engaged in biotech research. However, due to a chronic lack of funding and infrastructure, access to elite germplasm and resources for access to laboratory equipment the pace of research has been rather slow. Only prime institutions like NIBGE, NIAB and NCEMB etc possess highly trained personnel and developed labs (table 1).

Table 1

Development of Bt cotton varieties by different organizations in Pakistan

Company/ Research Institute	Developed Bt Variety
M/s Guard Agricultural Research Services, Raiwind Road, Lahore	GM-2085
M/s Ali Akbar Seeds, Multan	Ali Akbar-802, Ali Akbar-703,
M/s Nawab Gurmani Foundation, Kot Addu	Sitara-008
M/s Agri Farm Services, Multan	MG-6
M/s Neelam Seeds, Multan	Neelam-121
Cotton Research Institute, AARI, Faisalabad	FH-113
Nuclear Institute for Bio-Technology and Genetic Engineering (NIBGE)	IR-3701, IR-1524
Center of Excellence in Molecular Biology (CEMB),Lahore	CEMB-1, CEMB-2

Bt cotton is the only GM crop which was commercialized in 2011 and the production has been a record with cotton bales of 150 million. The adoption not only provided with socio economic benefit to farmers community but also gave a boost to the national exchequer.

MANAGERIAL PROBLEMS

The management of genetically modified crops comprises of their evaluation and adoption under a sound regulatory regime which monitors the interactions of a GM crop in its local and broad environment i.e. it covers the environmental and food/feed biosafety aspects. Similarly, the adoption of GM crops must not cause socio economic percussions for the growers in the receiving agricultural system.

The adoption of Bt cotton has not been straight forward and simple as there has been reports of illegal Bt cotton growing as far back as in 2002-2003.A large amount of germplasm was smuggled from other countries and hybridised with the local varieties. A rampant adoption of non-uniform Bt germplasm not only posed a threat of resistance build-up in target lepidopteran insects but also created intellectual property issues for the

germplasm imported for research purposes only. Major issues regarding the introduction and utilization of GMCs in Pakistan include the following (figure 3):

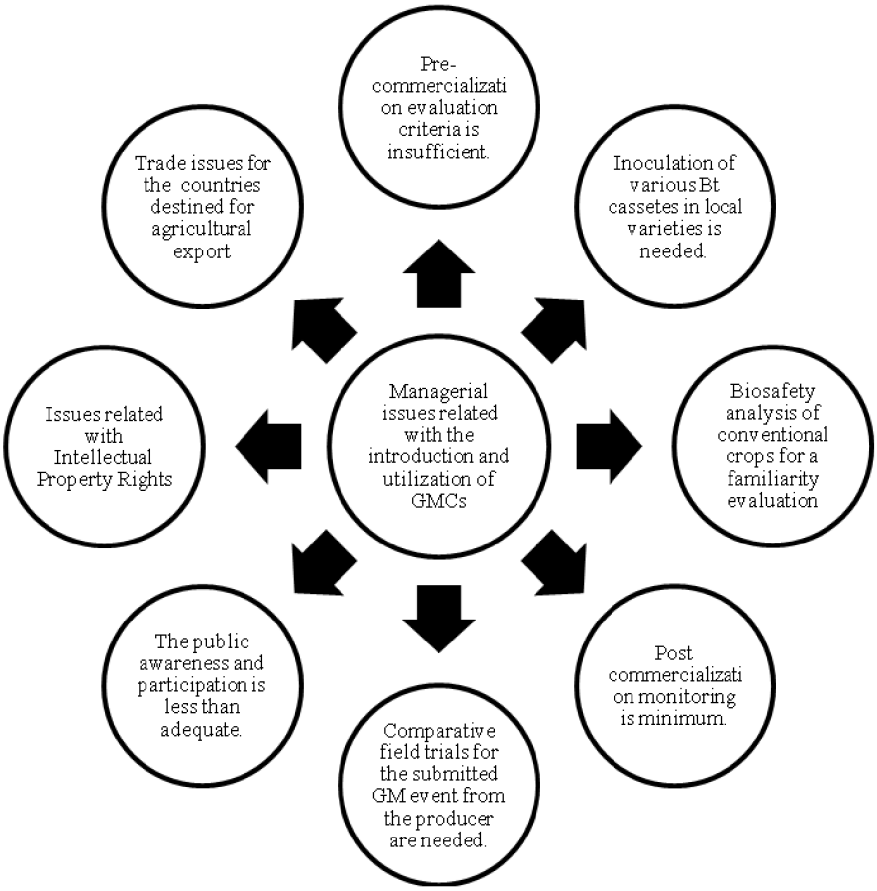

Figure 3:Issues related with the adoption of genetically modified crops in Pakistan

The approval and introduction of GM events in agricultural landscape of Pakistan is still in early stages.GM crop events are available from international and national research institutions; however due to a slow legislative process to determine the economic and ecological outcomes of commercialization, the rate of adoption and spread of GM events in the country is far from adequacy. If on one hand, there is a dearth of studies upon country wide farm level socio-economic studies after the formal nod to GM Bt cotton cultivation, ecological consequences of the introduction also need to be determined on well constructed environmental assessment criteria. Pre-introduction phase needs a comprehensive assessment criteria including food biosafety especially. It is so because today the new GM events submitted for wide scale cultivation are evaluated on productivity and economics with a relatively ignored position on safety of the crop for direct human and animal consumption. Moreover, the post commercialization monitoring is literally inexistent and no monitoring/ regulatory authority is responsible for conducting such studies. There are reported cases of increase in the frequency of resistance alleles in target pests in target in

countries where GM cotton was adopted with a disregard to proper environmental safety measures i.e. the refuge crop. In Pakistan, no refuge concept is being propagated either at the Government and private sector. The planting of Bt crops without it may lead to a catastrophic failure of the Bt technology in Pakistan with a re-introduction of biopesticides and ultimately chemical sprays. The case is further aggravated with the imminent release of GM crops with same or similar transgenes i.e. GM maize and GM rice.

The familiarity based concept for pre approval evaluation scheme holds significance where a GM crop is evaluated against a particular set of properties of its non-GM isogenic/near isogenic counterpart. Similarly the establishment of relationship between compositional difference among conventional and GM varieties in relation with monitoring of toxin variation (e.g. cry toxin in MON531) will lead to an ease in acceptance for GM technology. There are insufficient assessment criteria for new GM crops before approval for their commercial planting. But, because uncertainties exist, it is useful to establish some probabilistic framework and compare expected gains and costs. While considering the environmental and health costs of biotech crops, it is important to assess these costs relative to the costs that they exclude. The extra yield association with GM crops also has to be balanced against the extra costs. Therefore, using a more holistic approach to regulating biotech crops, by comparing all benefits and costs, may lead to elimination of some of the regulatory restrictions on these crops.

CONCLUSIONS

The ecological management under a sound legal regime coupled with biosafety analysis of the GM produce can lead to the improved welfare of the farming community. It is essential to integrate these aspects into existing monitoring/regulatory system. It will help prevent environmental contamination from undue /unwanted outcomes of the presence of GM foods and economic improvement of agricultural sector. The analysis of Pakistani monitory regulations reveals the need for implementation of following activities in Pakistan:

Constitution of the National Biosafety Framework to implement international agreements and regulations effectively and to align existing laws and regulations accordingly;

Assuring the legislative framework upon import/export of GMCs, their labeling and traceability in food and feed;

Constitution of a National Catalogue (also available online) of GMCs accepted in Pakistan for commercialization or trial evaluation;

Capacity building and infrastructure development of the laboratories specialized in detecting the GMCs;

Formulation and implementation of the legislation for Post commercialization monitoring of GMCs;

Assuring the functioning of institutional framework established through different bilateral and multilateral agreements;

Properly incorporated, the above management guidelines can pay the way to a rapid and safe introduction of GMCs. The crops will pose minimum ecological hazards and the socio economic dividends will be distributed in a justified manner.

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Vol. XVII (LIII) - 2012

ROOTING METHODS OF SHOOTS TO RASPBERRY AND BLACKBERRY THORN LESS OBTAINED IN VITRO

Sava Nina [1]

Key words: *shoot, peat, hydroponics*

ABSTRACT

The work includes a series of experiments the capacity of rooting and acclimatization of shoots of raspberry and blackberry without thorns derived from multiplying in vitro. Multiplication in vitro to produce a large number of shoots, which then can be rooted in pots kept in climatic chambers, in greenhouses and rooting substrate Hydroponic culture. The three methods are characterized by certain root and leads to different percentages of plants rooted and acclimatized. By choosing the correct method of rooting is obtained a large number of plants.

INTRODUCTION

The studies were performed to establish effective method of rooting shoots of blackberry and raspberry. Intended rooting shoots were small and came from in vitro multiplication. The study combines the modern with the traditional multiplication. We studied five varieties of raspberry and blackberry three varieties without thorns. We studied the ability of rooting buds and raspberries in growth room where there was a very good response and greenhouse conditions. Here the results were poor because growth factors can be controlled. Roots in the hydroponics system ensures the best results and rooted shoots was about 30 days.

MATERIALS AND METHODS

In vitro shoots were divided into two lengths to emphasize the influence of length on rooting ability. Rooting substrate was a mixture of peat and perlite. For this experience were made for each variety three repetitions with 30 pots for each repetition and category length of shoots, resulting in 90 shoots for each length of shoots, shoots and 180 shoots each variety in the length of 2 to 5 cm. Rooting methods were tested: the ex vitro rooting type Jiffy pots, covered with transparent foil, kept in growth chamber, the ex vitro rooting type Jiffy pots placed in the greenhouse, rooting Hydroponic culture. The varieties studied were varieties of raspberry Citria, Heritage, Willamette, Norna, Polka and blackberry varieties thorn less Silvan, Orkan, Thorn Free.

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RESULTS AND DISCUSSION

In ex vitro rooting Jiffy pots covered with transparent foil, kept in growth chamber. Shoots transferred to rooting and acclimatization in type Jiffy pots survived 100% in the first phase, and then the percentage of rooted plants had lower values results, depending on the method of rooting.

Table 1

Rooting and acclimatization ability of seedlings obtained from the 5 varieties of raspberry

Variety	Rooting capacity (%)		Acclimatization capacity (%)	
	shoots 2-3cm	shoots 3-5 cm	shoots 2-3cm	shoots 3-5 cm
Citria (Mt)	61	63	58	61
Heritage	55	55	52	50
Willamette	58	60	55	54
Norna	60	59	58	55
Polka	57	61	54	58

Table 2

Rooting and acclimatization ability of seedlings obtained from the three varieties of blackberry thorn less

Variety	Rooting capacity (%)		Acclimatization capacity (%)	
	l shoots 2-3cm	shoots 3-5 cm	shoots 2-3cm	shoots 3-5 cm
Silvan	62	64	60	62
Orkan	58	60	58	58
Thorn Free (Mt)	60	60	58	60

Due to the increasing use of camera and polyethylene film for extra moisture control rooting percentage ranged from 55% (Heritage) and 61% (Citria) for shoots of 2-3 cm long and between 55% and 63% for shoots of 3-5 cm length. This proves the high capacity of rootedness of this species under controlled conditions. Loss of plants during acclimatization were small, between 2% and 6% (Willamette, shoots of 3-5 cm), making the yield micropropagation of this species to be up (Table 1 and 2).

If blackberry varieties have obtained similar results, rooting percentage ranging from 58% (Orkan, shoots 2-3 cm) and 64% (Silvan, shoots of 3-5 cm length) .

In ex vitro rooting in Jiffy pots, placed in the greenhouse on the rooting platform. For this experience, conditioned on the two categories shoots length were planted in pots, which were then arranged on the platform directly rooted in greenhouse conditions.

Capacity in vitro rooting of shoots directly past the rooting substrate in the greenhouse decreased, rooting percentage ranging between 31% and 38%. From this time the results were not very dispersed, reaction raspberry varieties to be relatively similar rooting conditions.

Table 3

Rooting and acclimatization ability of seedlings obtained from the 5 varieties of raspberry

Variety	Rooting capacity (%)		Acclimatization capacity (%)	
	shoots 2-3cm	shoots 3-5 cm	shoots 2-3cm	shoots 3-5 cm
Citria (Mt)	32	33	30	30
Heritage	35	39	33	36
Willamette	38	34	35	33
Norna	31	38	30	35
Polka	37	30	36	30

Table 4

Rooting and acclimatization ability of seedlings obtained from the three varieties of blackberry
without thorns

Variety	Rooting capacity (%)		Acclimatization capacity (%)	
	shoots 2-3cm	shoots 3-5 cm	shoots 2-3cm	shoots 3-5 cm
Silvan	32	40	32	40
Orkan	28	36	27	25
Thorn Free (Mt)	44	31	40	38

Witness to one variety (Norna) showed significantly lower values of percentage of root while the root variety Willamette 6% more than in controls. Shoots from 3-5 cm long, rooting percentage ranged from 30% (Polka) and 39% (Heritage). The witness had average values (33%) for the categories of shoots. What was noticed was that 2-3 cm long shoots were rooted in a proportion lower than 3-5 cm long shoots from three varieties (Citria, Heritage, Norna).

This decrease in the percentage of rooting can be explained by a weak control of atmospheric humidity in the rooting medium, namely a more pronounced loss of water from the substrate. The blackberry varieties without thorns rooting percentage decreased from the first experience. Rooting percentage was between 31% and 44%, losses being 69% and 56%.

Hydroponic culture rooted. In this culture system, the first roots appeared after 10-12 days, and roots was fast, so after about a month shoots were rooted. After acclimatization seedlings were planted in pots with peat and perlite substrate.

Under this system the percentage of rooted plants was between 65% (Heritage) and 70% (Citria and Norna), if we refer to shoots of 2-3 cm length. In this time of the reference variety registered the highest percentage of rooting, being matched by the variety Norna.

Table 5

Rooting and acclimatization ability of seedlings obtained from the 5 varieties of raspberry

Variety	Rooting capacity (%)		Acclimatization capacity (%)	
	shoots 2-3cm	shoots 3-5 cm	shoots 2-3cm	shoots 3-5 cm
Citria (Mt)	70	66	70	63
Heritage	65	62	64	62
Willamette	68	70	67	65
Norna	70	70	65	62
Polka	67	68	62	68

Table 6

Rooting and acclimatization ability of seedlings obtained from the three varieties of blackberry
thorn less

Variety	Rooting capacity (%)		Acclimatization capacity (%)	
	shoots 2-3cm	shoots 3-5 cm	shoots 2-3cm	shoots 3-5 cm
Silvan	73	74	70	73
Orkan	70	71	65	69
Thorn Free (Mt)	72	73	70	70

CONCLUSIONS

Of the three methods studied best results were obtained from roots in Hydroponic Culture. Roots began to emerge after 10-12 days.

The rooting in the greenhouse gives the lowest percentage of rooting, because the vegetation factors can t be controlled. In the growth chamber to obtain good results, close Hydroponic culture.

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RASPBERRY PROPAGATION BY YOUNG SUCKERS

Nina Sava[1]

Key words: *suckers, raspberry, multiplication*

ABSTRACT

The raspberry bush is very easily multiply vegetatively, but many growers use during the breeding season. Number of suckers per plant formed as the plant grows older. Research has shown that harvesting suckers should be made when the plant is well developed. At the end of each variety of research has established the ability to form suckers and suckers to calculate the number of young people can be obtained per hectare. Mother plants were studied for 3 years under field conditions, and then set young suckers catching ability. Well-trained field facility equipped with wet grip is easy young suckers. The results were compared with other methods of multiplication.

INTRODUCTION

The studies were performed to clarify the effectiveness of conventional methods of multiplication of raspberry varieties. Research has shown that the methods applied and known can use other methods. In the literature there are few data on industrial multiplication raspberries, and this is because the consumption of fruit is becoming greater. Demand for planting material is increasing and therefore effective method specification is important in practical terms. Multiplication by young suckers was compared with another traditional method, but proved to be more demanding and apply for a short period of time. Multiply by young suckers is recommended to apply at the beginning of the third year of vegetation, the parent plant is well-formed.

MATERIALS AND METHODS

Biological material was represented by five raspberry varieties with different backgrounds. They studied two methods of multiplication, but the work refers to multiplication by young suckers. The studies were conducted under field conditions and the first two years were made observations on the capacity of each variety to form suckers. For this research were collected suckers with sizes from 5-15 cm long, were prepared for planting were planted under field conditions to specify the percentage of attachment.

This experience has been taken into account only suckers occurred while the fruiting stems were largely eliminated. This work was performed at the beginning of the

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third year of vegetation. Percentage was very high grip, which is due to adventitious roots suckers posed. Observations were made every two days to capture any changes that occur in the evolution of suckers.

REZULTS AND DISCUSSION

Multiplying by young suckers is less widely practiced, because many manufacturers raspberry seedlings of the species are insufficiently aware of the advantages of young suckers have no technical basis necessary.

This method does not exhaust the parent plant and the percentage is very high grip. Young plants derived from suckers can be planted immediately in soil well prepared. Watering is essential for successful trapping. To avoid dehydration suckers is recommended to be removed immediately after the mud, followed by immediate planting. New suckers so prepared can be stored refrigerated until planting.

In April, when suckers reached between 5-15 cm length proceeded to put their ranking on two categories of length, were the mire and were planted immediately.

Later observations were made on the percentage of grip and influence on the share suckers grip length.

Table 1

Number of suckers formed on the parent plant

Soiul	Nr.of suckers / plant			
	1 year plant	2 years plant	3years plant	**Average**
Citria (Mt)	4	9	21	**11,3**
Heritage	6	11	16	**11**
Willamette	7	14	18	**13**
Norna	5	10	20	**11,6**
Polka	6	10	23	**13**

Observations on the number of suckers formed by each variety revealed that the number depends on the variety and age of the mother plant.

In the first year of the parent plant vegetation formed by variety from 4 to 7 suckers, first place was owned by Willamette variety, while in two growing number of suckers emerged was much higher. At the beginning of the third year of vegetation each mother plant formed a large number of suckers by the evolution of the root system bearing adventitious buds. This year vegetative variety with the highest number of suckers was kind Polka (23), closely followed by varieties citrate (Mt) and Norna (20).

Analyzing the results of the average of three years of study shows that all varieties form suckers annually more than 11 varieties with the highest number of suckers as Norn and Willamette with 13 suckers each year.

After posting suckers from the parent plant and planting was mud their immediate place of production as planting distances. Observations have continued to indicate the

percentage of attachment of new suckers. It is remarkable that catching was very high and there was no dried plant. This high percentage of attachment was explained by the fact that each has suckers and root portion which prevents dehydration plant. The mud around the roots and watering after planting provides appropriate and sufficient moisture to maintain turgor suckers and promote growth of new roots.

Finally multiply by suckers from the beginning of the vegetation is an easy way not entirely destroy the parent plant, it being able to recover by year-end.

- But be given to speed of running operation suckers and mandatory collection should be mud.

- In addition to planting each plant must be wet with the amount of water.

- Not be neglected aspect of labor, which is hard to find and be trained accordingly.

- If don't planting immediately the suckers must be stratified in cold rooms in the sandbox, but not for long to prevent their etiolation.

Comparing the two methods of multiplication is found by multiplying the split bush offers more rooted plants per hectare applied to multiply by suckers growing season beginning immediately after the appearance of suckers on the surface.

This pit only be explained by gradual formation of a larger number of strains during the growing season.

Table 2

Annual average data on the number of suckers produced per hectare

Soiul	Nr. of suckers obtained		
	Annual average suckers/plant	Total suckers /3 years production	Total suckers /ha Annual average
Citria (Mt)	11,3	282.507	94.169
Heritage	11	274.989	91.663
Willamette	13	325.987	108.329
Norna	11,6	289.986	96.662
Polka	13	325.987	108.329
Media	**11,9**	**299891**	**99830**

The fact that the multiplication of the suckers they are removed immediately after their appearance on the surface prevents the emergence of new suckers. Otherwise, the two propagation methods ought to produce about the same number of plants per hectare.

Research shows a clear difference between the two methods, reflected by the number of plants per hectare, situated difference method for the separation bush. For all varieties studied bush separation leads to a greater number of plants per hectare. The average per hectare bush by the separation obtained with 44 830 plants / ha more than multiplying by suckers, establishing the quantity required about 3 hectares of productive plantation. The biggest difference between the methods of multiplication has been studied varieties Norn (69,998 plants) and Polka (63,330 plants).

Table 3

Planting material difference between the two conventional methods of multiplication

Soiul	Split bush	Multiply by suckers	Difference Plants/ha
	Total plants /ha Annual average	Total suckers /ha Annual average	
Citria (Mt)	113.328	94.169	19.169
Heritage	110.828	91.663	19.165
Willamette	160.826	108.329	52497
Norna	166.660	96.662	69998
Polka	171.659	108.329	63330
Media	**144.660**	**99830**	**44830**

After studies showed that multiplying by separating bush is more productive and advantageous in terms of efficiency of materials and labor. It can be applied in autumn after leaf fall, before freezing soil in early spring before the vegetation.

Multiply by suckers cant be applied only in early spring after emergence from the soil surface and suckers must be performed in a short time.

As a general conclusion we can say that raspberry varieties studied performs very well in copying whatever method applied. It is important that methods be applied at the right time.

CONCLUSIONS

Multiply by suckers raspberry bush is effective and easy to apply. The secret of this method is to protect young plants of dehydration plant as soon as possible by ensuring the necessary moisture.

Catching the young suckers out at 100%.

Average annual number of plants obtained per hectare is very high, regardless of propagation method applied.

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THE INFLUENCE OF ANY ANTIOXIDANTS AND OXIDANTS ACROSS THE LACTOPEROXIDASE'S CAW MILK

Petre Savescu[1], Cristian Vasile[2]

Keywords: *Lactoperoxidase, antioxidants, oxidants, caw milk*

ABSTRACT

The lactoperoxidase system plays an important role in the immune system by killing bacteria in milk and mucosal (linings of mostly endodermal origin, covered in epithelium, which are involved in absorption and secretion) secretions hence augmentation of the lactoperoxidase system may have therapeutic applications. The influence of antioxidants and oxidants present or added in to caw milk on any oxidoreductase - like as lactoperoxidase - are significant and the study on these peroxidase are normal and necessary.

INTRODUCTION

The research project are descript by the big volume of biochemical and electrochemical lot of analysis, and aimed to provide the improvement of redox processes that produced during the processing the caw milk [Savescu 2006].

Lactoperoxidase (EC 1.11.1.7) (LP) is a member of the peroxidase family, a group of natural enzymes, widely distributed in nature and found in plants and animals, including man[Kussendrager&Hooijdonk 2000].

Lactoperoxidase is a peroxidase enzyme secreted from mammary, salivary, and other mucosal glands that functions as a natural antibacterial agent [Tenovuo 1985, Pruitt & Reiter 1985]. Lactoperoxidase is a member of the heme peroxidase family of enzymes. In humans, lactoperoxidase is encoded by the *LPO* gene. [Dull et al. 1990, Kiser et al.1996]

Lactoperoxidase catalyzes the oxidation of a number of inorganic and organic substrates by hydrogen peroxide [Tenovuo&Pruitt 1985]. These substrates include bromide and iodide and therefore lactoperoxidase can be categorized as a halo-peroxidase. Another important substrate is thiocyanate. The oxidized products produced through the action of this enzyme have potent bactericidal activities. Lactoperoxidase together with its inorganic ion substrates, hydrogen peroxide, and oxidized products is known as the **lactoperoxidase system**. [Fweja et al. 2008]

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Lactoperoxidase (LPO) catalyzes the hydrogen peroxide oxidation of iodide:

$$2I^- + H_2O_2 + 2H^+ = I_2 + 2H_2O$$

The bovine milk enzyme is identical to that formed in bovine lacrimal and salivary glands (Morrison *et al*.; and Morrison and Allen, 1963) [http://www.worthington-biochem.com 2012]. It is possible that LPO may be important in controlling bacterial flora (Bjorck *et al*.; Gothefors and Marklund; and Morrison and Allen). LPO is useful for labeling proteins with radioiodine (Gow and Wardlaw; Holohan *et al*.; Morrison and Bayse; Bayse *et al*.; Frantz and Turkington; and Marchalonis). For membrane studies the large LPO molecule limits labeling to the exposed surface [http://www.worthington-biochem.com 2012].

The lactoperoxidase system is effective in killing a range of aerobic and certain anaerobic microorganisms [Fweja et al. 2008].

Lactoperoxidase is an effective antimicrobial agent and is used as an antibacterial agent in reducing bacterial micro flora in milk and milk products [Reiter&Härnulv 2010]. Activation of the lactoperoxidase system by addition of hydrogen peroxide and thiocyanate extends the shelf life of refrigerated raw milk [De Wit&van Hooydonk 1996]. It is fairly heat resistant and is used as an indicator of over-pasteurization of milk [Marks et al. 2008].

The biological significance of lactoperoxidase is its involvement in the natural host defense system against invading micro-organisms. In bovine milk it is one of the indigenous antimicrobial agents. In bovine milk LP is, next to xanthine oxidase, the most abundant enzyme. Its concentration is around 30 mg/l, constituting about 0, 5 % of the whey proteins [Kussendrager&Hooijdonk 2000].

The LP enzyme catalyses the peroxidation of thiocyanate and some halides (I_2, Br_2 but not C_{l2}) to generate products which kill or inhibit the growth of many species of micro-organisms. The reaction mechanisms are very complex. A summary of the pathways of the enzymatic mechanism with H_2O_2 and SCN_2 is presented by de Wit & van Hooydonk [10]. In brief the following reactions occur. The first step in the enzymatic mechanism is the initiation reaction of the resting LP (Fe3+) to its ground state, using H2O2, according to: $Fe^{3+} + H_2O_2 = Fe^{2+} + HO'_2$ followed by the propagation reactions[De Wit&van Hooydonk 1996].

Lactoperoxidase (LP) catalyzes the hydrogen peroxide (H_2O_2) oxidation of several acceptor molecules [De Wit&van Hooydonk 1996]:

reduced acceptor + H_2O_2 → oxidized acceptor + H_2O

Specific examples include:

$SCN^- + H_2O_2$ (reaction on presence of LP) = $OSCN^- + H_2O$ or

$2SCN^- + H_2O_2 + 2H^+$ (reaction on presence of LP) = $(SCN)_2 + 2H_2O$

$(SCN)_2 + H_2O = HOSCN + H^+ + SCN^-$

HOSCN (pKa=5,3) = $H^+ + OSCN^-$ (reversible reaction)

For these reasons are determined the concentration of reduced and oxidized form of Lactoperoxidase and the concentration of free SH groups.

MATERIAL AND METHODS

For the study using a raw milk (3,5% fat), unpasteurized, which was witness variant (Mt).Like as antioxidants have used a lot of vitamins (variants V1-V4 and V7) and selenium (V5).

Were set to the following experimental study:
V1 - milk + vitamin A (retinol acetate);
V2 - milk + vitamin C;
V3 - milk + vitamin E;
V4 - milk + coenzyme Q10;
V5 - milk + Se;
V6 - milk + 3% hydrogen peroxide (3% solution);
V7 - milk + ascorbic acid (5%);
V8 - milk + vitamin A + hydrogen peroxide;
V9 - milk + vitamin C + hydrogen peroxide;
V10 - milk + vitamin E + hydrogen peroxide;
V11 - milk + CoQ10 + hydrogen peroxide;
V12 - milk + Se + hydrogen peroxide.
V13 - milk + ascorbic acid + hydrogen peroxide
V14 - milk + hydrogen peroxide + ascorbic acid;
V15 - milk + hydrogen peroxide + vitamin E;
V16 - milk + hydrogen peroxide + ascorbic acid + vitamin E.

For dilution (1:40) and cleaning vats, the samples were prepared using double distilled water. Initial samples were centrifuged prior to a Sigma centrifuge at 7800 rev / minute for 5 minutes. To establish absorption spectra and measure of pick of absorbance are using a spectrophotometer UV / VIS UNICAM2 type with 2 nm bandwidth. He scanned the UV (190-400nm) and visible range (400 - 700 nm) and at 325 nm value is automatically changed Deuterium lamp with a Tungsten lamp.

The used hydrogen peroxide concentration was 3%, ultrapure (with concentrations of Al, As, B, Ba, Co, Cd, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Na, Ni, Pb, Sn by max. 1 ppm, residual 10ppt sec max.). It was used at a concentration of 3 cm^3 / L.

The L (+) ascorbic acid was used as aqueous 5%, with a density of 1.65 g/cm^3, molecular weight 176.13 g / mol, odorless white. The best dose was found to be the 3.5% (after being checked in cow's milk doses of 0.1%, 1%, 2%, 3%, 3.5%, 5% and 10% ascorbic acid aqueous 5%).

The E Vitamin (DL - alpha-tocopherol acetate) and A (retinol) have been used as solutions of alpha-tocopherol acetate. The used concentrations were 0.5 mg vit. A/100 mL milk and 30 mg vit. E/ 100 mL milk. The Vitamin E was used to protect polyunsaturated fatty acids, vitamin A and carotenoids from milk and thiol groups from enzymes that act as well into synergism with ubiquinone (coenzyme Q10) [Savescu 2006].

The Coenzyme Q10, 98% solution in oil (lipid soluble compound of each essential cell metabolic pathways that act for energy production, strong antioxidant) was introduced at a concentration of 15 mg/100 ml milk [Savescu 2006].

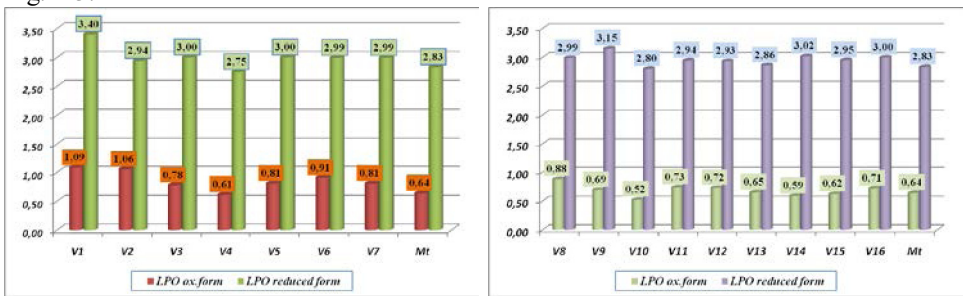
Selenium (used in milk for its antioxidant action, for to increase the resistance of body consumers in fight against the free radicals) [Savescu 2006], was used in a dose of 50 µg Se/100 mL milk. For V_5 and V_{12} variants, the used Selenium (52%) came from Wallmarck Co.

The hydrogen peroxide acid L (+) ascorbic vitamins E, D and ubiquinone were from Sigma Co.

