

ISSN-L=1454-7376  
(Print)-ISSN 1454-7376  
(Online)=ISSN 2069-8275  
(CD-ROM) = ISSN 2069 – 847X

UNIVERSITATEA DE ȘTIINȚE AGRICOLE  
ȘI MEDICINĂ VETERINARĂ  
“ION IONESCU DE LA BRAD” DIN IAȘI



**LUCRĂRI ȘTIINȚIFICE**

**Vol. 59**

**NR. 2**

**SERIA HORTICULTURĂ**

EDITURA “ION IONESCU DE LA BRAD”



**IAȘI 2016**



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ISSN-L=1454-7376  
(Print)-ISSN 1454-7376  
(Online)=ISSN 2069-8275  
(CD-ROM) = ISSN 2069 – 847X

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ISSN-L=1454-7376  
(Print)-ISSN 1454-7376  
(Online)=ISSN 2069-8275  
(CD-ROM) = ISSN 2069 – 847X

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## YIELD, SEED QUALITY AND RESIDUAL BIOMASS CHEMICAL COMPOSITION OF ORGANIC FABA BEAN AS AFFECTED BY FARMING SYSTEM AND PLANTING TIME

### CANTITATEA, CALITATEA SEMINTELOR ȘI COMPOZIȚIA CHIMICĂ A BIOMASEI REZIDUALE DE BOB ECOLOGIC OBTINUT ÎN FUNCȚIE DE SISTEMUL DE CULTURĂ ȘI EPOCA DE SEMĂNAT

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**Abstract.** *The effects of two farming systems (open field, greenhouse) and two transplanting times (11 October, 8 November) on plant growth, fresh pods yield, seed quality, and residual biomass quality for fuel production were assessed. The research was carried out in the years 2011-13 on faba bean in southern Italy. Crop dry weight, LAI and pods yield were higher in open field than in greenhouse and upon the 8 November transplant. Seeds to whole pod weight, yield and mean weight were better affected by the later planting time and by greenhouse, though farming system had no significant effect on yield. Seed fiber and proteins content attained higher values in greenhouse than in open field, whereas polyphenols and ascorbic acid showed the opposite trend; the later planting time better affected proteins and ascorbic acid, but polyphenols were more concentrated in earlier crops seeds. Residual biomass obtained in greenhouse and in earlier crops showed higher values of lignin and pectin, but lower cellulose, compared to open field.*

**Key words:** *Vicia faba* L., open field, greenhouse, plant growth, fiber, proteins, polyphenols, ascorbic acid.

**Rezumat.** *În studiul de față au fost evaluate efectele a două sisteme agricole (câmp deschis, seră) și două epoci de înființare (11 X; 8 XI) privind creșterea plantelor, producția de păstăi, calitatea semințelor, precum și calitatea biomasei reziduale pentru producerea de combustibil la bobul de grădină. Cercetarea a fost realizată pe perioada 2011-2013, pe bobul de grădină, din sudul Italiei. Greutate uscată, suprafața foliară și producția de păstăi au fost mai mari în câmp deschis comparativ cu sera pentru epoca de semănat 8 noiembrie. Greutatea semințelor din total greutate păstăi, producția de păstăi și greutatea medie a păstăilor au fost influențate de epoca de înființare mai târzie și locul de cultură (seră), deși sistemul de cultură nu a avut nici un efect semnificativ asupra producției. Conținutul total de fibre și proteine au atins valori mai mari în seră decât în câmp deschis, în timp ce polifenolii și acidul*

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*ascorbic, au arătat o tendință opusă; epoca de înființare mai târzie afectează conținutul de proteine și acidul ascorbic, dar în schimb polifenolii se concentrează în semințele obținute de la culturile timpurii. Biomasa reziduală obținută în seră și în culturile timpurii au arătat valori mai mari de lignină și pectină, celuloză mai redusă, în comparație cu câmpul deschis.*

**Cuvinte cheie:** *Vicia faba* L., câmp deschis, seră, creșterea plantelor, fibre, proteine, polifenoli, acid ascorbic

## INTRODUCTION

Faba bean (*Vicia faba* L.) is cultivated in many world areas, mainly located in China, Ethiopia, Mediterranean basin, central-northern Europe, Australia and South America, its surface extension accounting for 240,000 ha worldwide (FAOSTAT, 2014). Faba bean features fit a sustainable agriculture model (Nadal *et al.*, 2003), since this species benefits from symbiosis with *Rhizobium* bacterium to fix nitrogen from the atmosphere.

*Vicia faba* is mostly grown in the field, but greenhouse environmental conditions may be suitable to organic vegetables, which are more susceptible to the environmental unbalances caused by a less intensive management (Maynard, 1994). Planting time also affects crop performances and, in this respect, Khalil *et al.* (2010) reported that in Pakistan the highest yield is favored by early October sowing, compared to late summer or mid-autumn planting.

*Vicia faba* seeds represent a remarkable energy source and they are rich in fiber, proteins, mineral nutrients, vitamins and antioxidants (Crépon *et al.*, 2010). Notably, the high fiber concentration is beneficial for intestinal functions regulation as well as for blood glucose and cholesterol control (Macarulla *et al.*, 2001) and polyphenols act as antioxidants, also protecting the plants against ultraviolet radiations (Jansen *et al.*, 2001).

With the prospect of valorizing the whole crop system, the exploitation of residual lignocellulosic biomass for fuel production offers the advantage to use an added-value by-product (Schievano *et al.*, 2009).

In this research, we assessed the effects of farming system and planting time on crop growth, fresh pods and seeds yield, seeds quality, and residual biomass chemical composition.

## MATERIAL AND METHOD

**Plant material and growth conditions.** Research was carried out on faba bean (*Vicia faba* L. major Hartz) cultivar Agudulce supersimonia, grown under organic management in Naples, southern Italy, in 2011-2013 on a sandy-loam soil (87% sand, 11% silt, 2% organic matter). The highest mean temperature values were recorded in the first ten days of June (22.3 °C and 25.3 °C in open field and in greenhouse respectively) and the lowest in the first ten days of February (7.0 °C and 9.5 °C in open field and in greenhouse respectively).

Comparisons were made among four experimental treatments, obtained by the factorial combination of two farming systems (open field, greenhouse) and two planting times (11 October, 8 November). A split plot design with three replicates was

arranged, where each elementary plot was 6.20 m<sup>2</sup>; plants were transplanted in single rows spaced by 0.85 m from each other and the spacing was of 0.13 m along the rows, with an areal density of 9 pt·m<sup>-2</sup>.

Organic farming practices were performed in compliance with the EU regulation 834/2007. Each year the plants were supplied with 90 kg·ha<sup>-1</sup> of N, 75 kg·ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> and 200 kg·ha<sup>-1</sup> of K<sub>2</sub>O. A 30% fertilizers dose was given just before transplanting and the remaining 70% on dressing, by using the Bioilsa 6-5-13 manure. Drip irrigation was activated when the soil available water capacity (AWC) decreased to 80%.

Fruit harvests were performed from mid-February to early June, as an average of the two research years.

**General analytical methods.** Plant samples were randomly selected to assess the maximum leaf surface extension using a bench top LI-COR leaf area meter. Harvests of fresh pods were performed coinciding with maximum seed growth and determinations were made in each plot for: weight and number of undamaged pods classified as marketable; mean fruit weight on 30 unit samples; seeds weight on random samples including 20 pods. Cumulative plant biomass was calculated as the sum of the above ground plant biomass at the end of each crop cycle plus the total fruit production from the beginning of the harvest period. Dry weight was assessed after dehydration of the fresh samples in an oven at 70°C under vacuum until they reached constant weight. After harvest, residual biomass samples were randomly collected in each plot and transferred immediately to the laboratory, where they were dried in an oven at 70°C under vacuum. The samples were, then, carefully milled and the final material, composed of particles ≤ 1 mm diameter, was used for chemical analyses.

**Seed quality analyses.** Faba bean pods were randomly sampled in each plot in early April, then the seeds were extracted from the pods and the following laboratory determinations were performed.

Soluble solids, expressed as Brix at 20°C, were assessed using a Bellingham and Stanley digital refractometer, model RFM 81.

Fiber and proteins were determined using a chemometric method, applying near infrared spectroscopy (NIR) and principal component regression method (PCR). Spectra between 4000-10000 cm<sup>-1</sup> were run in reflectance on a Perkin Elmer Frontier infrared spectrophotometer equipped with a NIRA accessory, acquired in Spectrum program and chemometrically processed by Quant+ software, both from Perkin Elmer.

The total polyphenols determination were performed with Folin Ciocalteu reagent (Singleton and Rossi, 1965), and determined in a Lambda 25 UV-Vis (Perkin Elmer) spectrophotometer at 760 nm, using gallic acid as standard.

Ascorbic acid was assessed according to Kampfelgen *et al.* (1995).

Lignin, cellulose, crystalline cellulose, hemicellulose and pectin in the crop residual biomass were assessed as previously described (Ercolano *et al.*, 2015).

Data statistical processing was performed by analysis of variance and mean separations were performed through the Duncan multiple range test, with reference to 0.05 probability level, using the SPSS software version 21. Data expressed as percentage were subjected to angular transformation before processing.

## RESULTS AND DISCUSSIONS

**Plant growth and yield.** There were no significant differences between years in the experiment, and only the effects of farming system and planting time are reported.

The highest leaf area index (LAI) was recorded at full fructification stage and it was not significantly affected by farming system; moreover, LAI showed

higher value under the transplant performed on 11 October compared to the 8 November one (tab. 1). Different trends were recorded for plant dry matter, which attained higher value both with the later planting time and in open field conditions. In previous research (Confalone *et al.*, 2010), faba bean grown in open field in north-western Spain showed a similar trend in LAI to that observed in the present work.

Table 1

## Growth indexes and yield results of faba bean

Treatment	LAI (m <sup>2</sup> ·m <sup>-2</sup> )	Dry weight (g·m <sup>-2</sup> )	Crop cycle duration (days)	Pods Yield (t·ha <sup>-1</sup> )	Number (per plant)	Mean weight (g)	Seeds Weight/ pod (%)	Yield (t·ha <sup>-1</sup> )	Mean weight (g)
Farming system									
Open field	4.4	1257.6	144.5	15.7	6.8	22.6	26.3	4.1	1.44
Greenhouse	4.3	1087.1	130.3	13.6	6.0	21.8	29.1	4.0	1.67
	n.s.	*	*	*	*	n.s.	*	n.s.	*
Planting time									
11 October	4.5	1041.3	133.0	11.2	5.4	20.5	26.0	2.9	1.42
8 November	4.1	1303.4	141.8	18.1	7.4	23.9	29.4	5.2	1.69
	*	*	*	*	*	*	*	*	*

n.s. no statistically significant difference; \* significant difference at  $p \leq 0.05$ .

As shown in table 1, crop cycles were shorter both in greenhouse, by about two weeks compared to open field, and under the earlier transplant by about nine days. Pods yield attained a higher value in open field conditions than in protected environment and with the later planting time. These results were connected to the pods number per plant, which showed the same trends as those recorded for yield. The pod mean weight contributed to the production outcome only referring to the planting time effect, whereas no significant differences arose with regard to farming system. Both the ratio between seeds to pod weight and the mean seed weight were higher under protected environment than in open field and upon the later transplant. The latter better affected the seeds yield as well, but this variable did not change between the two farming system conditions.

Marcellos and Constable (1986) reported that sowing performed in early autumn in Mediterranean environments prolongs the crop cycle compared to later planting time, resulting in better growth and higher yields. In research carried out by Khalil *et al.* (2010) in open field in Pakistan (Peshawar region), the highest yield was obtained with early October sowing, compared both to late September and late October planting times. In south-western Australia, Mwanamwenge *et al.* (1999) reported that early planting time allows faba bean plants to avoid late spring high temperatures. Confalone *et al.* (2010) reported that in open field

grown faba bean the trend of pods yield as a function of planting time depended on both fruit number and weight.

In the sub-coastal area of Naples region, yield obtained from crop cycles transplanted on 11 October was adversely affected by temperatures exceeding faba bean tolerance threshold, which caused about 30 % plant mortality in greenhouse crops, compared to 15 % mortality recorded in open field. The phenological progress of the plants which survived was sub-optimal, as high temperature also caused earlier flowering and accordingly reduced pod set. The possible reason for this reduced number of pods is pollination deficiency, which is considered a major factor causing flower abortion (Chen *et al.*, 2006). Conversely, Adisarwanto and Knight (1997) found that in Mediterranean environment sowing time has low effect on the time span between plant emergence and flowering beginning. However, temperature affects flowering progress which optimally happens at 22-23°C (Ellis *et al.*, 1988), or even at lower temperatures for some cultivars.

**Seed quality and antioxidants content.** Dry residue and soluble solids of faba bean seeds were not affected by farming system (tab. 2), but the former variable attained a higher value upon the later planting time, whereas the soluble solids did not change between the two transplants. Moreover, farming system significantly affected fiber content, which was higher in greenhouse grown seeds than in the open field ones (tab. 2), whereas planting time did not have significant effect on this variable. Other authors reported lower fiber levels in faba bean seeds compared to those detected in our research (Hedley, 2001).

Table 2

Quality indicators and antioxidants in faba bean seeds

Treatment	Dry residue mg	Soluble solids °Brix	Fiber mg	Proteins mg	Polyphenols mg	Ascorbic acid µg
<u>Farming system</u>						
Open field	190	8.5	220	279	30.1	769
Greenhouse	193	8.9	238	293	25.4	489
	n.s.	n.s.	*	*	*	*
<u>Planting time</u>						
11 October	181	8.6	227	280	31.6	594
8 November	200	8.9	231	293	25.7	663
	*	n.s.	n.s.	*	*	*

Dry residue data are per g of fresh weight of seed tissue; other data are per g of dry weight of seed tissue.

n.s. no statistically significant difference; \* significant difference at  $p \leq 0.05$ .

As shown in table 2, the seed protein content was higher under protected environment compared to open field and it increased with the planting delay. Protein values obtained in our research fall within the range reported by other authors (Ofuya and Akhidue, 2005).

Total polyphenols content (tab. 2) was higher in open field grown seeds than in the greenhouse ones and in the seeds obtained upon the first transplant. Polyphenols content in faba bean seeds obtained in our research are consistent with the values reported by Oomah *et al.* (2011).

Ascorbic acid concentration (tab. 2) was higher in faba bean seeds grown in open field than in the greenhouse and it increased with the planting time delay.

**Residual biomass chemical composition.** In order to exploit the residual biomass of faba bean crops for fuel production, we assessed its chemical composition (tab. 3). Greenhouse crops showed significantly higher lignin content, hemicelluloses and pectin than the open field ones. Open field grown biomass showed higher cellulose content, whereas no difference in crystalline cellulose was detected between the two farming systems.

The planting time delay resulted in a decrease of lignin and pectin content, an increase of cellulose (both total and crystalline) percentage and no significant effect on hemicellulose.

Table 3.

Chemical composition of faba bean residual biomass

Treatment	Lignin	Total cellulose	Crystalline cellulose (%)	Hemicellulose	Pectin
<u>Farming system</u>					
Open field	12.8	49.4	12.9	13.6	10.9
Greenhouse	14.3	47.6	12.7	15.0	11.8
	*	*	n.s.	*	*
<u>Planting time</u>					
11 October	15.0	46.4	12.0	14.5	12.3
8 November	12.2	50.9	13.7	14.1	10.6
	*	*	*	n.s.	*

n.s. no statistically significant difference; \* significant difference at  $p \leq 0.05$ .

As the biomass quality for fuel production is mainly affected by cellulose, but hemicellulose also give an important contribution, in our research faba bean residual biomass showed valuable features for biorefinery application (tab. 3).

Consistently with our results, other authors (Xuan *et al.*, 2015) reported the significant effect of planting time on biomass quality in sweet sorghum.

## CONCLUSIONS

In the present research carried out on faba bean in Mediterranean area, open field crops benefited from more appropriate climate conditions than those grown in greenhouse and the transplant performed in mid-autumn resulted in better yield performances than the early autumn one. The latter led to higher pod ripeness precocity, as well as the protected environment conditions enhanced the plants phenological progress compared to the open field.

The open field climate conditions also enhanced seeds quality, in terms of higher antioxidants content and lower fiber percentage. As for crop residual biomass, open field growing resulted in the highest yield as well as the 8 November transplant and they both showed higher cellulose fraction, the latter being the main quality component in biorefinery application prospect.

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## PHYSIOLOGICAL EFFECTS OF THIOUREA ON BIOLOGICAL PERFORMANCE OF PLANTS IN DROUGHT CONDITIONS: I. INCREASE OF ANTIOXIDANT PROTECTION

### EFFECTUL FIZIOLOGIC AL TIUREEI ASUPRA PERFORMANȚELOR BIOLOGICE A PLANTELOR ÎN CONDIȚII DE SECETĂ: I. MAJORAREA PROTECȚIEI ANTIOXIDANTE

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**Abstract.** *The greenhouse experiments were conducted to evaluate the effect of grain presoaking and foliar application of cytokinin (CK), thiourea (TU) and combination (TU+C) - thiourea with Composite preparation (pat. MD 813) containing micronutrient, on the changes of antioxidant enzymes activities, photosynthesis and grain yield of maize plants under normal water content and drought stress conditions. Treated with TU and TU+C plants resulted in great increases in the activity of SOD, CAT, APX, GR. The higher antioxidant enzyme activity in pre-treated plants was associated with the lesser MDA. Positive physiological effects of TU and TU+C were confirmed by the higher level of assimilating pigments and photosynthesis. Concentration of carotenoids was affected by drought, but spraying with TU and TU+C alleviated drought effects. It was concluded that combined application of TU+C as seed treatment and foliar spray was more effective than cytokinin in improving the Zea mays performance.*

**Key words:** plants, drought, thiourea, cytokinin, antioxidant enzymes

**Rezumat.** *În experiențe de vegetație s-a studiat efectul pre-tratării semințelor pentru semănat și aparatului foliar al plantelor de porumb cu citokinină (CK), tiouree (TU) și tiouree + Compozit (TU+C), - preparat, care conține micronutrienți, br.MD 813, asupra activității enzimelor antioxidante, fotosintezei, creșterii și productivității plantelor în condiții de umiditate optimă și de secetă. Pre-tratarea plantelor cu TU și TU+C condiționează majorarea activității superoxid dismutazei (SOD), catalazei (CAT), ascorbatperoxidazei (APX), glutationreductazei (GR) și glutationperoxidazei (GPX). Activitatea înaltă a enzimelor antioxidante este asociată cu diminuarea conținutului di-aldehidei malonice (DAM). Efectul fiziologic pozitiv al TU și TU+C este confirmat și de nivelul mai înalt al pigmenților de asimilație și fotosintezei. În concluzie: utilizarea combinației tioureei și compozitului pentru pre-tratarea semințelor pentru semănat și aparatului foliar este veridic mai efektivă pentru ameliorarea performanțelor biologice ale plantelor de Zea mays prin majorarea activității enzimelor antioxidante, fotosintezei și productivității plantelor.*

**Cuvinte cheie:** Plante, secetă, tiouree, citokinină, enzime antioxidante

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## INTRODUCTION

In connection with climate warming on Earth, the problem of water use by plants and physiological consequences of drought, remains the most important impediment to agriculture and food production on a global scale.

The developing and implementing in practice of innovative methods related to tolerance induction, impact mitigation, and stabilization of plant productivity open big prospects in agriculture country located in the area of climate risk, such as and the Republic of Moldova. Therefore, the development that would increase plant resistance to unfavorable factors is a necessary and current requirement for contemporary agriculture.

A wide variety of physiologically active substances are involved in the regulation of plant growth and development under stress. It is known a positive correlation between plant hormones content, including their exogenous application, and antioxidant protection, and plant drought tolerance formation (Asada, 2006; Parisa Sharifi *et al.*, 2012; Merewitz *et al.*, 2015; Ștefiriță *et al.*, 2015). On the other hand, a series of compounds with thiol groups are involved in maintaining the redox status (SH / -SS-), exhibiting antioxidant properties (Astir *et al.*, 2013; Pandey *et al.*, 2013). It is known the adaptogenic effect of urea derivatives – thiourea, diphenylurea, and difeniltiourea, known as compounds with similar to natural cytokinin action, due to attenuation of oxidative lesions of plant cells (Ламан *et al.*, 2010).

The objective of the research was the comparative analysis of antioxidant properties of cytokinin and thiourea to develop an innovative method to increase plant tolerance to drought.

## MATERIAL AND METHOD

As the study objects served plants *Zea mays* L., cultivar (cv.) P458. The greenhouse experiments were performed on plants grown in Mitcherliih containers with a capacity 30 kg absolutely dry soil under controlled water content conditions. The scheme of experiences provided the following treatments: a) control plants, grown on permanent soil water content - 70% from the total water capacity of soil (TWC); b) plants exposed to the drought (30 % TWC) for 10 days. The parallel treatments included the exogenous pre-treated plants with CK (0.0001%), TU (0004%) and TU (0.0004%)+C (0.0001%) in 1:1 ratio. The analyses were performed after 10 days of water stress at the initial stage of growth and during flowering of plants. Homogenization of the plant material, fixed in liquid nitrogen, and extraction were performed as described (Keshavkant and Naithani, 2010). The values of the total antioxidant status have been evaluated by the degree of modification of the content of malone dialdehyde (MDA) and of the antioxidant enzymes activity. The superoxide dismutase (SOD) activity was measured according to the method (Чевари, Чабѧ, Секей, 1985); the ascorbate peroxidase (APX) activity was assayed by the method (Nakano and Asada, 1981); the catalase activity was determined by the method (Chance B. and Machly A., 1955); glutationreductase (GR) - by reduction of oxidized glutathione in the presence of NADH H<sub>2</sub>, λ 340 nm (Schadle, Bassham, 1977); Glutation peroxidase (GPX) - by oxidation of reduced glutathione, 260 nm (Полесская,

Каширина, Алехина, 2004). The intensity of CO<sub>2</sub> assimilation, transpiration, stomatal conductivity, and water use efficiency were determined by using a portable LCA-4 gas analyzer in experiments performed under the same conditions of temperature and soil water content. The contents of chlorophyll *a* and *b* and carotenoids were determined, spectrophotometrically in 80% acetone extract. Statistical analysis of results was performed using the computer program "Statistics 7".

## RESULTS AND DISCUSSIONS

It has been known, that water stress tolerance was associated with the induction of antioxidant defence systems, including reactive oxygen species (ROS) scavenging enzymes such as SOD, CAT, APX, GPX, GR, and non-enzymatic antioxidants such as ascorbic acid, glutathione,  $\alpha$ -tocopherol, and carotenoids. Drought stress induced high production of ROS and caused damages by increasing lipid peroxidation (MDA) of the cells (Sairam and Saxena, 2000; Mittler, 2002, 2006). According to our research results, plants pre-treated with the CK, TU and TU+C reduced the impact of the oxidative stress caused by drought (tab. 1).

Table 1

**The influence of pre-treatment of maize seed with PhAS on antioxidant enzyme activity in leaves of plants, exposed to drought in the early stages of ontogenesis**

Parameters	Control, optimal	Control, drought	TU, drought	TU+C, drought
	M±m	M±m	M±m	M±m
MDA, mkmol · g <sup>-1</sup> f.w.	9.5±0.11	14.2±0.2	13.9±0.11	9.6±0.11
SOD, conv.un · g <sup>-1</sup> f.w.	126.8±2.1	145.6±2.9	197.9±3.1	199.0±2.7
CAT, mmol · g <sup>-1</sup> f.w.	0.45±0.01	0.38±0.02	0.50±0.001	0.50±0.004
APX, mmol · g <sup>-1</sup> f.w.	1.9±0.03	2.1±0.05	2.2±0.04	2.8±0.06
GR, mmol · g <sup>-1</sup> f.w.	55.7±0.77	60.6±1.0	63.5±1.23	73.4±1.12
GPX, mmol · g <sup>-1</sup> f.w.	26.0±0.54	27.8±0.5	28.9±0.31	31.0±0.28

The effect of TU and TU+C treatments on antioxidant enzymes activity had similar trend under normal soil water content and drought conditions. Namely, it resulted in considerable increases in the activity of SOD and CAT enzymes accompanied by great reduction in MDA content. Maximum growth of SOD and CAT activity was observed by treating plants with TU under drought conditions (56.1% and 11.1%, respectively). Pre-treatment of seeds with the combination of TU+C led to greater increases of APX, GPX and GR activities (43.8%, 19.2%, and 31.7%) compared to the degree of magnification in the activity of these enzymes induced by TU (16.7%, 11.0%, and 14.0%, respectively). Also, greater

antioxidant enzyme activities in pre-treated plants were associated with the lesser MDA content.

In field conditions the drought stress can be aggravated by high temperatures and intense solar radiation, which drastically reduces crop plants. The plants at reproductive growth stage are very sensitive to high temperatures and insufficient soil water content.

According to our previous research findings (Ștefirta *et al.*, 2015), the exogenous application of phytohormones to some extent neutralized the adverse drought effect. This action could be explained with improving the water status in the cells and activation of antioxidant enzyme system. Major effect occurred following the administration of exogenous CK. Taking into account the close connection between phytohormones and water status it could be assumed that phytohormones were capable to stabilize water homeostasis and to influence on the degree of formation of ROS. These results formed the basis of the idea that applying of substances with cytokinin activity would result in the optimization of the antioxidant systems in moderate drought conditions. Indeed, subsequent studies have demonstrated an activation of enzymatic antioxidant protection system in leaves of plants pre-treated with physiologically active substances such as cytokines (tab. 2).

Table 2

**Influence of CK, TU and TU+C on the antioxidant protection in the leaves of *Z. mays* plants under drought conditions**

Parameters	Control	CK	TU	TU+C
MDA, mkmol · g <sup>-1</sup> f.w.	<u>37.35±1.67*</u>	<u>34.43±1.21</u>	<u>27.67±1.1</u>	<u>25.34±1.2</u>
	44.30±0.98**	35.94±0.57	31.44±0.6	26.65±0.6
SOD, conv.un · g <sup>-1</sup> f.w.	<u>50.64±0.88</u>	<u>56.95±0.72</u>	<u>59.37±0.6</u>	<u>75.97±0.8</u>
	60.55±0.69	63.64±0.88	69.89±0.7	80.63±1.2
CAT, mmol · g <sup>-1</sup> f.w.	<u>0.83±0.003</u>	<u>0.93±0.002</u>	<u>1.17±0.006</u>	<u>1.19±0.009</u>
	0.76±0.006	0.90±0.008	1.03±0.008	1.08±0.009
APX, mmol · g <sup>-1</sup> f.w.	<u>2.48±0.03</u>	<u>3.81±0.04</u>	<u>5.10±0.03</u>	<u>4.22±0.05</u>
	2.55±0.03	5.33±0.07	5.70±0.06	5.92±0.0
Chlorophyll a+b, mg · 100 g <sup>-1</sup> f.w.	<u>200.46±2.98</u>	<u>239.54±3.5</u>	<u>292.12±3.1</u>	<u>249.35±3.2</u>
	187.91±2.16	220.87±2.3	242.17±2.2	221.13±3.1
Carotenoids, mg · 100 g <sup>-1</sup> f.w.	<u>33.62±0.98</u>	<u>44.87±0.76</u>	<u>46.75±0.65</u>	<u>44.12±0.45</u>
	30.25±0.59	39.44±0.45	40.21±0.21	43.50±0.34

\*- in optimal soil water content conditions; \*\*- in drought conditions

The treated plants had a more effective protection system, which ensured the possibility of their functioning under stress. It was evident, the plants pre-

treated with CK, TU, and especially with TU+C, in drought conditions differed from control ones by significantly lower content of MDA, which was indicative of lesser degree of lipid peroxidation and oxidative destructions due to higher protection capacity of antioxidant enzymes SOD, CAT and APX. Positive physiological effect of TU and TU+C was confirmed by the higher level of assimilating pigments. Concentration of carotenoids was affected by drought, but spraying with TU and TU+C alleviated drought effects. Combined application of TU and TU+C as seed treatment and foliar spray was more effective than cytokinin in improving the *Z. mays* performance (tab. 3).

Table 3

**The effect of TU and TU+C use on biological performance of *Z. mays* plants in drought conditions**

Parameters	Control	CK	TU	TU+C
Height of plant, dm	$12.15 \pm 0.3$ *	$14.28 \pm 0.1$	$16.10 \pm 0.4$	$13.02 \pm 0.20$
	$9.85 \pm 0.13$ **	$11.2 \pm 0.12$	$10.7 \pm 0.17$	$12.23 \pm 0.21$
Leaf area, dm <sup>2</sup>	$37.82 \pm 0.35$	$39.04 \pm 0.28$	$39.19 \pm 0.19$	$39.34 \pm 0.41$
	$30.23 \pm 0.32$	$32.30 \pm 0.29$	$33.95 \pm 0.43$	$35.70 \pm 0.34$
Productivity, g/plant	$52.8 \pm 0.5$	$61.70 \pm 0.4$	$69.30 \pm 0.6$	$70.0 \pm 0.9$
	$34.81 \pm 0.5$	$38.94 \pm 0.3$	$37.72 \pm 0.6$	$39.74 \pm 0.9$

\*- in optimal conditions; \*\*- in drought conditions

## CONCLUSIONS

1. Cytokinin (CK), thiourea (TU) and thiourea in combination with Composite preparation, containing micronutrients, (TU+C), all used for *Zea mays* plants treatment through grain presoaking and foliar, provided the increase of the antioxidant capacity of cells, the content of the assimilating pigments, and the reduction of lipid peroxidation.

2. Treating plants with TU and TU+C resulted in considerable increases in the activity of SOD, CAT, APX, GR. The greater antioxidant enzyme activity in pre-treated plants was associated with the lesser MDA content.

3. The TU and TU+C increased the adaptive potential of plants, reduced the negative action of soil water content deficit, and optimized the processes of growth and productivity.

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## INVOLVEMENT OF SALICYLIC ACID DERIVATIVES IN PLANT WATER POTENTIAL ADJUSTMENT UNDER CONDITIONS OF INSUFFICIENT MOISTURE

### ANTRENAREA DERIVAȚILOR ACIDULUI SALICILIC ÎN REGLAREA POTENȚIALULUI APEI PLANTELOR ÎN CONDIȚII DE INSUFICIENȚĂ DE UMIDITATE

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**Abstract.** *The greenhouse experiments were conducted to evaluate the effect of salicylic acid and salicylate  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{++}$  on the possibility of adjusting the water status of the Zea mays, L. and Sorghum bicolor L. Moench plants in the drought conditions. It has been shown that drought causes significant changes in the stomatal conductance, intensity of transpiration, coupled with the change in the value of water potential ( $\Psi_w$ ) and the hydrostatic pressure ( $\Psi_p$ ) of the plants leaves of both species. Salicylic acid and salicylates  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{++}$  have the property of the plant water haemostatis by increasing the degree of hydration of the tissue, reducing the water deficits, increased turgidity organs and retention of water, adjusting the hydraulic and stomatal conductivity, thus ensuring the maintenance of a higher level  $\Psi_w$  in both favourable and at moderate deficiency moisture conditions.*

**Key words:** plants, drought, salicylic acid, derivate, water potential, resistance.

**Rezumat.** *În experiențe cu umiditatea dirijată s-a studiat posibilitatea reglării status-ului apei plantelor de Zea mays L. și Sorghum bicolor L., Moench în condiții de secetă moderată prin utilizarea acidului salicilic și salicilaților de  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{++}$ . S-a demonstrat că seceta condiționează schimbări semnificative atât în intensitatea transpirației apei, cuplate cu modificarea conductanței stomatelor, cât și în valoarea potențialelor apei ( $\Psi_w$ ) și presiunii hidrostatice ( $\Psi_p$ ) ale frunzelor plantelor. AS și salicilații de  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{++}$  au proprietatea de homeostatare a apei în organismul vegetal prin majorarea gradului de hidratare a țesuturilor, micșorarea deficitului de saturație, creșterea turgescenței organelor și capacității de reținere a apei, reglarea conductibilității hidraulice și stării stomatelor, ceea ce asigură menținerea  $\Psi_w$  la un nivel mai ridicat atât în condiții favorabile de umiditate, cât și la un deficit moderat de apă.*

**Cuvinte cheie:** plante, secetă, acid salicilic, derivați, potențialul apei, rezistență

## INTRODUCTION

It is known that living organisms are totally dependent on the presence of water, their predominant component. The smallest disturbance of the water balance in them are accompanied by serious changes in absolutely all life

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processes. The plants, being sedentary organisms, are constantly exposed to fluctuating of hydro-thermal conditions, often extreme ones. Agriculture as a priority branch of the country is highly vulnerable to risk factors, both to the ecological ones of climatic origin, such as lengthy droughts, early autumn or late spring frosts, scorching heat, etc., and to the fluctuation in their extreme range during ontogeny.

In this connection it is particularly important to develop the methods of induction of tolerance to reduce the impact of drought and stabilize plant productivity. In this aspect along with improving agricultural technique and application of new technologies in complex outcome measure, an important role is assigned to the creation and implementation in practice of new varieties with high potential for productivity and resistance, not only tolerant to drought, cold, etc., but also to fluctuation in wide range of external environmental factors. It is widely accepted that the change in the water status is the primary reaction of the organism to changing of environmental conditions, and is a trigger of cascade of protection reactions, as well as, is a commutator of cell functional activity from normal regime to one, characteristic of stress conditions. Given the fact that the regulation and coordination of growth, development and productivity of plants come under the control of plant hormones, more and more researches in phyto-physiology are geared toward elucidating of mechanisms of their action and to explore ways of exogenous regulating of functional status of plants under moderate drought. Lately particular attention was given to such effect of salicylic acid (SA). There have data that demonstrated the role of SA in maintaining of homeostasis by enhancing water absorption and stomatal apparatus reactivation (Raskin, 1992; Shachirova *et al.*, 2003). It was established that exogenous SA in high concentrations ( $10^{-3}$  M) conditioned the closing of desmotubule of plasmodesmata (Лялин *et al.*, 1993), and SA in low concentrations ( $10^{-5}$  -  $10^{-7}$  M) - the unlocking of water channels (Безрукова, Сахабутдинова, Шакирова, 1999). But few researches have been undertaken to elucidate the effect of AS and its derivatives on adjusting of plants water status in dry conditions. It was assumed that SA is able to change turgor-dependent valve diameter of desmotubule of plasmodesmata in a short time (Wilkinson and Davies, 2002). These data motivated the study of the SA effect on the peculiarities of regulating of the water status in plants to ensure adaptation, tolerance and productivity both in optimal conditions and under moderate drought.

## MATERIALS AND METHODS

As the study objects served plants *Zea mays* L., cultivar (cv.) P459 and *Sorghum bicolor* L., cv. Piscevoi 1, grown in Mitcherlii containers with a capacity 30 kg absolutely dry soil under controlled water content conditions. The scheme of experiences: 1 - control plants grown on permanent soil water content - 70% from the total water capacity of soil (TWC); 2 - plants exposed to the drought (30 % TWC) for 10 days. The parallel treatments included the plants pre-treated through grain soaking and foliar with an aqueous solution of SA and salicylates  $\text{NH}_4^+$ ,  $\text{K}^+$ .

Hydraulic conductivity of plant was calculated from the diurnal relationship of the water potential gradient of leaves and stems; and transpiration rate – according to equation:

$F = L_p (\Psi_{w\text{ fr}} - \Psi_{w\text{ tulip}})$ , where  $F$  - the rate of transpiration, or water flow;  $L_p$  - a proportionality factor defined as hydraulic conductivity of the plant (Blatt, 2000; Moreshet *et al*, 1990). Given that the water flow through the plant is maintained due to soil water potential gradient and leaves site, which transpires, just its value was considered as transpiration rate (Steudle *et al*, 1987). Water potential ( $\Psi_w$ ) was determined by the compensation method (Колесник and Еропов, 1989).

Turgor potential ( $\Psi_p$ ) was obtained from the difference of water and osmotic potential:  $\Psi_p = \Psi_\pi - \Psi_w$ . Cellular juice concentration was determined by refractometer type Carat (1989) according to Larcher (1976). The intensity daytime transpiration of different leaves that differed by age was performed using transpirometer (Копецкы 1981). Data on the water status parameters in organs have been shown as mean value  $\pm$  standard error of 5 reproductions, and as the average modification degree in 3-5 experiments. The results were statistically analyzed using the software package "Statistics 7" for computers.

## RESULTS AND DISCUSSION

The research results have shown that plants treated with SA and salicylates exhibited more pronounced self-regulating properties of water exchange, optimization of the degree of hydration, and turgidity of tissue, both in optimal and moderate deficit of soil water content conditions (tab. 1).

Table 1

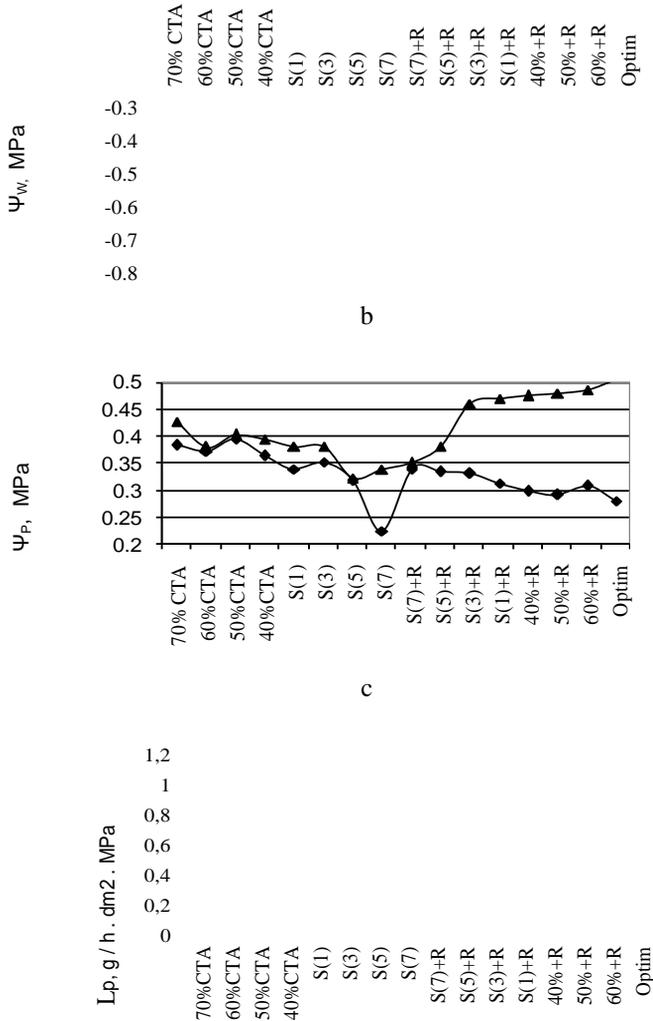
**Water status parameters modification of maize and sorghum plants under the influence of salicylic acid and moderate deficit of soil water content**

Treatments	Water content (WC), g 100 g <sup>-1</sup> f. m.		Saturation deficiency (SD), % of full saturation		Intensity of transpiration (IT), Mg dm <sup>-2</sup> h <sup>-1</sup>	
	M $\pm$ m	$\Delta$ , % of control	M $\pm$ m	$\Delta$ , % of control	M $\pm$ m	$\Delta$ , % of control
<i>Zea mays</i> L., cv. P459						
Control	<u>74.9 <math>\pm</math> 0.5</u> 69.4 $\pm$ 0.4	-7.3	<u>7.8 <math>\pm</math> 0.24</u> 28.4 $\pm$ 0.32	264.1	<u>828.3 <math>\pm</math> 23.0</u> 482.8 $\pm$ 12.0	-41.71
SA	<u>77.0 <math>\pm</math> 0.3</u> 73.5 $\pm$ 0.1	-4.6	<u>3.8 <math>\pm</math> 0.04</u> 21.1 $\pm$ 0.22	170.5	<u>894.0 <math>\pm</math> 10.8</u> 556.2 $\pm$ 7.6	-32.85
Salicylates NH <sub>4</sub> <sup>+</sup> , K <sup>+</sup> .	<u>80.12 <math>\pm</math> 0.6</u> 77.72 $\pm$ 0.9	-3.0	<u>2.0 <math>\pm</math> 0.03</u> 18.12 $\pm$ 0.09	132.1	<u>976.64 <math>\pm</math> 22.4</u> 622.32 $\pm$ 12.8	-24.86
<i>Sorghum bicolor</i> (L.) Möench, cv. Pișcevoi 1						
Control	<u>72.68 <math>\pm</math> 0.38</u> 69.55 $\pm$ 0.23	-4.31	<u>8.66 <math>\pm</math> 0.11</u> 30.55 $\pm$ 0.21	252.77	<u>636.38 <math>\pm</math> 22.87</u> 460.06 $\pm$ 25.34	-27.71
SA	<u>74.40 <math>\pm</math> 0.15</u> 73.98 $\pm$ 0.60	-0.56	<u>7.18 <math>\pm</math> 0.14</u> 23.71 $\pm$ 0.34	230.25	<u>1015.18 <math>\pm</math> 30.45</u> 874.58 $\pm$ 13.46	-13.85
Salicylates NH <sub>4</sub> <sup>+</sup> , K <sup>+</sup> .	<u>76.0 <math>\pm</math> 0.54</u> 75.78 $\pm$ 0.7	-0.29	<u>5.98 <math>\pm</math> 0.15</u> 19.81 $\pm$ 0.21	128.75	<u>1128.18 <math>\pm</math> 29.27</u> 992.85 $\pm$ 27.14	-11.99

\*- in optimal soil water content conditions; \*\*- in drought conditions

Under moderate deficiency of soil water content, the WC in the leaves of the plants *Zea mays* L. cv. P459 treated with SA was 5.97% greater than of the control plants at the same condition. SA assured the reduction of SD with 25.7% of the value of this parameter in untreated maize plants and with 22.5% - in sorghum plants. Due to the increase of tissue turgidity, the water consumption in transpiration process of

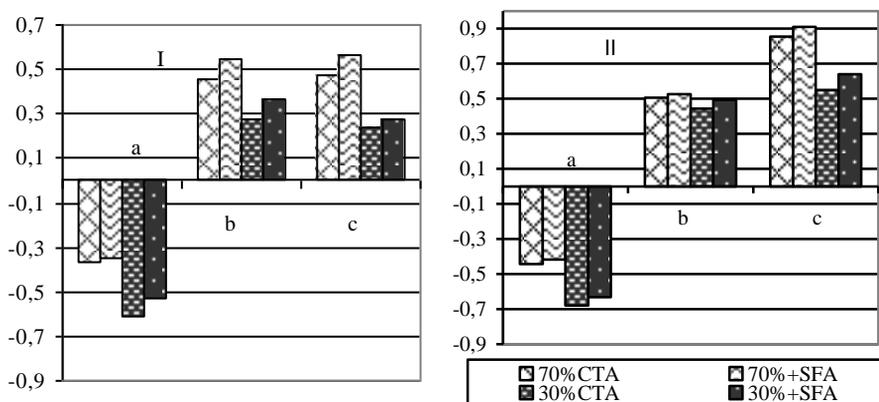
treated plants maintained at the highest level both in optimal soil water content conditions and in insufficient ones (tab. 1). The IT in the plants, treated with SA and salicylates, under moderate drought conditions was greater compared to control plants by 73.4 and 139.7 mg dm<sup>-2</sup> h<sup>-1</sup> for maize and by 414.5 and 532.8 mg dm<sup>-2</sup> h<sup>-1</sup> –for sorghum plants. The plant was able, to some extent, compensate for water loss due to the increase of hydraulic conductivity and transport the water from the roots to the leaves (Raschke, 1987).



**Fig. 1** Changing  $\Psi_w$ ,  $\Psi_p$  (MPa) in leaves and hydraulic conductivity ( $L_p$ ) in connection stem-leaf of maize plants, cv. P459 and sorghum cv. Pișcevoi (II) under fluctuation of water content and water stress of various durations. Legend: 1 - sorghum plants; 2 - maize plants.

The investigation results (fig. 1) have shown that in spite of higher tolerance of anisohydric plants and they were prone to hydraulic deficiencies because of the limit of hydraulic system narrowed during drought conditions.

Decreasing of soil water content below the critical threshold prompted changes both in the degree of hydration and in value of the water potentials and hydrostatic pressure of plant leaves. The reduction of the water flow to the leaves (IT) and the increase of gradients  $\Psi_w$  and  $\Psi_p$  under insufficient soil water content were associated with reduced hydraulic conductivity of the tissues - most significant for isohydric plants (fig. 1). Typical anisohydric plants were able to maintain hydraulic conductivity and water supply to the leaves at a relatively constant level. As soil water content decreased and tissues were dehydrated the stomatal resistance increased and their conductivity to water vapor decreased. The plants treated with the combination of salicylic acid with  $\text{NH}_4^+$ ,  $\text{K}^+$  under drought conditions kept the hydraulic conductivity at a higher level as compared with untreated ones (fig. 2).



**Fig. 2** Influence of water stress on water potential,  $\Psi_w$ , MPa (a), turgidity,  $\Psi_p$ , MPa (b) and hydraulic conductivity,  $L_p$ , g / h.dm<sup>2</sup>.MPa (c) in segments "stem - leaves" of plants *Zea mays* L., cv. P459 (I) and *Sorghum bicolor* L. Moench, cv. Pișcevoi 1 (II), treated with the combination of the salicylic acid with  $\text{NH}_4^+$ ,  $\text{Mg}^{2+}$ ,  $\text{K}^+$ .

Due to the fact that the stomatal conductance in plants treated with SA and salicylates was reduced to a lesser extent under water content deficit, the flow of water continued to keep, which ensured the maintenance of a higher level of  $\Psi_w$ . Under optimum water content a trend of intensified transpiration of treated plants can be expected, which, incidentally, can be also a consequence of the optimization of water potential and turgor pressure (fig. 2).

The experimental results led to the conclusion that salicylic acid reduced the impact of water stress, caused by insufficient water content, by adjusting the hydraulic conductivity, stomatal conductance, and water consumption in the process of transpiration during the action of hydric stress, and by a compensatory

reaction and repair of physiological parameters after improving water content conditions. Salicylates increased the  $\Psi_w$  and ability of water attraction.

## CONCLUSIONS

1. Drought conditioned the significant changes both in the intensity of transpiration of water, coupled with the change in stomatal conductance, and in the values of water potential ( $\Psi_w$ ), and the hydrostatic pressure ( $\Psi_p$ ) of the plant leaves.

2. Salicylic acid and salicylates  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{2+}$  demonstrated the property of maintaining of water homeostasis in the vegetal organism by increasing the degree of hydration of the tissue, reducing the saturation deficiency, increasing turgidity of organs and capacity of water retention, adjusting the hydraulic conductivity, and the stomata state, both at the favourable water content conditions and at a moderate water deficiency.

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## COMBINED EFFECTS OF PHOSPHORUS FERTILIZATION AND SOIL WATER DEFICIT ON LEAF DEVELOPMENT AND AMINO ACIDS CONCENTRATIONS IN TOW SOYBEAN (*GLYCINE MAX L. MERR*) CULTIVARS

EFFECTUL COMBINAT AL FERTILIZĂRII CU FOSFOR ȘI DEFICITULUI DE APĂ DIN SOL ASUPRA DEZVOLTĂRII FRUNZELOR ȘI CONCENTRAȚIEI DE AMINOACIZI LA DOUĂ CULTIVARE DE SOIA (*GLYCINE MAX L. MERR*)

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**Abstract.** The effects of phosphorus application ( $KH_2PO_4$ ) levels 0 mg P/kg (P0), 10 mg (P10), 20 mg (P20) and 100 mg P/kg (P100) on leaves development and amino acids concentrations in two cultivars of soybean under two soil moisture regimes: control as normal irrigation, 70% of water holding capacity (WHC) and moderate stress (35% WHC) were studied. Plants were subjected to water deficit conditions at flowering stage for two weeks. Leaves area and amino acids concentrations in leaves were measured in relation to P nutrition. Plant performance of Licurici was better than Zodiac under P deficiency (P0) and low P (P10) supply. Leaves growth of both soybean cultivars was significantly reduced when the soil moisture content was decreased from 70% to 35% WHC. The highest leaf area and dry matter accumulation by leaves was found in the treatment with 100 mg  $kg^{-1}$  of P application. The same trend of P supplementation influence was denoted in plants subjected to drought stress, but to a lesser extent than in well-watered plants. Total soluble amino acids concentrations in leaves rose under temporary drought irrespective of P nutrition levels. Cultivar Zodiac displayed higher response in amino acids accumulation at suboptimal moisture regime. Hence, the adequate P fertilization improved the ability of soybean plants to tolerate drought stress.

**Key words:** amino acids, *Glycine max.*, leaf area, moisture, phosphorus

**Rezumat.** S-au studiat efectele aplicării diferitor doze cu fosfor 0 mg P/kg (P0), 10 mg (P10), 20 mg (P20) și 100 mg P/kg (P100), asupra dezvoltării frunzelor și a conținutului de aminoacizi liberi la două cultivare de soia cultivate în două regime de umiditate a solului: martor, irigare la 70% CTA și stresul hidric moderat (35% CTA). Plantele au fost supuse deficitului de apă la faza de înflorire pentru două săptămâni. S-a determinat suprafața foliară și conținutul de aminoacizi liberi în funcție de nivelul de nutriție cu P. Creșterea plantelor cultivarului Licurici a fost mai bună decât a cultivarului Zodiac în condițiile deficitului de P (P0) și asigurării joase cu nutrient (P10). Dezvoltarea frunzelor la ambele cultivare s-a redus semnificativ când nivelul de umiditate din sol s-a micșorat de la 70% CTA la 35% CTA. Cea mai mare suprafață și greutate de substanțe uscate acumulate în frunze s-a stabilit la aplicarea 100 mg  $kg^{-1}$  de P.

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*Aceeași influență a suplimentării plantelor cu P s-a depistat și în condiții de secetă, dar la un nivel mai inferior comparativ cu plantele irigate normal. Concentrațiile de aminoacizi solubili din frunze s-a majorat sub acțiunea secetei temporare indiferent de nivelul de nutriție cu P. Cultivarul Zodiac a manifestat răspuns mai înalt la nivel de acumulare a aminoacizilor în condiții insuficiente de umiditate. Deci, s-a stabilit că fertilizarea adecvată cu P a îmbunătățit toleranța plantelor de soia la secetă.*

**Cuvinte cheie:** aminoacizi, fosfor, *Glycine max.*, umiditate, suprafața foliară

## INTRODUCTION

Phosphate (Pi) deficiency is a major agricultural production constraint in many soils involving up to 70% of global arable surface (Vance *et al.*, 2003). Phosphate is a component of nucleic acids and cellular membranes, and essential for a large range of metabolic processes (Raghothama, 1999; Vance *et al.*, 2003). Phosphate deficiency decreases plant growth and photosynthesis and thus biomass accumulation and yield. It was reported that insufficient P nutrition can affect cell division in growing tissues and restricts expansion growth of plant organs (Chiera and Rufty, 2002; Radin and Eidenbock, 1984).

Soybean is an important component of the agro-system due to its capacity to produce significant quantities of oil and protein-rich seed and to improve soil quality. However, its growth and productivity is very low due to many environmental constraints, particularly insufficient phosphorus nutrition and drought. According to research of Desclaux *et al.* (2000) soybean is very sensitive to water shortage during the stage of reproductive development. Water deficit often causes nutrient deficiency, particularly phosphorus (Haefele *et al.*, 2006). There is a general agreement that the ability of crops to cope with drought can be enhanced by adequate P nutrition (Gutierrez-Boem and Thomas, 1999). Studies by Burman *et al.* (2009) have demonstrated the beneficial impact of P application in reducing plant water stress in moth bean and cluster bean. Likewise, positive effects of P application on plant performance under stress environments have been reported for soybean (Devi and Sinclair, 2013). Under field conditions, crops are routinely exposed to a range of stress factors; therefore, understanding of the interaction between environmental factors such as P nutrition and soil moisture is critical for finite resources management of the sustainable agriculture. The goal of this study was to determine the effect of P application levels on leaf area development and amino acids contents in two soybean cultivars during exposure to water deficit conditions of soil.

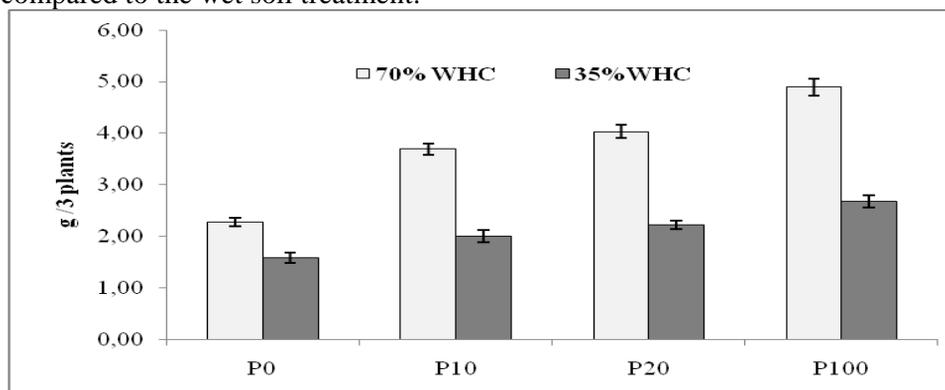
## MATERIAL AND METHOD

The pot experiment was conducted in a greenhouse at the Organic Plant Production and Agroecosystems Research in the Tropics and Subtropics Department, University of Kassel, Germany. Treatments included the factorial combination of four P levels, two soil water regimes (control and low water supply) and two soybean (*Glycine max. L.*) cultivars namely Zodiac and Licurici. The P fertilizer was  $\text{KH}_2\text{PO}_4$ , and net P

levels were 0, 10, 20 and 100 mg kg<sup>-1</sup> of soil as P0, P10, P20 and P100, respectively. The growth medium consisted of a soil-sand mixture 3:1 by volume fertilized with macro- and microelements. All pots with P application received potassium (K) as KCl to equivalent potassium level. The content of available phosphorus was 4,4 mg kg<sup>-1</sup> (CAL) (and 11,5 mg Olsen method), pH (CaCl<sub>2</sub>) 7,74, total N - 0,04% and C - 1,42%. Seeds were treated with *Bradyrhizobium japonicum* at sowing time. Two soil water treatments, well-watered and temporary drought, commenced at the initial flowering stage (lasting for two weeks). Mean night temperature ranged within 18-20 °C and mean day temperature varied 26-28 °C. Relative humidity varied between 60-65%. The pots were placed on tables and rotated every second day for random distribution in a greenhouse. At harvest, 6 weeks after sowing, plants were separated into leaves, stem plus petioles, roots and nodules and leaf area was determined. Leaf area was determined by leaf meter (LiCor model 3000). The dry weight of plant materials was determined after drying in an oven at 70 °C until constant weight was obtained. The procedures for the total free amino acids contents in plant tissues were described in detail by Yemm and Cocking (1955). Data of experiments were averaged and the respective standard error (SE) was calculated. To determine the significance of the means differences, least significant differences (LSD) were estimated at 5% probability level.

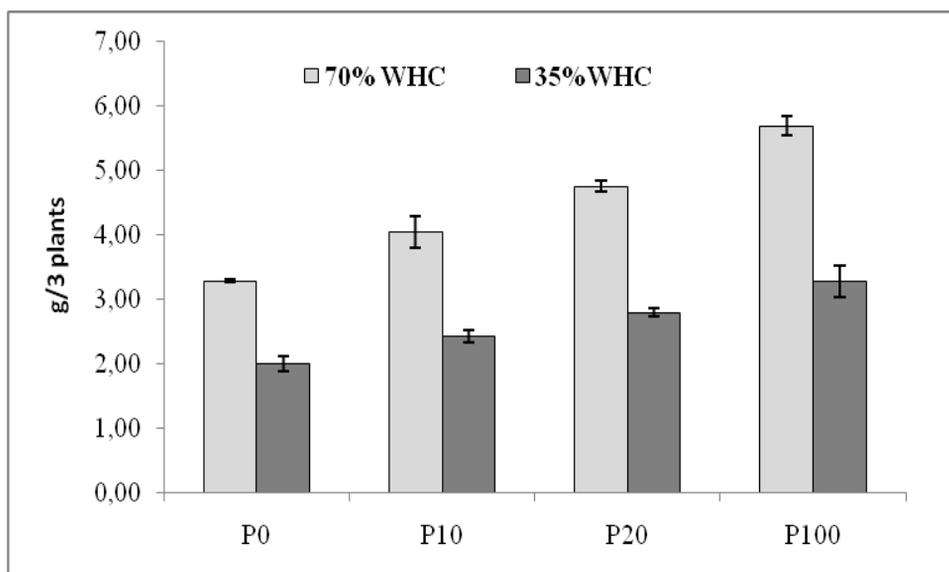
## RESULTS AND DISCUSSIONS

There is a general consensus about the importance of leaf area for plant growth and yield (Russel *et al.*, 1989). Many investigations have documented that leaf development is more susceptible to environmental stresses, particularly to P deficiency and drought than stems and roots. Since water shortage is more dangerous at flowering stage in grain legumes grown under rain-fed conditions, the present pot experiment focused at investigating the effects of different rates of P application on dry matter production of leaves and their area development as well as amino acids contents of plants subjected to low soil moisture regime at the beginning of flower bud initiation stage. The soil water regime influenced the leaf growth significantly and leaf parameters declined in the dry conditions compared to the wet soil treatment.



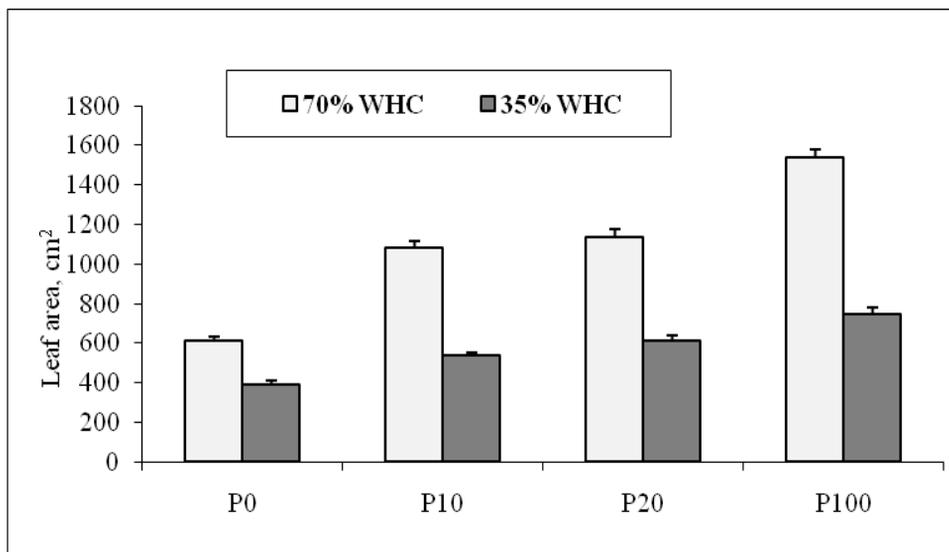
**Fig. 1** Effect of phosphorus supply and soil water regime on leaves dry matter accumulation (g/3 plants) of cv Zodiac soybean plants. Bars indicate standard error of the mean. P<0.05

Experimental results revealed that leaves growth of cvs Zodiac and Licurici was affected significantly ( $p < 0.05$ ) by P fertilization (fig. 1 and fig. 2). The plant dry weight and total leaf area followed similar trend across the treatments at harvest. It was established that the leaves dry matter (DM) of two soybean cultivars was significantly reduced by combination of low P supply and water deficit. The soybean cultivars displayed differences in the responses of leaf growth to P application at two water regimes. Leaves growth of Zodiac had more pronounced responses to supplemental P nutrition than Licurici.



**Fig. 2** Effect of phosphorus supply and soil water regime on leaves dry matter accumulation (g/3 plants) of cv Licurici soybean plants. Bars indicate standard error of the mean.  $P < 0.05$

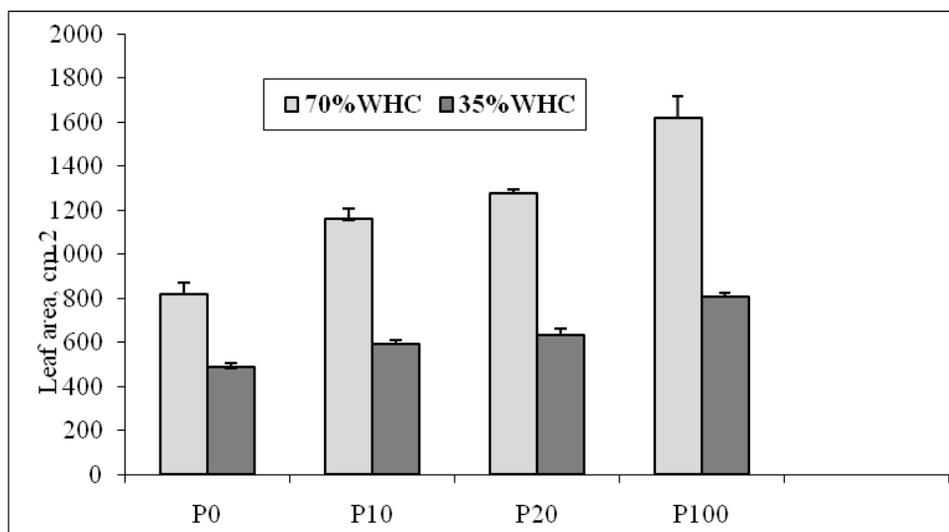
The most pronounced difference between cultivars was established at insufficient P nutrition (P0). Radin and Eidenbock, (1984) reported that phosphate deficiency limited cell expansion by reducing hydraulic conductance inside plants, which may lead to reduced plant size and leaf area expansion. Under P insufficiency, the reduction in DM with cv. Zodiac (114%) was greater as compared to that with Licurici (73.2%) under normal water conditions and adequate nutrient (P100) supply. Increasing P supply promoted plant growth and the difference between Zodiac and Licurici was reduced. Shen *et al.* (2013) also stated that appropriately manipulating P supply can enhance plant growth, nutrient uptake and the ability to resist various stresses, including water deficit.



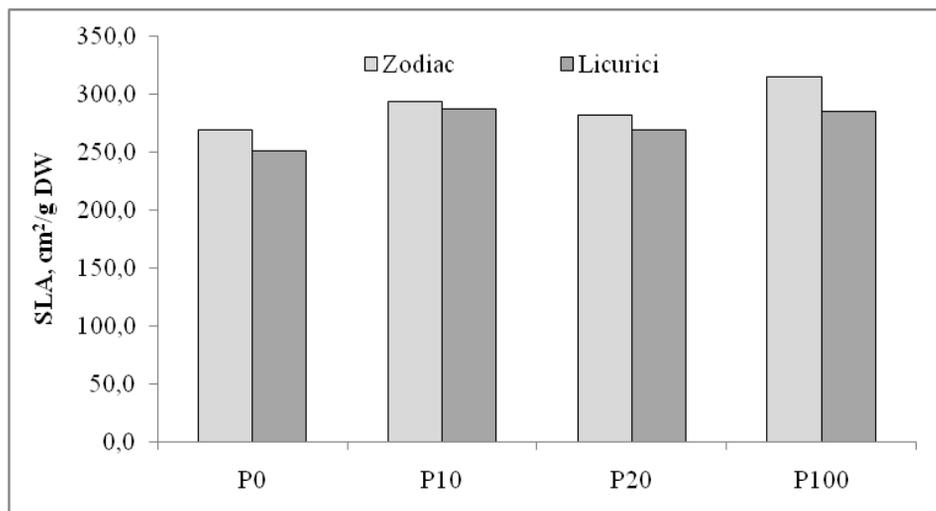
**Fig. 3** Interactions of P addition and soil water regime on surface area ( $\text{cm}^2/3$  plants) of cv Zodiac soybean plants. Bars indicate standard error of the mean

The significant response of plants to the low level of P application indicates that soil P was very scarce and, also, that soybean plants have a high requirement for fertilization. Our results are consistent with the data of Vadez *et al.*, (1999) which confirmed the sensitivity of *Phaseolus vulgaris* to P deficiency. It is necessary to note that the influence of P fertilization on this morphological trait was more pronounced under sufficient moisture level. The application of P at different rates increased leaf area by 21.8-65.7% in Licurici (fig. 4) and by 36-90% in Zodiac (fig. 3) under drought conditions. The lowest leaf area was observed under low P nutrition (P0) and Zodiac had a lower value by 150% and 90,4% under well watered and stress treatments respectively compared to high P treatment (P100). Therefore, the maximum leaf area under optimal and low soil moisture level was observed in treatment with the application of 100 mg of P per kg of soil. We consider that the greater susceptibility of cv. Zodiac to P deficiency than Licurici is partly due to higher vulnerability of leaf growth to insufficient P supply. Studies (Guterez-Boem and Thomas, 1999) have demonstrated that P insufficient supply reduced leaf area and the number of leaves as well as the relative leaf appearance rate.

It was established that the irrigation of plants to optimal soil moisture (70% WHC) increased the mean leaf area by 69% and dry matter accumulation by 64% of Licurici in treatment without fertilization compared with the ones grown at low moisture level (35% WHC). In cv Zodiac, the increases of these two parameters due to normal water supply were 55.8% and 44.3%, respectively compared to plants subjected to drought.



**Fig. 4** Interactions of P addition and soil water regime on surface area (cm<sup>2</sup>/3 plants) of cv Licurici soybean plants. Bars indicate standard error of the mean.

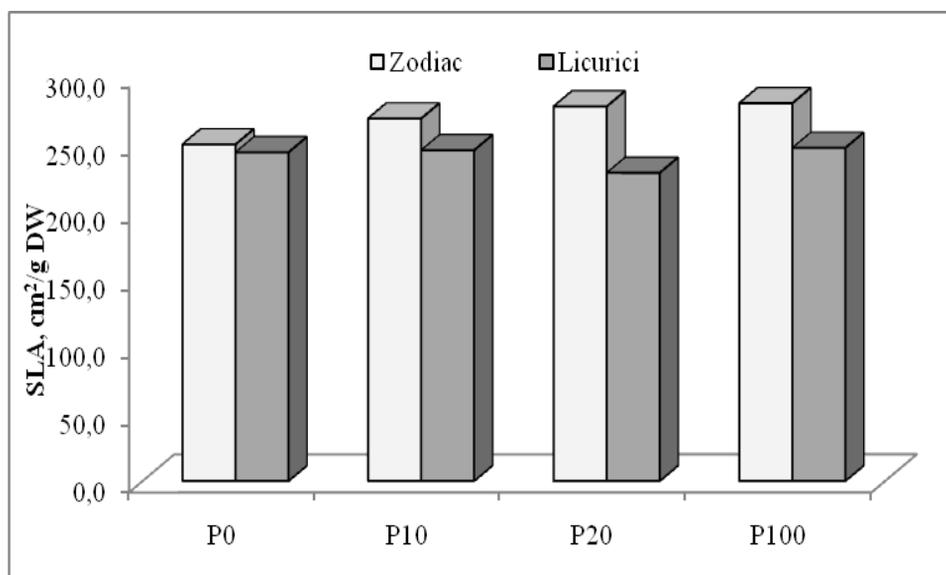


**Fig. 5** Effect of P supply on specific leaf area (SLA, cm<sup>2</sup> g<sup>-1</sup>), of soybean plants under normal water supply

Therefore, experimental data clearly demonstrated that P supplemental nutrition helps plants overcome water deficit stress probably by increasing the hydraulic conductance of water (Singh and Sale, 2000). The combination of these two constraints resulted in a greater reduction in total leaf area than in plant dry mass. So our data is consistent with the results of other researchers (Gutierrez-

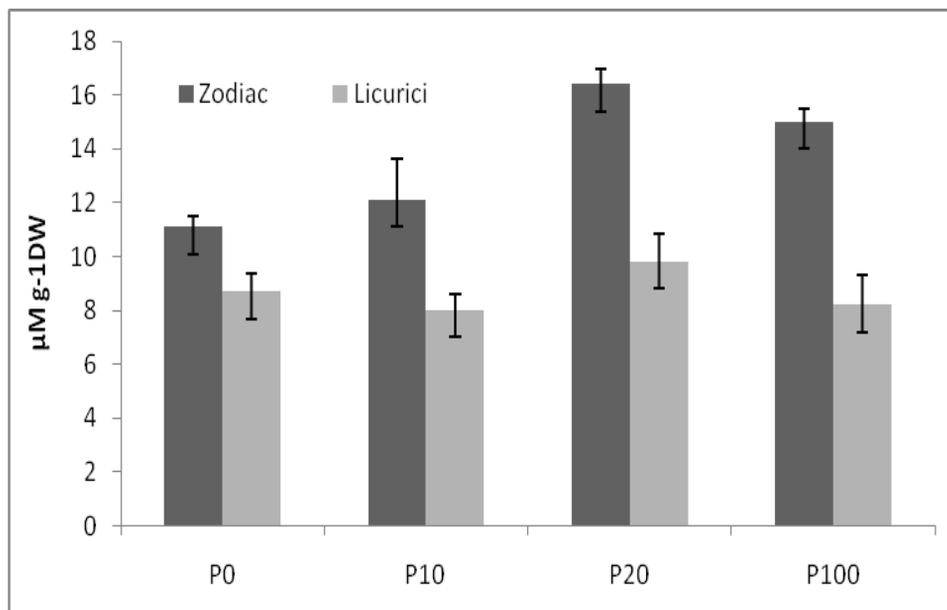
Boem and Thomas, 1999). The data presented in figure 5 shows that specific leaf area (SLA) did not change significantly in relation to P treatments under normal water supply. The higher P supply had only a tendency to improve this parameter. The same trend was observed in plants subjected to temporary drought (fig. 6). Under insufficient water regime Licurici displayed lower values than Zodiac in treatment with adequate phosphorus nutrition. Compared with those under well-watered condition (70% WHCC) treatments of drought stress (35% WHC) exhibited the lowest SLA.