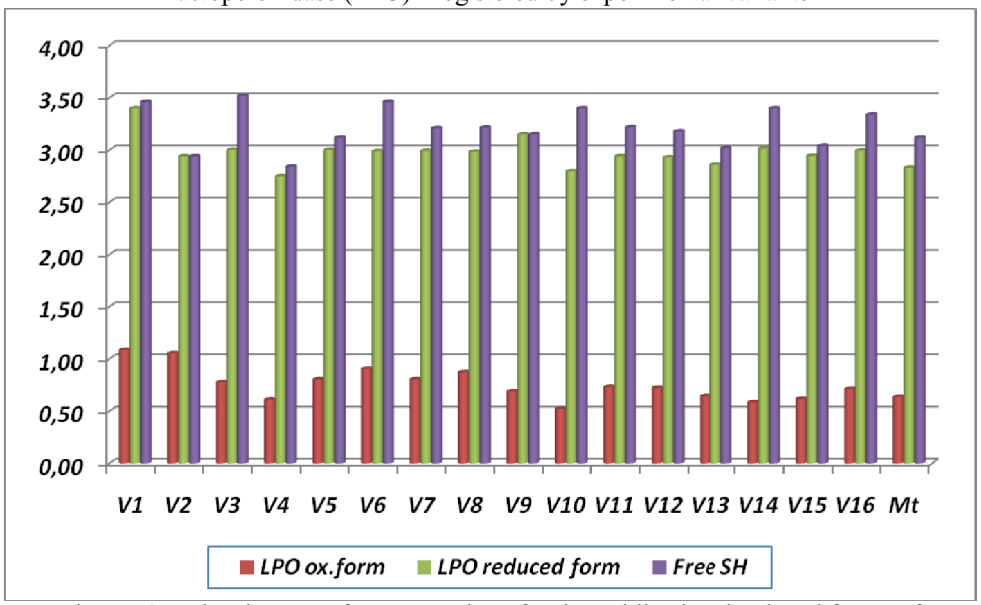
Following research using spectrophotometric methods, electrochemical method and Pure Analysis substances were determined wavelengths at which were recorded maximum absorption molecular spectra for reduced and oxidized forms of lactoperoxidase [Savescu 2006].

RESULTS AND DISCUSSIONS

After analyze of the experimental variants result the data showed into graphs of Fig. 1-3.



Figures 1and 2 – The changes of concentrations for the oxidized and reduced forms of lactoperoxidase (LPO) - registered by experimental variants



Figures 3 – The changes of concentrations for the oxidized and reduced forms of lactoperoxidase (LPO) and for the free SH groups- registered by experimental variants.

As shown in Figures 1, 2 and 3, the experimental variant V_{13} record the best ratio of oxidized and reduced forms of Lactoperoxidase, the ratio of oxidizing are registered the small difference for the value of recorded ratio in witness variant.

From the analysis of V_{13} and V_{14} variants we can see that the system is more protected from oxidation when added preventive antioxidant (the ascorbic acid). Thus, the V_{13} system it is safe like as witness variant; the hydrogen peroxide additions can produce just minor changes.

From the analysis of V_7 and V_9 variants can be seen the decrease of reduced forms concentration of lactoperoxidase in milk when adding an oxidant (hydrogen peroxide) for to extend shelf life.

Analyzing the action of antioxidants using in to V_8, V_9, V_{10}, V_{11}, and V_{12} variants was found that the best effect on the redox balance of lactoperoxidase was recorded when using the vitamin E (V_{10}).

CONCLUSIONS

From the graphs in figures can easily see that ascorbic acid (from V_{13}) protects best redox systems that involve vitamin A (especially in terms of reduced forms of vitamin A can thus protect these redox systems in milk and subsequent oxidation) .

From the analysis of V_{13} and V_{14} variants we can see that the system is more protected from oxidation when added preventive antioxidant (the ascorbic acid). Thus, the V_{13} system it is safe like as witness variant; the hydrogen peroxide additions can produce just minor changes.

From the analysis of V_7 and V_9 variants can be seen the decrease of reduced forms concentration of lactoperoxidase in milk when adding an oxidant (hydrogen peroxide) for to extend shelf life.

The analysis of reduced and oxidized forms concentrations of milk oxidoreductases (xantinoxidoreductase, superoxide dismutase, lactoperoxidase, Lactate dehydrogenase, P450 cytocromreductase), the flavoproteins, cytochromes a, b, c, c_1, of FMN coenzymes, NAD and riboflavin, the V_{13} is the best experimental variant. This experimental variant used as antioxidant ascorbic acid 5%, introduced before conservation agent (hydrogen peroxide).

In this version (V_{13}), the oxidized form of FMN and reduced form of NADH + H^+ concentrations were very close to those of similar witness. The differences in absorption maximum recorded values of each enzyme are similar to the untreated control, the experimental version is better, being close to the natural redox systems of milk;

The study of cow's milk redox processes can lead to improve the classic processing technology of milk, both by increase the storage time of milk and decrease the dietary risk by eliminating the toxicity of hydrogen peroxide.

Analyzing the action of antioxidants using in to V_8, V_9, V_{10}, V_{11}, and V_{12} variants was found that the best effect on the redox balance of lactoperoxidase was recorded when using the vitamin E (V_{10}).

The influence of antioxidant and oxidant place in order - across the redox balance of lactoperoxidase can be observed in analysis of V_{10} and V_{15} variants.

Even if V_{15} variant showed values very close to those of witness variant -in the rate of degradation of hydrogen peroxide, the V_{13} remains the best experimental variant; this variant can keep almost all redox systems at the closest level to the natural of witness control.

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THE INFLUENCE OF ANY ANTIOXIDANTS AND OXIDANTS ACROSS THE SUPEROXIDE DISMUTASE'S CAW MILK

Petre Savescu[1], Marius Vladu[1], Ana Maria Dodocioiu[1]

Keywords: *superoxide dismutase, antioxidants, oxidants, caw milk*

ABSTRACT

Superoxide Dismutase (SOD) is an enzyme that repairs cells and reduces the damage done to them by superoxide, the most common free radical in the body. SOD is found in both the dermis and the epidermis, and is the key to the production of healthy fibroblasts (skin-building cells).
The influence of antioxidants and oxidants present or added in to caw milk on any oxidoreductase - like as superoxide dismutase - are significant and the study on these oxido-reductase are normal and necessary. The work paper is part of a complex study able for improve the redox process that occur in the processing of food (soymilk, caw milk).

INTRODUCTION

The research project are descript by the big volume of biochemical and electro-chemical lot of analysis, and aimed to provide the improvement of redox processes that produced during the processing the caw milk [Savescu 2006].

Superoxide dismutase (EC1.15.1.1.), discovered in 1972 by McCord and Fridovich, is considered the most important enzyme characteristic for aerobic life, in terms of oxidative biochemical processes, and is present in all living cells. SOD shows the highest activity in the animal's blood. In case of pathological conditions (diabetes, cancer, inflammatory diseases, and cardiovascular diseases) enzyme activity is lower, either due to the appearance of inhibitors or due to a limited synthesis [Dejica 2000].

Milk contains low levels of SOD, 150 times lower than blood. The enzyme present in cow's milk has the same structure with SOD from bovine erythrocytes. The presence of this enzyme is important in maintaining the antioxidant stability of milk. Studies have shown that exogenous addition of SOD causes a reduction in lipid peroxidation processes, providing greater stability of milk [Fox&Kelly 2006].

Superoxide dismutase (SOD) is an antioxidant metalloprotein family involved in the defense system against reactive oxygen species (ROS). It converts superoxide radicals and hydrogen peroxide in water, which is then catalyzed in O_2 and H_2O by glutathione peroxidase and catalase.

Several common forms of SOD exist: they are proteins co-factored with copper and zinc, or manganese, iron, or nickel. Thus, there are three major families of superoxide

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dismutase, depending on the metal cofactor: Cu/Zn (which binds both copper and zinc), Fe and Mn types (which bind either iron or manganese), and the Ni type, which binds nickel.

Each type of SOD plays a different role in keeping cells healthy. Cu/Zn SOD protects the cells' cytoplasm and Mn SOD protects their mitochondria from free radical damage [Sentman et al. 2006].

Studies have shown that SOD acts as both an antioxidant and anti-inflammatory in the body, neutralizing the free radicals that can lead to wrinkles and precancerous cell changes. Researchers are currently studying the potential of superoxide dismutase as an anti-aging treatment, since it is now known that SOD levels drop while free radical levels increase as we age [Sentman et al. 2006].

This enzyme, called superoxide dismutase, contains 2 eq of copper per mole of enzyme. The copper may be reversibly removed, and it is required for activity. Superoxide dismutase has been shown to be identical with the previously described copper-containing erythrocuprein (human) and hemocuprein (bovine) [Kresge et al. 2006].

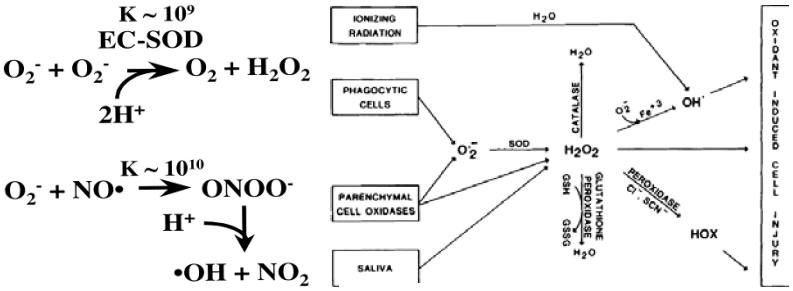
Superoxide dismutase (SOD) catalyzes the destruction of the O^{2-} free radical [http://www.worthington-biochem.com 2012]:

$$2O^{2-} + 2H^{+} = O_2 + H_2O_2$$

Milk is a polyphasic secretion of the mammary gland with significant antioxidant activity and high calcium content bound by protein micelles.

The removal of calcium caused disruption of casein micelles and significant decrease in SOD activity from 5 to 1.24-0.18 U/mg protein [Filipović 2003].

Assay of individual cow milk indicated no significant difference in superoxide dismutase concentration between night and morning milking. Also, superoxide dismutase concentration was affected by breed but not by stage of lactation or age. Superoxide dismutase decreased the minor pro-oxidant effect of xanthine oxidase. However, the concentration of superoxide dismutase in individual cow milk did not account for the large variability in oxidative resistance in raw milk that had not been exposed to light. The oxidative effect attributed for hydrogen peroxide in raw milk is significant [Marks et al. 2008].



(a) (b)

Fig. 1 Mechanism of action for SOD (a) and Sources and metabolism of reactive oxygen metabolites in gastrointestinal tract (b). [Granger et al. 1988].

Most important source is respiratory burst from phagocyte cells that secrete superoxide ($O_2^{-}\cdot$). $O_2^{-}\cdot$ is rapidly metabolized by superoxide dismutase (SOD) to H_2O_2. H_2O_2 may be metabolized to H_2O by 2 major oxidant defenses—catalase or glutathione peroxidase. Alternatively, H_2O_2 may be metabolized to hydroxyl ($OH\cdot$) in the presence of Fe^{3+} by Haber-Weiss reaction or it may be changed to hypothalamus acids (HOX) by myeloperoxidase. These various oxidants—$O_2^{-}\cdot$, H_2O_2, $OH\cdot$, and HOX—may induce

tissue injury but may also take part in tissue defense by killing microorganisms and initiating intestinal water and electrolyte secretion to wash them from intestine [Granger et al. 1988]

For these reasons are determined the concentration of reduced and oxidized form of Superoxide Dismutase (SOD) and the concentration of hydrogen peroxide.

MATERIAL AND METHODS

For the study using a raw milk (3, 5% fat), unpasteurized, which was witness variant (Mt). Like as antioxidants have used a lot of vitamins (variants V1-V4 and V7) and selenium (V5).

Were set to the following experimental study:

V1 - milk + vitamin A (retinol acetate);
V2 - milk + vitamin C;
V3 - milk + vitamin E;
V4 - milk + coenzyme Q10;
V5 - milk + Se;
V6 - milk + 3% hydrogen peroxide (3% solution);
V7 - milk + ascorbic acid (5%);
V8 - milk + vitamin A + hydrogen peroxide;
V9 - milk + vitamin C + hydrogen peroxide;
V10 - milk + vitamin E + hydrogen peroxide;
V11 - milk + CoQ10 + hydrogen peroxide;
V12 - milk + Se + hydrogen peroxide.
V13 - milk + ascorbic acid + hydrogen peroxide
V14 - milk + hydrogen peroxide + ascorbic acid;
V15 - milk + hydrogen peroxide + vitamin E;
V16 - milk + hydrogen peroxide + ascorbic acid + vitamin E.

For dilution (1:40) and cleaning vats, the samples were prepared using double distilled water. Initial samples were centrifuged prior to a Sigma centrifuge at 7800 rev / minute for 5 minutes. To establish absorption spectra and measure of pick of absorbance are using a spectrophotometer UV / VIS UNICAM2 type with 2 nm bandwidth. He scanned the UV (190-400nm) and visible range (400 - 700 nm) and at 325 nm value is automatically changed Deuterium lamp with a Tungsten lamp.

The used hydrogen peroxide concentration was 3%, ultrapure (with concentrations of Al, As, B, Ba, Co, Cd, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Na, Ni, Pb, Sn by max. 1 ppm, residual 10ppt sec max.). It was used at a concentration of 3 cm^3 / L.

The L (+) ascorbic acid was used as aqueous 5%, with a density of 1.65 g/cm^3, molecular weight 176.13 g / mol, odorless white. The best dose was found to be the 3.5% (after being checked in cow's milk doses of 0.1%, 1%, 2%, 3%, 3.5%, 5% and 10% ascorbic acid aqueous 5%).

The E Vitamin (DL - alpha-tocopherol acetate) and A (retinol) have been used as solutions of alpha-tocopherol acetate. The used concentrations were 0.5 mg vit. A/100 mL milk and 30 mg vit. E/100 mL milk. The Vitamin E was used to protect polyunsaturated fatty acids, vitamin A and carotenoids from milk and thiol groups from enzymes that act as well into synergism with ubiquinone (coenzyme Q10) [Savescu 2006].

The Coenzyme Q10, 98% solution in oil (lipid soluble compound of each essential cell metabolic pathways that act for energy production, strong antioxidant) was introduced at a concentration of 15 mg/100 ml milk [Savescu 2006].

Selenium (used in milk for its antioxidant action, for to increase the resistance of body consumers in fight against the free radicals) [Savescu 2006], was used in a dose of 50 µg Se/100 mL milk. For V_5 and V_{12} variants, the used Selenium (52%) came from Wallmarck Co.

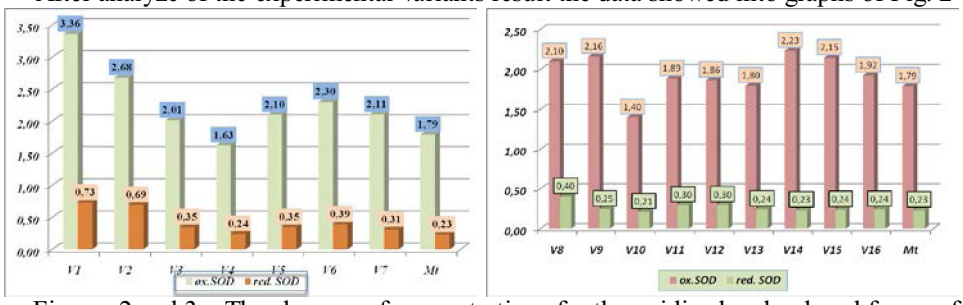
The hydrogen peroxide acid L (+) ascorbic vitamins E, D and ubiquinone were from Sigma Co.

Following research using spectrophotometric methods, electrochemical method and Pure Analysis substances were determined wavelengths at which were recorded maximum absorption molecular spectra for reduced and oxidized forms of Superoxide Dismutase [10]. Determination of blood SOD activity was performed using the kit RANSOD, from Randox Laboratories. For both enzymes the hemoglobin content in blood was measured and the activity expressed as Units/g Hemoglobin (U/g Hb).

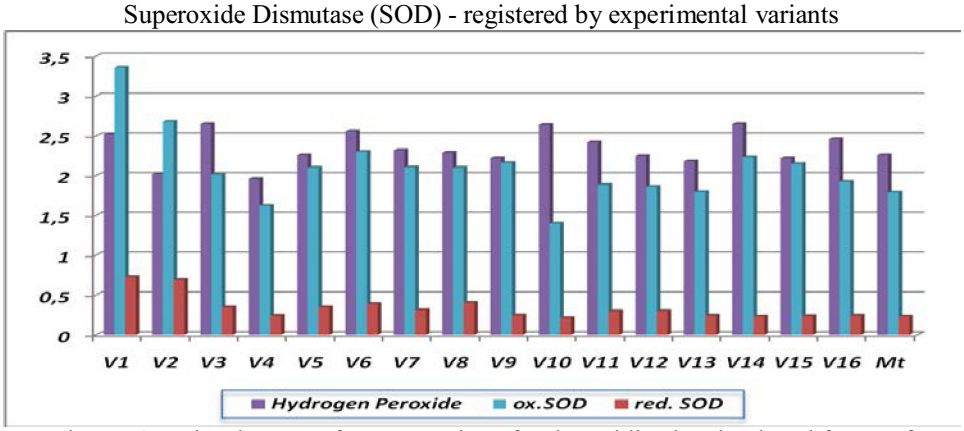
Determination of SOD activity in milk was realized by photometric methods. The first step in this analysis was the dissolution of casein micelles, a process known as clearing milk. This was achieved using a solution containing urea and a reducing agent - dithiotrietol. Determination of SOD activity was performed using RANSOD kit, from Randox Laboratories. For SOD enzyme activity was calculated and expressed in U/ml milk. Using the calibrate curve and the only added method it is establish the wavelength specifically for maximum of molecular absorption spectra for SOD and hydrogen peroxide.

RESULTS AND DISCUSSIONS

After analyze of the experimental variants result the data showed into graphs of Fig. 2-4.



Figures 2 and 3 – The changes of concentrations for the oxidized and reduced forms of Superoxide Dismutase (SOD) - registered by experimental variants



Figures 3 – The changes of concentrations for the oxidized and reduced forms of

Superoxide Dismutase (SOD) and for H_2O_2- registered by experimental variants

As shown in Figures 2, 3 and 4, the experimental variant V_{13} record the best ratio of oxidized and reduced forms of Superoxide Dismutase (SOD), the ratio of oxidizing are registered the small difference for the value of recorded ratio in witness variant.

From the analysis of V_{13} and V_{14} variants we can see that the system is more protected from oxidation when added preventive antioxidant (the ascorbic acid). Thus, the V_{13} system it is safe like as witness variant; the hydrogen peroxide additions can produce just minor changes.

Follow the Analysis of variant enriched with Coenzyme Q_{10} (V_4) observed that SOD activity is inhibited, the concentration of oxidized and reduced forms is minimized.

The lowest activity of SOD was recorded if we used vitamin E (V_{10}) this version was recorded the lowest concentration of SOD.

From the analysis of V_7 and V_{13} variants can be seen the decrease of reduced forms concentration of Superoxide Dismutase (SOD) in milk when adding an oxidant (hydrogen peroxide) for to extend shelf life.

Analyzing the action of antioxidants using in to V_8, V_9, V_{10}, V_{11}, and V_{12} variants was found that the best effect on the redox balance of SOD was recorded when using the vitamin E (V_{10}).

CONCLUSIONS

It is concluded that the high antioxidant content of the initial raw milk, as well as the technology of its processing are of great importance for the preservation of SOD activity in the final foodstuff product.

From the graphs in figures can easily see that ascorbic acid (from V_{13}) protects best redox systems that involve vitamin A (especially in terms of reduced forms of vitamin A can thus protect these redox systems in milk and subsequent oxidation) .

The lowest activity of SOD was recorded if we used vitamin E (V_{10}) this version was recorded the lowest concentration of SOD.

From the analysis of V_{13} and V_{14} variants we can see that the system is more protected from oxidation when added preventive antioxidant (the ascorbic acid). Thus, the V_{13} system it is safe like as witness variant; the hydrogen peroxide additions can produce just minor changes.

The analysis of reduced and oxidized forms concentrations of milk oxidoreductases (xantinoxidoreductase, superoxide dismutase, lactoperoxidase, Lactate dehydrogenase, P450 cytocromreductase), the flavoproteins, cytochromes a, b, c, c_1, of FMN coenzymes, NAD and riboflavin, the V_{13} is the best experimental variant. This experimental variant used as antioxidant ascorbic acid 5%, introduced before conservation agent (hydrogen peroxide).

In this version (V_{13}), the oxidized form of FMN and reduced form of $NADH + H^+$ concentrations were very close to those of similar witness. The differences in absorption maximum recorded values of each enzyme are similar to the untreated control, the experimental version is better, being close to the natural redox systems of milk;

The study of cow's milk redox processes can lead to improve the classic processing technology of milk, both by increase the storage time of milk and decrease the dietary risk by eliminating the toxicity of hydrogen peroxide.

The biggest release of hydrogen peroxide was achieved if it used Vitamin A (V_1) and Vitamin C (V_2).

The lowest production of hydrogen peroxide was recorded if we used vitamin E (V_{10}, V_{14}).

The variants were used Coenzyme Q_{10} (V_4), ascorbic acid (V_{13}), ascorbic acid and vitamin E (V_{16}) recorded the same production of hydrogen peroxide - very close to that of the variance Witness (Mt).

The influence of antioxidant and oxidant place in order - across the redox balance of SOD can be observed in analysis of V_{10} and V_{15} variants.

Even if V_{15} variant showed values very close to those of witness variant -in the rate of degradation of hydrogen peroxide, the V_{13} remains the best experimental variant; this variant can keep almost all redox systems at the closest level to the natural of witness control.

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RESEARCHES REGARDING THE CHEMICAL COMPOSITION OF THE YOUNG WINES OBTAINED FROM CHARDONNAY CLONES IN SAMBURESTI VINEYARD

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Key words: *wine, clones, Chardonnay, composition*

ABSTRACT

The Chardonnay type is the most well-known for the white wines worldwide being spread all over the continents. In our country, it used to be unknown and therefore almost uncultivated until 2000, but in the last decade, it has gained great reputation and extension. Therefore, it has got to be cultivated at Samburesti, as well, a vineyard designed, through excellence, for the production of red wines. The introduction of the Chardonnay type in the range of white wines represent an absolute innovation for the Samburesti vineyard because this type has not existed so far in the range of wines belonging to it.

INTRODUCTION

Primary aroma compounds are important mainly for young wines. These compounds originate from grape and are present in wine either in free or bound form. The free volatiles are responsible for the varietal character of the wines. The bound aroma compounds are the non-volatile and organoloptically inactive precursors of aroma compounds. Primary aroma compounds comprise also prefermentative volatile compounds, which are in the period between the grapes harvest and the beginning of the alcoholic fermentation (Petka J. and Farkas P., 2001a).Pre-fermentation practices determine the aromatic characteristics of white wine (Ribereau-Gayon P. et al.,2006). The aroma precursors are located mostly in the grape skins (Cabaroglu et al., 1997, Peinado et al., 2004, Ramey et al., 1986, Sanchez Palomo et al., 2006, Selli et al., 2006).

Pre-fermentative skin contact of crushed grapes or maceration are the most important enological practice used for increasing the aroma of wines (Cabaroglu et al., 2002, Romero-Cascales et al., 2008, Versini et al., 1981, Marais, 1987, Tamborra, 1992, Selli et al., 2003, Rodriguez Bencomo et al., 2008).

MATERIAL AND METHOD

The present paper has been elaborated having as a root the physical, chemical and organoleptical studies of the wines obtained from six clones of Chardonnay, harvested

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between 2010 and 2011 from Samburesti vineyard, where this wine has recently been introduced in the range. The wines were elaborated in conditions of micro wine-making in the Oenology Laboratory from the Horticulture Faculty of Craiova, in the same location having also been done all the studies regarding the main composition and organoleptical parameters. The six Chardonnay clones that have been studied are: 95/K5BB; 121/K5BB; 548/K5BB; 95/SO_4; 76/SO_4 and 548/SO_4.

The main compositional parameters that were studied were: the alcohol strength; the total acidity; the volatile acidity; the residual sugar; the glycerol; the free SO2 and the total SO_2. The methods used were the official ones, which are currently used in the Oenology Lab from our country.

The purpose of the researches was to evaluate up to what extent the newly introduced type in the Samburesti vineyard allows the acquiring of some quality wines and to evaluate the potential of each of the six clones with the purpose of optimizing the white wine making technology, taking into account the fact that we are talking about a new type that had not been cultivated so far in the vineyard and its capacity to adapt itself to the specific vineyard climate and soil conditions is yet unknown.

RESULTS AND DISCUSSIONS

In order to carry out an accurate study of the oenologycal potential of each of the six Chardonnay clones, the obtained results will be studied in accordance to the types of accomplished studies. The results regarding the chemical composition of wines are displayed in tables 1 and 2.

Table 1

The chemical composition of the Chardonnay wines, 2010, Samburesti

The clone	Alcohol % vol.	Total acidity g/l H_2SO_4	Volatile acidity g/l CH_3COOH	Residual sugar g/l	Glycerol g/l	SO2, mg/l	
						free	total
95/K5BB	11,8	3,35	0,33	5,1	7,6	12	116
121/K5BB	11,6	3,44	0,38	3,3	7,4	20	124
548/K5BB	12,1	3,28	0,30	3,1	8,1	18	120
95/SO_4	12,0	3,62	0,42	3,0	8,6	24	138
76/SO_4	12,3	3,82	0,45	2,8	8,8	16	118
548/SO_4	11,6	3,21	0,35	3,2	7,7	14	102

From table 1, we can observe that the alcoholic strength of the wines belonging to the 2010 harvesting was between 11,6 and 12,3% alcohol volume. The greatest content in alcohol was displayed by the wine obtained from clone 76/SO_4 with 12, 3% volume followed by the one obtained from clone 548/K5BB with 12,10% volume and the one coming from clone 95/SO_4 with alcohol contents beneath 12% volume, respectively 11,8% volume, clone 95/K5BB and 11,6% volume the clones 121/K5BB and 548/SO_4. So, all the six Chardonnay clones gave in 2010 11,5% volume, but none of them reached 12,5% volume. This is due to the rather early harvesting(September the 10^{th} 2010), but, inevitably, there is also a connection to the very immature age of the plantation, the grapes coming from a plantation that had not yet given fruits, which allowed the clones to expose all of their qualitative potential.

In 2011, all the six Chardonnay wines display contents between 14,1 and 15% alcohol volume. These are quite high levels, which are not to be frequently found in wines, so they are much more beyond the multi yearly media of wines from Samburesti vineyard. It is also important to underline the fact that the wines are coming from grapes harvested in

the middle of the harvesting campaign, in other words these are not wines obtained from the latest grapes harvested from a plantation. Another element that has to be taken into account in the discussions about the alcoholic strength of wines and the other parameters of the composition is that 2011 was for the whole south region of the country and especially for Oltenia region a wine-producing year characterized by great sugar accumulation in grapes, with a lot beyond the multi yearly medium values. It is also significant to mention the fact that the grapes from which there had been obtained the wines are coming from an immature plantation which is in the first year of production.

The greatest content in alcohol (15, 0% volume) was determined by the two wines obtained from clone 548 and by the one obtained from clone 95/K5BB. The other wine coming from clone 95 (on the stock SO_4) displayed the following content in alcohol (14,5% volume) and the clones 76 and 121 gave the wines with the alcoholic strength of 14,1% volume.

Table 2

The chemical composition of the Chardonnay wines, 2011, Samburesti

The wine	Alcohol % vol.	Glycerol g/l	Residual sugar g/l	Total acidity g/l H_2SO_4	Volatile acidity g/l acetic acid	SO_2 free mg/l	SO_2 total mg/l
76/SO_4	14,1	12,0	3,4	4,3	0,28	40	180
95/SO_4	14,5	12,4	3,0	3,9	0,32	32	172
95/K5BB	15,0	12,8	2,7	3,8	0,24	30	164
121/K5BB	14,1	12,1	3,6	4,2	0,34	34	168
548/K5BB	15,0	12,9	2,8	4,1	0,30	30	162
548/SO_4	15,0	12,8	2,6	4,3	0,32	33	160

The data regarding the total acidity of the wines from the 2010 harvest, at six months from obtaining, show important decreases, from 0,75g/l until 1,20g/l in comparison to the total acidity of the unfermented wines from which the respective wines had come. Under these circumstances, at six months of age, all the six Chardonnay wines resulted in being acid, the lack of acidity affecting them with regard to the organoleptic equilibrium. The lack of acidity has been obvious from the harvesting, taking into account the fact that the total acidity of the grapes was between 4,0 and 4,8g/l H_2SO_4 against the background of some sugar contents of 200-210 g/l.

The decrease of the wines total acidity, with medium values of 1g/H_2SO_4 is explained through the significant precipitation of the tartaric acid, especially under the form of acid salt- potassium bitartrate. The phenomenon took place as a result of the conjugated action of all the factors that favor it: alcohol, the temperature decrease and the presence of potassium cations. Regarding this last aspect, the important decrease of the wine total acidity is obviously connected to the pre fermentative pellicular maceration practiced at the early wine-making, a phenomenon that leads, among others, to the extraction of greater proportions of K cations from the grapes peel and implicitly to a more consistent decrease of the fixed wine acidity.

For the wines produced in 2011, the study of the total acidity values indicate the fact that the wines contain between 3,8 and 4,3 g/l H_2SO_4, thus existing rather small differences between them. Obviously, these values are slightly scarce considering the fact that they should start from 4,5g/l. Still, the slight shortage of acidity is acceptable, taking into account the fact that we are making reference to grapes which are extremely rich in sugar, thus very ripe. The greatest content in total acidity, 4,3g/l is to be met at two wines, one of these having the highest alcohol content (548/SO_4) and the other one the lowest

amount of alcohol (76/SO4). A total acidity of 4g/l has also been found at the wines obtained from clones 121/K5BB (4,2g/l) and 548/K5BB (4,1g/l). The lowest contents in total acidity are found at the wines 95/SO_4 (3,9g/l) and 95/K5BB (3,8g/l). Thus, of the two clones that have been grafted on both stocks, the clone 95 has lower levels of total acidity while clone 548 has a greater total acidity with 0,3g/l. Likewise, at both clones, the type grafted on stock SO4 has the acidity slightly greater than the one grafted on stock K5BB. In what concerns the volatile acidity of wines, the values that they display at six months are normal, not emphasizing any type of immediate danger for the biological stability of wines. Values of the volatile acidity comprised between 0,30 and 0,45g/l CH_3COOH are more inferior to the legal limit of 1,08g/l CH3COOH and cannot be perceived when tasting in order to influence the negative smell-taste balance of wines. The wines from 2011 have also displayed very low values of volatile acidity which do not endanger their biological stability.

The residual sugar content of the 2010 wines shows only one wine, the one obtained from clone 95/K5BB with more than 4 g/l, so a semi-dry wine, while at the other five types of wines display around 3g/l residual sugar, so they are dry wines, resulted from a complete alcoholic fermentation. The lowest content in residual sugar , of 2,8 g/l was recorded at the wine obtained from clone 76/SO4 the same that also displayed the highest content in alcohol (12,3 %volume), in other words the clone that offered the highest degree of the immature wine process of fermentation.

In 2011, all the wines had residual sugar beneath the value of 4g/l which means that the alcoholic fermentation took place in exceptional conditions, despite the fact that the grapes had been extremely rich in sugar. The use of the selected yeasts for the accomplishment of the alcoholic fermentation proved to be very useful from this point of view, too, because they lead the fermentation to dry and be carried on without fermentative incidents.

The following composition parameter that was studied was the wine content in glycerol. The glycerol is the second chemical constituent of the wine, from the quantity perspective, and at the same time, the main secondary product of alcoholic fermentation. The six wines coming from the 2010 Chardonnay clones display relatively modest glycerol contents, comprised between 7,4 and 8,8 g/l but in a certain balance with the alcohol contents, the most convenient connection between the contents in glycerol and alcohol being of 1/10.

Even though none of the six wines excel in glycerol content, it is absolutely obvious to take into account the experimental conditions more precisely the fact that the grapes come from a very young plantation.

On the other hand, at the wines coming from the 2011 harvesting, the glycerol contents of the six wines display high values which are met quite rarely in wines, but which are in a perfect accordance with the high contents of alcohol, taking into account the fact that while the alcohol is the main product of alcoholic fermentation, the glycerol is the main secondary product of fermentation. Under these circumstances, the six Chardonnay wines display glycerol contents which are comprised between 12,0g/l and 12,9g/l. the highest are met at the same three clones as in the ethylic alcohol: : 95/K5BB, 548/SO_4 and 548//K5BB the last one being the only one with 12,9g/l. The lowest level was displayed by clone 76/SO4 with 12,0g/l and clone 121/K5BB with 12,1g/l, these ones being also the ones with the lowest alcohol strength. The clone 95/SO_4 which displayed an alcoholic content which was of a medium size compared to the other clones displayed even in glycerol a medium content (12,4g/l).

The last studied composition parameter was the wine content in SO2, both free and total. Regarding this parameter, it has to be mentioned the fact that the wines took

advantage of the same sulphitation conditions until the first decanting but later, in accordance to the results of the cyclical wine tasting there has been decided the administration of some SO_2 doses for certain wines which lead to the fact that the results of the SO_2 administration to six months from the wine making to indicate certain differences between the wines regarding the total content of SO_2.

Under these circumstances, the six 2010 wines display total contents in SO_2 comprised between 102mg/l and 138mg/l, while the contents in free SO_2 are comprised between 12 mg/l and 24mg/l. In these conditions, the proportions of SO_2 combined from the total one and the free one are comprised between 82 and 90%. Considering the safety dose of SO_2 as being of 20mg/l SO_2, it is observed that four of the six wines display doses of free SO_2 beneath the value of 20mg/l, the lowest one being found at the wine from clone 95/K5BB (12mg/l), which has also one of the lowest doses of total SO2(116mg/l). Actually, of all of the six wines, this is the one with the lowest proportion of free SO_2 (10,3%). The highest dose of free SO_2 (24mg/l)is met at the wine coming from clone 95/SO4 which also has the highest content of total SO_2 (138mg/l) and at the same time, the highest percent of free SO_2 from the total one (17,3%).

Taking into account the fact that almost all the wines are dry, the legal maximum limit of the SO2 content for this type of wines being of 210mg/l, it is obvious the fact that all of them bear quite easily certain corrections of SO_2 doses, without the risk of reaching allowable maximum contents. At the same time, it is obvious the fact that under the conditions of an industrial wine making where all wine-making conditions are substantially superior to those from the micro wine-making, the assurance regime of the antiseptic and antioxidant protection of wines could be significantly improved.

For the 2011 wines, the wine contents in free SO_2 and total SO_2 situate between 30 and 40 mg/l, respectively 160-180m/l. these are values which situate in the legal limits, in other words, they do not exceed 210mg/l which is the limit imposed by the Romanian wine-making legislation for the dry white wines, just like in the case of the six Chardonnay wines which had been the subject of this study.

For the free SO_2, none of these six wines reaches the limit of 50mg/l imposed by the law or lowers beneath 20 mg/l, being thus stable and offering the guarantee that there is no risk of diseases or any other microbiological accidents on medium and short terms. The lowest levels of total SO_2 (162, respectively 160mg/l) are recorded to the two wines from clone 548 and the highest value of total SO_2 content recorded at clone 76/SO_4 (180mg/l). This clone also displays the highest content of free SO_2 (40mg/l) while the lowest content of free SO_2 (30mg/l) was observed at clones 95/K5BB and 548/K5BB.

CONCLUSIONS

The researches on the wines produced in 2010 and 2011 from the six Chardonnay clones lead to the following conclusions:

- The results obtained in the specific conditions of 2010 indicate the fact that even though there is an important discrepancy between the clones in what concerns the phases of the vegetative cycle, in the first part of this one, after the process of maturity and throughout the maturation, these discrepancies diminish in such a manner that the differences between the clones regarding the sugar and acidity contents are much lower at maturity than at the maturity process
- Of the six clones from the culture, two of them have remarked in the conditions of 2010: 548/K5BB from which it had been obtained the best wine from the sensorial aspect and 95/SO4 that remarked through compositional and organoleptiocal aspect.

- The description of quality of the 2011 Samburesti Chardonnay wines met a difficulty due to the fact that the plantation that gave the grapes from which the wines had come was very immature, being in the first year of production and a difficulty connected to the fact that the wine-making of 2011 was a completely different year from the previous one as it allowed the accumulation of some high proportions of sugar in grapes which is not to be usually found in the other years.
- The 2011 wines displayed contents in alcohol of 14-15% volume, unusually high and at the same time unjustified in these conditions and the glycerol contents of 4 g/l being also very high.(above the limit of 12g/l). under these circumstances, it is obvious that in the future, it is required as a first top priority that if the grapes accumulate a lot of sugar once again, to be harvested earlier in order to have less alcohol and more acidity in wine.
- The sensorial appreciation of the wines was met with difficulties by the application of a wine-making scheme and the unitary conditioning of the wines and especially by the fermentation of all the varieties with the same type of selected yeasts. Thus, the differences between the wines have been rather small thus it is difficult to come up with accurate conclusions regarding the differences between the clones. Still, the sensorial study emphasized the fact that at the two month tasting, all the wines have been better appreciated than at the six month tasting, the main cause being represented by the intensity and quality of the secondary flavor, which had been better at two month than at six.

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THE EFFECT OF PLANT GROWTH REGULATORS ON *IN VITRO* MICROPROPAGATION OF TWO INTERGENERIC HYBRIDS *FRAGARIA X POTENTILLA*

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Key words: *intergeneric hybrids, micropropagation, plant growth regulators.*

ABSTRACT

The feasibility of in vitro propagation of 'Pink Panda' and 'Serenata' genotypes (Fragaria x Potentilla) was tested by using six MS-based proliferation media supplemented with different combinations of BAP at 0.5, 1.0 or 2.0 mg l^{-1}, Kin at 0.5 mg l^{-1}, IAA at 0.5 or 1.0 mg l^{-1}, IBA at 0.1 or 0.2 mg l^{-1}, and GA_3 at 0.1 or 2.0 mg l^{-1}. Rate of micropropagation was the highest for 'Serenata' genotype, giving a maximum number of 23.65 shoots per explant on MS medium augmented with 0.5 mg l^{-1} BAP, 0.5 mg l^{-1} IAA and 0.1 mg l^{-1} GA_3. Multiple shoots were also induced, for both intergeneric hybrids, when combinations of 1.0 mg l^{-1} BAP and 0.1 mg l^{-1} GA_3, with either 0.2 mg/l IBA or 1.0 mg/l IAA, have been added in the basic media. Rooting was best induced in shoots grown on half-strength MS medium with IBA at 0.25 mg l^{-1} and GA_3 at 0.1 mg l^{-1} concentration. Plantlets were successfully acclimatized and established in soil.

INTRODUCTION

Fragaria and *Potentilla* are supposed to be closely related, but independent genera (Gerstberger, 1994; Leslie, 1995; Eriksson *et al*. 1998) and investigation of their crossability had a valuable contribution to intergeneric hybrids induction, with the first objective of improving the cultivated strawberry crop *Fragaria x ananassa*, by the introgression of genes from *Potentilla* (Jones, 1995). The success gained by the *Fragaria x Potentilla* hybrid forms resulted in a rapid increasing of the breeders interest for obtaining intergeneric hybrids presenting both the important traits of *Fragaria* species (fruit with optimal flovour) and some traits of *Potentilla*, mainly those associated with ornamental value, such as prolonged blossoming season, compact plant habit and attractive flowers (Ellis, 1962; Sukhareva, 1976; Niemirowicz-Szczytt, 1984; Macfarlane Smith and Jones, 1985, 2004; Abdullah and Hennerty, 1993; Sayegh and Hennerty, 1993; Silva and Jones, 1996; Bentvelsen and Bouw 2006). Therefore, a number of ornamental strawberry variety, knowing as 'Pink Panda' ('Frel'), 'Lipstick', 'Red Ruby', 'Serenata', 'Rosalyne' and 'Rosseberry' (Khanizadeh *et al*., 2002), have been released in last two decades.

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Since ornamental strawberries are highly heterozigous and segregation in their progenies is unforeseeable, the *in vitro* micropropagation of these elite plants provides an advantage for their multiplication, without sexual recombination and in a very short time. Thus, the objective of this study was to elaborate a reliable protocol for *in vitro* micropropagation of the 'Pink Panda' and 'Serenata' genotypes of ornamental strawberry. Analogous studies have not been reported previously.

MATERIAL AND METHODS

Two genotypes of ornamental strawberry (*Fragaria x Potentilla*), 'Pink Panda' (with pink flowers) and 'Serenata' (with red flowers), respectively, from the *Fragaria* Germplasm Collection of the Research Institute for Fruit Growing, Pitesti - Mărăcineni, Romania, were selected for the establishment of *in vitro* cultures.

For the initiation of shoot cultures, meristems with 2-3 leaf primordia, of 0.1- 0.3 mm in size, excised from runners formed by field grown stock plants were immersed successively in 94°etilic alcohol for 7 minutes and 6% calcium hypoclorite for 14 minutes, with the aim of disinfection. Runner tips were subsequently rinsed with sterile distilled water. Meristems with 2-3 leaf primordia, of 0.1-0.3 mm in size were cultured on LF basic medium (Lee and Fossard, 1977), supplemented with MS vitamins, dextrose at 40 g l^{-1} and agar at 7.0 g l^{-1} concentration. The pH was adjusted to 5.8.

Six treatments with different combinations of BAP at 0.5, 1.0 or 2 mg l^{-1}, Kin at 0.5 mg l^{-1}, IAA at 0.5 or 1.0 mg l^{-1}, IBA at 0.1 or 0.2 mg l^{-1}, and GA_3 at 0.1 or 2.0 mg l^{-1}, added to MS medium (Murashige and Skoog, 1962), were used in order to find an adequate medium for obtaining a high rate of micropropagation, while maintaining a good vigor of micropropagated shoots.

After four subcultures, shoots which regenerated from *Fragaria x Potentilla* explants were separated from the multiplication media, when they were approximately 2-3 cm long, and placed on a medium suitable for root growth. Root growth was stimulated by supplementing the solidified MS basal medium, containing half-strength macroelements and half LF microelements, with the auxins IBA at 0.25 or 0.5 mg l^{-1} and IAA at 0.5 mg l^{-1} concentration. In all treatments 0.1 mg l^{-1} of GA_3 was also added to the basal medium. As carbon source in all culture media was used dextrose, at 40 g l^{-1} concentration. The cultures have been incubated in a growth chamber at the temperature of 22-24°C, with a photoperiod of 16 hours light / 8 hours darkness, and a light intensity of about 40 µmol $m^{-2}s^{-1}$. *In vitro* rooting was followed by acclimatization to *ex vitro* conditions, plantlets being transfered in perlite in greenhouse conditions.

To avoid major statistical errors, at least 5 conical flasks (each with 30 ml of culture medium and closed with cotton-wool bungs and tinfoil) with 6 shoots per flask were used as repetitions in each of the experimental treatment investigated. In order to establish the efficiency of each treatment, the micropropagation rate, rooting rate, average root number and root length, were determined. The observations were carried out at every 4 weeks, respectively at the moment of subculturing the micropropagated shoots. Statistical analysis of the data obtained with 'Pink Panda' and 'Serenata' genotypes, were performed using Statistical Package for the Social Science (SPSS) statistical software (ver. 16.0), at $p < 0.05$.

RESULTS AND DISCUSSION

Irrespective of genotype, all apical meristems produced shoots on MS medium, irrespective of the combinations and concentrations of plant growth regulators.

However, analysis of variance test revealed that 'Serenata' and 'Pink Panda' genotypes, and plant growth regulators interacted significantly, with respect to the mean number of shoots formed per explant. After the first subculture, the highest rates of shoot proliferation calculated for 'Serenata' (19.3 shoots per explant) and 'Pink Panda' (10.9 shoots per primary explant) genotypes were achieved on medium supplemented with 1.0 mg l^{-1} BAP, 0.2 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3. As shown by the Duncan's multiple range test, in 'Serenata' genotype, a very closed rate of multiplication, of 19.2 shoots per explant, was induced also by the MS medium augmented with 2.0 mg l^{-1} BAP and 1.0 mg l^{-1} IAA, while in 'Pink Panda', an average number of 10.5 shoots formed per primery explant was calculated for the treatment with 1.0 mg l^{-1} BAP, 1.0 mg l^{-1} IAA and 0.1 mg l^{-1} GA_3 (Fig. 1).

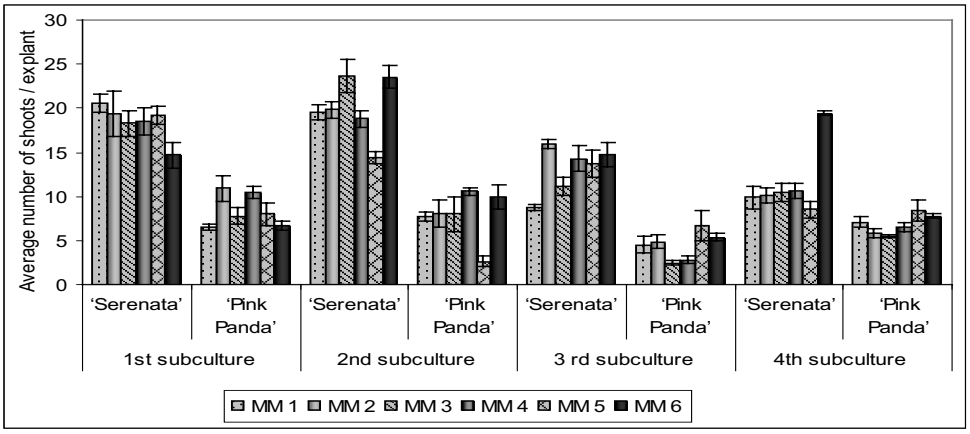

Figure 1. The micropropagation rate of the 'Serenata' and 'Pink Panda' genotype during the four subcultures on MS basic medium, supplemented with different combinations and concentration of plant growth regulators: **MM 1** – 0.5 mg l^{-1} BAP, 0.1 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3; **MM 2** – 1.0 mg l^{-1} BAP, 0.2 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3; **MM 3** – 0.5 mg l^{-1} BAP, 0.5 mg l^{-1} IAA and 0.1 mg l^{-1} GA_3; **MM 4** – 1.0 mg l^{-1} BAP, 1.0 mg l^{-1} IAA and 0.1 mg l^{-1} GA_3; **MM 5** – 2.0 mg l^{-1} BAP, 1.0 mg l^{-1} IAA; **MM 6** – 1.0 mg l^{-1} BAP, 2.0 mg l^{-1} GA_3 and 0.5 mg l^{-1} Kin. Error bars are ± SE.

An interesting response was observed after the subsequent subcultures, as the micropropagation rate decreased in 'Pink Panda' and 'Serenata' genotypes, on all the variants of media consisting of combinations of growth regulators added to the MS medium, which in previous subcultures promoted good rates of shoot multiplication, and increased for the explants subcultured on variants which previously resulted in lower rates of shoot multiplication. The overall results indicates that the micropropagation rate of 'Pink Panda' genotype increase, when 2.0 mg l^{-1} of BAP was used. It is relevant the fact that, excepting the treatment with 1.0 mg l^{-1} BAP, 2.0 mg l^{-1} GA_3 and 0.5 mg l^{-1} Kin (associated with an average number of 19.45 shoots per explant), no other combinations of growth regulators resulted in significantly different rate of shoot micropropagation, at the end of the fourth subculture (Fig. 1). In this respect, it worth mentioning that Kin have been used as a high

efficient cytokine, mainly for callus induction and plant regeneration in strawberry (Sayegh, 1989; Kaushal *et al*., 2006), and for direct somatic embryogenesis in *Fragaria x ananassa* cv. 'Chandler' (Husaini *et al*., 2008). The cumulative effect of the Kin and BAP, on successfully *in vitro* shoot multiplication, have also been reported in other species, such as *Citrus aurantifolia* (Al-Bahrany, 2002), *Capsicum frutescens* (Sanatombi and Sharma, 2007) or *Vitex agnus-castus* (Balaraju *et al*., 2008).

Although, o positive correlation between shoot proliferation and low levels of cytokines and auxins was observed in the first subcultures, good rate of micropropagation was induce in the lasts subcultures, when combinations of 1.0 mg l^{-1} BAP with either 0.2 mg l^{-1} IBA or 1.0 mg l^{-1} IAA have been added in the MS basic media. Similarly, other authors suggested 1.0 mg l^{-1} BAP + 1.0 mg l^{-1} IAA + 0.05 mg l^{-1} GA_3; 0.5 mg l^{-1} BAP + 0.1 mg l^{-1} IBA + 0.1 mg l^{-1} GA_3 (Boxus, 1999; Litwińczuk, 2004); 0.5 mg l^{-1} BAP + 0.1 mg l^{-1} IBA (Bozena, 2001) and 0.2 mg l^{-1}BAP +0.01 mg l^{-1} IBA + 0.1 mg l^{-1} GA_3 (Li *et al*., 2009) for strawberry micropropagation.

A positive correlation between shoot proliferation and genotype, have been revealed in 'Serenata', compared with 'Pink Panda'. The influence of the genotypes on *in vitro* micropropagation potential of strawberry plants have been reported by several authors (Landi and Mezzetti, 2006, Kikas *et al*., 2006; Debnath and Teixeira da Silva, 2007).

In all combination of plant growth regulators used, basal part of micropropagated shoots exhibited a pink pigmentation of anthocyanins, which was less pronounced in 'Pink Panda' genotype. Also, presence of the auxins in the basic culture media, promoted low rhizogenic activity, in all subcultures.

After four weeks in culture, the percentage of microshoots rooted, number of roots and length of roots per culture was influenced by the different types and concentrations of auxins added in the rooting expression media and hormonal composition of the basic media used for explants micropropagation.

In 'Pink Panda' genotype, rooting induction of the shoots started about 10 days after the initiation of culture. A 100% rooting rate, was recorded in all treatments (RM1, RM2, RM3), but only for those shoots micropropagated in basic medium supplemented with IBA and lower concentration of IAA, such as MM1, MM2, MM3. The rooting rate was higher for this genotype, no significantly differences being observed between the variants of culture medium used for root induction (Fig. 2). Also, the mean root number per shoot varied between 2.37 and 6.06, and the average length of the roots between 5.46 and 18.49 mm. Highest values were calculated for RM1 and RM3 rooting media, but only for those shoots micropropagated on MM2 (Tabel 1).

In 'Serenata' genotype rooting induction of the shoots started about 16 days after the initiation of culture and responded by a lower rate of rooting on all the three variants of culture media. Excepting the shoots obtained by treatment with 1.0 mg l^{-1} BAP, 0.2 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3, no other combinations of growth regulators added to the MS basic medium resulted in significantly different rate of rooting. The highest rooting rate (80%) calculated for 'Serenata' genotype was obtained on medium supplemented with 0.25 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3. Shoots obtained on MM3 failed to induce rhizogenesis (Fig. 2). The statistical analysis revealed that 6.19 roots per shoot, with highest average length of 9.96 mm, was induced in this genotype by the combination 0.5 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3, for those shoots obtained on MM1 variant of medium (Table 1). When increasing IBA concentration from 0.25 mg l^{-1} to 0.5 mg l^{-1}, or replacing this auxin with IAA, a decrease in the mean root number and average roots length, respectively, or an inability of rhizogenesis was observed for those microshoots obtained on MM4 variant of medium

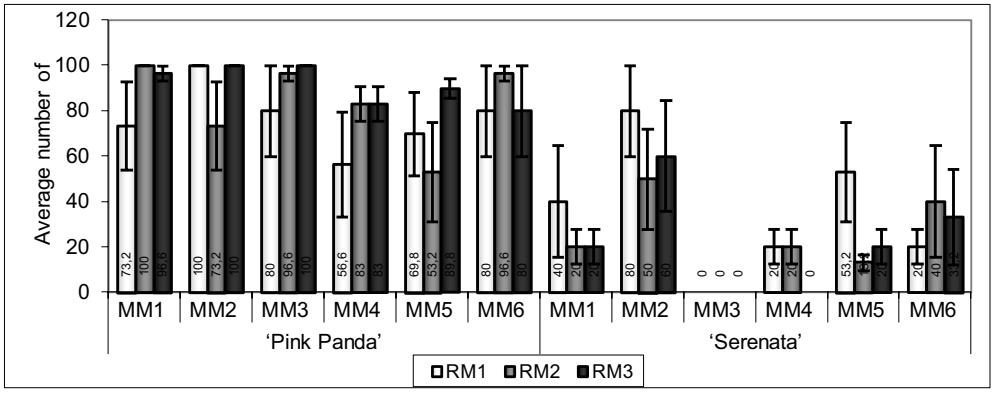

Figure 2. The rooting rate of 'Serenata' and 'Pink Panda' genotypes on half-strength MS basic medium containing 0.25 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3 (**RM 1**), 0.5 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3 (**RM 2**), 0.25 mg l^{-1} IAA and 0.1 mg l^{-1} GA_3 (**RM3**). Error bars are ± SE.

Table 1.

The average number of roots per shoots and the average length of the roots, recorded in 'Serenata' and 'Pink Panda' genotypes.

Shoots provenance		Mean number of roots per shoot			Average length of the roots (mm)		
		RM1	RM2	RM3	RM1	RM2	RM3
'Serenata'	MM 1 (MS)	2.13 ± 0.37^{abc}	6.19 ± 0.37^{a}	0.93 ± 0.03^{bc}	4.16 ± 2.5^{bc}	9.96 ± 1.01 ^{a}	1.43 ± 0.02^{bc}
	MM 2 (MS)	5.19 ± 0.49 ^{ab}	3.53 ± 0.49 ^{abc}	3.59 ± 1.08^{abc}	7.22 ± 2.13^{ab}	4.56 ± 2.19^{abc}	5.79 ±1.63^{abc}
	MM 3 (MS)	0	0	0	0	0	0
	MM 4 (MS)	2.4 ± 0.4^{abc}	1.46 ± 0.03^{abc}	0	1.7 ± 0.3^{bc}	1.7 ± 0.16^{bc}	0
	MM 5 (MS)	4.26 ± 1.78^{abc}	0.7 ± 0.02^{bc}	1.93 ± 0.46^{abc}	4.83 ± 2.61^{abc}	1.25 ± 0.42^{bc}	1.33 ± 0.12^{bc}
	MM6 (MS)	0.02^{bc}	3.26 ± 0.2^{abc}	2.87± 0.17 ^{abc}	1.33 ± 1.33^{bc}	4.46 ± 1.74^{abc}	3.38 ± 1.31^{bc}
'Pink Panda'	MM 1 (MS)	3.83 ± 0,98^{ab}	3.36 ± 0.94^{ab}	4.06 ± 0.46^{ab}	9.71 ± 2.07^{bc}	10.89 ± 2.78^{bc}	9.99 ± 2.15^{bc}
	MM 2 (MS)	6.06 ± 0.66^{a}	3.86 ± 1.01^{ab}	5. 0.63± 0.77^{a}	12.13 ± 1.92^{abc}	8.5 ± 2.19^{bc}	18.39 ± 0.98^{a}
	MM 3 (MS)	4.03 ± 1.04^{abcd}	3.99± 0.43^{ab}	5.03 ± 0.74^{a}	10.36 ± 2.69^{bc}	11.07 ± 2.15^{bc}	15.54^{ab} ± 0.56
	MM 4 (MS)	2.73 ± 1.16^{b}	3.67 ± 0.35^{ab}	3.67 ± 0.35^{ab}	8.4 ± 2.52^{bc}	8.88 ± 0.85^{bc}	8.51 ± 0.62^{bc}
	MM 5 (MS)	4.05 ± 1.2^{ab}	2.37 ± 1^{b}	4.08 ± 0.3^{ab}	10.45 ± 2.64^{bc}	7.51 ± 3.13^{c}	11.95 ± 1.97^{abc}
	MM6 (MS)	3.93 ± 0.98^{ab}	4.45 ± 0.46^{ab}	2.99 ± 0.92^{b}	7.83 ± 2.25^{bc}	11.67 ± 1.98^{abc}	5.46 ± 2.46^{c}

*Mean comparison (±SE): in each column values followed by different letters are significantly different, $p<0.05$. SE – standard error.

To induce rooting in the regenerated shoots the precise concentration of plant growth regulators is critical. Moreover, slightly lower rate of rooting was obtained with 'Pink Panda' genotype, when 0.5 mg l^{-1} IBA was added to the culture medium. Elhamdouni *et al*. (2000) have showed that auxins is only required during the initiation phase and becomes inhibitory for root out growth. The IBA added in MS medium, for root development of meristem derived plantlets, was reported by other authors (Kaur *et al*., 2005; Biswas *et al*., 2007). Furthermore, several workers reported that *in vitro* rooting was successfully induced on 1/4 MS or ½ MS medium (Yue *et al*., 1993; Moore *et al*., 1991). Also, satisfactory rooting can take place on full strength culture media, but is a very common practice to transfer shoots to be rooted from high strength media to less concentrated solution. The favourable effect of a diluted mineral solution on rooting can be explained by the reduction of nitrogen concentration (Driver and Suttle, 1987). As showed Bouza *et al*. (2004) rooting capacity of explants was influenced by a preliminary accumulation of endogenous auxins and cytokines, absorbed from the multiplication medium. In 'Pink Panda' genotype, shoots regenerated on medium containing BAP at 1.0 mg l^{-1} and IBA at 0.2 mg l^{-1} concentration responded by higher values for all three rooting characteristics analyzed, when rooting medium was supplemented with 0.5 mg l^{-1} IAA and 0.1 mg l^{-1} GA_3. In "Serenata" genotype, lower concentration of BAP and IBA in the multiplication medium was associated with significantly higher average root number and root elongation, when IBA concentration in the rooting medium was higher. Therefore, irrespective of the genotype, these results may be attributed to "carry over effect" from plant growth regulators in the regeneration medium, higher concentration of auxins and cytokines in the multiplication media favoring rhizogenesis of microshoots in those rooting media supplemented with lower concentration of auxins (Şuțan *et al*., 2009).

The plantlets were transferred in perlite and acclimatized gradually to green house conditions.

CONCLUSIONS

In both 'Pink Panda' and 'Serenata' varieties of *Fragaria x Potentilla*, the average number of shoots formed per primary explant was higher when the explants were subcultivated on the MS medium, rather than on LF medium Irrespective of the basic culture media, the micropropagation rate of both "Pink Panda" and "Serenata" varieties was demonstrated to be generally higher when combinations of 1.0 mg l^{-1} BAP, 0.2 mg l^{-1} IBA and 0.1 mg l^{-1} GA_3 are used.

In 'Pink Panda' and 'Serenata' genotype, half-strength MS medium with low concentration of IBA (0.25 mg l^{-1} IBA) was enough for maximum number of root per explant. In both intergeneric hybrids, higher concentration of auxins and citokinins in the multiplication media favoured rooting capacity of explants in those rooting media supplemented with lower concentration of auxins.

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EVOLUTION OF MICROAEROFLORA IN WORKSPACES DURING CATTLE SLAUGHTERING

D. Tibulca[1], Mirela Jimborean[1], C. D. Salagean[1], Laura Stan[1]

Key words: *slaughter room, total germs number, microaeroflora.*

ABSTRACT

The aim of this article is to monitor the evolution of microaeroflora during cattle slaughtering. Two microbiological parameters were determined: total mezofilic bacteria/m^3 air and total number of micromicetes/m^3 air. Research was performed in a cattle slaughtering plant and 3 experimental variants were taken into study.

Aforementioned parameters were evaluated and analyzed at the beginning of work and at the end of the slaughtering.

INTRODUCTION

The degree of air contamination in slaughtering rooms has a direct effect on contamination level of cow semi-carcases obtained.

The level of contamination reflects the hygienic conditions from slaughtering house; the composition of microflora reflects the contamination source and the efficiency of prophylactic measures against meat contamination.

The main contamination of meat with the highest significance for preservation and sanitation is during primary manipulation (bleeding, skinning, splitting, grooming) and an important role among other factors belongs to air (Bărzoi & Apostu 2002).

In Romania OMS 976/1998 is reglementing the maximum admitted limits (MAL) for a series of pathogenic microorganisms like total germ number TGN/ m^3 air and total yeast and mold number (micromicetes) TMN/ m^3 air from production and storage spaces.

Any deviation from these limits means low level of hygiene, disrespecting the GHP (Good Hygiene Practices) and implies immediate adoption of corrective actions (Dillon & Griffith 1995, Fose 2003, Mackey & Roberts 1993).

In research literature there are few data about level of air contamination in slaughtering spaces.

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MATERIALS AND METHODS

In order to determine TGN, TMN it was applied the method provided by Romanian Standard SR EN ISO 4833/2003 which is identical with European standard EN ISO 4833/2003 (Microbiology of food and animal feeding stuffs. Horizontal method for the enumeration of microorganisms. Colony-count technique at 30 degrees C).

In order to determine to total number of yeasts and molds it was applied the method provided by Romanian Standard SR EN ISO 7954/2001 which is identical with European standard EN ISO 7954/1987: "Microbiology-General guidance for enumeration of yeasts and moulds - Colony count technique at 25°C".

It was established a working protocol for the following experimental designs:

*V_1 – preoperational disinfection of slaughter room performed with NaOH 2,5% solution;

*V_2 – preoperational disinfection of slaughter room performed with Decontaminol 1% solution and implementation of HACCP plan;

*V_3 – preoperational disinfection of slaughter room performed with Decontaminol 1% solution and use of special instructed team for the purpose of sanitation –disinfection.

RESULTS AND DISCUSSIONS

Samples were collected and analyzed as follows:

* For V_1 - 46 samples;
* For V_2 - 47 samples;
* For V_3 - 35 samples.

The results obtained regarding the microaeroflora are presented in Tables 1 and 2.