Biochemical studies have shown that plants under environmental stresses accumulate a number of metabolites, termed compatible solutes because they do not interfere with metabolically reactions. The majority of such metabolites include carbohydrates and nitrogen-containing compounds, particularly amino acids.



**Fig. 6** Effect of P supply on specific leaf area of soybean plants under water deficit conditions (35% WHC)

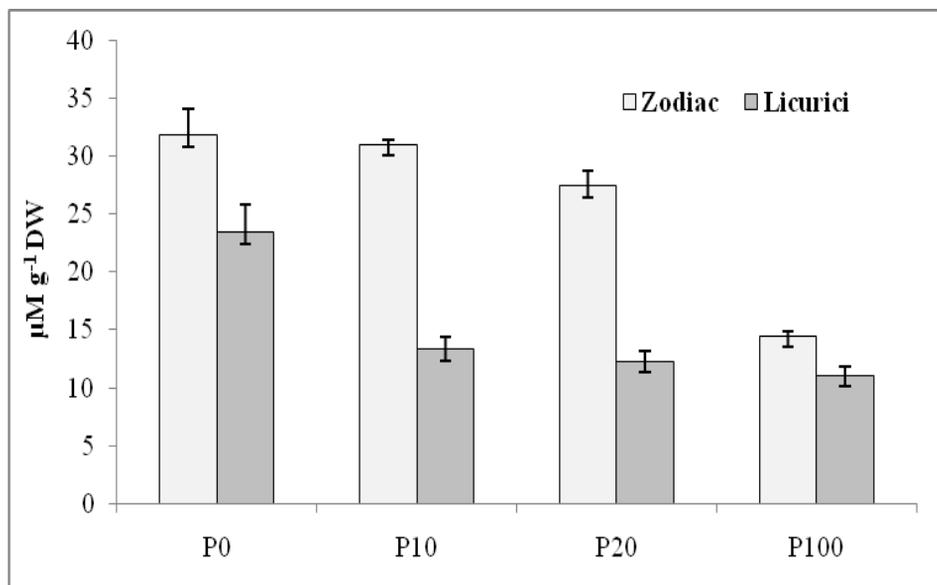
Water stress and/or P treatments caused significant changes in the content of free amino acids in the two soybean genotypes (fig. 7 and fig. 8). In this study, the concentration of free amino acids decreased in direct proportion with the levels of P application only under limited moisture conditions. Also, plants subjected to drought stress allocated more amino acids in their leaves than well-watered counterparts (fig. 7).



**Fig. 7** The effect of phosphorus supply on the concentrations of total free amino acids in leaves of soybean cultivars at the flowering stage under normal soil moisture (70% WHC). Data are means of four replicates  $\pm$  SE

Several explanations for the accumulation of free amino acids under stress have been suggested. These effects could be due to stimulated synthesis, impaired protein synthesis and/or enhanced protein degradation. This accumulation may play an important role in the osmotic adjustment of plants under stress as has been reported by Khavari-Nejad *et al.* (2008) and by Schubert *et al.* (1995). The P supplemental nutrition induced decreases of this physiological parameter in leaves in both cultivars under water stress conditions. Experimental data has shown that the application of P at high level (P100) decreased their concentration in leaves approximately by 2 times in plants of Licurici and Zodiac subjected to water deficit. We suggest that adequate P nutrition provided better conditions for nitrogen assimilation inclusive protein synthesis. Nevertheless, under drought stress the accumulation of these N-compounds in leaves was more evident in Zodiac than in Licurici, especially under insufficient nutrient supply. Hence, it was observed that supplemental P nutrition of plants subjected to drought decreased amino acids concentration in leaves and the lowest level (12-15  $\mu\text{M g}^{-1}$  DW) was observed in treatment with adequate P nutrition (fig. 8). No effect of P treatment was apparent on amino acid concentrations in leaves tissues of Licurici plants provided with moderate (P20) and high rate (P100) fertilization under suboptimal water regime. Altogether, adequate P nutrition increased leaf area development which in turn may improve plant photosynthesis and stress tolerance (Devi and Sinclair, 2013). The mechanisms of plant stress tolerance involve a

number of metabolic changes of cellular compounds, gene expression and protein modification.



**Fig. 8** The effect of phosphorus supply on concentrations of total free amino acids in leaves of soybean cultivars at the flowering stage under water deficit conditions (35% WHC). Data are means of four replicates  $\pm$  SE

Understanding the regulation of these processes will be essential to developing crops with higher tolerance to multiple stresses in particular to drought and nutrient deficiency which necessitates further investigations of the complex responses and biochemical pathways of plants exposed to unfavorable conditions.

## CONCLUSIONS

The phosphorus fertilization and soil moisture regime have significant influence on leaf development and amino acids content in two soybean cultivars.

Increased P rates increased the leaf area of both species, however, cv Zodiac displayed higher responses and it seems to be more susceptible to low P conditions than Licurici cultivar mainly due to poor leaf area development and higher free amino acids concentration.

Specific leaf area was not significantly affected by P treatment at flowering stage. Field studies are required to confirm the potential utility of P supplementation under suboptimal moisture conditions.

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## STUDY ON WOOD WASTE MATERIALS' CAPACITY TO RETAIN HEAVY METAL IONS FROM AQUEOUS SOLUTIONS, UNDER DYNAMIC CONDITIONS

### STUDIUL ASUPRA CAPACITĂȚII DEȘEURILOR LEMNOASE DE A REȚINE IONII METALELOR GRELE DIN SOLUȚII APOASE, ÎN CONDIȚII DINAMICE

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**Abstract.** Wood residues are commonly used in the furniture industry and in agriculture, as composts' ingredients or in different techniques as absorbents, having a large specific surface and a chemical composition appropriate for achieving adsorption processes. As a result of previous experiments it is found that the activation by boiling in acidic solutions of sawdust, in addition to increasing the adsorption capacity, it does not add any color to the treated solutions. In the present study we tested the retention capacity of two heavy metal ions (Cu and Co) in solutions of different concentrations, on sawdust processed by grinding, sieving and activation in hydrochloric acid. As main results, copper ions were retained between 63.2 – 91.16% and cobalt ions, between 79.9 – 91.16%, considering the initial concentrations of the solutions.

**Key words:** sawdust, heavy metals, pollution, water

**Rezumat.** Deșeurile lemnoase sunt de obicei folosite în industria mobilei și în agricultură, în compoziția composturilor sau în diverse procedee tehnice ca absorbanți, având o suprafață specifică mare și o compoziție chimică adecvată realizării proceselor de adsorbție. În urma unor experimente anterioare, s-a constatat că prin activarea la fierbere în soluții acide a rumegușului, pe lângă creșterea capacității de adsorbție, acesta nu imprimă nici o culoare soluțiilor tratate. În lucrarea de față s-a testat puterea de reținere a ionilor a două metale grele (Cu și Co) din soluții de concentrații diferite, pe material prelucrat prin măcinare, sitare și activare în acid clorhidric. Ca rezultat, ionii de cupru au fost reținuți între 63,2 – 91,16% și ionii de cobalt, între 79,9 – 91,16%, în funcție de concentrațiile inițiale ale soluțiilor.

**Cuvinte cheie:** rumeguș, metale grele, poluare, apă

## INTRODUCTION

Capitalization of bark and sawdust as ingredients for composting does not completely solve the problem of the large quantities of woodwaste, even if it is considered as a source of income for different enterprises in forestry domain.

Another way in which these wood waste materials are consumed is their use as absorbent material for different substances.

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Ion exchange is probably one of the major adsorption mechanisms for binding divalent metal ions to the maple sawdust. In a study conducted by Bin Y. and co-workers, they used batch techniques for the adsorption of copper on sawdust. The equilibrium adsorption level was determined to be a function of the solution pH, contact time and sorbate concentration (Yu *et al*, 2000).

The same researchers studied the equilibrium adsorption capacity of sawdust for lead, which was measured and extrapolated using linear Freundlich and Langmuir isotherms and compared with that for copper. Metal ions bounded to the sawdust could be stripped by acidic solution so that the sawdust can be recycled. They also conducted non-equilibrium experiments in glass columns. The results indicated the potentially practical value of this method in industries (Yu *et al*, 2001).

Sawdust and wood chips were also used to remove heavy metals from industrial and mining waste waters. Especially bivalent metals were retained on spruce wood chips – Cd up to 94%, Cu - up to 81% and Zn - up to 88% (Argun *et al*, 2008).

Šćiban and Klačnja tested the adsorption of two heavy metals on sawdust and concluded that the main adsorption sites for metal ions in wood are on lignin. Batch and column studies yielded almost equal ratio for the adsorption efficiencies of copper (II) and cadmium (II) (Šćiban and Klačnja, 2003).

Bulut and Tez tested the adsorption of lead, cadmium and nickel from aqueous solution by walnut sawdust, at different temperatures. They concluded that the adsorbent's selectivity order is  $Pb(II) \approx Cd(II) > Ni(II)$  (Bulut and Tez, 2007).

Another study performed on deciduous sawdust samples found that the kinetics of adsorption is relatively fast, reaching equilibrium in less than 20 min. The maximum adsorption capacity (7-8 mg/g of sawdust) was achieved at a pH between 3.5 and 5 for all the studied types of sawdust (Božić *et al*, 2009).

In this paper, we used alder sawdust processed by grinding, sieving and activation in hydrochloric acid as adsorbent material for different concentrations of copper and cobalt solutions.

## MATERIAL AND METHOD

The adsorbent material was alder sawdust, grinded and sieved in order to keep only the smaller particles. The variants used the sawdust activated by half hour boiling in hydrochloric acid 0.1 n, followed by filtration and pH correction to low acidic to neutral values. We chose the acid activated sawdust due to previous studies conducted on the adsorption of nitrates.

The heavy metal solutions had initial concentrations from 50 to 250 mg  $Cu^{2+}/L$  respectively from 50 to 250 mg  $Co^{2+}/L$ . For each variant, we used 0.5 g of adsorbent material per 50 ml of each solution, stirred continuously for 15 minutes, then filtered. We used spectrophotometric methods to determine the amount of metal ions in the filtrate of each variant.

## RESULTS AND DISCUSSIONS

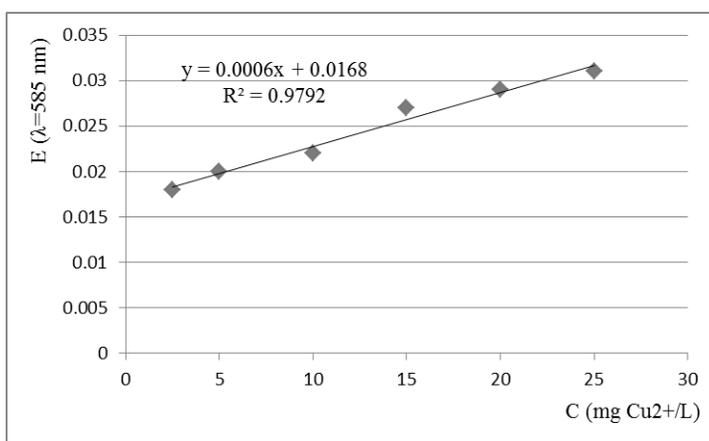
The considered adsorbent retained both metal ions from the aqueous solutions, the adsorption rate decreasing at higher concentrations of the heavy metal ions' solutions.

We presented the data regarding the calibration curves established for the two considered ions in table 1 and figures 1 and 2.

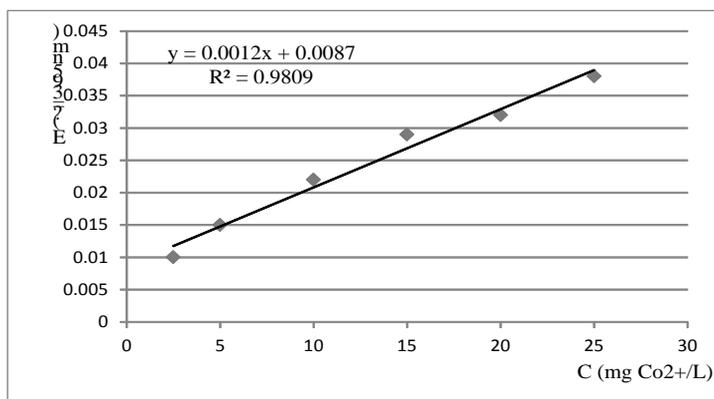
Table 1

**Calibration curves data for copper and cobalt ions**

Ion/E	Conc.	2.5	5	10	15	20	25
	mg/L						
Copper E <sub>(λ=585 nm)</sub>		0.018	0.02	0.022	0.027	0.029	0.031
Cobalt E <sub>(λ=395 nm)</sub>		0.01	0.015	0.022	0.029	0.032	0.038



**Fig. 1** Calibration curve for copper



**Fig. 2** Calibration curve for cobalt

For each heavy metal ion we calculated the adsorption coefficients related to the initial concentrations, establish the correlation between their values (tab. 2, fig. 3, fig. 4) and also determined the percentage of metal ions retained per one gram of adsorbent (tab. 2, fig. 5).

Table 2

Adsorption coefficient values

Ion	Conc.	50	100	150	200	250
		mg/L	mg/L	mg/L	mg/L	mg/L
Copper	Final concentration	5.33	18.667	38.667	53.667	92.0
	Adsorption coefficient	89.333	162.667	222.667	292.667	316.0
	% of retained ion	94.50	81.333	74.222	73.167	63.20
Cobalt	Final concentration	2.75	6.083	10.25	20.05	50.25
	Adsorption coefficient	94.5	187.833	279.5	359.5	399.5
	% of retained ion	94.50	93.917	93.167	89.875	79.90

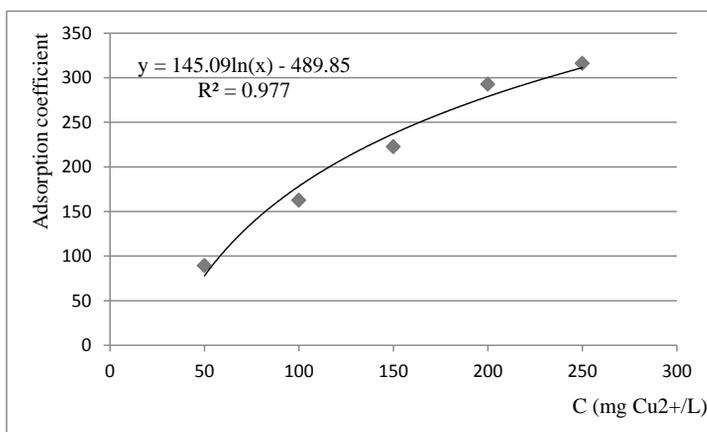


Fig. 3 Correlation between initial copper concentrations and adsorption coefficients

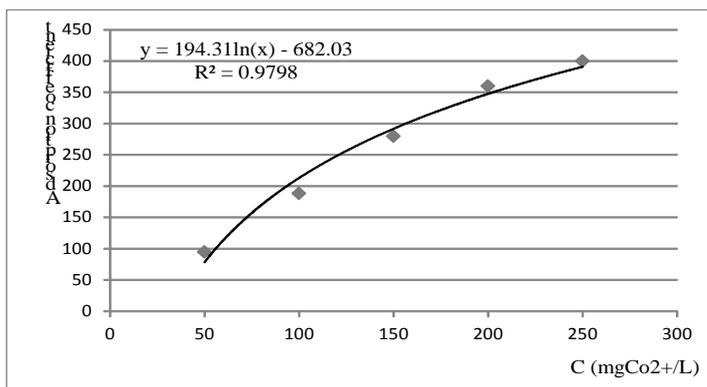
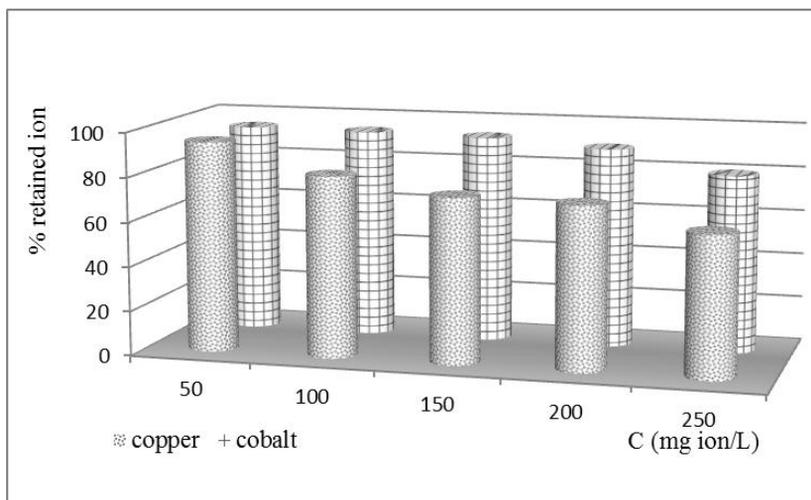


Fig. 4 Correlation between initial cobalt concentrations and adsorption coefficients



**Fig. 5** Percentages of heavy metal ions removed by acid activated sawdust

The activation process with acidic solutions does not require high concentrations and long reaction time, being available for large-scale applications. The actual absorption process is very simple and efficient; working with small samples, we preferred stirring the adsorbent with the tested solutions, but for larger quantities the process can develop in continuous flow, in columns, under minor pressure or statically in large chambers filled with adsorbent sawdust.

## CONCLUSIONS

1. Alder sawdust can serve as adsorbent material for divalent copper and cobalt ions from aqueous solutions;
2. The activation procedure, which proved to be increasing the adsorption capacity, is simple and quick;
3. Copper ions were adsorbed on sawdust in percentages from 94.5 (from 50 mg/L solution) to 63.2 (from 250 mg/L solution);
4. Cobalt ions were adsorbed on sawdust in percentages from 94.5 (from 50 mg/L solution) to 79.9 (from 250 mg/L solution);
5. The method proved effective for decreasing heavy metal ions levels in solutions and can develop in continuous flow or static conditions.

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## THE INFLUENCE OF SALT STRESS ON FOLIAR WATER CONTENT LEAVES OF SOME TOMATO CULTIVARS FROM N-E ROMANIA

### INFLUENȚA STRESULUI SALIN ASUPRA CONȚINUTULUI DE APĂ LA NIVEL FOLIAR A UNOR GENOTIPURI DE TOMATE DIN NORD-ESTUL ROMÂNIEI

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**Abstract:** Water is an important factor for the distribution of plant species on Earth, under stress leaves are the organs which are observed first symptoms. The research was conducted under greenhouse condition. The biological material was represented by ten local tomatoes populations collected from areas with saline soils from Moldavia region and compared with commercial type salt-tolerant tomato. Tomato genotypes in the study were subjected to salt stress for a period of 30 days is constantly wetted with saline solution to a concentration of 100 mM and 200 mM. There have been a number of quantitative investigations in the foliar ascertaining the fact that the genotypes subjected to salt stress is a reduction in the content of free water and to increase the amount of water bound thereby increasing the capacity of the biological tolerance of such local populations of tomatoes.

**Key words:** free water, bound water, tomato cultivars, salinity, stress

**Rezumat:** Apa este un factor important pentru repartiția speciilor vegetale pe glob, în condiții de stres frunzele sunt organele la nivelul cărora se observă primele simptome. Au fost luate în studiu 10 genotipuri de tomate colectate din solurile saline ale Moldovei și un soi comercial rezistent la salinitate. Acestea au fost expuse stresului salin pe o perioadă de 30 de zile, fiind udate constant cu soluții saline de concentrație 100 mM și 200 mM. S-au realizat o serie de investigații cantitative la nivelul aparatului foliar constatându-se, faptul că la genotipurile supuse stresului salin are loc o reducere a conținutului de apă liberă și o creștere a cantității de apă legată mărindu-se capacitatea biologică de toleranță a acestor populații locale de tomate.

**Cuvinte cheie:** apă liberă, apă legată, genotipuri de tomate, salinitate, stres

## INTRODUCTION

High salinity is the most widespread abiotic stress and constitutes the most stringent factor in limiting plant distribution and productivity (Iqbal and Ashraf, 2005; Yıldırım *et al.*, 2009; Qin, 2010). The main negative effects of high salinity that influence plant growth and development are photosynthesis inhibition water deficit, ion toxicity

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associated with excessive  $Cl^-$  and  $Na^+$ , interference with nutrition leading to nutrient imbalance (Qin, 2010). Water is the environment for conducting biochemical reactions, solutes participating in circulation, regulate temperature, maintaining and increasing plant turgidity (Ciobanu and Șumălan, 2009; Șerbănescu-Ionescu, 2012; Ivan, 2015). Given the vital functions that it performs water in living organisms, knowledge of plant water regime of economic interest has great practical significance.

The amount of water in the plant body in a state of active life ranging from 60-90 % depending on the internal factors (species, organ, tissue, developmental stage) and external factors (atmospheric humidity and soil, temperature, air movements), reducing generally the advance older plants (Trifu and Bărbat, 1997).

Water absorption is intense when the soil solution is hypotonic to the cell vacuole absorbent juice. The usual concentration of soil solution is 0.5-1.5 % the plants grow and develop normally. Soil solution becomes more concentrated in drought conditions, according to the excessive application of chemical fertilizers in arid and salty soils where (Heller *et al.*, 1989).

Free water and bound water is liquid water fractions of a particular biological importance, having functional role in different periods of life of the organism. In most cases predominate the free water. In dry conditions as in winter it decreases the amount of free water and the bound water increases, which results in a higher resistance of the plant. From the quantitative point of view, the ratio of the two kinds of water in all cases is in the favor of free water (Sand, 2001).

## MATERIAL AND METHOD

The research was conducted under greenhouse condition from USAMV Iași during the years 2014-2015.

The biological material was represented by local tomatoes populations collected from areas with saline soils from Moldavia region and compared with commercial type salt-tolerant tomato (*Ursula F<sub>1</sub>*) from Israel.

The bifactorial experience was conducted in a pots experiment in randomized blocks with four repetitions. Ten tomato genotypes (*Moșna 2*, *Șcheia 1*, *Dorohoi 6*, *Dorohoi 8*, *Copălău 2*, *Copălău 3*, *Copălău 4*, *Copălău 5*, *Dorohoi 4*, *Moșna 3*) studied were subjected to salt stress for a period of 30 days is constantly wetted with saline solution to a concentration of 100 mM and 200 mM.

Water lost after 24 hours, the relative percentage of the initial weight of the plant material is considered free water. The same plant material is then dried in an oven at a temperature of 105 °C, up to constant weight. The amount of water lost through the drying oven is regarded as bound water. The amount of free water and is linked to the total water.

## RESULTS AND DISCUSSIONS

Analysis of total water and forms was determined after 30 days to the salt stress. After 30 days there is observed an increase in the total water content for all genotypes at the three variants, it oscillating between 76.2 % and 91.2 %. We highlight that after this time the saline treatment application, the total water content is maintained close to that

of control version, which indicates that these genotypes have adapted to osmotic stress. Also percentage values of total water content of local cultivars are quite close to those of cultivar and salinity resistant *Ursula*  $F_1$  (fig.1.)

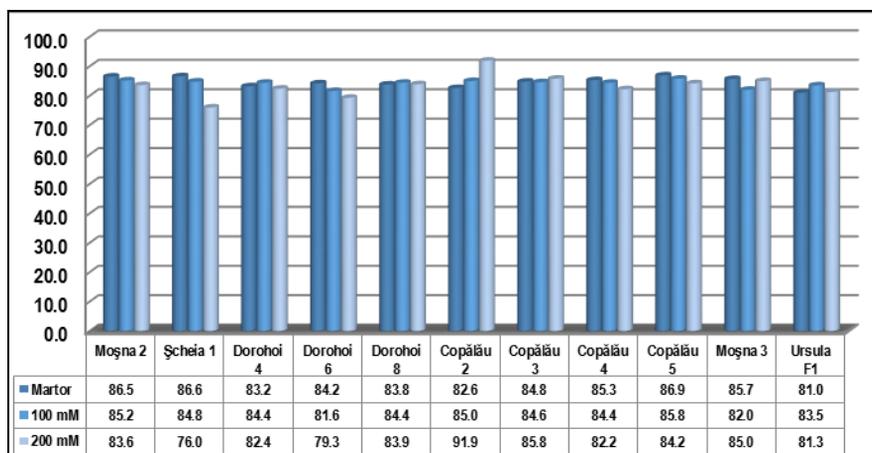


Fig. 1 Effect of salt stress on the total water content % after 30 days

Statistical analysis showed that after 30 days of treatment application saline genotype and saline concentrations differ statistically insignificant, this can be explained by the fact that the percentage values of variants 100 mM and 200 mM are very close to those of version control. Genotypes were adapted well from this point of view salt stress conditions (tab. 1)

Table 1.

**The variance analysis of the tomato genotypes under stress with NaCl for a period of 30 days on the total wather content**

Variance source	SP	GL	MS	F	P-value	F crit	Influence
Genotype	71.03	10	7.10363	1.01584	0.46427	2.3478	NS
Concentration	10.63	2	5.31848	0.76056	0.48045	3.4928	NS
Error	139.8	20	6.99281				
Total	221.5	32					

**Anova Two- Factor:** <sup>NS</sup> insignificant statistical differences ( $p \geq 0.05$ ); \* significant statistical differences ( $p \leq 0.05$ ); \*\* distinctly significant statistical differences ( $p \leq 0.01$ ); \*\*\* highly significant statistical differences ( $p \leq 0.001$ ),  $F > F$  crit null hypothesis rejected

Analysis of water content free after 30 days of treatment application saline shown in figure 2, highlights a lower free water compared to the control for all genotypes variant 100 mM and variant 200 mM water free, which shows, according to data presented in the literature (Jensen et Collins, 1985; Șumălan, 2009) that reduce free water content increases the capacity of biological tolerance of plants to abiotic stress conditions. Of the all genotypes on the 100 mM solution cultivar *Moșna* are registered a 1.1% difference compared to the control and

genotypes on the 200 mM variant cultivar *Copălău*<sub>4</sub> showed a difference of 2.6%. This shows a good ability for this genotypes to adapt on the salt stress.

Under stress, the plant vital activity is reduced, there is an increase in the amount of bound water, thereby ensuring the survival in such conditions. This was seen with cultivars studied in this experiment.

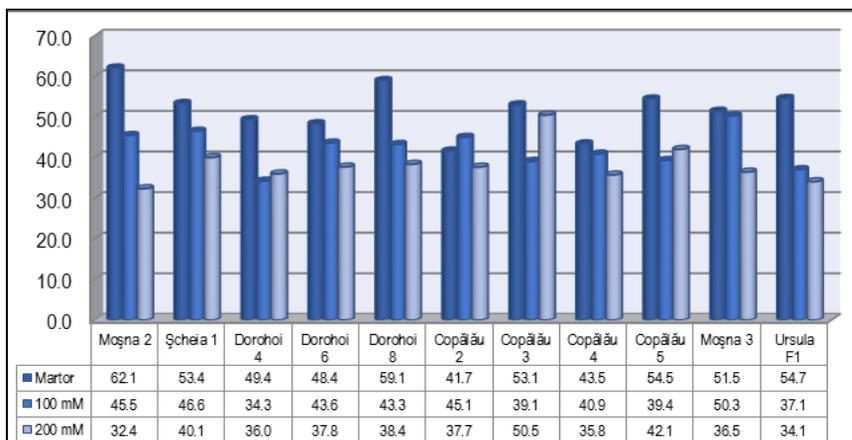


Fig. 2 Effect of salt stress on the free water content % after 30 days

After 30 days of exposure to stressful conditions values within 100 mM and 200 mM variants increase significantly compared to the control (fig. 3), which shows resistance to salinity genotypes studied.

The cultivars *Moșna*<sub>3</sub>, *Copălău*<sub>2</sub> and *Moșna*<sub>2</sub> shows a higher degree of adjustment due to the large amount of bound water, treated under the conditions of a high concentration salt stress (200 mM).

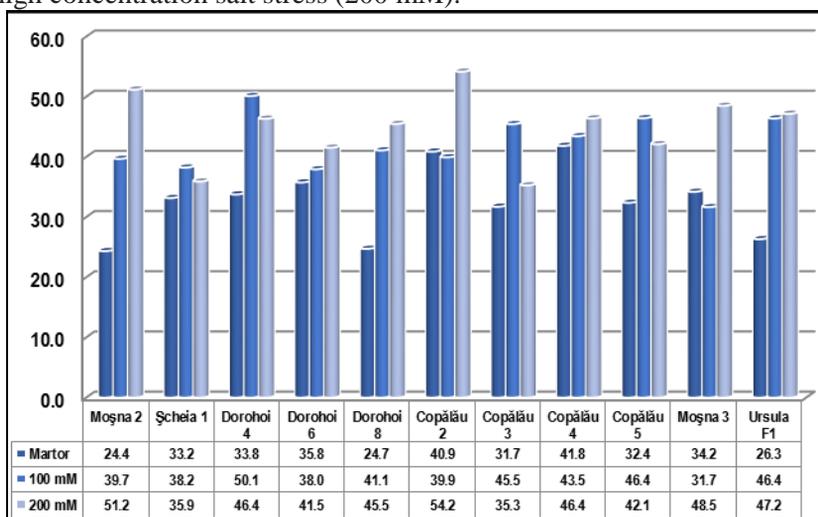


Fig. 3 Effect of salt stress on the bound water content % after 30 days

Statistical analysis shows that after 30 days concentration of salt has a lot of influence significantly the content of bound water to local populations tomato studied, which highlights the adaptive high that presents these genotypes in conditions of prolonged stress saline (table 2).

Table 2

The variance analysis of the tomato genotypes under stress with NaCl on the bound wather content

Source of variance	SP	GL	MS	F	P-value	F crit	Influence
Genotype	277.999	10	27.7999	0.83256	0.60404	2.3478	NS
Concentration	897.133	2	448.566	13.4337	0.0002	3.4928	***
Error	667.818	20	33.3909				
Total	1842.95	32					

Anova Two- Factor: <sup>NS</sup> insignificant statistical differences ( $p \geq 0.05$ ); \* significant statistical differences ( $p \leq 0.05$ ); \*\* distinctly significant statistical differences ( $p \leq 0.01$ ); \*\*\* highly significant statistical differences ( $p \leq 0.001$ ),  $F > F$  crit null hypothesis rejected

The ratio of free and bound water plant ensures survival in stressful conditions such as frost, drought, salinity. In this context, the free water content decreases, and the content of bound water increases the biological resistance to dehydration (fig. 4).

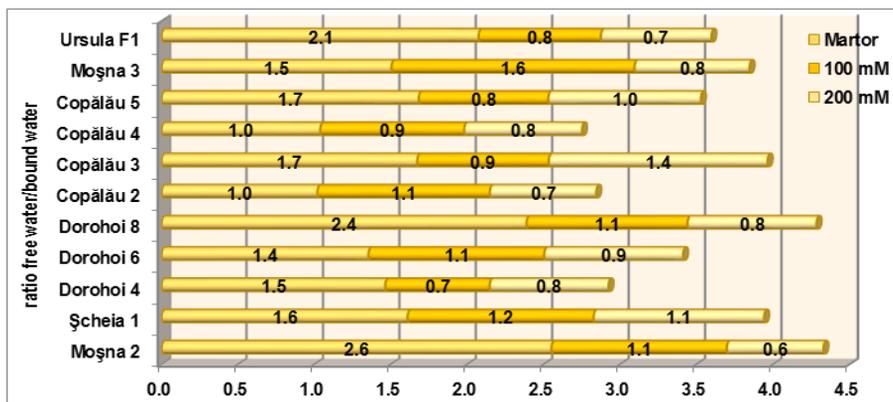


Fig. 4 Effect of salt stress on the report free water/bound water after 30 days

## CONCLUSIONS

1. On the genotypes of variant 100 mM and 200 mM it reduces of the content of free water, increasing the ability of biological tolerance.
2. The genotypes *Moșna 3*, *Copălău 4* and *Moșna 2* it has a higher degree of adjustment because of the large bound water, treated under the conditions of a high concentration salt stress (200 mM).
3. The ratio of free and bound water plant ensures survival in stressful conditions as salinity.

**Acknowledgement:** *This work was supported by a grant of the Romanian National Authority for Scientific Research, CNDI-UEFISCDI, project number PN-II-PT-PCCA-2011-3.1-0965.*

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## EXPERIMENTAL RESULTS IN THE OBTAINING OF NEW PERSPECTIVE LINE IN TOMATO

### REZULTATE EXPERIMENTALE ÎN OBTINEREA UNEI POPULAȚII NOI DE TOMATE

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**Abstract.** *The present paper is focused on the main morpho-physiological characteristics of a new tomato line, suitable for large scale cultivation and raw consumption, obtained at the Institute of Genetics, Physiology and Plant Protection of the Academy of Sciences from Moldavia. The newly obtained line, which will be forwarded for omologation, was studied in a CPCT with 10 perspective lines obtained in the tomato improvement programe, during 2010-2012, then in a CC along with 2 cultivars from The Republic of Moldavia as controls, during 2013-2015. The research highlights the newly created line – L-28 – which complements the tomato assortment cultivated in the Republic of Moldova.*

**Key words:** *Lycopersicon esculentum* L., cultivar, Comparative Perspective Crops Trial (CPCT), Competitive Crop (CC).

**Rezumat.** *Lucrarea de față se axează pe prezentarea principalelor caracteristici morfo-fiziologice a unei linii noi de tomate pentru industrializare și consum în stare proaspătă obținută la Institutul de Genetică, Fiziologie și Protecția Plantelor din cadrul Academiei de Științe a Moldovei. Linia nou obținută, ce va fi propusă spre omologare, a fost studiată într-o CCO cu zece linii de perspectivă obținute în programul de ameliorare a tomatelor, în perioada 2010-2012, apoi într-o CC cu două soiuri cultivate în Republica Moldova ca martori, în perioada 2013-2015. Cercețările au evidențiat linia nou creată – L-28, linie ce completează sortimentul de tomate cultivat în Republica Moldova.*

**Cuvinte cheie:** *Lycopersicon esculentum* L., cultivar, Cultură Comparativă de orientare (CCO), Cultură de Concurs (CC).

## INTRODUCTION

Tomato crop is one of the most important of the vegetable plants spectrum, as demonstrated by the cultivated area of about 3.2 million hectares and a delivery of about 95 million tons. Also, in terms of food, tomatoes are products with a high nutraceutical value, based on a sugar content of 4-6%, 0.5-0.6% ash and vitamins, of which 4.5-8 mg / 100 g provitamin A (Patron, 1992; Munteanu, 2003).

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Tomato fruits are preferred by consumers due to the multiple uses and accessible price. Also, they are considered profitable economic crops by growers.

In the practice of an efficient technology that ensure competitiveness, an important role is to choose a valuable cultivar (Munteanu, 2000). Knowing this, the Institute of Genetics, Physiology and Plant Protection of the Academy of Sciences from Moldavia, a high priority should be given to tomatoes breeding, in particular, those with determinated growth for industrial processing. The new achievements shall correspond to market requirements and have a high level of novelty.

In this regard, we improve our program aimed at obtaining cultivars for processing and consumption of fresh yellowish-orange fruits, less known vegetable market in the Republic of Moldova.

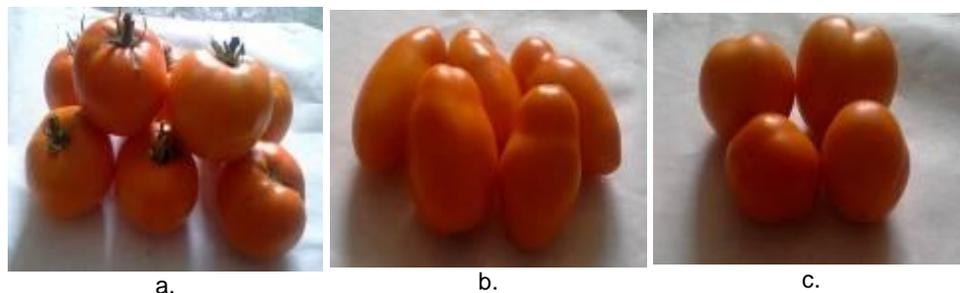
The objectives of the research reported in this paper were (1) evaluation of biological material culture in a comparative perspective crops trial (CPCT) and study of biological material promoted in CPCT (2) in a competitive crop (CC).

## MATERIAL AND METHOD

The research was carried out at the Institute of Genetics, Physiology and Plant Protection Academy of Sciences from Chisinau, during 2010-2015, in the laboratory of Genetics, Physiology and Plant resistance.

CPCT consists of ten perspective lines achieved in tomato breeding program in 2010-2012.

CC was accomplished with one of the lines promoted in CPCT (L-28) and two cultivars grown in Moldova as controls (Luci and Slava Moldavii), 2013-2015 (fig. 1).



**Fig. 1** Tomato cultivars in CC  
a. Line L-28; b. Luci cultivar; c. Slava Moldavii cultivar

CPCT and CC evaluations were made based on observations and biometric indicators on the main morphological, physiological and agronomical features.

Cultivation technology in both fields was the recommended literature (Atherton and Rudich, 1994; Chaux and Foury, 1994; Munteanu, 2003). Crop establishment was carried out by the seeding product in open field beds (about 500 seeds/m<sup>2</sup>), sown on 18-20 of April. The seedlings of 30 days old, having 3-4 true leaf stage were planted in the field at 20-25 May, following planting scheme 70 x 30 cm, with a density of approximately 50,000 plants/ha.

## RESULTS AND DISCUSSIONS

Our results regarding lines from CPCT revealed high diversity of evaluated biological material (tab. 1).

Table 1

The morphological characterization of the CPCT lines

Nr. crt.	Line	Type of growth	Vigour	Foliage colour	The joint peduncle	The fruit						
						Size	The h / d ratio	Fruit shape	Number of vacuoles	Green spot in the peduncular area	Color before maturity	Color at maturity
1.	L-28	*	medium-high	green	articulated	medium-high	1,1	spherical-high	4	green spot (unstressed)	green	yellow-orange
2.	L-04	*	medium-high	green	inarticulated	medium-high	1,2	ovoid	3-4	spotless	dark green	red
3.	L-14	*	medium-high	dark green	articulated	medium-high	1,1	spherical-high	3-4	spotless	dark green	dark red
4.	L-23	*	medium-high	dark green	inarticulated	medium-high	1,2	rectangular	2	spotless	dark green	light red
5.	L-48	**	medium-high	light green	inarticulated	medium-high	1,3	pear-shaped	2	spotless	light green	light red
6.	L-58	*	medium	green	inarticulated	medium	0,9	spherical	3-4	green spot	green	dark red
7.	L-63	*	medium	dark green potato	inarticulated	medium	0,9	spherical	4	green spot	dark green	pink
8.	L-77	*	medium-high	green	inarticulated	medium	1,2	ovoid	3-4	spotless	green	red
9.	L-83	*	medium-high	dark green	articulated	medium	1,3	pear-shaped	2-3	green spot	dark green	red
10.	L-95	*	medium	light green	articulated	medium	1,0	spherical	4	spotless	light green	red

\* – determinated; \*\* – undetermined

Growth type of increase shows that nine of the ten studied lines of have determinated growth pattern, which shows that the line of research is directed to cultivars intended for cultivation in open field for processing, but also to fresh consumption.

Vigour of the plants is variable, four lines having a medium to medium-to a medium-high. Three lines have a medium vigour (L-58, L-63 and L-95) and high-medium are the other seven lines (L-28, L-04, L-14, L-23, L-48, L-77, L-83).

Foliage color is green of different shades: dark green - four lines, light green - two lines and green-usual - four lines.

The joint peduncle is particularly important, introduced relatively recently (20-30 years) in the world assortment of tomatoes for processing. In our CPCT four lines have joint peduncle (original character, ancestral) and six ones have

jointless peduncle (mutant character, mutation).

The fruit is the most important organ for the estimation the agronomic characters cultivars of CPCT: size, ratio h/d (height/diameter), shape, number of vacuoles, green spot in the peduncular area, color before maturity and color at maturity.

In terms of size, the assortment comprises five lines with fruits medium-large (L-28, L-04, L-14, L-23 and L48) and five fruit average (L-58, L-63, L-77, L-83 and L-95). The h / d ratio and fruit shape are mutually correlated characters. Considering these characters, lines from assortment have fruits of the following shapes: spherical (3) spherical-high (2), ovoid (2) pear-shaped (2), rectangular (1).

Green character at the peduncular area divides the range into two groups: green spot (4) and spotless (6).

Fruit color is green until normal physiological maturity it is green - three lines, dark green - five lines and light green - two lines. When they have reached physiological maturity, the fruits consolidate a number of features, but the color is more noticeable. Most lines are red of different shades - eight lines, one is yellow-orange and one is pink.

Regarding the physiological characterization (tab. 2), this refers to the specific period of three distinct phenophases: blooming, first fruit ripening (start ripening fruit) and fruit ripening last (end of the growing season).

Table 2

**Physiological characterization of lines in CPCT**  
(number of days in the changes of phenological events, from the East )

Nr. crt.	Line	Blooming	Maturation of the first fruit	Maturation of the last fruit
1.	L-28	57	125	170
2.	L-04	58	130	176
3.	L-14	55	126	187
4.	L-23	48	120	178
5.	L-48	53	123	179
6.	L-58	55	109	193
7.	L-63	48	116	163
8.	L-77	43	106	186
9.	L-83	53	134	178
10.	L-95	44	90	165

The period from plant raising to bloom lasts under CPCT from 43 days (L-77) to 58 days (L-04). The importance of this characteristic is that it is directly involved in earliness of cultivars. The first fruit matures in about 90 days after emergence, L-95 is the earliest line in the assortment. The delayed lines are L-04 and L-83, the first fruit maturation being carried out for 130 or 134 days after emergence.

The ripening of the final fruit provides the information on the length of the ripening period and the length of the growing season. Time of ripening lasts between 45 to 84 days, and the growing season lasts between 163 and 193 days.

A great variability may be noted, with large limits: fruit maturation period has an amplitude of 39 days (84-45 days), and the vegetation has an amplitude of 30 days (193-163 days). These features indicate that the studied lines are intended both for processing and for fresh consumption.

During 2013-2015, the new created line L-28, considered for perspective, was compared with two varieties grown in The Republic of Moldova, Luci and Slava Moldovii considered as control in the culture of competition (CC). These control were chosen because they have similar color and fruit are part of the same group of precocious line proposed for plant introduction (tab. 3, tab. 4).

Table 3

## Morphological characteristics in CC

Nr. crt.	Cultivar	Type of growth	Vigour	Foliage colour	The joint peduncle	The fruit						
						Size	The h / d ratio	Fruit shape	Number of vacuoles	Green spot in the peduncular area	Color before maturity	Color at maturity
1.	L-28	*	medium-high	green	articulated	medium-high	1,1	spheric-high	4	green spot (unstressed)	green	orange-yellow
2.	Luci	*	medium	green	inarticulated	medium	1,3	elongated	3	spotless	green	yellow
3.	Slava Moldovii	*	medium	green	inarticulated	medium	1,0	spheric	4	green spot	green	yellow

Distinguishing features of the line L-28 are: plant vigor is medium-high, higher than of the control ones, the fruits have joint peduncle, size being medium to high, shape is spherical-high, the green spot is present, but unstressed and the fruit is an orange-yellow colour.

Regarding the physiological characteristics (Table 4) of the CC cultivars we can see that all belong to the same group of precocity - semilate, first fruits ripening during the period from 2 to 8 August, after 105-111 days from germination. The growing season is between 165 and 170 days, and maturation period between 59 and 61 days.

Table 4

## Physiological characterization in CC

Nr. crt.	Cultivar	Blooming		The maturation of the first fruit		The maturation of the last fruit	
		date	n.z.	date	n.z.	date	n.z.
1.	L-28	15 VI	57	08 VIII	111	6 X	170
2.	Luci	13 VI	55	04 VIII	107	4 X	168
3.	Slava Moldovii	08 VI	50	02 VIII	105	01 X	165

\*n.z. – number of days from the East

Also, agroproductivity ability was evaluated expressed by major quantitative characters: the average number of fruits per plant (NFP), average fruit weight (AFW) and average fruit weight per plant (AFWP) (tab. 5).

Table 5

#### Agro-productive capacity in CC

Nr. crt.	Cultivar	NFP	AFW (g.)	AFWP (g.)
1.	L-28	10-12	190-210	2100
2.	Luci	14-16	90-110	1500
3.	Slava Moldovii	13-14	120-130	1700

In line L-28 NFP was lower versus control in the study (10-12 fruits, compared with 13-14 or 14-16 fruit per plant), but AFW was superior, with L-28 values between 190-210 g compared to 120-130, respectively, 90-110 g, the control, while AFWP also registered higher values (2100 g/plant), in comparison with controls (1700 g or 1500 g).

### CONCLUSIONS

Research in a comparative perspective crop trial and a culture of competition highlighted the perspective line L-28 that complements the assortment of tomatoes grown in Moldova;

Line L-28 is characterized by medium-high vigour, medium-large fruit size, spherical-tall yellow-orange with a production of 2100 g/plant.

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## EVALUATING HAIL STORM DAMAGE TO CORN PRODUCTIVITY

### EVALUAREA DAUNELOR PROVOCATE DE FURTUNI ÎNSOȚITE DE GRINDINĂ ASUPRA PRODUCTIVITĂȚII PORUMBULUI

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**Abstract.** In Romania, more than half of hail storm are in the period from May to July. They are responsible for small losses of productions they occur in the early stages of vegetation. Significant losses occur when hail storm occur during flowering-silking period. James V. Vorst appreciates that monetary losses caused by hail storm reach \$ 52 million annually in the USA. Starting from this consideration, but also in terms of attracting attention on providing corn crops, in the spring of 2015 we established a two-way experience, in Lovrin, Timisoara. The first experimental factor, corn cultivar has been represented by 18 maize hybrids of FAO 350-550 maturity groups. The second factor was the degree of defoliation on plants corn produced artificially in various stages of vegetation. Results indicate losses of up to 5% when maize has six leaves, 1-10% when corn has 10 leaves, of 9-62% during flowering-silking and 6-41% when milk line was distinguishable.

**Key words:** hail storm, enable recovery, corn, productivity

**Rezumat.** În România, peste jumătate din ploile însoțite de grindină sunt în perioada Mai – Iulie. Acestea sunt responsabile de pierderi de producții mici când apar în primele faze de vegetație. Pierderile semnificative apar atunci când ploile însoțite de grindină apar în perioada înflorit-mătăsit. James V. Vorst aprecia că pierderile bănești provocate de ploile cu grindină ajung la \$52 milioane dolari anual numai în SUA. Plecând de la acest considerent, dar și din prisma atragerii atenției asupra asigurării culturilor, în primavara anului 2015 am înființat o experiență bifactorială, în localitatea Lovrin, Timișoara. Cultivarul de porumb reprezentat de 18 hibrizi de porumb din grupele de maturitate FAO 350-550 au reprezentat primul factor. Al doilea factor a fost reprezentat de gradul de defoliere produs artificial asupra plantelor de porumb, în diferite stadii de vegetație. Rezultatele indică pierderi de până la 5% când porumbul are 6 frunze, de 1-10% când porumbul are 10 frunze, de 9-62% în perioada înflorit-mătăsit și de 6-41% când se poate distinge linia de lapte.

**Cuvinte cheie:** grindină, capacitate refacere, porumb, producție

## INTRODUCTION

Conform to Klein and Shapiro, 2011, corn plants are not affected or less affected by hail storm just after vegetative emergence (VE). At VE stage, the

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growing point is still in the soil and remain there for at least 3 weeks (up to 5-7 fully visible leaves, V5-V7). About 3 weeks after emergence, all nodes and internodes plant are developed; growing point is above ground due internodes elongation. In the next 4-5 weeks, the growth rate is faster and also increasing the vulnerability of plants when rain with hail (Lauer, 2014).

During silky-flowered when developing reproductive organs occurs, rain hail can destroy in whole culture, the risk is 100% when the tassel is broken (Mangen and Peter, 2001; Lee, 2007). After pollination has occurred, risk of loss of production decreases as the plants approaching physiological maturity (Thomison, 2013). After physiological maturity plants begin to lose water of the grain, the risk is very low, except torrential rains accompanied by hail very large ear that can lead to detachment from the plant (Nielsen *et al.*, 2015).

According to Hicks and Naeve, 2009, in Minnesota more that \$ 10 million is spent annually to insure crops against heavy hail. Losses farmers varies from year to year, but insurers pay \$ 6 million in compensation for farmers who losses. The effect of hail not only affects production in a certain percentage, but may even reach the decision to sow again if it is required.

## MATERIAL AND METHOD

Hybrids analyzed are from group of maturity FAO 260-510. Of these, 15 hybrids are dent grain and 3 are flint grain. At the 15% moisture were harvested 10 cobs of each variant for analyzing biometric cob (ear length, no of rows/ear and ear diameter). The yield values has been evaluated at 14% moisture in t/ha.

For each of the studies hybrids were scored for the number of days from vegetative emergence to flowering – silking and number of days from vegetative emergence to milk line apparition.

The experimental protocol on ice storm simulated on corn it was as follow:

1. Transitions field at the each V stage of developement to simulate a ice storm.  
Using a commercial cnife, we exfoliated all leaves visible at that moment.
2. For each variant we made the same protocol, trying to simulate the real ice storm.
3. The experimental variants are ice storm simulated at:  
**V1** – 6 visible leaves (V6)  
**V2** – 10 visible leaves (V10)  
**V3** – flowering – silking time  
**V4** – milk line apparition.

## RESULTS AND DISCUSSIONS

Evaluating the results on the number of days from VE to flowering-silking, in V3 and V4, it was considered more logical treating them together (silking date has not varied in the case of these variants). Given this, we can confirm that heavy hail in vegetative phases lead to more activity of auxins stimulate cellular developement. Thus, until 6 leaves visible (V1), flowering-

silking date is up to 1.5 days earlier than in V2 and up to 3.5 days compared with V3 and V4.

Interesting results were obtained and concerning occurrence of milk line apparition. Thus, in embodiments where the hail storm was simulated at 6 leave visible, the hybrids were developed line of the milk to about 106 days from the VE, while to the control in this case (V4) was 118 days. The difference 12 days is the influence of heavy hail in the development of accelerated plant to reach physiological maturity. The overall conclusion in this case is that as the rains hail occur early during vegetative, the development time further shortens. The conclusion is confirmed by the results in variants V2 and V3, 108 days and 114 days, and also by Robertson *et al.*, 2011.

Regarding moisture at harvest time it is observed as no significant differences between variants V1, V2 and V3. Instead, rain accompanied by hail in the phase of developing the milk line, leading to faster loss of moisture of the grain, the average difference being about 7%.

Stem lignification in stalks may explain the plants lodging in V4, which is the percentage of plants fallen is 2.25%. However, the data is interesting in that the plants develop a sensitivity in 6-10 leaves phase (V1 and V2, where the percentage of lodging is 17% and 26%. Generative period corresponds to a downward trend of this factor, the registered values in V3 and V4.

We will not focus more on the cobs biometric indicators in time to harvest, but conclude that the differences between variants are not significant in terms of the number of rows of kernels/cob and its diameter. In terms of length ear, general trend is observed with a rain hail delayed.

Regarding the production capacity, the most important indicator to watch in this experience, we find that the rains accompanied by hail at 10 leaves visible (V2) and during flowering-silking (V3) are the most damaging (there is a risk of a production diminished when the tassel is full destroyed).

Direct proportion to the period of bloom is the production obtained in that shortening the vegetative lead to an extension of the generative period. Obviously, productions can be much lower compared to checks where such rain occurs during milk line apparition.

According to literature and especially the results obtained by Balas Baconschi *et al*, 2015, late hybrids exceed productions of early hybrids, and dent hybrids are more yielding than flint ones.

Table 1

Evaluating the silking – flowering date and milk line apparition from VE, harvest moisture and lodging plants at harvest time

Genotype	FAO	Silking date			Milk line (days from VE)				Harvest moisture (%)				Lodging plants (%)			
		V1	V2	V3-V4	V1	V2	V3	V4	V1	V2	V3	V4	V1	V2	V3	V4
Hyb1S	290	58	60	62	105	107	113	117	19,8	20,4	21,6	15,7	16,1	25,3	8,4	4,3
Hyb2E	350	59	61	64	107	108	115	119	20,4	20,2	20,8	12,9	16,7	25,9	7,6	1,5
Hyb3P	350	60	61	65	108	109	116	120	20,2	20	20,6	12,7	16,5	25,7	7,4	1,3
Hyb4L	380	61	62	64	107	110	115	119	20,8	20,6	21,2	13,3	17,1	26,3	8	1,9
Hyb5D	450	57	60	63	106	106	114	118	18,8	18,6	19,2	11,3	15,1	24,3	6	0,1
Hyb6O	430	58	61	62	105	107	113	117	20,7	20,5	21,1	13,2	17	26,2	7,9	1,8
Hyb7M	400	60	63	61	104	109	112	116	21,8	21,6	22,2	14,3	18,1	27,3	9	2,9
Hyb8SA	490	58	61	60	103	107	111	115	20,2	17,7	15,8	10,6	16,5	25,7	2,6	0,8
Hyb9M	400	59	63	60	103	108	111	115	22,1	21,9	22,5	14,6	18,4	27,6	9,3	3,2
Hyb10P	310	61	62	61	104	110	112	116	21,5	22,1	23,3	17,4	17,8	27	10,1	6
Hyb11K	350	59	63	63	106	108	114	118	21,8	21,6	22,2	14,3	18,1	27,3	9	2,9
Hyb12M	400	60	62	65	108	109	120	120	21,4	21,2	21,8	13,9	17,7	26,9	8,6	2,5
Hyb13O	400	63	63	66	109	112	117	121	22,2	22	22,6	14,7	18,5	27,7	9,4	3,3
Hyb14A	490	59	63	65	108	108	116	120	21,7	19,2	17,3	12,1	18	27,2	4,1	0,7
Hyb15L	510	58	61	67	110	107	118	122	20,7	18,2	16,3	11,1	17	26,2	3,1	0,3
Hyb16C	260	56	60	62	105	105	116	117	18,9	19,5	20,7	14,8	15,2	24,4	7,5	3,4
Hyb17E	350	60	60	64	107	109	115	119	19,1	18,9	19,5	11,6	15,4	24,6	6,3	0,2
Hyb18R	340	62	61	63	106	111	114	118	19	19,6	20,8	14,9	15,3	24,5	7,6	3,5

Table 2

Evaluating the yeild, ear length, no of orows/ear and ear diameter

Genotype	FAO	YIELD (q/ha)				EAR LENGTH (cm)				NO OF ROWS/EAR				EAR DIAMETER (cm)			
		V1	V2	V3	V4	V1	V2	V3	V4	V1	V2	V3	V4	V1	V2	V3	V4
Hyb1S	290	7,59	6,52	5,41	8,45	21	20,9	22,6	27,2	16	18	18	18	41	41,5	44,4	44,8
Hyb2E	350	7,78	6,68	6,04	8,1	18,2	21,8	23,5	28	16	18	18	18	44,7	44,7	47,5	47,5
Hyb3P	350	7,15	6,54	6,22	7,81	18	20,8	24,6	27,3	16	18	18	18	41,9	43,5	43,9	46,8
Hyb4L	380	8,1	7,21	6,84	8	18,6	21,4	25,2	29,4	16	20	20	22	45,1	45,2	45,7	46,9
Hyb5D	450	9,52	8,41	7,94	9,51	16,6	19	27,5	28,7	18	18	20	20	46,4	47	49,5	50,4
Hyb6O	430	8,14	7,34	6,81	8,68	18,5	22,5	27,4	29,7	18	18	20	20	43,7	43,6	47,6	48
Hyb7M	400	9,82	8,65	7,48	10,14	19,6	23,7	28,4	29,9	16	18	18	20	42,7	42,5	48	48,1
Hyb8SA	490	9,68	9,12	8,49	11,43	15,9	23,4	25,4	30,1	18	18	20	20	40,3	42,4	47,3	49,5
Hyb9M	400	8,72	7,81	7,19	8,4	19,9	22,8	26,5	30,4	16	18	18	20	44	44	47,2	47,2
Hyb10P	310	7,75	7,64	6,89	8,06	22,7	24,1	24,9	28,4	16	18	18	18	44,7	44,7	45,1	45
Hyb11K	350	5,11	4,12	3,78	5,5	19,6	24	26,8	28,6	16	18	18	18	43,8	44,5	44,6	46,7
Hyb12M	400	8,33	8,14	7,34	9,01	19,2	23,1	24,8	29,7	18	20	20	20	45,1	45,6	47,5	48,3
Hyb13O	400	8,64	7,87	7,02	9,11	20	23	26,7	31,2	18	20	22	22	41,4	42,5	42,9	46,8
Hyb14A	490	8,34	7,58	6,86	8,48	17,4	23,1	25,3	32,5	18	18	20	20	44,6	43,5	45,8	46,2
Hyb15L	510	11,3	9,48	8,79	12,7	16,4	24,4	24,9	33,8	18	18	20	20	45,7	46,5	49,8	51,2
Hyb16C	260	6,61	6,5	5,75	6,92	20,1	21,4	26,8	26,8	16	18	18	18	40,2	41,8	44,8	43,9
Hyb17E	350	8,61	7,51	6,87	8,93	16,9	24,5	27,1	29	16	18	18	18	42,8	45,2	45,7	46,8
Hyb18R	340	8,63	8,52	7,77	8,94	20,2	23,8	27,2	28,6	16	18	18	18	44,1	42,7	46,5	46,2

## CONCLUSIONS

1. In terms of the ice storm simulation, also in differentiating the genotypes the data obtained after one year of experimentation has been conclusive. The commercial hybrids tested reacted differently for yield, silking date and milk line apparition.

2. We can easily see that in V4 for yield are the best values. A ice storm at flowering – silking time is the worst for corn and we think that the results are not really with reality, because in our simulated we did not touch the panicle, male inflorescence, which can be destroyed 100% in a strong ice storm. We can easily differentiate the values in stage 6 and 10 visible leaves and we can conclude that the effect of ice storm in these situations are not very big influenced in yield.

3. Very interesting results are for moisture harvest and lodging plants. We scored that in V1 and V2 variants are the highest values for lodging plants; it means that the ice storm can induce a susceptibility for lodging and harvest moisture compare with other situations.

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## STUDY CASES REGARDING LANDSCAPING DESIGN IN FAMILY VEGETABLE GARDENS

### STUDII DE CAZ REFERITOARE LA FOLOSIREA DESIGN-ULUI PEISAGER ÎN CULTURA LEGUMELOR

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***Abstract.** The paper presents an analysis of Romanian household vegetable gardens, based on ornamental criteria and specific vegetable growing technologies characteristic to these plant displays in cultivation. The weak and strong points of the studied vegetable gardens were marked out through a SWOT analysis and based on it solutions were elaborated to improve them on an aesthetic, technologic and workable level in accordance with specialized literature. The obtained results offered unity, balance and an increasing aesthetic value to the plant display, participating in an active matter to the enlargement of their sustainability degree.*

**Key words:** Vegetable garden, plant display, sustainability

***Rezumat.** Lucrarea prezintă o analiză a grădinilor legumicole de tip familial de pe teritoriul țării noastre, realizată pe baza criteriilor de design ornamental și pe baza tehnologiei de cultură specifică acestor dispozitive de dispunere a plantelor în cultură.*

*Prin intermediul analizei de tip SWOT au fost analizate punctele forte ale grădinilor legumicole luate în studiu, dar și cele slabe și au fost elaborate soluții pentru îmbunătățirea acestora din punct de vedere estetic, practic și tehnologic conform literaturii de specialitate.*

*Rezultatele obținute au oferit unitate, echilibru și o valoare estetică mărită dispozitivelor de dispunere a plantelor în cultură, contribuind activ la creșterea gradului de sustenabilitate a acestora.*

**Cuvinte cheie:** grădină legumicolă, dispunerea plantelor, sustenabilitate

## INTRODUCTION

Nothing compares with the benefits that a garden can give you. Growing fruits, vegetables, flowers and herbs combines sport with the pleasure of working in fresh air, relaxing and admiring your work. Seeing your garden develop brings a smile on your face and an achievement of happiness, that only working in a garden can give you (Greenwood, 2008).

From a piker to a grower, man over time has passed many steps, helped by the evolution of crop technology, the large number of species in our days and accessibility to information.

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According to landscaping design specialists, the family vegetable garden, is an important element that must be designed in the same style as the main ensemble. The proposed species must be complementary with the main buildings, in the same stylistic lines, when it comes to color and shape (*habitus*). By applying landscape design rules the vegetable garden is given an ornamental character, with many functions (Sima, 2009; Iliescu, 2008).

As time passed family vegetable gardens have evolved due to the desire to grow more species and terms such as crop rotation are indispensable for a true grower. Stan *et al.* (1999, 2003) describes the importance that crop rotation has in family vegetable gardens due to its multiple advantages, reducing chemical use.

The garden “represents an ensemble of heterogenic elements bound by different relations between them, it’s a structure” (Constantinescu, 1992). Combining flower and crop technology with landscape design rules represents one of our objectives in designing sustainable ornamental vegetable gardens using an intercropping system.

The present study case takes into analysis three family vegetable gardens from different parts of the country, with the objective of increasing their ornamental value and sustainability, by applying the correct crop technology.

## MATERIAL AND METHOD

For achieving the purpose and objectives of this paper, three family vegetable gardens from Bistrița-Năsăud county and Cluj county were studied. Due to the less favorable location for vegetable growing in the process of designing the vegetable gardens, geological, climatic and cultural factors were taken into consideration by owners. The surface of the family vegetable gardens is from 100 to 260 m<sup>2</sup>.

The biological material used by the owners in the vegetable garden was represented by common and local species, such as tomatoes, peppers, white cabbage, carrots and many more.

Based on landscape design rules and vegetable growing technological criteria’s aspects such as fertilization, carrying works for plants and ornamental value were analyzed.

The family vegetable gardens were analyzed using SWOT analyses, which underlined the strengths and weakness of the studied gardens giving possibility to enhance the ornamental value by applying to correct technological plant requirements.

The applied working methods for evaluating the studied family gardens were case study, observation and SWOT analysis.

## RESULTS AND DISCUSSIONS

### a. Case study 1.

Based on technological plant requirements, landscape design rules and functionality of the family vegetable garden, using SWOT analysis (tab. 1) for the existing garden, solutions for enhancing the ornamental value of the first garden were elaborated.

SWOT analysis based on the existing garden in case study 1.

Strengths	Weaknesses
<ul style="list-style-type: none"> <li>• Many vegetable species</li> <li>• Bio vegetables</li> <li>• Usesown fertilizers</li> <li>• Crop rotation</li> <li>• Intercropping system</li> <li>• Using trap crops</li> <li>• Systematized garden</li> </ul>	<ul style="list-style-type: none"> <li>• Low ornamental value</li> <li>• Planting distances not respected</li> <li>• High degree of weed seeds in the soil due to neglect from the neighbours gardens</li> <li>• Leaving to many suckers on the tomato bush</li> </ul>
Opportunities	Threats
<ul style="list-style-type: none"> <li>• Appling an intercropping system – increasing sustainability of the garden</li> <li>• Increasing the activity of the soil microorganisms by using organic fertilizers</li> <li>• Increasing ornamental value by applying design rules</li> </ul>	<ul style="list-style-type: none"> <li>• Due to incorrect planting distances the possibility of disease attack is much higher</li> <li>• Small planting distances can clog caring works in the garden</li> <li>• The many sucker left on the tomatoes can reduce sun light and produce fruits much smaller than normal</li> </ul>

Due to its mixt style, the family vegetable garden, gives the opportunity to create different points of attraction, using different colors, volume and textures that the proposed species have, which will enhance the unity of the garden, unifying the three visual plans (fig. 1).



**Fig. 1** Perspective of the proposed solutions

The habitus of the species proposed in the vegetable garden, in an intercropping system, pepper + garlic, celery + leaf cabbage + garden nasturtium, lettuce + onion + lavender, creates a “game” of color, enhancing the ornamental value.

By applying an intercropping system that respects the technological requirements for vegetable species and design rules, the vegetable garden has new functionalities such as aesthetic and social ones.

Intercropping vegetables with different floral species, such as French marigold + tomatoes + lavender, can have a beneficial effect on their

development, a positive reaction, known as allelopathy, reducing the number of pests attack (fig. 2).



**Fig. 2** Details of the proposed solutions

**b. Case study 2.**

The second garden studied from a technological point of view, was better developed, with simple lines, realized in a geometrical style, with a small slope (3-4 %) that gives the opportunity to create different levels of perception, creating the opportunity to enhance its ornamental value and the impact that it has on the environment (tab. 2).

*Table 2*

**SWOT analysis in case study 2**

<b>Strengths</b>	<b>Weaknesses</b>
<ul style="list-style-type: none"> <li>• Vegetable garden watered using drip irrigation</li> <li>• Local varieties used in garden</li> <li>• Crop rotation</li> <li>• Organic fertilizer was used</li> <li>• The tomatoes were correctly trimmed</li> <li>• Systematized garden</li> <li>• Using successive crops</li> </ul>	<ul style="list-style-type: none"> <li>• Low ornamental value</li> <li>• Planting distances not respected</li> <li>• The usage of insecticides</li> <li>• No support for cucumbers</li> <li>• A high degree of weeds in the surrounding areas</li> </ul>
<b>Opportunities</b>	<b>Threats</b>
<ul style="list-style-type: none"> <li>• Increasing the ornamental value by intercropping vegetables with flowers, which can lead to a low degree of pest and disease attack</li> <li>• Increasing soil fertility by using an intercropping system</li> <li>• Increasing ornamental value by applying design rules</li> </ul>	<ul style="list-style-type: none"> <li>• A lower number of bees due to the usage of insecticides</li> <li>• Small planting distances can cause an inconsonant development</li> <li>• Due to incorrect planting distances the possibility of disease attack is much higher</li> </ul>

Based on the existing SWOT analysis by applying aesthetic and compositional principles and by using an intercropping system a compositional unity and a visual balance can be created. Intercropping vegetable species with flowers, tomato + purple basil + lettuce, creates harmony and decreases the number of diseases and pest in the vegetable garden (fig. 3).



**Fig. 3** Detail of the proposed design for the vegetable garden in case study 2.

### c. Case study 3.

Situated in the 3<sup>rd</sup> vegetable growing area in the country, which is less favorable for the development of vegetables, the family garden is systemized, shaped as a square (10 x10 m), surrounded by fruit trees, which actively contribute to the biodiversity of the area.

The vegetable family garden was well taken care of, based on plant requirements that assured a good development of the species that were present. From an ornamental point of view the studied vegetable garden was designed respecting general landscaping principals and functions, presenting potential for improvement of the aesthetic value (tab. 3.).

*Table 3*

#### SWOT analysis for the existing vegetable garden

Strengths	Weaknesses
<ul style="list-style-type: none"> <li>• Many local species used</li> <li>• Bio vegetables</li> <li>• No fertilizer used</li> <li>• Crop rotation</li> <li>• Systematized garden with a slight ornamental value</li> <li>• Snails and slugs eliminated using running Indian ducks</li> <li>• Correct planting distances</li> <li>• Well cared plants</li> </ul>	<ul style="list-style-type: none"> <li>• Low ornamental value</li> <li>• A high degree of weeds in the surrounding areas</li> </ul>
Opportunities	Threats
<ul style="list-style-type: none"> <li>• Applying an intercropping system – increasing sustainability of the garden</li> <li>• Increasing the activity of the soil microorganisms by using organic fertilizers</li> <li>• Increasing ornamental value by applying design rules</li> </ul>	<ul style="list-style-type: none"> <li>• In time the soil resources will decrease if no organic fertilizers are used</li> </ul>

By applying an intercropping system, the number of vegetables found in the garden will grow, the different habitus and color of the species will increase

the ornamental value, giving originality to the composition and a higher degree of sustainability for the garden (fig. 4).



**Fig. 4** Detail of proposed intercropping system

Introducing flowers and herbs in the garden will decrease the number of pest and disease attacks, increasing in the same time the ornamental value of the garden, providing educational and health functions to it.

The studied family vegetable gardens are systemized with a low ornamental value. By applying the proposed solutions, based on landscape design rules and vegetable growing technology, they will become unifying aesthetic elements of the family garden.

## CONCLUSIONS

Attractive perspectives were created through the usage of visual cues, of colors and volumes, which lead to an increase in compositional unity and also regarding the sustainability of the family vegetable gardens.

The proposed solutions for the three case studies increase the functionality, the technical and ornamental values of the garden.