Table 1

TGN/m^3 air from slaughtering room during cattle slaughtering

Process step	Before slaughtering process			At the end of slaughtering process		
Design experimental	V_1	V_2	V_3	V_1	V_2	V_3
Mean $\overline{X} \pm s_{\overline{X}}$	1559,56 ± 141,10	1178,45 ± 67,86	1310,00 ± 72,85	7143,75 ± 3252,35	3156,25 ± 634,74	2463,88 ± 116,74
Variability coefficient, %	61.36	39.48	32.90	45.44	69.67	14.22

Therefore, total germs number/ m^3 air is:

* in V_1 the increase is 5584.19 cfu/m^3 air, which represents 4.58 times the initial value;
* in V_2 the increase is 1977.8 cfu /m^3 air, which represents 2.68 times the initial value;
* in V_3 the increase is 1153.88 cfu /m^3 air, which represents 1.88 times the initial value.

Table 2

TMN /m^3 air from slaughtering room during cattle slaughtering

Process step	Before slaughtering process			At the end of slaughtering process		
Design experimental	V_1	V_2	V_3	V_1	V_2	V_3
Mean $\overline{X} \pm s_{\overline{X}}$	1018,75 ± 100,80	828,46 ± 75,37	793,21 ± 82,21	2848,33 ± 1015,96	1737,50 ± 259,80	1661,11 ± 54,02
Variability coefficient, %	67.11	62.37	61.31	61.14	51.80	9.76

Therefore, total moulds and yeasts number/ m^3 air is:

* in V_1 the increase is 1829.58 cfu/m^3 air, which represents 2.8 times the initial value;
* in V_2 the increase is 909cfu/m^3 air, which represents 2.1 times the initial value;
* in V_3 the increase is 867.9 cfu/m^3 air, which represents 2.1times the initial value.

From data presented in the two tables it can be seen that in all 3 experimental designs, the amount of total germs is increasing from the moment of starting the slaughtering to the end of it.

These results are explained by the fact that a series of microbiological polluting elements lead to increase of microaeroflora. These elements are: animal hygiene (hooves, hair, skin, horns), gastro-intestinal content, milk from udder, sanitation state of working personnel, equipments and tools, floors and walls (Bărzoi 1985).

The highest contamination degree takes place in first experimental design (V_1) when NaOH is used for sanitation.

Analysis of microaeroflora evolution in the three experimental designs shows the next conclusions:

For total germs number/ m^3 air is:

*in the beginning of the process there is a parameter decrease from 1559.56 cfu/ m^3 air (V_1) to 1178.45 cfu/ m^3 air (V_2) which is a normal evolution, followed by a increase to 1310 cfu/ m^3 air (V_3) which implies a microbiological contamination after sanitation step;

*in the final step of process there is a parameter decrease from 7143.75 cfu/ m^3 air (V_1) to 3156.25 cfu/ m^3 air (V_2), followed by a decrease to 2463.88 cfu/ m^3 air (V_3).

For total yeasts and moulds number/ m^3 air is:

*in the beginning of the process there is a parameter decrease from 1018.75 cfu/ m^3 air (V_1) to 828.46 cfu/ m^3 air (V_2) followed by a decrease to 793.21 1310 cfu/ m^3 air (V_3);

*in the final step of process there is a parameter decrease from 2848.33 cfu/ m^3 air (V_1) to 1737.5 cfu/ m^3 air (V_2), followed by a decrease to 1661.11 cfu/ m^3 air (V_3).

CONCLUSIONS

During bovine slaughtering process there is an increase of microaeroflora in the working space. This is determined by the animal sanitation being slaughtered, by the sanitation state of equipments and tools as well as sanitation state of floors and walls and

working personnel, efficiency in removal of legs, horns, stomachs and intestines, skins, blood, meats and wastes.

Massive multiplication of microorganisms and obvious contaminations of air happen in first experimental design with significant results in bacterial multiplication which evolve more than yeasts and molds.

Replacing the sanitation solution and application of hygienic measures leads to a significant decrease of air contamination (exception third experimental).

Comparing the results with recommendations applied from order 976/1998 it can be noticed that MAL for V_1 (7143.75 cfu/m^3 air) and V_2 (3156.25 cfu/m^3 air) compared to 3000 cfu/m^3 air for TGN, TYMN at the end of slaughtering process. For micromicetes there are deviations for V_1 (2848.33 cfu/m^3 air) compared to 2000 cfu/m^3 air in the same process step. There no recordings of deviations for other experimental designs applied in this study.

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VARIATION OF HEAVY METAL CONTENT FROM RAW MILK ACCORDING TO SEASON

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Key words: *heavy metals, raw milk, mercury, lead*

ABSTRACT

Heavy metals can get into breast milk in different ways, some of them can reach the animal and human body in quantities above certain limits and cause toxic conditions.

Milk can directly contaminate with some toxic metals during processing, during storage, transport,or from packaging.

INTRODUCTION

Heavy metals are elements which they own or their compounds in concentrations above certain levels in food are recognized as substances harmful to human body: lead, cadmium, tin, mercury, copper, zinc.

Potentially harmful heavy metals (Pb, Hg, Cu, Sn, etc..) can get into food and feed in various ways, but the most important source is the industrial emissions. They can also get into the food by administration of food additives (Cu, As) which helps to increase the weight and also by some compounds with drug role (salts of Zn, Cu). (V. Stanescu, 2010)

Mercury (Hg) is among the most harmful pollutant elements and is responsible for important and tragic consequences for humans and animals. Major sources of food contamination issues are the industrial, fossil fuel and organochlorine fungicides (Enache et al., 1994).

Lead is a cumulative toxic. One of the most common sources of environmental contamination with lead is the exhaust gases that contaminate the plants from surrounding streets (V. Stanescu, 2010).

Copper (Cu) is found naturally in all foods in very small quantities. In the body plays a role in oxygen biochemistry, tissue metabolism, synthesis of phospholipids.

Zinc (Zn) is a necessary bioelements both animal and vegetable kingdom, being involved in numerous metabolic processes in zinc-dependent enzymes (Popa, G. et al., 1986).

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Excess zinc can come from many sources: drug overdose or nutritional, industrial shows, use of fungicides with zinc, zinc or galvanized containers. The longer the milk is kept in galvanized pots the higher the zinc content is.

MATERIALS AND METHODS

The heavy metals content is determined using the atomic absorption spectrophotometry according to the standards:

- SR EN 14082/2003: Food. Determination of microelements. Determination of lead, cadmium, iron and chromium by atomic absorption spectrometry (AAS) after calcination.
- SR EN 14083/2003: Food. Determination of microelements. Determination of lead, cadmium, chromium and molybdenum by atomic absorption spectrometry with graphite furnace (GFAAS) after pressure digestion.
- SR EN 13805/2003: Food. Determination of microelements. Pressure digestion.
- SR EN 13804/2003- Determination of microelements. Performance criteria, general considerations and sample preparation.

Atomic absorption spectrophotometry (AAS) is based on the concentration of a chemical element by measuring the absorption of electromagnetic radiation of a specific wavelength to pass it in a medium containing as vapors evenly distributed, free atoms of the investigated element. The electromagnetic radiation crossing the medium is provided by a hollow cathode lamp (hollow) HCL emitting a narrow spectral line which is characteristic to the analyzed element.

The means of evaporation and atomization in AAS are the flame and the electrothermal evaporation.

In case of flame method there is the Flame Atomic Absorption Spectrometry (FAAS). The sample in liquid form is introduced into a high temperature (about 2500 ° C) flame of a mixture of air-acetylene aerosol generated by a nebulizer and suffering successive processes of atomization, ionization and excitation. The light signal transmitted through the flame is transformed by a photodetector in an electrical signal which is then amplified. After demodulation, the signal is recorded and converted into a digital size which is displayed. Hollow cathode lamp is selected depending on the analyzed element.

In electrothermal atomization the graphite furnace is used, the technique being symbolized GF-AAS. The sample, of microliter - size, is discontinuously introduced in the atomizer, which is subjected to thermal processing program. The entire quantity of sample is atomized and maintained a relatively long time (over 1 s) to the optical beam.

RESULTS AND DISCUSSIONS

Statistical analysis of the content of heavy metals in raw milk is shown in Table 1.

Statistically analyzing the variation of heavy metals, the seasons (Table 2) shows the following:

* for **mercury**:

➢ during autumn it recorded a mean ($\overline{X} \pm s_{\overline{X}}$) of 0.002467 ± 0.0004866 ppm and a variation coeffcient of 76.41%. The determined values ranged between 0.001 and 0.006 ppm.

➢ during winter it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.0022 ± 0.0004386 ppm and a variation coefficient of 77.22%. The determined values ranged between 0.001 şi 0.006 ppm.

➢ during spring it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.002067 ± 0.0003712 ppm and a variation coefficient of 69.56%. The determined values ranged between 0.001 and 0.006 ppm.

➢ during summer it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.002133 ± 0.0002557 ppm and a variation coefficient of 46.43%. The determined values ranged between 0.001 and 0.004 ppm.

Table1

Statistical analysis of the heavy metals detected in raw milk

Parameter	Mean, $\overline{\mathbf{X}}$	Standard deviation of the mean	Standard error of the mean, $\mathbf{s}_{\overline{\mathbf{X}}}$	Variation coefficient, %
Mercury	0,002217	0,001508	0,0001947	68.03
Lead	0,005983	0,005835	0,0007533	97.53
Copper	0,06723	0,07132	0,009207	106.07
Zinc	0,7700	1,002	0,1294	130.16

* for **lead**:

➢ during autumn it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.005067 ± 0.001236 ppm and a variation coefficient of 94.5%. The determined values ranged between 0,0 and 0.014 ppm.

➢ during winter it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.003733 ± 0.001322 ppm and a variaton coeffcient of 137.13%. The determined values ranged between 0.0 and 0.018 ppm.

➢ during spring it had a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0.005067 ± 0.001399 ppm and a variation coeffcient of 106.93%. The determined values situated between 0.0 şi 0.019 ppm.

➢ during summer it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.01007 ± 0.001631 ppm and a variation coefficient of 62.77%. The determined values ranged between 0.0 and 0.017 ppm.

* for **copper**:

➢ during autumn it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) of 0.05087 ± 0.01095 ppm and a variation coefficient of 83.4%. The determined values situated between 0,0 and 0,1 ppm.

➢ during winter it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0,06873 ± 0,02558 ppm and a variation coefficient of 144.15%. The determined values situated between 0,0 and 0,4 ppm.

➢ during spring it had a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0.088 ± 0.02245 ppm and a variation coefficient of 98.8%. The determined values situated between 0,0 and 0,3 ppm.

➢ during summer it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0.06133 ± 0.009898 ppm and a variation coefficient of 62.5%. The determined values situated between 0.0 and 0.1 ppm.

* for **zinc**:

➢ during autumn it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0.7333 ± 0.1953 ppm and a variation coefficient of 103.17%. The determined values situated between 0.0 and 3.0 ppm.

➢ during winter it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0.8667 ± 0.3425 and a variation coefficient of 153.05%. The determined values situated between 0.0 and 4.0 ppm.

➢ during spring it had a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 0.42 ± 0.1147 ppm and a variation coefficient of 105.79%. The determined values situated between 0.0 and 1.0 ppm.

➢ during summer it recorded a mean ($\overline{\mathbf{X}} \pm \mathbf{s}_{\overline{\mathbf{X}}}$) de 1.06 ± 0.3132 ppm and a variation coefficient of 114.43%. The determined values situated between 0.0 and 4.0 ppm.

Table 2

The limits of variation of heavy metal content

Nr. crt.	Season	Heavy metals (ppm)								
		Mercury			Copper		Zinc		Lead	
		<0,005	0,006-0,01	0-5	> 5	>0,5	0-5	> 5	0-0,02	>0,02
1.	A	13	2	15	-	-	15	-	15	-
2.	W	13	2	15	-	-	15	-	15	-
3.	S	14	1	15	-	-	15	-	15	-
4.	S	15	-	15	-	-	15	-	15	-
TOTAL		55	5	-	60	-		60	-	-
%		91,7	8,3	-	100	-		100	-	-

Key words: A/ autumn;WI/ winter; S/ spring; S/ summer

Lead poisoning has been known since antiquity. The main food sources of pollution are some mining and the related industries that pollute water plants and direct change of the lead in milk from vessels plumbifer tinsmith tin, lead in paint and varnish waste, batteries, flour from millstones grouted with lead (Stanescu, 2010; Enache et al., 1994).

According to the current regulations, harmonized with the European Union's laws, the maximum allowable mercury content in milk is 0.01 ppm (mg / kg product) (Reg. EC 1881/2006).

Regarding the milk contamination, there is the danger of exposing the cows to unwanted mercury levels, following administration of treated seeds, of fishmeal or ointment with mercury and the possibility that the mercury can pass in the milk (Stanescu, 2010; Enache et al., 1994).

The plants cannot take in the mineral circuit lead compounds, but lead concentration increases with exposure to large foliage plants. Plants and animals accumulate higher doses than man. One of the most common sources of environmental contamination with lead is the exhaust gases that contaminate the surrounding streets plants (Stanescu, 2010).

Maximum limit of lead residue in milk is 0.02 ppm (Order 106/2004).

Sources of food contamination with pesticides are pesticide treatments containing copper. Also, copper passes into milk during processing and storage because of the tools made from copper due to the corrosion processes (Costin, 2008; Popa et al., 1986).

Maximum limit of copper residue in milk is 0.5 ppm.

Zn is the responsible element for regulating the concentration of metal ions in the blood by its implication in melatinonine activites (Costin, 2008).

Maximum limit of zinc residue in milk is 5 ppm.

CONCLUSIONS

The heavy metals values (mercury, lead, copper, zinc) in the analyzed milk, situated under the maximum allowed limit (MAL) which is: 0.01 ppm (Hg), 0.02 ppm (Pb), 0.5 ppm (Cu), 5 ppm (Zn).

The heavy metals in the raw milk statistically had the following values:

- **Mercury**, a mean ($\overline{X} \pm s_{\overline{X}}$) of 0.002217 ± 0.0001947 ppm and a variation coefficient of 68.03%. The determined values ranged between 0.001 and 0.006 ppm.
- **Lead,** a mean ($\overline{X} \pm s_{\overline{X}}$) of 0.005983 ± 0.0007533 ppm and a variation coefficient of 97.53%. The determined values ranged between 0.0 and 0.019 ppm.
- **Copper**, a mean ($\overline{X} \pm s_{\overline{X}}$) of 0.06723 ± 0.009207 ppm and a variation coefficient of 106.07%. The determined values situated between 0.0 and 0.4 ppm.
- **Zinc**, a mean ($\overline{X} \pm s_{\overline{X}}$) of 0.77 ± 0.1294 ppm and a variation coefficient of 106.07%. The determined values situated between 0.0 and 4.0 ppm.

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RIPENING EFFECT OVER THE PHYSICAL CHARACTERISTICS OF DIFFERENT TYPE OF CHEESE

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Key words: *cheese, analysis, factors, interactions, quality*

ABSTRACT

The ripening technology has a major influence on several aspects of cheese manufacture. The presence of certain components, formed by the ripening, influence the taste and flavor of the cheese, the parameters that make a given cheese desirable (flavor, body, texture, melt and stretch properties). Analyses were made in the laboratories of the Faculty of Agricultural Sciences, Food Industry and Environmental Protection, Sibiu over three different assortment of cheese during 21 days. From this study we observed that the content of condensate substance, salt and acidity increased during the 21 days and the moisture content of cheese decreased. The highest values for this parameter were registered at the cheese Assortment 1. The pH value increases during maturation in the first stage and towards the end it drops. Cheesemaking technology should always tries to make a step forward in leading to cheese with more consistent composition and quality.

INTRODUCTION

Several studies have shown seasonal variations in composition of milk (Guinee, O'Brien et all., 2010). The gross composition of cheese milk, especially the concentrations of protein, casein and fat, has a major influence on several aspects of cheese manufacture, including rennet coagulability, gel strength, curd syneresis, cheese composition, yield and quality (Tita, 2005; Chintescu et all., 1982; Fox et all, 2004). The efficient manufacture of high-quality cheese consistently is a highly complex biotechnological process involving controlled destabilization and gelatin of the milk protein, fermentation of the milk sugar lactose to lactic acid, dehydration of the gel to obtain cheese curd and maturation of the curd to a ripened cheese with the desired quality attributes (sensory, aesthetic, usage, safety, convenience, wholesomeness, value for money) required by the consumer (Guinee, O'Brien et all., 2010, Albillos et all., 2006). The taste and flavor of the cheese depends not only on the presence of certain components, which is formed by the ripening, but needed a "balanced mix" of different substances that result from transforming the composition of this mixture through a botched, ripening causes the appearance of abnormal tastes from various

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sorts of cheese. In the areas of ageing should be ensured a certain temperature and relative air humidity of each specific assortment of cheese. (Costin, 2003; Euston et all, 2002)

MATHERIAL AND METHODS

Analyses for the study were made in the laboratories of the Lucian Blaga University, Faculty of Agricultural Sciences, Food Industry and Environmental Protection, Sibiu over three different assortment of cheese during 21 days (from 7 in 7 days). The technology of cheese making has two overriding goals: firstly, to establish the parameters that make a given cheese desirable (flavor, body, texture, melt and stretch properties); and, second, to develop a manufacturing and ripening protocol that will routinely reproduce these parameters every time this cheese is made. (Guinee, O'Brien, 2010; Moraru, 2003).

The physical or characteristics of cheese are governed by interactions between casein molecules (Tita, 2005). Factors that influence these interactions and which were analyzed in this study are the following:

- acidity
- pH
- sodium chloride
- dry matter and water.

The methodology used for the examination of cheese is:

1. Determination of acidity

The acidity of a certain volume of sample prepared for analysis is neutralized by titration with sodium hydroxide solution 0, 1N, in the presence of phenolphthalein as indicator.

2. Determination of pH.

Determination of the pH can be carried out using the paper indicator or pH meter. Procedure for the analysis of the sample is obtained from about 5 g cheese with 5 ...10 ml distilled water. If indicator solutions are used, a couple of drops are added in the sample to be analyzed and compare the color obtained with standard scale (consisting of test tubes with solutions with known pH). In the same way, it can be used that indicator papers and in the case of soft and fresh cheese can be applied directly onto a fresh section cut. The determination of pH is made by introducing an electrode into the sample to be analyzed, followed by reading directly the pH value on the scale.

3. Determination of sodium chloride content.

Chlorides are extracted from the sample with warm water (70 ...80°C), and chlorine ions are titrated with a solution of silver nitrate in the presence of potassium chromate as indicator.

4. Determination of the dry mix and water was made with moisture Analyzer-50 ML at a temperature of 105 ° C.

RESULTS AND DISCUTIONS

Cheeses are most consumed dairy products, especially the seasoned ones, such as cheeses in brine, cheese with semi-hard cheese and cheese paste with scalded cream paste-cheese. These sorts after the period of ripening are in smaller chunks (100-250 grams), packed in polyethylene film and then sent for marketing. It is well known that during

storage in retail centers appear changes of physical-chemical parameters. The aim of the paper is to see the evolution of these parameters in sales at those kinds of cheese the most consumed and how these are affected by movement.

The evolution of **acidity** was made for a period of 21 days (from 7 in 7 days) for all three kinds of cheese and the results are represented in the table below.

Table 1

Variation of acidity in three different assortment of cheese

No.	Type of cheese	Analysis period		
		7 Days	14 Days	21 Days
1	Cheese type 1	90°T	100°T	155°T
2	Cheese type 2	220°T	225°T	235°T
3	Cheese type 3	240°T	255°T	260°T

During this period of maturation the acidity increased for all kinds of cheese, as follows:

- The Cheese Assortment 1 acidity increases from 90°T to 155°T;
- The Cheese Assortment 2 acidity increases from 220 °T at 235°T;
- The Cheese Assortment 3 acidity increases from 240°T to 260°T.

Notice that in the last analysis period (days 14-21) there is a stronger acidification for Cheese Assortment 1 with 55 ° T, by transforming milk lactose into lactic acid.

Using indicator paper we can observe that the **evolution of pH** of the three types of cheese changed. Analyses were made for a period of 21 days, the analysis being made of the 7 in 7 days. The results are presented in the following figure.

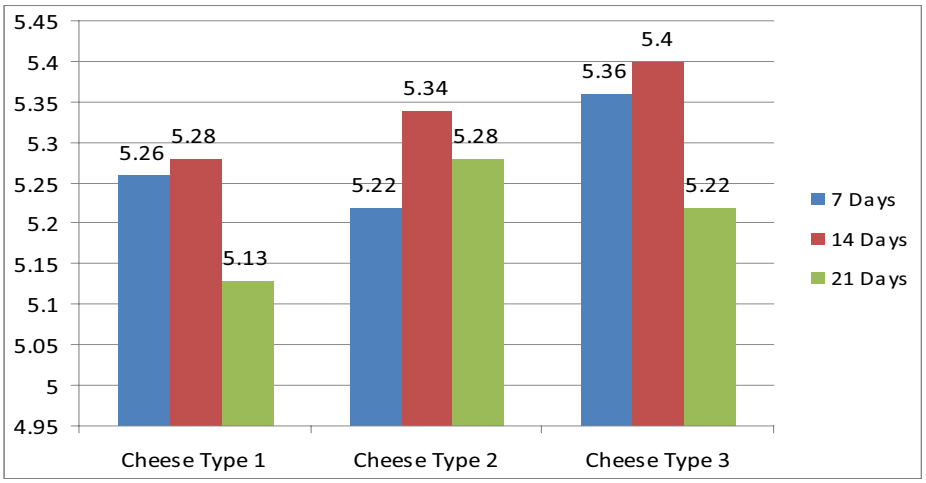

Fig. 1 Evolution of pH in the range of cheese studied

Also, we make determination of **sodium chloride content** over 21 days. Sodium chloride content was determined by titration method. The results obtained are shown in the following figure.

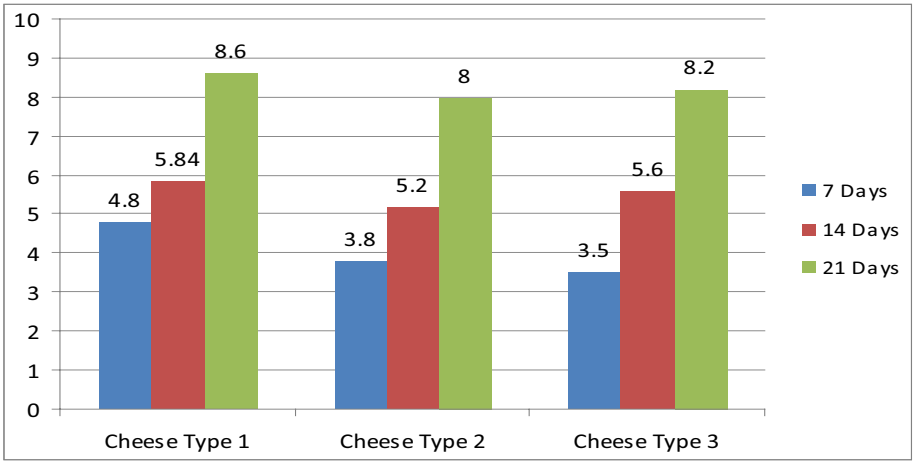

Fig. 2 The evolution of sodium chloride content, three kinds of cheese

During maturation, it causes a diffusion of the salt by the middle part of cheese. After a certain period of time it is noticed almost an equalization of the salt content in cheese mass. Thus the salt content increased during the 21 days at all three types of cheese. Finally the content of salt was: Cheese Assortment 1 with 8.6% NaCl, cheese assortment 2 with 8% NaCl and cheese Assortment 3 with 8.2% NaCl.

The evolution of the **dry mix and water** has been observed and the analyses were performed using moisture Analyzer ML-50. The results thus obtained are represented in the following figure:

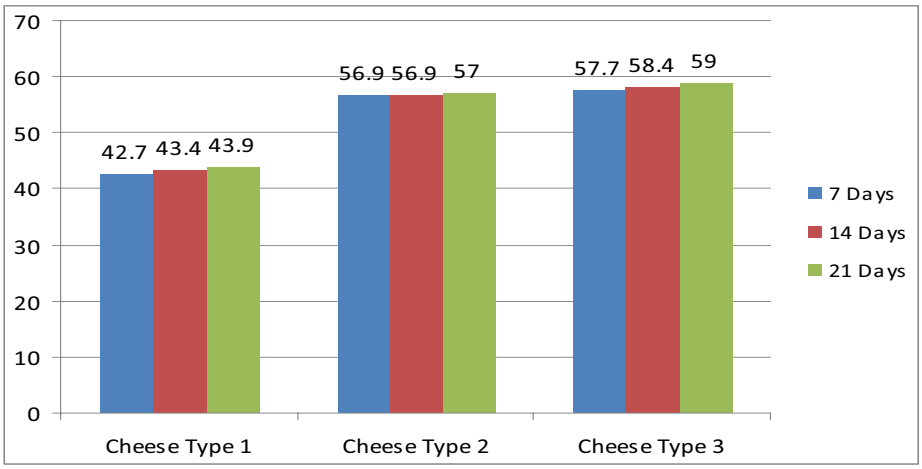

Fig. 3 Evolution of dry mix and water during 21 days over the three assortments of cheese

The content of the dry mix and water grew over the period of review for all three assortment of cheese. The percentage of condensate substances in the 21 days growth: 1.2% for Cheese Assortment 1, 0.1% for Cheese Assortment 2 and 1.3% for Cheese Assortment 3. Moisture content of cheese decreased: the Cheese Assortment 1 humidity has decreased by 1.2% after 21 days, the Cheese Assortment 2 with 0.1% and by 1.3% for Cheese Assortment 3.

CONCLUSIONS

Following study content of condensate substance, salt content and acidity increased during the 21 days, and the moisture content of cheese decreased. The highest values for this parameter were registered at the cheese Assortment 1.

The pH value increases during maturation in the first stage, and towards the end it drops.

During the storage of the products in outlet centers do not appear essential changes of physico-chemical parameters that could adversely influence the quality of the cheese. Increasing or decreasing values of physical-chemical parameters is the citadel in the requirements of standards in force.

The main conclusion of this study is in accordance with Guinee, who said „cheesemaking technology always tries to make a step forward in leading to cheese with more consistent composition and quality.

The impetus towards more consistent cheese is driven by consumer demands for consistent sensory characteristics on repeat purchase and in having more knowledge in relation to the intake of specific nutrients".

In modern cheese factories, minimizing such in-process and product variation is a key goal of the quality assurance function.

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THE IMPACT OF CLIMATIC CHANGES ON GRAPEVINE

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Key words*: grapevine, climatic changes, pulp maturity, phenolic maturity*

ABSTRACT

Climate changes are a reality today and a challenge for scientists who need to evaluate them and to identify the effects and impact on socio-economic level of each social and financial component. Although it was acclimatized over times in different viticultural climates, grapevine is a plant which has its optimal limits in physiological and metabolic meaning. The researches performed at the IC-DVV Valea Calugareasca have shown that over the past 50 years, the climate has changed in the meaning that heating resources increased and the rainfall resources decreased. Changes were lower in the period between 1960-2000 and bigger in the last 10 years. On an annual basis for 50 years the temperature has increased with 1.9°C. It has registered, also, years with hot summers which significantly influenced the grapevine. During hot summers, vine phenology has changed in that way in which the period between two phenophases was shortened greatly. Also, grapevine evolved under conditions of heat and hydric stress. The impact was relatively easy to assess at the grapes level at harvest maturity. The maturity of the pulp was much evolved, showing a very high glucoacidimetric index due to its very low acidity. Black grapes skins, which were not exposed to direct sunlight, were very well matured. The maturity of the seeds showed a great variability depending on the variety.

INTRODUCTION

The viticultural climate of terroir influences the physiology of grapevine and the ripening of the grapes. In the last 20 years it has registered significant changes due to climate changes. Various studies have been aimed at assessing the impact of climate changes on the grapevine in order to obtain knowledge which can be used to substantiate new cultural models adapted to the present viticultural climate.

In the year 2005, based on research conducted by Jones et al., it was shown that in "17 vineyard regions in the world the warming is significantly in the years 1950-1999". Other researches have shown that global warming has strongly influenced the vegetative cycle of the grapevine (Barbeau et al., 2007). Grapevine phenology was modified, all phenological stages are shorter, except bud burst which became late. In these conditions, harvest time has recorded 15-30 days earlier (Sequin, 2009). The shorter period of ripening grape berries has changed the composition of grape berries. The concentration in sugar has significantly increased and the acidity decreased. In Alsace, in the last 30 years, the alcoholic potential wines increased by 2.5° (Duchere et al., 2005).

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MATERIAL AND METHODS

The first objective. The description of viticultural climate evolution of DOC Dealu Mare Valea Calugareasca area in the 1961-2010 period.

In order to achieve this objective, it was used the information of wine climate from the database of IC-DVV Valea Calugareasca. The grapevine's parameters of the annual climate and of the vegetation stages for the 1971-2012 period were compared with the average registered in the 1961-1970 period.

The second objective. The characterization of viticultural climate of excessive drought years

The viticultural climate of the very dry harvests was characterized based on the following monthly parameters: monthly average temperature, the absolute maximum and medium values, the precipitation and air hygroscopicity. For each summer month the number of the days with the air temperature bigger than 30°C was calculated, this being the threshold at which the vine is significantly affected.

The third objective. The assessing of the impact of climate changes on wine grapes at harvest maturity

The maturity of the grapes at harvesting was evaluated by pulp, skin and seeds analysis. The maturity of the pulp is directly proportional with glucoacidimetric index (sugar/acidity). The phenolic maturity is directly proportional with total phenolic potential, the anthocyanins potential and anthocyanin extractability (maturity of skin) and the potential of tannins seed (maturity of seeds). The determination of these parameters was performed according to the Glories method (Vivas et al., 1998).

RESULTS AND DISCUSSION

The climate evolution during 1961-2010 period

Over the past 50 years the climate of the DOC Dealu Mare-Valea Calugareasca area has registered significant changes.

In the 1961-1970 period the average annual air temperature was 11°C. This value increased with 0.4°C (3.6%) in the 1971-2010 period and with 2.0°C (18.2%) in 2011. The fluctuations are much higher in case of the average monthly temperature (Figure 1).

The lowest values were registered in the 1971-2010 period, ranging between -0.4°C (April) and +0.8°C (June and July). In 2011 the increase of the air temperature has ranged between 0.4°C (more) and 4.6°C (September). In 2012 the differences between the values of temperature were very high, with values over 4.0°C in June, July and September.

The monthly rainfall amounts are different in the 1971-2012 period, in comparison with those registered in the 1961-1970 period (Figure 2). The biggest negative differences in 2012 were recorded in July (-41.6 mm) and September (-45.9) in 2011 and in June (-59.4) in 2012.

The evolution of the variation of average monthly air hygroscopicity in the 1971-2012 vegetation period is presented in figure 3, in comparison with the average value registered in 1961-1970 years.

The hygroscopicity has tagged small variations in the 1971-2010 period and larger in 2011 and 2012. During year 2012, the air hygroscopicity was lower with 14% in August and with 13% in September.