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## CONTROLLING SEEDLING HEIGHT BY TREATING SEEDLINGS WITH PACLOBUTRAZOL ON SOME PLANT SEEDLINGS

### CONTROLUL ÎNĂLȚIMII RĂSADURILOR PRIN TRATAREA ACESTORA CU PACLOBUTRAZOL LA CÂTEVA SPECII DE PLANTE

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**Abstract.** *The objective of this study was to control seedling height by treating seedlings with paclobutrazol on some plant seedlings at plug growth period under in vitro and greenhouse conditions during first seedling cycle. Paclobutrazol was applied as foliar sprays to in vitro and greenhouse-grown seedlings [banana (*Musa acuminata*, cv. Dwarf Cavendish), tomato (*Lycopersicon esculentum* Mill., cv. T83-48F1) and potato (*Solanum tuberosum*, cv. Granola)]. Foliar spraying was done one week after transplanting at doses of 0, 100, 250, 500, 750 and 1000 ppm of Paclobutrazol solutions. To examine the effect of Paclobutrazol application on growth of seedlings in “in vitro” and greenhouse conditions, plant growth, leaf area, thickness of leave, leaf chlorophyll content, plant height, root length and internodes length were measured. Paclobutrazol had effect at all doses and plug growth was reduced. Plant height was shortened; leaf area and internodes distance and thickness of leaves were reduced. Paclobutrazol slightly decreased the internodes length. The leaf area was reduced with increasing dose of paclobutrazol and total leaf chlorophyll content increased. Results showed that paclobutrazol application positively affected seedling growth and increased the adaptation of the seedlings in the field conditions.*

**Key words:** plant regulator, *in vitro*, paclobutrazol, banana, *Musa acuminata*, tomato, *Lycopersicon esculentum*, potato, *Solanum tuberosum*, chlorophyll content

**Rezumat.** *Scopul acestei lucrări a fost de a studia dezvoltarea înălțimii răsadurilor prin tratarea acestora cu paclobutrazol. Răsadurile studiate au fost crescute atât în condiții “in vitro”, cât și în condiții de seră pe durata primului ciclu de viață. Paclobutrazolul a fost aplicat foliar răsadurilor crescute în condiții “in vitro” cât și în condiții de seră [banan (*Musa acuminata*, cv. Dwarf Cavendish), tomate (*Lycopersicon esculentum* Mill., cv. T83-48F1) și cartof (*Solanum tuberosum*, cv. Granola)]. Produsul a fost aplicat foliar după o săptămână de la transplantare în doze de 0, 100, 250, 500, 750 și 1000 ppm soluție de paclobutrazol. În vederea evaluării efectului aplicării de paclobutrazol asupra dezvoltării răsadurilor în condiții “in vitro” cât și în condiții de seră au fost analizate: dezvoltarea plantelor, suprafața frunzelor, grosimea acestora, conținutul în clorofilă a frunzelor, înălțimea plantelor, lungimea rădăcinilor și a internodurilor. Paclobutrazolul a avut efect pentru*

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*toate dozele aplicate și dezvoltarea plantelor a fost diminuată. Plantele au avut înălțimi mai mici, iar suprafața frunzelor, distanța internodală și grosimea frunzelor au scăzut. Paclobutrazolul a avut un ușor efect descrescător asupra lungimii internodurilor. Suprafața frunzelor s-a redus odată cu creșterea dozelor de paclobutrazol iar conținutul total de clorofilă din frunze a crescut. Rezultatele obținute arată ca aplicarea de paclobutrazol influențează în mod pozitiv dezvoltarea răsadurilor și crește capacitatea de adaptare a răsadurilor în condițiile din câmp.*

**Cuvinte cheie:** regulator de creștere, *in vitro*, paclobutrazol, banan, *Musa acuminata*, tomate, *Lycopersicon esculentum*, cartof, *Solanum tuberosum*, conținut de clorofilă

## INTRODUCTION

In Turkey, tomato, potato and banana plant cultivation has undergone changes mainly related to seedling production due to two strong reasons: the advance of disease and pest into disease-free areas and the formation of large export orchards. Use of micro-propagated seedling *in vitro* techniques and seedlings size control with growth regulators such as paclobutrazol led to the production of a large amount of seedlings with high phytosanitary standard within a short period of time. This was made possible by the use of micro-propagation techniques.

Plant size control is a major problem confronting the seedling industry. During seedling growth, large amounts of energy reserves are partitioned into vegetative growth, thereby potentially depleting reserves for seedling production. Redirecting energy reserves to reproductive seedling growth and reducing seedling size, for example by applying growth regulators, may increase seedling productivity and reduce irregular bearing. Triazole-type plant growth retardants such as paclobutrazol are effective on a wide spectrum of seedling plants (Jung *et al.*, 1986).

Paclobutrazol as a potent specific inhibitor of GA<sub>3</sub> biosynthesis (Aloni and Paskkar, 1987), restricts the GA<sub>3</sub> synthesis in leaves and fruits. Paclobutrazol, an inhibitor of gibberellin biosynthesis (Hedden 1983) has shown promise in reducing vigor of many plant species (Tukey, 1986; Li *et al.*, 1989). Paclobutrazol is slowly metabolized in plants (Sterrett, 1985) and is acropetally transported via the xylem which may act as a reservoir for paclobutrazol taken up by roots (Lever, 1986), thus prolonging persistence.

The main plant responses observed after paclobutrazol application are: reduced plant height and foliar area, smaller length of the new shoots, increased chlorophyll concentration and green color intensification. Paclobutrazol has an advantage over many plant growth regulator in that application can be effective when on the sprayed foliage or as a soil drench or when initially incorporated in the potting compost. Reports of paclobutrazol on horticultural plants have shown good potential in suppressing or reducing growth without reducing yield. Application of paclobutrazol significantly reduced the rate of extension of the

stem and, subsequently, the final plant height. Paclobutrazol has exhibited retardant effects in seedling. Paclobutrazol reduces vegetative growth on plants. Paclobutrazol, a PGR's has been reported to be very effective for dwarfing a wide range of crops including *Lolium perenne* (Hebhlethwaite *et al.*, 1982), tulips (Menhenett and Hanks, 1983), chrysanthemum (McDaniel, 1983) and carrots (Thomas *et al.*, 1982). Similar results were reported by Brigard *et al.*, (2006); Magnitskiy, (2004); Magnitskiy *et al.* (2006) on tomato; by El-Otmani *et al.*, (1992) and Maia *et al.*, (2009) on banana, and by Tekalign *et al.*, (2005) on potato. Paclobutrazol has been reported to reduce the vegetative growth and increase flowering and fruiting in apples (Greene, 1986). Leaf area reduction by paclobutrazol was reported on apple seedlings (Steffens *et al.*, 1986), cranberries seedlings (McArthur and Eaton, 1989). Paclobutrazol did not delay flower emergence or time of harvesting. Literatures indicate that application of paclobutrazol increased total yield of apples (Lever, 1986). Similar results reported by Lolaei *et al.*, 2012 on strawberry. In strawberry analyses McArthur and Eaton (1988) indicated a slight increase in flower number treated with paclobutrazol. Paclobutrazol was reported to have no effect on fruit quality of strawberries (Atkinson and Crisp, 1986; Lolaei *et al.*, 2012), grapes (Intrieri *et al.*, 1986) or citrus (Monselise, 1986). Fruit starch content was significantly increased by paclobutrazol applied as a foliar treatment. Others have shown that paclobutrazol had no effect on reduced yield of apples (Elfving and Proctor, 1986) or increased fruit size (Erez, 1986). These results indicate that paclobutrazol decreased vegetative growth caused by the growth retardants. Increasing paclobutrazol concentration increased total leaf chlorophyll content. Similar conclusions can be drawn for two components: chlorophyll *a* and chlorophyll *b*, with chlorophyll *a* being present in double quantities those of chlorophyll *b*. Wang (1985) reported that application of paclobutrazol by soil drench inhibited growth and increased chlorophyll content of leaf discs on cucumber and zucchini squash. Tekalign *et al.*, (2005) reported that paclobutrazol treated leaves were dark green due to high chlorophyll *a* and *b* in potato in accordance with the stimulation effect of paclobutrazol on cytokinin synthesis.

Some countries and institutions mandate maximum residual levels of paclobutrazol at 0.01 mg kg<sup>-1</sup> in stone fruits, 0.5 mg kg<sup>-1</sup> in dry beans, 0.5 mg kg<sup>-1</sup> in apples and small berries. These levels have been established based on analysis of plants subjected to foliar or soil of paclobutrazol applications (Singh and Ram, 2000). Magnitskiy *et al.*, (2006) reported that paclobutrazol residue was not detected in cucumber and tomato fruits harvested from plants grown from seed soaked in 1000-4000 ppm paclobutrazol for 180 minutes without accumulation of paclobutrazol residue in fruits. Assuming that if tomato, potato and banana plants could be kept small by the use of growth retarding chemicals (same as paclobutrazol) without appreciable reduction of yield per plant, per hectare plant populations can be increased and therefore total yield could be increased.

Purpose of the present investigation was to examine the retarding effects of paclobutrazol on the growth of tomato banana and potato seedling. This work

proposes to compare the effects of paclobutrazol on some development characteristics tomato, potato and banana seedling by the use of micro-propagation techniques in *in vitro* under greenhouse conditions.

## MATERIAL AND METHOD

For the experiment, the seedlings were propagated at the Plant Tissue Culture Laboratory of the Agricultural Biotechnology Department, Faculty of Agriculture, Süleyman Demirel University. Tomato seedlings were micro-propagated from tomato seed (*Lycopersicon esculentum* Mill., cv. T83-48F1), potato seedlings were micro-propagated from meristem culture of potato (*Solanum tuberosum*, cv. Granola) and banana seedlings were propagated (*Musa acuminata*, cv. Dwarf Cavendish) by the use of micro-propagation techniques in *in vitro*. Tomato seeds were placed in trays (each trays has in 250-cell) with a sheet of fibres on the liquid  $\frac{1}{4}$  MS aerated solution. Then trays (1000 seeds) were placed in an environmentally controlled growth box ( $21\pm 3^{\circ}\text{C}$ , 80% relative humidity, dark) in a completely randomized design. Tomato seeds were placed onto a fresh liquid  $\frac{1}{4}$  MS medium *in vitro* and seedlings were obtained in 25 days. MS medium was supplied uniformly for all treatments once a day through a mat aeration system. A liquid equal amount of liquid  $\frac{1}{4}$  MS medium was supplied to all treatments. Germination of tomato seeds was checked once a day *in vitro* conditions. Later, acclimatized tomato seedling plantlets were transferred to the in plug viols (in 45 celled transplant trays, filled with equal parts by volume of vermiculite, polystyrene granules and peat) after one week under greenhouse.

To examine the effect of paclobutrazol application on growth of seedlings under greenhouse conditions, the paclobutrazol assayed and non- assayed seedlings were used. The control consisted of untreated seedlings. After acclimatized seedlings were transferred to the in plug-mix compost (in 45 celled transplant trays, filled with equal parts by volume of vermiculite, polystyrene granules and peat) after one week. Tomato seedlings were sprayed until run-off with 0, 100, 250, 500, 750 and 1000 ppm of paclobutrazol solutions. In the foliar treatment, each plant received 4 ml solution using a hand sprayer, while the control plants received the same amount of distilled water. The grown seedlings were transferred to greenhouse conditions and maintained at  $21\pm 3^{\circ}\text{C}$ , 50-60 % relative humidity and  $50 \mu\text{mol m}^{-2} \text{s}^{-1}$  light intensity.

Propagated seedlings *in vitro* conditions from meristem were adapted under greenhouse conditions. After acclimatized plantlets were transferred to the in plug-mix compost after one week. At the end of three week period, seedlings were assessed according to their adaptation capacity (number of seedling plants). Adapted banana and potato seedlings were applied foliar sprays with various doses of paclobutrazol solutions (0, 100, 250, 500, 750 and 1000 ppm). The foliar treatments of banana and potato were followed the same procedure as in the foliar treatments of tomato seedlings. Foliar spraying was done one week after transplanting and in plug-mix compost in greenhouse-grown seedlings.

Seedlings of equal size were transplanted in cell plug flats containing same plug-mix compost. The flats were placed on greenhouse benches on a capillary mat and sub-irrigated. After 15 days, the flats were removed from the capillary mat and overhead. Banana, tomato and potato seedling were grown in plug-mix compost and fertilized with a water-soluble fertilizer 20N-8P-16K at a rate of 2.5 g/l every four irrigation until approximately 21 days of cultivation in a greenhouse.

The experiment included 4 replicates consisting of 25 seedlings per replication. One seedling was placed per cell plug flats. A hundred treated seedlings were used each

treatment in order to select the most suitable paclobutrazol doses. After application of paclobutrazol and 21 days of cultivation, growth of seedlings were observed for their seedlings of the surviving (survival rate) and usable plugs of banana, tomato and potato data were recorded. Five seedlings of each treatment replication were harvested to calculate mean of total seedling height, internodal length, leaf area, leaf thickness, leaf chlorophyll content (Chlorophyll *a* and *b*), stem diameter and root length. Fifteen leaves of the first, second and third leaves from the top shoots were collected for each paclobutrazol dose. Non-applied seedlings, bearing 15 leaves were used as control. Leaf area of the representative seedlings was measured by a digital planimeter (Placom KP-90N). Plant chlorophyll content was measured after the treatment with paclobutrazol. Non-applied seedling plants of leaf samples were used as control to compare with the applied material. The chlorophyll content analysis of paclobutrazol applied leaf samples was carried out using 10 leaves for each dose, plus 10 non-applied as control plants. For measurement of chlorophyll concentration, vigorous and uniform leaves were sampled and crude leaf chlorophyll extracts were made using 80% (v/v) acetone for 24 h for 5 plants per treatment. Total chlorophyll concentration was determined by measuring absorbance of the extracted solution at 645 nm and 663 nm with a spectrophotometer (Uvikon 922, Kotron) according to the procedure developed by Arnon (1949).

Chlorophyll concentration ( $\mu\text{g}\cdot\text{mg}^{-1}\text{fw}$ ) =  $[(20.29 \times A_{645}) + (8.02 \times A_{663})] \times [\text{volume of acetone (ml)} \div \text{fresh weight (mg)}]$ , where  $A_{645}$  and  $A_{663}$  are absorbance at 645 and 663 nm, respectively.

After foliar application of paclobutrazol, five seedlings of each treatment replication were destructively harvested for residual of paclobutrazol effects, and surface dried, placed in paper bags, and dried at 30°C for 3 day. Dried seedling plants were stored at -18°C for further measurements of paclobutrazol residues.

The test was performed in an entirely factorial scheme and a complete randomised plots experimental design with three replications of 15 plants, each replication represented by ten seedling plants and 45 seedlings per dose were used. The statistical one-way analysis of variance was used to analyse data. The mean values were subjected to analysis using the Tukey-Kramer (JMP, Version 5) to determine the dose of paclobutrazol necessary to reduce or increase the above parameters for a comparison of the averages. Mean separation between treatments by LSD was 5 percent level.

## RESULTS AND DISCUSSIONS

At all doses of paclobutrazol, growth in plug (seedling height, root length, leaf area and internodal length) decreased but leaf thickness increased. Seedlings in the concentration trial were more responsive to paclobutrazol than those in the formulation test as shown by the final growth reduction. Most reduction in growth was obtained from 500-750 ppm of paclobutrazol applications. Least reduction in seedling growth was from a foliar application at the lower rate of 100 ppm.

Survival rate of the seedling was affected by concentrations of paclobutrazol after planting. Results showed that paclobutrazol application positively affected seedling growth and increased the adaptation of the seedlings in the greenhouse (tab. 1, tab. 2 and tab. 3).

A high correlation was obtained between paclobutrazol rate and seedling plant height was shortened. Data on final total seedling height of banana, tomato and potato

plants showed highly significant results (tab. 1, tab. 2 and tab. 3). All the treatments (100-750 ppm) of paclobutrazol greatly reduced the seedling height as compared to control but the differences between 750-1000 ppm were non-significant. Paclobutrazol treatment resulted in shorter (total seedling height = plant height and length of internodes) and stem diameter compared to the control seedling plant (tab. 1, tab. 2 and tab. 3). Seedling internodal length after application was restricted by paclobutrazol application (tab. 1, tab. 2 and tab. 3). Internodes length was most restricted between 100-1000 ppm paclobutrazol. All treatments significantly reduced seedling height extension of greenhouse-grown banana, tomato and potato seedlings. Paclobutrazol rate of 750 ppm being extremely inhibitory resulted in almost complete suppression of internodal length growth. At the tomato, the mean seedling height was reduced from 138.9 cm (control) to 40.7 cm (750 ppm) in response paclobutrazol treatment while stem diameter was increased (15.1 mm at 750 ppm) over the control (7.9 mm) (tab. 1).

At the banana, the mean seedling (pseudo-stem) height was reduced from 38.4 cm (control) to 10.1 cm (750 ppm) in response paclobutrazol treatment while pseudo-stem diameter was increased (17.6 mm at 750 ppm) over the control (9.1 mm) (tab. 2). At the potato, the mean seedling height was reduced from 123.4 cm (control) to 50.5 cm (750 ppm) in response paclobutrazol treatment while stem diameter was increased (13.6 mm at 750 ppm) over the control (5.7 mm) (tab. 3). Paclobutrazol's the magnitude of the growth inhibitory effect increased as the rate of application increased. The results of Brigard *et al.* (2006), Magnitskiy (2004) and Magnitskiy *et al.*, (2006) were in agreement with the results obtained for tomato, and results by Tekalign *et al.*, (2005) were in agreement with the results obtained for the potato who reported that the paclobutrazol reduced the height of the seedling. Research by Passian and Bennet (2001), Pill and Gunter (2001), and Still and Pill (2003) suggested the need for higher paclobutrazol concentrations for effective long-term height control.

Leaf area was affected by high paclobutrazol concentration of the foliar spraying. Paclobutrazol treatment, leaf area of leaves was reduced but leaf thickness increased. The leaf area was reduced with increasing dose of paclobutrazol. Analysis of variance revealed highly significant differences between all the treatments in respect of leaf area (tab. 1, tab. 2 and tab. 3). It reflects that the leaf area decreased proportionately with the increasing dose of paclobutrazol. The leaf area was maximum in (control) with an area of 97.9 cm<sup>2</sup> followed by 100 ppm and 1000 ppm with a size of 37.7 cm<sup>2</sup> and 18.4 cm<sup>2</sup>, respectively for tomato (tab. 1); the leaf area was maximum in (control) with an area of 59.9 cm<sup>2</sup> followed by 100 ppm and 1000 ppm with a size of 28.3 cm<sup>2</sup> and 14.5 cm<sup>2</sup>, respectively for banana (tab. 2), the leaf area was maximum in (control) with an area of 73.8 cm<sup>2</sup> followed by 100 ppm and 1000 ppm with a size of 22.2 cm<sup>2</sup> and 13.9 cm<sup>2</sup>, respectively for potato (tab. 3). These observations are in accordance with those of Ramina and Tunutti (1985); Assem (1986) and Maia *et al.*, (2009). As in the species differences in response to paclobutrazol, adjusted leaf area for seedling were decreased by paclobutrazol. The results of Atkinson *et al.*, (1985) were in confirmity at the species.

Table f

The effect of foliar application of paclobutrazol doses applied twenty-one days after adaptation on total seedling height, internodes length, survival rate, leaf area, leaf thickness, leaf chlorophyll content, usable plugs, stem diameter and root length on potted tomato at the propagated seedlings *in vitro* conditions

Paclobutrazol doses (ppm)	Survival rate number of the seedling (%)	Usable plugs (%)	Total seedling height (cm)	Stem diameter (mm)	Length of internodes (cm)	Total leaf area (cm <sup>2</sup> )	Total leaf thickness (μm)	Leaf chlorophyll content (μg mg <sup>-1</sup> fresh weight)		Root length (cm)
								a	b	
0.0	98.6 <sup>a</sup> ±1.2	91.7 <sup>a</sup> ±1.5	138.9 <sup>a</sup> ±1.0	7.9 <sup>a</sup> ±3.4	6.1 <sup>a</sup> ±0.5	97.9 <sup>a</sup> ±1.3	201.12 <sup>a</sup> ±0.3	0.58 <sup>a</sup> ±0.07	0.34±0.04	28.0 <sup>a</sup> ±0.9
100.0	98.4 <sup>a</sup> ±1.6	84.4 <sup>b</sup> ±2.0	96.4 <sup>b</sup> ±1.2	10.3 <sup>b</sup> ±3.6	4.7 <sup>b</sup> ±0.3	37.7 <sup>b</sup> ±1.1	269.88 <sup>b</sup> ±0.6	0.75 <sup>b</sup> ±0.04	0.46±0.06	27.9 <sup>b</sup> ±1.2
250.0	98.2 <sup>a</sup> ±1.9	87.6 <sup>ab</sup> ±1.8	81.6 <sup>b</sup> ±1.6	12.8 <sup>b</sup> ±4.5	3.3±0.5	24.8 <sup>b</sup> ±1.5	275.75 <sup>b</sup> ±0.4	0.80 <sup>b</sup> ±0.08	0.53±0.02	26.1 <sup>b</sup> ±0.7
500.0	97.1 <sup>a</sup> ±1.4	83.9 <sup>b</sup> ±1.5	47.3 <sup>b</sup> ±1.3	13.4 <sup>ab</sup> ±3.9	2.9±0.7	20.5 <sup>b</sup> ±1.5	283.16 <sup>b</sup> ±0.6	0.84 <sup>b</sup> ±0.06	0.57±0.05	24.4 <sup>b</sup> ±1.4
750.0	99.2 <sup>a</sup> ±1.5	90.5 <sup>b</sup> ±1.4	40.7±1.7	15.1 <sup>a</sup> ±4.1	2.4±0.4	17.3 <sup>b</sup> ±1.6	304.33 <sup>b</sup> ±0.4	0.92 <sup>b</sup> ±0.04	0.59±0.04	25.8 <sup>b</sup> ±0.8
1000.0	97.8 <sup>a</sup> ±1.7	85.3 <sup>b</sup> ±1.6	41.9±1.8	16.4 <sup>a</sup> ±3.7	1.8±0.8	18.4 <sup>a</sup> ±1.3	395.91 <sup>b</sup> ±0.7	0.87 <sup>b</sup> ±0.05	0.55±0.07	26.3 <sup>b</sup> ±0.6

Significant differences between treatment effects were analyzed by regression analysis.

<sup>a</sup>/values ± mean standard deviation

Means followed by the same letters do not differ significantly at 5% level of significance.

Table 2

The effect of foliar application of paclobutrazol doses applied twenty-one days after adaptation on total seedling height, internodes length, survival rate, leaf area, leaf thickness, leaf chlorophyll content, usable plugs, seedling (pseudo-stem) height, stem (pseudo-stem) diameter, and root length on potted banana at the propagated seedlings *in vitro* conditions

Paclobutrazol doses (ppm)	Survival rate number of seedling at the (%)	Usable plugs (%)	Total seedling (pseudo stem) height (cm)	Stem diameter (pseudo-stem) (mm)	Length of internodes (cm)	Leaf area (cm <sup>2</sup> )	Total leaf thickness (μm)	Leaf chlorophyll content (μg mg <sup>-1</sup> fresh weight)		Root length (cm)
								a	b	
0.0	98.9 <sup>a</sup> ±1.2	97.8 <sup>a</sup> ±1.3	38.4 <sup>a</sup> ±1.9	9.1 <sup>a</sup> ±3.7	14.9 <sup>a</sup> ±0.9	59.9 <sup>a</sup> ±1.3	356.4±0.2	0.40±0.06	0.19±0.04	43.7 <sup>a</sup> ±1.1
100.0	98.6 <sup>a</sup> ±1.4	94.3 <sup>a</sup> ±1.4	25.6 <sup>a</sup> ±1.6	12.5 <sup>a</sup> ±4.5	10.8 <sup>a</sup> ±1.3	28.3 <sup>a</sup> ±0.9	384.7 <sup>a</sup> ±0.2	0.52 <sup>a</sup> ±0.03	0.28±0.06	41.5 <sup>a</sup> ±1.3
250.0	96.2 <sup>a</sup> ±1.2	96.7 <sup>a</sup> ±1.1	22.7 <sup>a</sup> ±1.9	14.9 <sup>a</sup> ±3.9	8.1±0.8	25.6 <sup>a</sup> ±1.1	403.9 <sup>a</sup> ±0.4	0.57±0.05	0.35±0.08	40.2 <sup>a</sup> ±0.7
500.0	95.8 <sup>a</sup> ±1.3	94.9 <sup>a</sup> ±1.2	16.5 <sup>a</sup> ±1.5	15.7 <sup>a</sup> ±4.4	7.4±0.9	20.4 <sup>a</sup> ±0.9	427.1 <sup>a</sup> ±0.1	0.64 <sup>a</sup> ±0.07	0.39±0.05	39.8 <sup>a</sup> ±0.8
750.0	97.9 <sup>a</sup> ±1.5	95.2 <sup>a</sup> ±1.2	10.1 <sup>a</sup> ±1.4	17.6 <sup>a</sup> ±3.8	6.6 <sup>a</sup> ±1.2	16.9 <sup>a</sup> ±1.0	451.8 <sup>a</sup> ±0.3	0.77 <sup>a</sup> ±0.04	0.41±0.08	37.6 <sup>a</sup> ±1.1
1000.0	97.7 <sup>a</sup> ±1.2	95.6 <sup>a</sup> ±0.9	7.8 <sup>a</sup> ±1.5	18.0 <sup>a</sup> ±4.2	5.9±1.3	14.5 <sup>a</sup> ±1.2	507.5 <sup>a</sup> ±0.2	0.79 <sup>a</sup> ±0.06	0.47±0.04	36.4 <sup>a</sup> ±0.9

Values ± mean standard deviation. Means followed by the same letters do not differ significantly at 5% level of significance.

Table 3

The effect of foliar application of paclobutrazol doses applied twenty-one days after adaptation on total seedling height, internodes length, survival rate, leaf area, leaf thickness, leaf chlorophyll content, usable plugs, stem diameter and root length on potted potato at the propagated seedlings *in vitro* conditions

Paclobutrazol doses (ppm)	Survival rate number of seedling at the (%)	Usable plugs (%)	Total seedling height (cm)	Stem diameter (mm)	Length of internodes (cm)	Total leaf area (cm <sup>2</sup> )	Total leaf thickness (μm)	Chlorophyll content (μg mg <sup>-1</sup> fresh weight)		Root length (cm)
								a	b	
0.0	92.9 <sup>a</sup> ±1.3	87.5 <sup>a</sup> ±2.0	123.4 <sup>a</sup> ±1.7	5.7 <sup>a</sup> ±3.9	4.2 <sup>a</sup> ±0.8	29.8 <sup>a</sup> ±1.6	188.75 <sup>a</sup> ±0.9	0.46±0.06	0.25±0.07	20.4 <sup>a</sup> ±1.1
100.0	90.3 <sup>a</sup> ±1.1	86.9 <sup>a</sup> ±1.8	98.9 <sup>a</sup> ±1.4	8.9 <sup>a</sup> ±4.4	3.2±0.8	22.2±2.2	242.67 <sup>a</sup> ±1.1	0.64 <sup>a</sup> ±0.08	0.34±0.09	18.7 <sup>a</sup> ±1.2
250.0	91.7 <sup>a</sup> ±1.5	87.3 <sup>a</sup> ±1.7	83.3 <sup>a</sup> ±1.5	10.7 <sup>a</sup> ±3.8	3.0±0.9	17.9 <sup>a</sup> ±1.9	255.63 <sup>a</sup> ±0.9	0.68 <sup>a</sup> ±0.09	0.42±0.06	17.9 <sup>a</sup> ±0.9
500.0	89.6 <sup>a</sup> ±1.3	86.1 <sup>a</sup> ±1.7	59.6±1.9	11.1 <sup>a</sup> ±4.3	2.9 <sup>a</sup> ±1.0	14.3±1.5	268.12 <sup>a</sup> ±1.0	0.73 <sup>a</sup> ±0.03	0.45±0.07	18.5 <sup>a</sup> ±1.3
750.0	91.5 <sup>a</sup> ±1.7	89.8 <sup>a</sup> ±1.6	50.5±1.4	13.6 <sup>a</sup> ±4.1	2.2±1.2	10.5±1.7	281.45 <sup>a</sup> ±0.7	0.81 <sup>a</sup> ±0.09	0.48±0.09	17.2 <sup>a</sup> ±1.4
1000.0	90.2 <sup>a</sup> ±1.5	86.5 <sup>a</sup> ±1.4	52.8 <sup>a</sup> ±1.8	12.8 <sup>a</sup> ±4.0	2.3±1.2	13.9±1.9	279.14 <sup>a</sup> ±0.8	0.76 <sup>a</sup> ±0.05	0.46±0.05	19.6 <sup>a</sup> ±1.2

Values ± mean standard deviation

At the 750 ppm paclobutrazol treatment, leaf thickness increased from 201.12  $\mu\text{m}$  (control) to 395.91  $\mu\text{m}$  in tomato (tab. 1), from 356.4  $\mu\text{m}$  (control) to 507.5  $\mu\text{m}$  (1000 ppm) in banana (tab. 2) and from 188.75  $\mu\text{m}$  to 279.14  $\mu\text{m}$  in potato (tab. 3). The results of Brigard *et al.* (2006), Magnitskiy (2004) and Magnitskiy *et al.*, (2006) were in agreement with the tomato, and by Tekalign *et al.*, (2005) were in agreement with the potato who reported that the paclobutrazol reduced the height of the seedling.

Paclobutrazol slightly decreased the internodal length. Measurement of the internodal length (tab. 1, tab. 2, tab. 3) exhibited a highly significant effect of paclobutrazol on the reduction of internodal length. With the increasing dose of paclobutrazol, reduction of internodal length was observed at the tomato, potato and banana seedling. The internodal length of tomato was maximum in control (6.1 cm) followed by 1.8 cm (at 1000 ppm) and 2.4 cm (at 750 ppm) (tab. 1). The internodal length of banana was maximum in control (14.9 cm) followed by 5.9 cm (at 1000 ppm) and 6.6 cm (at 750 ppm) (tab. 2). The internodal length of potato was maximum in control (4.2 cm) followed by 2.2 cm (at 750 ppm) and 2.3 cm (at 1000 ppm) (tab. 3). All these treatments differed from each other significantly.

Application of paclobutrazol at the seedling growing stage increased chlorophyll content. Increasing paclobutrazol concentration resulted in significant increase in total leaf chlorophyll content. The total leaf chlorophyll content increased with increasing dose of paclobutrazol. At the 750 ppm Paclobutrazol treated leaves were dark green due to high chlorophyll *a* (0.92  $\mu\text{g mg}^{-1}$  fresh weight) and chlorophyll *b* (0.59  $\mu\text{g mg}^{-1}$  fresh weight) contents for tomato (tab. 1); and high chlorophyll *a* (0.77  $\mu\text{g mg}^{-1}$  fresh weight) and chlorophyll *b* (0.41  $\mu\text{g mg}^{-1}$  fresh weight) contents for banana (tab. 2); and high chlorophyll *a* (0.81  $\mu\text{g mg}^{-1}$  fresh weight) and chlorophyll *b* (0.48  $\mu\text{g mg}^{-1}$  fresh weight) contents for potato (tab. 3). Leaves of control treatment contained 0.58 and 0.34  $\mu\text{g mg}^{-1}$  fresh weight for tomato, 0.40 and 0.19  $\mu\text{g mg}^{-1}$  fresh weight for banana, 0.46 and 0.25  $\mu\text{g mg}^{-1}$  fresh weight for potato chlorophyll *a* and chlorophyll *b*, respectively. This is in agreement with results obtained by Senoo and Isoda (2003), Elfving and Proctor (1986), and Steffens and Wang (1986). Application of paclobutrazol at 100-1000 ppm by foliar spraying inhibited seedling plants height and increased chlorophyll content of leaf discs of banana, tomato and potato. This is in agreement with results obtained by Wang (1985). The higher chlorophyll content of treated banana, potato and tomato leaves may be related to the influence of paclobutrazol on endogenous cytokinin levels. It has been proposed that paclobutrazol stimulates cytokinin synthesis that enhances chloroplast differentiation, chlorophyll biosynthesis, and prevents chlorophyll degradation (Fletcher *et al.* 2000).

Results of average root length (tab. 1, tab. 2 and tab. 3) were highly significant. The average root length of paclobutrazol treated plants were 25.8 cm

(750 ppm) shorter than 28.0 cm of the control for tomato (tab. 1), and shorter 17.2 cm (750 ppm) than 20.4 cm of the control for potato (tab. 3).

At the banana seedling, paclobutrazol applications were followed by an increase in the number of lateral shoots that was reduced at higher paclobutrazol levels. These results indicate that paclobutrazol application decreased seedling growth. This is in agreement with results obtained by Brigard *et al.*, (2006), Magnitskiy (2004).

## CONCLUSIONS

It seems that paclobutrazol could play a useful role in future management of the seedling industry.

Experimental use of paclobutrazol under greenhouse conditions are to confirm the data obtained in this greenhouse experiment. In conclusion, the growth of tomato, potato and banana seedlings was efficiently regulated by paclobutrazol 750 mg l<sup>-1</sup> (by foliar spraying) treatment.

These results should be helpful in allowing plant nurseries to control the growth of tomato, potato and banana seedling. While the basic knowledge on the use of paclobutrazol in vegetable and ornamental crops is more advanced, this technique may be effective for use on other species which experience excessive plant height during plug production.

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## ROOTSTOCK AND INTERSTOCK EFFECTS ON CARBOHYDRATE AND N, P, K CONCENTRATIONS OF APPLE NURSERY TREES

### EFECTELE CONCENTRAȚIEI CARBOHIDRAȚILOR ȘI N, P, K ASUPRA PORTALTOIULUI DINTR-O PEPINIERĂ DE MĂR

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**Abstract.** *Different interstock combinations have been investigated for different purposes in apple growing so far. Some of them are still being applied in practice and positive results are observed. This study has been conducted to determine the effects of different variety/interstock/rootstock combinations on some physiological characteristics in apple nursery trees. For this purpose, Fuji (vigor) and Red Chief (spur) apple varieties have been grafted using three different interstock/rootstock combinations (MM106/M9, MM106/seedling, M9/seedling). The cultivar 'Fuji' had higher total leaf area than 'Red Chief'. Interstock/rootstock combinations did not have a significant effect on the leaf area. Root carbohydrate was detected as higher concentration than leave in both cultivars. On the other hand, N concentration of roots was lower than the leaves. Generally CH:N of leaf was high in nursery trees grafted on seedling. There were no significant differences among the interstock/rootstock combinations in terms of the total carbohydrates, C:N ratio, chlorophyll. However, it has been determined that cultivars have significant effects on these parameters.*

**Key words:** M9, MM106, seedling, interstock

**Rezumat.** *Diferite combinații de altoi au fost studiate până în prezent pentru diferite scopuri în cultivarea mărului. Câteva dintre acestea sunt încă aplicate în practică și au efecte pozitive. Acest studiu s-a realizat în vederea determinării efectelor diferitelor combinații varietate/altoi/portaltoi asupra unor caracteristici fiziologice ale merilor din pepiniere. Pentru acest scop s-au folosit varietățile de măr Fuji (pentru vigoare) și Red Chief (pentru ramificație) care au fost altoite folosindu-se trei combinații diferite (MM106/M9, MM106/răsad, M9/răsad). Cultivarul 'Fuji' a avut suprafața totală a frunzelor mai mare decât cea a cultivarului "Red Chief". Combinațiile altoi/portaltoi nu au avut efecte semnificative asupra suprafeței frunzelor. Carbohidrații din rădăcini au avut concentrații mai mari decât în frunze pentru ambele cultivare. Pe de altă parte concentrația de azot din rădăcini a fost mai scăzută decât cea din frunze. În general raportul CH:N din frunze a fost mai ridicat la pomii din pepiniere care au fost altoiți cu răsaduri. Nu s-au înregistrat diferențe între combinațiile de altoi/portaltoi în ceea ce privește carbohidrații, raportul C:N, clorofila. Cu toate acestea s-a determinat că aceste cultivare au efecte semnificative asupra acestor parametri.*

**Cuvinte cheie:** M9, MM106, răsad, altoi

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## INTRODUCTION

Size controlling of apple tree using rootstock are well-known practice to effective, modern high density orchard-management systems. Also, dwarf rootstocks can provide to size control, labor shortages and enhanced tolerance to abiotic and biotic stress factors. Interstock using in the world has been applied to provide disease-pest control (Jackson, 2003), to overcome soil-borne adversities (Hrotko and Magyar, 2004), to avoid cold damage (Balkhoven *et al.*, 2000). Combination of semi-vigorous rootstocks, that are resistant to woolly apple aphid infection of roots, with a dwarfing interstem to obtain small manageable trees is studied in New Zealand (Palmer *et al.*, 1997). Dwarfing interstems, such as M9, on semi-vigorous rootstocks MM106 can be efficient small trees (Carlson, 1978, Ferree and Carlson, 1987).

On the other hand, rootstock can affect the bearing, productivity and orchard lifetime. Vigor rootstocks have some advantages such as, strong anchorage drought resistance etc. In the specific ecology as windy, wet-heavy soil or drought climate, growers can request an appropriate rootstock. In this situation a dwarf interstock grafted between vigor rootstock and scion to get a dwarf and strong-free-standing tree. These trees combine strong root system and early bearing. Many researches have confirmed that interstock controls size and increases yield and quality (Westwood, 1993; Wertheim 1998, Özongun, 2014).

The interactive relation with scion and interstock-rootstock is not fully explained, but many researchers have produced many hypotheses such as nutrient and water transportation, carbohydrate distribution, hydraulic and hormone communication (Jackson, 2003; Özongun, 2014). The aim of this work was to test some rootstock and interstock combination influences on carbohydrate (C), SPAD and N, P, K concentration of apple nursery trees. The experiments were assessed during two consecutive year period with consistent results; the data presented here are from the second experimental year.

## MATERIAL AND METHOD

The trial was conducted at the Fruit Research Institute situated in Isparta province (lat. 37° 48' 52.16" N and long. 30° 52' 39.66" E; altitude: 920 m) of Turkey. 'Red Chief' (spur) and 'Fuji' (vigour) varieties were used as scion in the trials. The interstocks (M9 and MM106) were grafted with whip graft at 10 cm height of rootstocks (*Malus sylvestris* seedling and M9) in late February (tab. 1). Then, these materials were planted in graft plots and were sprouted in same vegetation season for T-budding. In August, scion varieties budded onto interstock above 30 cm.

**Rootstock and interstock combinations of trail**

Treatments	Rootstock	Interstock	Number of graft
MM106/Seedling	Seedling	MM106	2
M9/Seedling	Seedling	M9	2
MM106/M9	M9	MM106	2
Seedling	Seedling	-	1
MM106	MM106	-	1
M9	M9	-	1

Total leaf area was assessed on the 10 leaves randomly collected from each block in the last week of July. 10 leaves for each selected tree were measured using a digital roller-type planimeter (Placom KP-90N, Koizumi, Japan). The chlorophyll meter Spad-502 Plus portable device (Konica Minolta, Osaka, Japan) was used for measure to leaf greenness. The non-destructively measures of leaf greenness were assessed with using 30 leaves per replicate. This experiment was applied afternoon in a day. Leaves samples for the physiological analyzes *i.e.*, total carbohydrates and nutrient contents, were collected in the last week of July. The root sample collected from defoliated nursery plants. Initially, leave and root samples were decontaminated and rinsed with tap water, 2 N HCl and distilled water. Then, samples were dried at 35 °C during 1 day and at 70°C till constant weight, and then ground for sieving. Total carbohydrates were determined with a spectrophotometer (UV-1800, Shimadzu, Kyoto, Japan) according to the anthrone method described by Kaplankiran (1992). The total nitrogen (N) was determined using Kjeldahl method with a distillation unit (Gerhardt, Königswinter, Germany). The ratio of the total carbohydrate to N (C:N) was calculated. Concentrations of phosphorus (P), potassium (K) were determined with an inductively coupled plasma spectrometer (Perkin-Elmer, Optima 2100 DV Optical Emission Spectrometer, Shelton, CT 06 484, USA). The concentrations of total carbohydrates and some macro elements (N, P and K) in leaf and root tissues were expressed on a dry-mass basis.

The treatments employed in a randomized complete block design with three replicates and each replicate contains 25 plants. The variables were subjected to an analysis of variance (ANOVA) using SAS-JMP software version 7.0. Results were expressed at the  $P < 0.05$  level of significance.

## RESULTS AND DISCUSSIONS

Through the propagation method in this research, nursery plants with interstock were obtained in two vegetation period as well as classic nursery propagation. Also, the graft success has been remarkable high although these nursery plants have 2 graft unions.

The vigour cultivar ‘Fuji’ had higher total leaf area than ‘Red Chief’. The differences in cultivar’s vigour affected directly leaf area, and this result might be attributed to nutrition reserve of scion cultivar (Atay and Koyuncu, 2013). Jackson

(1997) reported that the extent of leaf area depends on the cultivar. Interstock/rootstock combinations did not have a significant effect on the leaf area for both cultivars. However, M9 was able to produce more leaf area for ‘Fuji’ (42.4 cm<sup>2</sup>) and ‘Red Chief’ (34.2 cm<sup>2</sup>) (tab. 2) compared to other rootstocks. Similar findings were reported by Gjamovski and Kiprijanovski (2011) which ‘Grany Smith’ trees grafted on Jork 9, Pajam 2 and M.9 EMLA have the highest leaf area per tree.

A positive linear relationship has been demonstrated between SPAD measurements and total extracted chlorophyll (TCHL) for a range of plant species however, models differed between field and greenhouse grown trees (Richard *et al.*, 1990). So, in our trail there was no significant effect of rootstocks on SPAD values. Also, the SPAD values of two cultivars were almost same, and have no significant. Atay and Koyuncu, (2013) points out that leaf area and SPAD of apple trees are strongly evaluated with together for explicable as physiological. Generally, there is close relationship between leaf area and photosynthesis (Kviklyš *et al.*, 2008).

The total carbohydrate and C:N ratio values of root were higher than the leave of both ‘Fuji’ and ‘Red Chief’ cultivars (tab. 2). Root reserves have a role as an important and perhaps the major source of substrates for the subsequent year’s early respiration, growth, and development. The sensitivity of root reserves to late-season stresses may disproportionately affect plant performance and yield, particularly for early flowering and fruiting (Wayne *et al.*, 1990). Nonetheless, the root system nearly always contains higher concentrations of nonstructural carbohydrates than any other portion of the tree, and therefore has been considered the main site of carbohydrate storage. Several kinds of data support this conclusion for apple (Abusrewil *et al.*, 1983; Chong, 1971; Grochowska, 1973). There were no evident differences among the interstock/rootstock combinations in terms of the total carbohydrates and C:N ratio. In overall, C concentration of root and leaf of nursery plant with interstock was lower than the without interstock. In consideration rootstocks and interstocks generally, C and CH:N ratio of roots were detected high values in all MM106 combination and followed by seedling. The leaf C and CH:N values of apple nursery trees which grafted on seedling were generally high. Study of rootstock and scion interactions demonstrated that certain rootstocks resulted in greater dry weight of both roots and shoots, but scion effects on the rootstock were complex (Brown *et al.*, 1985). Further work in this area may clarify the role of root reserves in rootstock and scion performance.

Table 2

**Effects of rootstock/interstock combinations on mean ( $\pm$ s.d.) total leaf area (TLA), SPAD values, Total carbohydrate concentration and C:N of 'Red Chief' and 'Fuji' apple cultivars**

Cultivar	Rootstock	TLA (cm <sup>2</sup> )	SPAD	Total carbohydrate (%)		C:N	
				Root	Leaf	Root	Leaf
Red Chief	MM106/Seedling	33.63 <sup>ns</sup>	56.33	9.10	2.14	9.72	0.72
	M9/Seedling	32.00	57.60	8.28	2.86	11.15	0.96
	MM106/M9	32.14	58.30	8.64	2.75	7.87	1.17
	Seedling	29.00	55.28	10.56	4.32	9.96	1.42
	MM106	32.20	58.38	12.87	3.16	14.63	0.96
	M9	34.20	56.72	9.62	4.35	8.91	1.35
	<i>Mean</i>	31.79 $\pm 1.7$	57.18 $\pm 1.34$	9.85 $\pm$ 1.68	3.26 $\pm 0.89$	10.37 $\pm 2.35$	1.10 $\pm 0.19$
Fuji	MM106/Seedling	40.85	56.43	8.38	2.40	12.36	0.72
	M9/Seedling	41.79	55.78	7.92	2.86	8.32	0.91
	Seedling	36.00	57.65	11.73	4.41	16.29	1.25
	MM106	35.10	57.00	12.64	3.34	19.45	1.04
	M9	42.40	57.99	8.39	3.53	8.93	1.02
	<i>Mean</i>	39.22 $\pm 3.41$	56.97 $\pm 0.89$	9.81 $\pm 2.19$	3.31 $\pm 0.75$	13.07 $\pm 4.77$	0.99 $\pm 0.19$

ns: nonsignificant  $p \leq 0.5$

On the other hand, N concentration of roots was lower than the leaves for two cultivars (tab. 3). There was no significant difference between all rootstocks and interstock. Nitrogen metabolism and carbohydrate metabolism are interrelated, because carbon assimilation depends on N metabolism to provide the photosynthetic machinery, and N assimilation requires carbohydrate input for the carbon skeleton and energy supply (Cheng and Fuchigami, 2002). Because the reserve nutrients are necessary to support initial growth and development the following spring, the reserve of nursery plant nitrogen (N) and carbohydrates by the end of the growing is more important. This is supported by some investigates of Titus and Kang (1982), Tromp (1983), Oliveira and Priestley (1988), Loescher *et al.* (1990). In addition, Cheng and Fuchigami (2002), reported that an understanding of whether reserve N or reserve carbohydrate limits growth in the spring has important practical implications for managing reserve N and reserve carbohydrates to improve apple tree performance.

There was not a significant effect of interstock/rootstock combinations on P and K nutrients, and nearly the same effect were recorded in both treatments. While root P concentration of 1-yr-old-apple nursery tree at the end of the

growing season was similar to laves, root K was evident low than those of leaves. Given that most cultivated deciduous fruit trees are complex genetic systems with rootstocks chosen in many cases for growth control, nutrient uptake characteristics, and, ultimately, such factors as fruit quality and yield efficiency (Westwood, 1993), there is surprisingly little information on root reserves in different rootstocks. Study of rootstock and scion interactions demonstrated that certain rootstocks resulted in greater dry weight of both roots and shoots, but scion effects on the rootstock were complex (Brown *et al.*, 1985). Further work in this area may clarify the role of root reserves in rootstock and scion performance.

Table 3

**Effects of rootstock/interstock combinations on mean ( $\pm$ s.d.) leaf and root macro element concentration of 'Red Chief' and 'Fuji' apple cultivars**

Cultivar	Rootstock	N (%)		P (%)		K (%)	
		Root	Leaf	Root	Leaf	Root	Leaf
Red Chief	MM106/Seedling	0.84 <sup>ns</sup>	2.97	0.17	0.24	0.52	1.98
	M9/Seedling	0.81	3.00	0.21	0.26	0.46	1.96
	MM106/M9	1.10	2.52	0.12	0.28	0.43	2.04
	Seedling	1.06	3.05	0.17	0.26	0.50	2.31
	MM106	0.88	3.28	0.18	0.27	0.52	2.00
	M9	1.08	3.22	0.19	0.24	0.42	2.09
	Mean	0.99 $\pm$ 0.13	3.01 $\pm$ 0.27	0.17 $\pm$ 0.01	0.26 $\pm$ 0.02	0.48 $\pm$ 0.04	1.96 $\pm$ 0.33
Fuji	MM106/Seedling	0.68	3.31	0.19	0.28	0.45	1.98
	M9/Seedling	0.95	3.16	0.16	0.27	0.57	1.99
	Seedling	0.72	3.54	0.19	0.17	0.51	1.74
	MM106	0.65	3.20	0.18	0.21	0.49	1.80
	M9	0.94	3.47	0.15	0.23	0.46	2.26
		Mean	0.79 $\pm$ 0.15	3.34 $\pm$ 0.17	0.17 $\pm$ 0.02	0.23 $\pm$ 0.04	0.50 $\pm$ 0.05

ns: nonsignificant  $p \leq 0.5$

## CONCLUSIONS

Some of the shortcomings of rootstocks can be (partly) eliminated by inserting a third cultivar between the rootstock and the scion. The rootstocks have different characteristics and may adapt to different ecological conditions. Therefore, it is necessary to thoroughly research their behavior in the adequate agro-ecological conditions related to characteristic of cultivar and growing technology. The scion-rootstock relationship is fundamental for optimal growth, water and nutrient uptake and transport. A deficiency in these physiological statuses might cause suppressed growth the scion cultivar. Therefore, rootstock and interstock selection for intended purpose (stress, etc.) must be made carefully. Based on our results interstock has no any unfavorable affect about C, C:N and some macro elements concentration of apple nursery trees. However, it has been determined that cultivars have significant effects on these parameters.

**Acknowledgements:** *This work was supported by The Scientific Research Projects Coordination Unit of Süleyman Demirel University of Turkey [Project No: 2783-D-11]. The authors thank TAGEM (Republic of Turkey Ministry of Food, Agriculture and Livestock, General Directorate of Agricultural Research and Policy).*

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## EFFECTS OF SOME POSTHARVEST TREATMENTS ON QUALITY OF FRESH-CUT 'BRAEBURN' APPLE DURING COLD STORAGE

### EFECTELE UNOR TRATAMENTE POST-RECOLTARE ASUPRA CALITĂȚII MERELOR 'BRAEBURN' TĂIATE PE DURATA STOCĂRII LA RECE

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**Abstract.** *The aim of this research is to determine the effects of Lovastatin, 1-MCP, and hot water treatments on quality of fresh-cut "Braeburn" apple during cold storage. Fruits picked at optimum harvest time were transported to the Postharvest Physiology Laboratory of Horticulture Department immediately. Fruits treated with Lovastatin (1.25 mmol/l), 1-MCP (1  $\mu\text{L L}^{-1}$ ), hot water (50°C for 60 s), and control group were sliced with an apple slicer device. Sliced apples were packaged in plastic boxes and stored at 0°C and 90±5 relative humidity during 14 days. Weight loss, fruit flesh firmness, titratable acidity, fruit flesh colour, respiration rate, ethylene production and microbial activity were determined at the beginning and after 7 and 14 days of storage. As a result, 1-MCP treated apple slices had a little higher titratable acidity and L\* values than those of the other treatments. Lovastatin treated apples gave better results in terms of microbial activity compared to other applications. Fresh-cut 'Braeburn' apple could be stored at 0°C and 90±5 relative humidity for 7 days without significant quality losses.*

**Key words:** Lovastatin, 1-MCP, hot water, fresh-cut apple, storage

**Rezumat.** *Scopul acestei lucrări a fost de a determina efectele induse de tratamentul cu Lovastatin, 1-MCP, și apă fierbinte asupra calității merelor "Braeburn" proaspăt tăiate pe durata depozitării la rece. Fructele au fost culese în perioada optimă de recoltare după care au fost imediat transportate către Laboratorul de Fiziologia plantelor din cadrul Departamentului de Horticultură. Fructele au fost tratate cu Lovastatin (1,25 mmol/l), 1-MCP (1  $\mu\text{L L}^{-1}$ ) și apă fierbinte (50°C timp de 60 s), apoi lotul martor a fost tăiat cu ajutorul unui dispozitiv de feliat. Merele feliate au fost ambulate în pungi de plastic și depozitate la 0°C și o umiditate relativă de 90±5 pe durata a 14 zile. Pierderile în greutate, consistența feliilor de măr, aciditatea titrabilă, culoarea feliilor de măr, rata de evaporare, producția de etilenă precum și activitatea microbiană au fost determinate la începutul perioadei de stocare cât și după 7 respectiv 14 zile. Ca și rezultat s-a observat că feliile de măr tratate cu 1-MCP au avut valorile pentru aciditatea titrabilă și L\* ceva mai mari decât cele obținute la folosirea altor tratamente. Merele tratate cu Lovastatin au avut rezultate mai bune în ceea ce privește activitatea microbiană decât în cazul*

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*aplicării altor metode de tratare. Merele 'Braeburn' proaspăt tăiate pot fi depozitate la o temperatură de 0°C și o umiditate relativă de 90±5 pentru 7 zile fără pierderi calitative semnificative.*

**Cuvinte cheie:** Lovastatin, 1-MCP, apă caldă, mere proaspăt tăiate, depozitare

## INTRODUCTION

Apple, a well-known fruit, is produced and consumed in many parts of the world. Turkey is also one of the main producer countries of apples in the world with 2.890.000 tons, following China and America (FAO, 2016). Thus, the susceptibility of apple and their products to enzymatic browning during postharvest, handling, and processing operations continues to be an important topic from the food science and technology (Eissa *et al.*, 2006; Bayındır *et al.*, 2013). Consumer demand is increasing for convenient, ready-to-use and read-to-eat fruits and vegetables with a fresh-like quality (Rupasinghe *et al.*, 2005). However, little has been done to evaluate the effects of some applications (lovastatin, hot water, some natural products and their derivatives) and techniques such as modified atmosphere packaging (MAP), controlled atmosphere (CA) and dynamic controlled atmosphere (DCA) on fresh-cut produce quality in Turkey. Fresh-cut fruits undergo mechanical injuries caused by fresh-cut processing such as peeling, shredding, cutting etc. The physical injury attendant to fruit processing initiates a series of events such as increased respiration and ethylene production, and increased enzyme activities (King and Bolin, 1989). Blankenship and Dole (2003) indicated that different physical and chemical techniques have been developed to extend the shelf life of fresh-cut produce: refrigeration, disinfection, ethylene absorbers, gamma irradiation, edible coating, chemical dipping and controlled/modified atmosphere.

This study aimed to investigate the effects of lovastatin, 1-MCP and hot water treatments on quality of fresh-cut 'Braeburn' apple during cold storage.

## MATERIAL AND METHOD

Apples cv. 'Braeburn' picked at the optimum harvest date in two experimental years from a commercial orchard in Isparta (Turkey) were transported to laboratory, immediately. Fruits were randomly selected and exposed to four different treatments. In the first treatment, fruits were placed in sealed plastic bags and exposed to 1-MCP ( $1 \mu\text{L L}^{-1}$ ) for 24 h at 20 °C. In the second treatment, an oil solution of  $1.25 \text{ mmol L}^{-1}$  was prepared by dissolving lovastatin in chloroform and then diluting with commercial sunflower oil. Apples were wiped with cheese-cloth dipped in oil containing lovastatin (Ju and Curry 2000). In the third treatment, fruits were dipped into hot water at 50°C for 60 s as Kupferman (2001) reported. For fourth application, the apple of same amount in per treatment was used as control fruit. After treatments apples were sliced with an apple slicer, and packaged in plastic boxes. Packaged apple slices were stored at 0°C and  $90 \pm 5\%$  Rh for 14 days in normal (air) atmosphere ( $21\% \text{ O}_2 + 0.03\% \text{ CO}_2$ ). Weight loss (%), fruit flesh firmness (N), titratable acidity (%), fruit flesh colour (CIEL\*a\*b\*), respiration rate ( $\text{ml CO}_2/\text{kg h}^{-1}$ ), ethylene production (ppm) and microbial activity (log cfu) were determined at the beginning and after 7 and 14 days of storage.

Statistical analyses were performed with General Linear Model (GLM) using SPSS software package (v.16.0). The differences among means were analyzed by Duncan's multiple range test at a significance level of 0.05.

## RESULTS AND DISCUSSIONS

The weight losses of all treated and sliced apples increased slightly at the end of 14 days storage. The effect of treatments on weight losses of stored slices in both years was not significant, but only the storage period had significant effect on fruits stored in first year. Lovastatin and 1-MCP treatments relatively limited the weight loss of fresh-cut apples compared to other treatments (tab. 1). It was thought that this positive effect of lovastatin may be due to the barrier effect of sunflower oil applied with lovastatin against moisture vapor permeable. Calderon-Lopez *et al.* (2005) reported similarly that 1-MCP treatment had positive effects on the weight loss of apple slices.

Table 1

### Weight loss (%) of cold stored apple slices after different postharvest treatments

Storage period (days)						
First Year				Second year		
Treatments	7	14	Mean	7	14	Mean
Control	0.04	0.08	0.06 <sup>2</sup>	0.02	0.04	0.03 <sup>2</sup>
Hot water	0.04	0.09	0.07	0.00	0.03	0.02
1-MCP	0.05	0.08	0.06	0.00	0.02	0.01
Lovastatin	0.03	0.06	0.05	0.00	0.02	0.01
Mean	0.04 b <sup>1</sup>	0.08 a		0.01 <sup>2</sup>	0.03	

<sup>1</sup> Means followed by different letters with in the same row and column are significantly different at  $P < 0.05$ , <sup>2</sup> not significant

The average flesh firmness of apple slices increased slightly (23.8 N and 22.3 N) in both years at the end of 14 days storage compared to initial (23.3 N and 21.6 N) values (tab. 2). This result may be due to difficulty of penetration of probe to elastic apple slices after 14 days storage. Otherwise, it is difficult to say that the firmness of apple slices increased with increasing storage period. In fact, Koyuncu *et al.* (2010) found that the flesh firmnesses of apple slices treated with hot water, 1-MCP and lovastatin were higher at the end of 14 days storage compared to initial values. In this research, the best result in terms of firmness was obtained from apple slices treated with 1-MCP. Fresh-cut fruit firmness is an important quality attribute that can be affected by cell softening enzymes present in the fruit tissue and by decreased turgor due to water loss (Varoquaux *et al.*, 1990).

Table 2

**Flesh firmness (%) of cold stored apple slices after different postharvest treatments**

Storage period (days)								
First Year					Second year			
Treatment	0	7	14	Mean	0	7	14	Mean
Control	22.5	24.1	24.9	23.8 <sup>2</sup>	23.0	21.0	22.4	22.1 <sup>2</sup>
Hot water	23.4	24.6	23.8	23.9	21.3	21.2	21.9	21.5
1-MCP	24.1	24.0	23.5	23.9	21.1	21.9	21.9	21.6
Lovastatin	23.2	25.0	22.8	23.7	21.0	21.6	22.9	21.8
Mean	23.3b <sup>1</sup>	24.4a	23.8b		21.6ab <sup>1</sup>	21.4b	22.3a	

<sup>1</sup> Means followed by different letters with in the same row and column are significantly different at  $P < 0.05$ , <sup>2</sup> not significant

There were not much variations in the average titratable acidity contents of apples at the end of storage period compared to initial values. However, the highest titratable acidity contents (0.68% and 0.53%) were found in apple slices treated with 1-MCP in both years (tab. 3). Koyuncu *et al.* (2010) found that 1-MCP treated apple slices showed the highest (0.57 %) titratable acidity content throughout the cold storage. Perera *et al.* (2003) also indicated that 1-MCP treatment was significant in terms of titratable acidity in fresh-cut apples.

Table 3.

**Titratable acid content (%) of cold stored apple slices after different postharvest treatments**

Storage period (days)								
First Year					Second year			
Treatment	0	7	14	Mean	0	7	14	Mean
Control	0.65	0.65	0.71	0.67ab <sup>1</sup>	0.59	0.41	0.56	0.52 <sup>2</sup>
Hot water	0.66	0.69	0.66	0.67 ab	0.54	0.40	0.52	0.49
1-MCP	0.66	0.71	0.67	0.68 a	0.55	0.41	0.63	0.53
Lovastatin	0.61	0.61	0.61	0.61 b	0.53	0.35	0.46	0.45
Mean	0.65 <sup>2</sup>	0.67	0.66		0.55a <sup>1</sup>	0.39b	0.54a	

<sup>1</sup> Means followed by different letters with in the same row and column are significantly different at  $P < 0.05$ , <sup>2</sup> not significant

The effects of storage period on flesh colour of apple slices were significant (data are not showed) except for first-year  $L^*$  value ( $P < 0.05$ ). Generally,  $L^*$  values of apple slices declined during the storage period. The  $L^*$  value is useful indicator of darkening during storage, either resulting from browning reactions or from increasing pigment concentrations (Rosaj-Graü *et al.*, 2006). A decrease in  $L^*$  value indicates a loss of whiteness (brightness), and a more positive  $a^*$  value indicates browning, whereas a more positive  $b^*$  indicates yellowing (Buta *et al.*, 1999). In the first year, while 1-MCP treated apples had the highest  $L^*$  value (72.09), in the second year apple slices treated with hot water (73.52) and 1-MCP (73.38) had more brightness than the other groups. Fan *et al.* (2005) reported that calcium treated slices were whiter than those of non-treated. Similar results in  $L^*$  values were found by Raybaudi-Massilia *et al.* (2007). As expected, a significant increasing in  $b^*$  values was observed throughout storage for all treatments. Correspondingly, Koyuncu *et al.* (2010) and Bayındır *et al.* (2013) found that the  $b^*$  values of sliced apples increased clearly during cold storage.

The effects of storage period and treatments on respiration rate of apple slices were significant in first year. But, respiration rate of apple slices was not affected by storage period and treatments during storage in second year ( $P < 0.05$ ). The average respiration rate values increased after 14 days storage (6.10 and 8.57 ml CO<sub>2</sub>/kg h<sup>-1</sup>) compared to initial (5.02 and 6.50 ml CO<sub>2</sub>/kg h<sup>-1</sup>) values (tab. 4). Similar results were found by Koyuncu *et al.* (2010), Chung and Moon (2009) and Calderon-Lopez *et al.* (2005). Apple slices treated with 1-MCP and lovastatin had lower respiration rates than those of the hot water and control groups at the end of storage period (tab. 4). Likewise, Koyuncu *et al.* (2010) found that lovastatin and 1-MCP treated “Granny Smith” slices gave the lowest respiration rates towards the end of their storage life.

Table 4.

**Respiration rate (ml CO<sub>2</sub>/kg h<sup>-1</sup>) of cold stored apple slices after different postharvest treatments**

Storage period (days)								
First Year					Second year			
Treatment	0	7	14	Mean	0	7	14	Mean
Control	5.60	3.29	6.77	5.22 b <sup>1</sup>	7.43	11.87	10.77	10.0 <sup>2</sup>
Hot water	4.86	4.88	9.00	6.25 a	2.64	8.56	8.48	6.56
1-MCP	5.21	5.84	5.04	5.36ab	4.26	7.76	8.46	6.83
Lovastatin	4.43	3.08	3.60	3.70 b	11.69	9.50	6.56	9.25
Mean	5.02ab <sup>1</sup>	4.27b	6.10a		6.50 <sup>2</sup>	9.42	8.57	

<sup>1</sup> Means followed by different letters with in the same row and column are significantly different at  $P < 0.05$ , <sup>2</sup> not significant

Throughout the storage, the changes in the ethylene production of the stored apple slices were shown in the Tables 5. Ethylene production decreased significantly ( $P < 0.05$ ) with increasing storage period. The average ethylene values were 0.75 ppm (first year) and 2.06 ppm (second year) at the beginning of the storage. These values decreased to 0.21 ppm and 0.96 ppm, respectively, after 14 days storage. Likewise, Perera *et al.* (2003) and Bayındır *et al.* (2013) reported that there was a general decline in ethylene productions of apple slices throughout storage period. The effect of treatments on ethylene production of apple slices was not significant, however, lovastatin treated apples had the lowest values (0.11 and 0.31 ppm) at the end of storage in both years. Koyuncu *et al.* (2010) indicated similar effects of lovastatin and 1-MCP.

The effects of storage period and treatments on microbial activity of apple slices were significant (data are not showed) in both years ( $P < 0.05$ ). The number of total bacteria on fresh cut apple slices increased with increasing storage period in all treatments. Similarly, the number of yeasts and molds increased steadily during storage period. The lowest average yeast and mold counts were determined in apple samples treated with lovastatin. While hot water was limiting application for bacteria in first year, 1-MCP gave better results in second year compared to other treatments. It can be said that storage duration was more effective than treatments in terms of microbial quality of stored apple slices. Similar results were obtained by Muthuswamy *et al.* (2008) in fresh cut apples during storage. Bett *et al.* (2001) also reported that storage duration was important factor for microbial activity of apple slices.

Table 5.

**Ethylene production (ppm) of cold stored apple slices after different postharvest treatments**

Storage period (days)								
First Year					Second year			
Treatment	0	7	14	Mean	0	7	14	Mean
Control	0.69	0.10	0.17	0.32 <sup>2</sup>	2.68	3.32	1.07	2.36 <sup>2</sup>
Hot water	0.88	0.19	0.24	0.44	0.98	2.49	1.65	1.71
1-MCP	0.67	0.25	0.32	0.41	1.11	2.01	0.80	1.31
Lovastatin	0.76	0.13	0.11	0.33	3.46	1.95	0.31	1.91
Mean	0.75a <sup>1</sup>	0.17b	0.21ab		2.06a <sup>1</sup>	2.44a	0.96b	

<sup>1</sup> Means followed by different letters with in the same row and column are significantly different at  $P < 0.05$ , <sup>2</sup> not significant

## CONCLUSIONS

1-MCP treated apple slices had a little higher titratable acidity and L\* values than those of the other treatments. Generally, lovastatin and 1-MCP treated apple slices gave better results for microbial activity than those of hot water and control applications. It can be said that fresh-cut 'Braeburn' apple could be stored at 0°C and 90±5 relative humidity for 7 days without significant quality losses.

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## THE STUDY OF A TOMATO ASSORTMENT WITH MULTI-USE (FOOD, MEDICINAL AND ORNAMENTAL)

### STUDIUL UNUI SORTIMENT DE TOMATE CU MULTIPLE UTILIZĂRI (ALIMENTARE, MEDICINALE ȘI ORNAMENTALE)

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**Abstract.** *The study was conducted in Bacau County, Podiș Farm, during 2015 - 2016, under open field conditions. The fruits stood out by (were noted for) the content of the anthocyanin type antioxidants (Indigo™ 'Apple', Indigo™ 'Blue Beauty', Indigo™ 'Blue Berries'), the excellent taste of all the cultivars and the aesthetic appearance: indigo and red: Indigo™ 'Apple', Indigo™ 'Blue Berries', indigo, red and pink: Indigo™ 'Blue Beauty', indigo, yellow and red: Lucid Gem, indigo and yellow: Indigo 'Blue Gold', Indigo 'Goldberries', Indigo™ 'Sun' F1.*

**Key words:** tomato, open field, multiple use

**Rezumat.** *Studiul s-a efectuat la Ferma Podiș din județul Bacău, în perioada 2015 – 2016, în condiții de câmp. Fructele s-au remarcat prin conținutul de antioxidanți de tip Antocianină (Indigo™ 'Apple', Indigo™ 'Blue Beauty', Indigo™ 'Blue Berries', Indigo 'Blue Gold'), gustul foarte bun al tuturor soiurilor și provocarea estetică a culorilor: mov cu roșu Indigo™ 'Apple', Indigo™ 'Blue Berries', mov cu roșu roziiu: Indigo™ 'Blue Beauty', indigo cu galben și roșu: 'Lucid Gem', indigo și galben: Indigo 'Blue Gold', Indigo 'Goldberries', Indigo™ 'Sun' F1.*

**Cuvinte cheie:** tomate, câmp, utilizare multiplă

## INTRODUCTION

The tomato metabolites are recognized as having health benefits in cardiovascular diseases and cancer, due, in part, to the bioactive properties of the plant derived compounds like polyphenolics and carotenoids (Kris-Etherton *et al.*, 2002, Gonzali *et al.*, 2009). The anthocyanins produced by the fruits of the studied tomato cultivars are determined by two genes which have been initially transferred from two wild tomato varieties to a cultivated tomato (Boches and

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Myers, 2007; Mes *et al.*, 2008). Tomato plant is an attractive plant, and it's true that the sprawling nature of indeterminate varieties can pose an aesthetic challenge. Such anthocyanin varieties produce beautiful fruits, and they can bring color and an aesthetic value to the garden (Crist, 2012).

## MATERIAL AND METHOD

During 2015 - 2016, open field experiments were performed at Podis Farm Bacau - Romania, in order to evaluate an assortment of cultivars for food, medicinal and ornamental use: Indigo™ 'Apple', Indigo™ 'Blue Beauty', Indigo™ 'Blue berries', Indigo 'Blue Gold', Indigo 'Goldberries', Indigo™ 'Sun' F1, 'Lucid Gem'.

The collection of improved cultivars with purple fruit with red, pink or yellow colors was placed in the field in permaculture system.

Phenological observations and biometric measurements were carried on the food, medicinal and decorative qualities of tested cultivars. All the pictures presented below are genuine.

## RESULTS AND DISCUSSIONS

### Food qualities study of the tomato assortment

The results of the quantitative and qualitative characteristics of the studied biological material are presented in table 1.

Table 1

Qualitative and quantitative determinations of studied cultivars

Variety/ Hybrid	Bloomed starting	Maturity of the fruit	Fruit weight (g)	Fruit shape	Fruit taste	Yield (kg/plant)
Indigo™ 'Apple'	05.06. 2016	28.07. 2016	90.0	Round	Sweet	2.1
Indigo™ 'Blue Beauty'	05.06. 2016	01.08. 2016	180.0	Beef stake	Sweet	2.8
Indigo™ 'Blue Berries'	06.06. 2016	02.08. 2016	7.0	Cherry	Very sweet	0.9
Indigo™ 'Blue Gold'	07.06. 2016	04.08. 2016	70.0	Round	Sweet	1.9
Indigo™ 'Goldberries'	10.05. 2016	05.08. 2016	8.0	Cherry	Very sweet	1.1
Indigo™ 'Sun' F1	01.06. 2016	25.07. 2016	15.0	Cherry	Sweet	1.3
'Lucid Gem'	10.05. 2016	05.08. 2016	150.0	Slightly flat	sweet	2.4

The data presented in the above table show that:

- Indigo™ Apple variety (fig. 1, fig. 2), the first fruits have ripened 131 days after the seeds sprouted, having an average weight of 90 g, a spherical shape and a sweet, flavored taste. The average yield of a single plant is 2.1 kg.



**Fig. 1** Indigo™ 'Apple' (side view)



**Fig. 2** Indigo™ 'Apple' (top view)

- Indigo™ 'Blue Beauty' variety (fig. 3, fig. 4), the first fruits have ripened 135 days after the seeds sprouted, having an average weight of 180g per single fruit, a flattened round shape and a very good, sweet, and aromatic taste. The average production of a single plant is 2.8 kg.