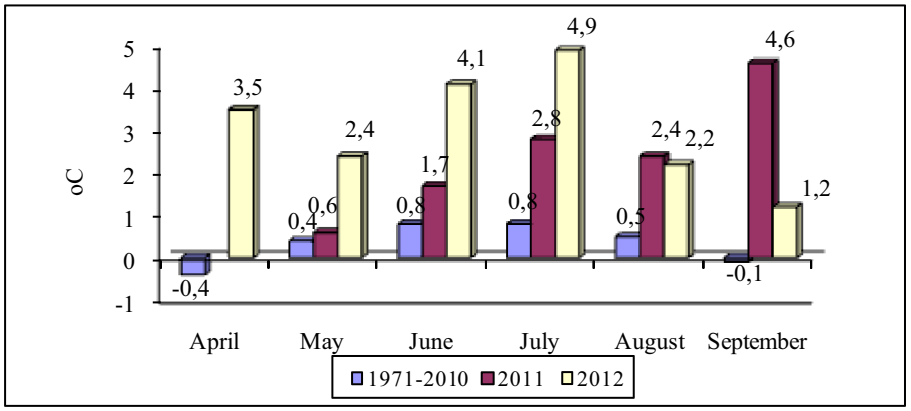

Fig. 1. The variation of the monthly average air temperature in the 1971-2012 period in the DOC Dealu Mare-Valea Calugareasca area (in comparison with the 1961-1970 period)

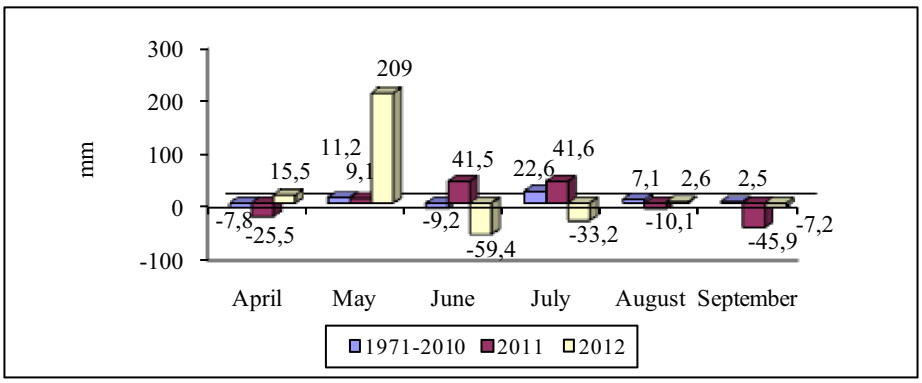

Fig. 2. The variation of the monthly rainfall in the 1971-2012 period in the DOC Dealu Mare-Valea Calugareasca area (compared to the period 1961-1970)

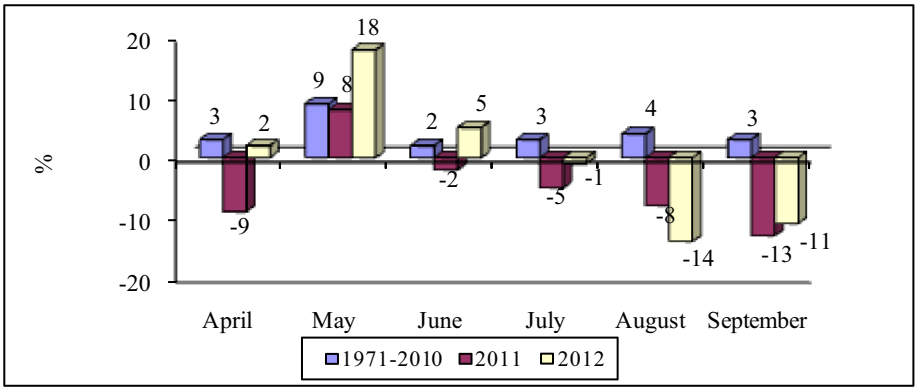

Fig. 3. The variation of the monthly hygroscopicity in the 1971-2012 period in the DOC Dealu Mare-Valea Calugareasca area (compared to 1961-1970 period)

The analysis of climatic monthly parameters showed the fact that 2012 was an excessive dry wine year.

The characterization of the viticultural climate during the drought years

The intensity of the drought in the year 2012 was determined function of the length of drought periods (10 consecutive days). In July the days with temperatures exceeding 30°C were in number of 29 and in August, in number of 16.

21 consecutive days had hot temperature. Other indices and climatic parameters, compared with those of the 1961 to 1970 period are presented in table 1.

Table 1

Parameters and climatic indices of the summer months of 2012 year

Month	The period/ year	Huglin Index	The duration of sunshine (hours)	Rainfall (mm)	Higrosco-picity (%)	The number of the days with the air temperature > *30°C*
July	1961-1970	472.3	289	62.0	63.0	*9*
	2012	652.8	351	29.4	63.6	*29*
August	1961-1970	472.3	287	60.0	65.0	*11*
	2012	*565.8*	*326*	*63.1*	*50.2*	*16*

The impact of climatic changes on grapes at harvest maturity

At Valea Calugareasca viticultural center, in the normal viticultural years, the glucoacidimetric index has the next values, specific for every variety: for red wines 36 (Cabernet Sauvignon) and 53 (Pinot noir); in case of white wines is 26 (Feteasca regala) and 89 (Muscat Ottonel), (Table 2).

Table 2

The glucoacidimetric index of the grapes for specific wines in normal viticultural years (DOC Dealu Mare-Valea Calugareasca area)

The indicator	*Red wines varieties*				
	Burgund mare	Cabernet Sauvignon	Feteasca neagra	Merlot	Pinot noir
The glucoacidi-metric index	40	36	44	42	53
	White wines varieties				
	Chardonnay	Feteasca regala	Muscat Ottonel	Riesling italian	Sauvignon
	44	26	89	56	41

In 2012, a drought excessive year, the glucoacidimetric index of wine grapes was significantly higher than in normal viticultural years, the relative differences ranging between 4% (Muscat Ottonel) and 53% (Pinot noir) (Figure 4).

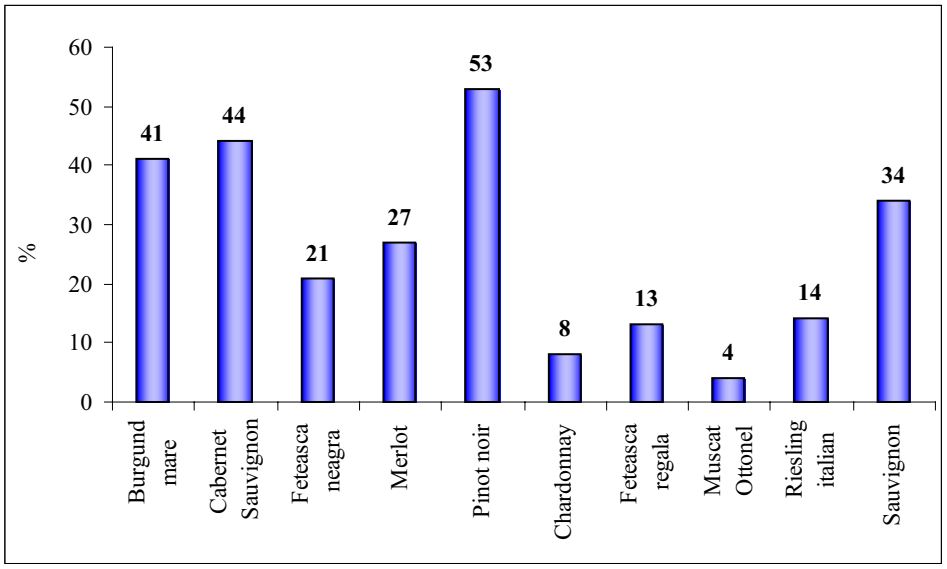

Fig. 4 The relative differences of the glucoacidimetric index for wine grapes (2012/the average of normal viticultural years)

The smallest differences were registred in case of Muscat Ottonel and Chardonnay varieties and the highest at Burgund mare, Cabernet Sauvignon and Pinot noir varieties.

The results of the phenolic maturity analysis for the 2012 wine grapes harvest, are presented in table 3.

Table 3

The parameters of phenolic maturity of the red wines, 2012 harvest (DOC Dealu Mare-Valea Calugareasca area)

The variety	The total phenolic potential (UA)		The anthocyanin potential (mg/l)		The tannin potential of seeds (UA)		*Total points*
	Value (UA)	The grade	Value (mg/l)	The grade	Value (UA)	The grade	
Burgund mare	47	2	1132	2	24	2	***6***
Cabernet Sauvignon	46	2	1437	1	8	1	***5***
Feteasca neagra	42	2	1119	2	7	1	***5***
Merlot	46	2	1281	1	15	1	***4***
Pinot noir	62	1	480	5	48	4	***9***

The obtained experimental data put into evidence that the Merlot variety registered an excellent phenolic maturity, Feteasca neagra and Cabernet Sauvignon varieties registered a very good maturity. Pinot noir variety showed an average maturity because of the incomplete maturity of the seeds.

The extractibility of anthocianins was excellent in case of Feteasca neagra variety (88%), very good at Pinot noir variety (74%), good at Cabernet Sauvignon (67%) and Merlot (61%) varieties and average at Burgund mare variety (Figure 5).

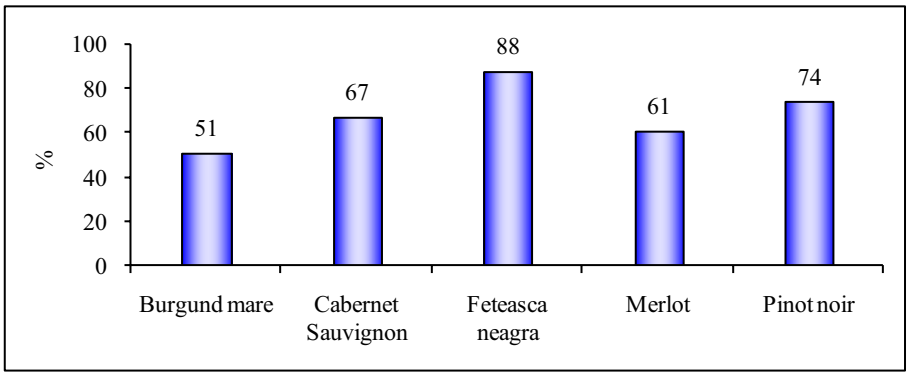

Fig. 5. The relative differences of the extrability of anthocianins for wine grapes (2012/the average of normal viticultural years)

CONCLUSIONS

The climate of the viticultural DOC Dealu Mare-Valea Călugărească area has changed over the past 5 decades, recording the increase of the average temperature, the diminishing of the pluviometrical regime and hygroscopcity of the air; the changes were significant during the summer months.

The year 2012 has been excessively a dry year, being characterized by a heliothermic surplus in the period of vegetation of the vine, when the number of the days with temperatures bigger than 30°C was far greater than average multi-annual; the rainfall was reduced and unevenly distributed.

In specific climatic conditions with excessive drought years, the maturity of the pulp is unusual; the musts are unbalanced by taste.

The phenolic maturity varieties for red wines are very good at the skin level and variable, depending on the variety, at the seeds level.

The extractibility of antocianins was excellent at Feteasca neagra (88%), very good at Pinot noir (74%), good for the Cabernet Sauvignon (67%) and Merlot varieties (61%) and average at Burgund Mare.

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THE VARIABILITY OF COWPEA CHARACTERS UNDER THE INFLUENCE OF SOME BIOACTIVE EXTRACTS IN LUVOSOIL CONDITIONS FROM ARDS SIMNIC

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Key words: *Vigna unguiculata, cowpea, character, bioactive extract*

ABSTRACT

Cowpea (Vigna unguiculata) L. Walp. is an important edible legume crop which play an important nutritional role in the world generally and in Africa particularly. Two natural bioactive extracts and their combination (Yucca schidigera extract, Salix babylonica extract, Yucca+Salix), applied in three treatments (T1-Water, Yucca, Salix, Yucca+Salix), T2-(Water, Yucca, Salix, Yucca+Salix)+dried lives alfalfa extract, T3-(Water, Yucca, Salix, Yucca+Salix)+soybean extract, were evaluated for their potential influence to cowpea characters variability. Comparatively with the control (water treatment) all studied cowpea characters recorded higher values when were applied bioactive natural extracts, but the differences weren`t statistically assured. For all treatments only Yucca extract determined a medium variability of plant height. The leaves number was strongly influenced by Yucca extract and Yucca+Salix extract application recording high variation coefficients, while the branches number was strongly unstable regardless treatment or time of application.

INTRODUCTION

Cowpea *Vigna unguiculata* (L.) Walp. is an important edible legume crop gaining recognition in the world generally and in sub-saharan Africa, Asia, Central and South America particularly, known as "poor man`s meat" because of its high protein content (Ehlers and Hall, 1997, Singh et al., 1997, Phillips et al., 2003). The precise location of the center of species origin is rather difficult to determine. Previous studies on the origin and domestication were done, suggesting that Nigeria is the most important (Ng and Maréchal, 1985, Vaillancourt and Weeden, 1992, Ng, 1995). According to Singh and Eaglesfield (2000), cowpea seed can be grown under various production systems including rainfed and irrigated environments as well as in areas of poor soil in low rainfall regions. Many cowpea varieties are tolerant to acidic soils, particularly poor soil fertility and shading by other crops (Summerfield and Roberts, 1985).

Previous studies regarding the influence of natural bioactive extracts to the control of different pests and root/soil borne fungal pathogens show that these extracts can be effective (Abiala et al., 2010, Killiani et al., 2011, Lodama, 2011, Shukla and Dwivedi, 2012) but little work has done regarding the influence of natural bioactive extract to the variability of cowpea characters. Thus, the aim of this study was to determine this previous

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aspect testing different natural plant extracts applied to cowpea in the natural conditions of luvosoil from Simnic area.

MATERIAL AND METHODS

In 2012 year the autochthonous cowpea variety Aura was evaluated in natural conditions on luvosoil from Simnic Agricultural Research Station regarding the influence of bioactive natural extracts to main characters variability. The experience was designed as a complete split block with four variants and 4 replications. The size of each plot was 25 m^2. The trail had two factors as follows:

Factor A – natural extract

a1- water (control), a2- *Yucca schidigera* extract

a3- *Salix babylonica* extract, a4- Yucca + Salix extract

Factor B – treatments

I – only with natural extracts, II – natural extracts + alfalfa extract

III- natural extracts + enzyme extract of soybean

The extracts preparation was done according with the following procedure:

Preparation of *Yucca schidigera* extracts solution

The plants roots were washed previously by soil residue and then peeled and cut in small cubs and put into a flask with distilled water (1:20 ratio, 100 g Yucca roots and 2l distilled water) to be turned into pulp. The mixture was left to stand 24 hours at 40 ^oC and then the insoluble material was separated by vacuum filtration. Thus was obtained shock solution for which freshness preservation was put into plastic bags and frozen. Working solution was made by dissolving two cubes of frozen shock solution in two liters of water.

Preparation of *Salix babylonica* extracts solution

For extract preparation were used *Salix babylonica* branches (1-2 cm thick) cut to 1-2 cm. There were used 100 g cut branches to 2 liters of cold distillate water and rest 48 hours, filtered and frozen. Working solution was made by dissolving two cubes of frozen shock solution in two liters of water.

Unused solution must be discarded.

Preparation of dried lives alfalfa extracts solution

Concentration of this solution was 1:25 (100 g of alfalfa dried leaves with 2500 ml distillated water heated to 40 ^oC for 48 hours. After 24 hours the aqueous mixture was filtered under vacuum. After filtration process to keep its freshness the extract was put into plastic bags and frozen. When using was thawed the cubes required for 100 ml aqueous extract. 100 ml of this solution was added to each of the above extracts.

Preparation of soybean extracts solution

Soybeans were ground very fine and the oil content was removed with a solution of petroleum ether and Soxhelt device. The soybean flour was dried to 30-45 minutes then dried in oven at 80 ^oC for one hour. After drying, it was prepared a solution with deionized water in ratio of 1:10. Solution was corrected to pH 7 by 2 N NaOH. To stabilize the solution this was mixed for 1 hour at room temperature. To remove insoluble material the solution was centrifuged at 6000 rot/min. Insoluble material recovered was corrected with 2 NHCl and heated in a water bath at 32 ^oC. Was added 5% enzyme solution (1 ml for each 200 ml insoluble material) and allowed to clot. The crud was separated by a screen similar to that used for cheese. Drained crud is an enzyme hydrolyzed soybean which has high content of cysteine and tryptophan in biological active form „1". For watering plants was made solutions with deionized water for ratio of 1:20.

During the vegetation period was determined number of plants/plot, growing rhythm, noting plants high, number of leaves, number of branches, number and length of

internodes to 10 plants in each variant and replication. Foliar area was determined with Area Meter AM300 device. Area and leaf area were performed on three leaves of each variant and replication. The characters variability was calculated based on biometric determinations calculating arithmetic mean, standard deviation and variation coefficient. Setting the variation coefficient was done according with Ceapoiu (1968).

RESULTS AND DISCUSSION

For the first treatment plants height ranged between 23,73 cm (control) and 26,7 cm when was applied one treatment with Yucca extract. This character was very stable when plants were watered with Yucca+Salix extract solution (Table 1).

Table 1

Plants height and its variability according to the first treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	23,73±2,72	11,48%	Mt.
	Yucca schidigera	26,07±2,93	11,26%	2,34
	Salix babylonica	24,89±2,92	11,74%	1,16
	Yucca+Salix	25,43±2,03	8,00%	1,70

Variability: <10% = low, 10-20% = medium, >20 = high

DL 5% = 3,4, DL 1% = 4,89, DL 0,1 = 7,18

For the second treatment plants height values were higher than in the first treatment and ranged between 24,72 cm (Yucca+Salix extract) and 25,5 cm (Yucca extract). Stability of this character was more pronounced than for the first treatment because for three variants (water, Salix, Yucca+Salix) the variation coefficients were below 10% (Table 2). Very small deviations comparatively with the control showed no differences statistically assured.

Table 2

Plants height and its variability according to the second treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	24,78±1,89	7,62%	Mt.
	Yucca schidigera	25,50±3,09	12,10%	0,72
	Salix babylonica	25,47±1,91	7,48%	0,69
	Yucca+Salix	24,72±2,14	8,64%	-0,06

Variability: <10% = low, 10-20% = medium, >20 = high

DL 5% = 3,67, DL 1% = 5,28, DL 0,1 = 7,77

As plants grow the height values increased ranging between 28,39 cm (control) and 30,33 cm (Salix extract) (Table 3). For the third treatment the variability coefficients exceeded 10% indicating a medium variability of this character. Even in this case the differences among tested variants were not statistically assured.

Table 3

Plants height and its variability according to the third treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	28,39±3,64	12,83%	Mt.
	Yucca schidigera	30,05±5,28	17,56%	1,66
	Salix babylonica	30,33±5,69	18,76%	1,94
	Yucca+Salix	29,40±4,07	13,83%	1,01

Variability: <10% = low, 10-20% = medium, >20 = high

DL 5% = 6,94, DL 1% = 9,99, DL 0,1 = 14,69

For the first treatment the number of leaves ranged between 7,8 (control) and 8,51 leaves when was applied Salix extract solution (Table 4). After one week new determinations were done this character ranged between 8,77 leaves (Yucca +Salix extract) and 9,63 (Yucca extract) (Table 5). As the number of leaves increased as a result of advancement in plant vegetation the character stability changed.

The variation coefficients showed that this character had a medium variability for all tested variants, excepting the first treatment with Yucca and Salix when the variation coefficient was <10%.

Table 4

Leaves number and its variability according to the first treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	7,80±1,04	13,29%	Mt.
	Yucca schidigera	8,51±1,34	15,72%	0,71
	Salix babylonica	8,15±1,07	13,13%	0,35
	Yucca+Salix	8,30±0,71	8,53%	0,50

Variability: <10% = low, 10-20% = medium, >20 = high
DL 5% = 0,82, DL 1% = 1,17, DL 0,1 = 1,72

Table 5

Leaves number and its variability according to the second treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	9,12±1,53	16,78%	Mt.
	Yucca schidigera	9,63±1,81	18,84%	0,51
	Salix babylonica	9,55±1,16	12,15%	0,43
	Yucca+Salix	8,77±1,24	14,15%	-0,35

Variability: <10% = low, 10-20% = medium, >20 = high
DL 5% = 1,72, DL 1% = 2,48, DL 0,1 = 3,64

For the third treatment leaves number ranged between 11,48 (control) and 13,13 leaves (Salix extract) (Table 6). This character was strongly influenced by Yucca extract and Yucca +Salix extract application recording high variation coefficients. The highest influenced in this treatment with enzyme extract of soybean was observed for control variant (water).

Table 6

Leaves number and its variability according to the third treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	11,48±3,48	30,28%	Mt.
	Yucca schidigera	12,74±2,72	21,31%	1,26
	Salix babylonica	13,13±2,36	17,94%	1,65
	Yucca+Salix	12,18±2,44	20,02%	0,70

Variability: <10% = low, 10-20% = medium, >20 = high
DL 5% = 4,07, DL 1% = 5,85, DL 0,1 = 8,60

The branches number was strongly unstable regardless treatment or time of application. For the first treatment the branches number ranged between 5,65 (control) and 6,48 (Yucca+Salix extract) (Table 7).

In case of treatment with Salix and Yucca extract the number of branches had a significantly higher value than what the control had indicating that this character was influenced by the treatment applied.

Table 7

Branches number and its variability according to the first treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	5,65±0,94	16,56%	Mt.
	Yucca schidigera	6,30±1,22	19,41%	0,65
	Salix babylonica	6,29±1,77	28,07%	0,64
	Yucca+Salix	6,48±0,99	15,30%	0,83*

Variability: <10% = low, 10-20% = medium, >20 = high
DL 5% = 0,75, DL 1% = 1,08, DL 0,1 = 1,58

When the second treatment was applied the number of branches was relatively uniform ranging between 7,44 (Yucca+Salix extract) and 7,90 (Yucca extract) (Table 8). Small differences recorded between the treatments didn`t show statistically deviations.

Table 8

Branches number and its variability according to the second treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	7,54±1,38	18,33%	Mt.
	Yucca schidigera	7,90±1,51	19,09%	0,36
	Salix babylonica	7,64±2,12	27,79%	0,10
	Yucca+Salix	7,44±1,41	18,92%	-0,10

Variability: <10% = low, 10-20% = medium, >20 = high
DL 5% = 1,30, DL 1% = 1,87, DL 0,1 = 2,75

For the third treatment the variability of this character was high (>20%) for all treatments applied (Table 9). This aspect is emphasized also by high limit differences pointing out statistically, although the values ranged between 11,08 (control) and 14,78 (Salix extract).

Table 9

Branches number and its variability according to the third treatment applied

Species	The Extract	$\bar{x}\pm s_x$ (cm)	s%*	Diff.+Signif.
Cowpea	Water	11,08±3,30	29,79%	Mt.
	Yucca schidigera	12,23±2,87	23,46%	1,15
	Salix babylonica	14,78±6,05	40,91%	3,70
	Yucca+Salix	11,71±2,42	20,69%	0,63

Variability: <10% = low, 10-20% = medium, >20 = high
DL 5% = 5,35, DL 1% = 7,70, DL 0,1 = 11,32

CONCLUSIONS

Generally, for all studied characters recorded values for water treatment were lower than those recorded when *Yucca schidigera*, *Salix babylonica* and Yucca+Salix treatments were applied. Recorded positive or negative differences compared to the control in most of them were not statistically assured, which shows wick influence of natural bioactive extract treatments to main morphological characters of cowpea. There were exceptions, mainly due to the association of Yucca extract with Salix extract in case of branches number for the first treatment.

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RESEARCH ON THE INFLUENCE OF NATURAL BIOACTIVE EXTRACTS TO FEW MORPHOLOGICAL CHARACTERS OF COWPEA

Tută Claudia Elena[1]

Key words*: cowpea,bioactive extract, plant height, branche, pod, internode*

ABSTRACT

Cowpea (Vigna unguiculata) L. Walp. is an important edible legume crop which play an important nutritional role in the world generally and in Africa particularly. Two natural bioactive extracts and their combination (Yucca schidigera, Salix babylonica, Yucca+Salix) were applied using a three treatments scheme T1(water, Yucca schidigera, Salix babylonica, Yucca+Salix), T2 (water, Yucca schidigera, Salix babylonica, Yucca+Salix)+alfalfa extract, T3 (water, Yucca schidigera, Salix babylonica, Yucca+Salix)+soybean extract. Generally, all studied characters (plant height, branches number, internodes number, pods number, pods length) weren`t significantly affected by the application of natural bioactive extracts for all treatments applied and all differences recorded comparatively with the control weren`t statistically assured.

INTRODUCTION

Vigna unguiculata is a diploid species, with 2n = 22. All cultivated cowpeas are grouped under *V. unguiculata* subspecies *unguiculata* which is subdivided into four groups namely *unguiculata* (cowpea), *biflora* (catjang), *sesquipedalis* (yard-long or asparagus bean) and *textilis* (Maréchal et al., 1978, Ng and Maréchal, 1985, Menéndez et al., 1997). Cowpea, *Vigna unguiculata* (L.) Walp. is a widely adopted, stress tolerant grain legume, vegetable and fodder crop grown in warm to hot regions of Africa, Asia and Americas (Ehlers and Hall, 1997, Singh et al., 1997). As a legume, cowpea is valued for the high protein content of its grains but also for the vitamins and minerals present in the young leaves, pods and peas (Nielsen et al., 1997, Phillips et al., 2003).

Previous studies regarding the influence of natural bioactive extracts to the control of different pests and root/soil borne fungal pathogens show that these extracts can be effective (Abiala et al., 2010, Killiani et al., 2011, Lodama, 2011, Shukla and Dwivedi, 2012) but however little work has done regarding the influence of natural bioactive extract to cowpea characters. Thus, the aim of this study was to determine the influence of these natural extracts to the most important morphological characters of cowpea.

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MATERIAL AND METHODS

In 2012 year the autochthonous cowpea variety Aura was evaluated in natural conditions on luvosoil from Simnic Agricultural Research Station regarding the influence of bioactive natural extracts to the most important morphological characters of cowpea. The experience was designed as a complete split block with four variants and 4 replications. The size of each plot was 25 m^2. The trail had two factors as follows:

Factor A – natural extract

a1- water (control)

a2- *Yucca schidigera* extract

a3- *Salix babylonica* extract

a4- *Yucca* + *Salix* extract

Factor B – treatments

I – only with natural extracts

II – natural extracts + alfalfa extract

III- natural extracts + enzyme extract of soybean

The extracts preparation was done according with the following procedure:

Preparation of *Yucca schidigera* extracts solution

The plants roots were washed previously by soil residue and then peeled and cut in small cubs and put into a flask with distilled water (1:20 ratio, 100 g Yucca roots and 2l distilled water) to be turned into pulp. The mixture was left to stand 24 hours at 40 ^oC and then the insoluble material was separated by vacuum filtration. Thus was obtained shock solution for which freshness preservation was put into plastic bags and frozen. Working solution was made by dissolving two cubes of frozen shock solution in two liters of water.

Preparation of *Salix babylonica* extracts solution

For extract preparation were used *Salix babylonica* branches (1-2 cm thick) cut to 1-2 cm. There were used 100 g cut branches to 2 liters of cold distillate water and rest 48 hours, filtered and frozen. Working solution was made by dissolving two cubes of frozen shock solution in two liters of water.

Unused solution must be discarded.

Preparation of dried lives alfalfa extracts solution

Concentration of this solution was 1:25 (100 g of alfalfa dried leaves with 2500 ml distillated water heated to 40 ^oC for 48 hours. After 24 hours the aqueous mixture was filtered under vacuum. After filtration process to keep its freshness the extract was put into plastic bags and frozen. When using was thawed the cubes required for 100 ml aqueous extract. 100 ml of this solution was added to each of the above extracts.

Preparation of soybean extracts solution

Soybeans were ground very fine and the oil content was removed with a solution of petroleum ether and Soxhelt device. The soybean flour was dried to 30-45 minutes then dried in oven at 80 ^oC for one hour. After drying, it was prepared a solution with deionized water in ratio of 1:10. Solution was corrected to pH 7 by 2 N NaOH. To stabilize the solution this was mixed for 1 hour at room temperature. To remove insoluble material the solution was centrifuged at 6000 rot/min. Insoluble material recovered was corrected with 2 NHCl and heated in a water bath at 32 ^oC. Was added 5% enzyme solution (1 ml for each 200 ml insoluble material) and allowed to clot. The crud was separated by a screen similar to that used for cheese. Drained crud is an enzyme hydrolyzed soybean which has high content of cysteine and tryptophan in biological active form „1". For watering plants was made solutions with deionized water for ratio of 1:20.

During the vegetation period was determined number of plants/plot, growing rhythm, noting plants high, number of leaves, number of branches, number and length of internodes to 10 plants in each variant and replication. Foliar area was determined with Area Meter AM300 device. Area and leaf area were performed on three leaves of each variant and replication. The characters variability was calculated based on biometric determinations calculating arithmetic mean, standard deviation and variation coefficient. Setting the variation coefficient was done according with Ceapoiu (1968).

RESULTS AND DISCUSSION

For the first treatment plants height ranged between 32,65 cm (*Salix babylonica* extract) and 36,15 cm for the control (water). Comparatively with the control all natural extracts applied didn`t influence plants height and the differences were negative and not statistically assured. The highest difference was recorded when plants were treated with *Salix babylonica* extract (-3,45 cm). For the second treatment when was applied alfalfa extract all natural extract determined increases of plants height, the highest value (+1,60 cm) being recorded when plants were treated with *Yucca schidigera* extract. For the third treatment (enzyme extract of soybean) plants height increased with 5,60 cm when was applied Yucca+Salix extract (Table 1).

Table 1

Influence of treatment and natural extract to plants height

Treatment	Natural bioactive extract	Plants height (cm)	Diff.+Signif.
T1	water	36,15	Mt
	Yucca schidigera	34,40	-1,70
	Salix babylonica	32,65	-3,45
	Yucca+Salix	35,15	-0,95
T2	water	31,95	Mt
	Yucca schidigera	33,55	1,60
	Salix babylonica	32,85	0,90
	Yucca+Salix	32,65	0,70
T3	water	33,75	Mt
	Yucca schidigera	33,15	-0,60
	Salix babylonica	33,35	-0,40
	Yucca+Salix	39,35	5,60

DL 5% = 8,45 cm, DL 1% = 11,80 cm, DL 0,1 = 16,56 cm

For all treatments and natural extracts applied the branches number increased comparatively with the control (water), but the differences weren`t statistically assured. The highest branches number was recorded when plants were treated with enzyme extract of soybean + *Yucca schidigera* (4,55) (Table 2).

Generally, the internodes number was negatively influenced by the application of natural bioactive extracts comparatively with the control (water). Only for the second and the third treatments positive differences comparatively with the control were recorded when

were applied *Yucca schidigera*, *Salix babylonica* and Yucca+Salix extracts, but these weren`t statistically assured (Table 3).
For the first treatment all bioactive extracts determined a lower pods number comparatively with the control (water), while for the second treatment the alfalfa extract solution + natural bioactive extracts determined higher pods number, but not statistically assured. For the third treatment (soybean extract solution + natural bioactive extracts) it was observed that *Salix babylonica* extract determined a lower pods number (-1,20) comparatively with the control (water) (Table 4).