**Fig. 3** Indigo™ 'Blue Beauty' (side view)



**Fig. 4** Indigo™ 'Blue Beauty' (top view)

- Indigo™ 'Blue Berries' variety (fig. 5, fig. 6), the first fruits have ripened 129 days after the seeds sprouted, having an average weight of 7g per single fruit, a round shape and a quite great amount of sugar when very well ripened. The average production of a single plant is 0.8 kg.



**Fig. 5** Indigo™ 'Blue Berries' (side view)



**Fig. 6** Indigo™ 'Blue Berries' (top view)

• Indigo™ 'Blue Gold' (fig. 7, fig. 8), the first fruits have ripened 138 days after the seeds sprouted, having an average weight of 70g per single fruit, a round shape and a sweet taste. The average production of a single plant is 1.9 kg.



**Fig. 7** Indigo™ 'Blue Gold' (side view)



**Fig. 8** Indigo™ 'Blue Gold' (top view)

• Indigo™ 'Goldberries' variety (fig. 9, fig. 10), the first cherry fruits have ripened 136 days after the seeds sprouted, having an average weight of 8.0g per single fruit, a round shape and a very sweet and rich taste when very well ripe. The average production of a single plant is 1.0 kg.



**Fig. 9** Indigo™ 'Goldberries' (side view)



**Fig. 10** Indigo™ 'Goldberries' (top view)

• Indigo™ ‘Sun’ F1 hybrid (fig. 11, fig. 12), the first cherry fruits have ripened 127 days after the seeds sprouted, having an average weight of 15.0g per single fruit, a round shape and a sweet taste when very well ripe. The average production of a single plant is 1.3 kg.



**Fig. 11** Indigo™ ‘Sun’ F1 (side view)



**Fig. 12** Indigo™ ‘Sun’ F1 (top view)

• ‘Lucid Gem’ Tomato (fig. 13, fig. 14), the first fruits have ripened 135 days after the seeds sprouted, having an average weight of 150g per single fruit, a slightly flattened round shape and a sweet, balanced taste. The average production of a single plant is 2.4 kg.



**Fig. 13** ‘Lucid Gem’ Tomato (side view)



**Fig. 14** ‘Lucid Gem’ Tomato (top view)

### **Medical qualities study of the cultivars**

The medical qualities of the analyzed varieties, indicate the presence of antioxidants like anthocyanin, carotene and lycopene as presented in table 2.

It is noticed the fruits of all the 7 varieties of tomatoes studied contain anthocyanins,  $\beta$ -carotene and flavonols. Moreover, the fruit of Indigo™ ‘Apple’ Tomato, Indigo™ ‘Blue Beauty’ Tomato and Indigo™ ‘Blue Berries’ Tomato contain lycopene.

Table 2

**The study of the antioxidants presence in the cultivar's fruits**

No.	Variety/hybrid	Antioxidants		
		Lycopene	Anthocyanins	$\beta$ -carotene and flavonols
1	Indigo™ 'Apple'	Yes	Yes	Yes
2	Indigo™ 'Blue Beauty'	Yes	Yes	Yes
3	Indigo™ 'Blue Berries'	Yes	Yes	Yes
4	Indigo™ 'Blue Gold'	-	Yes	Yes
5	Indigo™ 'Goldberries'	-	Yes	Yes
6	Indigo™ 'Sun' F1	-	Yes	Yes
7	'Lucid Gem'	Some	Yes	Yes

**Ornamental qualities study of the cultivars**

**1. INDIGO™ 'APPLE'** (fig. 15). It's an indetermined variety which produces dark indigo fruits on their shoulder and dark red on the blossom end. The fruits exposed to sun become almost black due to the high level of anthocyanins. The leaves are quite purple. The immature fruits have the same indigo color on the stem-end. It is a great looking garden plant due to its decorative clusters of 7 – 8 dark fruits.

**2. INDIGO™ 'BLUE BEAUTY'** (fig. 16). It's an indetermined variety which is the result of a crossing between 'Beauty King' variety and a blue tomato. The fruits are also indigo on the upper side where the sunlight strikes. When they ripe, they become red-pink on the blossom end. The fruits are big, pulpy, resistant to sunburn, holding well on the vine for a long time, thus offering a very pleasant decorative view.

**3. INDIGO™ 'BLUE BERRIES'** (fig. 17). It's an unusual indetermined cherry variety which has almost black with a brick red blossom end fruits, very high levels of anthocyanin, and has the fruits arranged as 8 in a cluster. The plants, with pulpy leaves, are vigorous and quite resistant against diseases.

**4. INDIGO™ 'BLUE GOLD'** (fig. 18). It's an indetermined variety created by Brad Gates from Wild Boar Farms, which produces fruit of average weight, dark indigo and yellowish when ripened. The first two clusters produce

many fruits, up to 8-12 in a bunch. The fruits are sunburn resistant and have a very decorative effect.



**Fig. 15** Indigo™ ‘Apple’



**Fig. 16** Indigo™ ‘Blue Beauty’



**Fig. 17** Indigo™ ‘Blue Berries’



**Fig. 18** Indigo™ ‘Blue Gold’

**5. INDIGO™ ‘GOLDBERRIES’.** It’s an indetermined cherry variety. The long clusters are packed with beautiful indigo to bright yellow cherry fruits. The plants are very productive, being attractive in the garden.

**6. INDIGO™ ‘SUN’ F1** (fig. 19). It's an indetermined cherry type hybrid with small (15 g), purple fruit which can be dark yellow on the blossom end. It has thin and fewer leaves, the decorative effect of the plant being assured by the beautiful clusters packed with 7 – 8 outstanding fruits.

**7. ‘LUCID GEM’.** It's an indetermined variety, which produces big and very beautiful, decorative fruits. They may range from dark purple to orange and yellow. Inside they may have small red lines. The plant is very productive, can tolerate heat, the fruits do not crack and they can be kept for a long time after harvesting.



**Fig. 19** Indigo™ ‘Sun’ F1

## CONCLUSIONS

The study was conducted in Bacau County, Podiș Farm, during 2015 - 2016, under open field conditions. The fruits stood out by (were noted for) the content of the anthocyanin type antioxidants (Indigo™ ‘Apple’, Indigo™ ‘Blue Beauty’, Indigo™ ‘Blue Berries’), the excellent taste of all the cultivars and the aesthetic appearance: indigo and red: Indigo™ ‘Apple’, Indigo™ ‘Blue Berries’, indigo, red and pink: Indigo™ ‘Blue Beauty’, indigo, yellow and red: ‘Lucid Gem’, indigo and yellow: Indigo™ ‘Blue Gold’, Indigo™ ‘Goldberries’, Indigo™ ‘Sun’ F1.

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## CHANGES DURING CHERRIES RIPENING AND THEIR QUALITY AT ADRIANA, FERROVIA AND SKEENA VARIETIES, GRAFTED ON GISELA 6

### MODIFICĂRI ÎN TIMPUL MATURĂRII CIREȘELOR ȘI CALITATEA LOR LA SOIURILE ADRIANA, FERROVIA ȘI SKEENA, ALTOITE PE GISELA 6

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**Abstract.** Changes during ripening fruit are among the most important characteristics in the training and quality of the harvest cherries. This research was undertaken to examine the yield formation ( $n = 32$ ) at cherry varieties Adriana, Ferrovia and Skeena, grafted on Gisela 6. Regardless of variety, branches bunch cherries mass decreases from 8.69 - 8.89 g annual branches to 7.19 - 7.49 g branches of three years. Harvest is uniformly biennial branches (27.27-39.21%), branches bunch of 1 year (from 27.27 to 31.22%) and branches bunch of 2 years (from 27.35 to 31.2%).

**Key words:** cherry, variety, branches of fruit, harvest

**Rezumat.** Modificările în timpul maturării fructelor sunt printre cele mai importante caracteristici în formarea recoltei și calității cireșelor. Această cercetare a fost întreprinsă pentru a examina formarea recoltei ( $n=32$ ) la soiurile de cireș Adriana, Ferrovia și Skeena, altoite pe Gisela 6. Indiferent de soi, masa cireșelor pe ramuri buchet se micșorează de la 8,69-8,89 g pe ramurile anuale la 7,19-7,49 g pe ramurile de 3 ani. Recolta este uniform repartizată pe ramuri bienale (27,27- 39,21 %), pe ramuri buchet de 1 an (27,27-31,22 %) și pe ramuri buchet de 2 ani (27,35-31,2 %).

**Cuvinte cheie:** cireș, soi, ramuri de rod, recoltă

## INTRODUCTION

Production potential and cherry fruit quality are determined by biological factors, agro technical, environmental, and also the time of harvest, collection and management practice after harvest (Yiannis *et al.*, 2013; Milošević *et al.*, 2015). Agro-technical measures of crop management, one of the most important is proper selection of variety but is equally important to succeed, that is the rootstock combination properly, and you find your own management system and cutting trees. (Milatović, 2013; Long *et al.*, 2014)

This is one of the main reasons for selecting the correct varietal and fruit growing technology. Rationale practice and perfecting the technology for

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producing high quality cherries, competitive market, and the efficient use of labor becomes an issue of great value to modern orchards.

## MATERIAL AND METHOD

They were studied, in terms of formation and development of trees, some new varieties of cherry crop in Moldova: Ferrovia, Adriana and Skeena, but widespread in the European Union (EU). The orchard was planted in 2010, varieties were grafted on Gisela 6 vegetative rootstock (*Prunus Cerasus vulgaris x canescens*). The distance between rows is 4m and between trees is 2m (1250 trees / ha). The experience includes 4 repetitions of 8 trees each (n = 32). On the other hand, measurements were made in field and laboratory conditions in accordance with the approved research methods in fruit growing.

Fruit harvesting was carried out at the stage of maturation, as skin color, as the color sheet CTIFL and soluble solids content. Establishing harvest for each variety was conducted by weighing individual fruit per trees in all 32 versions. Distribution cherry harvest (kg/tree) and fruit formations inside the crown were studied in four identical trees of every kind in the stage of full maturity of the fruit. Fruits were harvested separately biennial branches and branches bunch aged 1, 2 and 3 years. The average weight of the fruits was determined by weighing and counting them, a sample of 1 kg of cherries per each repetition.

The diameter and weight of cherries during the development and maturation of the fruits were identified using the template provided with openings 26, 28, 30, 32, 34 and 36 mm, corresponding to 8.5 mass; 10; 11.5; 13; 14.5; and 16 g accordingly. These analyzes were made on four samples in the cherry, 20 the same of every kind (n = 80).

## RESULTS AND DISCUSSIONS

Cherry plantations productivity and fruit quality is correlated in time during harvest. During the days preceding the harvest of the first fruits and cherries have a sharp increase in both volume and weight while carrying taste and qualities. Unlike other species, cherry processes do not continue ripening after harvest, that must be gathered when ripe. Industrial harvesting of cherries is done in a single pass, although staggered maturation occurs in 5-6 days. The work of harvesting is 70-80% of annual labor.

Analysis of experimental data (tab.1) shows that in the 10 days prior to harvest cherries diameter increased at Adriana variety from 21.5 to 28.3 mm, at Ferrovia variety from 22.8 to 29.5 mm and at Skeena variety from 22.4 to 29.5 mm or an average of 26.3 to 31.7%. Growth in diameter between fruit ripening cherries was different. When fruits start to mature and skin color turns from green to yellow transparent growth rate is higher compared to the last period of maturation. For example, at Adriana variety, 10 days before harvest, cherries diameter was 21.5 mm and 5 days before harvest - 25.8 mm or 20% more. In the next 5 days fruit diameter increased by 3.5 mm or only 11.6%. The same regularity was registered at Ferrovia and Skeena varieties.

The harvest is closely correlated with fruit weight and diameter of fruit. The data presented demonstrate that in the days preceding the harvest cherries mass increase considerably. From the moment they start to mature, cherries fruit weight increased by 88.3% at Adriana variety, 80.2% at Ferrovia variety and 88 5% to Skeena, representing 10.11 g, 10.29 g and respectively 10.53 g. Hence, the determination of the optimal period to harvest cherries is the first step in obtaining quantitative and qualitative fruit

production. When harvested early pre considerably, decreases the harvest, the cherries are immature, small and tasteless specific to variety characteristics. Obviously, delayed harvesting of cherries cause great damage to fruit.

Table 1

**The diameter and mass cherries during fruit development and ripening**

Date, month, year	Variety					
	Adriana		Ferrovia		Skeena	
	mm	g	mm	g	mm	g
05.06.2016	21.5	5.37	-			
09.06.2016	25.8	8.43	-			
14.06.2016	28.3	10.11				
10.06.2016	-		22.8	5.71	22.4	5.60
14.06.2016	-		26.5	8.66	26.6	8.70
19.06.2016	-		28.8	10.29	29.5	10.53

Physiologically size of the fruit is in close correlation with age branches bunch (tab. 2). It is known that fruits formed on the biennial branches mature earlier than the fruit from on the bunch branches. Also fruit on older branches bunch ripen later than fruit bouquet younger branches.

Table 2

**The diameter and mass cherries depending on age branch bunch, mm**

Variety	Branches bunch old						LSD 5%	
	1 year		2 years		3 years			
	mm	g	mm	g	mm	g	mm	g
Adriana	26.2	8.69	23.7	7.35	22.7	7.42	3.24	1.17
Ferrovia	27.2	8.89	25.1	8.21	22.9	7.49	4.15	0.98
Skeena	27.1	8.88	25.0	8.17	22.0	7.19	3.74	1.23

The results show that on the diameter, 3-year-old branches bunch cherries form smaller fruit compared branches bunch of 1 and 2 years. Thus, the studied varieties, diameter branches constituted 26.2 to 27.2 mm annual bouquet while the branches of three years was only 22.0 to 22.9 mm, or less than 13.4 -18.8%. Fruit weight is also interdependent by age branches bunch. With increasing age, decreases branch bunch fruit weight. Regardless of variety, cherries mass decreases from 8.89 to 8.69 g and from 7.49 to 7.19 g annual branches of three years.

The bouquet branches of cherry dominate, these are up to 75-80% from flowering buds and only 20-25% on the branches medium and ventures.

The results obtained highlight the fact that Adriana crop varieties, Ferrovia and Skeena, grafted on Gisela 6 during fruiting and growth is broadly based on biennial branches and branches bunch aged 1-3 years (tab. 3). Regardless of variety, 27.27- 39.21% of harvest was formed on the biennial branches, from 27.27 to 31.22% - branches bunch of one year, from 27.35 to 31.2% - 2 years branches bunch and 8.7 to 13.88% - 3 years branches bunch.

Cherry trees, grafted on rootstock Gisela 6 began to fructify the 4th year after grafting. The first harvest was the level of 625-1125 kg/ha (tab. 4).

Table 3

## Distribution cherry harvest on the fruit formations, kg/tree

Variety	Biennial branches		Branches bunch age						kg/tree
			1 year		2 years		3 years		
	Kg	%	Kg	%	Kg	%	Kg	%	
Adriana	4.99	27.27	5.46	29.84	5.50	29.56	2.35	13.33	18.30
Ferrovía	5.40	27.55	6.12	31.22	5.36	27.35	2.70	13.88	19.60
Skeena	5.57	39.21	5.78	27.27	6.92	32.65	2.92	8.70	21.19

Table 4

## Cherry fruit production according to the sort, kg/ha.

Variety	Year 2013	Year 2014	Year 2015	Year 2016	Average (2013-2016)
Adriana	625	4375	11875	8455	6332.5
Ferrovía	1125	4875	13250	10327	7394.3
Skeena	625	4250	16000	7680	7138.8
DL 5%		435.2	971.8	1205.9	

In two fructification, productivity was increased and amounted to 4250-4875kg/ha. In 2015 fruit harvest was increased significantly and amounted to 11875 kg/ha at Adriana variety up at 16000 kg/ha at Skeena variety. In the 4th year of fruiting the crop was reduced as compared to the previous year and varied from 7680 kg/ha at Skeena variety, to 10327 kg/ha at Ferrovía variety. This decrease is due to adverse weather crop conditions during the flowering trees, expressed in precipitation and cloudiness. On average the first 4 years of fruiting varieties Ferrovía (7394.3 t/ha) and Skeena (7138.8t/ha) were more productive than the variety Adriana (6332.5 t/ha).

## CONCLUSIONS

Research on quality training cherries have shown that bigger fruit, crisp handling and more resistant to scratches and transport are obtained from the branches and branches bunch biennial aged 1-3 years.

Fruit sizes are interdependent to age branches bunch. Regardless of variety, branches bunch cherries mass decreases from 8.69 - 8.89 g annual branches to 7.19 - 7.49 g branches of three years. Adriana, Ferrovía and Skeena crop varieties, grafted on Gisela 6, during fruiting and growth of the tree, is uniformly formed on biennial branches (27.27 - 39.21%), branches bunch of 1 year (from 27.27 to 31.22%), branches bunch 2 years (from 27.35 to 31.2%) and branches bunch of 3 years (8.7 to 13.88).

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## THE BEHAVIOUR OF SOME APRICOT VARIETIES GRAFTED ON GENERATIVE ROOTSTOCKS AT S.C. OLMA S.R.L. COM. BALTATI, JUD. IASI

### COMPORTAREA UNOR SOIURI DE CAIS ALTOITE PE PORTALTOI GENERATIVI LA S.C. OLAMA S.R.L. COMUNA BĂLȚAȚI, JUD IAȘI

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**Abstract.** *Production of new planting material for apricot is difficult because in Romania we don't have valuable vegetative rootstocks to ensure easy propagation and to allow establishment of intensive and superintensive orchards. In this specific segment of nursery generative rootstocks are used obtained from the seeds of *Prunus cerasifera*, apricot or even almond. Thanks to this element, we can set up intensive orchards, being necessary a relatively large space for spatial formation and development of the tree crown.*

**Key words:** grafting, varieties, generative rootstocks

**Rezumat.** *Producerea materialului săditor la specia cais este dificilă deoarece în România nu există portaltoi vegetativi valoroși care să asigure o înmulțire facilă (marcotaj vertical, orizontal etc) și care să permită înființarea de livezi intensive și superintensive. În acest segment specific al pepinierii se folosesc portaltoi generativi (franc) obținuți din semințele de mirobolan, zarzăr sau chiar migdal. Datorită acestui element, plantațiile care se pot înființa sunt de tip intensiv, fiind necesar un spațiu relativ mare pentru formarea și dezvoltarea spațială corespunzătoare a coroanei pomilor.*

**Cuvinte cheie:** altoire, soiuri, portaltoi generativi

## INTRODUCTION

Apricot, one of the species highly valued for its fruit, currently receiving great attention both from researchers and from growers.

Thus assortment of apricot has known a constant regeneration over time, each year adding new varieties, more productive and with outstanding quality fruit (Istrate, 2007).

Iasi district is the north-eastern limit of culture for this species, and for apricot, is a constant demand for planting material used for setting up new orchards and from smallholders who cultivate this species for their own consumption (Dascălu *et al.*, 2010).

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The presented material we study nursery behavior of four varieties of apricot (Goldrich, Umberto, Favorit and N.J.A 19) grafted on two rootstocks (*P. armeniaca* and *P. cerasifera*).

## MATERIAL AND METHOD

The experience was made from year 2014, at S.C. Olma S.R.L., Baltati, Iasi, and we used 4 apricot varieties (Goldrich, Umberto, Favorit, N.J.A. 19) engrafted on *Prunus armeniaca* and *Prunus cerasifera* rootstocks.

Each variety was an experimental variant, and for each was taken 5 repetitions.

Biometric measurements were made on grafting grip, number of buds start in vegetation in first nursery field, number of plants lost in second field and total new trees obtained in second year of nursery field (Gradinariu *et al.*, 1998).

## RESULTS AND DISCUSSIONS

In table 1 can be seen that the Goodrich variety, gripping the bud grafted on two rootstocks was 88% from 83% to *P. armeniaca* and *P. cerasifera*, and in the second nursery field losses were 12% and 17% respectively.

Variety Umberto had an identical behavior in both rootstocks being used, gripping was 86% and losses from the in the second nursery field around 14%.

Favorite variety has obtained the lower grip to grafting, which is 85% in the case of rootstock *P. armeniaca* and 80% at *P. cerasifera*. Also, the percentage of dying in the in the second nursery field was 15% and 20% respectively .

The N.J.A 42 grafted on to *P. armeniaca* gripping the grafting was 86%, all buds grafted starting in vegetation, and the losses were 14%. The same variety grafted on *P. cerasifera* recorded a grip rate of 79%, the number of buds lost in the second nursery field being 21%.

Table 1  
Behavior in nursery of four apricot varieties grafted on different rootstocks

Grafted variety	Rootstock used	Grafting grip (%)	Buds started in vegetation (%)	Scions lost in second field (%)	Standard (STAS) trees obtained from those grafted (%).
Goldrich	<i>P. armeniaca</i>	88	88	12	84
	<i>P. cerasifera</i>	83	83	17	79
Umberto	<i>P. armeniaca</i>	86	86	14	82
	<i>P. cerasifera</i>	86	86	14	82
Favorit	<i>P. armeniaca</i>	85	85	15	81
	<i>P. cerasifera</i>	80	80	20	76
N.J.A 42	<i>P. armeniaca</i>	86	86	14	81
	<i>P. cerasifera</i>	79	79	21	75

Table 2 analysis it can be seen that in the field, to a height of 80 cm, the diameter of grafting section was 4 mm at N.J.A 42 and Goldrich varieties, 3.5

mm at Favorit variety and 5 mm at Umberto variety. Tree height at the time of stratification was between 1.80 m for Favorite variety and 2.8 m at Goldrich and Umberto varieties.

Of the total of 400 trees review (100 for each variety grafted) 327 were obtained from trees, gripping STAS being between 81-84% for Favorit, Umberto and Goldrich varieties, and the N.J.A. 42 has obtained 80%.

Tabel 2

**Scion increase after gripping on the rootstock *P. armeniaca***

Grafted variety	Rootstock used	Diff. between rootstock and scion diameter (mm)	Scions behavior		Standard trees obtained	
			Diameter at 80 cm height (mm)	Height (m)	Pcs.	% from total
<b>Goldrich</b>	<i>P. armeniaca</i>	4.7	4	2.80	100	84
<b>Umberto</b>	<i>P. armeniaca</i>	3.2	5	2.80	100	82
<b>Favorit</b>	<i>P. armeniaca</i>	4.5	3.5	1.80	100	81
<b>N.J.A 42</b>	<i>P. armeniaca</i>	4.6	4	2.70	100	80
<b>Total</b>					<b>327</b>	

In the case of grafting on *P. cerasifera* (tab. 3) may find that the tree diameter at 80 cm above ground level was 4 mm to 5 mm for Favorite, Umberto N.J.A.42 varieties and 6 mm at Goldrich variety.

The height of the trees obtained was between 1.60 m from Favorite variety and 2.50 m in N.J.A. 42 and Umberto varieties.

Of the total of 400 trees taken into study were obtained at the end of the second year, 303 trees, lowest value on the N.J.A. 42, and highest value on the Umberto variety.

Table 3

**Scion increase after gripping on the rootstock *P. cerasifera***

Grafted variety	Rootstock used	Diff. between rootstock and scion diameter (mm)	Scions behavior		Standard trees obtained	
			Diameter at 80 cm height (mm)	Height (m)	Pcs.	% from total
<b>Goldrich</b>	<i>P. sativa</i>	4	6	2.40	100	79
<b>Umberto</b>	<i>P. sativa</i>	5	5	2.50	100	80
<b>Favorit</b>	<i>P. sativa</i>	6	4	1.60	100	76
<b>N.J.A. 42</b>	<i>P. sativa</i>	5	5	2.50	100	70
<b>Total</b>					<b>303</b>	

## CONCLUSIONS

All varieties grafted onto the that two rootstock recorded a percent closely to the standard (STAS).

Technical conditions for 1 year grafted trees emphasized that both systems (radicular and aerial) fall within the quality limits for apricot propagated material.

Both rootstocks can be used successfully used in orchards in NE Romanian area.

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**PHOTOSYNTHETIC EFFECTS OF LIGHT-EMITTING DIODE (LED) ON IN VITRO-DERIVED STRAWBERRY (*FRAGARIA X ANANASSA* CV. FESTIVAL) PLANTS UNDER IN VITRO CONDITIONS**

**EFECTELE FOTOSINTETICE ALE DIODEI ELECTRO-LUMINISCENTE (LED) ASUPRA PLANTELOR DE CĂPȘUN (*FRAGARIA X ANANASSA* CV. FESTIVAL) OBTINUTE ȘI CRESCUTE ÎN CONDIȚII IN VITRO**

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**Abstract.** Purpose of this study was to investigate effect of different photosynthetic photon flux density (PPFD) conditions using LED lamps on culture of shoot explants derived from in vitro shoots of *Fragaria x ananassa* cv. Festival. To examine the combined effect of 55% red LED, 15% far red LED, 10% blue LED, 15% green LED and 5% warm light LED light on in vitro growth of plantlets, fresh and dry plant biomass, plant height, leaf area, number of shoot, shoot length, number of root and percentage of rooting and various growth of micro-propagated plants were assessed under four light intensities (25, 50, 75 and 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD; TRT2, TRT3, TRT4, TRT5, respectively). Un-rooted strawberry shoots were cultured in the "Culture Pack"-rockwool system with MS medium under CO<sub>2</sub>-enriched condition. The best response for regeneration of shoots and root induction was observed for shoot explants obtained on MS supplemented with BAP 1mg/l and NAA 0.5 mg/l at TRT3 (50  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD). Chlorophyll and net photosynthesis were optimal in plants grown under TRT4 (75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD). Stomatal resistance and Fv/Fm values were highest at low light irradiance (TRT2). The highest efficiency and high frequency of shoot formation occurred after 30 days. Elongation of shoot buds was obtained at TRT4 (75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD) and TRT5 (100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD) on the same MS medium. Regenerated shoots rooted best on the same medium of elongation. Irradiance at 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD was suitable for the acclimatization of strawberry plants. We concluded that the present protocol can be efficiently used for mass propagation of the strawberry.

**Key words:** Strawberry, *Fragaria*, LED lamp, light intensity, micro-propagation, direct organogenesis

**Rezumat.** Scopul acestui studiu a fost de a investiga efectul diferitelor densități ale fluxului fotonilor fotosintetici (PPFD) în condițiile folosirii lămpilor LED asupra lăstarilor transplantați care provin din lăstari "in vitro" de *Fragaria x ananassa* cv. Festival. Pentru a examina efectul combinat al luminii compuse din 55% LED roșu, 15% LED roșu pal, 10% LED albastru, 15% LED verde și 5% LED cu lumină caldă asupra dezvoltării "in vitro" a plăntuțelor, a producției de biomasă proaspătă și uscată, a înălțimii plantelor, a suprafeței frunzelor, a numărului de lăstari, a lungimii acestora, a numărului de rădăcini și a procentului de înrădăcinare s-au folosit patru intensități luminoase diferite (25, 50, 75 și respectiv 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD; TRT2,

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*TRT3, TRT4, TRT5). Lăstarii de căpșun fără rădăcină au fost cultivați folosind sistemul "Culture Pack"-cu vată minerală într-un mediu MS îmbogățit cu CO<sub>2</sub>. Cel mai bun răspuns pentru regenerarea lăstarilor și a rădăcinilor a fost observat pentru lăstarii crescuți într-un mediu MS suplimentat cu BAP 1mg/l și NAA 0.5 mg/l pentru TRT3 (50 μmol m<sup>-2</sup> s<sup>-1</sup> PPFD). Clorofila și fotosinteza au fost optime la plantele crescute sub TRT4 (75 μmol m<sup>-2</sup> s<sup>-1</sup> PPFD). Rezistența stomatelor și raportul Fv/Fm au avut cele mai mari valori în condițiile unei iradiere luminoase scăzute (TRT2). Cea mai mare mare eficiență și frecvență a formării lăstarilor a fost obținută după 30 de zile. Elongația lăstarilor a fost obținută pentru TRT4 (75 μmol m<sup>-2</sup> s<sup>-1</sup> PPFD) și TRT5 (100 μmol m<sup>-2</sup> s<sup>-1</sup> PPFD) pe același mediu MS. Regenerarea rădăcinilor lăstarilor a fost cea mai bună la utilizarea aceuiași mediu ca și în cazul elongației. Iradierea la 75 μmol m<sup>-2</sup> s<sup>-1</sup> PPFD a fost benefică pentru aclimatizarea plantelor de căpșun. Concluzionăm că prezentul protocol poate fi utilizat cu eficiență în cazul plantelor de căpșun.*

**Cuvinte cheie:** căpșun, *Fragaria*, lampă LED, intensitate luminoasă, micropropagare, organogeneză directă

## INTRODUCTION

According to Food and Agriculture Organization (Anonymous, 2012) of the United Nations, world production of strawberries has exceeded 4 million tons since 2007 (Wu *et al.*, 2012). Currently, Turkey stands second in the world for strawberry production with an annual production of 353 173 tones (Anonymous, 2012). The future of strawberry production and sales is very positive. Increased demand in the future from European countries should increase export demand on Turkish growers. Strawberry production, sale price, trade (import and export), and consumption are expected to continue to increase over the next several years.

The conventionally propagated plants and propagated with tissue culture plants of differences in performance have been investigated for strawberry. The strawberry propagated plants with tissue culture were shown to have greater flower number and higher yield, smaller fruit size, more runners and increased vigor as compared to conventionally propagated plants. Methods to reduce input costs and increase productivity of *in vitro* propagation are needed to improve *in vitro* culture conditions for successful strawberry micropropagation. Numerous studies described an *in vitro* method for mass propagation of strawberry through axillary shoot multiplication (Boxus, 1974; Cameron and Hancock, 1986). The micropropagation of strawberry was achieved and now it seems to be a routine task in many commercial laboratories. Numerous studies have been published regarding field behaviour of micropropagated strawberry. Adventitious (stipular) shoots occurred spontaneously, and were common in strawberry *in vitro* shoot cultures. The special conditions during *in vitro* culture result in the formation of strawberry plantlets of normal anatomy, morphology and physiology.

Plants require temperature, light, CO<sub>2</sub>, water and nutrients at optimal level to grow and develop (Yamori *et al.*, 2010). The light is the factor influencing the growth of green plants in *in vitro* growing. If lighting is too excessive, light generates oxygen radicals and causes photoinhibition. However, if lighting is too

weak, etiolating symptoms appear and photosynthesis cannot work efficiently. Both phenomena strongly limit primary productivity (Barber and Andersson, 1992; Solymosi and Schoefs, 2010). Light intensity and quality have been characterized as important factor for plant development and growth *in vitro* (Macedo *et al.*, 2011). At high light intensity, an increase in photosynthetic carbon fixation can occur, which varies depending on light intensity and may lead to different susceptibilities to photo-inhibition. But, under conditions of excess light absorption, the chloroplast lumen becomes acidic in nature, which reduces the electron transport chain, and excitation energy accumulates within the chloroplast which can lead to the generation of reactive oxygen species (ROS) such as superoxide and hydrogen peroxide. The accumulation of ROS may lead to lipid peroxidation and to reduced accumulation of enzymatic antioxidants which exist as a defense system in plants (Asada, 1999). High photosynthetic flux of photoautotrophically micopropagated plant, cultured under high CO<sub>2</sub> concentration and high photosynthetic photon flux (PPF) has been observed in some plant species. Many studies have clearly showed that modulation in light photoperiod, quantity and quality can affect plant development and growth (Zuchi and Astolfi, 2012). Plants react to light mainly via photomorphogenetic, photosynthetic, and phototropic response (Kaur, 2015). These responses depend on the photoperiod, light quality and photon flux density (PFD) (Taiz and Zeiger, 1991). The Fv/Fm value, a measure of the intrinsic or maximum efficiency of PSII i.e. the quantum efficiency if all PSII centres were open, decreased significantly in plants grown and development. The decrease in this parameter indicates the down regulation of photosynthesis or photo-inhibition.

The fluorescent lamps were the main light source commonly used for *in vitro* cultivation of plants. However, conventional light sources (such as fluorescent light (especially cool-white), metal halide, high pressure sodium, incandescent and fluorescent lights) have some limitations due to their short lifetime, high electrical consumption and heat emission. Fluorescent lamps have fixed emission spectra composed of many bands in the wavelength range from 320 to 800 nm without the possibility of varying illumination of spectrum and time characteristic parameters. One of the disadvantages of fluorescent lamps is the difficulty of controlling light quality, which has been shown to have significant influences on plant morphogenesis.

To considerable interest for general plant photo-physiology, these responses are of commercial importance for *in vitro* plant cultivation. In terms of both sustainability and economics, new lighting technologies such as light-emitting diodes (LEDs) thus were necessary to be developed (Sheng, 2013). In recent years, the use of LEDs as an irradiation source for plants growing and propagation has attracted attention due to its vast potential for commercial application (Bula *et al.*, 1991). LEDs are particularly suitable for plant growth chambers, because of their light weight, small volume and long life (Yeh and Chung, 2009). Narrow bandwidths and wavelength specificities of LEDs have been

used in morphogenesis (Robin *et al.*, 1994), photosynthesis (Tennessen *et al.*, 1994), photo-biological research (Tripathy and Brown, 1995) and algal photobioreactor (Lee and Palsson, 1994). Among artificial lighting systems, LEDs present the maximum photosynthetically active radiation (PAR) efficiency (80–100%). LEDs emitting blue, green, yellow, orange, red and far red are available and can be combined to provide either high fulence (Bula *et al.*, 1991). LEDs, which generate virtually no heat, have very low energy consumption and estimated lifetimes of several years offer a simple and economic solution to the problem of creating controlled environments. Compared to conventional fluorescent lamps, LED based illuminators have improved features, including longer life-time, solid state construction, tailored spectrum, smaller mass and volume (Brown *et al.*, 1995; Bula *et al.*, 1991). Because of these culture of plants in a tightly controlled environment such as space-based plant culture systems. Thus, LEDs-based illuminators provide an alternative to fluorescent lamps as a light source with controllable spectrum that can be used for plant cultivation (Bula *et al.*, 1991; Barta *et al.*, 1992; Brown *et al.*, 1995; Žukauskas *et al.*, 2002; Tamulaitis *et al.*, 2005).

The LEDs technology is predicted to replace HID and fluorescent lamps in horticultural systems and to revolutionize controlled growth environments (Eva *et al.*, 2015). The LEDs have features which are far better than the commonly used irradiation source. Investigation of the effect of illumination spectrum and photon flux density (PFD) has been carried out by applying LED-based illumination to some species of plants on plant growth *in vitro*. The efficiency of 650–665 nm wavelengths of red LEDs on plant growth is fit with the absorption peak of chlorophylls (Schoefs, 2002) and phytochrome, while the supplemented blue light introduced the idea that growth under natural light could be mimicked using red and blue LEDs. In addition to providing an excitation of the different types of photoreceptors, the red+blue combination allowed a higher photosynthetic activity than that under either monochromatic light (Sabzalian *et al.*, 2014). The supplementation of blue+red LEDs could also be complemented with green (460–475 nm) LEDs. Illumination with containing up to 24% green LED light enhanced in plant growth (Kim *et al.*, 2006). Hypocotyl elongation could be prevented by adding at least  $15 \mu\text{mol m}^{-2} \text{s}^{-1}$  of blue light (Hoenecke *et al.*, 1992). Under red LEDs illumination, phytochrome stimulation is especially high as far red light is not provided.

The attractive features of the LEDs system, and the acquired knowledge about the effect of light quality on plant photosynthesis, morphogenesis and chlorophyll synthesis studies on growth and development of strawberry plants under different LEDs systems are very limited (Nhut *et al.*, 2003), whereas are more advanced on horticultural plant species (Miyashita *et al.*, 1995; Tanaka *et al.*, 1998; Hahn *et al.*, 2000; Lian *et al.*, 2002; Nhut *et al.*, 2003; Kim *et al.*, 2004; Jao and Fang, 2004; Heo *et al.*, 2006; Stutte *et al.*, 2009; Li *et al.*, 2012). Micro-propagation of strawberry shoots grown using LEDs were previously reported by Nhut *et al.* (2003). Nhut *et al.* (2003) showed that the best growth of strawberry plantlets

cultured *in vitro* was observed under LED-based illumination, and the optimal total photon flux density (PFD) were found to be  $60 \mu\text{mol m}^{-2} \text{s}^{-1}$  with 70% red and 30% blue spectral components. Tripathy and Brown (1995) showed that wheat seedlings accumulated chlorophyll content under red LED light at  $100 \mu\text{mol m}^{-2} \text{s}^{-1}$ , but not at  $500 \mu\text{mol m}^{-2} \text{s}^{-1}$ . This inhibition of chlorophyll accumulation under high fluence red LED light could be avoided by the supplementation of blue LED light at  $30 \mu\text{mol m}^{-2} \text{s}^{-1}$ .

This research was aimed to examine the effects of LED lighting on the growth and development of strawberry plantlets in *in vitro* and on the changes in morphology and physiology of *in vitro* cultured strawberry explants, which were cultured *in vitro* under illumination system with four photon flux densities using LEDs. We tested the application of traditional fluorescent lighting and LED lighting in relation to growth in strawberry in *in vitro*.

## MATERIAL AND METHOD

For the experiment, the strawberry plantlets were multiplied at the Plant Tissue Culture Laboratory of the Agricultural Biotechnology Department, Faculty of Agriculture, Süleyman Demirel University. Strawberry plantlets were micro-propagated from culture of shoot explants derived from *in vitro* shoots of *Fragaria x ananassa* cv. Festival.

The axillary shoots (micro-shoots) were obtained from a mass of shoots cultured *in vitro* on Murashige and Skoog (MS) basal medium. The axillary shoots were excised and placed on solid MS medium for *in vitro* culture. These axillary shoots were then transferred onto solid  $\frac{1}{2}$  MS medium supplemented with 6-benzylaminopurine (BAP; 2 ppm), indole-3-acetic acid (IAA; 0.1 ppm), ascorbic acid (80 ppm), TDZ (2.0 ppm), giberrellic acid ( $\text{GA}_3$ ; 0.2 ppm), phloroglucinol (100 ppm), sucrose (30 g/l) and gelrite (3 g/l) at pH 5.6. The root medium was composed of  $\frac{1}{2}$  MS nutrients, supplemented with gelrite (3 g/l), sucrose (20 g/l),  $\text{GA}_3$  (0.2 ppm) and IBA (1.0 ppm) at pH 5.6 before autoclaving at  $121^\circ\text{C}$  for 20 min. Axillary shoots were transplanted into 250 ml Maganta vessels with 50 ml medium in each vessel. Five axillary shoots were placed in each vessel onto a fresh proliferation medium and subcultures were obtained in 30 days. Five axillary shoots cultured in one vessel and 9 vessels were used for each treatment. The experiment included 3 replicates with consisting of 15 axillary shoots per replication. Vessels were sealed with a rubber stopper that had a hole with permeable film to provide ventilation.

These culture vessels were placed in the grown chamber in which the  $\text{CO}_2$  concentration was maintained at  $1000 \text{ mg m}^{-3}$ . For comparison, they were also placed on the shelf under PFD ( $45 \mu\text{mol m}^{-2} \text{s}^{-1}$ ) (control) and different photosynthetic photon flux density (PPFD) of LEDs light in the same grown chamber.

To elucidate the effect of different PPFD of LEDs light conditions at the *in vitro* growth of strawberry plantlets, the cultures were established and grown under different light densities of PPFD. The light was provided by LED lamps and cool-white [photon flux density (PFD) of  $45 \mu\text{mol m}^{-2} \text{s}^{-1}$  (for control)] (TRT1) fluorescent lamps placed under of shelf in the growth chamber. The cultures were kept in a growth chamber and the cultures of *in vitro* plantlets were illuminated using four different PPFD of LEDs light intensities in order to select the most suitable LED light intensity. Experiments conducted in growth chambers were programmed on a 24-h cycle; irradiance was set at a PPFD of LEDs light [25, 50, 75,  $100 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD (TRT2, TRT3, TRT4, TRT5, respectively)] and the combined effect

of 55% red LED (peak wavelength 640 nm), 15% far red LED (735 nm), 10% blue LED (450 nm), 15% green LED (510 nm) and 5% warm light LED and were tested. Irradiance was measured routinely at the centre of vessels (at the top of the plants of in the vessel) with a quantum sensor. The irradiation level measured at the center of vessel was kept constant at PPF of LEDs light and PFD of fluorescent lamps light (control). The cultures were exposed in a photoperiod of 16 h light and 8 h dark. Culture vessels were transferred to grown chamber and maintained at  $24\pm 2^{\circ}\text{C}$ , and 60-70 % relative humidity (RH). RH was controlled by an ultrasonic fog system. Air temperature and relative humidity were measured with the JUMO Humitherm TDAC-70.

The apparatus which was used in LED experiments was an aluminium panel [width: 35 length: 90 cm; 3 pieces per panel: 25 (width), 30 (length) cm]. As a light source, the LED panel were arranged and were mounted on the 30 cm height from vessel. A total of seventy five culture vessels were placed on five layers of shelves in a growth chamber, 15 vessels per shelf. For fast micro-propagation, during cultivation, the explants were transferred to multiplication solid medium (same as above) by dividing the axillary shoot of each sample. After cultivation, axillary shoots of the surviving plantlets were allowed to elongate, and each axillary shoot was divided and transferred on to a proliferation medium. Multiple axillary shoots were produced one month. These axillary shoots were each separated, and sub-cultured onto a fresh solid MS medium. Finally, 45 axillary shoots (~2 cm long) were cut off and transferred onto a rooting medium. Rooted strawberry plantlets were transferred to growth chamber for acclimatization. The test was performed in an entirely randomized experimental design with three replications, each replication represented by vessels with fifteen axillary shoots and 45 axillary shoots per light intensity were used. Five axillary shoots was placed per vessel.

Three culture vessels were taken out to measure the growth characteristics of strawberry plantlets. The samples were obtained by random. For measure, the plantlets were harvested at 30 day in the middle of the light period, washed to remove medium particles and separated into leaves, shoot and roots. End of 30 days of cultivation were observed for their survival rate, total number of axillary shoots per explant, length of axillary shoots (cm), fresh and dry plant biomass, plant height, leaf area, number of shoot, shoot length, number of root and percentage of rooting. Plant growth at the cool-white fluorescent light *in vitro* derived axillary shoots was used as control. At the end of four week period, axillary shoots were assessed according to their regeneration capacity (number of axillary shoots). The samples were prepared from young leaves of *in vitro* propagated axillary shoots. Fifteen leaves from the top axillary shoots with a 2.5-5.0 cm length of axillary shoot were collected for each light intensity. Leaves were cut out from the axillary shoots for evaluating extended. PGF of cool-white fluorescent light on strawberry leaf samples were used as controls to compare with the LEDs light intensities material. The LEDs light assayed and non- assayed plantlets were used. The control consisted of untreated plantlets. Growth of plantlets were observed for their plantlets of the surviving (survival rate) data were recorded.

Microropogated strawberry plantlets *in vitro* conditions from axillary shoots were adapted under grown chamber conditions. After one month acclimatized plantlets were transferred to the in plug-mix compost (in 45 celled transplant trays, filled with equal parts by volume of vermiculite, polystyrene granules and peat) and maintained at  $24\pm 2^{\circ}\text{C}$ , 50-60 % relative humidity. The trays were placed on shelves in a growth chamber on a capillary mat and subirrigated. Strawberry plantlets were fertilized with a water-soluble fertilizer 20N-8P-16K at a rate of 2.5 g/l every four irrigation until approximately 30 days of cultivation in a grown chamber.

Five explant of each light treatment replication were harvested to calculate

mean of total plantlet height, fresh plantlets and dry plantlets biomass, leaf area, number of shoot, shoot length, number of root and percentage of rooting. Fresh weights of strawberry plantlets as well as those of stems, leaves and roots were measured. For determination dry weights of whole plantlet were dried at 65°C until constant weight was reached. The plantlets were dried in a hot air oven for three days and the weight at ambient temperature was recorded as the total dry weight.

Leaf area of the representative strawberry plantlets was measured by a digital planimeter (Placom KP-90N).

The chlorophyll *a+b* contents in the fully expanded young leaves of 4-week-old plants was measured. PFD of fluorescent lamps light treatment of leaf samples were used as control to compare with the PPFD of LEDs light treated material. Total chlorophyll *a+b* content were quantified in samples formed by five leaf discs (1,283 cm<sup>2</sup>) cut from in the youngest fully expanded leaf (Porra *et al.*, 1989; Wellburn, 1994; Schoefs, 2002). At the chlorophyll *a+b* contents of leaflets were calculated (in chlorophyll fluorescence) and determinations were subsequently expanded leaf using 100 mg fresh weight extracted with 10 ml of 80% cold acetone at 4°C and determining the absorbance at 645 and 663 nm, calculations were made using the spectrophotometrically according in Serret *et al.* (1996) and Porra *et al.* (1989). The minimum chlorophyll fluorescence yield in dark (F<sub>0</sub>) was elicited. The maximum chlorophyll fluorescence yield in dark (F<sub>m</sub>) and in light (F<sub>m</sub>) were induced by a flash of intense white light from the light source PPFD of LEDs light and PFD of fluorescent lamps light. From chlorophyll *a+b* contents, fluorescence kinetic parameters, variable to maximum fluorescence ratio (F<sub>v</sub>/F<sub>m</sub>) was measured in strawberry plantlets grown *in vitro* conditions 30 days under 1000 mg m<sup>-3</sup> CO<sub>2</sub> concentration. The maximum photochemical efficiency was calculated in 30-min-dark-adapted leaves as (F<sub>m</sub>-F<sub>0</sub>)/F<sub>m</sub>=F<sub>v</sub>/F<sub>m</sub>. Variable to maximum fluorescence ratio (F<sub>v</sub>/F<sub>m</sub>) measured originally photoautotrophically grown plantlets *in vitro* in irradiance leaves adapted Kadleček *et al.* (1998). [(F<sub>v</sub>/F<sub>m</sub> = Variable chlorophyll fluorescence / maximum chlorophyll fluorescence) [maximum photochemical efficiency F<sub>v</sub>/F<sub>m</sub>; F<sub>v</sub> (F<sub>m</sub>-F<sub>0</sub>; variable fluorescence), F<sub>0</sub> (minimum fluorescence), F<sub>m</sub> (maximum fluorescence), and F<sub>v</sub>/F<sub>m</sub> (variable / maximum ratio fluorescence)].

The photosynthetic rate (P<sub>G</sub>) was measured at the strawberry plantlets grown in the *in vitro* conditions under 1000 mg (CO<sub>2</sub>) m<sup>-3</sup> concentration of the in-flow air. CO<sub>2</sub> concentration was increased to 1000 mg m<sup>-3</sup> concentration during the light period. At the CO<sub>2</sub> level in the growth chamber was measured by the CO<sub>2</sub> supply apparatus. A portable LI-6400 apparatus (LI-COR Inc., Lincoln) was used to measure net assimilation of CO<sub>2</sub> (A<sub>CO<sub>2</sub></sub>) in response to increasing levels of light. The CO<sub>2</sub> was set to 1000 mg m<sup>-3</sup> CO<sub>2</sub> (A<sub>CO<sub>2</sub></sub>) during the light period in all treatments. Net photosynthetic rate (P<sub>N</sub>) at the P<sub>N1000</sub> CO<sub>2</sub> concentration measured at 1000 mg (CO<sub>2</sub>) m<sup>-3</sup> in strawberry plantlets grown *in vitro* conditions under 1000 mg m<sup>-3</sup> CO<sub>2</sub> concentration. CO<sub>2</sub> concentration was increased to 1000 mg m<sup>-3</sup> during the light period. Adaxial stomatal resistance (cm s<sup>-1</sup>) was measured in strawberry plantlets grown *in vitro* conditions. Leaf temperature was 25°C and RH was 70%.

The experiment had one factorial design with four PPFD of LEDs light intensities levels and one PFD of fluorescent lamps light intensity, each replication represented by 45 explants per light intensity treatment were used. The statistical one-way analysis of variance was used to analyze data. The mean values were subjected to analysis using the Tukey-Kramer (JMP, Version 5) to determine the PPFD of LEDs light intensities the above parameters for a comparison of the averages. Mean separation between treatments by LSD was 5 percent level.

**RESULTS AND DISCUSSIONS**

Effects of LEDs light intensities on the morphology and physiology (leaf structure, root structure and photosynthesis) of strawberry plantlets grown *in vitro* were studied using an illumination system consisting of four different of light-emitting diodes (LEDs) intensities [25, 50, 75, 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light] and the combined effect of 55% red LEDs (peak wavelength 640 nm), 15% far red LEDs (735 nm), 10% blue (450 nm) LEDs, 15% green LEDs (510nm) and 5% warm light LEDs light of LEDs. Taking into account all differences in number of the transplanted plantlet, survival rate number of the plantlet, usable plugs, plantlet height, number of root, percentage of rooting, leaf area, elongation of axillary shoot length, total number of axillary shoot, axillary shoot regeneration, number of leaf, net photosynthetic rate ( $P_G$ ), stomatal resistance, Fv/Fm values, fresh plant biomass, total dry plant biomass, leaf chlorophyll content and elongation of root length from meristem of strawberry, observed while changing the PPFD of LEDs light intensities, the optimal total LEDs intensity for growth of strawberry plantlets *in vitro* was estimated. For 16 h photoperiod and the optimal total PPFD of LEDs light was found to be 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$ .

During *in vitro* culture, strawberry plantlets grew under four different PPFD of LEDs light intensities conditions in relatively air-tight cultivation vessels. To determine the effects of LEDs irradiation level, cultures were compared irradiation level at 45  $\mu\text{mol m}^{-2} \text{s}^{-1}$  cool-white fluorescent lamps. The strawberry (*Fragaria x ananassa* cv. Festival) plantlet growth was best at 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light intensity. The optimal PPFD of LEDs light intensity was at 75–100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light favored the growth of strawberry plantlets in *in vitro* (tab. 1, tab. 2).

The axillary shoot regeneration, leaf area, percentage of rooting, plantlet height, usable plugs, survival rate number of the plantlet (tab. 1); fresh plant biomass, photosynthetic rate ( $P_G$ ), net photosynthetic rate ( $P_N$ ), stomatal resistance and leaf chlorophyll *a+b* content (tab. 2) of plantlets in *in vitro* growth of strawberry plantlets under 75–100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light were significantly greater than low PPFD of LEDs light (25  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) of PPFD of LEDs light intensity (tab. 1, tab. 2). The number of root, elongation of axillary shoot length, total number of axillary shoot (tab. 1), total dry plant biomass, fluorescence ratio (Fv/Fm), number of leaf and elongation of root length (Table 2) of plantlets in *in vitro* growth of strawberry plantlets under 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light was not significantly lower than low PPFD (25  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light) of LEDs light intensity (tab. 1, tab. 22). A high PPFD of LEDs light at 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  had severely negative effect on plantlets.

The exposure of strawberry plantlets to high irradiance may be more optimal for survival rate number of the plantlet (89.2 % at 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) of strawberry plantlets than 87.1 % at 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  level of PPFD of LEDs light. But, at 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  light intensity level, the plantlets had smaller leaves (11.91  $\text{cm}^2$ ) than 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  level of PPFD of LEDs light (12.04  $\text{cm}^2$ ) and compact texture. At the 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  level of PPFD of LEDs light had better quality of pot plants than at other light intensities. In the survival rate number of the plantlet and usable plugs (for *ex vitro*

acclimatization), most of the significant differences were found at the different PPFD of LEDs light intensity and PFD light (control) *in vitro* culture conditions in *in vitro* plantlets (day 30), from to in the grown chambers. Most of the strawberry plantlets survived until the end of the *in vitro* growth. By the end of *in vitro* culture, strawberry plantlets had developed 89.2 - 93.4 % survival rate. The usable plugs *in vitro* growth in the grown chambers was different for the different PPFD of LEDs light intensities. Irradiance at  $75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD was suitable for the acclimatization (tab. 2).

Elongation of shoot buds was obtained at TRT4 ( $75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LED) and TRT5 ( $100 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LED).

The strawberry plantlets were taller, had higher dry mass of leaves, axillary shoots and roots, and larger leaf area. The formation of percentage of rooting during *in vitro* growth in the grown chambers was different for the different LED light intensities. The best response for regeneration of shoots and root induction was  $50\text{-}75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD (TRT3 and TRT4) of LED (tab. 1, tab. 2). The highest number of roots per plantlet (14.3 - 13.4) and percentage of rooting (65.4 - 68.6 %) were found under  $50 - 75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs (respectively), whereas a significantly lower number of roots per plantlet (13.2). Results showed that adventitious root formation was highest in plantlets cultured under  $75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light (68.6 %), whereas poor rooting was observed in plantlets grown under  $45 \mu\text{mol m}^{-2} \text{s}^{-1}$  cool-white fluorescent lamps light (45.4 %) (tab. 1). Similar results were reported in chrysanthemum (*Chrysanthemum morifolium*) (Kurilcik *et al.*, 2008) and anthurium (*Anthurium andreaeanum*) (Budiarto 2010), where red LEDs were also found to stimulate root formation.

The total dry plant biomass (plant dry) weight to leaf area increased in the PPFD of LEDs light intensities during *in vitro* grown in chambers while it remained more changed in the higher LED lights. Total dry mass of strawberry plantlets of grown PPFD of LEDs light intensities were higher than PGF light intensity. The total dry plant biomass (plant dry) weight of plantlets irradiated by the at  $50 - 75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light intensity were significantly higher than these grown under the other light treatments. The stimulatory effect of red + blue LEDs on leaf growth is similar to those reported in strawberry (*Fragaria ananassa* cv. Akihime) (Nhut *et al.*, 2003). Total dry plant biomass were significantly higher in 1.6 g/plant ( $75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LED light) than 1.3 g/plant (control,  $45 \mu\text{mol m}^{-2} \text{s}^{-1}$  PGF light) indicating a lasting effect of the different LED light intensities availability during in the *in vitro* growth (tab. 2).

Leaves from *in vitro* plantlets of strawberry had a normal development in leaves. At the end of growth in the growth chambers, 6.2 number of leaf ( $75 \mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LED light) had significantly higher leaf numbers than the 7.4 number of leaf ( $45 \mu\text{mol m}^{-2} \text{s}^{-1}$  PGF light) control plantlets, showing that this PPFD of LEDs light intensity treatment had been the most favourable for *in vitro* development (tab. 2).

Net photosynthetic rate ( $P_N$ ) in strawberry plantlets increased in the *in vitro* grown. Similar results were found by Baroja *et al.* (1995) and Van Huylenbroeck and Debergh (1996).  $P_N$  and in consequence biomass accumulation were increased at the  $1000 \text{ mg (CO}_2\text{) m}^{-3}$  had effect on strawberry plantlets growth *in*

*vitro* conditions 30 days under; this increase was more marked under higher LED light intensities. Similar results were found by Desjardins (1995).

Stomatal resistance and Fv/Fm values were highest at low light irradiance TRT2 (25  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light). Our study showed that the 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  highest PPFD of LEDs light intensity increased the plantlet extension and formation of roots; and simultaneously stomatal resistance and the Fv/Fm ratio were decreased; and content of chlorophyll *a+b* were increased (tab. 2). The highest stomatal resistance (0.72 – 0.69  $\text{cm s}^{-1}$ ) was recorded at 50 - 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (respectively). On the other hand, low stomatal resistance observed in strawberry plantlets increased after transfer to *ex vitro* conditions (tab. 2).

Net photosynthetic rate ( $P_N$ ) was increased in strawberry plantlets in the *in vitro*. The net photosynthetic rate was increased (tab. 2). Lee *et al.* (1985) reported that increasing PPFD of LEDs light to a high level could improve photosynthesis if other conditions necessary for a maximum photosynthetic rate were sustained, while a moderate PPFD of LEDs light level.

In strawberry plantlets cultivated to *in vitro* conditions under LEDs light intensity in an *in vitro* where irradiance varied during the daily illumination maximum was usually less than that needed for photosynthesis, no photoinhibition occurred: Fv/Fm was in the range typical for non-stressed plants and did not change during cultivation. Variable to maximum fluorescence ratio Fv/Fm were decreased at the highest PPFD of LEDs light intensities application of strawberry plantlets to *in vitro* conditions (tab. 2). The Fv/Fm value were decreased (0.554 - 0.539) significantly in strawberry plantlets grown and development in *in vitro* at 75 - 100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (respectively) (tab. 2). The decrease in this parameter indicates the down regulation of photosynthesis or photoinhibition.

Changes in plant grown were dependent on the PPFD of LEDs light intensity in the grown chamber during *in vitro* culture. In plantlets grown *in vitro* PPFD of LEDs light intensity, chlorophyll *a+b* content increased, in plantlets grown *in vitro*. Strawberry plantlets in growth chamber at low irradiance (25  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD) chlorophyll *a+b* content 279  $\text{mg g}^{-2}$  were found at low content, but chlorophyll *a+b* content was increased from the 50-100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LED light intensity. On the contrary, during cultivation at high irradiance (100  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LED) plant growth were much higher. The changes were most pronounced for 287  $\text{mg g}^{-2}$  chlorophyll content high light plantlets which had a considerably increased chlorophyll *a+b* content. Chlorophyll *a+b* content (291  $\text{mg g}^{-2}$ ) and photosynthetic rate (PG) (15.18  $\mu\text{g}(\text{CO}_2) \text{m}^{-2} \text{s}^{-1}$ ) were higher at 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light than at low PPFD of LEDs light (tab. 2).

Our results suggest that 75  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PPFD of LEDs light intensity are suitable as a light source to promote root (elongation of root length and percentage of rooting), leaf formation (number of leaf and leaf area), fresh plant biomass, total dry plant biomass, photosynthetic rate, net photosynthetic rate, stomatal resistance, leaf chlorophyll *a+b* content, plantlet height, elongation of axillary shoot length and axillary shoot regeneration in strawberry plantlets in *in vitro*.

Table 1

The effect of different LEDs light intensities on number of the transplanted plantlet, survival rate number of the plantlet, usable plugs, plantlet height, number of root, percentage of rooting, leaf area, elongation of axillary shoot length, total number of axillary shoot, axillary shoot regeneration from axillary shoots of strawberry (*Fragaria x ananassa* cv. Festival) at the *in vitro* conditions

Treatments	Number of the transplanted plantlet	Survival rate number of the plantlet (%)	Usable plugs (%)	Plantlet height (cm)	Number of root	Percentage of rooting (%)	Leaf area (cm <sup>2</sup> )	Elongation of axillary shoot length (cm)
TRT1	100.0	93.4 <sup>a</sup> ± 1.9	87.7 <sup>a</sup> ± 1.4	6.5 <sup>f</sup> ± 0.1	13.2 <sup>b</sup> ± 2.1	45.4 <sup>a</sup> ± 2.2	6.85 <sup>a</sup> ± 1.5	1.6 <sup>bc</sup> ± 0.6
TRT2	100.0	88.4 <sup>b</sup> ± 2.2	74.4 <sup>d</sup> ± 1.7	7.4 <sup>e</sup> ± 0.3	12.9 <sup>c</sup> ± 1.9	59.2 <sup>c</sup> ± 1.8	7.97 <sup>b</sup> ± 1.3	1.7 <sup>c</sup> ± 0.7
TRT3	100.0	88.2 <sup>b</sup> ± 1.5	77.6 <sup>c</sup> ± 1.5	8.3 <sup>d</sup> ± 0.4	14.3 <sup>a</sup> ± 1.7	65.4 <sup>a</sup> ± 1.7	9.19 <sup>a</sup> ± 1.8	1.8 <sup>a</sup> ± 0.5
TRT4	100.0	87.1 <sup>b</sup> ± 1.8	73.9 <sup>d</sup> ± 1.3	8.9 <sup>d</sup> ± 0.2	13.4 <sup>bc</sup> ± 2.3	68.6 <sup>a</sup> ± 1.5	12.04 <sup>a</sup> ± 1.1	1.7 <sup>c</sup> ± 0.5
TRT5	100.0	89.2 <sup>bc</sup> ± 2.1	70.5 <sup>d</sup> ± 1.9	9.0 <sup>d</sup> ± 0.4	13.9 <sup>a</sup> ± 1.6	63.1 <sup>bc</sup> ± 2.3	11.91 <sup>a</sup> ± 1.4	1.5 <sup>c</sup> ± 0.8

Significant differences between treatment effects were analyzed by regression analysis.

Values represent means ± SE followed by the different superscript letters show significant differences by Tukey HSD test (p<0.05).

Means within a column followed by the same letters are not significantly different at P<0.05 according to Duncan's multiple range test.

Table 2

The effect of different LEDs light intensity on plant dehydrins, number of leaf, net photosynthetic rate (P<sub>n</sub>), stomatal resistance, Fv/Fm values, fresh plant biomass, total dry plant biomass, leaf chlorophyll content and elongation of root length from axillary shoots of strawberry (*Fragaria x ananassa* cv. Festival) at the *in vitro* conditions

Treatments	Fresh Biomass (g plant <sup>-1</sup> )	Total dry plant biomass (g plant <sup>-1</sup> )	Photosynthetic rate (P <sub>n</sub> ) [μg(CO <sub>2</sub> )/m <sup>2</sup> ·s <sup>-1</sup> ]	Net photosynthetic rate (P <sub>n</sub> ) [μg(CO <sub>2</sub> )/m <sup>2</sup> ·s <sup>-1</sup> ]	Stomatal resistance (cm s <sup>-1</sup> )	Fluorescence ratio (Fv/Fm)	Number of leaf	Leaf chlorophyll a+b content (mg g <sup>-1</sup> )	Elongation of root length (cm)
TRT1	308.5 <sup>a</sup> ± 0.3	1.3 <sup>a</sup> ± 0.02	6.52 <sup>a</sup> ± 7.05	237.75 <sup>a</sup> ± 1.53	0.52 <sup>a</sup> ± 0.07	0.589 <sup>a</sup> ± 0.05	5.4 <sup>a</sup> ± 0.1	279 <sup>a</sup> ± 0.08	7.9 <sup>bc</sup> ± 0.4
TRT2	312.2 <sup>a</sup> ± 0.2	1.4 <sup>a</sup> ± 0.03	11.91 <sup>a</sup> ± 5.22	252.34 <sup>a</sup> ± 1.41	0.72 <sup>a</sup> ± 0.05	0.583 <sup>a</sup> ± 0.08	5.7 <sup>a</sup> ± 0.1	284 <sup>a</sup> ± 0.06	8.2 <sup>a</sup> ± 0.3
TRT3	314.7 <sup>a</sup> ± 0.2	1.5 <sup>a</sup> ± 0.04	12.75 <sup>a</sup> ± 7.48	261.20 <sup>a</sup> ± 1.58	0.69 <sup>a</sup> ± 0.04	0.576 <sup>a</sup> ± 0.07	5.9 <sup>bc</sup> ± 0.2	289 <sup>a</sup> ± 0.05	8.3 <sup>a</sup> ± 0.4
TRT4	317.9 <sup>a</sup> ± 0.1	1.6 <sup>a</sup> ± 0.02	15.18 <sup>b</sup> ± 8.12	269.23 <sup>a</sup> ± 1.03	0.63 <sup>bc</sup> ± 0.05	0.554 <sup>a</sup> ± 0.05	6.2 <sup>a</sup> ± 0.3	291 <sup>a</sup> ± 0.08	8.3 <sup>a</sup> ± 0.5
TRT5	318.6 <sup>a</sup> ± 0.2	1.6 <sup>a</sup> ± 0.03	16.41 <sup>b</sup> ± 6.31	275.14 <sup>a</sup> ± 1.60	0.58 <sup>b</sup> ± 0.06	0.539 <sup>a</sup> ± 0.06	5.9 <sup>bc</sup> ± 0.2	287 <sup>a</sup> ± 0.07	7.6 <sup>b</sup> ± 0.7

Values represent means ± SE followed by the different superscript letters show significant differences by Tukey HSD test (p<0.05).

Means followed by the same letters do not differ significantly at 5% level of significance.

## CONCLUSIONS

For photosynthetic parameters it seems very important at which conditions *in vitro* plantlets have been grown.

According to this, PPF of LEDs light intensities can be increased in photosynthetic parameters.

Further, an increase in chlorophyll content and net photosynthetic rate is usually observed in dependence on the PPF of LEDs light intensities during *in vitro* grown.

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## GROWTH AND PRODUCTIVITY OF PLUM TREES DEPENDING ON THE FORM OF THE CROWN AND VARIETY

### CREȘTEREA ȘI FRUCTIFICAREA POMILOR DE PRUN ÎN FUNCȚIE DE FORMA DE COROANĂ ȘI SOI

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**Abstract.** *In order to reduce the growth vigor of plum trees and increase productivity, it was studied the improved bush crown form.*

*Trees with 3, 4 and 5 main branches took part in the experiment. The control group was presented by the “mixed pyramid” crown that is recommended for plum trees in Moldova. Twelve years of research have shown that the improved bush crown contributes to the rapid development of the vegetative mass of trees when they are young, the reduced size of the crown (by 32-40%) and to almost doubled increased orchard productivity. The most productive was the option of improved bush with 5 main branches.*

**Key words:** plums, growing power, form of the crown, variety, yield

**Rezumat.** *Cu scopul reducerii vigoriei de creștere a pomilor de prun și majorarea productivității, a fost studiată forma de coroană tufa ameliorată. În studiu au fost luați pomii conduși cu 3, 4 și 5 ramuri de schelet. Ca martor au servit pomii conduși după forma de coroană piramida mixtă recomandată pentru Republica Moldova. În rezultatul a 12 ani de studii s-a determinat că forma de coroană tufa ameliorată favorizează acumulării rapide a masei vegetale în perioada de creștere, diminuarea parametrilor coronamentului cu 32-40% și majorarea productivității plantației de aproximativ de două ori. Cea mai productivă s-a adevărit a fi varianta unde pomii conduși după forma de coroană tufa ameliorată cu 5 ramuri de schelet.*

**Cuvinte cheie:** prun, vigoarea de creștere, forma de coroană, soi, productivitate

## INTRODUCTION

Productivity and economic efficiency of plum trees in present are considerably lower than those of apple trees, the intensification level of which is the highest among fruit species. The major problem is the large size of plum trees due to the lack of reliable dwarf rootstocks, which leads to a significant reduction in fruit productivity because of increased costs of pruning, and especially harvesting (Cimpoieș, 2000; Журавель *et al.*, 2007; Grădinaru and Istrate, 2009). Large crown parameters also constrain the increase in the density of plantations, which ultimately affects lower crops in plum orchards (Cociu *et.al.*, 1997; Sitarek, 2006).

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At present, such factors as the growing power, the form of the crown and varieties fruiting particularities are not taken into account. Therefore, the search for agronomic techniques is the main way to reduce the growing power of plum trees and, consequently, to increase the density of plantations (Журавель *et al.* 2007; Rîbințev, 2012; Popa *et al.*, 2015).

The development of crowns with a rational deployment of branches in the space, which allows us to improve the access to light and to reduce the size of plants, is one of these techniques. The improved bush forming that has been subject to studies for 12 years in Moldova is very interesting in this regard.

## MATERIAL AND METHOD

In spring 2005 we founded an experimental garden to study the improved bush form of crowns at the experimental station "Criuleni". The garden included 1-year old trees of the Kabardinskaia ranniaia and Stanley plum varieties, grafted on seedling rootstock (*P. cerasifera*), which differ in the nature of growth and fruiting.

When we created an improved bush crown form, trees were shortened after planting at the height of 20-25 cm from the soil surface and during the vegetation period there were created variants with 3, 4 and 5 main branches. All the branches that were growing inside the crown were cut for sunlight rays and the crown looked open centre. The plantation scheme is 5 x 2 m.

The control trees were formed according to the naturally-improved system (a mixed pyramid) and planted under the scheme 6 x 4 m, which is recommended for plum trees in the Republic of Moldova.

The experiment was founded in 3 repetitions, each repetition representing 12 fruit trees.

The research was carried out, applying conventional methods that are widely used when we work with fruit crops. Crown parameters, the average and the total shoot length were measured at the end of the growing season, the crop yield and the number of fruits were counted 2 weeks before harvest, in 3 registered trees for three times.

The statistical processing of the data obtained in the study was done according to B. Dospekhov (1985), with the use of computer programs.

## RESULTS AND DISCUSSIONS

The conducted research showed that the vegetative mass of young plants increases faster and more intensively if they are formed according to the improved bush system.

The correlation between the root and the shoot systems in improved bush is almost completely removed. The total length of annual shoots, as one of the indicators that characterizes processes of growth and their intensity in plum trees, which are younger than 6 years old, was bigger in improved bush crowns (tab. 1).

Thus, the total shoot length in the variant with 5 main branches exceeded the control group by 14.5 m in Stanley variety and by 19.2 m in Kabardinskaia ranniaia variety. Since 2011, the total length of shoots is higher in the control group, where the trees are formed according to the naturally improved system.

This is explained by the fact that by the age of 7 years, processes of growth in the improved bush form are constrained by a density plantation and a more restrictive pruning of trees.