Table 2

Influence of treatment and natural extract to branches number

Treatment	Natural bioactive extract	Branches number	Diff.+Signif.
T1	water	3,10	Mt
	Yucca schidigera	3,80	0,70
	Salix babylonica	3,45	0,35
	Yucca+Salix	4,35	1,25
T2	water	2,85	Mt
	Yucca schidigera	3,95	1,10
	Salix babylonica	3,80	0,95
	Yucca+Salix	4,20	1,35
T3	water	4,10	Mt
	Yucca schidigera	4,55	0,45
	Salix babylonica	3,70	0,45
	Yucca+Salix	4,45	0,35

DL 5% = 1,56, DL 1% = 2,19, DL 0,1 = 3,10

Table 3

Influence of treatment and natural extract to internodes number

Treatment	Natural bioactive extract	Internodes number	Diff.+Signif.
T1	water	2,97	Mt
	Yucca schidigera	1,88	-1,09
	Salix babylonica	2,03	-0,94
	Yucca+Salix	1,84	-1,13
T2	water	1,99	Mt
	Yucca schidigera	2,17	0,18
	Salix babylonica	2,06	0,07
	Yucca+Salix	1,94	-0,05
T3	water	2,13	Mt
	Yucca schidigera	1,96	-0,17
	Salix babylonica	2,10	-0,03
	Yucca+Salix	2,23	0,10

DL 5% = 0,75, DL 1% = 1,04, DL 0,1 = 1,44

Table 4

Influence of treatment and natural extract to pods number

Treatment	Natural bioactive extract	Pods number	Diff.+Signif.
T1	water	5,50	Mt
	Yucca schidigera	4,35	-1,15
	Salix babylonica	4,55	-0,95
	Yucca+Salix	5,40	-0,10
T2	water	3,45	Mt
	Yucca schidigera	4,65	1,20
	Salix babylonica	5,15	1,70
	Yucca+Salix	5,65	2,20
T3	water	5,35	Mt
	Yucca schidigera	5,40	0,05
	Salix babylonica	4,15	-1,20
	Yucca+Salix	6,00	0,65

DL 5% = 2,70, DL 1% = 3,78, DL 0,1 = 5,33

The first and the second treatments influenced favorable pods length for all natural bioactive extract applied, while for the third treatment application of soybean extract solution decreased pods length for all tested extracts comparatively with the control (water) (Table 5).

Table 5

Influence of treatment and natural extract to pods length

Treatment	Natural bioactive extract	Pods length (cm)	Diff.+Signif.
T1	water	10,05	Mt
	Yucca schidigera	10,88	0,83
	Salix babylonica	10,05	0,00
	Yucca+Salix	10,44	0,39
T2	water	9,68	Mt
	Yucca schidigera	10,34	0,66
	Salix babylonica	9,90	0,22
	Yucca+Salix	10,76	1,08
T3	water	10,80	Mt
	Yucca schidigera	10,69	-0,11
	Salix babylonica	10,71	-0,09
	Yucca+Salix	10,19	-0,61

DL 5% = 1,68 cm, DL 1% = 2,35 cm, DL 0,1 = 3,31 cm

CONCLUSIONS

Generally, all studied characters weren`t significantly affected by the application of natural bioactive extracts for all treatments applied. For the third treatment (enzyme extract of soybean) plants height increased with 5,60 cm when was applied Yucca+Salix

extract. The highest branches number was recorded when plants were treated with enzyme extract of soybean + *Yucca schidigera*. For the third treatment (soybean extract solution + natural bioactive extracts) it was observed that *Salix babylonica* extract determined a lower pods number (-1,20) comparatively with the control (water). The first and the second treatments influenced favorable pods length for all natural bioactive extract applied, while for the third treatment application of soybean extract solution decreased pods length for all tested extracts comparatively with the control (water).

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RESEARCH ON RESPIRATION INTENSITY IN MECHANIZED PRODUCED TOMATO SEEDLINGS

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Key words*: respiration, seedling, tomato, seedling trays, mechanized*

ABSTRACT

To increase the culture quality it is absolutely necessary to improve the methods of obtaining the seedlings. This paper refers to the respiration processes of tomato seedlings produced in different types of seedling trays. For the development of experimental model we used seeds from five tomato hybrids: Buran, Platus, Magnus, Maximus and Shanon. The seeds were sown in Biolan peat in two types of seedling tray, with 144 cells and 160 cells. There have resulted 10 experimental variants cultivated in three repetitions. We measured the respiration intensity with a CO2 analyser and the obtained data were statistically interpreted to highlight the differences that occur between studied variants.

INTRODUCTION

The issue of container size is extremely important to both transplant producers and transplant consumers. In the latest years, there were made researches for finding new, modern solutions regarding the economic efficiency insurance, optimum seeds germination, the reduction of seeds lost, early crop, the elimination of some costs and the reduction of hand work (Uleanu et al., 2008). A trend among many commercial transplant producers is toward more cells per tray, which increases the number of plants produced, while reducing the need to develop more transplant production space (Vavrina, 1995).

Plants undergo many physiological and morphological changes in response to reduced rooting volume, which can affect transplant quality and performance. Root and shoot growth, biomass accumulation and partitioning, photosynthesis, leaf chlorophyll content, plant water relations, nutrient uptake, respiration, flowering, and yield all are affected by root restriction and container size (Nesmith & Duval, 1998).

Transplants for vegetable and crops are produced in a number of various sized containers or cells. Varying container size alters the rooting volume of the plants, which can greatly affect plant growth (Muşa et al., 2011). Container size is important to transplant producers as they seek to optimize production space. Transplant consumers are interested in

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container size as it relates to optimum post-transplant performance (Nesmith & Duval, 1998).

This paper was made as a comparative study of the tomato seedlings obtaining methods (in different type's seedling trays) concerning their recommendation into the small, medium and big farms. The purpose of this study is the prominence of the differences of the tomato seedlings obtaining methods for the protected culture.

MATERIAL AND METHODS

The researches were conducted in year 2008 and 2010 at Brăila, S.C. Petrosu S.A. For the realization of experimental model we used tomato seeds: A:

- A1-Buran - is an early hybrid with indeterminate growth, vigorous, with an open port allowing good light penetration. Fruits are large, 200-230 g, very firm and uniform, round, flattened, dark red at maturity. Highly productive well adapted to difficult growing conditions, suitable for culture in protected and field;
- A2-Platus - is the most cultivated hybrid in Europe, ideal for early crops and has a short period of ripening fruit. Fruits have a nice colour, are firm and weighing 130-140g. The plant is ventilated and requires a small workload, can be conducted on one or two arms. It can be grown all year round in greenhouses, solariums or field and is ideal for two crops per year;
- A3-Magnus - is one of the most important hybrids grown in eastern and southern Europe, due to very good resistance to disease and fruit quality. The plant is released, vegetative growth is stronger than at Platus, which allows cultivation and three branches in the autumn. Fruits have a weight of 140-160 g. Recommended for greenhouses and solariums poorly heated or unheated, and allowing two cycles of culture;
- A4-Maximus - is grown under the same conditions as Platus with a high quality of the fruit (colour, shape, firmness). Average weight of fruit is 130 g;
- A5-Shanon - is an early tomato hybrid with indeterminate growth, with large fruits (150-170 g), very uniform, suitable for growing in open fields or winter crops with a very high resistance to cold.

The seed were sown mechanized with MAS horticulture machine in Biolan peat in two types of seedling trays, B:

B1- with 160 cells (28 ml volume) ;

B2- with 144 cells (16 ml volume) (Photo 1). The seedlings were properly cared throughout their production period.

The determination of respiration intensity was made with S151 CO2 analyzer (Photo 2). It measures the concentration of CO2 in the air is passed through the assimilation chamber in which the seedlings are arranged, compared with CO2 in the air before passing through the assimilation chamber. The device comes with storage bag, hence the air that will be used to determine (to avoid influences caused by people around). Air is pumped into the unit using an electric pump, which can adjust air flow (in our case, 400 ml/min).

Photo. 1. Seeding machine (original).

Photo. 2. S151 CO2 analyzer with tomato seedlings. (original).

The experimental variants are represented each by ten plants. There have resulted 10 experimental variants cultivated in three repetitions.

RESULTS AND DISCUTIONS

The intensity of respiration was studied due to the influence of the variety and the seedling tray type. For statistical capitalization, all primary results were systematically listed in a table (Ardelean et al., 2002).

The tomato studied varieties respiration intensity ranged between 2.76 to 6.59, its highest recorded level of 6.59 being registered in case of V9. (Table 1).

Variance analysis establishes to ensure statistical differences between variants (Jităreanu, 2006). From the variance analysis for variant and error it results differences significance, established after calculating the factor F of Fisher's exact test.

Calculated F factor is greater than the theoretical F factor for p<5% (2.46) and 1% (3.60), Fisher's exact test is significant, differences go beyond errors, and researched factors contribute greatly to variation in results.

We continued the calculations for determining the error differences (differences standard deviation), limit differences for all significance thresholds (DL 5%, DL 1% and DL 0.1%) and the significance of differences between variants and experience average (Figure 1).

Table 1.

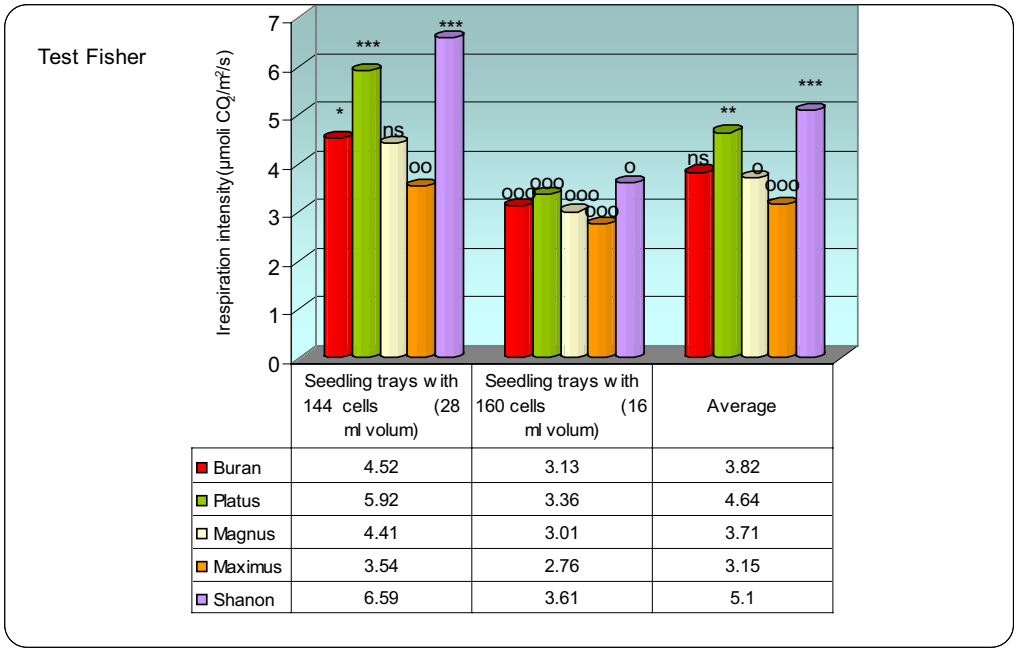
Respiration intensity average (µmol CO2/m2/s)

Variant	Respiration intensity average (µmol CO2/m2/s)
V1:A1,B1	4.52
V2:A1,B2	3.13
V3:A2,B1	5.92
V4:A2,B2	3.36
V5:A3,B1	4.41
V6:A3,B2	3.01
V7:A4,B1	3.54
V8:A4,B2	2.76
V9:A5,B1	6.59
V10:A5,B2	3.61

Table 2.

Tomato respiration intensity variance analysis for variants and error.

Variability cause	SP	GL	The variance (s^2)	Fisher's exact test	Ft	
					F 5%	F 1%
Total	45.39	29	1.565	-	-	-
Repetition	0.16	2	0.081	-	-	-
Variants	44.49	9	4.943	119.88>2.46;3.60	..46	3.60
Error	0.74	18	0.041	-	-	-



	Seedling trays with 144 cells (28 ml volum)	Seedling trays with 160 cells (16 ml volum)	Average
Buran	4.52	3.13	3.82
Platus	5.92	3.36	4.64
Magnus	4.41	3.01	3.71
Maximus	3.54	2.76	3.15
Shanon	6.59	3.61	5.1

Figure 1. The significance of differences between variants and experience mean for tomato respiration intensity.

The seedlings sown in seedling trays with 28 ml volume of cell had higher values of that process than the other with 16 ml volume. Platus presents very significant differences than the experience average for the two types of seedling trays and for the average too. Shanon presents very significant differences than the experience average for the seedling trays with 144 cells (28 ml volume).

CONCLUSIONS

The seedlings sown in seedling trays with 28 ml volume of cell had higher values of that process than the other with 16 ml volume.

The seedling tray cell volume is important for the intensity of the respiration process; the seedlings sown in seedling trays with 28 ml volume of cell had higher values of that process than the other with 16 ml volume.

Small cell restrict root growth with implications on aerial part, respiration being affected.

Platus presents very significant differences than the experience average for the two types of seedling trays and for the average too.

Shanon had the highest intensity of respiration, followed by Platus. Maximus has recorded the lowest intensity of respiration.

I recommend using seedling tray with larger alveolar cell at the expense of small ones and hybrids Platus and Shannon.

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APPLE HARVEST BASED ON THE VARIETY AND THE FRUIT THINNING METHOD

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Key words: *spindle-shaped crown, thinning fruit, variety, weight, chemical thinning.*

ABSTRACT

Investigations were carried out during 2009 - 2011 in apple orchard in SA Zubresti planted in spring 2003 with 4x2 m planting scheme trees are led by amiliorat thin spindle-shaped crown. Apple fruit thinning was studied by chemical thinning, manual and mixed in 3 varieties Golden Delicious, Idared and Florina, grafted on rootstock M26. Fruit weight was determined amount of fructela a tree as well as a unit area. The variety Idared as the variety Golden Delicious in 2009 the amount of fruit on a tree and at one hectare is higher in variant control 18.1 kg / tree respectively 22.7 t / ha. In 2010 the large amount of fruit on a tree and per hectare was recorded in variant 4 with manual thinning fruit when they were 16-18 mm in diameter after physiological fruit drop (27.6 t / ha) in 2011 has been 29.7 t / ha, followed by option 3 with 29.1 t / ha.

INTRODUCTION

The correct setting of intensive orchards of fruit load is an essential measure for obtaining stable production and quality (Babuc, 2012; Cimpoieş, 2012). This goal can be achieved through standardization and manual cleaning rod load correlated with maintenance and fructification, pruning, fertilization and irrigation (Balan ET others, 2008).

Thinning effect varies depending on climate and tree species growing conditions. (Black 1995, Sally 1991, Stopar 1999, 2001). For thinning has tested various chemicals substances such as: NAA, BA, NAD + NAA, Carbaryl. Chemical fruit thinning advantages are: Gaining a high quality production in terms of size, color, etc. increase fruit quality, and price realization default, increases labor productivity in collecting, sorting and packaging, as the number of fruits is lower; prevent breakage and split the branches, maintaining productive crown volume coming years and increased prevents alternation of fruitfulness, increase resistance to disease and frost trees due to store a sufficient reserve substances, ensuring the formation of annual shoots that formations will form fruit for years to come (Stopar 2001). At the same time the apple chemical thinning problem has been dealt with thoroughly, and the results are diverse and contradictory, which demonstrates the complexity of this topic in relation to plant requirements at different stages of vegetation (Gonda, 2003; Balan et others 2010, 2011).

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MATERIALS AND METHODS

Investigations were conducted during 2009 - 2011 in apple orchard firm "Zubresti" SA, planted with trees grafted 2 years old near the village Zubresti, Straseni. A plantation was conducted in spring 2003 varieties Golden Delicious, Florina, Idared, grafted on rootstock M 26. Distance 4x2 m planting trees

We studied 4 variants with fruit thinning:

Variant 1 - untreated control

Variant 2 - Administration chemicals when central fruit have diameter of 10-12 mm inflorescence are Bioprzerzedzacz 060 SL preparation in a concentration of 0.075%.

Variant 3 - Administration chemicals when central fruit diameter of 10-12 mm inflorescence are Bioprzerzedzacz 060 SL preparation in a concentration of 0.075% + Hand thinning of fruit.

Variant 4 - fruit thinning is performed manually by physiological fall when fruits reach 16-18 mm in diameter.

Yield was determined for each tree individually, weighing production from 24 trees and making arithmetic mean. The average weight of a fruit was determined by weighing the 100 fruit and dividing by 100.

RESULTS AND DISCUSSION

One fruit apple quality is their weight. After splashing fruit chemical thinning of fruit weight increases, so in 2009 the variety Golden Delicious fruit weight was 100 g in variant control and depending on their method of thinning fruit weight was 2 and 4 variants from 138 to 125 g

Depending on the Idared variety of biological specificity and fruit thinning method according to the research each variable, a fruit weight was within 137 g and 150 g in the control variant in variant 2 with chemical fruit thinning. In other variants fruit weight was 148 g in variant 3 with mixed fruit thinning and 149 g variant with manual thinning them.

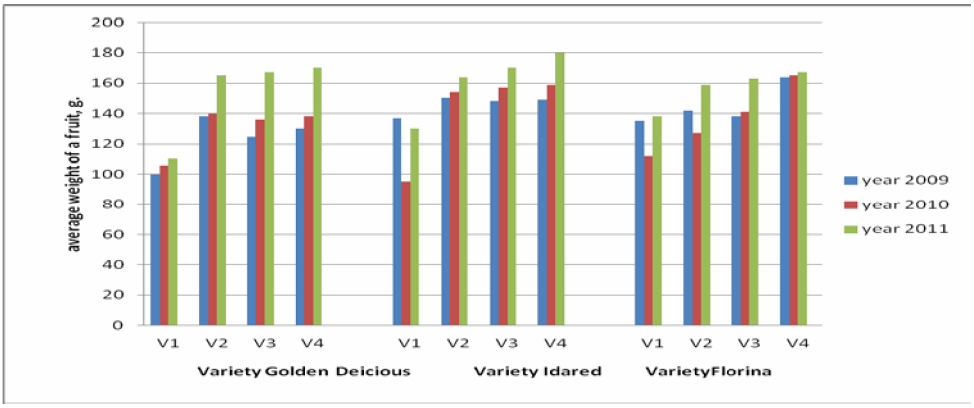

Figure1. The average weight fruits in depending of the variety and method of fruit thinning.
(M26 rootstock, planting distance 4x2 m, SA "Zubresti", 2009-2011).

In 2009 the cultivar Florina highest fruit weight were recorded in variant 4 with manual thinning fruit 164 g fruit or lowest recorded in the control variant weighing 135 g

In 2010 over 2009 fruit weight increased. The variety Golden Delicious lowest fruit weight was recorded a variant without thinning largest fruit 106 g as in 2009 occurred in variant 2 with 140g, fruit thinning in other variants recorded a weight of 136 g mixed fruit thinning variant and 138 g option with manual thinning fruit.

The variety Idared lowest fruit weight was recorded in variant control with 95 g and fruits were larger variant with manual thinning fruit 159 g chemical fruit thinning variant of a fruit weight was 154 g, and thinning in mixed-variant 157 grams.

In 2010 the cultivar Florina fruit weight ranged from 112 g to 165 g option without thinning and thinning in variant with manually fruit thinning.

In 2011 the weight of fruit is growing from year 2009 to 2010 and ranged in variant control at 110 g up to 170 g in variant 4. In the other 2 variants of a fruit average weight was 165 g and 167 g variant 2 and variant 3.

The variety Idared in 2011 the average weight of a fruit was: the biggest fruits were recorded in variant 4 180 g and the lowest as in previous years in variant control.

In 2011 the cultivar Florina in variant control fruits recorded an average weight of 138 g, which was applied to slow variant 4 manual fruit thinning fruit of a fruit average weight was 167 g.

Amount of fruit from a tree is result number of fruits and their weight.

The variety Golden Delicious amount of fruit from a tree in 2009 was from 11.2 kg in variant 4 and most fruits from a tree and at one hectare were recorded in variant control. In the years 2010 - 2011 the amount of fruit in variant control decreases, but instead increase the amount of fruit with both thinning. Otherwise in 2010 the smallest amount of fruit was recorded in variant control 16.5 kg, and with both the amount of fruit thinning fruit on a tree and as a hectare rose the highest index in variant 4 of 19 , 3 kg and 24.2 t / ha. In 2011 the amount of fruit in variant control is further decrease 16.2 kg, or 20.3 t / ha and fruit thinning variants showed indices to 23.2 t / ha in variant 2 to 27.9 t / ha in variant 3.

The variety Idared as the variety Golden Delicious in 2009 the amount of fruit on a tree and at one hectare is higher in variant control 18.1 kg/tree respectively 22.7 t/ha. In 2010 the large amount of fruit on a tree and per hectare was recorded in variant 4 with 22.1 kg /tree and 27.6 t/ha. In 2011 the highest amount of fruit also occurred in variant 4 with 23.8 kg / tree respectively 29.7 t/ha, followed by option 3 with 23.3 kg / tree and 29.1 t/ha.

The variety Florina in 2009, the largest amount of fruit from a tree was in variant control to 20.3 kg/tree, in variant 2 17.9 kg / tree. The small amount of fruit was in variant 3 with chemical thinning to perform manual as well as fruits (13.8 kg/tree) and variant 4 with manual fruit thinning 15.6 kg / tree. In 2010 the amount of fruit in variant control decreases because of too much fruit in previous annual and 17.4 kg /tree with a volume of 21.8 t/ha. The largest amount of fruit from a tree in 2010 was in variant 4 with 19.5 kg/tree and 24.4 t/ha. The other 2 variants recorded 18.6 kg /tree in variant 2 and 18.9 kg / tree in variant 3. In 2011 the amount of fruit was growing to the years 2009-2010 as both a tree and a unit area greatest amount of fruit was in variant 4 with 21.6 kg/tree and 27.0 respectively t/ha.

Table1

The fruits production in function of the different thinning method.
(M26 rootstock, planting distance 4x2 m, SA "Zubresti", 2009-2011)

Variant	Kg/tree			t/ha		
	year 2009	year 2010	year 2011	year 2009	year 2010	year 2011
Soiul Golden Delicious						
V1	19,0	16,5	16,2	23,8	20,6	20,3
V2	13,8	18,2	18,6	17,3	22,8	23,2
V3	11,3	18,7	22,3	14,1	23,4	27,9
V4	11,2	19,3	22,1	13,9	24,2	27,6
$DL_{0.5\%}$	0,98	1,33	0,75	1,55	1,84	1,78
Soiul Idared						
V1	18,1	15,4	17,8	22,7	19,3	22,2
V2	13,5	18,4	20,6	16,9	23,0	25,7
V3	12,9	21,7	23,3	16,1	27,2	29,1
V4	13,4	22,1	23,8	16,7	27,6	29,7
$DL_{0.5\%}$	1,24	1,07	0,75	1,35	1,57	3,74
Soiul Florina						
V1	20,3	17,4	19,2	25,3	21,8	24,0
V2	17,9	18,6	20,1	22,4	23,3	25,1
V3	13,8	18,9	20,7	17,3	23,6	25,9
V4	15,6	19,5	21,6	19,7	24,4	27,0
$DL_{0.5\%}$	0,79	0,83	0,78	1,67	1,41	3,59

CONCLUSIONS

The variety Idared lowest fruit weight was recorded in version control with 95 g and fruits were larger version with manual thinning fruit 159 g chemical fruit thinning version of a fruit weight was 154 g, and thinning in mixed-version 157 grams.

In the years 2010 - 2011 the amount of fruit in version control decreases, but instead increase the amount of fruits with both thinning. Otherwise in 2010 the smallest amount of fruit was in control variant 1 16.5 kg, and with both the amount of fruit thinning fruit on a tree and as a hectare rose the highest index in version 4 with respectively 19.3 kg and 24.2 t / ha.

The variety Idared as the variety Golden Delicious in 2009 the amount of fruit on a tree and at one hectare is higher in version control 18.1 kg / tree respectively 22.7 t / ha. In 2010 the large amount of fruit on a tree and per hectare was recorded in version 4 with 22.1 kg / tree and 27.6 t / ha. In 2011 the highest amount of fruit also occurred in version 4 with 23.8 kg / tree respectively 29.7 t / ha, followed by option 3 with 23.3 kg / tree and 29.1 t / ha.

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THE USE OF MANNITOL AS A RETARDANT AGENT IN THE GRAPE-VINE (*VITIS VINIFERA L.*) *IN VITRO* MULTIPLICATION

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Key words: *grape-vine germplasm, Initial G_0 material,* in vitro *maintenance*

ABSTRACT

The maintenance of the germplasm resources by using in vitro *plant cultures is considered as an alternative method for the preservation of the species with vegetative multiplication, including the grape-vine, too. The clones of Fetească neagră 6Şt and Cabernet Sauvignon 131Şt were used in the experiment. Apices and groups of 2-3 small shoots (of 0.5-1cm), were taken from the biological material regenerated* in vitro*, and were inoculated onto the environmental variants having various concentrations of mannitol in their composition (5, 10 and 20g/l, alone or in combination with 10g/l of sucrose). Mannitol was used to induce slow multiplication processes. The addition of mannitol in a concentration of 5-10g/l in combination with sucrose (10g/l), allowed maintaining the grape-vine in the* in vitro *cultures for a period of 150-180 days; the extension of the* in vitro *storage period for 6 more months, was performed by transferring them into fresh media in between two consecutive stages.*

INTRODUCTION

The conservation of the grape-vine is absolutely necessary, because there is the danger of extinction for a large number of old varieties of the global and domestic range and which can be used in the improvement process, as germplasm resources with a specifically valuable genetic potential, as well as for their reintroduction into plant culture. Generally, all the methods of *in vitro* plant conservation aim to a decrease or temporary cessation of the vital life processes, with the whole preservation of the hereditary endowment of the stored inoculums (Cachiţă, 1987, Witomska et al., 2008, Manoj et al., 2009, Akdemir et al., 2010). The main objective of this method is to maintain the biological material in a certain growth stage in order to ensure the availability of the plant material, free of pathogens, at any time and with the possibility of its immediate micro propagation.

The literature in the domain states that «additional mannitol added in the culture medium, regardless of its concentration, induces the maintenance of the cultures at an incipient stage of development. Mannitol proved to have a highly important role in the osmotic adjustment of the cells compared to other carbohydrates, thus suggesting a beneficial effect on the plant protection mechanisms against certain stress agents (lack of water, for example) » (Mitoi et al., 2009).

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The efficiency of the plant regeneration process from *in vitro* tissue cultures is the primary condition for the use of biotechnological methods in eliminating viruses and in the rapid multiplication of the grape-vine (Vişoiu et al., 2006). In this context, the conducted studies predominantly focused on the applied aspects concerning the multiplication and maintenance of valuable vine propagating material belonging to the Initial G_0 category.

MATERIAL AND METHODS

Two clones of Feteasca neagră (6Şt) and Cabernet Sauvignon (131Şt) were used for the multiplication in an *in vitro* culture. Apices and groups of 2-3 small shoots (of 0.5-1cm), were taken from the biological material regenerated *in vitro*. The explants were inoculated onto environmental variants containing various concentrations of mannitol. The common elements of the five environmental variants (M1, M2, M3, M4 and M5) were: the macro-elements, the micro-elements (M&S-1962), benzilaminopurine (BAP) - 1 mg/l, β indolilacetic acid (IAA) – 0.5 mg/l, agar 0.6% and a pH of 5.8. The varying components, sucrose and mannitol, were added alone or in combination in the culture medium. The addition of mannitol in the culture medium was at concentrations of 5 to 10-20g/l, alone or in combination with 10 g/l sucrose (the main energy support used in the multiplication in an *in vitro* culture).

The plant regeneration, multiplication and maintenance processes were conducted in growth chambers under controlled temperature (24 ± 1 ° C), light (3000-3500 lux) and photoperiodism (16 hours of light with 8 hours of darkness).

The observations and measurements were made periodically, after 1-2-6 months with maintenance of the original environment of inoculation. To highlight, as accurately as possible, the behaviour of the biological material in the environmental variants, two coefficients, which had been introduced by Lambardi et al. (1993), were considered:

a - CEM (the coefficient of explants multiplication) = (the average number of formations specific to the multiplication/initiated explant) x (% of explants which have multiplied)/100;

b - CES (the coefficient of elongation of shoots) = (the average number of shoots>1cm/ initiated explant) x (% of explants with elongated shoots)/100.

The obtained results were processed statistically using correlation and regression, and the values of the two coefficients were interpreted using the SPSS for Windows 10. From the multitude of the resulted correlations, the one chosen for illustration was that in which the differences between the M2 variant and the M3, M4, M5 variants were analyzed using One Way ANOVA - LSD, the significance being assessed at $P < 0.05$.

The biochemical determinations on the content of: dry matter (d.m.%), chlorophyll and anthocyanin pigments added to the visual observations meant to assess the quality of the biological material resulted in the cultures after a long period (of 5-6 months) of maintaining them into specific preservation media.

In order to determine the dry matter content, the plant material was dried at 105°C and it was weighed repeatedly until the constant mass was reached.

In order to determine the chlorophyll and carotenoid pigments, the biological material regenerated in culture media was used. Their extraction was performed by using 80% acetone and was followed by reading the optical density at three wavelengths: 440.5 nm, 644 nm and 662 nm. The pigment content, in mg/g of green substance, was determined by using the calculation formulas established by Holm (1954): Chlorophyll $a = 9.78 \cdot E662 - 0.99 \cdot E644$; Chlorophyll $b = 21.4 \cdot E644 - 4.65 \cdot E662$; Carotenoids $= 44.69 \cdot E440.5 - C(a+b) \cdot 0.268$.

RESULTS AND DISCUSSION

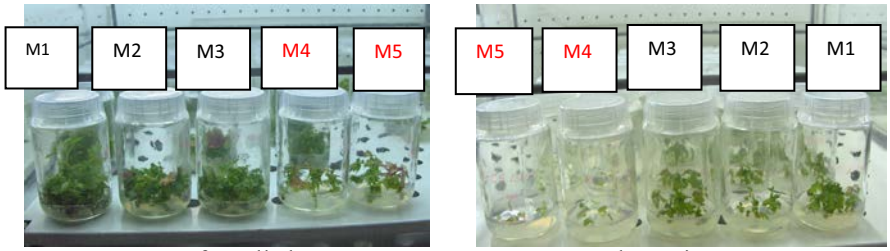
The conservation in an *in vitro* culture is considered as an alternative method for the preservation of the grape-vine germplasm resources. Maintaining the *in vitro* cultures in a certain stage of growth is meant to ensure the availability of the plant material, free of pathogens, at any time and with the possibility of its immediate micro propagation. Mannitol was used, in our studies, to induce slow multiplication processes for as long time as possible, but to maintain the viability of the explants and return to the initial regeneration potential immediately after its removal.