Table 1

The total length of shoots depending on the crown form and variety

Variety	The form of the crown	The number of branches in the bush	Years					
			2010	2011	2012	2013	2014	2015
Kabardinskaia ranniaia	Mixed pyramid (control)	-	74.4	97.4	62.7	64.3	66.7	61.9
	Improved bush	3	80.3	88.2	58.7	60.1	52.3	55.7
	Improved bush	4	88.7	92.3	60.4	52.7	61.4	57.3
	Improved bush	5	93.6	96.4	63.2	63.9	63.7	58.4
Stanley	Mixed pyramid (control)	-	87.7	108.2	74.8	83.1	92.6	79.5
	Improved bush	3	95.6	93.8	71.4	78.7	82.4	75.1
	Improved bush	4	98.7	96.8	73.7	80.2	82.9	77.4
	Improved bush	5	102.2	100.5	73.3	82.1	84.0	78.2
LSD <sub>0,05</sub>			2.36	7.07	2.34	2.40	2.45	2.21

Having analyzed the influence of the number of main branches on the processes of growth in plum trees, one should note that the more main branches we have, the bigger the total shoot length is regardless of the variety. Moreover, the difference between varieties is bigger in young plants; it diminishes as they grow older. For instance, in 2011 the difference between the variants with 5, 4 and 3 main branches was 4.1-8.2 m in Kabardinskaia ranniaia variety, while in 2015 it was only 1.0-2.7 m.

Having analyzed the processes of growth in plum trees by years, we can state that the maximum value of the total shoot length was observed in 2011 and ranged from 88.2 m/tree in Kabardinskaia ranniaia variety to 108.2 m/tree in Stanley variety. This indicator decreased in the following years. It was significant enough in 2012, as it was quite a dry year and the annual rainfall was only 146.1 mm. It was low in 2015 too, when we got the maximum yield of fruits.

Generally speaking, one should mention that the annual growth of shoots was stabilized by the 7<sup>th</sup> year and ranged from 52 to 64 m/tree in Kabardinskaia ranniaia variety and from 71 to 92 m/tree in Stanley variety that is more vigorous.

The analysis of the tree size (tab. 2) shows that in 2015 the height of trees in Stanley variety in case of the improved bush form system was by 140-156 cm lower in all the variants than in the control group, while it was 161-173 cm in Kabardinskaia ranniaia variety. It is first of all explained by the fact that this form of the crown has no standard and it has from 3 to 5 main branches of the same thickness and they redistribute nutrients among themselves and the overall height of trees is generally reduced.

It was also found out that the improved bush form promotes decreased width of the crown.

*Table 2*

**Crown parameters in plum trees, depending on the form of the crown and variety, the experimental station "Criuleni", 2015**

The form of the crown	The number of branches in the bush, pcs.	Kabardinskaia ranniaia		Stanley	
		The height of trees, m	The width of the crown	The height of trees, m	The width of the crown
Mixed pyramid (control)	-	4.35	3.15	4.56	3.92
Improved bush	3	2.74	2.10	3.16	2.31
Improved bush	4	2.75	2.10	3.08	2.36
Improved bush	5	2.62	2.15	3.00	2.42

For example, the width of the crown in Stanley variety with 3 main branches was 38% less and the one with 5 main branches was 41% less than in the control group.

The main efficiency indicator of the studied forms of crowns is their yield (tab. 3).

According to the data received during the experiment, the most productive were the trees with the improved bush form of crowns, regardless of the number of main branches; the productivity was on average 34-44% higher than in the control group. First and foremost, this is explained by a smaller size of trees with improved bush crowns and, therefore, a larger number of trees can be planted on a unit of area.

Having analyzed the impact of the number of main branches left in the bush, we have noticed that the more main branches we have, the higher the productivity is; the highest values are observed in the variant with 5 main

branches. So, the maximum harvest (34.5 t/ha) during the whole period of studies was observed in Stanley variety with 5 main branches in 2015.

Table 3

**The crop yield of plum plantations depending on the form of the crown and variety, t/ha**

Variety	The form of the crown	The number of branches in the bush / pcs.	Year					The average value
			2011	2012	2013	2014	2015	
Kabardinskaia ranniaia	Mixed pyramid (control)	-	8.4	6.4	16.3	5.9	10.2	9.4
	Improved bush	3	16.70	12.60	18.80	7.4	15.70	14.20
	Improved bush	4	18.90	13.70	20.50	7.80	17.20	15.60
	Improved bush	5	20.3	14.9	21.9	8.70	17.9	16.7
Stanley	Mixed pyramid (control)	-	10.5	6.2	16.2	9.1	17.8	12.0
	Improved bush	3	18.90	13.90	20.90	14.20	29.60	19.50
	Improved bush	4	20.60	15.20	21.50	14.80	33.30	21.10
	Improved bush	5	20.70	15.80	21.40	15.60	34.50	21.60

However, it should be noted that it is rather difficult to get a bush with 5 main branches in Kabardinskaia ranniaia variety as it branches out very slowly. Therefore, in the first year after planting we have to pinch the strongest runner to promote additional branching. Furthermore, as the variant with 5 main branches grows, its growth is strongly retarded and ultimately one branch is suppressed. All this suggests that the optimal variants include improved bush with 4 main branches and the one with 5 main branches in the varieties that easily branch out with a later removal of one of the branches as the growth of the tree is strongly retarded.

We have also noticed a significant change in the yield of plum trees by years. The years with an abundant harvest (2011, 2013 and 2015) alternated with a weaker harvest in 2012 and 2014. One should limit the yield of plum trees by 15-20 t/ha for a more stable fruiting by year. Irrigation as well as application of additional fertilizers is needed to obtain higher yields.

## CONCLUSIONS

1. The analysis of the received data makes it possible to conclude that the improved bush form of the crown promotes intensive growth of the vegetative mass of plants at a young age and earlier crops of fruit. The annual increment of shoots stabilizes at the age of 7 and ranges from 52 to 64 m/tree in Kabardinskaia ranniaia and from 71 to 92 m/tree in Stanley variety.

2. Improved bush reduces the height of the crown by 35-40% and its width – by 32-42% as compared to the naturally improved forming and helps to increase the plantation density of trees.

3. Reduced crown parameters and increased density of plantations almost doubles the yield of plum trees as compared to the forming practiced in present.

All the ideas mentioned above allow us to recommend the production of improved bush with 4-5 main branches, depending on the variety.

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## THE APPLICATION OF FRACTAL ANALYSIS TO THE COARNĂ SORTOGROUP

### APLICAREA ANALIZEI FRACTALE LA SORTOGRUPUL COARNĂ

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**Abstract.** *Coarnă neagră and Coarnă albă vine varieties were used as maternal genitor in order to obtain new romanian vine creations. Out of these were included in the study Coarnă neagră selecționată, Azur, Milcov, Gelu, Ozana and Mara varieties within Coarnă neagră sortogroup. After applying the fractal analysis, it was determined the differences between the parent variety and its descendants. It resulted the fact that the fractal dimension closest to Coarnă neagră variety it is owned by Coarnă neagră selecționată variety (1.0389) and the farthest it is owned by Ozana variety (1.1254). For the Coarnă albă sortogroup, the value closest to the genitor was 1.0855, at Miorița variety, and the furthest was at Muscat Timpuriu de București variety of 1.1124.*

**Keywords:** fractal analysis, Coarnă albă, Coarnă neagră, vine

**Rezumat.** *Soiurile de viță de vie Coarnă neagră și Coarnă albă au fost folosite ca genitor matern pentru a obține creații noi românești de viță de vie. Dintre acestea au fost luate în studiu soiurile Coarnă neagră selecționată, Azur, Milcov, Gelu, Ozana și Mara în cadrul sortogrupului Coarnă neagră. În urma aplicării analizei fractale s-au determinat diferențele dintre soiul mamă și descendenții săi. A rezultat faptul că dimensiunea fractală cea mai apropiată de soiul Coarnă neagră o are Coarnă neagră selecționată (1,0389), iar cea mai îndepărtată o are soiul Ozana (1,1254). La sortogrupul Coarnă albă, valoarea cea mai apropiată de genitor a fost de 1,0855, la soiul Miorița, iar cea mai îndepărtată, la soiul Muscat Timpuriu de București, de 1,1124.*

**Cuvinte cheie:** analiză fractală, Coarnă albă, Coarnă neagră, viță de vie

## INTRODUCTION

Coarnă neagră is an old grapevine variety with an oriental origin, cultivated for a long time in Romania, becoming a domestic variety. Being very well adapted to the local conditions, there were created new varieties based on it: Coarnă neagră selecționată, Azur, Milcov, Gelu, Ozana and Mara.

Coarnă albă is also a very old grapevine variety, cultivated especially in Moldova. It is assumed that was brought from Minor Asia (Turkey). On the basis of this variety, there were created other new varieties: Miorița and Muscat Timpuriu de București (Țârdea and Bosoi, 2010).

Given these ten grapevine varieties, using fractal analysis it will be determined the fractal dimension, in order to establish the degree of similarity of these two groups of grapevines (Mancuso, 1999; Secelean, 2002; Oancea, 2006).

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**MATERIAL AND METHOD**

The biological material necessary for this method was represented by 10 adult leaves for each variety of the 2 groups of grapevine (tab. 1). Every leaf has been scanned and then processed so in the end to have the leaf colored in black on white background. After that, this image will be loaded into the Harfa program which will give us the fractal dimension of each leaf (Zmeskal *et al.*, 2001). With these values we can determine the degree of similarity for each group of grapevine.

Table 1

**The genetic material studied**

Variety name	Genitors	Homologation year	Authors and the unity where has been approved
Coarnă neagră	Oriental origin	-	-
Coarnă neagră selecționată	Free fertilization of the Coarnă neagră variety	1970	Constantinescu Gh., Negreanu Elena, Agronomy Institute N. Balcescu, București
Azur	Coarnă neagră x Cardinal	1984	Lepădatu Victoria, Condei Gh., SCVV Drăgășani
Milcov	Coarnă neagră x Muscat de Hamburg	1988	Bădițescu Margareta, Varga N., Zaharia V., Coman Gh., SCVV Odobești
Gelu	Free fertilization of the Coarnă neagră variety whose hybrid seeds were irradiated with X rays	1997	Calistru Gh., Damian Doina, SCDVV Iași
Ozana	Free fertilization of the Coarnă neagră variety	1982	Dănulescu D. et al., SCVV Iași
Mara	SV 12303 x Ozana	2011	Damian Doina, Calistru Gh., Nechita Ancuța, Savin C., SCDVV Iași
Coarnă alba	Oriental origin	-	-
Miorița	Free fertilization of Coarnă alba variety	1980	Popescu Gh., Bădițescu Margareta, Varga N., Zaharia Victoria, SCVV Odobești
Muscat Timpuriu de București	Coarnă albă x Regina viilor	1970	Gh. Constantinescu, Elena Negreanu, Agronomy Institute N. Bălcescu, București

**RESULTS AND DISCUSSIONS**

The results obtained by Harfa program for Coarnă neagră group, are presented graphic in figure 1 and listed in table 2 and the results obtained for Coarnă albă group are listed in table 3 and represented graphic in figure 3.

Table 2

## Fractal dimensions for Coarnă neagră group

Grapevine variety Leaf number	Azur	Coarnă neagră	Coarnă neagră selecționată
1	1.0697	1.0581	1.0646
2	1.0899	1.0643	1.0274
3	1.0979	1.0160	1.0093
4	1.1100	1.0135	1.0480
5	1.0983	1.0245	1.0225
6	1.0845	1.0346	1.0467
7	1.0723	1.0198	1.0458
8	1.0786	0.9951	1.0258
9	1.0987	1.0515	1.0231
10	1.0721	1.0551	1.0755
<b>Average</b>	<b>1.0872</b>	<b>1.0333</b>	<b>1.0389</b>
			

Continue of table 2

## Fractal dimensions for Coarnă neagră group

Variety Leaf number	Gelu	Mara	Milcov	Ozana
1	1.0428	1.0229	1.0011	1.1343
2	1.0518	1.0325	0.9995	1.1310
3	1.0537	1.0095	1.0177	1.1112
4	1.0571	0.9951	1.0200	1.0992
5	1.0383	0.9883	1.0570	1.1526
6	1.0118	<b>0.9788</b>	1.0309	1.1220
7	1.0442	1.0048	1.0224	<b>1.1570</b>
8	0.9931	1.0183	1.0414	1.0868
9	1.0062	1.0048	1.0576	1.1356
10	1.0048	1.0122	1.0377	1.1239
<b>Average</b>	<b>1.0304</b>	<b>1.0067</b>	<b>1.0285</b>	<b>1.1254</b>
				

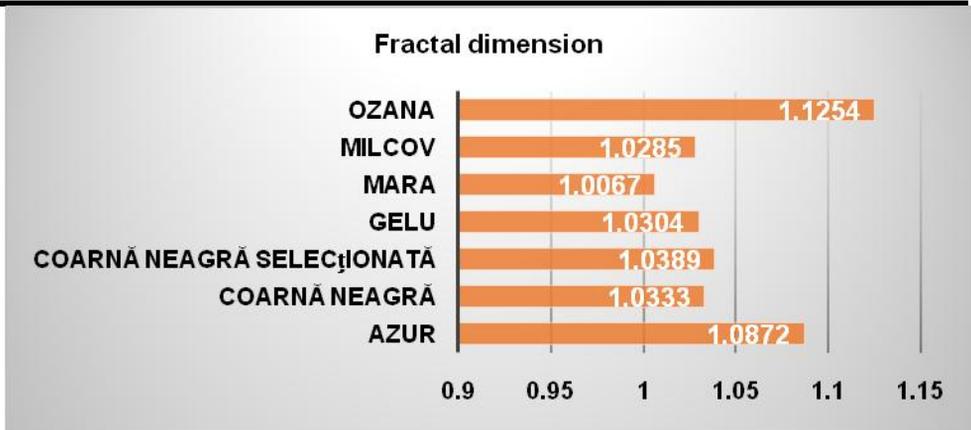


Fig.1 Fractal dimensions for Coarnă neagră group (averages)

From the analysis of data obtained is observed that the mean values of fractal dimension of the leaf vine varieties studied in Coarnă neagră group, varies between 0.9788 at Mara variety and 1.1570 at Ozana variety. Intermediate values of the fractal dimension had all the other five varieties, Azur (1.0697), Coarnă neagră (1.0643), Coarnă neagră selecționată (1.0646), Gelu (1.0571) and Milcov (1.0576). By comparing Coarnă neagră with his descendants, it can be seen that the varieties Coarnă neagră selecționată, Gelu and Milcov had values close to it, while Mara and Ozana have values significantly different from Coarnă neagră.

In figure 2 we can see the minimum fractal dimension obtained at Coarnă neagră group.

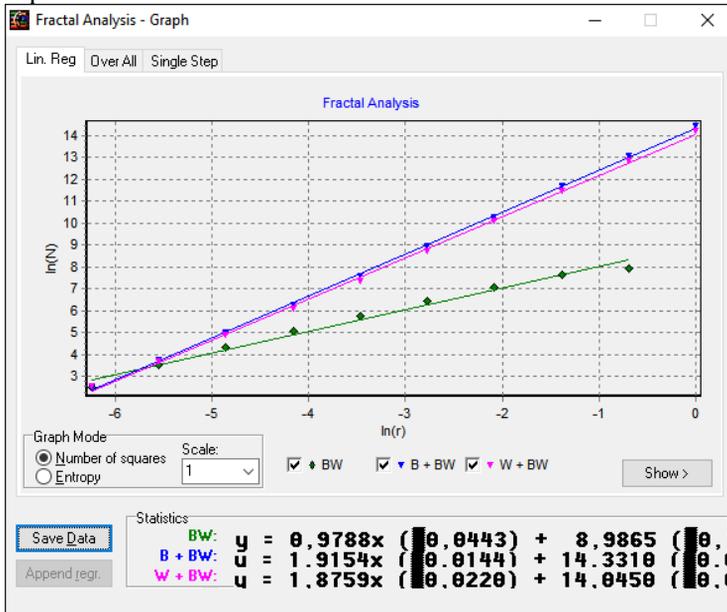


Fig. 2 Minimum fractal dimension for Coarnă neagră group

Fractal dimensions for Coarnă albă group

Grapevine variety Leaf number	Muscat Timpuriu de București	Miorița	Coarnă albă
1	1.1088	1.0537	1.0408
2	1.1168	1.0732	1.0584
3	1.0783	1.1295	1.0526
4	1.1073	1.1522	1.0560
5	1.1195	1.0691	1.0652
6	1.1029	1.0756	1.0436
7	1.0879	1.1116	1.0970
8	<b>1.1726</b>	1.0729	1.0653
9	1.0858	1.0497	<b>1.0292</b>
10	1.1442	1.0671	1.0322
<b>Average</b>	<b>1.1124</b>	<b>1.0855</b>	<b>1.0540</b>
			

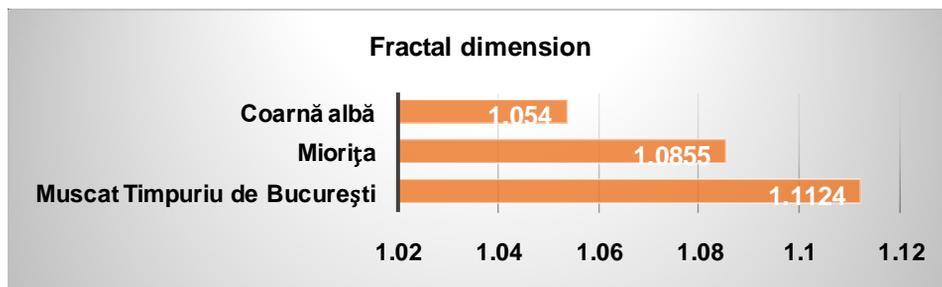


Fig. 3 Fractal dimensions for Coarnă albă group (averages)

The mean values of fractal dimension of the leaf vine varieties studied in Coarnă albă group, varies between 1.0292 Coarnă albă variety and 1.1726 at Muscat Timpuriu de București variety.

Intermediate values of the fractal dimension had all three varieties, Muscat Timpuriu de București (1.1029), Miorița (1.0497) and Coarnă albă (1.0970).

By comparing Coarnă albă with his descendants, it can be seen that the Miorița variety had a value very close to it, while Muscat Timpuriu de București variety it had an average value with a difference of over 0.5 against Coarnă albă variety.

In figure 4 we can see the minimum fractal dimension obtained at Coarnă albă group.

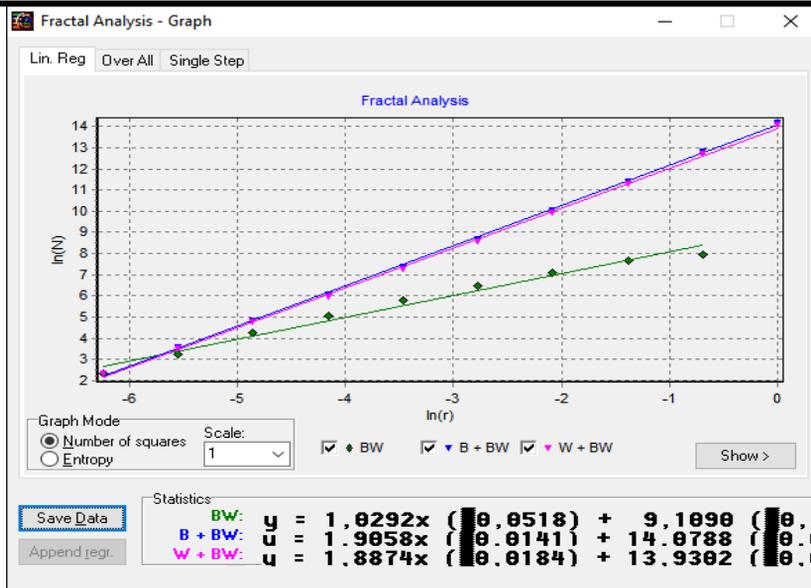


Fig. 4 Minimum fractal dimension for Coarnă albă group

## CONCLUSIONS

Fractal analysis in Coarnă neagră group revealed the following:

- average values of fractal dimension of leaves from Coarnă neagră vine varieties varies between 0.9788 at Mara variety and 1.1570 at Ozana variety;
- values close to the Coarnă neagră variety had Coarnă neagră selecționată, Gelu and Milcov, while Ozana and Mara had very different values.

Fractal analysis in Coarnă albă group revealed the following:

- average values of fractal dimension of leaves from Coarnă albă vine varieties varies between 1.0292 at Coarnă albă variety and 1.1726 at Muscat Timpuriu de București variety;
- values close to the Coarnă albă variety had Miorița variety, with 0.0315 more then, while Muscat Timpuriu de București had an average value with a difference of over 0.5 against Coarnă albă variety.

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**EFFECT OF TRACE ELEMENTS AND PGPB  
AZOTOBACTER CHROOCOCCUM AND PSEUDOMONAS  
FLUORESCENS APPLICATION ON GRAPE RESISTANCE  
TO THE WINTERING**

**EFFECTUL APLICĂRII MICROELEMENTELOR ȘI PGPB  
AZOTOBACTER CHROOCOCCUM ȘI PSEUDOMONAS FLUORESCENS  
ASUPRA REZISTENȚĂ VIȚEI DE VIE LA IERNARE**

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**Abstract.** *Vine resistance to the wintering is one of the main conditions of high productivity and longevity of vineyards. Realization of the genetically based potential of resistance may be enhanced by improving plant nutrient regime. Obtained results have shown that foliar fertilization of plants in the growing season with micronutrients adding metabolites and suspensions of bacteria Azotobacter chroococcum and Pseudomonas fluorescens is causing significant changes in metabolic processes during the growing season and period of dormancy, which contribute to increase of vine resistance to wintering.*

**Key words:** vine resistance, micro fertilizer Microcom-VA, bacteria, photosynthetic pigments, carbohydrates, bud viability.

**Rezumat.** *Rezistența viței de vie la iernare este una din condițiile principale a productivității și longevității podgoriilor. Realizarea potențialului genetic de rezistență poate fi sporită prin îmbunătățirea regimului nutritiv a plantelor. Rezultatele cercetărilor obținute au demonstrat, că fertilizarea foliară a plantelor în perioadă de vegetație cu microelemente cu adăugarea metaboliților și suspensiilor a bacteriilor Azotobacter chroococcum și Pseudomonas fluorescens induce schimbări esențiale în procesele metabolice pe parcursul perioadei de vegetație și în timpul iernării, ce contribuie la sporirea rezistenței a viței de vie la iernare.*

**Cuvinte cheie:** viță de vie, rezistență, microîngrășăminte Microcom-VA, bacterii, pigmenți fotosintetici, carbohidrați, viabilitatea mugurilor.

## INTRODUCTION

Resistance is a feature of plants wintering, influenced by a number of factors during the winter, first by frost and so-called winter drought (Cernomoreț *et al.*, 2000; Кондо, 1970; Погосян, 1975; Туманов, 1979). According to multiple data from the literature and field practices vine resistance to wintering depends largely on the plant mineral nutrition (Bratco, 1999; Veliksar and Toma, 2003; Waraich *et al.*,

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2012). Studies conducted over 10 years, showed that foliar fertilization of grape with a new set of trace elements Microcom-VA, created special for grape, increases the shoot growth and maturation, induces significant modifications in the processes of synthesis, accumulation, translocation, and storage of protective substances (sugars, free amino acids, phosphorus compounds) in the organs and tissues of the plants exposed to the action of low negative temperatures during winter. It contributes to the formation and a fuller manifestation of genetically based potential of frost and winter resistance, increase of plant productivity and longevity (Veliksar *et al.*, 2013).

In recent years appeared quite a lot publications about the influence of biological products, obtained on the basis of bacteria PGPB (plant growth promoting bacteria) on the metabolism of cultivated plants (Bhardwaj *et al.*, 2014; Шлабаев, 2012). Analysis of data on mechanisms of PGPB action on plants (Bordiec *et al.*, 2011; Bhardwaj *et al.*, 2014) confirms the necessity of deepen research in this area aimed at revealing of certain physiological aspects and genotypic peculiarities for achieving more complete realization of the plant adaptive potential and prevention of the environment contamination.

Our investigations showed opportunity to improve the nutritive regime of the vine by the simultaneous application of the halved dose of Microcom-VA and metabolites of bacteria strains PGPB. Application of suspension of *Pseudomonas fluorescens* and *Azotobacter chroococcum* together with micronutrients have increased the availability of nutrients to plants, the metabolic activity of some processes, improved the quality of planting material and decreased the quantity of applied nutrients (Veliksar *et al.*, 2014). The last gives possibility to reduce the negative impact of fertilizers excess on the environment and to increase viability of vineyards.

In this paper the results of the investigation of the trace elements and metabolites of the bacteria *Pseudomonas fluorescens* and *Azotobacter chroococcum* on the vine resistance to wintering are presented.

## MATERIAL AND METHOD

The researches have been performed in field conditions on the experimental plot of the Institute of Genetics, Physiology and Plant Protection of the ASM, cv. Codrinschi. The foliar fertilization of plants by the micro fertilizer (Microcom-VA-0,15%, that is 1/2 of the recommended doze) and metabolites or suspension of *Azotobacter chroococcum* and *Pseudomonas fluorescens* (titre -  $10^7$  CFU / ml) was conducted three terms: before flowering and twice - at the stage of intensive growth with an interval of 12 to 14 days. Water treated plants were used as control. The leaves for analyses were sampled during the vegetation period six days after the foliar treatment, thoroughly rinsed with water, allowed drying and used for analysis. The shoots and buds were sampled at the end of vegetation and on the end of deep dormancy periods. The following analytical methods were used: photosynthetic pigments content was measured using Chlorophyll Content Meter CCM-200 plus, the carbohydrate content - according to Bertan, proline content - according to Bates (1973). Evaluation of the winter resistance was carried out under field conditions in

accordance with the method developed by Cernomoreț (1985, 2000). The results were analyzed statistically according to Statistica-7.

## RESULTS AND DISCUSSIONS

Resistance of vine to wintering depends largely on the intensity of some metabolic processes of plants in vegetation period, growing and maturation of shoots. The high content of photosynthetic pigments in leaves and their active absorption capacity of PAR influences positive on the intensity of sugars accumulation - the primary products of photosynthesis, on the growth, development and productivity of plants in summer, as well as during the process of growth and hardening of vine, its preparation for winter (Жакотэ, 1974; Черноморец *et al.*, 1984).

Foliar fertilization of plants has contributed to increasing of summary content of photosynthetic pigments in the leaves of the vine in the second half of the growing season. On July 21 content of chlorophyll in the leaves of variants with application of halved dose of Microcom-VA plus suspension of microorganisms or metabolites was 13.27 and 14.22 mg/g fresh weight (tab. 1) - 117.7 and 119.7 % compared to control.

Table 1

**Total content of photosynthetic pigments in grape leaves under the influence of foliar fertilization by trace elements and bacteria, v. Codrinschi, mg/g f.w.**

Variants	09.06.2013	11.07.2013	21.07.2013
Control	8.59±0.19	13.45 ±0.16	11.88±0.48
Foliar fertilization by Microcom-VA, 0.5 doza	8.69±0.52	13.24 ±0.19	12.70±0.37
Foliar fertilization by metabolites of <i>Ps. fluorescens</i> + <i>Az.chroococcum</i>	9.02 ±0.26	12.62 ±0.33	12.51±0.54
Metabolites of <i>Ps. fluorescens</i> + <i>Az.chroococcum</i> + Microcom VA, 0.5	8.46 ±0.53	13.92 ±0.08	13.27±0.35
Suspensia of <i>Ps. fluorescens</i> + <i>Az.chroococcum</i> + Microcom VA, 0.5	9.17 ±0.29	14.47 ±0.52	14.22±0.043

It was revealed considerable increase in the degree of growth and maturation of shoots under the foliar fertilization. It has been confirmed the beneficial effect of micronutrient complex Microcom-VA on the growth of shoots - +4.92 % to the control (tab. 2). Effect of trace elements on the growth and maturation of the shoots increased in case of application of trace elements complex in combination with products of microorganisms, especially - in the form of suspension. The degree of the shoots maturation was increased from 3.09% to 9.11% compared to the control (tab. 2). Timely completion of the annual growth and maturation of shoots is one of the basic conditions for the formation of frost resistance of vine.

Table 2

**Effect of foliar fertilization on the growth and maturation of vine shoots,  
v. Codrinschii, 2013**

Variants	length of shoots, cm	length of matured shoots, cm	± to the control	grade of maturation %	± to the control
Control	134.13± 3.12	93.90±2.04	-	70.01	-
Foliar fertilization by Microcom-VA, 0.5 doza	165.40 ±2.74	123.93± 1.53	+30.03	74.93	+4.92
Metabolites of <i>Ps. fluorescens</i> + <i>Az. chroococcum</i>	158.93± 1.78	117.77± 1.67	+23.87	74.10	+3.09
Metabolites of <i>Ps. fluoresc.</i> + <i>Az. chrooc.</i> +Microcom VA, 0.5	169.07± 3.06	133.77± 2.79	+39.87	79.12	+9.11
Suspensia of <i>Ps. fluorescens</i> + <i>Az.chrooc.</i> + Microcom+VA, 0.5	171.37± 2.65	132.37 ±2.13	+39.47	77.24	+7.23

One of the essential moments for grape preparation at the wintering period is October. Suspending flow of carbohydrates and their translocation into the perennial shoots and roots in late October is stipulated by plants entrance to the dormancy period, occlusion of sieve tubes of callose. In October 2014 after the first autumn night frost ( $-5^{\circ}\text{C}$ ) content of carbohydrates in the leaves and shoots of vine was determined. The increased content of sucrose in the leaves and annual shoots was manifested in variants with plant fertilization by metabolites or suspension of bacteria and Microcom-VA – 112.65 and 117.74%.

Next selection of shoots for the analysis was performed at the end of the deep dormancy - January 27<sup>th</sup>, at the  $-7^{\circ}\text{C}$ . The content of monosaccharide and disaccharide, which are known as a compatible osmolytes, increased compared to the control, especially after the simultaneous treatment of plants by suspension of bacteria and compound of trace elements Microcom+VA. Sum of sugars was in that variant 5.82 % (tab. 3) or 123.83% compared to the control.

Table 3

**Effect of foliar fertilization on the sugars content in shoots, 27 January 2014,  
t.  $-7^{\circ}\text{C}$ , v. Codrinschii, %**

Variants	monosaccharide	disaccharide	sum of sugars	% to control
Control	3.10±0.11	1.60±0.56	4.70	100
Foliar fertilization by Microcom-VA, 0.5 doza	3.62±0.19	2.09±0	5.71	121.49
Metabolites of <i>Ps. fluorescens</i> + <i>Az. chroococcum</i>	2.65±0.32	1.72±0.02	4.37	92.98
Metabolites of <i>Ps. fluoresc.</i> + <i>Az. chrooc.</i> +Microcom VA, 0.5	3.42±0.56	1.87±0.02	5.29	112.55
Suspensia of <i>Ps. fluorescens</i> + <i>Az.chrooc.</i> + Microcom+VA, 0.5	3.90±0.34	1.92±0	5.82	123.83

There has been a significant increase of starch and less pronounced – hemicelluloses in variants fertilized with micronutrients and bacteria metabolites. Starch cleavage does not occur at the moment. After the literature date in the period of dormancy starch cleavage and formation of glucose, fructose and sucrose in vine strings

is more intense after the temperature of  $-10^{\circ}\text{C}$ . Starch reserve stored in xylem not are hydrolyzed because of strong cell walls lignification and blocking of enzymes, that's why it do not participate in the protection of vine from frost. The content of hemicelluloses in plants treated with micronutrients and product of bacteria increased compared to control. It is considered as a form of carbohydrate reserves, hydrolysis of hemicelluloses occurs at lower temperatures.

The obtained data allow us to assume that the treated plants accumulated more reserves carbohydrate and are able to resist lower negative temperatures. This assumption was confirmed at the influence of lower temperature on 31 January 2014 ( $-24^{\circ}\text{C}$ ) and subsequent determination of the number of dead buds in spring.

Simultaneously with the carbohydrates it was determined content of compatible osmolytes – proline - into the shoots and buds. It was established that the foliar fertilization of plants during the vegetation period has contributed to increasing of the content of proline into the shoots and buds during the dormancy, particularly after applying metabolites or suspension of bacteria with Microcom.

Determination of one of the main indicators of vine resistance to wintering - bud viability - on spring, after the period of dormancy, showed, that the number of viable buds grew evident under the foliar fertilization, from 3.48% to 9.92% compared to control (tab. 4). Least of all died buds were on the plants fertilized during vegetation period by suspension or metabolites of bacteria with complex of trace elements - 57.36 and 55.84% viable buds (control - 47.44%). Generally amount of died buds was higher than previous years due to very low temperature on the end of winter.

Table 4

**Effect of foliar fertilization and frost ( $-24^{\circ}\text{C}$  on 31 January 2014) on the state of grape buds on the spring, v. Codrinschii, %**

Variants	buds died, %		buds alive, %	
	M± m	± to control	M± m	± to control
Control	52.56± 1.23		47.44±1.47	
Foliar fertilization by Microcom-VA, 0,5 doza	47.31±1.13	-5.25	52.69±1.25	+5.25
Metabolites of <i>Ps. fluorescens</i> + <i>Az. chroococcum</i>	49.08±1.16	-3.48	50.92±1.34	+3.48
Metabolites of <i>Ps. fluoresc.</i> + <i>Az. chrooc.</i> +Microcom VA, 0,5	42.64± 0.92	-9.92	57.36± 1.13	+9.92
Suspensia of <i>Ps. fluorescens</i> + <i>Az.chrooc.</i> + Microcom+VA, 0,5	44.16±0.78	-8.40	55.84±0.81	+8.40

Highlighting the positive effect of trace elements and products of bacteria *Az. chroococcum* and *Ps. fluorescens* on the accumulation of carbohydrates, compatible osmolytes content (sucrose and proline) in the organs of plants under low temperature indicates about the stimulator role of the mentioned products in achieving greater degrees of grape potential to resistance realization. We suppose that plants fertilized with trace elements and suspension and metabolites of bacteria facilitate the activities of one of the specialized mechanisms of plant resistance to low temperatures - compatible osmolytes accumulation.

## CONCLUSION

It was revealed that the complex of trace elements Microcom-VA in halved dose and the products of bacteria *Azotobacter chroococcum* and *Pseudomonas fluorescens* (suspension and metabolites), which contain large amounts of biologically active substances, applied simultaneously for foliar fertilization of the vine thrice on the vegetation, induce changes in the metabolic processes during vegetation and dormancy periods, increase the growth and maturation of shoots, contributing to increasing of vine resistance to the wintering.

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## MONITORING THE STATE OF THE INSURANCE OF SOIL WITH THE MAIN TOTAL FORMS AND MOBILE NUTRIENTS UNDER THE INFLUENCE OF FERTILIZATION, IN THE CONDITIONS OF THE VINEYARD IAȘI

### MONITORIZAREA STĂRII DE ASIGURARE A SOLURILOR CU PRINCIPALELE FORME TOTALE ȘI MOBILE DE ELEMENTE NUTRITIVE SUB INFLUENȚA FERTILIZĂRII, ÎN CONDIȚIILE PODGORIEI IAȘI

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**Abstract.** *The problem of vineyards fertilization is especially complex because of the great amount of agents that interfere. Fertilization, in nowadays technical progress, must amplify and intensify the productive capacity of fertile vineyards in correspondance with the effort-effect-efficiency relation avoiding the risk of pollution of the crop or the environment. The influence of fertilization chemical on certain indicators of basic soil fertility (Nt, P-AL, K-AL), appears clearly in the conditions of experimentation and reflects the status of cultivation and fertility status of the soils with vocation wine. From this point of view, use of doses moderate fertilizers, administered especially in complex ternary (NPK), led in the study undertaken at the improving regime nutritional variety Aligote and respectively to a stabilization of its in the field of optimum on the entire range of vegetation analyzed.*

**Key words:** pollution, soil, fertilizer, protection, doses

**Rezumat.** *Problema fertilizării în viticultură este una complexă datorată numeroșilor factori care interferează. Fertilizarea, în condițiile actualului progres, trebuie să amplifice și să intensifice capacitatea de producție a plantațiilor de vita de vie, în concordanță cu relația efort-efect-eficiența, evitând riscul de poluare a recoltelor și mediului. Influența fertilizării chimice asupra unor indicatori de bază ai fertilității solului (Nt, P-AL, K-AL) apare în mod evident în condiții de experimentare și reflectă starea de culturalizare și de fertilitate a solurilor cu vocație viticolă. Din acest punct de vedere, utilizarea dozelor moderate de fertilizanți, administrate mai ales în complex ternar (NPK), a condus în studiul întreprins la îmbunătățirea regimului nutrițional al soiului Aligote și respectiv la o stabilizare a acestuia în zona domeniului optim, pe întreg intervalul de vegetație analizat.*

**Cuvinte cheie:** poluare, sol, fertilizant, protecție, doze

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## INTRODUCTION

The concept of “lasting agriculture “ is more and more promoted in agriculture practical, and the most important attribute of this system is the one which referres , to the soil and the environment quality in general. Mentaining and developing the quality of the soil, is an essential condition for the promotion of this kind of system, are workable through the increase of the energy and technology for upkeeping the soil. In this case, an essential role is maintaining and developing fertility of the soil by reducing the consumption of chemical fertilizer, consumer of energy.

An important source of pollution of the soil is consisted in irrational application of the fertilizer, wich big variety makes difficult in putting some templates of utilisation (Avarvarei, 1997). The effect of using the nitrate and phosphate in excess is known which has negative actions on microbiological flora from the soil, and leading to the accumulation into the vegetation of these elements, far beyond any tolerated limits (Borlan, 1994).

This study is made on these coordinations, the obtained results, being the foundation of data for practising in this area and a source of information for starting producers in the neighborhood (Volf, 2004).

## MATERIAL AND METHOD

Knowing the agro-eco-pedological characteristics of the wine-growing center of Copou- Sorogari (Cotea, 2000) allowed to organize the experience like the influence of the mineral fertilization on the evolution of sourshing elements from the soil. Primary and secondary ecological factors of biotope in interaction are distinguishable, making the agrofitotechnic base of viticulture.

This study was developed during 3 years in the viticol center Copou-Sorogari, which was owned by the old S.C Podgoria S.A Copou lasi from administratively point of view. Now this lands are privately owned.

Acording to the geographic aspect, this lands are part of Jijia-Bahlui depression, beeing located in the south part of this depression. Under the geomorphological aspect, the studied territory is situated at the contact of two large subunits of the Moldavian Plateau: The moldavian Plain in north, and the Central Moldavian Plateau in south.The stratigraphy of the region is represented by the natural layers damaged by the rocks, on which are layers of sedimentary rocks of various ages; on the surface being deposited conglomerates of clay and marl interposed with fine sand.

The pedological study indicates the presence of chernozem (poorly levigated and decarbonated), clayey-argillaceous, developed on loessial deposits and not irrigated.

The chemical structure of these soils presents: pH(H<sub>2</sub>O) 6.9 – 7.5, neutral reaction towards to poorly alkaline containing a medium concentration of loamy (2.4-2.5 % H) , a low to medium of nitrate concentration (1.9-3.9 mg NO<sub>3</sub><sup>-</sup>/100 g sol), a low concentration in mobile phosphorus (20.8-52.8 ppm P-AL); good gatering estate in mobile potassium (260-301 ppm K-AL).The content of 17.0 me % Ca<sup>2+</sup>, indicates an increased fertility. Among the changeable cations, the content of 2.06-2.88 me % Mg<sup>2+</sup> situates the soil into a middle class fertility.The Boron is situated at the inferior limit of a

middle assurance (0.4-0.5 ppm B), the Copper at >25 ppm values indicates an excess soil catering, the movable zinc is at high quota: 1.8-2.0 ppm Zn.

There was studied the Aligote variety, widespread, spliced on the Berlandieri × Riparia Kober 5 BB as mother as, planting distances being of 2.20x1.10. There were applied monotonous cutting tasks of 17 eyes/m<sup>2</sup> (41 eyes/log) to the logs.

The undertaken study was concretized through the establishment of an experience with two factors A and B, the arranging method in the terrain being that of the subdivided holding, repeated 4 times. In the experimental machine, the graduation of the variable factors was:

The A Factor – the compost administered dosage (kg s.a./ha)

a<sub>0</sub> – unfertilized (Witness); a<sub>1</sub> – 0.5 dosage; a<sub>2</sub> – 1.0 dosage; a<sub>3</sub> – 1.5 dosage

The B Factor – the combination of fertilizing elements

b<sub>0</sub> – NPK – witness; b<sub>1</sub> – N; b<sub>2</sub> – P; b<sub>3</sub> – K; b<sub>4</sub> – NP; b<sub>5</sub> – NK; b<sub>6</sub> – PK

In the present study there were suggested three fertilization levels referring to the dosage and 7 levels of utilization of the combination of fertilizing elements.

The following levels were established, taking into account the expended crop and the productivity soil estate:

- the 0.5 dosage level with : 50 kg/ha s.a N; 25 kg/ha s.a P<sub>2</sub>O<sub>5</sub>; 90 kg/ha s.a K<sub>2</sub>O
- the 1 dosage level with ; 100 kg/ha s.a N; 50 kg/ha s.a P<sub>2</sub>O<sub>5</sub>; 180 kg/ha s.a K<sub>2</sub>O
- the 1.5 dosage level with : 150 kg/ha s.a N; 75 kg/ha s.a P<sub>2</sub>O<sub>5</sub>; 270 kg/ha s.a K<sub>2</sub>O

This is how 7 fertilization levels resulted: exclusive fertilization with nitrogen (N); exclusive fertilization with phosphorus (P); exclusive fertilization with potassium (K); binary fertilization with nitrogen + phosphorus (NP); binary fertilization with nitrogen + potassium (NK); binary fertilization with phosphorus + potassium (PK); ternary fertilization with nitrogen + phosphorus + potassium (NPK).

As a range of chemical compost were used simple chemical compost for facilitating the dosage and reports calculus and also for the general economicity of other advantages. In this context was used the ammonium nitrogen, NH<sub>4</sub>NO<sub>3</sub> as a nitrogen nutrition source, with 34.5% s.a., as phosphorus source was proposed the utilization of *concentrated superphosphate* Ca(H<sub>2</sub>PO<sub>4</sub>)<sub>2</sub> · H<sub>2</sub>O, with 50% s.a. The potassium was administered as potassium salt 50%. The organical fertilization was assured by using the stable dirt 40t/ha.

Annually, during 3 vegetation moments : unbudging, booming and grapes maturing, there are taken soil samples on 2 depths 0-20 and 20-40 cm, from each variant, in order to determine the primary bidding major elements dynamics.

The analysis methods and techniques for the soil samples were the following:

- the P-AL și K-AL content, the Egner-Riehm-Domingo method, colorimetric dosage;

- the total nitrogen content - the Kjeldahl method.

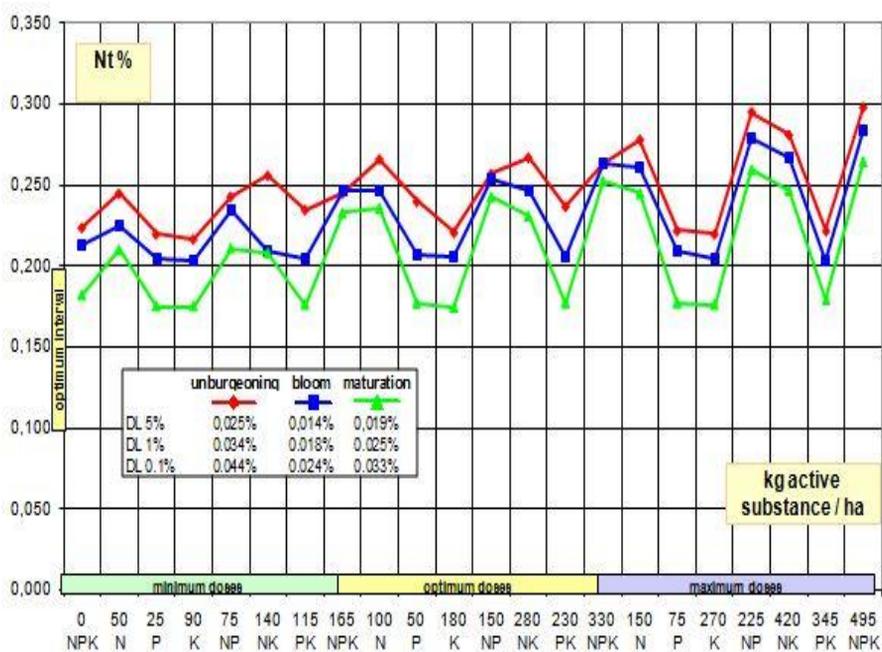
**RESULTS AND DISCUSSIONS**

The results obtained by compost administrating in dosages and different combinations, dignify the improval of nitrogen regime within soil (fig.1).

At unbuding the minimal dosages, no matter, the combination of fertilizing element, there are no statistically assured results after the first fertilization year, ever if the the nitrogen containt increases progressively with them .Only during the second year of research there is assured a soil content of 0.267% N<sub>t</sub> during unmuding in the 165 kg/ha s.a variant, bigger than the average of the years, statistically unasured.

During booming, the 165 kg/ha s.a. NPK variant concurs at a normal soil suply with this element, situating at 0.246% N<sub>t</sub> annual average, with a difference from the unfertilized witness of + 0.033%.The 330kg/ha s.a. NPK (1 dosage) and 495 kg/ha s.a. NPK (1.5 dosages) leads to a soil catering beyond the normal limits – luxury consumption .

During grape maturation, annual average, minimum dosages, at N, NK, NP combinations they dignify as statistically assured; the minimum dosage and the ternary combination, 165 kg/ha s.a. NPK, strenghtens the motivation of application in complex fertilizad elements, the total nitrogen being situated at 0.233% N<sub>t</sub> .



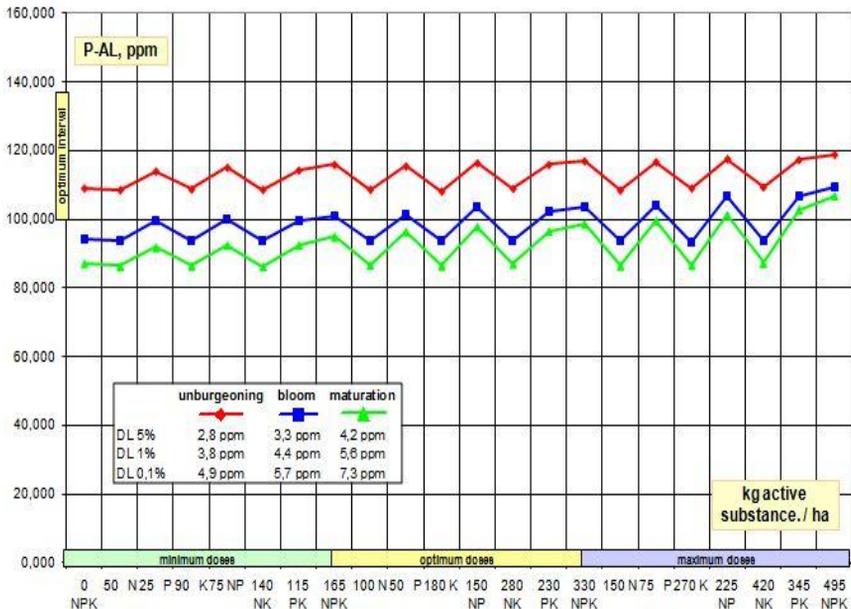
**Fig. 1** The influence of fertilizers combination x measure interaction towards the evolution of total nitrogen (Nt %) in soil at Aligote in 3 phenological phases: unburgeonig, bloom, maturation of grapes

Analyzing the phosphorus ensurance estate (fig. 2), accesible fraction, after aplying dosages fertilization and different compost combinations, during the whole vegetation period, we ascertain that they were improved.

In the unbuding phenophase, there is a obvious tendency of improving of the phosphatic nutritional diet, from one year to another, at every single level of fertilization (0.5 dosage, 1 dosage and 1.5 dosage and especially at the P, NP, PK and NPK combinations) .The minimum dosage variant 165 kgs.a./ha NPK ensures the soil with a 116.0 ppm content P-AL on annual average (statistically assured – very semnificative) comparable to 117.1 ppm P-AL for the 330 variant kg/fa s.a NPK and 119 ppm P-AL for the 495 kg/ha s.a NPK.

In the booming phenophase, although the values decrease from the previous phenophase, because of the vine consumption, they have an optimum phosphatic activity of the soil.The minimum dosages ensure the soil on the annual average 100.9 ppm P-AL (165 kg/ha s.a NPK variant – very significant).

At grapes maturation, the obtained results on the annual average, prove the efficacy of the progresseive application of dosages and binary but especially ternary combinations, of nutrient elements .The 165 kg/ha s.a. NPK variant ensures the soil with a content of 96.5 ppm P-AL, very significant ensurance, considered to be a medium phosphorus gatering of the soil (> 71 ppm P-AL).

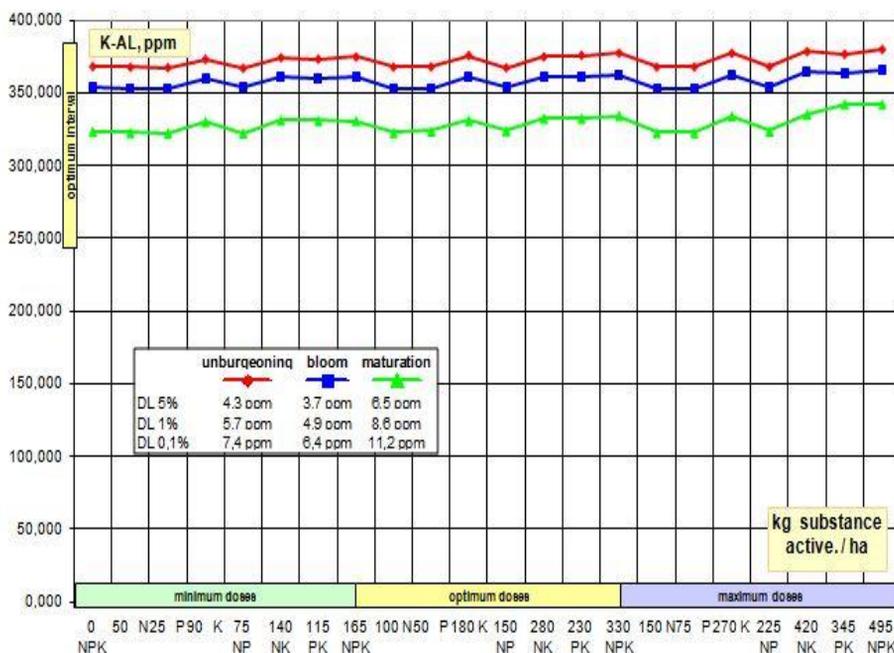


**Fig. 2** The influence of fertilizers combination x measure interaction towards phosphorus mobile forms' evolution (P-AL, ppm) in soil at Aligote in 3 phenological phases: unburgeoning, bloom, maturation of grapes

In the phenophase of unbudging, there are being assured normal levels (optimal) of accessible K in soil by utilizing the minimal dosage. The 90 kg s.a/ha K, 140 kg s.a/ha NK, 115 kg s.a/ha PK și 165 kg s.a/ha NPK variants ensures the soil in 373.1 ppm, 374.2 ppm, 373.3 ppm and 374.7 ppm K-AL contents. The last one is being ensured distinctively significant.

In the blossom moment the result for those 3 years of study regarding the fertilization impact for the evolution of accessible K, show that the administration of the fertilizers in increased dosages and in singular combinations, binary and ternary ensures an improvement of the K nutritional regime. The 140 kg s.a/ha NK and 165 kg s.a/ha NPK variant (minimal dosages) ensures to the soil 360.8 ppm K-AL, respectively 361.4 ppm K-AL very good in significance.

The results are showing that in maturation phenophase of the grapes, in soil, of all the years, it is satisfied the optimal level of providing the soil with accessible K, statistically assured significantly (ecological dosages), distinct significant (optimal dosages) and very significant (maximal dosages) (fig. 3)



**Fig. 3** The influence of fertilizers combination x measure interaction towards potassium mobile forms' evolution (K-AL, ppm) in soil at Aligote in 3 phenological phases: unbudging, bloom, maturation of grapes

## CONCLUSIONS

- ❖ Chemical fertilization of the soil in viticulture, broached in minimal usage of fertilizers, avoids the environmental pollution danger and assures the agronomical, economical and entropical efficiency.
  
- ❖ The statistical parameters of the total N content from the superior horizon of the soils with viticulture utilization, chemical fertilized, show a quite restrained interval of content, variable in rapport with the utilized compost and the nature of the fertilized element.
- Assigination of minimal dosages unilateral distributed (N), in binary complex (NP, NK) and in tertiary complex (NPK) has led, in all 3 phenophases, at accumulating N sleightly overoptimal, ensured statistically at different levels and comparable.
- The 165 NPK variant correlates with a total N content within 0.245% at unbud and 0.233% at maturation. The two times or three times increasing in compost dosages had favored N accumulations in soil much over the optimal limits, wich may induce the manifestation of some nutritional imbalance.
- The 330 NPK and 495 NPK variants achieve accumulations in the soil within 0.263% (unburgeonig) and 0.253% (maturation) and 0.298% and 0.265%, for the same interval of vegetation.
  
- ❖ The variant analysis showed a cumulative effect in the interaction of the 2 singular factors considered in the study (dosage x combination of fertilized elements) on the evolution of the phosphate fertility of soils.
- The annual chemical fertilization with N, P and K in minimal quantities (the 165 NPK variant) had determine an improving of the mobile forms of P in soil, assuring a medium to optimal supply: 116.0 ppm P-AL (unburgeonig)), 100.8 ppm P-AL (bloom) respectiv 95.1 ppm P-AL (maturation) very significant.
- The doubling and tripling of the fertilizers dosages in NPK relationship, does not enrich the soil with mobile phosphates spectacularly, the differences between the utilization of the minimal dosages are insignificant and relatively pumpy.
  
- ❖ The interaction between the dosage factors x fertilizing element combination was good for the evolution of the potassium nutritional diet of the soil, put across different ensurance optimum estates especially at the fertilization in minimum dosages.
- At the level of utilization of minimum fertilizers dosages detanches the 165 NPK variant which realizes in soil values statistically ensured for “accessible K”: 374.7ppm K-AL at unbuding, 361.4 ppm K-AL at booming and 330.9 ppm K-AL at maturation, comparable to the values obtained by using the optimum dosages, the 330 NPK variant.

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## DETERMINATION AND ANALYSIS OF THE TOTAL FORMS OF NUTRIENTS FROM THE PLANT MATERIAL OF THE VARIETY ALIGOTE, INTO AN EXPERIENCE STATIONARY WITH FERTILIZERS, IN THE CONDITIONS OF THE VINEYARD IASI

### DOZAREA ȘI ANALIZA FORMELOR TOTALE DE ELEMENTE NUTRITIVE DIN MATERIALUL VEGETAL LA SOIUL ALIGOTE, ÎNTR-O EXPERIENȚĂ STAȚIONARĂ CU ÎNGRĂȘĂMINTE, ÎN CONDIȚIILE PODGORIEI IAȘI

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**Abstract:** Foliar diagnosis of nutrition by chemical analyses is an extended method, by which are analyzed some vegetative parts (especially leave, but also nervures, petiole, young buds, or branches of one year old), determinate the feeding stage of the analyzed plants with nutrition elements, to see the existence of excess or lack situations, to predict the lacks of nutrition during vegetation period, to establish the fertilization recommendations, to evaluate the exports of nutritive elements with yield and to keep the soils' fertility and also for yields prediction. The paper presents the results obtained by the foliar diagnosis, at the leaves of vines of the variety Aligote, of the content of macroelements, in the three moments of vegetation, using a scheme of fertilization specify. Use of doses minimum of fertilizers manages to attract a proper content of NPK in the plant material, which confirm the need for optimization of doses of fertilizers and also the choice assortment of these fertilizers. This practice, can converge to a steady nutritional optimal, with the avoidance of any excesses, evidenced by the emergence of diseases of nutrition, default by lowering the yields.

**Key words:** diagnosis foliar, nutrition, macroelements

**Rezumat:** Diagnoza foliară a nutriției prin analize chimice este o metodă extinsă, prin care sunt analizate unele organe vegetative (în special frunze dar și nervuri, pețiol, lăstari tineri sau mai mari de un an ca vârstă), determinându-se alimentarea cu elemente nutritive în anumite fenofaze a plantelor analizate, pentru a investiga existența exceselor sau carențelor, pentru previzionarea dereglărilor de nutriție în timpul perioadei de vegetație, pentru stabilirea recomandărilor de fertilizare, pentru evaluarea exporturilor nutritive o dată cu recoltele și pentru menținerea stării de fertilitate a solurilor și deasemenea pentru prognozarea recoltelor. Lucrarea prezintă rezultatele obținute prin diagnostic foliar, la frunzele de viță de vie a soiului Aligote, a conținutului de macroelemente, în trei momente de vegetație, folosindu-se o schemă de fertilizare specifică. Utilizarea dozelor minime de fertilizanți, reușește să atragă un conținut adecvat NPK în materialul vegetal, ceea ce confirmă necesitatea optimizării dozelor de îngrășămintă dar și alegerii sortimentului de fertilizanți.

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*Această practică, poate converge către un echilibru nutrițional optim, evitându-se eventualele excese, materializate prin apariția bolilor de nutriție, implicit prin scăderea producțiilor.*

**Cuvinte cheie:** diagnoză foliară, nutriție, macroelemente

## INTRODUCTION

Diagnosis by chemical analyze of nutritive elements (classic diagnosis) reflects the accumulation and supplying of the plant during the whole vegetation period, from its starting till harvest moment of the sample and less the momentum stage of supplying (Davidescu, 1980) This diagnosis method is the most used one to confirm or infirm the nutrition malfunctions and to correct fertilization system. Also, this type of diagnosis is very good for researches of nutritive elements accumulation dynamics in plants during vegetation period, case in which are analyzed those elements in each moment. (Volf, 2004) From this point of view, plants' chemical analyses serves also to monitor different types of fertilization and also to establish fertilizations recommendations, in according with soil analyses on samples from the same placement, with climatic conditions, with phyto-sanitary stage of the crop and with information regarding the used technology.

So, the prognosis of possible modifications of the nutrition stage in negative way (lacks, excesses, un-balance) by foliar diagnosis, offers the possibility of reviewing and adapting of integrated fertilization system in time and on pheno-phases, for a good supplying of nutritive elements in soil-plant system. At the level of the obtained results and on the line of capitalization of the results of this experience could be made some conclusions for a good practice of the farmers.

## MATERIAL AND METHOD

Experience took place on a three years period, in the Copou-Șorogari vineyard, Iași county, Romania and the experimental polygon belonged to the private owners. Geographically, the studied area is placed in the Jijia-Bahlui depression, taking in study the *Aligote* kind, grafting on port-graft *Berlandieri* × *Riparia Kober 5 BB*, planting distance being  $2.20 \times 1.10$ . Vine stocks presents uniform cut of 17 eyes/m<sup>2</sup> (41 eyes/vine stock). *Aligote* kind is included in the cultivars of 61 vineyards from 10 counties and is on the "recommended kind" position in 29 areas (including in the vineyards of Iași County). This sot of grape is for dry wines, which function of the area and years' climatic conditions, could be placed in the ordinary wines category (usual consumption) or in the category of superior quality. In some areas (Iași for example), *Aligote* wines could reach the quality level of the ones with origin denomination (D.O.C.).

The experimental scheme focused the establish of a bi-factorial experience, the method of placement in field, being the one of sub-divided plots, in four repetitions, and the graduation of the variable factors being the following:

Factor A – fertilizers dose (kg a. s/ha)

Factor B – combination of fertilizers elements

a<sub>0</sub> – un-fertilized (control)b<sub>0</sub> – NPK – controla<sub>1</sub> – 0.5 doseb<sub>1</sub> – Na<sub>2</sub> – 1.0 doseb<sub>2</sub> – Pa<sub>3</sub> – 1.5 doseb<sub>3</sub> – Kb<sub>4</sub> – NPb<sub>5</sub> – NKb<sub>6</sub> – PK

In the present paper are proposed three levels of fertilization as regarding the dose and 7 levels of usage of the fertilizers elements' combination. Function of the predicted yield, production ways and soils' fertility stage was established:

- 50 kg/ha a. s N
- level 0.5 dose – 25 kg/ha a. s P<sub>2</sub>O<sub>5</sub>
- 90 kg/ha a. s K<sub>2</sub>O
- 100 kg/ha a. s N
- level 1 dose – 50 kg/ha a. s P<sub>2</sub>O<sub>5</sub>
- 180 kg/ha a. s K<sub>2</sub>O
- 150 kg/ha a. s N
- level 1.5 dose – 75 kg/ha a. s P<sub>2</sub>O<sub>5</sub>
- 270 kg/ha a. s K<sub>2</sub>O

Fertilizers elements were singular administrated or in combination so in this way resulted 7 levels of fertilization.

- fertilization exclusive with nitrogen (N)
- fertilization exclusive with phosphorous (P)
- fertilization exclusive with potassium (K)
- fertilization binary nitrogen + phosphorous (NP)
- fertilization binary nitrogen + potassium (NK)
- fertilization binary phosphorous + potassium (PK)
- fertilization ternary nitrogen + phosphorous + potassium (NPK).

Were used as chemical fertilizers, simple chemical fertilizers such as *ammonium azotize*, NH<sub>4</sub>NO<sub>3</sub>, with 34.5% a. s, as nutrition source of nitrogen. As phosphorous source we propose the usage of *concentrate super-phosphate* Ca(H<sub>2</sub>PO<sub>4</sub>)<sub>2</sub> H<sub>2</sub>O, with 50% a. s. Potassium was administrated as fertilizers under the form of *potassium salt* 50%. Organic fertilization was assured by administration of manure in a norm of 40 t/ha. For tracking the elements' mobility, their movement from soil to plant and the dynamics of the their accumulation during vegetation, for foliar diagnosis were taken leave samples at three moments – budding, flowering and maturation – from each experimental variants, from the control vine stocks. The working methods were in according with the ones elaborated by I.C.P.A. Romania for agrochemical analyses.

Were made the following determinations:

- dosage of total nitrogen, variant mineralized with sulfuric acid, distillation and titration with H<sub>2</sub>SO<sub>4</sub>;
- dosage of total phosphorous – mineralization on wet way, with ammonium molibdate and reduction with stanium colure, colorimetric dosage (after Nicolov, 1976);
- dosage of total potassium, by mineralization with a mixture of sulfuric acid and percloric acid and dosage by flame photometry.

## RESULTS AND DISCUSSIONS

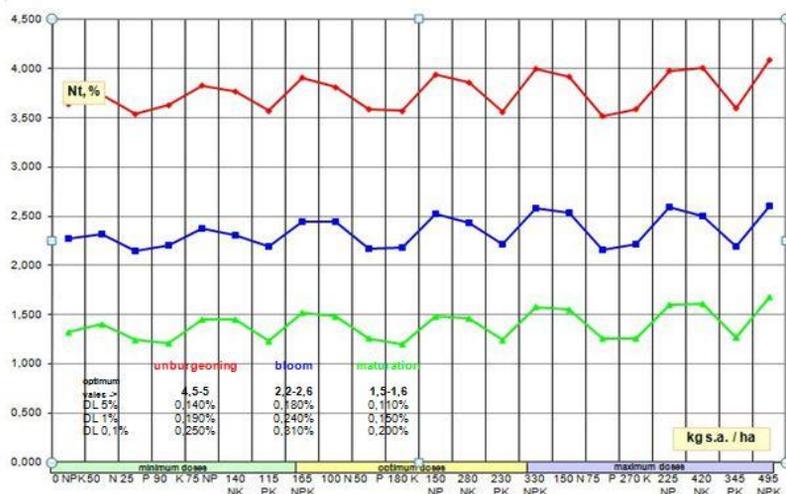
***The influence of interaction dose × combination of fertilizers elements, on the evolution of total nitrogen from plant (N<sub>t</sub>, %)***

Interaction of the factors dose × combination of fertilizers elements on the evolution of total nitrogen content in plant shows the following situations. In the budding phenol-phase, fertilization variant 165 kg a. s NPK, assures to the vineyard a content in total nitrogen of 3.91% N<sub>t</sub> (very significant), with a difference face to the un-fertilized control of 0.27% N<sub>t</sub>, smaller that at the ternary variant in optimal doses (330 kg a. s NPK) with only 0.09% N<sub>t</sub> and with only 0.1% smaller than the ternary variant in maximal doses (495 kg a. s NPK).

At flowering, could be observed on years average, that the minimal doses no matter what was the combination of used fertilizers element, did not succeed in statistical assuring of supply with nitrogen of vineyard, but the values of this index are in the optimal interval of supplying (2.2-2.6%) as the correct values of optimal and maximal doses.

At grapes maturation, supplying with total nitrogen is optimal for vineyard by using the variant 165 kg a. s NPK (1.52% N<sub>t</sub> – value distinct significant). Face to the variant 330 kg a. s NPK (optimal doses, ternary combination), variant 165 kg a. s NPK is smaller with only 0.06% N<sub>t</sub> and face to variant 495 kg a. s NPK (maximal doses, combination ternary) with only 0.16% N<sub>t</sub>.

Figure 1 put in light the variation of total nitrogen from plants under the impact of using the three levels of doses, in different combinations with fertilizers elements. At the three moments of vegetation, the top for total nitrogen supply in small doses, is recorded at variant 165 kg a. s NPK, very close to the top for total nitrogen supply in optimal doses (variant 330 kg a. s NPK).



**Fig. 1** The influence of fertilizers combination x measure interaction towards the evolution of total nitrogen (N<sub>t</sub> %) in leaves at Aligote in 3 phenological phases: unburgeoning, bloom, maturation of grapes

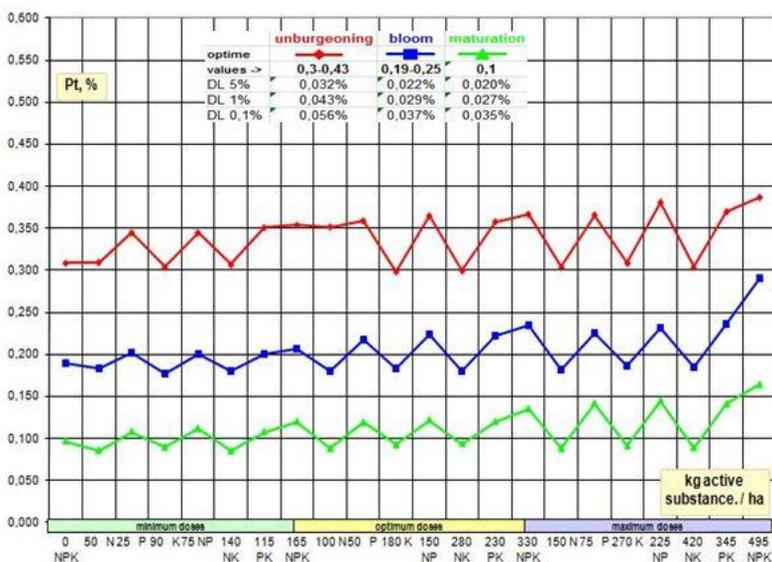
***The influence of interaction dose × combination of fertilizers elements, on the evolution of total phosphorous from plant ( $P_t$ , %)***

In the budding pheno-phase, variants “minimal doses” in combinations P, NP, PK and NPK offers to the vineyard a stage of optimal supply with phosphorous (optimal interval of supplying 0.30-0.43%  $P_t$ ). Variant 165 kg/ha a. s NPK realized in plant a supply of 0.354 %  $P_t$ , superior to the un-fertilized control with 0.045%  $P_t$  (distinct significant).

In flowering phenol-phase, variant 165 NPK on the average of the years taken in study, shows that the values of this index are not statistic assured, superior to the control and in the optimal interval, recording for this variant a content of 0.207%  $P_t$ .

At grapes maturation, variant “minimal doses”, and combination ternary – 165 NPK – realized in leave a level of 0.120%  $P_t$ , value superior to the control with 0.023%  $P_t$  (significant). Variant 330 NPK (optimal dose, combination ternary) realized an accumulation of 0.135%  $P_t$  in leave, only with 0.015%  $P_t$  more that at the previous cited variant.

Figure 2 express suggestive that at the level of all three moments of vegetation, the usage of minimal doses drives to accumulations of total phosphorous in leave at optimal parameters (variants 165 NPK, 115 PK, 75 NP and 25 P). Variant 495 NPK (maximal dose, combination ternary) leads for flowering and grapes maturation, at over-passing of the optimal doses of phosphorous in leave.



**Fig. 2** The influence of fertilizers combination x measure interaction towards phosphorus mobile forms' evolution ( $P_t$  %) in leaves at Aligote in 3 phenological phases: unburgeoning, bloom, maturation of grapes

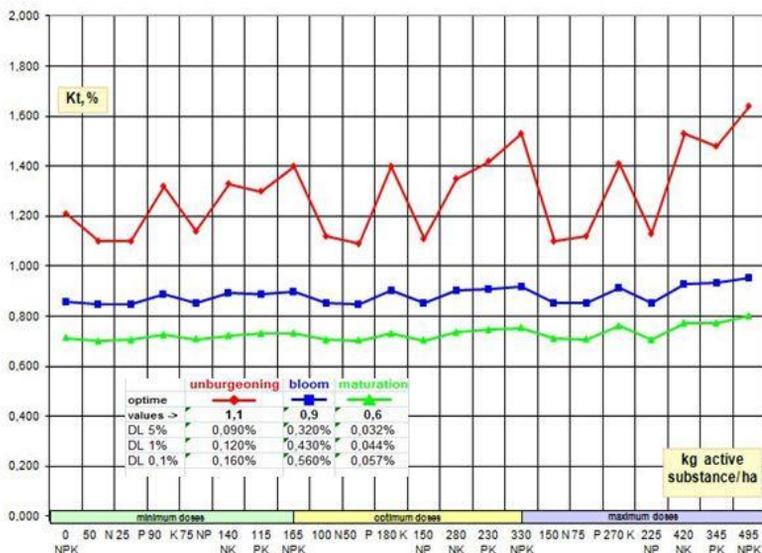
**The influence of interaction dose × combination of fertilizers elements, on the evolution of total potassium from plant ( $K_t$ , %)**

In budding phenol-phase, the minimal doses in binary and ternary combinations assure a very good content of leave in total potassium (fig.3).

Variant 115 kg a. s K realized 1.30%  $K_t$ , variant 90 kg a. s K realized 1.32%  $K_t$ , variant 140 kg a. s NK realized 1.33%  $K_t$ , all statistic assured, significant. Variant ternary in minimal doses - 165 kg a. s NPK, realized in plants an accumulation of 1.40%  $K_t$  (very good supply), value superior to the control with 0.19%  $K_t$  (very significant). Variant 330 kg a. s NPK realized 1.53%  $K_t$ , more above the optimum and only 0.13%  $K_t$  face to the variant 165 kg a. s NPK.

At flowering, variant 165 kg a. s NPK, even if it is statistic assured – significant – is very close to reach the optimal for this phenol-phase, records for this variant the value of 0.899%  $K_t$ . Variants 330 kg a. s NPK and 495 kg a. s NPK reaches optimal values of the total potassium respectively 0.917%  $K_t$  and 0.954%  $K_t$  (very significant).

In maturation pheno-phase, even if are reached levels of a very good supply with total potassium in plant (> 0.6%  $K_t$ ) at all the levels of dosage and in all combinations of fertilizers elements, only at the level of maximal doses (1.5 dose) values are statistical assured. Could be observed that the variant 495 NPK realized 0.801%  $K_t$ , with 0.2%  $K_t$  above optimum, being considered “luxury consumption”, exist the possibility of manifestation of the excess stage, by appearing external signs.



**Fig. 3** The influence of fertilizers combination x measure interaction towards potassium mobile forms' evolution ( $K_t$  %) in leaves at Aligote in 3 phenological phases: unburgeoning, bloom, maturation of grapes

## CONCLUSIONS

As a result of chemical fertilizers could be observed in a significant manner, the cumulative effect of the interaction of the studied factors and from this point of view the following things are observed:

1. Variant 165 NPK assures accumulation in  $N_t$  superior to unfertilized control, for all the three pheno-phases analyzed, placed as being a little bit sub-optimal at budding (3.91%  $N_t$ ), optimal for flowering (2.44%  $N_t$ ) and maturation (1.52%  $N_t$ ); doubling or tripling the administrated doses in ternary complex (variants 330 NPK and 495 NPK) did not solve the supplying in optimum with  $N_t$  of plants at budding (values  $< 4.5\%$   $N_t$ ), are at optimal at flowering (interval 2.2-2.6%  $N_t$ ) and optimal to an easy excess at maturation (1.58%  $N_t$  and respectively  $> 1.6\%$   $N_t$ ).

2. The rhythm of absorption and accumulation of the forms of total phosphorous is specific to the studied pheno-phases and are different in range with the fertilized variants: at budding variants 25 P, 75 NP, 115 PK and 165 NPK realized optimal accumulations of  $P_t$  (values  $> 0.30\%$   $P_t$ , assured statistic), in flowering pheno-phase the same variants accumulates optimal quantities of  $P_t$  (values in interval 0.19-0.25%  $P_t$ , un-assured statistic), and at the grapes' maturation, only variant 165 NPK to realize an optimal stage of  $P_t$  accumulation (value  $> 0.10\%$   $P_t$ , significant); usage of maximal doses applied in tertiary complex, variant 495 NPK leads to optimal accumulation of  $P_t$  at budding and over-optimal at flowering and grapes' maturation, showing the probability of appearance of phosphorous excess, but not manifested physiologic.

3. Analytical dates regarding the total potassium content in plant ( $K_t$  %) shows a clear influence of NPK relation on the evolution of this indicator: at budding, variant 165 NPK realized an accumulation over optimal in  $K_t$  (value  $> 1.1\%$   $K_t$ ), optimal in flowering pheno-phase (0.9%  $K_t$ ) and also over-optimal in maturation pheno-phase (values  $> 0.6\%$   $K_t$ ).

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**CONTRIBUTIONS TO THE STUDY BIOLOGY CHESTNUT  
MOTH *CAMERARIA OHRIDELLA* DESCHKA & DIMIČ,  
UNDER CONDITIONS IN THE HUSI AREA, VASLUI  
COUNTY**

**CONTRIBUȚII LA STUDIUL BIOLOGIEI MOLIEI MINIERE A  
FRUNZELOR DE CASTAN *CAMERARIA OHRIDELLA* DESCHKA &  
DIMIČ ÎN ZONA HUȘI, JUDEȚUL VASLUI**

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**Abstract.** Through this study bring the new contributions to the knowledge of the biology chestnut leaf mining moth. These observations were made in terms of Husi, Vaslui area and consisted of systematic recording of data on the conditions of the studied area, appearance and flying insect oviposition, completing the larval stage, the transformation in pupa stage and re-start every generation, observations what we've done using pheromone traps, "Atra-CAM" and using the Agroexpert program we calculated the sum of effective temperatures required at each stage of development. Order to better establish during each stage in the field I chose trees that have below branches, so as to be able to reach them and chose 10 whole leaves that we meant by a thread, and on them we follow all stages from egg to pupa and adult. We watched practically the egg stage, larva and pupa to butterfly leaving the mine by the same leaves and folioles.

**Key words:** *Cameraria ohridella*, "Atra-CAM", leaf mining moth.

**Rezumat.** Prin acest studiu se aduc noi contribuții la cunoașterea biologiei moliei miniere a frunzelor de castan. Aceste observații s-au realizat în condițiile zonei Huși, județul Vaslui și au constat în înregistrarea sistematică a datelor privind condițiile ecologice din zona studiată, apariția și zborul insectei, depunerea pontei, parcurgerea stadiului larvar, împuparea și reluarea ciclului fiecărei generații, observații ce s-au realizat cu ajutorul capcanelor cu feromoni "Atra-CAM" și cu ajutorul programului Agroexpert am calculat suma temperaturilor efective necesară fiecărui stadiu de evoluție. Pentru a stabili cât mai bine durata fiecărui stadiu în câmp s-au ales arborii care aveau ramurile mai jos, astfel încât să se poată ajunge la ele și, de pe aceste ramuri, am selectat 10 frunze întregi pe care le-am marcat cu un fir de ață, iar pe ele s-au urmărit toate stadiile de la ou până la pupă și adult. În mod practic am urmărit stadiul de ou, larvă și pupă, până la părăsirea minei de către fluture pe aceleași frunze și foliole.

**Cuvinte cheie:** *Cameraria ohridella*, "Atra-CAM", molia miniera a frunzelor

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## INTRODUCTION

In Romania the mining moth *Cameraria ohridella* Deschka & Dimic have first appeared in the west of the country, in Timisoara, in 1996, in 1998 is seen in the center of the country of Cluj-Napoca (Ilanovici, 2012; Perju T, 2001), and in 2005 the insect was appeared in Vaslui.

## MATERIAL AND METHOD

The purpose of the research subject of this paper was to know the biological cycle, flight dynamics, effective temperature sum necessary stages of development of chestnut mining moth *Cameraria ohridella* Deschka & Dimic, according to the Husi area, Vaslui county.

A view to achieving research on species bioecology *Cameraria ohridella* Deschka & Dimic, pheromone traps were used adhesive type "atra-CAM" by their reading of 3 in 3 days. It was used by a trap / location, and change baits were made monthly from May to September and out of 2012 to 2016.

Life cycle of the species have been established based on observations in the field, both visually where they were chosen 10 leaves that were marked with a thread and, and on the leaves of "observation" 3 in 3 days, I followed and I note laying and still other stages: larva, pupa and adult (Beratlief, 1998; Perju, 2004).

Observations on chestnut mining moth *Cameraria ohridella* Deschka & Dimic were performed in parks and chestnut alignments.

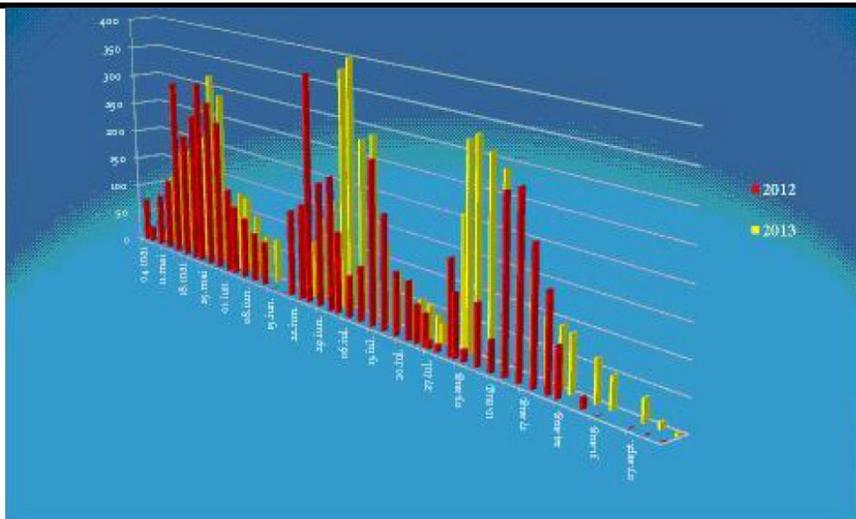
## RESULTS AND DISCUSSIONS

In the mining moth flight dynamics has been found that many insect has three flights, one for each generation, with one or two maximum one (fig. 1, fig. 2).

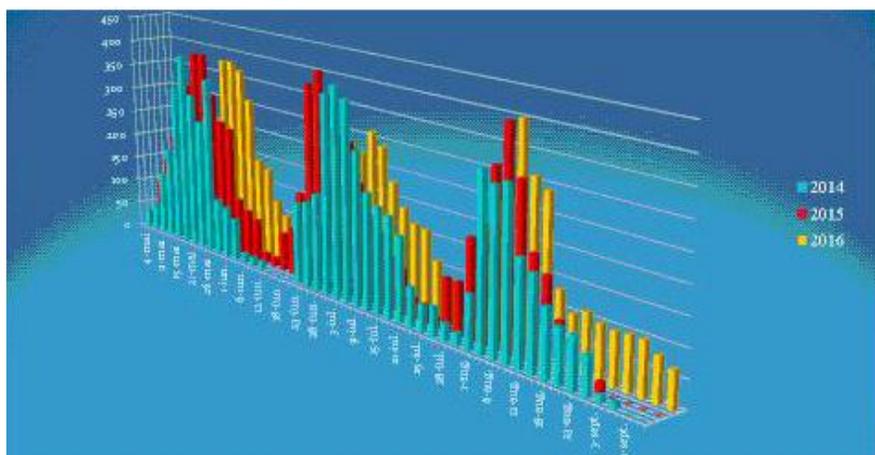
The first flight of the generation G3 large (hiemal) II begins decade of May until the end of May - early June and lasts between 15 and 25 days and maximum flight between 7 and 10 days.

The second flight numerous corresponding generation I, G1, takes place in late June - early July to the second decade of July for 17 - 24 days.

The third flight of the insect large corresponds generation II, G2, occurs in the first and second decade of August and lasts between 10 and 23 days and the maximum flight between 6 and 10 days.



**Fig. 1** The flight dynamics species *Cameraria ohriddella* Deschka & Dimic 2012-2013



**Fig. 2** The flight dynamics species *Cameraria ohriddella* Deschka & Dimic 2014-2016

The investigations carried out between 2012 - 2016 on the biological cycle, each year insect that has three generations per year (tab. 1) and winter in the pupa in a cocoon silky, made from the leaf epidermis.

After the appearance, butterflies climbs the trunks of trees where mating takes place. Sexual maturation and copulation duration is between 7 and 10 days.

Oviposition occurs on the surface of the lamellae between the leaf veins. According to the literature a female produces an average of 20-40 eggs (Georgescu, 2005).

Eggs are always deposited on the leaves normally developed, mature.

Incubation lasts 3 -10 days depending on temperature and humidity. After hatching larva immediately penetrates between the epidermis leaflet starts to feed. At first larval gallery is small and round, then it becomes erratic and extends between the ribs leaflet.

Larva in its development passes through five age, and two phase feeding: a phase-consuming and one unconsumer plant tissue, which occurs in the cocoon made in leaf.

Larval stage lasts depending on the temperature between 20 and 25 days.

Having reached complete development larva builds a mine between the leaf epidermis (cocoon) covered with a whitish below which turns into pupa. Pupal stage lasts about 8 to 10 days.

Table 1

The life cycle of *Cameraria ohridella* Deschka & Dimic the mining moth of chestnut

Year	Generation		
	I	II	III
2012	12 may – 20 june	28 june - 31 iulie	10 august – 5 may
2013	12 may – 17 june	29 june – 04 august	08 august – 10 may
2014	20 may – 29 june	05 july – 11 august	17 august – 08 may
2015	15 may – 21 i june	27 june - 01august	09 august – 09 may
2016	19 may – 26 june	01 july– 03 august	10 august -

$\Sigma(tn-t_0)$

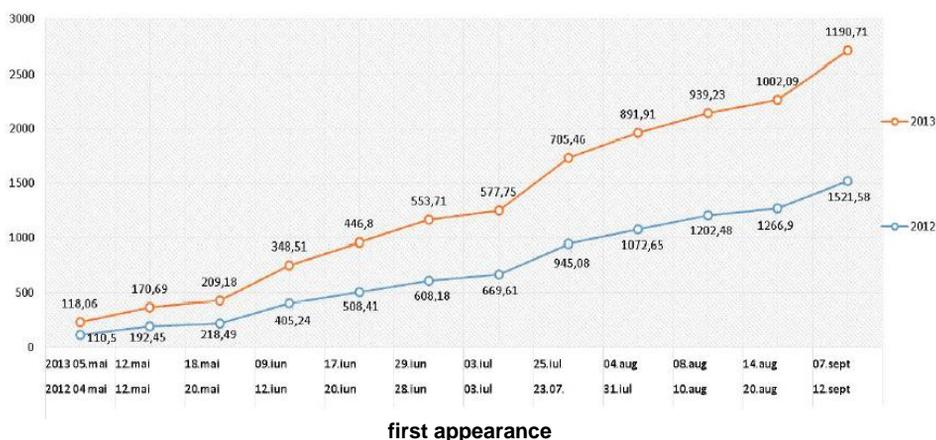


Fig. 3 The graphical representation of the first appearances of the development stages of mining moth *Cameraria ohridella* Deschka & Dimic, in 2012 – 2013



Fig. 4 The graphical representation of the first appearances of the development stages of mining moth *Cameraria ohridella* Deschka & Dimic, in 2014 – 2016

First generation adult emergence occurs at temperatures carry amount,  $\Sigma (t_n - t_o) = 400 \pm 500C$ , and for adults to II- generation of  $\Sigma (t_n - t_o) = 850 \pm 500C$  (fig. 3, fig. 4).

The length of a generation in weather conditions in the area Napa, is between 33 and 40 days. Usually it takes longer generation I of 38-40 days, and the second generation of shorter than 33-35 days, depending on temperature and humidity.

### CONCLUSIONS

Following comments made on species *Cameraria ohridella* Deschka and Dimic in terms of area Vaslui Huși- find the following conclusions:

- Under the conditions the area Husi in the four years of observations *Cameraria ohridella* species Deschka & Dimic, has three generations per year and overwinters in the pupal stage.
- The length of a generation ranged from 40 days to generation I and 33-35 days in generation II.
- The first generation adult emergence occurs at temperatures carry amount,  $\Sigma (t_n - t_o) = 400 \pm 500C$ , and for adults to II- generation of  $\Sigma (t_n - t_o) = 850 \pm 500C$ .
- Incubation takes between 3-10 days depending on temperature and humidity.
- The last larval stage, depending on the temperature and the relative humidity between 20 and 25 days, and pupated 8-10 days.
- In the flight dynamics mining moth it was found that numerous insect has three flights, one for each generation, with 1 or 2 maximum flight.

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**OBSERVATIONS ON WHEAT CROPS ENTOMOFAUNA  
COLLECTED AT SOIL TRAPS TYPE BARBER**

**OBSERVAȚII ASUPRA ENTOMOFAUNEI DIN CULTURILE DE GRÂU  
COLECTATĂ LA CAPCANELE DE SOL**

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**Abstract** The increase grain production is possible through the application of technology that involves the use of varieties with high yield, soil fertilization to provision according to their requirements, full mechanization of work and measures to prevent and combat specific pests and weeds. Regarding pests of grain crops grasses, they can cause annual havoc 13.8% compared to 11.6% produced by pathogens and weeds due to 9.5%. This paper presents the research on the epigenous entomofauna of wheat crops in 2016, in Focșani area used the traps type Barber and collecting biological material was conducted from May to July. The collection of the biological material was performed periodically on the following dates: 23.05; 07.06; 23.06; 07.07 using 18 traps belonging to the three experimental variants: •Variant 1 - wheat after sunflower; • Variant 2 - wheat after wheat; • Variant 3 - wheat after rape. The most commonly species found in this period are following the order: Coleoptera, Hymenoptera, Orthoptera, Heteroptera, Diptera, Colembolla etc. They were collected in addition to insects, and species belonging to the Crustacea class with the Isopods order, and the Arachnida class with the Araneae order.

**Key words:** entomofauna, wheat , traps type Barber

**Rezumat:** Creșterea producției de cereale este posibilă prin aplicare unor tehnologii care presupune folosirea soiurilor cu mare capacitate de producție, fertilizarea corespunzătoare a solurilor în funcție de cerințele acestora, mecanizarea completă a lucrărilor și prin măsuri de prevenire și combatere a buruienilor și dăunătorilor specifici. În ceea ce privește dăunătorii din culturile de cereale graminee, aceștia pot produce pagube anuale de 13,8% față de 11,6% produse de către agenții patogeni și față de 9,5% datorită buruienilor. În lucrarea de față sunt prezentate cercetările asupra entomofaunei epigeice din culturile de grâu în anul 2016, din zona Focșani, cu ajutorul capcanelor de tip Barber, iar colectarea materialului biologic s-a realizat din luna mai până în luna iulie. Colectarea materialului biologic s-a realizat periodic la următoarele date: 23.05; 07.06; 23.06; 07.07 utilizând 18 capcane ce aparțin celor trei variante experimentale: •Varianta 1 – grâu după floarea soarelui , • Varianta 2 – grâu după grâu, • Varianta 3 – grâu după rapiță. Speciile cel mai frecvent întâlnite în această perioadă aparțin următoarelor ordine: Coleoptera, Hymenoptera, Orthoptera, Heteroptera, Diptera, Colembolla etc. Au mai fost

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*colectate în afară de insecte, și specii aparținând clasei Crustacea, ord. Izopoda, dar și clasei Arachnida, ord. Araneae.*

**Cuvinte cheie:** entomofauna, grâu, capcane de tip Barber

## INTRODUCTION

Regardless of the agricultural production potential agricultural losses caused by diseases, insects, nematodes and weeds are less than 30% in countries with developed agriculture (average loss of 35% is considered) (Perju and Ghizdavu, 2001). If crops or in a forest, great damage is caused by a wide range of pests, which often are not specific to a single plant or plant families, and species that are found particularly in an ecosystem, which actually produce great damage (Antonescu *et al*, 2012). We must also remember that a number of pests, consider hunger, even if not all species of farm attacks, are common and cause damage of economic importance to a large number of crops.

## MATERIAL AND METHOD

To collect material were used Barber soil traps. They were placed in wheat area from Tișița in Vrancea country, 6 traps in each variant, namely:

- Untreated wheat for consumption during the growing season, only seed;
- Wheat for consumption on treatments that were applied during the growing season against pathogens and pests;
- Wheat seed which was applied technology to do so.

The traps were placed in two rows, 3 each in turn to the distance between them between 6 and 8 m and between two rows distance of about 10 m. To capture the species collected using a solution of formalin into the item concentration of 4 -5 %. Traps have worked from May until late June (Tălmăciu *et al*, 2007). The harvesting collected material was done at intervals of between 10 and 20 days during 2016. At each harvest species collected from each arm and traps were placed in gauze, previously eliminating crop residues or soil particles other impurities. Such evidence is in this way was then labeled, the label specifying:

- Date of collection;
- Variant;
- Trap number.

The material was then brought to the laboratory after being washed under running water species has been identified or counted specimens collected per trap, the variants and species.

The determination was made using the German Identification Manual (Reitter, 1908), another manual for identification (Panin, 1951; Manolache *et al*, 1982), and another source for the internet to compare different species pictures.

## RESULTS AND DISCUSSIONS

In total, the 3 variants were collected 5694 specimens belonging to 115 species (taxa). On variants situation is as follows (tab. 1, fig. 1):

- The consumption of untreated wheat were collected copies 2656 belonging to a 69 species;

- Wheat consumption treated specimens were collected in 1534 in total, belonging to a number of 64 species (taxa);

- Wheat seed were collected in 1504 samples in total, belonging to a number of 76 species (taxa);

A total of 32 species collected were common in the 3 experimental variations. These include: *Pentodon idiota* Hbst, *Opatrum sabulosum* L., *Pseudocleonus cinereus*, *Diptere*, *Dermestes lanarius* L., *Arahnide*, *Phyllotreta nemorum*, *Epicometis hirta* Poda, *Agriotes lineatus* L., *Amara aenea* Dejean, *Himenoptere* (wasp), *Tanymecus dilaticollis* Gyll., *Formicomus pedestris* Rossi, *Pleurophorus caesus* Panz., *Pterostichus marginalis* Dejean, *Colembole*, *Himenoptere* (ants), *Gryllus campestris* Gyll., *Harpalus distinguendus* Duft, *Homoptere* (cycads), *Blaps mortisaga* L., *Conosoma bipunctata*, *Ortoptere* (locust), *Homoptere* (aphids), *Anthicus antherinus* L., *Heteroptere* (bedbugs), *Armadillidium vulgare* Latreille, *Coccinella 7 punctat*, *Anthicus floralis*, *Colodera nigrita* Mnnh., *Hypnoidus pulchellus* Linnaeus, *Pteryngium crenatum* Fabricius.

A number of 69 species were collected in one variant, they were: *Ceuthorrhynchus punctiger*, *Pseudophonus rufipes*, *Pterostichus lepidus*, *Cassida nobilis*, *Anthicus humilis*, *Cryptophagus dentatus*, *Orchestes fagi*, *Otiorrhynchus singularis*, *Calosoma inquisitor*, *Necrophorus antennatus*, *Callistus lunnatus*, *Anthicus gracilis*, *Eurygaster integriceps*, *Anisoplia segetum*, *Oxyporus rufus*, *Coccinella quatuordecimpustulata*, *Crypticus quisquilius*, *Broscus cephalotes*, *Coccinella 5 punctata*, *Onthophagus taurus*, *Stomodes gyrosicollis*, *Anobium punctatum* Deg., *Aphodius fimetarius*, *Staphylinus caesareus*, *Staphylinus caesareus*, *Calathus rufipes*, *Astenus filiformis*, *Mycetophagus populi*, *Cephus pygmaeus*, *Oulema melanopa* L., *Psammobius porcicollis* Illiger, *Chrysopa perla* L., *Anisodactylus binotatus* F., *Ophonus azureus*, *Harpalus smaragninus* Duft, *Paramecosoma melanocephalum* Hrbst, *Tanymecus palliatus* F., *Atomaria fuscicollis*, *Bidessus geminus* F., *Amara eurynota* Panz., *Hister quadrimaculatus* Illiger, *Brachynus explodens* Duft, *Ophonus sabulicola* Panz., *Harpalus cupreus* Dejean, *Cantharis fusca*, *Calathus fuscipes* Goeze, *Tachyusa constricta*, *Scirtes hemisphaericus* L., *Anthicus quadriguttatus* Haldeman, *Selatosomus latus* F., *Cercyon lateralis* Marsh., *Cryptophagus dorsalis* Sahlberg, *Cartodere ruficollis* Marsh, *Paederus limnophilus* Erichson, *Cerylon ferrungineum* Steph., *Paradons quadrisignatus*, *Melanotus brunnipes* Germ.

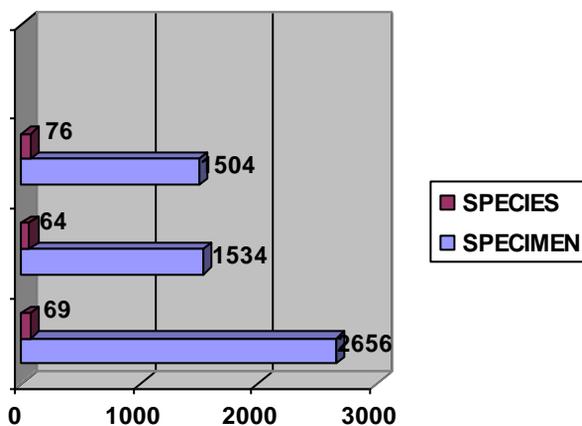
The structure and abundance of the collected entomofauna from the wheat crops

No.	Name of species (taxa)	Variant			Total
		Untreated wheat for consumption	Wheat consumption	Wheat for seed	
1	<i>Pentodon idiota</i>	29	35	9	73
2	<i>Cetonia aurata</i>	1	1	-	2
3	<i>Opatrum sabulosum</i>	69	47	27	143
4	<i>Pseudocleonus cinereus</i>	2	4	1	7
5	Diptere	50	88	53	191
6	<i>Dermestes lanarius</i>	32	22	56	110
7	<i>Ceutorhynchus punctiger</i>	2	-		2
8	<i>Drasterius bimaculatus</i>	115	31	-	146
9	Arahnide	198	287	150	635
10	<i>Phyllotreta nemorum</i>	21	112	6	139
11	<i>Epicometis hirta</i>	78	99	18	195
13	<i>Agriotes lineatus</i>	11	9	12	32
14	Himenoptere (wasp)	25	25	13	63
15	<i>Amara aenea</i>	1	4	2	7
16	<i>Tanymecus dilaticollis.</i>	25	15	12	52
17	<i>Pedinus femoralis</i>	23	6		29
18	<i>Formicomus pedestris</i>	41	22	37	100
19	<i>Pleurophorus caesus</i>	4	8	14	26
20	<i>Pterostichus marginalis</i>	1	-	259	260
21	<i>Pseudophonus rufipes</i>	6	-		6
22	Colebole	1198	150	68	1416
23	<i>Anthicus humeralis</i>	13	-	4	17
24	<i>Metabletus truncatulus</i>	5	5		10
25	Himenoptere (ants)	128	230	210	568
26	<i>Gryllus campestris</i>	5	21	17	43
27	<i>Pterostichus lepidus</i>	3	-		3
28	<i>Harpalus distinguendus</i>	16	10	7	33
29	<i>Cassida nobilis</i>	2	-	2	4
30	<i>Anthicus humilis</i>	6	-	3	9
31	Miriapode	2	2	-	4
32	<i>Haplothrips tritici</i>	140	5	-	145
33	Homoptere (cycads)	204	32	19	255
34	<i>Cryptophagus dentatus</i>	21	-	-	21
35	<i>Blaps mortisaga</i>	2	1	1	4
36	<i>Pyrrhocoris apterus</i>	-	51	2	53
37	<i>Conosoma bipunctata</i>	11	4	233	248
38	<i>Corticaria longicornis</i>	3	-	3	6
39	<i>Orchestes fagi</i>	7	-	1	8
40	<i>Aphthona euphorbia</i>	10	-	2	12
41	<i>Otiorrhynchus laevigatus</i>	6	2	-	8
42	<i>Otiorrhynchus singularis</i>	2	-	1	3
43	<i>Pterostichus aterrimus</i> var. <i>niger</i>	1	1	-	2

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44	<i>Calosoma inquisitor</i>	1	-	1	2
45	<i>Necrophorus antennatus</i>	-	1		1
46	<i>Callistus lunatus</i>	-	1	-	1
47	Homoptere (aphids)	20	5	3	28
48	Ortoptere (locust)	9	2	1	12
49	<i>Anthicus antherinus</i>	13	57	13	83
50	Heteroptere (bedbugs)	8	9	7	24
51	Himenoptere(parasit wasp)	7			7
52	<i>Anthicus gracilis</i>	11			11
53	<i>Ityocara rubens</i>	1	-	12	13
54	<i>Harpalus tardus</i>	4	2		6
55	<i>Armadillidium vulgare</i>	5	25	21	51
56	Curculionide	1			1
57	<i>Eurygaster integriceps</i>	2			2
58	<i>Anisoplia segetum</i>	1			1
59	<i>Idiochroma dorsalis</i>	1	-	8	9
60	<i>Coccinella 7 punctata</i>	9	5	1	15
61	<i>Anthicus floralis</i>	5	5	28	38
62	Himenoptere (bees)	2	1		3
63	<i>Colodera nigrita</i>	4	30	15	49
64	<i>Hypnoidus pulchellus</i>	4	5	1	10
65	<i>Pteryngium crenatum</i>	8	9	59	76
66	<i>Oxyporus rufus</i>	9			9
67	<i>Zabrus blapoides</i>	1	1		2
68	<i>Coccinulla quatuordecimpustulata</i>	2			2
69	<i>Tachyporus ruficolis</i>	3		8	11
70	<i>Crypticus quisquilius</i>	1			1
71	<i>Emphilus glaber</i>	1	1	2	4
71	<i>Brosicus cephalotes</i>	1			1
72	<i>Coccinella 5 punctata</i>	1			1
73	<i>Silpha obscura</i>	-	1	12	13
74	<i>Onthophagus taurus</i>	-	1		1
75	<i>Phyllotreta atra</i>	-	7	5	12
77	<i>Stomodes gyrosicollis</i>	-	3		3
78	<i>Anobium punctatum</i>	-	5		5
79	<i>Aphodius fimetarius</i>	-	2		2
80	<i>Phyllotreta nodicornis</i>	-	5	5	10
81	<i>Staphylinus caesareus</i>	-	1		1
82	<i>Microletes maurus</i>	-	7	5	12
83	Acari (spider)	-	4	3	7
84	<i>Calathus rufipes</i>	-	1		1
85	<i>Astenus filiformis</i>	-	1		1
86	<i>Mycetophagus populii</i>	-	1		1
87	<i>Cephus pygmaeus</i>	-	1		1
88	<i>Pterostichus cupreus</i>	-	-	1	1
89	<i>Oulema melanopa</i>	-	1		1
90	<i>Sipalis circularis</i>	-	1	1	2
91	<i>Psammobius porcicollis</i>	-	1		1
92	<i>Chrysopa perla</i>	-	-	2	2

93	Anisodactylus binotatus	-	-	3	3
94	Ophonus azureus	-	-	1	1
95	Harpalus smaragninus	-	-	3	3
96	Paramecosoma melanocephalum	-	-	3	3
97	Tanymecus palliatus	-	-	3	3
98	Atomaria fuscicollis	-	-	1	1
99	Bidessus geminus	-	-	1	1
100	Amara eurynota	-	-	3	3
101	Hister quadrimaculatus	-	-	1	1
102	Brachynus explodens	-	-	5	5
103	Ophonus sabulicola	-	-	4	4
104	Harpalus cupreus	-	-	1	1
105	Scirtes hemisphaericus	-	-	1	1
106	Anthicus quadriguttatus	-	-	2	2
107	Selatosomus latus	-	-	1	1
108	Cercyon lateralis	-	-	7	7
109	Cryptophagus dorsalis	-	-	1	1
110	Cartodere ruficollis	-	-	2	2
111	Paederus limnophilus	-	-	1	1
112	Melanotus brunnipes	-	-	1	1
113	Metabletus foveatus	-	-	1	1
114	Harpalus spp.	-	-	1	1
115	Zabrus tenebrioides	-	-	1	1
Total species		2656	1534	1504	5694



**Fig.1** The number of collected specimens and species at the three variants

The largest number of collected specimens from the 3 variants, over 50 specimens have played a total of 14 species. These were (tab. 2): *Conosoma bipunctata*, 248 samples representing 4.35% of the total; *Pterostichus marginalis*, with a total of 260 specimens, representing 4.56% of the total; *Epicometis hirta*

Poda, with a total of 195 specimens, representing 3.42 % of the total; *Opatrum sabulosum* L., with a total of 143 specimens, representing 2.51% of the total; *Haplothrips tritici*. with a total of 145 specimens, representing 2.54% of the total; *Drasterius bimaculatus* Rossi, with a total of 146 specimens, representing 2.56% of the total; *Phyllotreta nemorum*, with a total of 139 specimens, representing 2.44% of the total; *Dermestes lanarius* L., with a total of 110 specimens, representing 1.93% of the total; *Formicomus pedestris* Rossi, with a total of 100 specimens, representing 1.75% of the total; *Pentodon idiota* Hbst, with a total of 73 specimens, representing 1.28% of the total; *Anthicus antherinus* L., with a total of 83 specimens, representing 1.45% of the total; *Pteryngium crenatum* Fabricius, with a total of 76 specimens, representing 1.33% of the total; *Pyrrhocoris apterus* Firebug, with a total of 53 specimens, representing 0.93% of the total; *Colodera nigrita*., with a total of 49 specimens, representing 0.86% of the total.

Table 2

**The structure of species (taxa) with the largest number of specimens collected**

No.	Name of species (taxa)	Total	%
1	<i>Conosoma bipunctata</i>	248	4.35
2	<i>Pterostichus marginalis</i>	260	4.56
3	<i>Epicometis hirta</i>	195	3.42
4	<i>Opatrum sabulosum</i>	143	2.51
5	<i>Haplothrips tritici</i>	145	2.54
6	<i>Drasterius bimaculatus</i>	146	2.56
7	<i>Phyllotreta nemorum</i>	139	2.44
8	<i>Dermestes lanarius</i>	110	1.93
9	<i>Formicomus pedestris</i>	100	1.75
10	<i>Pentodon idiota</i>	73	1.28
11	<i>Anthicus antherinus</i>	83	1.45
12	<i>Pteryngium crenatum</i>	76	1.33
13	<i>Pyrrhocoris apterus</i>	53	0.93
14	<i>Colodera nigrita</i>	49	0.86
<b>TOTAL</b>		<b>5694</b>	-

## CONCLUSIONS

1. In the 3 variants were collected a number of 5694 samples belonging to a total of 115 invertebrate species of wheat. The situation, on the variants is as follows:

- In the variant, the consumption of untreated wheat were collected in a total of 2656 samples 69 belonging to a number of species (taxa);
- The variant number 2 treaty wheat consumption have been collected 1534 specimens belonging to a number of 64 species (taxa);
- The variant number three, wheat for seed the samples were collected from to 1504 belonging to 76 one species (taxa).

2. During the period of observations, a number of 32 species were common to the three variants while a total of 69 species were collected from a single experimental variant.

3. A number of 14 species had more than 50 specimens. The species with the highest number of specimens were *Conosoma bipunctata*, with 248 specimens, representing 4.35% of the total; *Pterostichus marginalis*, with 260 specimens, representing 4.56% of the total; *Epicometis hirta* with 195 pieces, representing 3.42 % of the total, *Opatrum sabulosum* 143 specimens, representing 2.41% of the total, with 145 samples *Haplothrips tritici*, representing 2.54% of the total; *Dermestes lanarius*, with 110 specimens, representing 1.93% of the total and *Phyllotreta nemorum* 139 specimens, representing 2.44% of the total.

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## EFFECT OF PLANTING DISTANCES ON THE GROWTH AND YIELD OF RHUBARB

### EFFECTUL DISTANȚELOR DE PLANTARE ASUPRA CREȘTERII ȘI PRODUCȚIEI DE REVENT

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**Abstract.** *The aim of the present research was to study the influence of planting distances on the growth and yield on rhubarb, cultivars of Victoria, Glanskin's perpetual and local population. Applying differential cultivation technology, rhubarb yield varies according to crop density. The highest production was obtained in case of Victoria cultivar at density of 13.330 pl.ha<sup>-1</sup> (0.75 m x 1 m). Statistically assured yields were also obtained at the density 10.000 pl.ha<sup>-1</sup> (1 m x 1 m) on the same cultivar. Total yield varied within wide limits according to planting distances, ranging from 24.480 kg.ha<sup>-1</sup> to 41.460 kg.ha<sup>-1</sup>.*

**Key words:** plant density, growth and yield, cultivars, rhubarb

**Rezumat.** *Scopul cercetării de față a fost acela de a studia influența distanțelor de plantare asupra creșterii și producției la revent, în cazul cultivarelor Victoria, Glanskins perpetual și o populație locală "de Moldova". Prin aplicarea diferențiată a tehnologiei de cultivare, producția de revent variază în funcție de distanțele de plantare. Cea mai ridicată producție s-a înregistrat în cazul în cultivarului Victoria, când plantarea s-a făcut la distanțe de 0,75 m x 1,0 m. Producții, de asemenea, asigurate statistic au mai fost obținute și în cazul distanței de plantare de 1 m x 1 m la același cultivar. Producția totală a variat în limite foarte largi în funcție de distanța de plantare, variind de la 24,480 kg/ha la 41,460 kg/ha.*

**Cuvinte cheie:** densitate, creștere și producție, cultivare, revent

## INTRODUCTION

The rhubarb (*Rheum rhabarbarum* L.) is a less known and spread crop in Romania. It is a perennial vegetable species, adapted to cold temperate climate (Ciofu *et al.*, 2004; Indrea *et al.*, 2007).

Rhubarb is originated in the Himalayas, where its root was an important medicine believed to purge the body of ill humors (Stan *et al.*, 2003).

In our country rhubarb is more cultivated in the western part of the country and it is used for compot, jam and other dessert (Treptow, 1985).

In the last time rhubarb products are spread by the supermarket on all over the country. So it is a good opportunity for farmers to cultivate this species also in other parts of the country, not only in the traditional ones.

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For this reason, our research was focused to evaluate the possibilities to cultivate rhubarb in the environmental condition of the Eastern part of Romania.

To achieve this good our objective was to study the influence of the cultivar and planting distances on the crop and, mainly, on the yield (Stoleru, 2013).

Distance between plants in the row and between rows is a technological factor influencing crop density, which is number of plants per unit area. This technological factor, determined directly from the feeding soil surface, light regime etc. (Loughton, 1969).

## MATERIAL AND METHOD

Management of experiment. To achieve the goal and objectives of this research work, an experimental was done at “V. Adamachi” Experimental Station of the Agronomic University, using root cuttings of Glanskin’s perpetual (fig.1), local population (fig. 2) and Victoria (fig.3) cultivars. Harvested area of experimental plots covered the 5 plants.

Considering the importance studying factors in the growing technology, their ability to change and taking into account the possibilities of organizing experience, it was established hierarchy of factors, as follows:

1. A factor – cultivars, with three graduations: Glanskin’s perpetual, local population and Victoria;
2. B factor – planting distances, with two graduations: 0.75 x 1.00 m and 1.00 x 1.00 m.



**Fig. 1** Rhubarb – Glanskin’s perpetual (original)



**Fig. 2** Rhubarb – Local Population (original)



**Fig. 3** Rhubarb – Victoria (original)

**Collection and processing the experimental data.** The experimental data collection was carried out observations and weight measurements, according to the experimental technique used in experiments. During 2015 were made a total of eight harvesting: 04.04, 11.04, 20.04, 28.04, 08.05, 15.05, 26.05 and 18.06.

The experimental variants were compared with the experimental mean, using the percentage reporting and differences. The influence of experimental factors was assessed using ANOVA. The significance of differences was assessed on the basis of LSD (least significant difference) for three degrees of confidence (95%, 99%, 99.9%).

## RESULTS AND DISCUSSIONS

Applying differential cultivation technology, rhubarb production varies according to cultivar and crop density.

Regarding to the influence of planting distances and cultivar, during 2015, it ranged from 24.48 t/ha at Glanskin's perpetual cultivar with 1.00 x 1.00 m planting distance to 41.46 t/ha at Victoria cultivar with 0.75 x 1.00 m planting distance (tab. 1).

Table 1

**Influence of cultivar and planting distances at rhubarb crop**

Variants	Total yield (t/ha)	% to the average	Difference to average (t/ha)	Semnificance of differences
c <sub>1</sub> d <sub>1</sub>	28.76	89.07	-3.53	00
c <sub>1</sub> d <sub>2</sub>	24.48	75.81	-7.81	000
c <sub>2</sub> d <sub>1</sub>	34.89	108.05	2.60	**
c <sub>2</sub> d <sub>2</sub>	29.13	90.21	-3.16	00
c <sub>3</sub> d <sub>1</sub>	41.46	128.40	9.17	***
c <sub>3</sub> d <sub>2</sub>	35.03	108.49	2.74	**
<b>x (Average)</b>	32.29	100.00	0.00	-

LSD 5% = 1,01 t/ha; LSD 1% = 2,05 t/ha; LSD 0,1% = 7,41 t/ha

c<sub>1</sub> – Glanskin's perpetual; c<sub>2</sub> – Local population; c<sub>3</sub> – Victoria; d<sub>1</sub> – 0,75 x 1,00 m; d<sub>2</sub> – 1,00 x 1,00 m

### **Influence of planting distances on rhubarb yield**

Very negative differences significantly, compared to the average have been obtained when Glanskin's perpetual cultivar is planted at distances 1.00 x 1.00 m.

Positive differences compared to the average have been obtained when

Glanskin's perpetual cultivar planted at distances of 0.75 x 1.00 m, and local population cultivar planted at distances of 1.00 x 1.00 m.

The differences obtained between experimental variants and experimental mean average, ranged from -7.81 t/ha for Glanskin's perpetual cultivar planted at 1.00 x 1.00 m, up to 9.17 t/ha for Victoria cultivar planted at 0.75 x 1.00 m.

The total yield in case of rhubarb crop ranged from 24.48 t/ha, for Glanskin's perpetual cultivar planted at distances 1.00 x 1.00 m, to 41.46 t/ha, for Victoria cultivar planted at distances 0.75 x 1.00 m.

## CONCLUSIONS

1. Regarding the influence of planting distances and cultivar on rhubarb total yield during 2015, it ranged from 24.48 t/ha for Glankin's perpetual cultivar planted at distances 1.00 x 1.00 m, to 41.46 t/ha, for Victoria cultivar planted at distances 0.75 x 1.00 m.

2. Very negative differences significantly, compared to the average have been obtained when Glankin's perpetual cultivar is planted at distances 1.00 x 1.00 m.

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## CONTRIBUTIONS TO THE IMPROVEMENT OF THE FERTILITY REGIM ON A SWEET PEPPER CROP

### CONTRIBUȚII LA ÎMBUNĂȚIREA REGIMULUI DE FERTILIZARE LA O CULTURĂ DE ARDEI GRAS

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**Abstract.** *The research conducted within this study aimed to evaluate the influence of fertilization simultaneously with drip irrigation in a crop of peppers established in a tunnel. Trials were organized in a tunnel belonging to the Vegetable discipline, located in "V. Adamachi" Didactic Farm, in Iasi. The purpose of the research was to evaluate the effect of the fertilization method, using conventional fertilizers, applied to the soil surface by spraying, and based on micro-organisms, simultaneously with drip irrigation, compared to the unfertilized version. Thus, after fertigation, there were found differences concerning the plant height, the number of flowers and fruits on a plant, the average weight of fruit and the production per hectare. The production obtained by the Brilliant F1 pepper cultivar in the fertigation version was of 85.850 kg / ha.*

**Keywords:** fertigation, sweet pepper, yield, tunnel

**Rezumat.** *Cercetările efectuate în cadrul acestei lucrări au avut drept scop evaluarea influenței fertilizării concomitent cu irigarea prin picurare într-o cultură de ardei amplasată într-un solar. Experiențele au fost organizate într-un solar aparținând disciplinei de Legumicultură, situat în Ferma Didactică "V. Adamachi", din județul Iași. Scopul cercetărilor a fost de a evalua efectul metodei de fertilizare, folosind îngrășăminte clasice, aplicate prin împrăștiere pe suprafața solului, și pe bază pe microorganisme, concomitent cu irigarea prin picurare, comparativ cu varianta nefertilizată. Astfel, după fertirigare, s-au constatat diferențe în ceea ce privește înălțimea plantelor, numărul de flori și fructe pe plantă, greutatea medie a fructelor și a producției la hectar. Producția obținută de cultivarul de ardei Brilliant F1, la varianta fertirigată, a fost de 85,850 kg/ha.*

**Cuvinte cheie:** fertirigare, ardei, producție, solar

## INTRODUCTION

Fertigation, a modern concept of agricultural technique, is the method by which the fertilizing substances are distributed to the plants simultaneously with the irrigation water (Biolan *et al.*, 2010).

Fertigation is usually associated with localized irrigation, although it may be associated with any other irrigation method. By fertigation, a synergistic effect

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is obtained, the water improving the absorption of the fertilizer, at the same time, this one making the use of water efficient (Țenu, 2004).

The basics of an efficient and rational fertigation are knowing the crop's nutritional requirements (minerals and hydro), the soil's fertility and the quality of the irrigation water, combined with an efficient management of the irrigation technique.

Fertigation, as a method of fertilizer application, is supported by a number of advantages, such as: the increase of the nutrient absorption by plants, the reduction of the amount of nutrients, the reduction of water consumption due to the increase of the roots volume capacity to retain and store water; the application of nutrients can be controlled according to the phenophase growth; the reduction of the root risk of contracting communicable diseases through the contaminated soil and the elimination of the soil erosion. The bell pepper (*Capsicum annuum L.* convar. *grossum L.*) is a vegetable plant, rich in antioxidants, phenolic compounds, especially PCA - p-coumaric acid (Dimitriu *et al.*, 2016), vitamin C (150-300 mg/100g) and carotenoids (1.8 to 4.5 mg/100g) (Hoble, 2010).

Pepper has high water requirements during the growing season, due to the poorly developed root system and the abundant biomass exposure to the strong perspiration (Tanaskovik, 2013). In the case of sweet pepper, the amount of fertilizer has a distinctly significant influence on the differences in the total yield (Stan *et al.*, 2006).

The cultivar is an important factor for a successful fertilization, along with drip irrigation. This one must be suitable for the salt stress conditions (Stoleru *et al.*, 2012).

## MATERIAL AND METHOD

The research was carried out in a semicircular tunnel, located in the "V. Adamachi" Didactic Farm, within USAMV Iasi, with an area of 270 m<sup>2</sup>. The studied plants belong to the *Brillant F1* sweet pepper cultivar and are grouped into four experimental versions (tab. 1), on tapes, the distance between the tapes being of 90cm and the distance between the plants in a row, of 45 cm, resulting a density of 31740 plants/ha (fig. 1). The protection tape for the experiment was established with the same, unfertilized hybrid.

Table 1

Experimental versions (2016)

Experimental version	Fertilization methods
V <sub>1</sub>	Drip fertigation, using soluble fertilizers
V <sub>2</sub>	Classic fertilization, by spreading on the soil surface and incorporated
V <sub>3</sub>	Microorganisms fertilization, by spraying on the soil and incorporated
V <sub>4</sub>	Unfertilized (Mt)

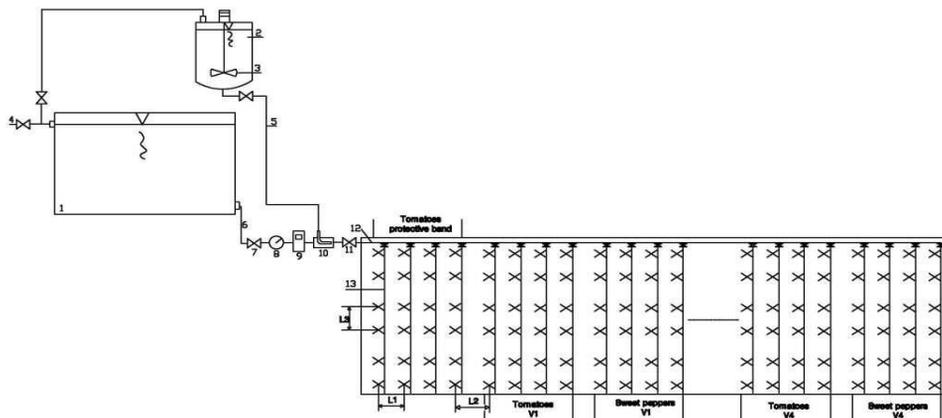
For the irrigation of the plants, a drip irrigation line was used, with the tapes' diameter of 16 mm and the distance between the drainers of 10 cm. During the growth cycle, it has been administered an amount of water equivalent to 5600 m<sup>3</sup>/ha, the watering being carried out every other day, in the time range 8<sup>00</sup> - 10<sup>00</sup> or 7<sup>00</sup> - 9<sup>00</sup>, based on the recorded temperature.

In order to perform the experiments, a fertigation facility was designed and built within the disciplines of Agricultural Engineering, consisting of a water tank with constant

watering, with a capacity of 20 tonnes; a tank for preparing the concentrated solution of fertilizer, an automatic system of watering programming and a watering line (fig. 1).

The plants in the  $V_1$  version were fertilized along with drip irrigation, twice a week, the fertigation being carried out in the morning. It was used Nutrispore<sup>®</sup> fertilizer - NPK (MgO) 10/30/10 - 300 kg / ha, NPK Nutrispore<sup>®</sup> (MgO) 10/30/15 - 425 kg/ha and NPK 12-48-8 Nutrispore<sup>®</sup> - 400 kg/ha. In order to have a good fertigation, the plant nutrition has been carried out periodically, twice a week, between two consecutive waterings.

The plants in version 2 were chemically fertilized, by spreading, on the surface of the soil, in each plant area, in the equivalent of 200 kg/ha NPK 20-20-20 Cristaland<sup>®</sup>, applied during basic fertilization; 250 kg/ha Cristaland<sup>®</sup> NP 15-50 + 2MgO, applied in the floral button phase (the first inflorescence) and 200 kg/ha of NPK 9-18-27 + 2 MgO Cristaland<sup>®</sup>, applied in the first fruit phenophase.



**Fig. 1** The scheme of trial organization

1- buffer water tank; 2- tank for the preparation of the fertilizer watering solution; 3- stirrer; 4 – connection for supplying the buffer water tank; 5 – connection for supplying the fertilizer solution; 6 – connection for supplying the irrigation facility; 7 and 11 – valves; 8 – flowmeter; 9 – programmer; 10 – ejector for mixing water - solution of fertilizer; 11 – watermeter; 12 – main pipeline; 13 –dropping pipeline; L1 – distance between rows on a tape (60 cm); L2 – distance between tapes (80 cm); L3 – distance between plants in a row (45 cm).

The plants in the  $V_3$  version were fertilized with a micro-organism based fertilizer, Micoseed<sup>®</sup> MB, by spreading, in each plant area, in the equivalent of 60 kg/ha, applied in order to prepare the ground, 2-3 days before planting the peppers. According to the data from the specialty literature, Micoseed MB<sup>®</sup> is a fertilizer based on *Glomus sp.*, *Beauveria sp.*, *Metarhizium sp.* and *Trichoderma sp.* (Stoleru *et al.*, 2014). In this version, during the growing season, there were also applied two fertilizations with Nutryaction<sup>®</sup> 2, in the amount of 5 L/ha, to stimulate the biological activity of the microorganisms.

In the  $V_4$  control version, there have been applied drip irrigations, under the same conditions as in the  $V_1$ ,  $V_2$  and  $V_3$  versions.

Weekly, biometric measurements were performed, by which the dynamics of the plant growth has been determined, following the height of the plant, the number of flowers and the number of related flowers/fruits.

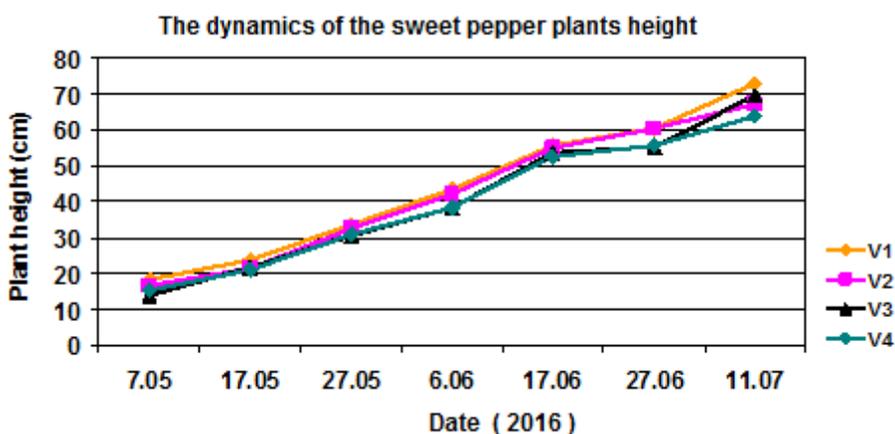
The sweet pepper plants were carried for according to data from the specialty literature (Ciofu *et al.*, 2004; Stoleru *et al.*, 2014). The processing of the experimental data was carried out using the analysis of the version (ANOVA), by calculating the limit differences (Leonte, 1997).

## RESULTS AND DISCUSSIONS

### Experimental results concerning the sweet pepper plant growth indicators

The dynamics of the sweet pepper plant height in 2016 is shown in figure 2. Seven biometric measurements were performed every 7 ... 10 days, starting with the third week after planting. The *Brilliant F1* sweet pepper cv. presents an indeterminate growth, being used for two crops. The average height obtained after the measurements was of 42.2 cm.

From figure 2 it can be seen that, at the beginning of the growing season (7-27/05/2016), although the growth was constantly in all the four versions, the most significant increase of the *Brilliant F1* hybrid occurred in the fertiligated version, V<sub>1</sub>, registering a value of 33.6 cm.



**Fig. 2** The graph of plant height dynamics

The data presented in Fig. 2 shows that, in the unfertilized version, V<sub>4</sub>, the plant height was lower at the beginning of the growing season compared to the fertilized versions.

In the second half of the growing season, it can be seen that in the V<sub>1</sub> (fertiligation) and V<sub>2</sub> (classic), the growth remained constant and close in value, with a medium value of 55.6 cm or 55 cm. In the V<sub>3</sub> and V<sub>4</sub> versions, the plant height was lower, with medium values of 53.6 and 52.6 cm.

At the end of the growing season, the average height of sweet pepper plants ranged from 63.6 cm in the unfertilized version - V<sub>4</sub>, till to 72.8 cm in the fertiligated version - V<sub>1</sub>.

The results concerning the productivity of the sweet pepper plants are given in table 2. The average number of flowers on sweet pepper sprout ranged from 1.52, registered in the control version ( $V_4$ ), till to 4 flowers, obtained in the fertigated version ( $V_1$ ).

Concerning the average number of fruits per plant, it ranged from 4.45 in the unfertilized version -  $V_4$ , up to 11.30, in the fertigated version -  $V_1$ . Medium values close to the version 1 were made in the classic fertilization version -  $V_2$  (9.55 cm).

Regarding the average weight of the *Brilliant F1* sweet pepper fruit, it ranged from 100.45 g in  $V_4$  (unfertilized) to 131.81 g in  $V_1$  (fertigated).

Table 2

**Results concerning the dynamics of the pepper plant development (2016)**

Experimental version	Average number of flowers on the sprout	Average fruit number in a plant	Average fruit weight (g)
$V_1$	4	11.3	131.81
$V_2$	3.52	9.55	102.22
$V_3$	2.96	4.85	101.15
$V_4$	1.52	4.45	100.45

**Experimental results concerning to the yield of sweet pepper (2016)**

The results concerning the yield of pepper in 2016 are shown in Table 3. The production of sweet pepper within the experiment ranged very widely, from 55,870 kg/ha in the control version -  $V_4$ , up to 85,850 kg/ha in the fertigated version -  $V_1$ .

Table 3

**Results concerning the production of sweet pepper (2016)**

Experimental version	Total production (kg/ha)	Relative production (%)	Difference compared to the control (kg/ha)
$V_1$	85850	153.7	29980***
$V_2$	64814	116.0	8944*
$V_3$	56530	101.2	660 <sup>ns</sup>
$V_4$	55870	100.0	0

DL 5% = 4896 kg/ha; DL 1% = 15520; DL 0,01% = 22472

The difference compared to the control version, of 29980 kg/ha, is considered positive very significant. A positive significant difference was also achieved in the classic fertilized version, of respectively 8944 kg/ha.

The microorganism fertilized version -  $V_3$ , realised a non significant difference from the control version, respectively of 660 kg/ha.

## CONCLUSIONS

1. At the beginning of the growing season, the highest values of plant height were obtained in the  $V_1$  version, of 33.6 cm, and, by the end of the growing season, the biggest increases in the height of plants, of 72.8 cm, were recorded in

$V_1$  and in  $V_3$ , of 69.6. This is mainly due to phenophases uniform distribution of nutritional elements.

2. The average number of fruits per plant ranged from 4.45, in the unfertilized version  $V_4$ , till to 11.3, in the fertigated version - $V_1$ . Medium values close to  $V_1$  were realised in the classic fertilized version -  $V_2$  (9.55).

3. The weight of the *Brillant F1* sweet pepper fruit ranged from 100.45 g in  $V_4$  till to 131.81 g, in  $V_1$ .

4. The yield of sweet peppers within the experiment ranged from 55.870 kg/ha in the control version -  $V_4$ , up to 85.850 kg/ha in the fertigated version - $V_1$ , which indicates that the fertigated version made a better distribution of the fertilizers in time and space, in the root system, the distribution being performed uniformly during the growing season.

### **Acknowledgement**

*Research presented in this paper has been developed with financial support of UEFISCDI (Executive Unit for Financing Higher Education, Research, Development and Innovation) under PCCA 2013 Programme, Financial Agreement no. 158/2014.*

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## IN VITRO SCREENING OF GERMINATION CAPACITY OF SEEDS AT SOME IMPORTANT VEGETABLES GENOTYPES PRESERVED IN SEED BANKS

### REALIZAREA UNUI SCREENING IN VITRO PRIVIND CAPACITATEA GERMINATIVĂ A SEMINTELOR UNOR GENOTIPURI IMPORTANTE DE LEGUME MENȚINUTE ÎN BĂNCI DE GENE

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**Abstract.** Conservation of seeds in gene banks for long periods has the disadvantage of loss in germination capacity. Culture of seed or embryos in vitro is a method to overcome this limitation by forcing hormonal seed germination, resulting in direct production of plants. The experiments carried out in developing this paper targets to determine the optimum in vitro temperature and culture medium for the germination of seeds at different important genotypes of vegetables.

**Key words:** vitro, cultivation, tomatoes, pepper

**Rezumat.** Conservarea semințelor în bănci de gene pentru perioade lungi de timp are dezavantajul de pierdere a capacității de germinare. Cultura semințelor sau a embrionilor in vitro, este o metodă care permite depășirea acestei limitări prin forțarea germinării semințelor prin diferite metode rezultând în producerea directă a plantelor. Experimentele realizate pentru dezvoltarea acestui deziderat s-au concentrat pentru a determina valoarea optimă a temperaturii și mediului de cultură in vitro pentru germinarea semințelor la diferite genotipuri importante de legume.

**Cuvinte cheie:** vitro, cultivare, tomate, ardei.

## INTRODUCTION

The aims of applied plant science research for agriculture are to enhance crop yields, improve food quality, and preserve the environment where human beings and other organisms live. The best way for conservation of plant biodiversity and its environment, would be to achieve high crop productivity per unit area.

Genetic improvement through biotechnology needs conventional breeding because (1) the elite cultivars will be the parents of the next generation of improved genotypes, (2) field testing across locations or cropping systems and over years will be needed to determine the best selections due to the genotype-by-environment interaction.

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It is well known that conservation of seeds in gene banks for long periods has the disadvantage of loss in germination capacity. Some collected seeds although valuable due their genetic dowry, can be characterized by their low viability. The viability of the seed is affected by the conditions before harvest, the maturity, the drying conditions, as well as physical factors (temperature, humidity) during the storing (Dilday *et al.*, 1994; Blackman *et al.*, 1996). During storage, the seeds begin to deteriorate and their resistance to environmental factors during germination and growth decreases at the seedling stage (Dilday *et al.*, 1994). Culture of seed or embryos *in vitro* is a method to overcome this limitation by forcing hormonal seed germination, resulting in direct production of plants. This method is also a powerful tool to define crop evolution and gathering new knowledge. Such information should be incorporated into genetic enhancement programmes, especially those with an evolutionary breeding scheme. Likewise, plant ideotypes for each crop should drive the work of plant breeders.

Thus, the experiments carried out in developing this paper targets to determine the optimum “*in vitro*” temperature and culture medium for the germination of seeds at different important genotypes of vegetables.

## MATERIAL AND METHOD

The biological material is represented by seeds belonging to two genotypes – one hybrid and one variety, for each species (tab. 1), originated from the Vegetable Research and Development Station Bacau seed collection.

Table 1

The vegetables genotypes tested concerning their germinative capacity

Nr. crt.	Genotype	Specification
Tomatoes		
1.	Bersola F1	Hybrid
2.	Siriana	Variety
Pepper		
3.	Apollo F1	Hybrid
4.	Dariana Bac	Variety
Cabbage		
5.	Flavius F1	Hybrid
6.	CO-BCO 7-9	Inbred line

The seeds have lost partial or total germination but have particular relevance for breeders due to the stock of genes that contain it. The seeds were surface sterilised by immersion in mercuric chloride solution ( $\text{HgCl}_2$ ) 0.1% for 10 minutes, followed by repeated washing with sterile distilled water. The sterile seeds were cultivated on a basic medium Murashige Skoog, 1962 and Quoirin – Leproive, 1977, supplemented with different plant growth regulators (tab. 2).

Components of different nutrient media for in vitro seed germination

	G1	G2	G3	G4	G5	G6	G7	G8	G9	G10
Macro-elements	MS 1*	MS 2*	MS3*	QL	MS 1*	MS 2*	MS 3*	MS 1*	MS 2*	MS 3*
GA3	-	-	-	-	0.3	0.6	0.8	-	-	-
KIN								0.3	0.6	0.8

\*MS1 - MS 100% concentration, MS2 - MS 75% concentration, MS3 - MS 50% concentration.

Under aseptic conditions, seeds were inoculated on basal medium containing 3% (w/v) sucrose and 0.8% agar. The pH was adjusted to 5.8 and autoclaved at 121°C (1.06 kg/cm<sup>2</sup>) for 25 min.

The cultures were incubated in culture chambers in dark and then transferred in conditions with controlled light, humidity and temperature control at 25°C, a 16-h photoperiod, and 5000 lx light intensity. Repeated sub cultures were done at an interval of 30 days and incubated under the same temperature as mentioned previously. The culture vessels showing signs of contamination were discarded. Day to day observation was carried out to note the responses.

For each genotype, 10 seeds were cultured and the experiment was repeated three times. The data were analyzed by ANOVA (analysis of variance), the means were compared with the control (seeds germinated in pots) using the Duncan multiple comparison test at  $P < 0.05$ .

## RESULTS AND DISCUSSIONS

Unlike the other species addressed in this study, tomato seed germination is positively influenced by a concentration of 100% of the amount of macro and micronutrients (fig. 1, fig. 2). Furthermore, kinetin has been found to support the reaction rate of the seeds of the tomato. The best results were obtained on variants G6 - G10, with the point on the G6 version. Seed germination is achieved gradually over the course of 7-14 days, most seeds are germinated in the ninth day (tab. 3).



Fig. 1 Tomatoes seeds on culture media "in vitro"

Table 3

The seed germination of tomatoes genotypes tested in vitro culture media 14 days after inoculation

Nr. crt.	Variant	Genotype Charlotte F1	Genotype Dacia
1.	G1	55.70	55.90
2.	G2	53.19	52.60
3.	G3	50.30	49.90
4.	G4	30.90	35.30
5.	G5	59.70	59.40
6.	G6	59.20	58.90
7.	G7	57.90	56.80
8.	G8	57.60	58
9.	G9	56.80	57.10
10.	G10	55.30	57.20

Seed germination of pepper – fig. 2 ,was carried out over a period of 8-14 days with a peak on the tenth day. Control variant, represented by germination of seeds in conditions "ex vitro", on the moistened filter paper, recorded low levels of germination, significantly below those obtained on media culture "in vitro", 12.6% at Apollo F1 genotype and 21.03% at Dariana Bac genotype, , while cultivated "in vitro" condition, 79.10% and 79.7% respectively of seeds germinated and reached the seedling stage.



Fig. 2 Pepper seeds on culture media "in vitro"

Table 4

The seed germination of pepper genotypes tested in vitro culture media 14 days after inoculation

Nr. crt.	Variant	Genotype Apollo F1	Genotype Dariana Bac
1.	G1	79.10	79.70
2.	G2	77.20	78.00
3.	G3	76.16	77.70
4.	G4	49.20	42.90
5.	G5	46.50	45.30
6.	G6	67.80	68.60
7.	G7	63.20	59.10
8.	G8	70.20	69.90
9.	G9	71.60	70
10.	G10	70.90	79.10

The cabbage seeds of the tested genotypes germinated four days after of their inoculation on media variants tested. As the emergence of young plants, the cultures were transferred to the light, so in addition to pursuing the percentage of germination and seed germination dynamics over time was monitored.

The speed of germination of the hybrid Flavius F1 was higher compared to that of the inbred CO-BCO7-9 (see table above). The dynamics of seed germination has also increased in both genotypes, the percentage reached 68.9% in the first genotype (Flavius F1) and 65.7% in the case of the second (tab. 5).

Table 5

The seed germination of cabbage genotypes tested in vitro culture media 4 days after inoculation

Nr. crt.	Variant	Genotype FLAVIUS F1	Genotype CO-BCO7-9
1.	G1	67.70	65.50
2.	G2	65.19	62.00
3.	G3	68.90	65.70
4.	G4	35	23
5.	G5	69.50	45.30
6.	G6	57.58	43.60
7.	G7	52.60	59.10
8.	G8	50.20	58.20
9.	G9	69.60	58
10.	G10	50	58.10

## CONCLUSIONS

The experimental results demonstrate the high efficiency of "in vitro" culture in the maintenance and recovery of seed germination at tested genotypes. This is highly important, due to the fact that in gene banks, seeds can lose vitality.

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## EFFECT OF MULCH AND CULTIVAR ON YIELD OF RHUBARB

### EFFECTUL MULCIRII ȘI A CULTIVARULUI ASUPRA PRODUCȚIEI DE REVENT

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**Abstract.** *This paper presents the influence of mulch and cultivar on yield of rhubarb. The biological material used was represented by three cultivars: Glanskin's perpetual, Victoria and a local population. Were tested three mulching methods: mulching with straw, mulching with black polyethylene film of 15 $\mu$ , mulching with black polyethylene film of 30 $\mu$  and unmulched variant. Applying differential cultivation technology, rhubarb yield varies according to cultivar and mulch. The research was carried out in a rhubarb crop, in a four year production. The highest production was obtained in case of Victoria cultivar mulching with straw. Statistically assured yields were also obtained at Victoria cultivar un-mulched and Glanskin's perpetual cultivar mulching with straw.*

**Key words:** mulch, cultivars, yield, rhubarb

**Rezumat.** *Lucrarea prezintă influența mulciului și a cultivarului asupra producției de revent. Materialul biologic utilizat a fost reprezentat de trei cultivare: Glanskin's perpetual, Victoria și o populație locală. Au fost testate trei metode de mulcire: mulcire cu paie, mulcire cu folie de polietilenă neagră de 15 $\mu$ , mulcire cu folie de polietilenă neagră de 30 $\mu$  și o variantă nemulcită. Prin aplicarea diferențiată a tehnologiei de cultivare, producția de revent variază în funcție de cultivar și sistemul de mulcire. Cercetările au fost efectuate pe o cultură de revent aflată în anul 4 de producție. Cea mai ridicată producție a fost obținută la cultivarul Victoria mulcit cu paie. Producții, de asemenea, asigurate statistic au mai fost obținute și în cazul cultivarelor Victoria nemulcit și Glanskin's perpetual mulcit cu paie.*

**Cuvinte cheie:** mulci, cultivar, producție, revent

## INTRODUCTION

The rhubarb (*Rheum rhabarbarum* L.) is a less known and spread crop in Romania. It is a perennial vegetable species, adapted to cold temperate climate (Ciofu *et al.*, 2004; Indrea *et al.*, 2007).

Rhubarb is originated in the Himalayas, where its root was an important medicine believed to purge the body of ill humors (Treptow, 1985).

In our country, rhubarb is more cultivated in the western part of the country and it is used for juice, jam and other dessert (Stan *et al.*, 2003).

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In the last time, rhubarb products are spread by the supermarket on all over the country. So it is a good opportunity for farmers to cultivate this species also in other parts of the country, not only in the traditional ones (Stoleru, 2013).

For this reason, our research was focused to evaluate the possibilities to grow rhubarb in the environmental condition of the North-East part of Romania.

The mulching is a technique through which the surface between cultivated plants is covered with a thin layer of different materials, a process which clearly shows a number of features highlighted by over time through experience and practice: preventing the crust and weeds emergence, keeping moisture in the soil and allowing faster soil warming, improving air system and soil porosity, keeping clean the edible parts in contact with soil, favorably influencing production, precocity and quality (Olson, 2005).

The materials for mulching used in vegetable or other horticulture crops are divided into two main categories: organic materials (straw, hay, leaves, bark, sawdust, compost, newspaper, peat, pine needles, chips of wood, corn stalks, achene of buckwheat, peanut shells, stems of tobacco plants, evergreen trees leaves, polyethylene etc.) and inorganic materials (gravel, crushed stone, sand, volcanic ash etc.) (Dumitrescu, 2001).

To achieve this objective was carried out a study to the influence of the cultivar and mulch on the crop yield.