The observations recorded after 1-2 months of culture, showed a pronounced multiplication process, with the development of formations specific to the multiplication (adventitious buds, primary shoots and small shoots of 1.5-2cm) on the M1 medium, significantly different depending on the explants and the genotype (Figure 1).

a- apices b-groups of small shoots

Figure1. Evolution of biological material on the standard culture medium (M1)

After 1-2 months since the initiation, the environmental variants including combinations of 5-10 g/l of mannitol and 10g/l sucrose (M2 and M3) showed a proliferation close to the standard medium (20g/l sucrose). The visual observations made, after the same period of culture, on the material initiated into media with 20 and 10 g/l of mannitol (M4 and M5) showed stagnation in the explant growth, with 30% of the leaves turning to red in the small shoot groups (Figure 2).



a-groups of small shoots b – apices

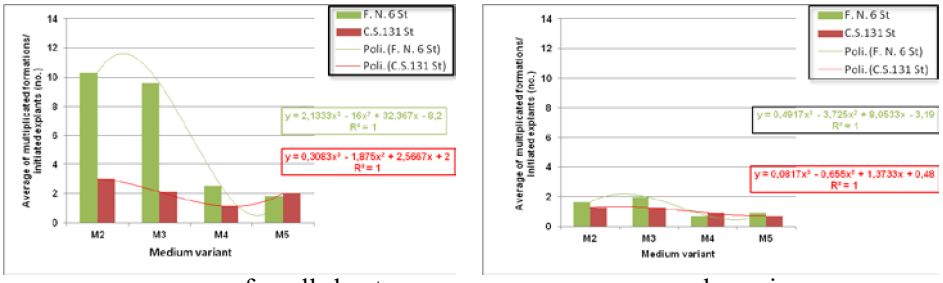
Figure 2. Evolution of explants in the medium variants, after 1-2 months (comparative analysis)

After 2 months of maintaining the multiplied biological material on the same substrate, in the standard culture medium (M1), the periodic observations showed a considerable elongation of shoots at the expense of multiplication, the appearance of the senescence phenomena (yellowing-necrosis of the basal leaves and multiple roots) and the degradation of the culture medium. In order to a further use of the biological material resulting from the multiplication of the two types of explants on the M1 culture medium,

the transfer to fresh media was necessary, after a severe selection, for both clones used in the experiment. The maximum quality and quantity were obtained from the biological material resulting from the multiplication after 30-35 days of culture, according to the *in vitro* grape-vine multiplication technology, regardless of the explant type.

As the diagram of the experiment shows, the assessments after 5-6 months were conducted only for the M2, M3, M4 and M5 variants, with the possibility to determine the quantitative and qualitative evolution of the biological material.

The results obtained after this experimental stage showed the ability to maintain a higher multiplication level (CEM=10.3 to 9.6 in Fetească neagră 6 Şt) for the culture media with combinations of 10 g/l of sucrose and 5-10 g/l of mannitol (M2 and M3). The fluctuations of the multiplication indices were mainly differentiated by the type of explant and less significantly based on the clone, during the storage period in the culture media (Figure 3).

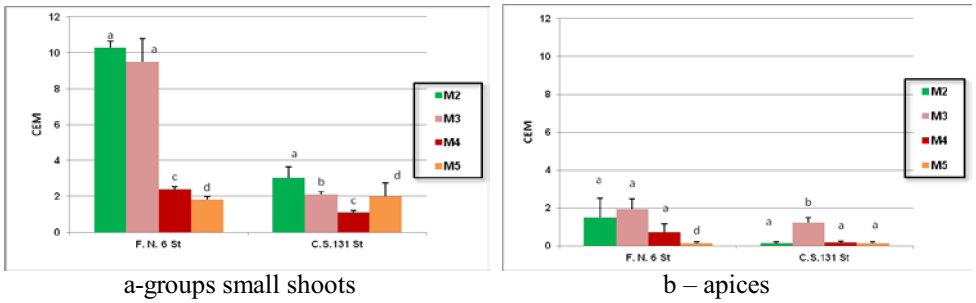


a-groups of small shoots b – apices

Figure 3. Explants reactivity after 150-180 days of maintenance in the same culture medium

The use of large explants (groups of small shoots), provided the possibility to maintain the multiplication capacity after the transfer into fresh media, too, without any addition of mannitol.

The results of the study, taken as a whole, on the assessment of the factors in multiplication and elongation of the small shoots > 1 cm, for the two types of explants, showed significant differences for the media that contain only mannitol (One Way ANOVA – LSD analysis, the significance being assessed at $P < 0.05$), compared to the M2 medium, with equal intake of sucrose and mannitol (Figure 4).



a-groups small shoots b – apices

Figure 4. Evaluation of the explant multiplication coefficient after 150-180 days of culture, without transfer into fresh media

The values are averages ± d.s. based on 3 repetitions, the bars indicate the standard deviations, the significant differences compared to M2, at $P < 0.05$.

The influence of mannitol, as a retardant agent, on the multiplication was obviously significant for apexes and especially to the clone of Cabernet Sauvignon 131 Şt.

The transfer into fresh media having the same composition, after the first stage of storage, brought to the maintenance of the plant cultures for an additional period of 4 months for the small shoot groups and of 3 months for the apexes in the M2 and M3 variants. The appearance of the senescence phenomena in the basal parts of the explants and the leaf deformation was seen during the second period of storage, especially in the M4 medium. The growth inhibiting effect, induced by mannitol in the apexes that were also transferred in the M4 and M5 media, led to 85-90% necrosis after 4 months. The selection of viable material and the immediate transfer into the specific grape-vine multiplication media (M1) resulted in the reactivation of the vegetative centres and the resumption of the regeneration processes.

The assessment of the content of dry matter accumulated in different time periods, highlights a stimulation in the vegetative mass growth, due to the clones, return to their initial regenerative potential when the biological material was transferred into a retardant free medium (Figure 5).

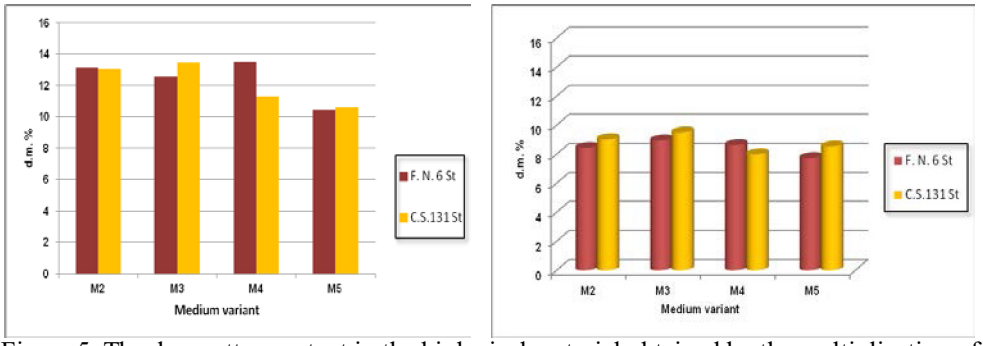


Figure 5. The dry matter content in the biological material obtained by the multiplication of small-shoot groups

The observations made on the material stored for 150-180 days showed that the appearance of deformed leaves and their turning to red to the base of the groups, to all the variants that contain mannitol and they were correlated with the anthocyanin accumulations for the M2 and M3 variants (Figure 6).

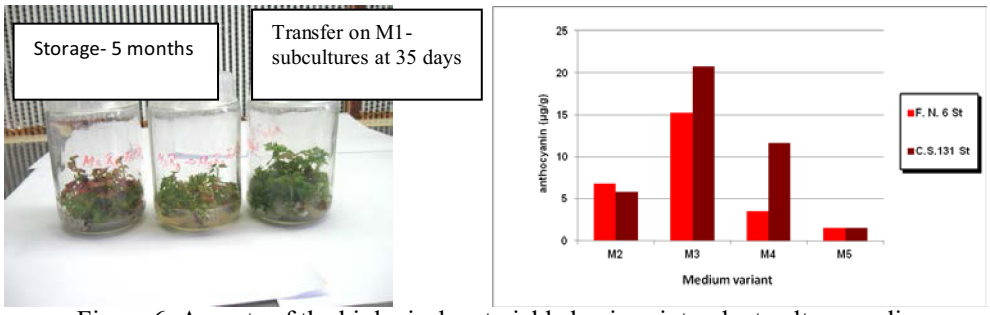


Figure 6. Aspects of the biological material behaviour into plant culture media - different time periods

The content of chlorophyll pigments (chlorophyll *a* and *b*) found in the biological material, stored for 5 months into the same medium, but also in the one transferred into mannitol free media (after 35 days of growth), was differentiated by genotype; accumulations were recorded after the return to the regenerative potential following the transfer into the standard medium, immediately after the first month of plant culture.

CONCLUSIONS

One of the methods used to maintain the *Initial* G_0 grape-vine biological material in an *in vitro* plant culture, supposes the use of some substances having a retardant role into the plant culture medium which slow down the multiplication on a medium-term (1-6 months) and a long-term (6-12 months), depending on the combination with other substances or their concentration;

The addition of mannitol in a concentration of 5-10 g/l in combination with sucrose (10 g/l), enabled the maintenance *in vitro* of the grape-vine cultures for a period of 150-180 days; the extension of the *in vitro* storage period, for other 6 months, was achieved by the transfer into fresh media in between two consecutive stages;

The presence of mannitol alone in the culture medium, in the amount of 10 - 20g/l led to: the inhibition of the regeneration processes and elongation of the shoots, the appearance of the senescence phenomena and the leaf deformation after 150-180 days; the return to their multiplication capacity and the decrease of the degrading phenomena in the biological material, were achieved by the periodic transfer (35 days) into standard multiplication media;

The quantitative and qualitative parameter assessment, performed periodically, showed the significant influence of the genotype and explant type, regardless of the culture medium that was used.

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IMPROVEMENT OF DOUGH RHEOLOGY OF DIFFERENT QUALITY WHEAT FLOURS THROUGH THE ADDITION OF BACTERIAL PROTEASE

Vizitiu Daniel[1], Danciu Ioan[2]

Key words: *wheat flour, Mixolab, Alveograph, rheology, bacterial protease*

ABSTRACT

Any argument regarding biscuits production should start with the wheat flours, which are the primary raw material for biscuits. Within the same type of flour, the flour quality may vary within very large limits, for various reasons. The effects of this phenomenon have an impact on the quality of biscuits. A natural and effective way is the addition of proteases. Our paper's purpose was the optimization of the different wheat flour's quality with the help of bacterial proteases using some of its beneficial properties. We have observed the effects of addition of bacterial proteases in six flours of the same type but different quality, on dough rheological properties. The results showed that using different quantities of bacterial proteases, depending on flour quality, it is possible to improve the quality of the flour for the production of biscuits.

INTRODUCTION

Romanian flours are characterized by a very good protein content which frequently exceeds 13% and presents a deformation index of gluten predominantly having lower values 5 mm. Very often, in practice, it is difficult to find suitable wheat flours with optimum gluten properties. The quantity and quality of the gluten have an important influence on the elasticity of the dough. That is why low protein wheat flours with weak gluten are usually chosen for biscuits production.

The processing of strong flours, with short gluten, creates problems due to the high resistant proteic network formed in dough. Whether a flour with low and weak protein is available or not, the use of elasticity-reducing agents will have benefits in all stages of the process. To improve the strong flours performance, proteases are very useful in the production of biscuits flours because proteases catalyze the cleavage of the polypeptidic chains (Stoica, 2007) and they are also recommended in literature. Small amounts of fungal proteases may be added to strong flours in order to reduce mixing times and improve dough extensibility. To enable the efficient, economical manufacture of quality products it is necessary to control the plasticity of the dough by modifying the quality of the gluten. Therefore, it is necessary to supplement dough with additives which may decrease the high resistance of gluten. For this purpose, a series of chemical agents is currently used in bread-

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making, mainly L-cysteine which, through the reduction of gluten proteins disulfide bonds, decreases the dough tenacity and elasticity. Similar results can be reached using proteases of different origins, although their mechanism of action in the dough is different compared to L-cysteine (Stauffer, 1990; Bordei, 2004). We chose for improving the quality of flours for biscuits production, bacterial proteases because they are faster than the fungal proteases but a little easier to control than bromelain or L-cysteine (Moodie, 2001). Rheological characteristics are important for the milling industry from the point of view of the prediction of the dough processing parameters and the end products quality (Jirsa et al., 2007). The different empirical methods are currently used to obtain data on rheological properties of flour (Dobraszczyk & Morgenstern, 2003). In order to characterize the quality wheat flours we selected one laboratory device to perform the rheological tests, Alveograph was use to analyze the rheological properties during biaxial stretching of a dough piece. The objective of this study was to describe the improving effect of bacterial proteases on the rheological properties of six different quality flours in order to optimize their quality and achieve constant quality flour for biscuits.

MATERIAL AND METHODS

The flours (T-550) were obtained from 6 samples of common wheat from 2011 crop. The wheat samples of wheat were milled on industrial roller mill with a capacity of 120t/24h. The flours were sampled according to the standard SR EN ISO 24333:2010. Chemical characteristics of wheat flours (Table 1.) indicated a wide variation in the quality characteristics.

Table1

Chemical characteristics of wheat flours

White flour type 550	1	2	3	4	5	6
Moisture, %	14,2	15	15	14,9	14,7	14,2
Ash content, %	0,454	0,46	0,51	0,53	0,504	0,47
Gluten index, %	94,7	97,5	90,3	94,7	90,4	96,8
Falling number, sec	272	367	341	382	362	372
Wet gluten, %	24	30,8	25,4	26,8	26,3	27,4

Bacterial protease (provided by Mühlenchemie GmbH & Co.KG) derived from *Bacillus subtilis*. The enzyme reduces the protein strength, relaxation times can be shortened or omitted without risking deformation by shirnkage, it is used in biscuits and cracker production. The activity of enzyme preparation is 100 u/g (casein hydrolysis, pH 8.0). Optimum activity of the enzyme is on pH 6-8 and optimum temperature at 40-60 ^{0}C.

The physical-chemical characteristics of the flours were evaluated as follows: the moisture content through the SR ISO 712:2010*; the ash content through the SR ISO 2171:2007*; the wet gluten content through the SR ISO 21415-1:2007*; the gluten index through the AACC Method 38-12 method (Sistem Glutomatic 2200)**; the falling number through the SR ISO 3093:2007* (Falling Number, model 1305). Rheological properties of wheat flour were determined with Alveograph according SR ISO 27971/2009*, analyzed factors were: P – the maximum over pressure needed to blow the dough bubble, expreses dough resistance, , P/L – alveograph ratio, W – the deformation energy, I_e – elasticity index.

RESULTS AND DISCUSSIONS

Some of the quality parameters required and recomended by literature and biscuits production units are presented in tabel 2. Part of these parameters may vary depending on the particularities of the production lines.

Table 2

Quality parameters of biscuits flour

Minerals, % dry. matt	0,55
Proteins (Nx5.7), %	7,5-9
Wet gluten, %	22-25
W – Deformation energy ,10^{-4} J	130-170
P/L – alveograph ratio	0,3-0,5
Stability, min	3-6
Soaking , U.F.	60-120
Falling number, s	>250

Depending on the quality parameters of witness flours we found the following dosages (Fig. 1, 2, 3, 4) for those six samples of flour, to achieve constant quality flour for biscuits.

As we can see in table 1 and table 2 the quality and the quantity of the gluten is much higher for biscuits flour requirements. In the following we present the analysis of protease effect on the rheological properties of dough compared to those of the whitness sample. The Chopin Alveograph provides information about extensional properties of dough by stretching the dough in two opposite directions (Dobraszczyk & Morgenstern, 2003). Overpressure (P), is considered an indicator of dough resistance to deformation and this indicator could be used as an index of dough stability (Amos, 1949). In fig. 1it can be observed the effect of protease to reduce the strength of the dough more obvious for sample number 3 where the amount of protease was higher compared to sample number 1 where the amount of the protease was smaller. The degree of the parameter is a function of the quantity of the added enzime. This is explainable because proteases reduce viscosity and P is correlated directly with the viscozity of the dough (Scott Blair and Potel, 1937).

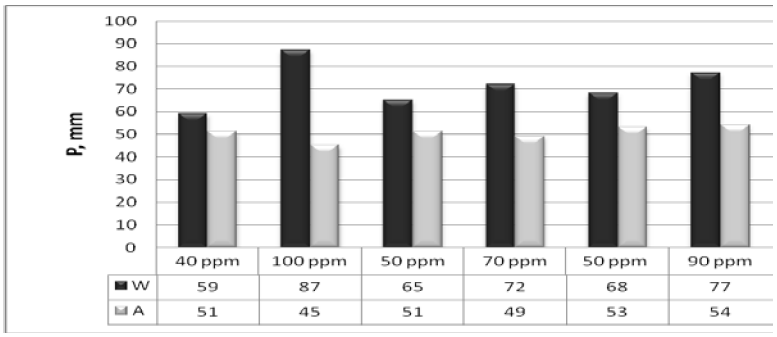
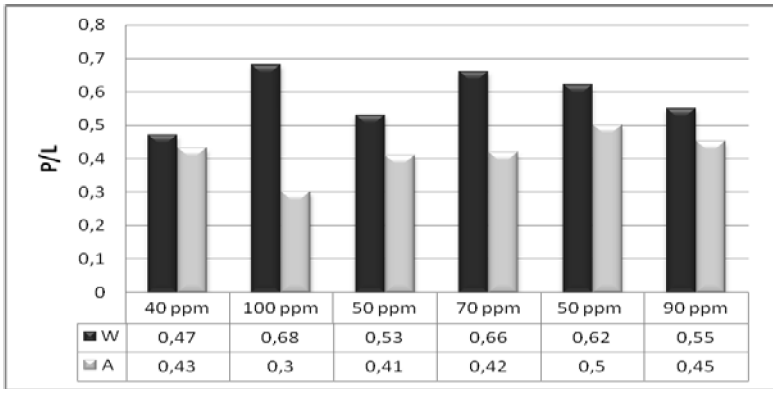


Figure 1. The influence of the protease addition on P (overpressure).

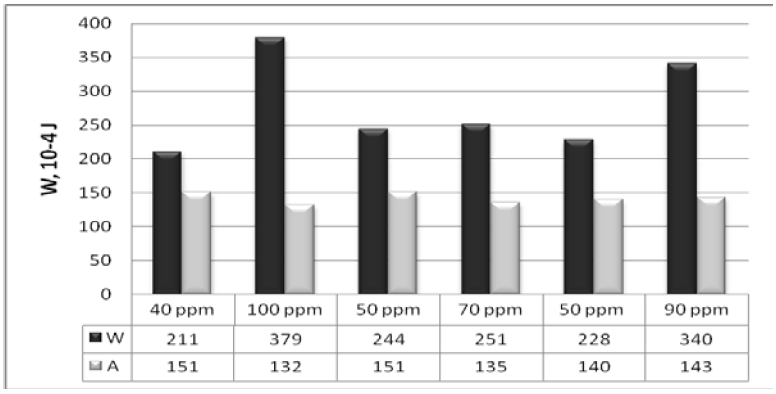
The configuration ratio (P/L) is the ratio of dough tenacity and dough extensibility (Chopin, 1962). Because the bacterial proteases affect the tenacity of the dough and very little the average abscisa to the rupture (L), the configuration ratio decrease is due to decreased of the dough toughness.



	40 ppm	100 ppm	50 ppm	70 ppm	50 ppm	90 ppm
W	0,47	0,68	0,53	0,66	0,62	0,55
A	0,43	0,3	0,41	0,42	0,5	0,45

Figure 2. The influence of the protease addition on configuration ratio P/L

The deformation energy (W) represents the energy needed to inflate the dough until it ruptures. The W value is considered to be closely related to the flour strength, and many users of the alveograph rely on this value in predicting the processing behaviour of the flour being evaluated. Because it is closely related to the quality of the gluten the degree of W value under the action of bacterial proteases is normal and as it can be seen in fig. 3, the decrease is dependent of the quantity of added enzyme.



	40 ppm	100 ppm	50 ppm	70 ppm	50 ppm	90 ppm
W	211	379	244	251	228	340
A	151	132	151	135	140	143

Figure 3. The influence of the protease addition on deformation energy, W

The elasticity index represents the elastic resistance of the dough and indicates the pressure inside the bubble. Since the amount of air inside the bubble is constant for each test, this pressure is directly linked to the bubble volume (Kitissou 1995). As well as P the elasticity index is dependent on the quantity of added protease. The elasticity of the gluten is decrease under the effect of protease and the relaxation of the gluten occurs.

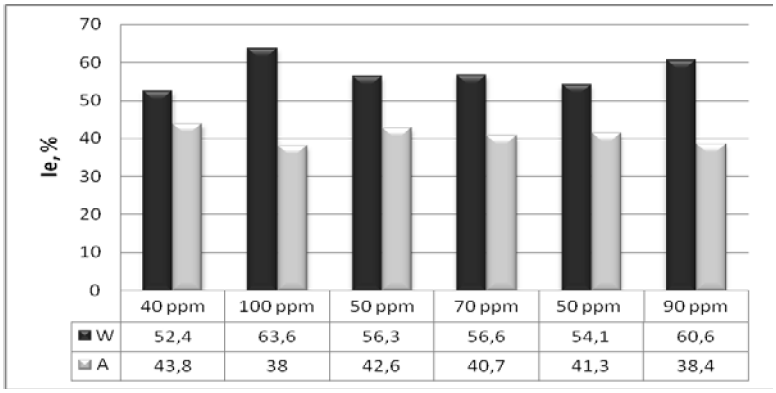


Figure 4. The influence of the protease addition on elasticity index, Ie

CONCLUSIONS

Because within the same type of flour, the flour quality may vary within very large limits quality of raw materials is an essential requirement for achieving good quality finished products. It is well known the effect of protease on dough properties where elasticity of the gluten is undesirable but extensibility prerequisite for proper processing of dough in the production of biscuits.The results showed that using different quantities of bacterial proteases, depending on flour quality, it is possible to improve the quality and also to optimize the quality differences between flours and achieve constant quality flour for the production of biscuits.

ACKNOWLEDGEMENTS

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RHEOLOGICAL EVALUATION OF DRY AND TEMPERED WHEAT ON LABORATORY SCALE, USING MIXOLAB DEVICE

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Key words: *wheat flour, Mixolab, rheology, Chopin CD1, milling*

ABSTRACT

Rheological testing is very effective to evaluate the breadmaking potential of wheat flours and an essential requirement for achieving high quality finished products. The wheat must be milled for testing; the added water before milling the wheat, influence the relevancy of wheat tests. The Chopin CD1 test mill was used to produce flour for rheological analyses in order to establish which of two methods is more reliable to give information about the wheat flour quality provided by industrial milling. The rheological behaviour of the wheat flours obtained in laboratory were compared with those of flours obtained in industrial mill using Mixolab device. For most of the parameters, significant correlations were established between the rheological evaluations of industrial and laboratory flour. The Pearson correlation coefficient varied between 0.62 and 0.94 for test ran on Mixolab device.

INTRODUCTION

Quality of raw materials is an essential requirement for achieving good quality finished products. Because wheat characteristics have an impact on the extraction of flour (Posner, 1988) and on the rheological quality of the flour obtained, millers are concerned with the quality of wheat and want to have it evaluated for quality potential before purchasing.

To generate appropriate qualitative evaluation of wheat, the wheat sample must be conditioned before milling. Conditioning before milling is a stage of great importance for miller from technical, flour-quality, and economic points of view (Posner and Hibbs 2005). Tempering is the process of adding water to wheat before milling to toughen the bran and mellow the endosperm of the kernel and thus improve the efficiency of flour extraction (Kweon et al., 2009). Adding water to the different parts exaggerates their differences and their behavior in reaction to the forces exerted upon them during milling (Bradbury et al., 1960). Among the cereal technologists, rheology is widely recognized as a valuable tool in quality assessment of

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flour. Numerous instruments have been devised to obtain objective data on dough properties in order to predict its behavior in the bakery. To investigate the rheological changes that occur in the gluten visco-elastic network during mixing and thermo-mechanical behavior of the dough we used Mixolab.The Mixolab technique allows the complete characterization of the flours in terms of proteins' quality by determining their water absorption, stability, elasticity and weakening properties; starch behaviour during gelatinization and retrogradation and enzymatic activity of the proteases, amylases (Banu et al. 2011). Because tempering is a time consuming step, most of the mills laboratory don't use tempering before milling their wheat samples for obtaining flour for laboratory tests, for that reason the objective of this study was to compare the rheological properties of wheat flours obtained by grinding tempered wheat and dry wheat on Chopin CD1 test mill in order to predict the rheological properties of industrial flours. For this purpose the rheological parameters of flours obtained by grinding ten different common wheats on Chopin CD1 test mill were compared with the parameters of industrial flour.

MATERIAL AND METHODS

White flour was obtained from 10 samples of common wheat from 2011 crop. The wheat samples were milled in an industrial roller mill with a capacity of 120t/24h, to an extraction rate of 79%, white flour (0.55% ash content) and black flour (1.25% ash content). Chemical characteristics of wheat samples (Table 1.) indicated a wide variation in the quality characteristics.

Table 1.

Chemical characteristics of wheat samples

Parameters	1	2	3	4	5	6	7	8	9	10
Umidity, %	11,8	12	10,9	11,5	11,3	11,5	11,3	12	12	13,2
Ash, % s.u.	1,6	1,6	1,24	1,57	1,55	1,58	1,71	1,59	1,38	1,47
Wet gluten, %	21,7	21,9	28,4	23,2	20,1	24,1	18,9	24	25	21
Gluten index,%	83,6	90,7	86,8	81	88,5	69,6	75	60,8	79,4	90,6
Falling number, sec	370	218	476	370	149	394	297	390	414	272

The obtained wheat mill streams consisted of 4 break flours (B_{1-2}, B_3, B_4 and B_5), 14 reduction flour fraction (C_{1a}-1, C_{1a}-2, C_{1b}, C_{2-3}-1, C_{2-3}-2, C_4, C_5-1, C_5-2, C_7-1, C_7-2, C_9-1, C_9-2, C_{10}-1, C_{10}-2) 1 break reduced fraction (Div). For white flour (0.55% ash content) the mill streams were (B_3, B_5, Div, C_{1a}-1, C_{1a}-2, C_{1b}, C_{2-3}-1, C_5-1, C_{10}-1). The flours were sampled according to the standard SR EN ISO 24333:2010. The flour for laboratory tests was obtained by grinding dry wheat and tempered wheat on Chopin CD1 test mill, according to the standard SR EN ISO 27971: 2009*. The physical-chemical characteristics of the wheat and flours were evaluated as follows: the moisture content through the SR ISO 712:2010*; the ash content through the SR ISO 2171:2007*; the wet gluten content through the SR ISO 21415-1:2007*; the gluten index through the AACC Method 38-12** method (Sistem Glutomatic 2200); the falling number through the SR ISO 3093:2007* (Falling Number, model 1305). Rheological

properties of wheat flour were determined with Mixolab using standard Chopin^{+} protocol and studied parameters were:

- ✓ C1 (Nm – maximum torque during mixing;
- ✓ C2 (Nm) – measures the protein weakening based on the mechanical work and temperature;
- ✓ C3 (Nm) – expresses the starch gelatinisation;
- ✓ C4 (Nm) – indicates the stability of the starch gel formed;
- ✓ C5 (Nm) – measures the starch retrogradation during the cooling stage.

RESULTS AND DISCUSSIONS

Mixolab is used to characterize the mechanical changes due to mixing and heating simulating the mechanical work as well as the thermal conditions that might be expected during the baking process (Rosell et al. 2007). The parameters obtained from the Mixolab curve and, also, the correlation of laboratory mills flours with industrial flours are presented in Table 3. The first part of Mixolab curve measures the characteristics of dough during mixing and kneading. The rheological changes, which occur in gluten structure during mixing, greatly determine the final product quality (Dobraszczyk and Morgenstern, 2003). Smith and Andrews (1957) showed that the water-soluble constituents of wheat flour have an appreciable effect upon the mixing characteristics of the flour. This first step of the test is a reflection of three basic processes: absorbtion of water, dough development and dough breakdown (Preston and Kilborn, 1984) At the beginning the mixolab curve is a reflection of the absorbtion of water by the flour components.

Water-absorption gives an indication about the potential of protein molecules to absorb the added water, and therefore is an indicator of baking quality. For the water absorption we obtained a high Pearson correlation coefficient for both methods but for tempered wheat the correlation with industrial flour was better.

Dough development time depends on the water absorption speed of flour constituents to form a smooth and homogenous appeareance, it reflects the time between the first addition of water and the time when the dough seems to have optimum elastic and viscous properties for the retention of gas. In this case the Pearson correlation coefficient was very good for tempered wheat and almost perfect for dry wheat.

Table 3.

Mixolab parameters of wheat flours

ixolab	Industrial flour		Chopin CD1 dry wheat		correlation coefficient	Chopin CD1 tempered wheat		correlation coefficient
	min	max	min	max	Pearson	min	max	Pearson
Water absorbtion, %	51,3	57	50,9	58,9	0,75	50,6	58,6	0,79
Development time,min	1,32	4,43	1,32	3,75	0,94	1,27	8,77	0,72
Stability, min	5,67	10,98	4,98	10,35	0,63	7,15	11,05	0,57
C2, Nm	0,37	0,53	0,28	0,47	0,67	0,36	0.55	0,75
C3, Nm	1,85	2,05	1,55	1,99	0,65	1,73	2.23	0,71
C4, Nm	1,34	1,85	0,89	1,8	0,76	1,08	2.1	0,85
C5, Nm	2,05	2,87	1,33	2,85	0,62	1,58	3.23	0,75

Dough stability is related to the period of time during which the dough is able to resist to the mechanical action of mixer kneading arms and to keep its optimum elastic and viscous properties for the gas retention. Stability is an indication of the flour's tolerance to mixing and stronger flours tend to be more stable (Miralbés, 2004). High correlations with industrial flour were obtained for both methods used for this study. A better value for Pearson correlation coefficient we obtained for dry wheat.

After the dough's stability period a decrease in dough consistency occurs as a result of the temperature increase. Actually, the temperature increase destabilises and unfolds the proteins, which become hydrophobic (Rosell et al. 2007). These changes in fibrillar structure of the gluten proteins under mechanical shear stress and the action of enzymes (protease and α-amylase) on the dough components involves the release of a large quantity of water and a decrease in torque until a minimum value C2. At the minimum torque (C2), the dough reaches the specific temperature for the beginning of starch gelatinisation. In the case of the flours obtained by grinding dry wheat the values for the minimum torque (C2) were smaller than that obtained for the flours obtained by grinding tempered wheat and suggests a better protein quality for flours obtained by grinding tempered wheat. A very high correlation with industrial flour was achieved for protein weakening parameter obtained by grinding tempered wheat. For dry wheat the Pearson correlation coefficient indicated a major correlation with industrial flour but smaller than that obtained for tempered wheat when protein weakening due to mechanical and thermal constraints was analysed.