## MATERIAL AND METHOD

**Management of experiment.** To achieve the goal and objectives of this research work, an experimental was done at "V. Adamachi" Experimental Station of the Agronomic University.

The research was conducted on a culture of rhubarb located on four year production. Harvested area of experimental plots covered the 5 plants.

Considering the importance studying factors in the growing technology, their ability to change and taking into account the possibilities of organizing experience, it was established hierarchy of factors, as follows:

1. A factor – cultivars, with three graduations: Glanskin's perpetual (Gp), local population (Lp) and Victoria (Vt);
2. B factor – mulching system, with three graduations: mulching with straw, mulching with black polyethylene film of 15  $\mu$ , mulching with black polyethylene film of 30  $\mu$  and an unmulched version.

**Collection and processing the experimental data.** The experimental data collection was carried out observations and weight measurements, according to the experimental technique used in experiments. During 2016, were made a total of ten harvesting: 07.04, 15.04, 22.04, 29.04, 06.05, 14.05, 22.05, 29.05, 7.06 and 16.06.

The experimental variants were compared with the experimental mean, using the percentage reporting and differences. The influence of experimental factors was assessed using ANOVA. The significance of differences was assessed on the basis of LSD (least significant difference) for three degrees of confidence (95%, 99%, 99.9%).

## RESULTS AND DISCUSSIONS

Applying differential cultivation technology, rhubarb production varies according to cultivar and mulching system.

Regarding to the influence of mulch and cultivar, during 2016, it ranged from 21.23 t/ha at Gp mulching with black polyethylene film of 30  $\mu$ , to 46.08 t/ha at Vt mulching with straw (tab. 1).

*Table 1*

**Influence of mulch and cultivar at rhubarb crop**

Variants	Total yield (t/ha)	% to the average	Difference to average (t/ha)	Significance of differences
a <sub>1</sub> b <sub>1</sub>	37.80	106.57	2.33	*
a <sub>1</sub> b <sub>2</sub>	28.07	79.14	-7.40	000
a <sub>1</sub> b <sub>3</sub>	21.23	59.85	-14.24	000
a <sub>1</sub> b <sub>4</sub>	32.90	92.75	-2.57	00
a <sub>2</sub> b <sub>1</sub>	40.01	112.80	4.54	***
a <sub>2</sub> b <sub>2</sub>	27.80	78.38	-7.67	000
a <sub>2</sub> b <sub>3</sub>	32.40	91.34	-3.07	00
a <sub>2</sub> b <sub>4</sub>	40.40	113.90	4.93	***
a <sub>3</sub> b <sub>1</sub>	46.08	129.91	10.61	***
a <sub>3</sub> b <sub>2</sub>	36.55	103.04	1.08	ns
a <sub>3</sub> b <sub>3</sub>	37.48	105.67	2.01	*
a <sub>3</sub> b <sub>4</sub>	44.96	126.76	9.49	***
<b>x (Average)</b>	35.47	100.00	0.00	-

LSD 5% = 1.56 t/ha; LSD 1% = 2.34 t/ha; LSD 0.1% = 3.79 t/ha

a<sub>1</sub> – Gp; a<sub>2</sub> – Lp; a<sub>3</sub> – Vt; b<sub>1</sub> – mulching with straw; b<sub>2</sub> – mulching with black polyethylene film of 15  $\mu$ , b<sub>3</sub> – mulching with black polyethylene film of 30  $\mu$ , b<sub>4</sub> – unmulched

### **The influence of the cultivar and mulch on yield of rhubarb.**

Negative significantly differences, compared to the average have been obtained when Gp cultivar is mulching with film of black polyethylene 30  $\mu$  (21.23 t/ha), Lp mulching with black polyethylene film of 15  $\mu$  (27.80 t/ha) and Gp cultivar when was mulching with black polyethylene film of 15  $\mu$  (28.07 t/ha).

Positive differences compared to the average have been obtained when Vt cultivar was mulching with straw (46.08 t/ha), Vt cultivar unmulched (44.96 t/ha), Gp cultivar unmulched (40.4 t/ha) and Gp cultivar mulched with straw (40.01 t/ha).

The differences obtained between experimental variants and experimental mean average, ranged from -14.24 t/ha for Gp cultivar mulching with black polyethylene film of 30  $\mu$ , up to 10.61 t/ha for Vt cultivar when was mulching with straw.

The total yield in case of rhubarb crop ranged from 21.23 t/ha, for

Gp cultivar mulching with black polyethylene film of 30  $\mu$ , to 46.08 t/ha, for Vt cultivar mulching with straw.

## CONCLUSIONS

1. Regarding to the influence of the mulching system and cultivar on total yield of rhubarb, during 2016, it ranged from 21.23 t/ha for Gp cultivar mulching with black polyethylene film of 30  $\mu$ , to 46.08 t/ha, for Vt cultivar mulching with straw.

2. In case of Gp cultivar the highest production was obtained for mulching with straw (37.80 t/ha).

3. In case of Lp cultivar the highest production was obtained for un-mulched variant (40.40 t/ha).

4. Very negative differences significantly, compared to the average have been obtained when Gp cultivar is mulching with black polyethylene film of 30  $\mu$  (21.23 t/ha) and Lp mulching with black polyethylene film of 15  $\mu$  (27.80 t/ha).

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## STUDIES ON THE BEHAVIOUR OF SOME APPLE VARIETIES CULTIVATED ON THE HORTICOL FARM „V. ADAMACHI” IAȘI

### STUDII PRIVIND COMPORTAREA UNOR SOIURI DE MĂR CULTIVATE ÎN FERMA HORTICOLA „V. ADAMACHI” IAȘI

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**Abstract** *Apple is the species which occupy in temperated areas first place as volume of production and surfaces.*

*In the period 2013-2015 were performed biometric measurements on three apple varieties (Idared, Generos and Florina), planted in intensive system from horticultural farm "V. Adamachi" Iași*

**Key words:** Apple, varieties, biometrics particularities

**Rezumat.** *Mărul este specia care ocupa în climatul temperat locul întâi atât ca volum al producției cât și ca suprafețe.*

*În perioada anilor 2013 – 2015 s-au efectuat masuratori biometrice asupra a trei soiuri de măr (Idared, Generos și Forina), plantate în sistem intensiv în cadrul fermei horticole „V. Adamachi” Iași.*

**Cuvinte cheie:** Măr, soiuri, particularități biometrice

## INTRODUCTION

Romania shows favorable natural conditions for apple culture. The culture is optimal to the Carpathian hills, in the area of beech and oak forests (Istrate, 2007).

Iasi County shows as a limitation for apple crop, the rainfall witch is lower in summer period for species requirements. However, apple production potential in the area offering a reasonable value, witch covers the investment during the year (Aiacoboaie *et al.*, 1994).

## MATERIAL AND METHOD

Experience was conducted in an orchard of apple set up in horticultural farm "V. Adamachi" Iași. The soil is a cernoziom loeossoid cambic clay formed on a terrace.

The average annual temperature in Iasi during the experimental years is 11.2°C, and the average annual rainfall is 520.8 mm.

Studied varieties (Idared, Generos and Florina) were grafted on rootstock MM106, trees being driven form of free palmettes, flattened by pruning.

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Planting distance was 4 x 3 m returning about 833 trees per hectare. The land was kept as black field by repeated manual and mechanical works

Each variety of the experience was a variant of experience, and within each variant were performed five repetitions (Grădinaru *et al.*, 1996).

## RESULTS AND DISCUSSIONS

Analyzing the height of trees (tab. 1) finds that the highest value is achieved during the experience at variety Florina with 4.20 m in 2013 and 4.12 m respectively in 2015.

Generos variety in 2013 had an average height of 3.61 m, 3.73 m and 3.85m in years 2014, 2015.

The control, variety Idared, gained 3.17 m in 2013, 3.14m in 2014 and 3.25m in 2015.

Table 1

The height of trees (m)

Variant name	The height of trees (m)		
	2013	2014	2015
<b>V1 Generos</b>	3.61**	3.73**	3.85**
<b>V2 Idared (control)</b>	3.17	3.14	3.25
<b>V4 Florina</b>	4.20***	4.05***	4.12***
LSD 5%	0.10	0.11	0.11
LSD 1%	0.19	0.21	0.19
LSD 0.1%	0.24	0.43	0.28

From table 2 it can be seen that the largest cross sectional area of the trunk was obtained from Florina variety, it recorded values between 198.2 - 201.5 cm<sup>2</sup>, followed by Generos variety with 146.6 to 149.8 cm<sup>2</sup> and witness, the variety Idared who recorded at this item 88.7 - 91.5 cm<sup>2</sup> during the experiment.

Tabelul 2

Cross-sectional area of the trunk (cm<sup>2</sup>)

Variant name	Cross-sectional area of the trunk (cm <sup>2</sup> )		
	2013	2014	2015
<b>V1 Generos</b>	146.6	148.0	149.8
<b>V2 Idared (control)</b>	88.8	89.7	91.5
<b>V3 Florina</b>	198.2	199.8	201.5
LSD 5%	38.3	36.5	37.3
LSD 1%	58.0	55.3	56.5
LSD 0.1%	93.2	88.8	90.8

Tree crown volume oscillated between 9.3 - 9.9 m<sup>3</sup> at variety Idared, followed by Generos variety with 12.0 – 12.4 m<sup>3</sup> in 2014 and 2013 respectively.

Florina variety recorded a crown volume variation limits between 14.3m<sup>3</sup> in 2013 and 2014 and 14.5 m<sup>3</sup> in year 2015 (tab.3).

Table 3

The crown volume (m<sup>3</sup>)

Variant name	The crown volume (m <sup>3</sup> )		
	2013	2014	2015
<b>V1 Generos</b>	12.4	12.0	12.2
<b>V2 Idared (control)</b>	9.3	9.4	9.9
<b>V3 Florina</b>	14.3***	14.3***	14.5***
LSD 5%	0.31	0.12	0.07
LSD 1%	0.52	0.18	0.13
LSD 0.1%	0.86	0.27	0.24

Table 4

## The production of fruit

Variety	Production of fruit (kg/tree)	Production of fruit (t/ha)
<b>V1 Generos</b>	38.41	32.00
<b>V2 Idared (control)</b>	36.8	30.65
<b>V3 Florina</b>	42.85	35.70

The cumulated fruit production during the experiment is analyzed in table 4. For Generos variety the average production in all three years was 38.41 kg/tree, respectively 32 t/ha. At V3 – Florina variety average production / tree was 42.85 kg/tree (35.7 t/ha). The smallest production is obtained at control V2 – Idared variety with 36.8 kg/tree (30.65 t/ha).

## CONCLUSIONS

Making annual technological interventions have the effect of maintaining tree crown in the designed parameters.

In some years more severe interventions are needed in the trees crown that fructify type III - Generos and Idared or type IV - Florina.

These interventions has as short-term effect a temporary reduction in the size of the crown, but for long term, these engineering works renew production capacity of trees, regenerating specific fructifying elements.

Fruit productions obtained are satisfactory considering the fact that rainfalls in the Iasi area are deficient for the apple species.

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## AGROBIOLOGIC POTENTIAL EVALUATION OF SOME NEW SWEET CHERRY VARIETIES IN ORDER TO IMPROVE UPON THE ROMANIAN ASSORTMENT

### EVALUAREA POTENȚIALULUI AGROBIOLOGIC AL UNOR SOIURI NOI DE CIREȘ ÎN SCOPUL AMELIORĂRII SORTIMENTULUI DIN ROMANIA

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**Abstract.** *In Romania, Iasi area is one of the most favorable for growing cherries. Regarding the zonal assortment, it suffered constant renewal through continuous selection of local biotypes valuable and not least by introducing new varieties and rootstocks of cherry trees from abroad. The experiment was organized as an intensive cherry plantation. Biological material used consisted of five new foreign cherry varieties: 'Kordia', 'Karina', 'Regina', 'Summit', 'Sunburst', grafted on 'Gisella', vegetative rootstock. Cherry plantation was established in 2010 with planting distances of 4 x 2 m, resulting in a density of 1250 trees/ha. As a form of leadership trees crown was chosen Leader of the Central Vogel (VCL). New cherry varieties studied were noted by productivity and fruit quality: 'Kordia' 12.1 t/ha in the fourth year after planting (10.3 g fruit average weight), 'Regina' 10.2 t/ha, (10.6 g), 'Karina' 10.2 t/ha (9.7 g) and 'Summit' 10.0 t/ha (9.8 g)*

**Key words:** new varieties, sweet cherry, assortment, cultivation area

**Rezumat.** *În România, zona Iași, este una dintre cele mai favorabile pentru cultura cireșului. În ceea ce privește sortimentul zonal, acesta a suferit o permanentă înnoire, prin selecția continuă de biotipuri locale valoroase și nu în ultimul rând prin introducerea de noi soiuri și portaltoi din străinătate. Experiența a fost organizată într-o plantație intensivă de cireș. Materialul biologic folosit cinci soiuri noi de cireș străine: Kordia, Karina, Regina, Summit, Sunburst, altoite pe portaltoiul vegetativ Gisella 5. Plantația de cireș a fost înființată în anul 2010, cu distanțele de plantare de 4 x 2 m, rezultând o densitate de 1250 pomi/ha. Ca formă de conducere a coroanei pomilor s-a optat pentru Liderul Central Vogel (VCL). Dintre soiurile noi de cireș luate în studiu s-au remarcat prin productivitate și calitatea fructelor: Kordia 12,1 t/ha în anul VI de la plantare (10,3 g greutatea medie a fructului), Regina 10,2 t/ha, (10,6 g), Karina 10,2 t/ha, (9,7 g) și Summit 10,0 t/ha, (9,8 g)*

**Cuvinte cheie:** soiuri noi, sortiment, cireș, zona de cultură

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## INTRODUCTION

Since the agricultural variety is the main mean of production, increasing the quantity and quality of the finished product is done by changing the variety and application upon him of the most suitable crop technologies that enhance the genetic potential and on the other hand to ensure the conservation and protection of natural ecological framework, namely the soil and environment. Each fruit-growing country cultivates its own range and it mainly consists of varieties well adapted to its own ecopedological conditions, fruit tree assortment adequate to specific growing areas.

Certainly, the most notable news obtained from breeders in Europe are early ripening varieties 'Primulat' and 'Early Bigi' (France), self-fertile 'Sweet Early' and 'Grace Star' (Italy), middle ripening 'Giorgia' (Italy) and 'Vera' (Hungary), medium-late 'Kordia', 'Vanda' and 'Techlovan' (Czech Republic) and 'Black Star' (Italy) and late ripening varieties 'Regina' (Denmark) and 'Alex' (Hungary) (Sansavini and Lugli, 2005).

In Romania, there is intense work on creating new varieties of trees, till present, corresponding to the general objectives such as different period of fruits ripening, comparing to the existing varieties, reduced vigor, self-fertility, high environmental adaptability, disease resistance, etc. In terms of assortment, it suffered constant renewal through continuous selection of local biotypes valuable and not least by introducing new varieties from abroad and new Romanian creations, obtained at Bistrita, Pitești and Iasi research stations (Balaci *et al.*, 2007; Budan *et al.*, 2007; Istrate *et al.*, 2008; Petre *et al.*, 2007).

The current study was conducted during 2015-2016 observing the production behavior of some new varieties of cherry grafted on Gisella 5 rootstock, in the climatic conditions of Iasi County.

## MATERIAL AND METHOD

The experiment was organized as an intensive cherry plantation. Biological material consisted of five new foreign cherry varieties: 'Kordia', 'Karina', 'Queen', 'Summit', 'Sunburst', grafted on 'Gisella 5' vegetative rootstock.

Sweet cherry plantation was established in 2010 with planting distances of 4x2m, resulting in a density of 1250 trees/ha.

As a form of leadership trees crown was chosen Leader of the Central Vogel (VCL).

The soil maintenance method was grassing, combined with mechanized work of intervals between rows.

The orchard is provided with fertigation system and it is also protected with hail cannon.

During the growing season there are applied specific maintenance works. Pest and disease control is performed by applying 6 to 8 phytosanitary treatments.

## RESULTS AND DISCUSSIONS

The main factors of the trees vigor evaluation were the growth of trunk sectional area and crown volume. Sectional area of the trunk at the studied cherry varieties, in the VI-th year after planting, recorded values between 34.19 cm<sup>2</sup> ('Karina') and 48.99 cm<sup>2</sup> ('Regina') (tab. 1). The highest increase growth in 2015 - 2016 period was registered by 'Regina' variety (14.8 cm<sup>2</sup>), followed by 'Sunburst' (11.31 cm<sup>2</sup>) and 'Karina' (11.31 cm<sup>2</sup>) that, evaluated as statistically significant differences compared with the control.

The smallest increase growth in the same period was recorded by 'Kordia' variety (7.91 cm<sup>2</sup>), with significant negative differences.

'Regina' variety (335.4 cm) recorded the highest values of trees height, followed by 'Kordia' (311.9 cm) and 'Summit' (309.0 cm) (tab. 1).

The longest annual increases (over 60 cm) were registered by 'Regina' and 'Kordia' varieties.

Table 1

**Trunk section area, tree height and length of annual increases at some new cherry varieties**

Variety	Trunk section area (cm <sup>2</sup> )		Tree height (cm)		Length of annual increases (cm)	
	2015	2016	2015	2016	2015	2016
Regina	34.19	48.99	311.6	335.4	38.4	62.4
Kordia	27.32	35.23	294.8	311.9	27.6	60.3
Karina	24.61	34.19	291.6	308.4	18.3	45.6
Summit	30.17	39.57	287.6	309.0	22.1	47.5
Sunburst	28.26	39.57	278.6	295.5	31.8	57.8

Table 2

**Tree vigor and crown volume at some new cherry varieties**

Variety	Tree vigor	Crown volume		
		D (m)	D (m)	H (m)
Regina	Medium	2.35	2.49	3.22
Kordia	Medium	2.48	2.69	2.96
Karina	Little-medium	2.19	1.95	2.94
Summit	Medium	2.59	2.67	2.81
Sunburst	Little-medium	2.11	1.98	2.91

Knowing the biological particularities of the cherry varieties and hybrids, allows us to adopt differentiated culture technologies according to: the ecological area, trees vigor and fructification type, crown shape and planting distance.

In the sixth year after planting, 'Kordia' and 'Summit' varieties achieved the highest values of the crown volume 8.21 m<sup>3</sup>/tree (10261 m<sup>3</sup>/ha) and 8.08m<sup>3</sup>/tree (10100 m<sup>3</sup>/ha) (tab. 3).

The lowest crown volume was obtained by 'Sunburst' (5.06 m<sup>3</sup>/tree) and 'Karina' (5.0 m<sup>3</sup>/tree) varieties. Reduced distance between trees per row (2 m) is fully covered by trees crown at the most varieties, achieving a continuous orchard fence.

Table 3

**Crown volume and fruit formations of some new cherry varieties**

Variety	Fruit formations	Crown volume	
		m <sup>3</sup> /tree	m <sup>3</sup> /ha
Regina	branch bouquet	7.84	9800
Kordia	branch bouquet	8.21	10261
Karina	branch bouquet	5.0	6250
Summit	branch bouquet	8.08	10100
Sunburst	branch bouquet	5.06	6325



**Fig. 1** Crown Vogel Central Leader (VLC)

<http://pmmg.org.ge/res/uploads/%E1%83%AE%E1%83%94.jpg>

Leading trees as Central Leader Vogel form was made as a result of trees scaffold leading and semi permanent branches trellising on the direction of the row (fig. 1).

Details on the way of fructification in cherry emphasize that all varieties predominantly fructify on May bouquet (tab. 3).

Fructification phenophases are specific to each specie biology and the starting date and duration are related to the climatic conditions of each year (tab. 4).

Data on fructification phenophases of some new cherry varieties

Variety	Early flowering	End of flowering	Flowering duration -days-	Fruit maturation	Nr. of days from flowering to harvest end
Regina	9-11.IV	17-20.IV	8-9	21-25.VI	65
Kordia	31 III- 4.IV	7-13.IV	8-9	6-10.VI	58
Karina	6-8.IV	14-17.IV	8-9	15-18. VI	61
Summit	2-6.IV	9-14.IV	7-8	9-12. VI	58
Sunburst	1-5.IV	7-13.IV	7-8	8-11. VI	57

The beginning of flowering at studied cherry cultivars during 2015-2016 was between 31<sup>st</sup> of March and 8<sup>th</sup> of April, and the end of blooming was between 7<sup>th</sup> and 20<sup>th</sup> of April (tab. 4). The earliest flowering was registered in 2015, and the latest in 2016.

Among the studied cherry varieties, 'Kordia' and 'Sunburst' showed an early flowering. 'Regina' and 'Karina' varieties showed blooming tardiness.

Flowering overlapped in all cherry varieties, providing favorable conditions for cross pollination.

For a proper pollination and fertilization process, are preferred moderate temperatures 15 - 17°C, which maintain the viability of the stigma and the pollen for a longer period of time and allow the flight of bees.

Fruitfulness precocity is a complex genetic trait that manifests itself by variety and has a practical significance, being a key criterion in the choice of varieties of cherry plantations and current super-intensive type.

Fruit production obtained depends on the fertility of variety, the interaction between variety, biotope conditions and applied crop technology. As the biotope conditions are closer to the biological requirements of the variety, the production level increases.

The average yields for the years V-th and VI-th after planting, at studied cherry varieties, were between 9.2 t/ha ('Sunburst') and 12.1 t/ha ('Kordia') (tab. 5).

Table 5

Fruit production obtained at some new cherry varieties

Variety	The production of fruit obtained during 2015-2016		Fruit average weight (g)	S.U. (%)	Fruits skin color
	Kg/tree	t/ha			
Regina	7.6-8.1	9.5-10.2	10.6	15.8	Brown-red
Kordia	8.9-9.6	11.2-12.1	10.3	14.4	Brown-red
Karina	7.7-8.1	9.7-10.2	9.5	14.9	Brown-red
Summit	7.8-8.0	9.8-10.0	9.2	15.3	Dark red
Sunburst	7.3-7.9	9.2-9.9	9.4	16.2	Dark red

In terms of productivity, it was noticed 'Kordia' variety (11.5 t/ha) by 2-year average yields.

Trees leadership as Vogel Central Leader in cherry has good results at medium vigor varieties that have reduced capacity of branching.

Fruit size is a genetic trait of the variety, which can manifest differences at the same variety, depending on the climatic conditions, production quantity, soil fertility etc.

Regarding fruit size, among the studied cherry varieties, were noted 'Regina' (10.6 g fruit average weight) and 'Kordia' (10.3 g) (tab. 5).

The chemical composition of the fruit was determined in each year of the experiment, at the most varieties, focusing on the elements that are most important for cherries (tab. 5).

Highest soluble solids content was recorded at 'Sunburst' (16.2%) and 'Regina' varieties (15.8%).

## CONCLUSIONS

- Climatic conditions of Serbesti area, Iasi County are favorable for growing cherry and may be enhanced by providing crop irrigation.
- Tree crown volume is very much influenced by the shape of the crown taken in conjunction with the varieties fructification type; planting distances and agricultural technique there were applied.
- 'Kordia', 'Summit' and 'Regina' cherry varieties, registered, in terms of planting distances of 4 x 2 m, a growth in volume, VI-th year after planting, of over 9800 cubic meters/ha, which explains the increased production obtained.
- In terms of productivity, stood by the average yields of 2 consecutive years (V-th and VI-th year after planting) the varieties: 'Kordia' (11.5 t/ha), 'Regina' (9.8 t/ha) and 'Karina' (9.7 t/ha).

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## RESEARCH ON MICROSPOROGENESIS AND THE DEVELOPMENT OF MICROSPORES IN SOME ROMANIAN VARIETIES OF PEACHES AND NECTARINES FRUIT TREES

### CERCETĂRI PRIVIND MICROSPOROGENEZA ȘI EVOLUȚIA MICROSPORILOR LA UNELE SOIURI ROMANEȘTI DE PIERSIC ȘI NECTARIN

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**Abstract.** *In this study is investigated microsporogenesis and evolution of young microspores both three varieties of Romanian peach Amalia, Congress and Splendid and two Romanian varieties of nectarine Tina and Michaela. Within these species it was considered that at the choice of the varieties to be included the all three ripening periods of the fruits (early, middle and late). The aim of the research consists in the knowledge of the maturing microspores steps at peaches and nectarines, regarding the development of sporoderma by reaching the final dimensions beginning from young microspores, to mature pollen. Also we had in view the selecting of peaches and nectarines varieties with resistance to very low temperatures in winter. It was studied the effect/impact of negative temperatures in winter on the successive stages of maturation microspores starting with CMP (pollen mother cells) to binucleat mature pollen of flowering buds. From the biological perspective the microsporogenesis was not disturbed by external factors and tetrad and young microspores had a normal aspect for the development phase. The pollen maturation went under normal physiological and undisturbed conditions in the line with the changing weather.*

**Key words:** pollen, young microspores, external factors

**Rezumat.** *În prezenta lucrare se cercetează microsporogeneza și evoluția microsporilor tineri, atât la trei soiuri românești de piersic Amalia, Congres și Splendid, cât și la două soiuri românești de nectarin Tina și Mihaela. În cadrul acestor specii s-a avut în vedere ca la alegerea soiurilor să fie cuprinse toate cele trei perioade de coacere (timpurie, medie și tardive). Scopul cercetării constă în cunoașterea etapelor de maturare a microsporilor la piersic și nectarin, privind evoluția sporodermei, prin atingerea dimensiunilor finale, începând cu microsporii tineri până la polenul matur, de asemenea cât și selectarea soiurilor de piersic și nectarin cu rezistența la temperaturile scăzute din iarna. S-a studiat microscopic efectul /impactul temperaturilor negative din iarna asupra stadiilor și etapelor succesive de maturare ale microsporilor începând cu CMP (celulele mama polinice) până la inclusiv polenul matur binucleat din mugurii floriferi. Din punct de vedere biologic, microsporogeneza nu a fost perturbată de factorii externi iar tetradele și microsporii tineri au avut aspect normal pentru etapa de dezvoltare. Maturarea polenului a decurs în*

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*condiții fiziologice normale și neperturbate în concordanță cu evoluția condițiilor meteorologice.*

**Cuvinte cheie:** pollen, microspori tineri, factori externi

## INTRODUCTION

The developments of the microsporogenesis biological processes are dependent both genetic and the thermic factors. All the microsporogenesis steps to the pollination represent "critical phenophases" in comparison with to resistance to much lower temperatures below 0 Celsius degrees (Balan, 2008). These stages mark the transition from the winter resting buds to the optional (Bordeianu, 1961; Tarnavski, 1963). Both the tetrad stage, the uninucleate microspore and the pollen stage binucleat, all these stages depend on "the necessary cold" that the buds compulsory sprout during the optional quiescence (Baciu, 1971). The period from December to January is very important for fruit trees, because the low temperatures creates by the continuity of their flowering buds, an accumulation of cca.200 hours of cold with temperatures below  $-7^{\circ}\text{C}$  (Ivascu, 2001). The winter stability protects flowering buds (Cociu, 1993). Both unfavorable and harmful weather conditions and the pronounced fluctuations of low/very low temperature in winter correlated with increased sensitivity of the flowering buds to extremes of temperature, can usually produce profound physiological disturbance in the microsporogenesis at some fruit tree varieties. All these can also cause disparities in the steps of the sporoderma pollen stratification, that affects the further development of the pollen tube, which ultimately it manifests in the orchard, through early fall fruit in large proportion. Also in other cases it manifests by disrupting physiological of fertilization (fertility and then through the weakly fruit binding (Iordache, 2010). The objective of this research is to select the peach and nectarine sorts and the resistance at very low temperatures in winter.

## MATERIAL AND METHOD

They were evaluated 5 (experiments) - Romanian fruit tree varieties with different ripening periods (early, middle and late): - for peaches it was analyzed three varieties Amalia, Congress and Splendid, and for nectarine varieties they were evaluated two Tina and Michaela varieties. Suitable to microsporogenesis and maturation stages of pollen in the SCDP Baneasa, they were weekly collected in the months (February, March, April 2011 ), flowering shoots in stages bud swell, bud burst, calyx red, white bud first/full bloom. The fastening of the taken flowering buds, was made in a Carnoy solution and ethanol 70% vol., after the method for the preserving buds (Andrei and Paraschivoiu, 2003). The microspores were released at each sample, in part by cutting the anthers for observing the early stages of microsporogenesis process (microspore mother cells, dyads, tetrads, monads in various stages and mature pollen). For microscopic examination was used optical microscope IOR ML4-M type. The examination was done in transmitted light and phase contrast with ocular 10x and objectives 10x20x40x.

## RESULTS AND DISCUSSIONS

The peaches flowering buds and nectarines sampled in from the second decade of February (21.02.2011) period: by their cutting were obtained very young anthers with normal looking and consistency, as a result of undisturbed organogenesis by climatic factors. There were not tetrades in the anthers because in this time the microsporogenesis process was not relased (started). In the first decade of March at flowering buds of nectarine, they were revealed microspore mother cells and tetrade in training (fig. 1). The nectarine flowering buds sampled period of 21 March 2011: they were already found tetrade very mature 21 March 2011, at flowering peach buds were highlighted microspore mother cells and tetrade very young (fig. 3). At the random sampling of the flowering buds in the period 25 March 2011, observed that Amalia peach varieties and Splendid had very mature tetrades and at Congress variety, were also found buds wich contained young tetrades (fig. 4).

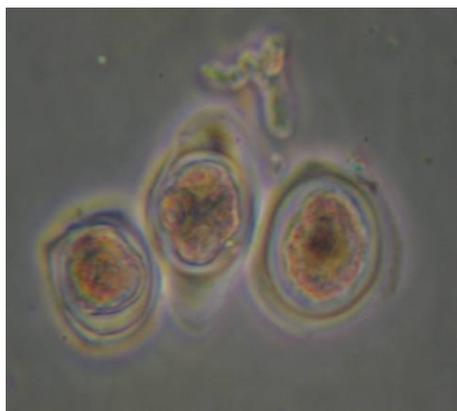
At the flowering nectarines buds were sampled in 25 March 2011 period, it is noted that in the sporoderma level begin to differentiate fine ornamentations and the shaping of apertures (fig. 5). It should be noted that in the terms of climate, the winter period December 2010 -January -February 2011 (fig. 6), was relatively mild with negative temperatures, but not excessive, that create harmful differences so as to disrupt the natural rhythm adaptation of the flowering buds. So the average temperature in December 2010 was (-0.4 ° C) and the annual average (-0.1°C).

The average temperature in January 2011 was (-2°C) and the annual average (-3.1°C). The average temperature in February 2011 was (-1.9°C) and the annual average (-0.7°C). In the last decade of January, the minimum temperature dropped from (- 10°C) to (-14°C) and continued in the first decade of February with T°C minimum at (-17°C) to (-8°C).

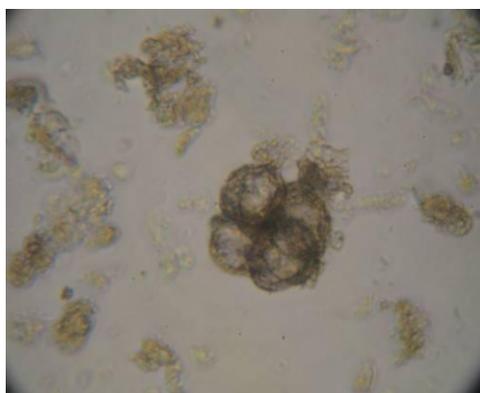
This prolonged moderate interval of cold, had favorable consequences, because by its thermal values did not present a risk of harm (because it has not reached -20°C). On the other hand, the uninterrupted cold protected by fading (decălire) the flowering buds and the risk of accelerated preparation for entrance into vegetation; on the other hand the uninterrupted cold maintained the cellular respiration at a low level as reserves and consumption of starch. In this period was actually created a stabilization of the winter that protect of the flowering buds. March and April 2011 have been positive temperatures with average (5°C) and (+10°C). In conclusion, from the examination of microscopic preparations obtained from samples of flowering buds, from the period 21.02.2011-18.04.2011, results that the stages of microsporogenesis process was held at peach , during/in period 21.03.-18.04.2011 and at nectarin during/in period 08.03.-13.04.2011, so:

1. **At peach** tetrad stage was held from 21.03.- 27.03.2011;
2. The very young microspores stage and undergoing maturation during 27.03-07.04.2011;

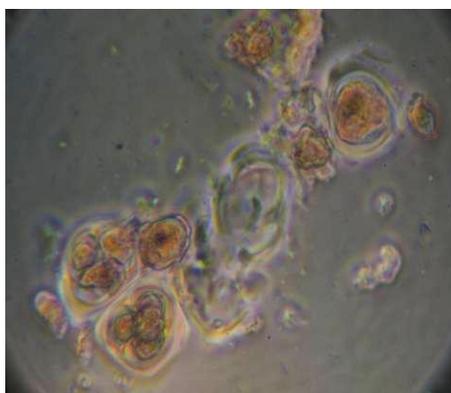
3. The mature microspores appeared starting with approx 10 days before anthesis;
4. The flowering peach this year started on 18.04.2011;
5. The status of tetrad **at nectarine** was held from 08.03. - 21.03.2011;
6. The stage of very young microspores and undergoing maturation during 21.04. - 01.04.2011;
7. The mature microspores appear starting from 01.04. - 12.04.2011 with approx. 10 days prior to anthesis;
8. The flowering of the nectarine this year started on 13.04.2011 with 5 days before peach.



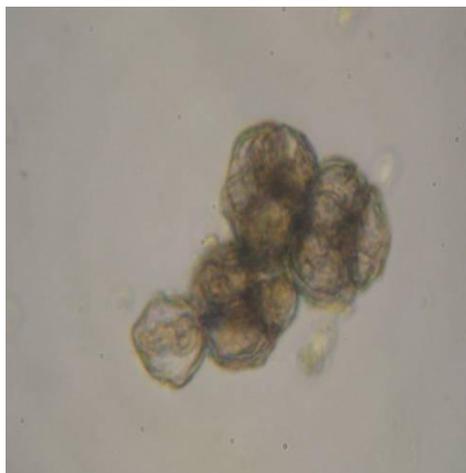
**Fig. 1** The Tina and Michaela nectarine varieties, microspore mother cells and tetrads in training (08. 03. 2011) in contrast phase and transmitted light.



**Fig. 2** The nectarine varieties Michaela, monads in detachment, ob.10x, 20x (21.03 2011)



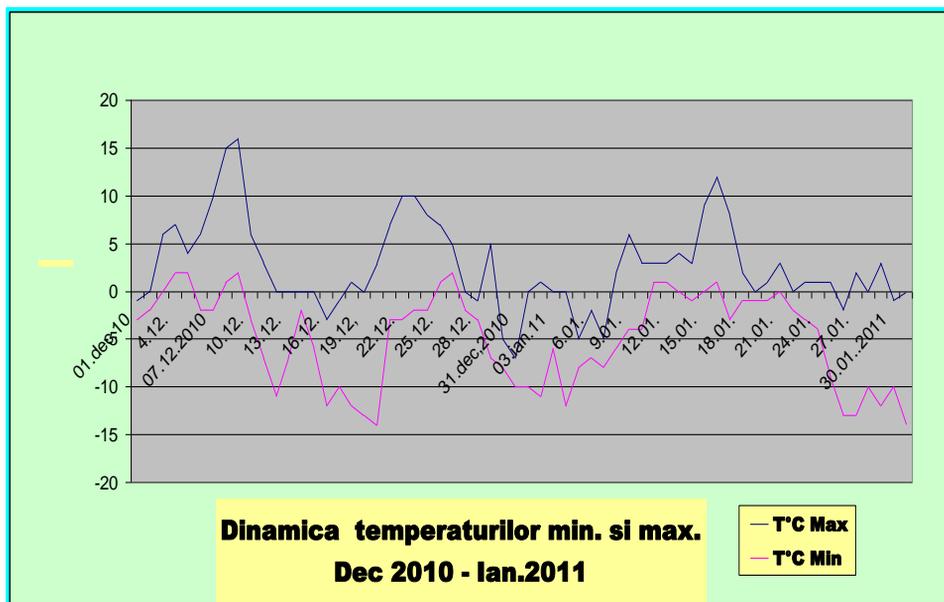
**Fig. 3** The peach varieties, Congress and pollen mother cells and very young tetrads in very young in contrast phases, ob.20x, 40x (21.03.2011)



**Fig. 4** Tetrades very mature at peach varieties Amalia (25.03.2011)



**Fig. 5** Variety nectarine Tina ob.20x, transmitted light (25.03.2011)



**Fig. 6** The dynamic of the minimum and maximum temperatures for the December 2010 and January 2011

## CONCLUSIONS

As a result the carried out researches in February-April 2011, in which has been evaluated the microsporogenesis and the young microspores pollen development at the 5 Romanian varieties of peach and nectarine we come to the conclusion that:

1. From the point of biological view, the microsporogenesis had a normal evolution undisturbed and unaffected by external factors;
2. Tetrades and microspores had normal aspect for the development phase;
3. The pollen maturation went in normal and undisturbed conditions, in accordance with the evolution of the favorable weather condition;
4. Both peach and nectarine have the advantage that their microsporogenesis process starts much later (approx. 13 days later than the other species of fruit trees such as apricots for example) and therefore the possibility that microsporogenesis steps may be disrupted due to adverse weather conditions is much lower at peach and nectarines.

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## CORALIS - A NEW SWEET CHERRY CULTIVAR FOR COMMERCIAL PLANTATIONS

### CORALIS – UN NOU SOI DE CIREȘ PENTRU PLANTAȚIILE COMERCIALE

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**Abstract:** *The aim of the paper is to improve the autochthonous sweet cherry assortment with new cultivars with late maturation and high quality fruits. The sweet cherry assortment from Romania contains many cultivars with maturation in the medium season. The current tendency is to create a balance by reducing the number of the cultivars with medium season maturation and by increasing the number of the cultivars with very early and late season maturation. Analyzing the main phenological stages for the two cultivars it was noticed that the new sweet cherry cultivar 'Coralis' is late both for the beginning of flowering time and for fruits maturation. Regarding the average productions on four years (2011-2014) from the statistical point of view, 'Coralis' cultivar (20.0 kg/tree) recorded positive production differences compared to the control cultivar (19.8 kg/tree). Under the aspect of fruits weight and equatorial diameter 'Coralis' (9.1 g and 24.7 mm) recorded significant differences and distinct positive significant differences compared to the control cultivar (6.9 g and 21.8 mm).*

**Key words:** assortment, sweet cherry, cultivar, late ripening, fruit production

**Rezumat:** *Scopul lucrării este de a îmbunătăți sortimentul de cireș autohton cu soiuri de cireș cu maturare târzie, cu fructe de calitate. Sortimentul de cireș din România, cuprinde o abundență de soiuri cu maturare medie a fructelor. Tendința actuală este de a realiza un echilibru, prin reducerea ponderii soiurilor cu epocă mijlocie de maturare a fructelor și a sporirii ponderii soiurilor extratimpurii și târzii. Analizând principalele stadii fenologice la cele două soiuri, s-a constatat că noul soi de cireș 'Coralis' este tardiv atât în ceea ce privește începutul înfloritului cât și cel al maturării fructelor. Referitor la producțiile medii pe patru ani (2011-2014), din punct de vedere statistic, soiul de cireș 'Coralis' (20.0 kg/pom) a înregistrat diferențe de producție pozitive față de soiul martor (19.8 kg/pom). Sub aspectul greutatei fructelor și a diametrului ecuatorial, 'Coralis' (9.1 g și 24.7 mm) a înregistrat diferențe semnificative (g) și distinct semnificative pozitiv (mm) față de soiul martor (6.9 g și 21.8 mm).*

**Cuvinte cheie:** sortiment, cireș, soi, maturare târzie, producție de fructe

## INTRODUCTION

The obtaining of new cultivars with improved features and characteristics is a permanent requirement both from the growers and from the fruits consumers side. This is also the main objective of the genetic breeding and

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controlled or natural selection which have been happening for centuries (Branște *et al.*, 2007).

Regarding the sweet cherry assortment accepted for the commercial trees nursery in Romania, it contains both autochthonous and foreign cultivars, verified through studies and long time observations in the regional research stations whose results are validated by National Institute for Variety Testing and Registration Bucharest and Ministry of Agriculture and Rural Development - Romania.

From the maturation and fresh fruits consumption point of view, all these cultivars cover a long period (46-50 days) between May (for very early cultivars) to July (for the very late cultivars) (Petre, 2006; Iurea, 2015).

The research concerning the assortment improvement for the sweet cherry tree, the cultivars quality and the avoidance of market overcrowding with fresh cherries on short term are important objectives. The tendency is to create a balance, assured by reducing the number of the cultivars with maturation in the medium season of sweet cherry and by increasing the number of the very early and late sweet cherry cultivars (Budan and Grădinariu, 2000). Therefore in March 2016 a new cherry cultivar obtained at RSFG Iași, was registered under the name '*Coralis*'.

The aim of the paper is to improve the autochthonous sweet cherry assortment with new cultivars with late maturation and high quality of the fruits.

## MATERIAL AND METHOD

The studies have been done during 2011-2014, on the sweet cherry cultivar '*Coralis*' (HC 885302) and the comparison has been done with the regional control cultivar '*Boambe de Cotnari*'.

The fruit-growing trees can be found in the experimental plots, grafted on mahaleb and planted at a distance of 5 x 4 m, with free flattened palmette crown shape on the direction of the trees row, without a sustaining system or irrigation system. On the row with trees, the cultivar was worked with the lateral disk with feeler and between the trees rows the soil was heated. Diseases and pests control was done applying phytosanitary treatments.

In the experimental field there have been done observations and determinations concerning the trees vigour, the resistance to anthracnose and monilia and the main phenological stages (Cociu and Oprea, 1989).

**The physical characteristics of the fruit** were determined this way:

- the fruit/stone weight (g) by weighting 10 fruits/stones in three repetitions with the electronic scale (Radwag, sensitivity 0,01g);
- the fruit/stone dimensions (mm) were determined with the digital calliper Luumytools for 10 fruits/stones in three repetitions; the fruit's equatorial diameter (D) and stone/fruit ratio;
- the fruit's colour, the pulp firmness and stone adherence to pulp was determined in accordance to the UPOV TG/35/7 questionnaire.

**The chemical and quality characteristics** of the fruits were determined this way:

- the soluble dry substance was determined refractometric, using a hand refractometer Zeiss;

- the resistance of the fruits to cracking was determined by immersion 100 fruits from each cultivar in distilled water and after 6 hours it was verified the number of cracked fruits, determining this way the percentage of fruit's cracking per cultivar (Cociu and Oprea, 1989).

- the productivity was determined by weighting the fruits per tree, per variants and per repetitions.

The experimental data was statistically interpreted by analysing the variance.

## RESULTS AND DISCUSSIONS

The two studied genotypes are of middle tree vigour. Concerning the diseases resistance, the year 2013 was a rainy year (in the first 6 months of the year there have been accumulated 446.5 mm of precipitations), that being very favourable for the evolution of the pathogens, therefore the sweet cultivars taken to the study manifested a low sensitivity both to anthracnose (attack frequency was between 3.1 to 3.8%) and to monilia (attack frequency was between 2.0 to 2.1 %) (tab. 1).

Table 1

The characteristics of the tree for two sweet cherry cultivars (RSFG Iași; average 2011-2014)

Genotype	Tree vigour*	Resistance to:					
		Anthracnose** (on leaves) ( <i>Coccomyces hiemalis</i> Higg.)			Monilia** (on fruits) ( <i>Monilinia fructigena</i> )		
		F%**	I%	A.D. %	F%	I%**	A.D.%
Coralis (HC 885302)	5	3.1	5	0.06	2.0	5	0.04
Boambe de Cotnari (control)	5	3.8	5	0.08	2.1	5	0.04

\*- tree's vigour degree on a scale between 1 to 9: 1= very weak; 3= weak; 5 = medium; 7= strong; 9= very strong (\*\*\*, 2006).

\*\*F% - attack frequency (%); I% - intensity degree on a scale between 1 to 6: 1 = 3% attacked surface; 3 = 25%; 4 = 50%; 6 = 100%; A.D. % - attack degree (Cociu and Oprea, 1989).

During 2011-2014, the flowering beginning for the 'Coralis' cultivar was recorded between 10<sup>th</sup> and 25<sup>th</sup> of April and for the control cultivar 'Boambe de Cotnari' it was recorded between 8<sup>th</sup> and 21<sup>st</sup> of April. Analysing the phenological stages for these two cultivars it was noticed that the new sweet cherry cultivar 'Coralis' blooms time was a couple days later than the control cultivar 'Boambe de Cotnari' (tab. 2). The late flowering time presents a great importance to avoid the damage caused by late spring frost (Milatović *et al*, 2011).

Regarding the harvesting maturity this oscillates in large limits each year. Thus, it can be noticed that '*Coralis*' were ripening time in the 3<sup>rd</sup> 10-days of June or the first 10-days of July, a week later than '*Boambe de Cotnari*' (tab. 2).

Regardless to all the climatic conditions of the year, the order in which the sweet cherry cultivars reach the ripening time is always the same, the difference being that the time period between two successive cultivars which is larger or shorter.

Table 2

**The main phenological stages for two sweet cherry cultivars  
(RSFG Iași; 2011-2014)**

Phenological stages	Year	Genotype	
		<i>Coralis</i> (HC. 885302)	<i>Boambe de Cotnari</i> (control)
Flowering beginning data	2011	25.04	20.04
	2012	21.04	15.04
	2013	24.04	21.04
	2014	10.04	8.04
	<b>Average</b>	<b>20.04</b>	<b>16.04</b>
Fruits maturity data	2011	4.07	18.06
	2012	20.06	13.06
	2013	24.06	22.06
	2014	30.06	25.06
	<b>Average</b>	<b>27.06</b>	<b>20.06</b>

Analysing the average productions on four years (2011-2014) from the statistical point of view it is noticed that '*Coralis*' (20.0 kg/tree) registered statistically positive differences compared to the control cultivar '*Boambe de Cotnari*' (19.8 kg/tree) (tab. 3).

The fruit's quality is determined by the fruit's size, epidermis' colour, stone's size and the easy detachment of the stone from the pulp.

The two studied genotypes registered an average weight of the fruit between 6.9 g for the control cultivar '*Boambe de Cotnari*' and 9.1 g for the cultivar '*Coralis*' existing positive significant differences (tab. 3). A continuous growth of the fruit's weight determines also a corresponding growth of the equatorial diameter (D). Thus, the cultivar '*Coralis*' (24.7 mm) registered distinct positive significant differences compared to the control (21.8 mm) (tab. 3).

For the size of the stone, the cultivars registered a weight between 0.30 – 0.34 g this being a middle size of stone according to the UPOV questionnaire.

The fruit/stone ratio for the cultivar '*Coralis*' (30.33) was superior compared with the control cultivar (20.29). As a percentage of the stone from the fruit's weight it registered negative significant differences from the statistical point of view compared to the control (4.93%) (tab. 3).

Table 3

**Fruits production and physical characteristics for two sweet cherry cultivars  
(RSFG Iași; average 2011-2014)**

Genotype	Fruits' production (kg/tree)	Fruit's weight (g)	Stone's weight (g)	Fruit/stone ratio	Stone / fruit's weight (%)	Fruit equatorial diameter (mm)
Coralis (HC 885302)	20.0	9.1*	0.30*	30.33	3.29 <sup>0</sup>	24.7**
Boambe de Cotnari (control)	19.8	6.9	0.34	20.29	4.93	21.8
DL 5%	7.3	1.3	0.04	12.21	1.46	1.3
DL 1%	13.4	2.3	0.08	22.42	2.68	2.4
DL 0.1%	29.7	5.2	0.18	49.68	5.95	5.3

Note: - plantation year: 2000, plantation distance 5 x 4 m

Fruits' colour is bicoloured for the cultivar '*Boambe de Cotnari*' and dark red for the cultivar '*Coralis*' (tab. 4).

The pulp firmness is an important quality feature especially for the fruits that are for fresh consumption (Kappel *et al.*, 2000). The two studied genotypes ('*Coralis*' and '*Boambe de Cotnari*') have firm pulp.

The content in dry substance is very important for sweet cherries, because the taste of the fruits depends a lot on it. Regarding the studied cultivars, the values of this parameter were between 16.1% ('*Boambe de Cotnari*') to 19.5% ('*Coralis*') (tab. 4).

The studied cultivars registered good resistance to the fruit's cracking with values under 21% (tab. 4).

Table 4

**The physico-chemical and quality features of the fruits for two  
sweet cherry cultivars (RSFG Iași; 2011-2014)**

Genotype	Skin colour	Pulp firmness	Fruit's shape	DS* (%)	Stone's adherence to pulp	Fruits cracking after 6 hours (%)
Coralis (HC. 885302)	Dark red	Firm	Kidney-shaped	19.5	Non-adherent	20.0
Boambe de Cotnari (control)	Half yellow, half red	Firm	Heart-shaped	16.1	Non-adherent	20.7

\*DS – dry substance

## CONCLUSIONS

1. The cherry cultivar ‘*Coralis*’ it’s according with the current objectives concerning the tree’s vigour, the flowering lateness, the productivity, the fruit’s quality and ripening time placed at the extremity of the sweet cherries harvest season.

2. The new cultivar with late fruit’s maturation could extend the sweet cherry season for fresh consumption or processing with 7-15 days.

3. ‘*Coralis*’ was registered in March 2016 in the National List of Variety and submitted for patent.

*Acknowledgments.* This study has been partially financed by the Ministry of Agriculture and Rural Development, Grant No. ADER 3.2.2. /2015, with title ‘Tree species breeding to increase safety and food security’.

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LUCRĂRI ȘTIINȚIFICE SERIA HORTICULTURĂ, 59 (2) / 2016, USAMV IAȘI  
**PRELIMINARY RESULTS REGARDING WEED  
MANAGEMENT IN FRUIT TREE NURSERY REGARDING  
MONOCOTYLEDONOUS POACEAE WEEDS AT APPLE  
SPECIES (*MALUS DOMESTICA*, BORKH)**

**REZULTATE PRELIMINARE PRIVIND COMBATEREA  
BURUIENILOR MONOCOTILEDONATE DE TIP GRAMINEE ÎN  
PEPINIERA POMICOLĂ LA SPECIA MĂR  
(*MALUS DOMESTICA*, BORKH)**

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*Abstract.* The field trial was established at Fruit Research Station Bistrita in a sandy-loam site, with optimum rainfall and thermal conditions in 2016. The main objective of the study was the applying of integrated weed management techniques in fruit nursery. The experimental design was polyfactorial and consisted in the application of two non selective herbicides with active compounds quizalofop p-tefuryl 40 g/L and glifosate acid 360 g/L at rootstocks M106, M26, M9 using two different leaf fertilizers in first year of rootstock development (Field 1). Main observed weeds were: *Avena fatua*, *Lolium temulentum*, *Echinochloa crus galli*, *Setaria glauca*, *Agropyron repens*. Control variant consisted in the applying of perforated black agro-textil mulch in the rootstock zone. Herbicides were applied using a conical PVC protection not to damage the main rootstocks during sprays. Research results showed that the applied herbicides and the control variant had different effects on rootstock growth.

**Key words:** fruit nursery, non selective herbicides, rootstock development, agro-textil mulch

*Rezumat.* Experiența a fost efectuată la Stațiunea de Cercetare Dezvoltare pentru Pomicultură Bistrița într-un sol luto-nisipos având condiții optime de temperatură și precipitații în anul 2016. Obiectivul central a fost aplicarea combaterii integrate a buruienilor în pepiniera pomicolă. Schema experimentală a fost polifactorială și a constat în aplicarea a două erbicide neselective având substanța activă quizalofop p-tefuryl 40 g/L și glifosat acid 360 g/L aplicate la portaltoi M106, M26, M9 utilizând doi fertilizanți foliari în câmpul I. Cele mai importante buruieni au fost *Avena fatua*, *Lolium temulentum*, *Echinochloa crus galli*, *Setaria glauca*, *Agropyron repens*. Varianta martor a constat în aplicarea unui mulci agrotexil negru, perforat în zona portaltoilor. Erbicidele au fost aplicate utilizând o protecție conică din PVC pentru a nu deteriora plantele. Rezultatele cercetării au arătat faptul că erbicidele aplicate și varianta martor au avut o influență diferită asupra creșterii portaltoilor.

**Cuvinte cheie:** pepiniera pomicolă, erbicide neselective, creșterea portaltoilor, mulci agrotexil

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The high quality demands of the certified planting material imposes an efficient weed management control (Wertheim *et al.*, 2003). In the fruit nursery the certified planting material should grow in good agrotechnical conditions without weed infestation. Growing process in the nursery field for the first year rootstock development is deeply influenced by the competition between the weeds and the rootstocks. The weeds influence negatively the growing process, development is shifted, the wood is not matured and the resulting planting material does not achieve the standard dimension for selling. Researches effectuated by researchers in Lithuania (Kviklys *et al.*, 2009) for a series of herbicides showed different effects on the nursery plants. Some of the applied pesticides affects the leaves with scorching, burning when directly applied, or causing development problems in the rootstock or young grafted trees (Altland, 2005). Some of the pesticides behaved well regarding the weed eradication but reduced the plants vigor. Some of the herbicides applied in the past can not be applied in the present due to legislation, or the regulations are different from country to country, some of the pesticides are not registered for nursery application just for vegetables, or other cultures, this fact makes more complicated the weed management system (Rankova, 2011). Thus the application of only herbicides in the fruit nursery is still a difficult problem. The objective of our research was to study and observe the behavior of two nonselective herbicides having the active compounds quizalofop p-tefuryl 40 g/L and glifosate acid 360 g/L applied in the first year of development of the rootstocks at apple (M106, M26, M9) and using other weed management techniques like agro textil mulching.

## **MATERIAL AND METHOD**

Researches were effectuated at the experimental micro-nursery of Fruit Research and Development Station Bistrita in 2016. The micro-nursery is located near Bistrita river, establishment of the first year rootstocks was on a sandy-loam site, well drained, with good thermal and rainfall conditions in 2016. The experiment followed a completely randomized block design with 3 repetitions per variant. Factor A was represented by rootstock type with graduations M106, M26, M9, factor B was represented by herbicide application (quizalofop p-tefuryl 40 g/L with a dose of 2l/ha and glifosate acid 360 g/L with a dose of 4l/ha both doses calculated for the research plot surface), factor C – foliar fertilizer application (Foliar fertilizer 1-NPK 19:19:19-dose:0.5% and Foliar fertilizer 2-0.2%-composition for 1L/ NPK: 0.2%:0.4%:0.02%+microelements Mg, Zn, Cu, Mn, B, Ca, Mo,+biostimulators). Foliar fertilizers were applied separately with another spraying equipment. Herbicides were applied with great care with a conical PVC protection not to damage the rootstocks. The herbicides were applied between the rows. The control variant was represented without herbicide and foliar fertilizer applications just only with agro-textil mulch application. At the final of the growth period in 2016 there were measured the shoot development length, there were counted the number of weeds, there were calculated the weed infestation amount and efficiency of herbicide application. The weed amount evaluation was effectuated based on the scientific literature (Rusu *et al.*

LUCRĂRI ȘTIINȚIFICE SERIA HORTICULTURĂ, 59 (2) / 2016, USAMV IAȘI 2012). The weed species were identified based on visual characteristics. The counting of weeds were effectuated using of measuring frame with a surface of 0.25 m<sup>2</sup>. Data obtained were registred in an observation card regarding the average number of weeds/m<sup>2</sup> for every species, making an average of the counted weeds at every repetition and dividing the sum at the total repetition number. The obtained value was multiplied with 4 in the case of measuring frame of 0.25 m<sup>2</sup> to obtain the number of total weeds per square meter. The registered data was statistically analyzed by the analysis of variance test and differences of limits Duncan's test.

## RESULTS AND DISCUSSIONS

Meteorological conditions were relatively optimum in the beginning of the growing period. Table 1 show that thermal conditions were ascendent in the spring during april-june 2016, average temperatures were between 12.4-19.8 °C, being relatively constant until the last part of summer (20.3-19.9). Lack of balance can be observed in the rainfall conditions, this factor fluctuated during spring and summer. In june 2016 a great amount of rainfall was observed (147mm), rootstocks had a good development and growth in the period of june and july (64 mm). In august 2016 rainfall amount was reduced, only 22 mm were registered. Maximum temperatures fluctuated between 25.8-32.5°C during april-august 2016 and were relatively constant.

Table 1

**Meteorological conditions registered at the meteorological station SCDP Bistrita**

2016	I	II	III	IV	V	VI	VII	VIII
<b>Average temperature in the air °C</b>	-2.8	4.5	5.6	12.4	14.1	19.8	20.3	19.9
<b>Average minimum temperature</b>	-5.3	1.4	2.2	7.2	8.8	14.5	14.6	14.8
<b>Absolute minimum temperature</b>	-14.9	-6.5	-5.6	0.4	4.3	7.9	7.9	7.6
<b>Average maximum temperature</b>	0.7	8.8	11.3	19.3	20.2	26.1	26.8	26.3
<b>Absolute maximum temperature</b>	9.1	17.5	18.8	25.8	27.6	32.5	32.5	31.5
<b>Relative humidity %</b>	85.6	82.0	66.6	66.3	69.5	79.9	72.2	68.7
<b>Rainfall (mm)</b>	66.7	76.1	21.0	54.2	48.2	147.4	64.3	22.0

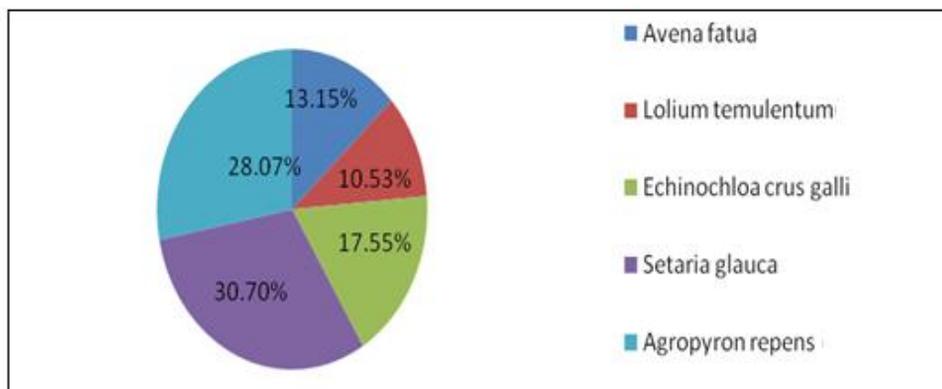
In the spring period there were effectuated the identification and evaluation of weed infestation degree and the counting of weeds per species and category. In table 2 there are shown synthetically the number of weeds per weed category. Based on the registered data the most important species is the couch grass (*Setaria glauca*) with a 30.7 % degree infestation.

Table 2

**Analysis of main weed species, counting the average number of weeds, degree of weed infestation in may 2016 at SCDP Bistrita micro-nursery before herbicide application**

Identified weed	Number of weeds	
	Number of weeds /m <sup>2</sup>	Weed infestation degree %
<b>Annual monocotyledonous weeds with early germination</b>		
1. <i>Avena fatua</i> (wild oat)	15	13.15
2. <i>Lolium temulentum</i> (darnel)	12	10.53
<b>Annual monocotyledonous weeds with late germination</b>		
1. <i>Echinochloa crus galli</i> (barnyard grass)	20	17.55
2. <i>Setaria glauca</i> (yellow foxtail)	35	30.70
<b>Perennial monocotyledonous weeds</b>		
1. <i>Agropyron repens</i> (couch grass)	32	28.07
<b>TOTAL</b>	<b>114</b>	<b>100</b>

After the most important yellow foxtail weed (*Setaria glauca*) follows closely the couch grass (*Agropyron repens*) with a 28.07% and the barnyard grass (*Echinochloa crus galli*) with 10.53% infestation degree and finally the wild oat (*Avena fatua*) 13.15% .



**Fig. 1** Compositional distribution of annual and perennial monocotyledonous weeds in the experimental plot

In figure 2 it can be observed the high degree of weed infestation between the rows, and the control variant with the perforated agro-textil mulch. Close to the mulched row it can be observed the plentiful weed vegetation.



**Fig.2** Aspect from the fruit nursery before herbicide application, plentiful yellow foxtail (*Setaria glauca*) weed infestation

In the frame of the researches the most important research parameter was the average height of rootstocks after a year of growing and development. In the research block of M106 rootstock the research results showed that in repetition 1 and repetition 3 there were no significant differences but in repetition 2 there were significant differences between the variants.

Table 3

**Average of M106 rootstock height (cm) of research variants**

Experimental variants	R1	R2	R3
V1- M106/ quizalofop p-tefuryl 40 / Foliar 1	84.250 a	87.750 a	86.250 a
V3- M106/ glifosate acid 360 g/l / Foliar 1	84.500 a	84.500 a	84.500 a
V2- M106/ quizalofop p-tefuryl 40 g/Foliar 2	80.750 a	83.750 a	89.750 a
V4- M106/ glifosate acid 360 g/l /Foliar 2	81.750 a	76.250 b	85.250 a
V5-Control variant	82.750 a	82.500 ab	83.750 a
Pr > F	0.850	0.020	0.316
Significant	No	Yes	No

In repetition 2 the greatest height was achieved by variant 1 by using quizalofop p-tefuryl 40 g/L and the applying of foliar 1 fertilizer. In this variant the quizalofop p-tefuryl 40 g/L presented a better result than glifosate acid with foliar fertilizer 2. Intermediate result was achieved with the control variant

LUCRĂRI ȘTIINȚIFICE SERIA HORTICULTURĂ, 59 (2) / 2016, USAMV IAȘI regarding the height of plants. This is an important result because the agro-textil mulch allows the water to penetrate the material and do not allows the weeds to grow. The microclimate under the agro-textil mulch is improper for the development of weeds , photosynthesis being not possible due to the black not transparent material so, the achieved average height of the plants in this variant being 82.5 cm.



Fig. 3 Control variant with agro-textil mulch

Table 4

Average of M26 rootstock height (cm) of research variants

Experimental variants	R1	R2	R3
V6-M26/ quizalofop p-tefuryl 40 g/L Foliar 1	67.250 ab	66.000 a	67.750 a
V7-M26/ quizalofop p-tefuryl 40 g/LFoliar 2	62.000 c	65.750 a	64.750 a
V9-M26/ glifosate acid 360 g/L /Foliar 2	68.500 a	62.750 a	64.500 a
V10-Control variant	64.500 abc	64.250 a	64.000 a
V8-M26/ glifosate acid 360 g/L / Foliar 1	63.500 bc	63.500 a	61.750 a
Pr > F	0.049	0.455	0.361
Significant	Yes	No	No

Analyzing table 4 we can conclude that in the case of rootstock M 26 lower heights were measured, due to the vigor of the rootstocks and just in the first repetition were observed significant differences between the variants. The highest plants were obtained at V9 variant with glifosate acid and foliar fertilizer 2 followed by the V6 variant M106/quizalofop p-tefuryl 40 g/L Foliar 1. Decreased heights were observed at the other variants. At M26 rootstock it seems

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 that the glifosate acid had a better result. This variant sprayed with foliar fertilizer 2 had a good development result.

Table 5

Average of M9 rootstock height (cm) of research variants

Experimental variants	R1	R2	R3
V11- M9/quizalofop p-tefuryl 40 g/L / Foliar 1	33.500 a	38.250 a	38.500 a
V14- M9/ glifosate acid 360 g/L /Foliar 2	34.500 a	34.500 ab	33.500 bc
V15- Control variant	32.500 a	32.500 b	36.500 ab
V12- M9/ quizalofop p-tefuryl 40 g/L /Foliar 2	32.250 a	33.500 b	34.250 bc
V13- M9/ glifosate acid 360 g/L /Foliar 1	32.500 a	33.250 b	31.000 c
Pr > F	0.219	0.052	0.007
Significant	No	No	Yes

The measurement results at rootstock M26 in the nursery in repetition 3 showed that there are significant differences between the variants, higher values of height were achieved in variant 11 with quizalofop p-tefuryl 40 g/Foliar fertilizer 1. The second rank was occupied interestingly by the control variant V15 with agrotexil mulch (36.5 cm). It seems that at the low vigor rootstock M9 the presence of the agrotexil mulch helps the elimination of weeds and contributes probable for the maintaining of a relative humidity favorable for the development of the plants. Intermediate results were obtained at V12 - quizalofop p-tefuryl 40 g/L-Foliar 2 (34.2 cm) and V14 M9/ glifosate acid 360 g/L /Foliar 2 (33.5 cm).

## CONCLUSIONS

1. The variants quizalofop p-tefuryl 40 g/L and Foliar fertilizer 1 influenced significantly the growth and development of the plants by removing the monocotyledonous weeds in the research plot.

2. At the applying of herbicides very careful precautions were made, the studied herbicides were total and non selective. The herbicides were not applied directly on the plants just between the rows using a special conical PVC protection not to damage the plants, being careful that sprays does not achieve the leaves of the rootstocks.

3. The glifosate acid 360 g/L and Foliar fertilizer 2 treatments was successful at M26 rootstock

4. The perforated black agro-textil mulch gave also good results by removing the weeds non directly underneath the textil material.

5. When accidentally herbicide drops achieved on rootstock leaves, there observed burning, discoloration and finally the plants dried out. Thus, the studied herbicides are hard to apply, present research results are preliminary and further

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researches will be made in order to make conclusions on possibility of applying these integrated weed management techniques in the field.

***Acknowledgements.** This work was supported by the Romanian Ministry of Agriculture and Rural Development, Project: ADER 3.3.3. – Modern technological solutions regarding the obtaining of fruit tree planting material according European quality standards.*

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## THE CONTROLLED TECHNOLOGY FOR OBTAINING AROMATIC QUALITY WINE FROM MUSCAT OTTONEL VARIETY AT THE RESEARCH AND DEVELOPMENT STATION FOR VINE AND WINEMAKING FROM TÂRGU BUJOR

### TEHNOLOGIE CONTROLATĂ DE OBȚINERE A VINULUI AROMAT DE CALITATE DIN SOIUL MUSCAT OTTONEL LA SCDVV BUJORU

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**Abstract.** To implement the control technology it was used the technology developed infrastructure segment, namely: the harvesting of grapes in plastic crates, the transportation of grapes using the laboratory equipment, the electronic quantitative and qualitative evaluation to determining the sugar content and total acidity; the sorting and selection of grapes; the removing of grapes from cluster and the crushing of grapes with a mini smashing instrument made of stainless steel; the maceration before the fermentation made in small capacity pneumatic press for 12 hours; the controlled fermentation with specific enzymes in 500 L tanks with controlled-temperature cooling jackets. The must (unfermented wine) was obtained by stopping the fermentation process when the desired alcohol content was achieved, preserving a certain quantity of sugar specific for this type of wine. For the current research for producing this aromatic wine with this type of flavour, enzymatic extracted flavours from the grapes were used in the before fermentation stage during the cold maceration. The alcoholic fermentation was carried out in the presence of selected yeasts and nutrients with a complex composition.

**Key words:** flavored wine, pre-fermentation maceration, controlled fermentation, flavors

**Rezumat.** Pentru implementarea tehnologiei controlate s-a apelat la infrastructura dezvoltată pe segmentul tehnologic și anume: culesul strugurilor în ladițe din plastic, transportul cu autolaboratorul, evaluarea cantitativă electronică și calitativă prin determinarea zahărului, acidității totale; trierea și selectarea strugurilor procesați pe bandă de selecție; desciorchinarea și zdrobirea strugurilor cu minizdrobitor din inox; macerația prefermentativă în presa pneumatică de mică capacitate timp de 12 ore; fermentația dirijată cu temperatură controlată în cisterne cu manta de răcire de 500 L și fermenți specifici. Vinul brut s-a obținut prin sistarea fermentației la concentrația de alcool dorită și a prezervării unei cantități de zaharuri specifice acestui tip de vin. În cercetările actuale pentru obținerea vinului aromat de tip muscat s-au folosit preparate enzimatice de extracție a precursorilor de arome varietale din epicarpul strugurilor în etapa macerației peliculare prefermentative la rece.

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*Fermentația alcoolică s-a derulat în prezența drojdiilor selecționate și a nutrienților cu o compoziție complexă.*

**Cuvinte cheie:** vin aromat, macerație prefermentativă, fermentație dirijată, arome

## INTRODUCTION

The aromatic spectrum of Romanian wines has been the main interest of a number of Romanian researchers from different geographical areas like Buia, 2001 (vineyard Târnavă) and Heroiu, 1998 (vineyard from Stefănești-Argeș). Research on the flavour compounds in general and the specific varietal aroma compounds of Muscat Ottonel in particular, have been made at Dealu Bujorului by Ciubucă, Postolache, Miclean (2010). Internationally, the flavours of the grapes and wine are linked by a series of research pertaining primarily to Schneider's Guerin (2001), Rapp and Version (1995).

## MATERIAL AND METHOD

During this research there were used grapes from the Muscat Otonel variety which were harvested at technological maturity, having a content of sugar of 226 g/L, a tartaric acidity of 6.8 g/L and a pH of 3.36.

The processing of the harvest was made using the modern technology of the research laboratory. The harvesting of the grapes was made manually using crates which were transported with the laboratory equipment. The quantitative evaluation was made using electronic scales, the grapes being sorted before removing from the cluster and smashed. The obtained must using a smashing instrument of low capacity of only 53.7% was fermented in a stainless steel tank using enzymatic extracted flavours and selected yeasts, and then it was cleared, filtered and bottled.