As the temperature increases, the role of the proteins goes to a secondary place, whereas the changes in the starch granules are the responsible of further torque variations. The dough heating coupled with the water released by the thermally denaturated proteins causes the starch gelatinisation. The starch granules swelling and hydration induce the dough consistency increase.This process is stopped when the mechanic shear forces and temperature lead to the physical division of the granules (Rosell et al. 2007). The maximum consistency of the dough was higher for the flours obtained by grinding tempered wheat compared with the consistency of the flours obtained by grinding dry wheat because of the decrease of the α-amylases activity (Banu et al. 2011). The maximum consistency of the dough had a high Pearson correlation coefficient for dry wheat and for tempered wheat a very high correlation for the dough viscosity during heating with industrial flour was registered.

The mechanic shear forces and temperature lead to the physical division of the granules and a reduction in viscosity is observed, dough's consistence decreases as the flour has an increased α-amylase activity. So, lower consistency (C4) was obtained for flours obtained by grinding dry wheat compared with de flours obtained by grinding tempered wheat (Table 3). For flours obtained by grinding tempered wheat we obtained a very high Pearson correlation coefficient and also for the flours obtained by grinding tempered wheat.

During the period of cooling the starch gel, the decrease in the temperature causes an increase in the consistency of dough. This stage is related to the retrogradation of starch molecules and the final torque C5 correspond to the end of the starch retrogradation period and higher values were recorded for flours obtained by grinding tempered wheat compared with de flours obtained by grinding dry wheat, because of the lower α-amylase activity. (Table 3). For the torque after cooling stage the Pearson correlation coefficient recorded for flours obtained

by grinding dry wheat indicated a major correlation with industrial flours. After the baking process the cooling of the crumb occurs and for the final torque the Pearson correlation coefficient for tempered wheat was higher.

CONCLUSIONS

Because water-soluble fraction of wheat flours is the principal factor responsible for determining the mixing requirements of wheat flour (Mattern and Sandstedt, 1957), for the characteristics of dough during mixing and kneading tested with Mixolab we found better correlations with industrial flours for the flours obtained by grinding dry wheat perhaps of the negative influence of added water to the flours obtained by grinding tempered wheat. The changes of the Mixolab curve after the increase of the temperature depeded on the value of the falling number and the flours obtained by grinding tempered wheat were better correlated with industrial flours. Taking into account of the results obtained we may conclude that the flour obtained by grinding tempered wheat is more reliable to give information about the wheat flour quality provided by industrial milling.

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THE INFLUENCE OF TECHNOLOGICAL, BIOLOGICAL AND BIOCHEMICAL FACTORS OVER THE CONTENT OF ANTHOCYANS IN THE MERLOT WINE OBTAINED ON STARMINA VINEYARD CENTRE

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Keywords: *anthocyans, vinification, biological and biochemical factors, maceration, optical density units.*

ABSTRACT

The study of the quality and productivity characteristics and also of efficiency of wine production is the premise of defining the oenological potential, being essential elements in assessing the possibilities of obtaining wines appropriate to current requirements. Particular attention should be paid to the development of phenolic compounds, those conferring specificity to red wines, with substantial importance on the organoleptic characteristics. Anthocyanin extraction speed, selectivity of the extraction process and its effectiveness depend on many factors, pectolytic enzymes having a decisive role in this respect due to the formed tannin-anthocyans complexes, essential to stabilize wine color. Addition of selected yeasts that take on possession fermentation environment from the beginning enables the oenologist to achieve and direct fermentation to obtain wines depending on desired goals.

INTRODUCTION

The use of macerating enzymes has become a very common practice. The use of maceration enzymes led to wines that are richer in anthocyanin compounds and show better chromatic characteristics (Moreno-Perez A. et al., 2010). Macerating enzymes may help in phenolic extraction and, at the same time, may modify the stability, taste and structure of red wines, because it is not only the anthocyanins that are released from skins, but also tannins bound to the cell walls (Bautista-Ortin A.B. et al., 2007).

The grape skin cell wall is a limiting barrier that prevents the release of polyphenols into the must during fermentation. Polyphenol extraction obviously requires the middle lamella walls to be degraded in order to release the cells, and that cell walls be broken to allow their contents to be extracted or diffused into the wine (Amrani Joutei K. and Glories Y., 1995).

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The sensory perception of wine is complex and involves the interaction of both volatile and non-volatile components (Robinson A.L. et.al., 2011).

Environmental variables are considered the most influential factors on grapevine production and berry composition (Montes C. et al., 2012). The existing components of the physical environment (e.g. climate, soil properties, topography and geology) can lead to the characteristic expression in different grapevine cultivars, resulting in temporal and spatial variations in wine quality and typicality (Winkler et al. 1974, Gladstones J.S., 2004).

The influence of climate on wine quality is well known, through the effect of both regional and local-scale climatic conditions during the growing season, and by its interannual variability, which generates variations in grapevine growth and then in berry composition (Jones G.V.et al. 2005, Soar C.J. et al. 2008).

MATERIAL AND METHODS

The present study was made starting from an experience through which the dynamics of anthocyanin extraction from Merlot pulp was watched in the wineyard center Starmina, Mehedinti county. To achieve the objective, the most important factors of primary wine obtaining process able to determine the reflection of the true potential of the grapes variety and of the area were taken into account and experienced, thus concluding that wines should not be produced by chance. Thus in the experimental model adopted has been included biological and biochemical factors (native and selected yeasts; pectolytic enzymes) and different maceration times (0-96 hours) - as variable factors and carbohydrate content, acidity, anthocyanins, SO_2, temperature of fermentation-maceration process and pulp phase mixing regime - as invariable factors within varieties and variants involving yeasts, enzymes and fermentation-maceration duration.

Wine anthocyanins were determined through spectrophotometry, by pH difference, knowing that the difference between the optical densities read at 520 nm of a solution at two different pH values is proportional to the amount of anthocyans in wine sample (method Ribera Gayon - Stonestreet - 1968).

For the determination of anthocyans monomers and polymers was used the spectrophotometric method over solutions consisting of 1 ml wine and HCl 1N, diluted 1:100 and 1 ml acetic acid, the same dilution at OD 520 nm and OD 320 nm.

RESULTS AND DISCUSSION

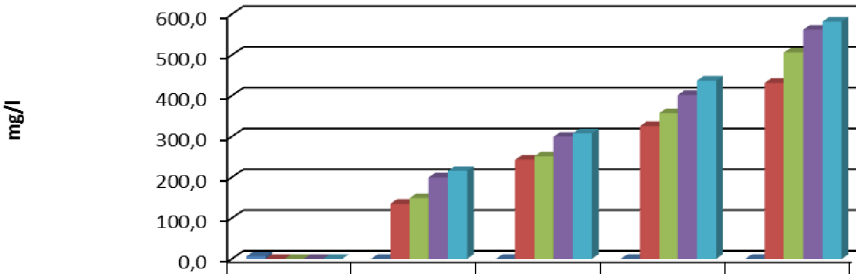
The main physico-chemical characteristics of Merlot wine obtained through fermentation-maceration under the action of indigenous and selected yeasts as singular factors or in combination with pectolytic enzymes on different periods of contact between pulp phases and compared with the white vinification version are presented in Table 1.

Table 1.

The main physico-chemical parameters of red Merlot wines from Starmina-Orevita based on biological and biochemical factors of primary wine and duration of maceration

Variants	Maceration durations hours	Alcohol % vol.	Total acidity g/l H_2SO_4	Volatile acidity g/l H_2SO_4	Glycerol g/l	Unreductible extract g/l	Ash g/l	Tartaric acid g/l
White vinification (LI)	0	12,68	4,11	0,40	9,8	22,9	2,08	2,43
Classical maceration (LI)	24	12,56	4,12	0,44	9,9	23,6	2,18	2,51
	48	12,50	4,09	0,46	9,8	24,9	2,38	2,62
	72	12,55	4,10	0,46	9,9	25,5	2,45	2,44
	96	12,45	4,12	0,47	9,9	26,7	2,58	2,55
Maceration with indigenous yeasts + pecto-lytic enzymes (LI+EP)	24	12,71	4,13	0,43	10,1	24,5	2,30	2,30
	48	12,70	4,10	0,45	9,9	25,1	2,40	2,44
	72	12,66	4,09	0,45	9,8	26,3	2,50	2,61
	96	12,59	4,05	0,46	10,1	27,4	2,68	2,51
Selected yeast fermentation-maceration (LSA)	24	12,88	4,08	0,34	10,1	25,2	2,40	2,45
	48	12,80	4,09	0,36	10,0	26,3	2,53	2,53
	72	12,79	4,11	0,39	10,3	27,4	2,70	2,49
	96	12,81	4,10	0,40	10,4	28,6	2,82	2,66
Fermentation-maceration with LSA +EP	24	12,82	4,06	0,32	10,4	26,3	2,51	2,71
	48	12,83	4,03	0,41	10,8	27,1	2,66	2,55
	72	12,79	4,10	0,43	10,9	28,3	2,81	2,72
	96	12,84	4,10	0,45	10,9	28,9	2,90	2,66

Depending on the biotechnological variants used for primary wine obtaining process and on the maceration-fermentation length, the anthocyans content of the obtained Merlot wine is presented in Figure 1.



	0	24	48	72	96
Vinified in white (LI)	6,0	0	0	0	0
Maceration-fermentation with LI	0	135,0	243,4	325,6	431,6
LI + pectolytic enzymes	0	148,9	251,6	357,0	506,1
Fermentation-maceration with LSA	0	199,7	299,6	401,6	562,0
Fermentation-maceration: LSA+EP	0	215,6	307,7	437,0	582,1

Maceration times - hours

Fig. 1. The total of anthocyans content in wines

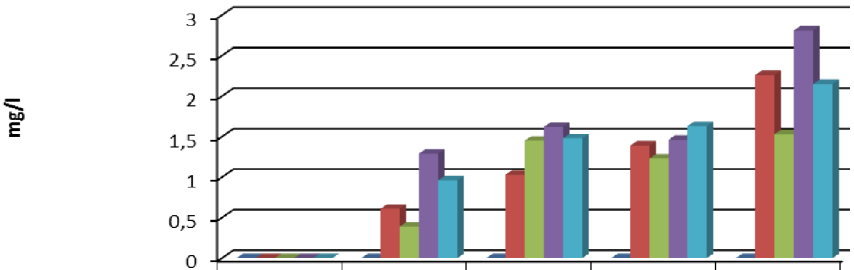
It can be seen that Merlot wines obtained through white vinification of grapes incorporate small amounts of anthocyans (6 mg / l) and can imprint a "stained" character to the product. Another aspect observed is that concerning easier release of colouring matter from the antocyanoplasts from the Merlot grapes epidermis.

The anthocyans content evolves into two directions: depending on the biotechnological factors of the primary wine obtaining process in the framework of the four variants with biological and biochemical interventions and depending on the length of contact between the phases of the pulp.

In the first 24 hours of contact between the phases of the pulp, anthocyanins extraction rate determines the difference between the biotechnological variants and the rate of accumulation of the colouring matter in wine.

The contents in monomers anthocyans determined at 60-65 days after wine obtaining vary depending on the biotechnological interventions from the primary wine obtaining process and the length of fermentation-maceration process (Fig. 2).

It can be set a specific order for all biotechnological variants, depending on the duration of the contact between the phases of the pulp subject to fermentation-maceration process. For the same age of wines, the monomer anthocyanins ratio increases in direct relation with the duration of the maceration-fermentation length of the primary wine obtaining process. For example: for 96 hours duration of fermentation-maceration on variant LSA + Ep ratio is 2.15.

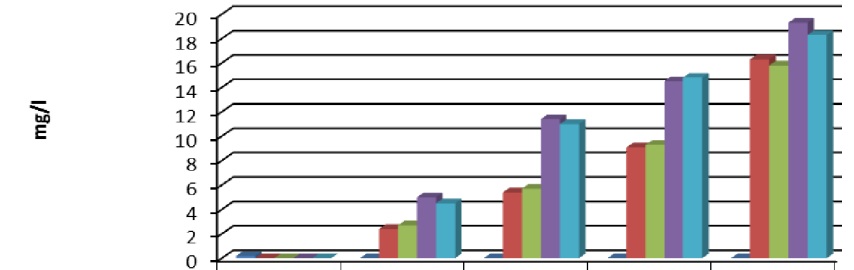


	0	24	48	72	96
Vinified in white (LI)	0	0	0	0	0
Conventional maceration (LI)	0	0,61	1,03	1,39	2,26
LI + pectolytic enzymes	0	0,39	1,45	1,23	1,53
Fermentation-maceration: LSA	0	1,29	1,62	1,46	2,81
Fermentation-maceration: LSA+EP	0	0,96	1,48	1,63	2,15

Maceration times - hours

Fig. 2. Content of monomers anthocyans in wines

The content of the polymers anthocyans (Fig. 3) shows an ascending trend proportional with the increasing of the maceration duration.



	0	24	48	72	96
Vinified in white (LI)	0,19	0	0	0	0
Conventional maceration (LI)	0	2,4	5,4	9,1	16,3
LI + pectolytic enzymes	0	2,7	5,7	9,3	15,8
Fermentation-maceration: LSA	0	5,0	11,4	14,5	19,3
Fermentation-maceration: LSA+EP	0	4,5	11,0	14,8	18,3

Maceration times - hours

Fig. 3. Content of polymers anthocyans in wines

In relation with the type of yeast and the presence of pectolytic enzymes on the composition of biotechnological variants as well as in the case of monomers, a certain order is not possible, since after growth, there will be a decrease and vice versa. Thus, in the case of a maceration for 72 hours, the contents of polymers anthocyans range from 9.1 (on the variant exposed to fermentation with indigenous yeasts - LI) and 14.8 (on the version LSA + Ep).

CONCLUSIONS

As it is known, the specific character of red wines is conferred by the presence of anthocyanins, along with other polyphenols. To define the colouring potential of red wines, together with the anthocyanin content, the quality of the anthocyanin complex has a considerable importance. Using the same raw material and the same technological method of grapes processing, the phenolic composition varies depending on the biotechnological factors involved in the primary wine obtaining process as follows:

- for the same length of the maceration-fermentation process, the largest proportions of anthocyanins are extracted under the combined action of selected yeasts and pectolytic enzymes. When biological and biochemical factors are used, anthocyanin contents increase with the duration of the maceration-fermentation process;
- the contents of monomers anthocyanins and polymers anthocyanin, at well defined moments during the storage time, present proportions that are higher when the duration of the fermentation-maceration process is more important;
- generally, for the same duration of contact between the pulp phases, the contents of the above-mentioned chromatic compounds are higher with the variants in which LSA and LSA + Ep acted.

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RESEARCH ON THE SUITABILITY OF SOME LOCAL VARIETIES AND BIOTYPES OF GRAPEVINE FOR ORNAMENTAL VITICULTURE

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Keywords: *varieties and biotypes, ornamental viticulture, arch, half an arch*

ABSTRACT

The research was performed in 2011 on private plantations, yards and gardens in Buziaș – Silagiu, where the plants were in various stages of their multiannual biologic cycle. The aim of our research was to identify and turn to profit the productive potential of some local varieties and biotypes cultivated in Banat area, especially in Buziaș-Silagiu area, as compared with the control varieties; depending on their growth vigour, they are suitable for pruning into ornamental shapes.

The research methods we used were in accordance with our objectives: observations, visual examinations, measurements, weighing, determinations, and eventually data calculation, centralization and interpretation. During our field trips, we identified certain local varieties and biotypes in people's yards and gardens, which, besides having good productive potential, grow vigorously, thus being suitable for pruning into high shapes (arbours, trellises, gazebos).

After completing our research we concluded that, due to their rusticity and high vigour, local varieties and biotypes require few treatments, and that is the reason why we recommend them for use in landscaping projects

INTRODUCTION

In the period when all plants were wild, grapevine was an element of the spontaneous flora, undergoing an evolution process brought about exclusively by natural selection. Man's interference consisted only in fruit gathering and removing the trees that cast shade over the ones that supported the grapevine. (Jukovski P., 1950, quoted by Oprea A.and Adriana Indrieș, 2000, Pop Nastasia, 2010).

Traditional Romanian varieties constitute valuable genetic heritage. They can represent an important source of variability and germplasm in the process of grapevine improvement. For wine growers, they can be an important source of income, by yielding good quality wine products, which are at the same time typical and authentic, bearing the imprint of their place of origin.

In viticulture areas in Banat, as in all viticulture regions in Romania, there are many old local varieties and biotypes, the potential of which has not been explored to full extent and which are suitable for ornamental viticulture.

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For the cultivation of grapevine near the house, people should choose varieties with mixed function (table/wine). The grapes should be medium-size and semi-crunchy, with a pleasant, sour-sweet taste. These vines provide table grapes for the family's consumption and the house wine is made of whatever quantity of grapes is left.

During our field trips, we identified certain local varieties and biotypes in people's yards and gardens, which, besides having good productive potential, grow vigorously, thus being suitable for pruning into high shapes (arbours, trellises, gazebos).

In most households we visited, we found arrangements in which grapevine was the central piece (varieties and biotypes existing in the reference areas), pruned as arches, semi-arches, but also on arbours and espaliers.

MATERIAL AND METHOD

The research was performed in 2011 on private plantations, yards and gardens in the region Buziaş – Silagiu, where the plants were in various stages of their multiannual biologic cycle. The aim of our research was to identify and turn to profit the productive potential of some local varieties and biotypes cultivated in Banat area, especially in Buziaş-Silagiu area, as compared with the control varieties; depending on their growth vigour, they are suitable for pruning into ornamental shapes.

During our field trips, we identified and sampled over 40 varieties from the plantations in localities Buziaş and Silagiu. Of these, 26 local varieties and biotypes were considered interesting and were kept under study. In order to establish the name of the local varieties and cultivars, we used several criteria: local name, if there is one, the name of the locality where they were discovered, predominant ampelographic and technological attributes, etc.

The research methods we used were in accordance with our objectives: observations, visual examinations, measurements, weighing, determinations, and eventually data calculation, centralization and interpretation.

RESULTS

In the case when the grapevine is high-trained, the most often used type of pruning is Lenz Moser.

Formative pruning is similar to the Cazenave type pruning, (in canes, with small shoots and spurs), the difference being that the trunk will be projected to 1-1.2 m.

Pruning for fruit production is made the same way as Cazenave pruning, leaving on the cordons the fruiting shoots formed of fruit bearing nodes (4-6 nodes) and replacement buds, placed very close to the cordon. On high shapes, where the trunk is 1-1.2 m high, we can apply cane pruning (stems of 10-12-14 buds), and spur pruning, as in Sylvoz pruning, rational Pergola etc.

In the case of horizontal cordon pruning (spur pruning, Cazenave pruning, Lenz-Moser pruning), we presented ways of forming bilateral cordons, but trunks can also be formed with only one cordon; in this case, the length of the cordon will be equal to the distance between two neighbouring trunks (1 - 1.20 m). Still, the presence of bilateral cordons diminishes the risk of declining health of the cordons and facilitates their restoration

Training grapevine on espaliers

In some private plantations, we found various forms of training grapevine on spaliers- fig 1.

Figure. 1. Training grapevine on espaliers

Training vine in the "halanga" type

This type of pruning implies the formation of one or more trunks from which cordons are trained to the eaves of the house, vertically at the beginning, and then obliquely. During the first years after plantation, the trunk is propped up, until the multiannual wood can support itself, and the vegetation is supported by wire netting, where the wire is Ø= 2.2 – 2.8 mm. In the places where the espalier has to hold up cordons with multiannual wood, thicker wire will be used; and where the annual cordons and stems are placed, people will use thinner wire- fig 2.

Figure. 2. Training vine in the "halanga" type

Training the vine in semi-arch shape

For this type of vine training, the upper part of the metallic poles which, together with the wire, are the support for the vine, is arched. Semi-arches are a decorative element near fences, walls, etc. In this case, the cordons are trained similarly to those pruned over arches.

Training the vine over an arch

The arch is the most widely used artistic way of grapevine training in the households under research. More often than not, the house is linked to the street by an asphalt alley or driveway which is covered by a grapevine archway, approximately 3.5 - 4 m wide and 2.5 - 3m high.

The arbour is made up of arches of various shapes, made from iron pipes, fixed in the ground by concrete in order to gain more stability. The arches are covered in green paint for aesthetic purposes, namely to look good in the periods when the cordons are left without leaves. In the lateral parts of the arch, wires are fastened, 40 to 80 cm apart, on which the grapevine is trained – fig 3.

Figure.3.Training the vine over an arch

Training grapevine in the shape of a garland

Garland-shaped grapevine seems to be an appropriate system along alleys, between lampposts. Two stocks are planted near each lamppost; under the lamp, there is a horizontal wire (or a chain, which will be curved), fixed between two poles, that supports the weight of the vine. The shoots will hang from the sides of the cordon, looking aesthetically pleasing.

Building a gazebo shaded by grapevine

The gazebo is a wooden construction of an octagonal shape. Inside, there are a round table with benches and chairs. The grapevine is planted on the outside of the gazebo, at a distance of 0.75-1.25m. – fig 4

The entrance to the gazebo is through one of the sides. The vegetation and fruit are supported by the horizontal wires fixed on the wooden frame of the gazebo.

In family gardens, such constructions are as pleasant as they are useful, the cosy atmosphere inside prompting enjoyable discussions among family members or friends

Figure.4. Building a gazebo shaded by grapevine

Training grapevine in the shape of a pergola

Pergola is a practical and decorative way of training all varieties of table grapes in family gardens. The frame for the **simple pergola** consists of vertical poles 2.4 m tall. On the interval between the rows, every pole has a 1.2 m long arm fixed by a wire at an angle of 50^0. The arm is installed 1.5 m above the ground and it is at its base that the first wire of the support system is placed; this wire has to be thicker, because it has to support the cordons. Other three wires are fixed on the arm, 40 cm away one from the other; their role is to support the vegetation. The trunk is supported vertically by a pole; the stocks are planted at a distance of 1.5 m from each other, and on the cordons the fruiting elements are the stems with 5-7 fruiting buds- figure 5

Figure. 5. Training grapevine in the shape of a pergola

CONCLUSIONS

In regards to the identification of ornamental methods of grapevine training, we can conclude that the following were the most widely used:

The archway made of grapevine was identified in almost all households in the area under research, at the entrance to the house or yard. Of the local varieties and biotypes identified in this type of training, the following were most widely used: Coarnă albă, Coarnă neagră, Cornă vânătă, Răşchirată albă and Ochiul boului;

The gazebo represents an artistic element that we found in many family gardens. Generally, the local varieties and biotypes used as elements of this types of construction were represented by the mixed types Mărcovață and Coada oii, as well as the wine grapes Arămiu.;

The garland made of grapevine was another artistic element found in some yards, along alleys or driveways, between lamppoles or simply between wooden poles used as decorative elements.

Most local varieties and biotypes of table grapes that we identified were trained (for practical and decorative purposes) in the shape of a **pergola** in family gardens.

Training grapevine on **espaliers** was very common in the gardens in the area where we made our research. Many of the local varieties and biotypes are suitable for this type of training, but the ones which we found especially suitable for that are: Auriu of Silagiu, Alb aromat of Silagiu; in their cases, their total growth was greater than that of the control variants.

"Halânga" type was only identified in two households. It was made from the local varieties and biotypes of table and mixed grapes characterized by vigorous growth.

Due to their rusticity and high vigour, local varieties and biotypes require few treatments, and that why we recommend them for use in landscaping projects.

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OLTEAN NEW TABLE GRAPE VARIETIES CREATED AT THE UNIVERSITY OF CRAIOVA

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Key words: *grapevine, descriptors, productivity, variety*

ABSTRACT

This paper presents research results on the assessment of the biological, agrobiological and technological potential of the new table grape variety Oltean created at the University of Craiova.

In order to assess the bioproductive and qualitative potential of the new variety under study, several specific observations were made in accordance with the O.I.V. and I.C.V.V. methods: the morphological descriptors (young shoot, ampelometry: leaf, bunch, berry), the phenology, agrobiological descriptors (the proportion of fertile shoots, the absolute and relative fertility indexes, the relative and absolute productivity index), and the quantity and quality of grape production.

INTRODUCTION

The range diversification and improvement of table grape variety conveyer through the creation of new varities that are better adjusted to the environmental conditions in our country, with various maturation stages, superior in terms of productive and qualitative characteristics and more resistant to unfavourable environmental factors, as well as to main diseases and pests, represent key objectives for viticultural betterment and research. (Dejeu L.C., 2010; Sestraş et al. 2004; This et al. 2006). At the same time, the promotion of a new grape variety calls for a good cognition of their bioproductive and quality characteristics and their environmental exigency with view to establishing the possibility of expansion in various viticultural areas. (Cichi Daniela & Costea D.C., 2008; Damian et al., 2011).

MATERIAL AND METHOD

Research activities are focused on the results obtained during 2008-2011in the competition comparative cultivars at Banu Maracine winemaking centre centre, which is situated in the southernmost section of the Getic Plateau between 44°19' northern latitiude

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and 23°48' eastern longitude, about 6 km away from the city of Craiova at an average altitude comparable to the sea level of 176-190 m (Giugea &.Olteanu 2001).

Oltean varieties resulted from the controled pollination of *Dattier de Saint Vallier X Victoria genitors.* They were obtained at the University of Craiova by a team of authors including Olteanu I., Cichi Daniela Doloris, Giugea N., Popa Camelia, Costea D.C. and were homologated by ISTIS in 2011.

Kober 5 BB was used as mother plant, the plantation distances were 2 x 1.2 meters, which meant 4,166 rootstocks / hectar, by using unprotected cultivation systems with semi-tall training, double Guyot pruning and 14 buds long canes.

Observations and determinations were focused on : morphological descriptors, agrobiological descriptors (proportion of fertile shoots, the absolute and relative fertility coefficients, relative and absolute productivity index) and technological descriptors in accordance with O.I.V. and I.C.V.V.

RESULTS AND DISCUSSIONS

Climatic resources studied by comparison with multiannual average values and with the demands of table grapes, as well as the climatic conditions imposed by the area cultivation of table grape varieties in Romania show that Banu Maracine winemaking centre belongs to the type of areas favourable for the cultivation of such varieties.

Considering Huglin's heliothermal index values, the Banu Mărăcine viticultural centre fits the category of *warm temperate climate* (IH4), with no heliothermal restrictions on the maturation of grape varieties from the entire variety conveyer of our country (Cichi Daniela Doloris et al. 2011).

In terms of morphological characteristics (table1), Oltean variety shows flowers with fully developed stamens and fully developed gynoecium. Leaf is three-lobed, average sized with circular kidney shape and open petiole sinus with U-shaped base and V-shaped superior lateral sinuses (table 1, figure 1 and figure2). Fruit cluster has conic shape, bunch medium density, medium-long length (peduncle excluded), with average weight of 545 g (table 2). Berry is ovoid, with average weight of 6.61 g and green yellow colour of skin covered with pruina of average size, no particularity of flavor, with normally developed seeds (figure 2).

In what concerns the agrobiological characteristics, the fertility of shoots is average, fertility coefficients having average values of 0.79 for relative coefficient (c.f.r.) and 1.38 for absolute coefficient (c.f.a.), with yet high productivity, being set-off by the average weight of fruit clusters, average productivity indexes being 430 in case of I.p.r. and 683 pentru I.p.a. (table 2). Their vigour is average-high with yet well maturated wood and good resistance to cold, downy mildew and grey rot.

Under the conditions of Banu Maracine winemaking centre gemmation starts from the second decade of April, blooming from the second decade of June while ripening begins in the last decade of August.

Oltean varities reach their full maturation in the second half of September providing high production per rootstock of 5.78 kg / vine in average, out of which over 83% finished products. In terms of quality, Oltean accumulates in average 164 g/l of sugars in the context of a total average acidity of 4.89 g/l H_2SO_4.

Grapes have long storage duration per rootstock and good resistance during transport.

Figure 1. Oltean variety- Rootstock appearance

Figure 2. Oltean variety – details of leaf, fruit cluster, grape berry

Table 1

Sinthesis of basic ampelographic descriptors in Oltean varieties

N° code OIV/UPOV/ Bioversity	Group of characteristics	Characteristic	Notes
001/3/6.1.1	Young shoot: opening of the shoot tip	Half open	3
065/19/6.1.21	Mature leaf: size of blade	Short-medium	3-5
068/ 23/ 6.1.23	Mature leaf: number of lobes	Three	2
069	Mature leaf: colour of the upper side of blade	Medium green	5
079 / 26 /6.1.30	Mature leaf: degree of opening / overlapping of petiole sinus	Open	3
080	Mature leaf: shape of base of petiole sinus	U-shaped	1
083-1	Mature leaf: shape of the base of upper lateral sinuses	V-shaped	3
082	Mature leaf: degree of opening / overlapping of upper lateral sinuses	Open	1
151/ 18 / 6.2.1	Flower: sexual organs	Fully developed stamens and fully developed gynoecium	3
223 / 40/ 6.2.6	Berry: shape	Ovoid/Obtuse-ovoid	6-7
225/ 41/ 6.2.8	Berry: colour of skin	Green yellow	1
226	Berry: uniformity of colour of skin	Uniform	2
227	Berry: bloom	Medium	5
231/ 44/ 6.2.9	Berry: intensity of the anthocyanin coloration of flesh	None	1
236/ 47/ 6.2.12	Berry: particularity of flavour	None	1
241/ 48/ 6.2.7	Berry: formation of seeds	Complete	3
303/ 35/ 7.1.4	Time of beginning of berry ripening (veraison)	Late	7

Table 2

The agrobiological and technological descriptors of Oltean variety grapes in winegrowing centre Banu Mărăcine (2008-2011)

Group of characteristics	Descriptors		
	Proportion of fertile shoots (%) Min. – Max. Average	The fertility coefficients Min. – Max. Average	
		Absolute (c.f.a.)	Relative (c.f.r.)
Fertility potential	55 – 81 67	1.00 – 1.66 1.38	0.54 – 1.00 0.79
Productivity index	Relative productivity (I.p.r.) Min. – Max. Average	Absolute productivity (I.p.a.) Min. – Max. Average	
	295 – 540 430	545 – 778 683	
Production	Production (kg/vine) Min. – Max. Average	Berry (g) Min. – Max. Average	
	4,35 – 6,32 5,78	4.69 – 8.60 6,61	
Quality of production	Sugar content of must (g/l) Min. – Max. Average	Total acidity of must (g/l H_2SO_4) Min. – Max. Average	
	147 – 176 164	3.43– 5.78 4.89	

CONCLUSIONS

Oltean varieties are distinguished by superior quality characteristics, such as big grape clusters and berries, pleasant commercial aspect, balanced taste, high productivity and finished products, high resistance to diseases and pests. They are suitable for cultivation expansion in Southern areas with sufficient heliothermal resources.

It is required the continuation of the research to establish the most convenient technological measures providing their bio-productive, quality and economic efficiency.

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