## RESULTS AND DISCUSSIONS

The process of winemaking from the Muscat-Otonel variety involved working with a technological process to extract and preserve the aromatic compounds of the wine. To reach this goal 3g/hL of enzymes of the Zymoclaire Pro Ice type were used together with the sulfitation of the must which happened 4 hours after the enzymes were added, the surface maceration taking place at 17 °C for a period of 24 hours. The must was pressed pneumatically in the press using the Macera programme. The surface maceration is an important stage because this is when the extraction and the diffusion of the aromatic compounds happens.

**The harvest processing.** The evaluation of the harvest includes both the quantity and quality aspects. The first involves the economic aspects like kg/ha, the financial assessment of the harvest, and the second involves the monitoring of the grapes' ripening through a periodical analysis of the sugar content, acidity and pH of the must, which may provide useful information for future necessary corrections.

The processing of the harvest involves sorting removing the grapes from the cluster, the separation of the rachis, the smashing and the cooling of the must. The sorting of the harvest involves the removal of leaves and other debris while the grapes are on the conveyer belt which must be set at a very low speed of 5 m/min.

**Antioxidant protection.** For healthy harvests a treatment with Antioxin W is recommended. This is a mixture of ascorbic acid and potassium metabisulfite of 20 g/hL which will release more than 50 mg/L SO<sub>2</sub> leading to an antioxidant result of the ascorbic acid ten times bigger than the result obtained through separate usage or in association with SO<sub>2</sub>.

The careful smashing of the grape ensures the obtaining of the must. In modern winemaking, the smashing is removed from the process and the grapes removed from the cluster are placed directly in the pneumatic press (fig. 1).



Fig. 1 Pneumatic press

The cooling of the must at 14-15 °C before the enzymatic treatment is compulsory because it ensures the protection of the smelling potential of the grapes.

*The enzymatic treatment in association with cold surface maceration before the fermentation.* Both operations involve the extraction of odorous substances from the skin of the grapes using a natural process of enzymatic maceration before the fermentation. The enzymatic treatment represents the most important procedure and has direct consequences on the sensory profile of the future wine. In the case of healthy harvests with a low maturity stage it is required the enzymatic treatment with enzyme extracts. The surface maceration only happens in the case of healthy crops with an advanced stage of maturity of the grapes. It is recommended to administer in the must an enzymatic compound from the Zymoclaire variety after the pneumatic press is loaded, making sure that the temperature is above 15 °C (preferably 17-18 °C).

The assembly of the must fractions from the first smashing is done using the must (unfermented wine) obtained without pressing the grapes.



Fig. 2 Controlled fermentation tanks



Fig. 3 Storage tanks

The settling of the must; this process is ensured either through the enzymatic treatment with Antioxin W or  $\text{SO}_2$ , or by lowering the temperature to 10-12 °C, followed by a gradual rise in temperature until the recommended temperature of 18 °C before adding the selected yeasts to start the alcoholic fermentation process. The intensity of the settling process is connected with the process of removal of the big burble and the preserving of the medium and fine burble which are natural nutritional resources necessary for the multiplying of the yeasts. NTU (Nephelometric Turbidity Unit) must be between 100-150 and 200-250 NTU.

**Directing and controlling the fermentation process.** The fermentation was controlled by adding yeasts of the Fermactive type of about 10 g/hL. The fermentation process was monitored on a daily basis and after five days the density of the must lowered to the value of 1.030 with a sugar content of 58 g/L (fig. 2). In the following stage the wine was cleared by adding 1 g/L of bentonite and 180 mg/L  $\text{SO}_2$  as antioxidant protection.

During the stage after the fermentation several operations of improving the aromatic profile were done by adding 4 g/hL of Zymovarietal Aroma G enzymes

at a temperature of 18 °C. This type of enzymes have a high level of  $\beta$ -glucosidase which cleaves the aromatic compounds connected with the sugar. The evaluation of the completion of the alcoholic fermentation i.e. the winetesting reveals the result. The raw wine was stored in stainless steel tanks (fig. 3).

The essential stages in winemaking from the Muscat Ottonel variety are the administration of the enzyme treatment during the cold surface maceration before the fermentation until the must is obtained, the adding of nutritional compounds with complex composition and selected yeasts of Fermactive blanc aromatique type for the fermentation. The compulsory second enzyme treatment releases the flavours and ensures a high level of fruity and floral character of the wine. Also very important is the monitoring of the temperature, the density, the alcoholic concentration, the oxygen level and the titre of the yeasts. Temperature variations higher than 1-2 degrees must be avoided.

The stage after the fermentation refers to the improvement of the of the smelling qualities of the wine. The enzyme treatment which releases the smelling compounds is done at the end of the alcoholic fermentation process or immediately after the first decanting.

If the enzyme treatment is done at the end of the alcoholic fermentation process, there is a risk of starting FML, a process which can be balanced by correcting the acidity level to the value of 6.3-6.6 g/L tartaric acid (using 2/3 tartaric acid and 1/3 citric acid).

At higher temperatures than 15 °C, it is recommended an enzyme treatment with 5g/hL of Zymovarietal aroma or 4g/hL of Endozym  $\beta$ -Split. The treatment must be administered at least 2-3 weeks to ensure maximum efficiency; the tanks must be full and antioxidant protection must be achieved by adding CO<sub>2</sub> or inert gas.

The monitoring of the enzyme treatment. It is recommended to test the wine which is being treated at least 2-3 times a week in order to establish the quality and the optimal flavours because of the gradual release of this flavours through a enzyme hydrolysis process. After the alcoholic fermentation is over and after the completion of the second enzyme treatment it is recommended to lower the temperature to 14-15 °C.

The characteristics of the wine when bottled and stored (fig. 4, fig. 5).

From the organoleptic and physico-chemical composition point of view the wine is ballanced. It is a semi-sweet wine with an alcohol content of 11% vol., a volatile acidity of 0.18 g/L CH<sub>3</sub>COOH, 24 mg/L free SO<sub>2</sub> and 163 mg/L total SO<sub>2</sub>. It also has 33 g/L reducing sugars, a non-reducing sugar content of 35 g/L and a pH of 3.25 with an intense muscat flavour, which assured this wine the gold medal at the "Bachus-Focșani" national wine contest this year.



Fig. 4 Bottling line



Fig. 5 Bottled wine

## CONCLUSIONS

1. To obtain a flavoured wine specific to the wine variety it is necessary to follow all the correct technological stages together with a firm monitoring of all the processes of extraction and aromatic preservation under the action of enzymes and those of alcoholic fermentation.

2. The obtained wine through this technology has good quality organoleptic features of composition which recommend this wine.

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## METHODS FOR ASSESSING THE MYCOTOXINS POTENTIAL IN OENOLOGY INDUSTRY

### METODE DE EVALUARE A POTENȚIALULUI ÎN MICOTOXINE DIN INDUSTRIA OENOLOGICĂ

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**Abstract:** *Recent studies have confirmed that mycotoxins such as aflatoxins are present in foods and are responsible for most cases of liver cancer and modification of human DNA. These forms on foods where conditions of handling, transportation and storage promote mould growth, such as high temperature and humidity. There are four types of aflatoxins in food isolates (B1, B2, G1, G2) with B1 the most toxic. The study aims to assess methods of analysis for these important substances in human health and quality of wine. The maximum authorized concentration of this class of mycotoxins is 2 ppb. The determination of aflatoxins in the wine can be achieved by high performance liquid chromatography, thin layer chromatography, gas chromatography and ELISA. The purpose of this work is to stimulate risk awareness to the improper selection of harvest quality and the need to invest in the product safety.*

**Key words:** grapes, mycotoxins, toxicity

**Rezumat:** *Studiile recente au confirmat faptul că micotoxine precum aflatoxinele sunt prezente în alimente și sunt responsabile de cele mai multe cazuri de apariție a cancerului hepatic precum și denaturarea ADN-ului la om. Acestea se formează pe alimente în cazurile în care condițiile de manevrare, transportare și depozitare favorizează apariția mucegaiului, precum umiditate și temperatură ridicată. Există patru tipuri de aflatoxine izolate din alimente (B1, B2, G1, G2), iar B1 are toxicitatea cea mai mare. Studiul își propune evaluarea metodelor de analiză pentru aceste substanțe importante pentru sănătatea umană și calitatea vinului. Concentrația maximă autorizată ale acestei clase de micotoxine este 2 ppb. Determinarea aflatoxinelor din vin se poate realiza cu cromatografia de lichide de înaltă performanță, cromatografia în strat subțire, tehnica ELISA și cromatografia de gaze. Scopul lucrării este conștientizarea riscului pe care îl reprezintă selecția necorespunzătoare a recoltei și necesitatea investițiilor în cea ce privește calitatea produsului.*

**Cuvinte cheie:** struguri, micotoxine, toxicitate

## INTRODUCTION

The grape is one of the most popular fruit in the world, intended mostly for wine making, namely 71% of its production, 27% is consumed fresh and 2% is converted into dried fruits (raisins). Regarding the quality of the wine we

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recognise two aspects: first the external quality that refers to the sensory characteristics and secondly that domestic quality is given by the chemical composition, hygiene factors related to wine toxicology.

In the last twenty years we have studied a lot of wine toxicity makers, with particular interest in mycotoxins. For these reason government agency tightened harvesting and processing conditions for winemakers and wanted to develop new techniques for identifying, quantifying and reducing mycotoxins in wine (Somma *et al.*, 2012).

Mycotoxins are metabolic by-products formed by moulds (Somma *et al.*, 2012), hence their name (Online Etymology Dictionary). Estimates worldwide show that at least a quarter of crops are affected by mould, which implies the risk of mycotoxin poisoning and possible economic losses (Cazzaniga *et al.*, 2001; SommaS. *et al.*, 2012). There are several types of fungi that lead to mycotoxins, but the most common include *Aspergillus*, *Alternaria*, *Penicillium* (Valero *et al.* 2005; Neacșu, 2012). Diseases caused by these toxins are called generally "mycotoxicoses" (Bennett and Klich, 2003). The most important classes of mycotoxins due to toxicity and their influence on human health and their correspondence in oenology are: Aflatoxins, Ochratoxins, Citrinin, Fumonisin, Patulin, Trichothecenes and Zearalenone.

Aflatoxins are produced by the fungus *Aspergillus flavus*, from which they were given the name (A -fla -toxin) (Sargeant *et al.*, 1963). They were first discovered in 1960 when an aviary pandemic occurred to 100,000 young turkeys and 20,000 ducks, pheasants and partridges. They all were strongly infected with *Aspergillus flavus* that developed as fodder came into contact with birds. Samples were taken from the feed and were analysed by Thin-Layer Chromatography (TLC). Thus they were observed on pads by UV fluorescence where two components one blue and one colour green, which were named "Aflatoxin B" and "Aflatoxin G" (Jacobsen *et al.*, 1993; Rustom, 1997; Devero, 1999; Sargeant *et al.*, 1963). It was later found about twenty compounds, but the most common are aflatoxins B1, B2, G1, G2 and two B1 metabolites, M1 and M2 (Agag, 2004). Aflatoxin B1 is 60-80% of the total aflatoxins (FDA, 1979) and is the most toxic natural carcinogen (Jones *et al.*, 1994; Jović *et al.*, 2009). "International Agency for Cancer on Research" framed aflatoxins in group I, i.e. carcinogenic compounds for the human body (IARC, 2016), targeting mainly the liver (Agag, 2004).

Contamination of wine and thus grapes can occur in two ways. First when developing culture is affected physically (by insects, birds, mammals, etc.) or mechanical (by hail, storm) and subsequently subjected to stress caused by heat and drought conditions (Cotty and Lee, 1990; Dowd, 1998). The optimum temperatures for the development of *A. flavus* species are between 25-32 °C (Lillehoj, 1983). Thus, were created favourable conditions develop mildew on grapes after due improper sorting before processing, are a major risk factor for wine quality. Secondly, the healthy grapes which after once harvested are not deposited, transported or handled appropriately and due higher temperatures and

humidity, the grapes are mould growth predisposed that causing toxins (Cotty, 1991; Russell *et al.*, 1976). In these conditions it is necessary that the time from harvest to processing to be minimized.

Ochratoxins were first discovered in 1965, as produced by *Aspergillus ochraceus* (Jović *et al.*, 2009). Currently known several types but the most popular from the point of view of toxicity is "Ochratoxin A". It has been classified by the "International Agency for Cancer on Research" in group 2B carcinogen (substance possibly carcinogenic to humans) (IARC, 2016). According to estimates *Codex Alimentarius* approximate 15% of the total ochratoxin ingested by humans comes from wine and especially from wine red which is the second source of ochratoxin, according to statistics of FAO and *Codex Alimentarius* (Jović *et al.*, 2009; *Codex Alimentarius* Commission). Ochratoxins have hepato-nephrotoxic actions but is not a very high risk to human health (Somma, 2012). They were detected in grapes before harvest (Aziz and Moussa, 2002). Maximum permitted quantity of aflatoxins and ochratoxins in grape juice and wine, according to the regulations E.U. is 2ppb (Vicom, 2016).

**Fumonisin** was first time studied in 1988 (Bezuidenhout, 1988; Gelderblom, 1988) as the product of a number of species such as *Fusarium*, particularly *Fusarium verticillioides* (Rheeder, 2002). When weather conditions are unfavourable and the culture is attacked by insects and pests, on the stem grow rot and seedlings are affected by downy mildew (Nelson, 1993). IARC classified the toxin in group 2B carcinogen.

**Patulin** was first isolated in 1940 from *Penicillium patulum* and used for its antibacterial and antiviral properties, but later was found to contain toxicity to both plants and animals. In 1960 it was classified as mycotoxin and the W.H.O. has established a tolerable upper limit of 0.4 mg/kg bw/day. The most common form in which we find in fruits is "blue mould" caused by *Penicillium expansum* (Bennett and Klich, 2003).

**Trichothecenes** are produced by several types of fungi such as *Fusarium*, *Myrothecium*, *Phomopsis*, *Stachybotrys*, *Trichoderma*, *Trichothecium*. Trichothecenes intoxications causing gastrointestinal bleeding, vomiting, damage to the small intestine and direct contact with mold can cause dermatitis. Trichothecenes can be found in grapes and wine but in very rare cases (Jović *et al.*, 2009; Bennett and Klich, 2003; Somma, 2012).

## MATERIAL AND METHOD

First it is necessary to realize a more accurate sampling. On this line we must have in mind that grapes are contaminated with mycotoxins heterogeneous and then we should use correct sampling techniques to achieve a representative analysis.

To prepare samples for analysis we can take into account several extraction techniques. The clean-up for the samples is done generally by Solid Phase Extraction (SPE), but can try more advanced techniques, such as Solid Phase MicroExtraction (SPME) or we can provide better recovery and higher sensitivity with Stir Bar Sorption Extraction (SBSE) (SBSE, Gerstel). In order to remove interference the desired compounds are concentrated in a solvent of choice to improve selectivity (Rahmani, *et al.*, 2008).

Actual analysis of mycotoxins requires the separation of interest compounds and their detection. The most effective are chromatographic techniques. Thin layer chromatography (TLC) is the first method by which identified aflatoxins and remains one of the most reliable for common laboratories. It was adopted as official method and may to identify and quantify concentrations below 1 ng/g. Currently is developed a technique more advanced by using smaller particles layers for better selectivity High Performance Thin Layer Chromatography (HPTLC). It has a maximum thickness of layer of 100  $\mu\text{m}$  and particle size of stationary phase is between 2-10  $\mu\text{m}$ , contributing to an advanced separation in a much shorter time. This makes the method to be comparable in terms of effectiveness with HPLC and ELISA methods. Another method developed for aflatoxin analysis is Over Pressured-Layer Chromatography (OPLC) which unlike HPLC require less mobile phase and compared to TLC provides better resolution and more compact spots (Rahmani *et al.*, 2008).

High Performance Liquid Chromatography (HPLC) is able to detect all types of mycotoxins and is the most commonly used method for its small amount of volatile solvent consumption. For different types of detectors: UV has a limit of detection 10 ng/g, DAD (PDA) has a limit of detection at 0.03 ng/g, and fluorescence detector (FLD) technique is the best spectrometric method. Citrinin could be detected by this method with a detection limit of 10 ng/g. Coupling Liquid Chromatography with Mass Spectrometry (LC-MS) results a more sensitive and selective. In this case, new techniques have been developed for better identification and quantification. For example, Trichothecenes were determined using Atmospheric Pressure Chemical Ionization (APCI), aflatoxins and ochratoxins by Electro Spray Ionization technique (ESI). A great sensitivity for low levels of ochratoxin concentration in the wine was obtained using clean-up with SPE C18 combined LC-ESI-MS/MS. For fumonisins LC-MS has 40 pg/Kg and most sensitive and accurate LC-MS/MS yielding a detection limit at 12 pg/Kg. To determine mycotoxins the best analysers are ion trap, time-of flight and triple quadrupole mass analysers (Rahmani *et al.*, 2008).

Wine mycotoxins can also be determined by Gas Chromatography coupled with mass spectrometry (GC-MS) or a flame ionisation detector (GC-FID) to obtain the better results. By this technique ochratoxins have a quantification limit of 2  $\mu\text{g/L}$  (Rahmani *et al.*, 2008).

Also common methods used in the analysis of mycotoxins, particularly aflatoxins are the ELISA tests (Enzyme linked immunosorbent assay). These clinical methods have good recoveries between 91% and 104% with a limit of detection at 0.01  $\mu\text{g/L}$  (Rahmani *et al.*, 2008).

## RESULTS AND DISCUSSIONS

Whichever technique of analysing mould toxins is used, it must be specific enough, fast and it must help us get low detection and quantification limits. One the right concentration of the mycotoxins present in food are identified the manufacturers must design a strategy to reduce the level of contamination.

The safety measures for obtaining a safe wine must be applied starting from the planting of the vine until harvest time. In this case it must be taken into account the climate type, avoiding soils with high humidity, protecting the crops from pests and potential diseases (Jović *et al.*, 2009). The selection of the grapes during harvest and before processing must be done in a way such that the risk of contamination is reduced as much as possible. This can be done easily with the new grape sorting technologies. One choice would be using the machine that uses

a blowing mechanism to select the grape by its specific weight which would help remove the dry or mould damaged grape (e.g. Delta RFlow). Another choice would be optical selection using the machine which selects the grape by its colour, being able to remove mould damaged grapes, whose appearance is changed (e.g. Delta Vistalys by BUCHER).

Once the selection was done qualitatively the next step is to store, transport and process the grapes in good hygienic conditions keeping them away from humidity and high temperatures. In case the product is contaminated in this step, there are a few treatments that can still be applied to the wine to lower the mycotoxin concentration: a yeast with good absorbing properties could be used during the alcoholic fermentation process; clearing fabrics that are able to absorb mycotoxins could be used, such as those based on silicagel or cellulose, but all of these also have disadvantages and can badly influence the sensorial properties (Jović *et al.*, 2009).

## CONCLUSIONS

Currently the requirements regarding wine quality and human health need investment and involvement both from the producers, with modern harvesting and processing technologies, and from the analysts, with faster and more sensitive mycotoxin determination and quantification methods.

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## THE ANTIOXIDANT CAPACITY OF NEGRU AROMAT WINES ENRICHED WITH ARONIA MACERATE

### CAPACITATEA ANTIOXIDANTĂ A VINULUI DE NEGRU AROMAT ÎMBOGĂȚIT CU MACERAT DE ARONIA

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**Abstract.** *This study was aimed to identify a source of antioxidants, compatible with the composition of the wine in order to be used for obtaining an antioxidant capacity of wines with a value of 4000 μmol Trolox/150 mL wine. The optimal dose of the macerate was established on the basis of the relationship between the dose of the macerate and total antioxidant capacity of the wine. For the enriched wine the polyphenolic and sensorial profile was established. The macerates of Aronia were made in wine and in synthetic wine solution. The maceration in wine, at a rate of 7, 15 and 25% was performed at room temperature. The total antioxidant capacity of wines was estimated using the TEAC method. The relationship between the dose of added macerate and total antioxidant capacity of the wine was linear and statistically. The lower dose 7% of Aronia macerate didn't modified the sensory profile and polyphenolic content of wines, while average doses of 15, 25% had improved the wines quality.*

**Key words:** antioxidant capacity, Aronia macerat, polyphenolic content

**Rezumat.** *Studiul a fost realizat pentru a identifica o sursă de antioxidanți, compatibilă cu compoziția vinului, care să fie utilizată pentru realizarea capacității antioxidante proiectate a vinului de 4000 μmol Trolox/150 mL vin. Doza optimă de macerat s-a stabilit pe baza relației dintre doza de macerat și capacitatea antioxidantă totală a vinului. Vinului îmbogățit i s-a definit profilul polifenolic și sensorial. Maceratele de Aronia au fost realizate în vin și în soluție sintetică de vin. Macerarea în vin s-a realizat la temperatura ambiantă. Maceratele au fost adăugate în vin în proporție de 7, 15 și 25% (vol/vol). Capacitatea antioxidantă a vinului a fost estimată prin metoda TEAC. Relația dintre doza maceratului adăugat în vin și capacitatea antioxidantă totală a vinului a fost liniară și asigurată statistic. Dozele mici (7%) de macerat de Aronia, nu au modificat profilul polifenolic și senzorial al vinului, iar dozele medii (15, 25%) au îmbunătățit calitatea vinului.*

**Cuvinte cheie:** capacitate antioxidantă, macerat de Aronia, conținut de polifenoli

## INTRODUCTION

The wine is a food whose biological value is given by its many therapeutic properties. Red wines contain high levels of antioxidants with an important role in

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preventing and repairing damages caused by free radicals (Elford, 1991). The beneficial effects of moderate consumption of wine, especially red wines, in the decrease on mortality risk caused by cardiovascular, neurodegenerative diseases and certain types of cancers have been highlighted by numerous epidemiological studies (Bagchi *et al.*, 2000; Briviba *et al.*, 2002; Katalinic *et. al.*, 2004; Rusoo, 2007). Consequently, the interest in developing the methods to obtain wines very rich in antioxidants has increased.

Antioxidants are compounds major, responsible for therapeutic effects of wine. As a consequence of increasing of concentration of antioxidant in wine, the biological value of the wine is greater. The total antioxidant capacity of grapes was valued at 2016  $\mu\text{mol Trolox}/100\text{ g}$ , corresponding to wine production potential rich or very rich in antioxidants.

Phenolics are responsible for many important properties of wine including color, bitterness, astringency and antioxidant capacity. The concentration and composition of phenolics in red wine are depend on environmental and management factors such as climate, soil conditions, canopy management factor (Jackson and Lombard, 1993).

## MATERIAL AND METHOD

The study was conducted during 2015 year, at the ICDVV Valea Călugărească Institute in microvinification conditions. Source of antioxidants dry bio *Aronia* fruit was used. The maceration was done in two variants, in synthetic solutions for wine and wine. The macerate in wine was performed at room temperature. The optimal duration of maceration was established as the duration of the total antioxidant capacity was maximum ratio of macerate. The ratio between fruit and wine was 125 g/1000 mL. The macerates were added to wine in a 7, 15 and 25% (vol/vol). Each experimental variant was conducted in three repetitions.

The total antioxidant capacity of wines was estimated using the TEAC method, based on the scavenging of ABTS<sup>+</sup>. The red wines were diluted with ethanol 10 or 100 times, depending on the obtained absorbance. Then 100  $\mu\text{L}$  of diluted sample, Trolox or ethanol were mixed with 2.9 mL diluted ABTS radical solution and after 3 min the absorbance was measured at 735 nm, the results being expressed as  $\mu\text{mol Trolox}/150\text{ mL wine}$ . The quantity of 150 mL is considered one serving of wine.

For the determination of the total content of polyphenolic compounds in wines the DO280 index was considered: wine was diluted with distilled water (1:100) and the absorbance was measured directly at 280 nm. The value of OD 280 index for each sample was given as the absorbance multiplied by the proper dilution rate (Ribereau-Gayon, Dubourdieu and Donèche, 2006).

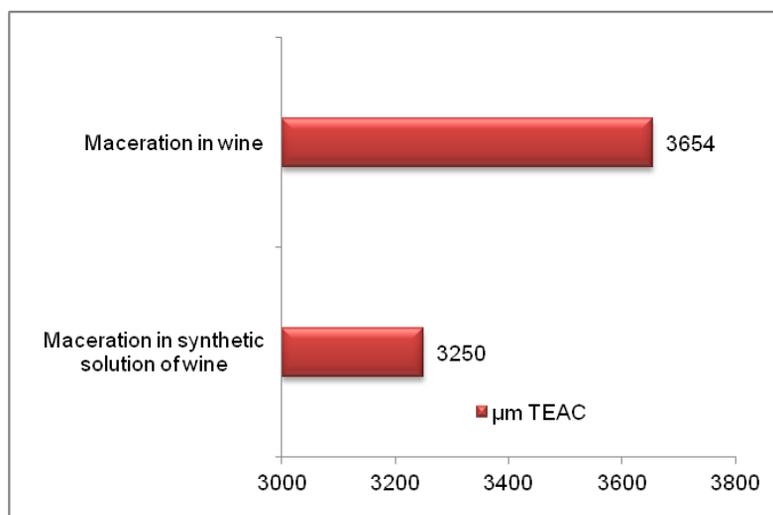
The determination of catechins (flavan 3-ols) is based on the reaction of the phloroglucinol ring with vanillin, that produces a red colour with a maximum absorption at 500 nm. For quantitative evaluation, a calibration curve is traced, by using a solution of (+)-catechin in etanol de 96% (Amerine and Ough, 1974).

The experimental estimation of tartaric esters is based on the specific absorbtion wavelength of 320 nm.

## RESULTS AND DISCUSSIONS

### 1. Evaluation of the total antioxidant capacity of dry *Aronia* fruits macerate and polyphenolic characterization of their composition

The maceration was done in Negru aromat wine, 2015 harvest, which had a total antioxidant capacity of 3654  $\mu\text{mol Trolox}/150\text{ mL}$  was observed in case of macerate in wine. *Aronia* dry fruit maceration in synthetic solutions of wine and in wine caused obtain extracts that were differentially by their total antioxidant capacity (fig. 1). Minimum value (3250  $\mu\text{mol Trolox}/150\text{ mL}$ ) the macerate at wine synthetic solution.

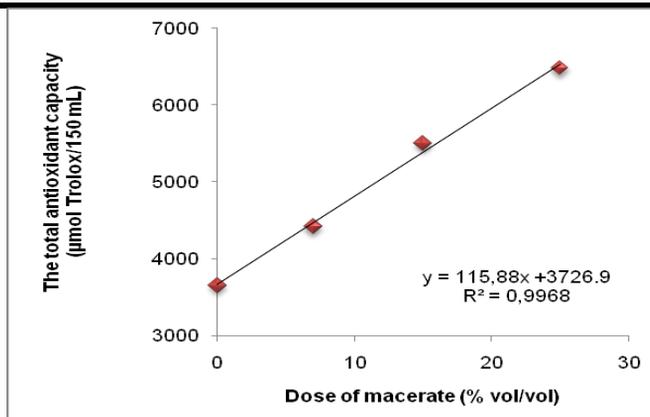


**Fig. 1** The total antioxidant capacity ( $\mu\text{mol Trolox}/150\text{ mL}$ ) of macerate from dry *Aronia* fruits

### 2. Modeling the relationship between the dose of *Aronia* fruits macerate and total antioxidant capacity of wine enriched in antioxidants

The relationship between the dose macerate for wine enrichment added antioxidants and total antioxidant capacity of wine was linear (fig. 2). The coefficient of determination was 0.9968.

$$Y=115.88x + 3726.91$$

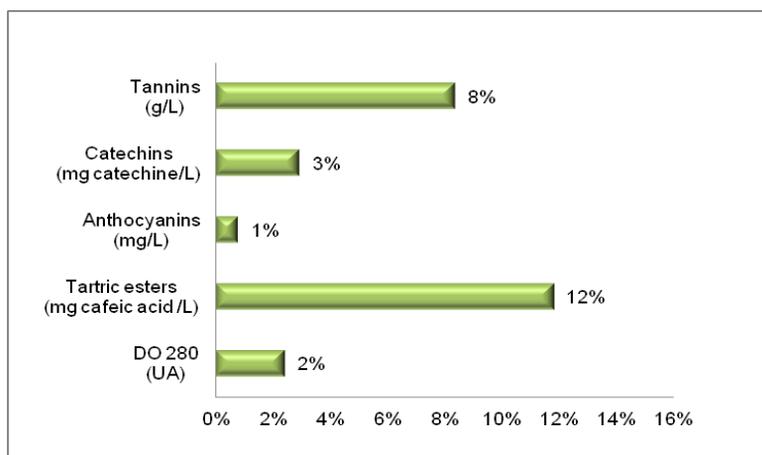


**Fig. 2** The influence of the type and the dose of the macerate on the total antioxidant capacity of the Negru aromat wine, 2015 harvest

Comparative analysis of total antioxidant capacity calculated based on the mathematical model and determined by chemical analysis allowed to assert with a 95% probability that the results obtained were reproductibility of 15 micromol Trolox/150 mL wine.

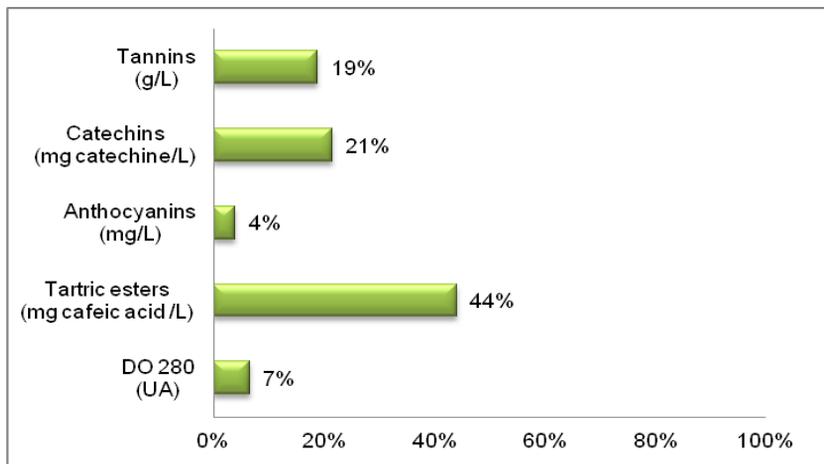
### 3. Polyphenolic profile of Negru aromat wine enriched in antioxidants

The structure of polyphenolic profile of Negru aromat wine was more and less modified. The addition of 7% macerate, regardless of type, caused smaller increases (<10%) to DO280, anthocyanins and catechins (fig. 3). Substantial increases (<15%) were recorded at the tartaric acid esters of phenolic acids and tannins.



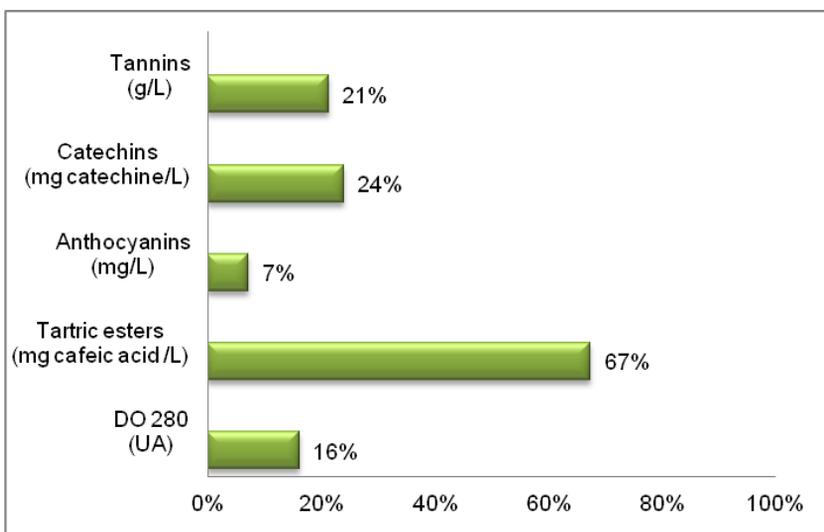
**Fig. 3** Modification of polyphenolic content of Negru aromat wine by enrichment with 7% macerate of *Aronia*

The dose of *Aronia* macerate 15% significantly modified the concentration of all polyphenolic compounds, with the exception of the anthocyanins which recorded increases of less than 5% (fig. 4).



**Fig. 4** Modification of polyphenolic content of Negru aromat wine by enrichment with 15% macerate of *Aronia*

When the macerate dose was 25% the full composition of the wine polyphenols has changed. Very high quantitative increase of acid esters of phenolic acids (67%), catechins (24%) and tannins (21%) were recorded (fig. 5).



**Fig. 5** Modification of polyphenolic content of Negru aromat wine by enrichment with 25% macerate of *Aronia*

## CONCLUSIONS

1. The structure of polyphenolic profile of Negru aromat wine was more and less modified. The addition of 7% macerate, regardless of type, caused smaller increases (<10%) to DO280, anthocyanins and catechins.

2. The dose of *Aronia* macerate 15% significantly modified the concentration of all polyphenolic compounds, with the exception of the anthocyanins which recorded increases of less than 5%.

3. When the macerate dose was 25% has changed the full composition of the wine polyphenols. Very high quantitative increase were recorded from tartaric acid esters of phenolic acids (67%), catechins 24%) and tannins (21%).

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## THE INFLUENCE OF LEAD ON THE GROWTH AND FLOWERING OF *HYACINTHUS ORIENTALIS* SPECIE

### INFLUENȚA PLUMBULUI ASUPRA CREȘTERII ȘI ÎNFLORIRII LA SPECIA *HYACINTHUS ORIENTALIS*

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**Abstract.** Experiment was conducted to investigate the growth and flowering at *Hyacinthus orientalis* exposed to lead toxicity. The experience was organized in five variants, each with three repetitions, and each repetition having 10 bulbs. The experience was organized in five variants, the watering process of the substrate was performed using distilled water for the control variant and Pb (NO<sub>3</sub>)<sub>2</sub> solutions for the other variants. The influence of lead on the plant growth and development has been assessed by plant height, number of flowers and the photosynthetic pigments content. The growth in height and capacity of plant flowering were inhibited by applying the lead, regardless of the dosage used.

**Key words:** *Hyacinthus orientalis*, lead, photosynthetic pigments

**Rezumat.** Scopul experimentului a fost de a studia influența toxicității plumbului asupra creșterii și înfloririi speciei *Hyacinthus orientalis*. Experiența a fost organizată în cinci variante, cu câte trei repetiții, iar fiecare repetiție având 10 bulbi. În cadrul celor cinci variante experimentale, umectarea substratului a fost realizată utilizând apă distilată pentru varianta martor și soluții de Pb(NO<sub>3</sub>)<sub>2</sub> pentru celelalte variante. Influența plumbului asupra creșterii și dezvoltării plantelor a fost evaluată prin înălțimea plantelor, numărul de flori și conținutul de pigmenți fotosintetici. Indiferent de doza utilizată, creșterea plantelor și capacitatea de înflorire a fost inhibată prin aplicarea plumbului.

**Cuvinte cheie:** *Hyacinthus orientalis*, plumb, pigmenți fotosintetici

## INTRODUCTION

In recent years, the concept of using plants to remediate the sites contaminated with heavy metals (phytoremediation) has gained a great deal of attention (Raskin *et al.*, 1994; Jarvis and Leung, 2002). Like any new approach, phytoremediation advocates for the implementation of phytoextraction as a technology to depollute the contaminated soils, with low-cost and aesthetic advantages, which allow the application of phytoremediation on wide surfaces, even in polluted areas (Vander Lillie *et al.*, 2001). Lead (Pb) is one of the heavy metals commonly found in large quantities since antiquity and

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it has gained considerable importance as a major pollutant of the environment. Although lead occurs naturally in the environment, the environment got contaminated with this metal as a result of the manufacture of gasoline, paints and explosives, as well as the elimination of sludge from municipal wastewater treated with lead (Chaney and Ryan, 1994). Some studies have shown that there is a correlation between the concentration of lead and decreased photosynthesis in plants, most probably caused by the closing of the stomata than by any direct effect of lead on the process of photosynthesis (Bazzaz *et al.*, 1975). According to Kosobrukhov and his collaborators (2004), the photosynthetic activity of plants is governed by many factors, cell size, number of stomata, stomatal conductance (Savari *et al.*, 2002).

## MATERIAL AND METHOD

In order to achieve the objectives of the experiment, it has been studied the species *Hyacinthus orientalis*, 'Blue Jacket' variety, which belongs to the category of rustic flowering geophyte. The studies were conducted on the Teaching Field of the Section of Floriculture within The Faculty of Horticulture, while analyses and calculations were performed within the Horticultural Research Centre of The Faculty of Horticulture within USAMV Iasi, Romania.

The biological material consisted of bulbs of *Hyacinthus orientalis*, the variety 'Blue Jacket', purchased from a firm specialized in the production of seeds and seedlings. The experiment was organized in containers with a capacity of 8 kg and consisted of 4 variants and three repetitions, each repetition containing 9 bulbs. In order to determine the influence of lead on plants' development, the contamination was done with solutions of  $Pb(NO_3)_2$  in different concentration (100ppm variant  $V_1$ , 300ppm in variant  $V_2$ , 500ppm in variant  $V_3$ , 700ppm in variant  $V_4$ ), while in the control variant the substrate was left uncontaminated. The influence of the heavy metal on the plants' growth was assessed through phenological observations, biometric measurements and physiological analyses.

## RESULTS AND DISCUSSION

The observations carried out in order to determine the influence of lead toxicity on the development of the vegetation pheno-phases in 'Blue Jacket' indicated a delay associated with increased concentration of lead. The vegetation started 5 days after the contamination in the plants of the control variant and in the variant with 100ppm lead concentration. The increase of lead concentration in the substrate resulted in a delay of the vegetation, with the most important differences being noted in the groups of plants belonging to variant  $V_3$  (10 days) and variant  $V_4$  (11 days) (table 1). Compared to the plants from the control group, the most important differences were obtained in plants of the variant  $V_3$  (4 days) and  $V_4$  (5 days). Comparing the results obtained in the three variants contaminated with lead did not show important delays in the beginning of flowering in the groups of plants contaminated with high concentrations of lead. In the control group, the flowering took place 4 days faster as compared to the groups of plants with a content of lead of 500ppm ( $V_3$ ) and 700ppm ( $V_4$ ).

Influence of lead on the vegetation pheno-phases in species  
*Hyacinthus orientalis* variety 'Blue Jacket'

Species / variety	Variant	Date of contamination	Starting vegetation	Beginning of flowering	Wilting flowers
<i>Hyacinthus orientalis</i> 'Blue Jacket'	control	27.02.2015	05.03.2015	06.04.2015	12.05.2015
	V <sub>1</sub>	27.02.2015	05.03.2015	12.04.2015	15.05.2015
	V <sub>2</sub>	27.02.2015	07.03.2015	12.04.2015	15.05.2015
	V <sub>3</sub>	27.02.2015	09.03.2015	13.04.2015	14.05.2015
	V <sub>4</sub>	27.02.2015	10.03.2015	16.04.2015	18.05.2015

The plants grown in conditions of lead pollution showed a reduction of the decorative period as compared to the plants of the control group, by 5 days in groups V<sub>3</sub>, V<sub>4</sub> and by 4 days in groups V<sub>1</sub>, V<sub>2</sub> (tab. 1). Determinations regarding the average height of the plants highlighted the influence of the concentration of lead on the growth and development of plants (tab. 2). The largest growth of the flowering rod was obtained in the control group (21.98 cm), while the lowest increase was obtained in the group of plants from variant V<sub>4</sub> (17.85). Compared to the control group, all experimental variants showed a decrease of the height of flowering rods, with the most significant ones recorded in the group of plants from V<sub>3</sub> (5.72 cm) and V<sub>4</sub> (6.53 cm). Regarding the height of the flowering rods of plants grown on substrate containing lead in 100ppm concentration, the differences compared to the control group were insignificant (1.21 cm). The statistical interpretation of the results regarding the average height of the flowering rods showed very significant negative differences in variants V<sub>4</sub> and V<sub>3</sub>, and significant in variant V<sub>2</sub> as compared to those obtained in the control group.

Statistical interpretation of the results showed negative differences in the variant V<sub>2</sub> and very significant negative values in variants V<sub>3</sub> and V<sub>4</sub>, as compared to the control group.

Table 2

Influence of lead on the growth of plants of *Hyacinthus orientalis* variety 'Blue Jacket'

Variant	Height of flowering rod (cm)	Leaf length (cm)	Length of the inflorescence (cm)
control	24.38 <sup>(Mt)</sup>	7.80 <sup>(Mt)</sup>	17.55 <sup>(Mt)</sup>
V <sub>1</sub>	23.17 <sup>(0)</sup>	7.40	16.33
V <sub>2</sub>	20.88 <sup>(00)</sup>	6.95 <sup>(0)</sup>	16.25 <sup>(0)</sup>
V <sub>3</sub>	18.66 <sup>(000)</sup>	6.55 <sup>(000)</sup>	15.56 <sup>(000)</sup>
V <sub>4</sub>	17.85 <sup>(000)</sup>	6.25 <sup>(000)</sup>	14.99 <sup>(000)</sup>
LSD 5%	0.6	0.4	0.3
LSD 1%	0.8	0.8	0.5
LSD 0.1%	1.2	1.5	0.7

The length of the inflorescence showed very significant negative differences in variants  $V_4$  and  $V_3$  when comparing the plants from the control group with the plants grown in substrate contaminated with different doses of lead. The smallest lengths were observed in the group of plants from  $V_3$  and  $V_4$ , which recorded a shorter inflorescence by 1.99 cm and 2.56 cm as compared to the control group. In the case of these experimental variants ( $V_3$  and  $V_4$ ), the statistical interpretation of the results highlighted very significant negative differences as compared to the control group. The lowest average number of flowers was obtained in the group of plants contaminated with 700ppm Pb, where there was a decrease of 11 flowers as compared to the control group. The statistical interpretation of the results obtained in conditions of lead pollution showed significant negative differences in variant  $V_3$  and very significant negative differences in variant  $V_4$  (tab. 3).

Table 3

**The mean number of inflorescence and flower length of Blue Jacket 'variety**

Variant	Inflorescence flowers/number (pcs)	The length of the flowers (cm)
control	46 <sup>(Mt)</sup>	3.4 <sup>(Mt)</sup>
$V_1$	44	3.2
$V_2$	42	3.3
$V_3$	41 <sup>(00)</sup>	4.0 <sup>(x)</sup>
$V_4$	35 <sup>(000)</sup>	4.2 <sup>(x)</sup>
LSD 5%	3.4	0.3
LSD 1%	4.4	0.4
LSD 0.1%	6.6	0.6

Observing the growth trend of the flowers in the inflorescences, we can notice that the variants that have been contaminated with higher doses of lead registered a slight elongation of the flower as compared to the uncontaminated group. At first sight we can say that the presence of lead in the substrate stimulate the growth of flowers on inflorescences in 'Blue Jacket ' variety. Nevertheless, analysing things in detail and consulting the specialty studies, the elongation of flowers in the inflorescences was ascribed to the toxic effects of lead on the plants. Thus, an increase in the levels of lead in the substrate caused an elongation of the flower with 4.0 cm in the plants from variant  $V_3$  and 4.20 cm in the plants from variant  $V_4$ , as compared to the control group (tab. 3). Determinations concerning the content of assimilating pigments in plants grown in lead-contaminated substrate have highlighted the potential for adaptation of the variety 'Blue Jacket' to lead polluted soil (fig. 1). The total content in chlorophyll pigments ranged between 2.74 mg/g s.p. in the control variant and 1.35 mg/g s.p. in the variant contaminated with a concentration of lead of 700ppm ( $V_4$ ). As compared to the control variant, the variants contaminated with lead showed major declines of the total content of assimilating pigments, thus confirming

that lead pollution in the species *Hyacinthus orientalis* the variety 'Blue Jacket' causes stress at physiological level.

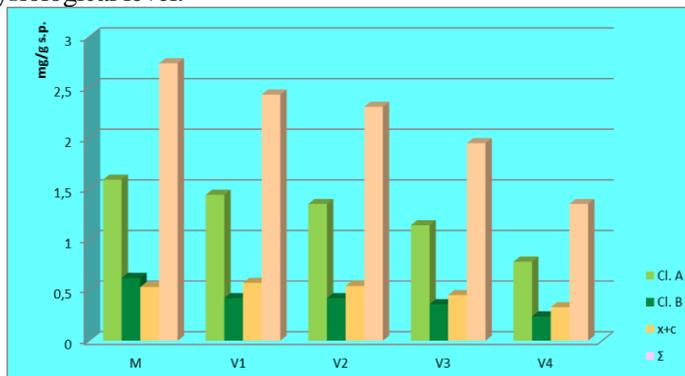


Fig. 1 Content of photosynthetic pigments in the variety 'Blue Jacket' in substrate contaminated with lead

Regarding the content of chlorophyll *a*, compared to the control variant, there can be observed a slight decrease in all experimental variants with values ranging between 1.59 mg/g s.p. in the control group and 0.78 mg/g s.p. in the group of plants with the highest dose of lead (V<sub>4</sub>). The values for the content of chlorophyll *b* have shown important differences, ranging from 0.62 mg/g s.p. in the plants from the control group and 0.24 mg/g s.p. in the group of plants from V<sub>4</sub>. The increase in the concentration of lead causes a decrease in the content of chlorophyll *b*. An important indicator in the assessment of plant adaptation to stress conditions is the ratio of chlorophyll *a*/chlorophyll *b* (Lichtenthaler *et al.*, 1981). In the case of the plants within the experimental variants grown in lead contaminated substrate, the ratio chlorophyll *a*/chlorophyll *b* varied between 3.43 mg/g s.p. in variant V<sub>1</sub> and 3.25 mg/g variant s.p. in V<sub>2</sub> (fig. 1). The results fall within the theoretical limits and the ratio of chlorophyll *a*/chlorophyll *b* is around 3:1 in all variants.

Table 4

The ratio of assimilating pigments obtained in the species *Hyacinthus orientalis*, the variety 'Blue Jacket' grown in substrate contaminated with different doses of lead

Variant	Σ	CL. a/CL. b	CL. a + b/x + c
control	2.74	2.56	4.17
V <sub>1</sub>	2.43	3.43	3.26
V <sub>2</sub>	2.31	3.21	3.28
V <sub>3</sub>	1.95	3.17	3.33
V <sub>4</sub>	1.35	3.25	3.09

Research results show that in the case of plants grown in conditions of lead pollution the ratio of chlorophyll pigments and carotenoids is between 3.26 mg/g s.p. in variant V<sub>1</sub> and 3.09 mg/g s.p. in variant V<sub>4</sub>. Comparing the results obtained

in the contaminated variants with those of the control variant, it is observed that the value of the ratio  $(CL. a + CL. b)/(c + x)$  is below the value of 4.2, indicating a stress caused by both climatic conditions and lead toxicity.

## CONCLUSIONS

1. In the species of *Hyacinthus orientalis*, the highest concentrations of lead have delayed the vegetation of plants by 10 days in the group of plants from V<sub>3</sub> and by 11 days in V<sub>4</sub>, as compared to the control group.

2. The presence of lead in the substrate affects the decorative period of the plants regardless of its concentration, as it has been revealed by a reduction of the number of days by 4 invariants V<sub>1</sub>, V<sub>2</sub> and by 5 days invariants V<sub>3</sub>, V<sub>4</sub>.

3. The statistical interpretation of the results regarding the average height of the flowering rods showed very significant negative differences in variants V<sub>4</sub> and V<sub>3</sub>, and distinctly significant in variant V<sub>2</sub>, as compared to the control group.

4. Statistical interpretation of the results regarding the size of the flowers in inflorescences showed a significant positive difference in plants contaminated with high doses of lead representing the level of intervention (V<sub>3</sub>) and those contaminated with doses above this level (V<sub>4</sub>).

5. The content of chlorophyll pigments from plants grown in contaminated substrate showed major decreases as compared to the control variant, which confirms that lead pollution in the variety 'Blue Jacket' causes stress at physiological level.

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## STUDIES REGARDING THE INFLUENCE OF THE PLANTING TIMES IN THE GROWTH AND DEVELOPMENT OF *ALLIUM* 'PURPLE RAIN'

### STUDII PRIVIND INFLUENȚA EPOCILOR DE PLANTARE ASUPRA CREȘTERII ȘI DEZVOLTĂRII LA *ALLIUM* 'PURPLE RAIN'

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**Abstract.** *The study presents some results regarding the influence of different bulbs planting times of the Allium 'Purple Rain' ornamental onion cultivar. The bulbs were planted in three different times: August 2015 (V<sub>1</sub>), October 2015 (V<sub>2</sub>) and March 2016 (V<sub>3</sub>). The plants were studied during the 2016 season. They were evaluated regarding the morpho-decorative characters, like leaves length, flowering stem height, inflorescence diameter and regarding the phenological aspects, like emergence time and flowering period. The aim of this study was to identify the planting time which favors the growth and development suitable for the studied cultivar. Compared with the experiment average, the results from every variant registred statistically ensured differences, for the most of the analysed characters. The most favorable planting times were the autumn and the end of the summer (V<sub>2</sub> and V<sub>1</sub>). The spring planting time (V<sub>3</sub>) is less recommended, regarding the decrease of the plants ornamental effect and the flowering delay.*

**Key words:** *Allium 'Purple Rain'*, planting times, ornamental characters

**Rezumat.** *Lucrarea prezintă rezultate privind influența diferitelor epoci de plantare a bulbilor de la cultivarul de ceapă ornamentală Allium 'Purple Rain'. Bulbii au fost plantați în trei epoci diferite: august 2015 (V<sub>1</sub>), octombrie 2015 (V<sub>2</sub>) și martie 2016 (V<sub>3</sub>). Plantele au fost studiate pe parcursul sezonului de vegetație din anul 2016. Plantele au fost evaluate atât din punct de vedere al unor caractere morfologice ornamentale (lungimea frunzelor, înălțimea tijeii florifere, diametrul înflorescențelor), cât și din punct de vedere fenologic, analizând data pornirii în vegetație și perioada de înflorire. Scopul lucrării a fost acela de a identifica la cultivarul luat în studiu perioada de plantare care favorizează creșterea și dezvoltarea corespunzătoare a plantelor. Comparate cu media experienței, rezultatele obținute la fiecare variantă au înregistrat diferențe asigurate statistic, la majoritatea caracterelor analizate. Cele mai favorabile epoci de plantat s-au evidențiat a fi toamna și la sfârșitul verii (V<sub>2</sub> și V<sub>1</sub>). Epoca de primăvară (V<sub>3</sub>) se recomandă mai puțin, având în vedere diminuarea efectului ornamental al plantelor și întârzierea înfloririi.*

**Cuvinte cheie:** *Allium 'Purple Rain'*, epoci de plantare, caractere ornamentale

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## INTRODUCTION

*Allium* is one of the most important crops all over the world. Most for the well known vegetables, and in the last decades also for the remarkable ornamental plants, which belongs to this gender. They became very popular worldwide like garden plants, they decorate from spring to autumn, because of the various species and cultivars of this gender. The ornamental *Alliums* are very hardy plants to the pests, diseases and also to the different environmental conditions (Frish and Frisen, 2002; Harding, 2004).

Even if they are so decorative and hardy, Davis (1992), talks about very few fallowers of the ornamental *Alliums*. Thus, it became very important to carry out in the establishment of a cultivation technology, adapted to wide areas, where these plants can be grown.

In Romania there are many wildly growing *Alliums* with ornamental qualities (*A. paniculatum* L., *A. ursinum* L., *A. saxatile* M. Bieb., *A. atrovioleaceum* Boiss), some of them studied and introduced in the ornamental plant culture and in the ornamental plants catalogs of the great markets (Frish, 2015).

Concerning the climatic changes from the last years, many late frosts can affect the early emergent plants, planted from late summer or autumn. So, this study aims to establish the bulbs planting date that can ensure the right growth and development of the plants and how the spring planting can influence the plants.

The planting dates can have very important impact over the growth and development of *Allium* species and cultivars. El-Helaly and Karan (2012), shown that the late autumn or winter planting can have a worse effect over the onion seed yield.

Regarding the flower production, many bulbous plants were tested by Armitage and Laushman, in 1990, to different planting times, for trying to extend the flowering period. The results have shown that the late planting can determine a later flowering period, but the plants will not develop properly.

## MATERIAL AND METHOD

The experiment was conducted in the period August 2015 – June 2016, in the field of Floriculture discipline, from the University of Agricultural Sciences and Veterinary Medicine of Iași, Romania.

The material was represented by a cultivar of ornamental onion, *Allium* 'Purple Rain'. This cultivar decorates thru its deep purple, star shaped flowers, grouped in simple spherical umbels. The leaves are wide, grown around the stem base. The stem can grow to about 60-80 cm height. The plants have formed every year new bulbs, which can be separated and replanted. This ornamental onion, flourish between April and June (fig. 1). The *Allium* 'Purple Rain' bulbs were planted in open field, in three different planting times, as **V<sub>1</sub> – August 2015**, **V<sub>2</sub> - October 2015** and **V<sub>3</sub> - March 2016**. The bulbs wich were planted in the spring were kept during the winter in about 15<sup>0</sup>C temperature conditions. The experiment was

organized in randomized blocs design, with three replications. A plot has a surface of 3.00 m<sup>2</sup> and it was planted with 30 bulbs.

Before planting, the bulbs were disinfected with Kaptan (1%) and Topsin (0.7%) solutions and during the frosty season the plots were mulched with hay and straw.



**Fig. 1** *Allium* 'Purple Rain' – general aspect (original photo)

The plants were studied thru the biometric measurements and determinations, regarding the main morpho-decorative characters, like leaves length, flower stem height, umbel diameter and regarding the phenological aspects, like the emergence time and flowering period. The experimental data was processed using analysis of variance, which established limits of probability for each planting times, compared with the control (average of experiment for each character). The significance of the differences was assessed by taking into account the LSD test (Săulescu and Săulescu, 1967).

## RESULTS AND DISCUSSIONS

The different planting times resulted to have a very important influence over the growing and development of the ornamental onion studied cultivar. The vegetation emergence and blooming period differences, between the variants (planting times), can guide the blooming time setting.

The results obtained by Armitage and Laushman (1990), at *Allium sphaerocephalon* L., show that late planting time can determine a later flowering, with positive effect on scaling flower production, but can have negative influence upon the ornamental characters of the plants. The planting time can influence not only the emergence and the blooming date, but also the morpho-decorative characters of the plants (leaves length, stem height and inflorescence diameter).

The vegetation emergence date was different between the three planting times. The first variant (August planting time) determined the most early vegetation emergence (mid-February) and the earliest blooming period (end of April) (tab. 1). The bulbs planted in October ( $V_2$ ), registered the vegetation emergence with around 5 days later than the previous planting time (August) and flourished with around 3 days later (tab.1). The bulbs planted in March ( $V_3$ ) registered the latest vegetation emergence, with 45 days later, than  $V_1$  (August) and with 40 days later than  $V_2$  (October). The same trend was observed regarding the blooming period. The plants from  $V_3$  (March) bloomed with 19 days later than  $V_1$  (August) and with 17 days later than  $V_2$  (October) (tab. 1).

Table 1

**The influence of planting time on the emergence and blooming period**

Planting time	Emergence date	Number of days from planting to vegetation emergence	Blooming period
$V_1$ (09 August 2015)	15.02.2016	190 days	28.04-02.06.2016
$V_2$ (20 October 2015)	20.02.2016	123 days	01.05-04.06.2016
$V_3$ (20 March 2016)	01.04.2016	12 days	17.05-08.06.2016

Regarding the leaves length, the  $V_2$  (October planting time) got the highest value (49.83 cm long leaves), a very significant positive results reported to the experiment average (control). The  $V_1$  (August planting time) got shorter leaves than  $V_2$  (47 cm), the result being distinctly significant positive (tab. 2).

The  $V_3$  variant (March planting time), had the worst influence on the leaves length of the studied cultivar. The leaves achieved only an average of 25.53 cm length, the result being very significant negative, reported to the control (average of the experiment) (tab. 2).

Table 2

**The influence of planting time on the leaves length**

Variants	Leaves length (cm)		d ( $\pm$ )	Significance of differences
	Absolute value	Relative value (%)		
$V_1$	47.00	115.3	6.22	**
$V_2$	49.83	122.2	9.06	***
$V_3$	25.53	62.5	-15.28	000
<b>Average (control)</b>	<b>40.78</b>	<b>100.0</b>	-	-

LSD 5% = 2.58 cm

LSD 1% = 4.25 cm

LSD 0.1% = 7.95 cm

The next studied character was the flower stem height. This one registered a decreasing trend of the values, from  $V_2$  to  $V_3$ . So,  $V_1$  (August planting time), achieved in average a 49.33 cm long stem, not significant result reported to the control (experiment average),  $V_2$  (October planting time), registered an average of 54.17 cm long stem, the result being positive significant and  $V_3$  (March planting time) got the lowest result, 33.33 cm long stem, a distinctly significant negative result, reported to the experiment average (control) (tab. 3).

Table 3

**The influence of planting time on the  
flower stem height**

Variants	Flower stem height (cm)		d ( $\pm$ )	Significance of differences
	Absolute value	Relative value (%)		
$V_1$	49.33	108.2	3.72	ns
$V_2$	54.17	118.8	8.56	*
$V_3$	33.33	73.1	-12.28	oo
<b>Average (control)</b>	<b>45.61</b>	<b>100.0</b>	-	-

LSD 5% = 6.26 cm

LSD 1% = 10.36 cm

LSD 0.1% = 19.39 cm

Concerning the inflorescence diameter, the planting time has determined very large differences between the variants. The most valuable results were obtained in case of  $V_1$  (August planting time), an average of 24.33 cm diameter, a distinctly positive significant result, reported to the control (experiment average) (tab. 4).

Table 4

**The influence of planting time on the  
inflorescences diameter**

Variants	Inflorescences diameter (cm)		d ( $\pm$ )	Significance of differences
	Absolute value	Relative value (%)		
$V_1$	24.33	111.2	2.44	**
$V_2$	24.00	109.6	2.11	*
$V_3$	17.33	79.2	-4.56	ooo
<b>Average (control)</b>	<b>21.89</b>	<b>100.0</b>	-	-

LSD 5% = 1.28 cm

LSD 1% = 2.12 cm

LSD 0.1% = 3.97 cm

The October planting time ( $V_2$ ), registered a significant value reported to the control (experiment average), but the difference to  $V_1$  (24 cm) was very low. In case of  $V_3$  (March planting time), the registered values were very significant negatives reported to the experiment average, the inflorescences got only 17.33 cm diameter, very low value, than the other two variants.

## CONCLUSIONS

1. The growth and development of *Allium* 'Purple Rain' cultivar varied in large limits between the three planting times. Both vegetation emergence and blooming period were earlier in case of V<sub>1</sub> (August planting time) and V<sub>2</sub> (October planting time) and later for the V<sub>3</sub> (March planting time).

3. Regarding the leaves length, V<sub>2</sub> (October planting time) got the highest value (49.83 cm long leaves), V<sub>1</sub> (August planting time) got shorter leaves than V<sub>2</sub>, (47 cm) and V<sub>3</sub> variant (March planting time) got only an average of 25.53 cm length leaves.

4. For the flower stem height, V<sub>1</sub> (August planting time) got in average 49.33 cm long stem, V<sub>2</sub> (October planting time) registered 54.17 cm and V<sub>3</sub> (March planting time) got the lowest result, 33.33 cm long stem.

5. The inflorescence diameter was also influenced by the planting time, as, V<sub>1</sub> (August planting time) got an average of 24.33 cm, the October planting time (V<sub>2</sub>) registered a 24 cm diameter and V<sub>3</sub> (March planting time) got only 17.33 cm diameter inflorescence.

6. Based on the results obtained in this study, it can be recommended for the cultivar *Allium* 'Purple Rain', in the N-E Romania, to be planted in late summer or autumn, without damages because of the late frosts in the spring. The spring planting can determine the faulty growth and development of the plants, but can extend in the same time the blooming period.

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## THE AIR FLOW INFLUENCE ON THE UNIFORMITY OF DISPERSION FOR A VINEYARD SPRAYING MACHINE

### INFLUENȚA DEBITULUI DE AER ASUPRA UNIFORMITĂȚII DE DISPERSIE PENTRU O MAȘINĂ DE STROPIT ÎN PLANTAȚII DE VIȚĂ DE VIE

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**Abstract.** Control of diseases and pests from vineyards is one of the most important technological links, because it makes the health of the culture, i.e. production of grapes. These treatments are applied with special machines, equipped with fans that can control drift droplets of phytosanitary substance, transporting them on target. For this reason, the determination of dispersion uniformity of the spray nozzles mounted on the machine is very important. In order to determine the influence of TARAL 200 PITON TURBO spraying machine fan airflow on the LECHLER's IDK 120-02 air absorption nozzles uniformity of dispersion, it was designed and built a vertical test stand with 18 troughs for collecting the substance from different heights. Analyzing the results obtained under laboratory conditions, it was found that the spraying machine fan air flow influences the uniformity of dispersion of the air absorption nozzles. The best uniformity has been achieved at the speed of 1400 rpm, being 90.20% for the pressure of 0.8 MPa.

**Key words:** air absorption nozzles, sprayers, uniformity of dispersion, axial fans

**Rezumat.** Combaterea bolilor și dăunătorilor din plantațiile viticole este una dintre cele mai importante verigi tehnologice, deoarece condiționează starea de sănătate a culturii, respectiv producția de struguri. Tratamentele sunt aplicate cu mașini speciale, dotate cu ventilatoare care pot controla deriva picăturilor de substanță fitosanitară, transportându-le pe țintă. Din acest motiv determinarea uniformității de dispersie a duzelor montate pe mașina de stropit este foarte importantă. Pentru a determina influența debitului de aer a ventilatorului mașinii de stropit TARAL 200 PITON TURBO asupra uniformității de dispersie a duzelor cu absorbție de aer IDK 120-02 LECHLER, s-a proiectat și realizat un stand de probe vertical conceput cu 18 jgheaburi pentru colectarea substanței de la diferite înălțimi. Analizând rezultatele obținute în condiții de laborator s-a constatat că debitul de aer al ventilatorului influențează uniformitatea de dispersie a duzelor cu absorbție de aer. Uniformitatea cea mai bună s-a obținut la turația de 1400 rot/min, fiind de 90,20% pentru presiunea de 0,8 MPa.

**Cuvinte cheie:** duze cu absorbție aer, mașină de stropi, uniformitate de dispersie, ventilator axial

## INTRODUCTION

Plant protection treatments applied in a timely and effective manner ensure a high production for vine plantations. Otherwise, production suffers very large

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losses, can reach over 30%, or may even be compromised. For this reason, the fight against pests and diseases represents a very important technological component, without which the production would not be sure and steady from year to year (Kamousantas *et al.*, 2000; Comeanu, 2003; Tomoiagă, 2013; Diaconu *et al.*, 2016).

For the application of phytosanitary treatments are used, in most cases, spraying machines with mechanical dispersion and pneumatic transport of the droplets. Optimal use of the air flow generated by the fan of these machines can improve the distribution of the droplets and the efficiency of the treatment on the surfaces of the plant (Viret *et al.*, 2003; Pergher and Petris, 2008). The incorrect air flow use, for a too strong or too weak jet, can produce irregularity of the quantity of liquid pesticide sprayed on the leaves surface (in some areas the amount of pesticide sprayed is in excess, and appear the leakage phenomenon, and in other areas, the surface sprayed is very small and the treatment is ineffective), so uneven distribution in the canopy, and on the other hand there is a risk of pollution increased environmental and in particular of the soil (Doruchowski *et al.*, 1997; Panneton *et al.*, 2005).

In order to determine the influence of the spraying machines fan air flow, in the context of this work, it has been designed and made a vertical stand and experimental research have been carried out in laboratory conditions.

## MATERIAL AND METHOD

In the Horticultural Laboratory Machinery of the University of Agricultural Sciences and Veterinary Medicine "Ion Ionescu de la Brad" from Iasi was designed and made a stand for the determination of the dispersion uniformity, at different working heights, using an universal machine for vines plantations pests and diseases combating with mechanical dispersion and pneumatic droplets transport, model TARAL 200 PITON TURBO. The sprayer was fitted with air absorption flat jet type IDK 120-02 nozzles, produced by LECHLER company. These nozzles prevent drift up to 90%, due to the fact that the spraying ramps generates large air-filled droplets, broken into small drops at their contact with the leaves surface the and improves the process of work, by covering a large area with liquid pesticide.

The stand is made of a vertical panel made of polycarbonate, with a length of 2350 mm and a width of 830 mm, on which were mounted the 18 inclined troughs, made of galvanized sheet. To determine the distribution uniformity of the substance dispersed throughout the panel, at the level of each trough there were mounted containers (fig.1). The substance collected for a minute, at the level of each trough in the containers was measured with a graduated cylinder.

The troughs were numbered starting at the base of the panel, from 1 to 18, trough number 1 being at the height of 300 mm from the ground, and number 18 at 2510 mm, the troughs being mounted on panel at 130 mm distance from each other.

The experimental tests were carried out in laboratory conditions, for different working pressures (0.2; 0.4; 0.6; 0.8; 1.0; 1.2 and 1.4 MPa) and speeds of the fan of 800, 1100 and 1400 rev/min. The height of the panels layout above ground was 300 mm, and the distance from the panel to the spraying machine axis of 1500 mm.

The dispersion uniformity of the pesticide liquid  $U_d$  (%), on the working height of the machine, must be, according to the data established in the literature, higher than 85% and is calculated with the relationship:

$$U_d = \left[ 1 - \frac{\sqrt{\frac{\sum_{i=1}^{i=n} (q_i - q_m)^2}{n(n-1)}}}{q_m} \right] * 100 \text{ (%),}$$

in which:  $q_i$  – the amount of liquid collected from each trough;  $q_m$  – the average amount calculated for all the collecting troughs;  $n$  – the number of troughs.



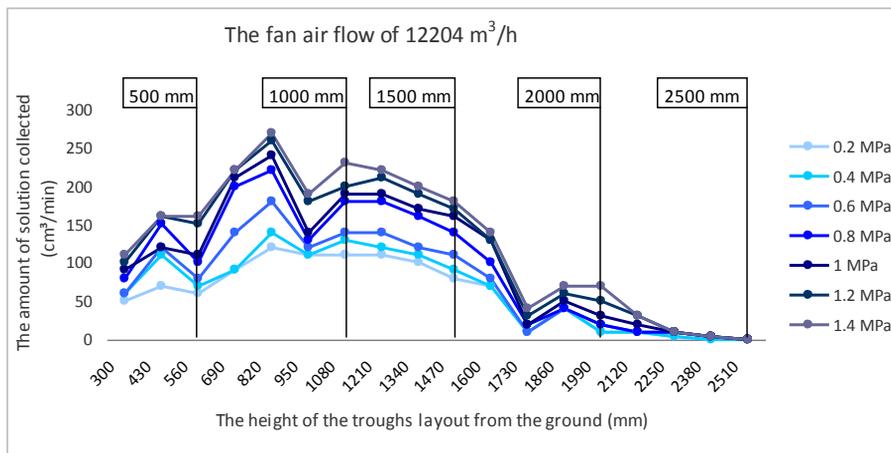
Fig. 1 Stand for determining the distribution uniformity of the dispersed pesticide fluid

## RESULTS AND DISCUSSIONS

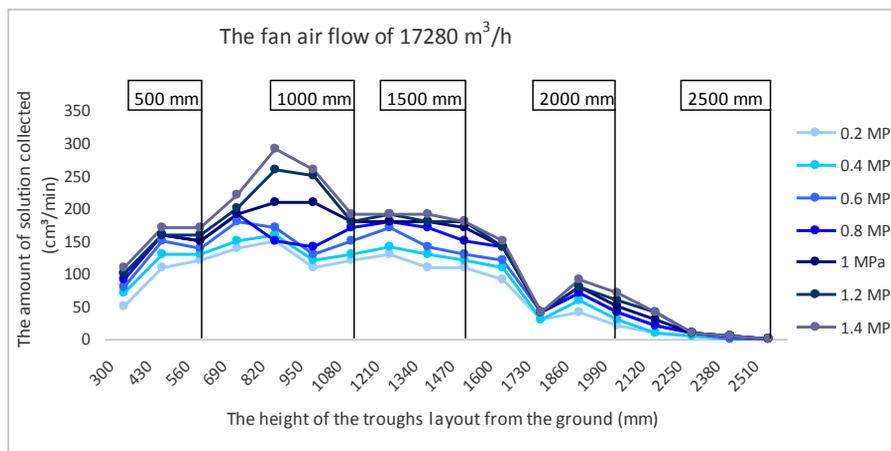
For the three speeds of the fan measurements have been made with an anemometer, in order to determine the velocity of the air flow discharge and air flow generated. The average speed measured at the level of the axial fan discharge section, respectively on the nozzle direction, was of 8.27 m/s for the speed of 800 rot/min, 11.71 m/s for 1100 rot/min and 14.32 m/s for 1400 rot/min. For these conditions it was calculated the fan air flow, after it was also determined the area of the discharge

section, being of  $0.41 \text{ m}^2$ , namely  $12204 \text{ m}^3/\text{h}$  at  $800 \text{ rot}/\text{min}$ ,  $17280 \text{ m}^3/\text{h}$  at  $1100 \text{ rot}/\text{min}$  and  $21132 \text{ m}^3/\text{h}$  at  $1400 \text{ rot}/\text{min}$ .

For all three flow rates of the airflow generated by the fan of the spraying machine is was obtained a greater amount of the liquid collected in the trough at the height level of  $820 \text{ mm}$  from the ground (fig. 2, 3 and 4).



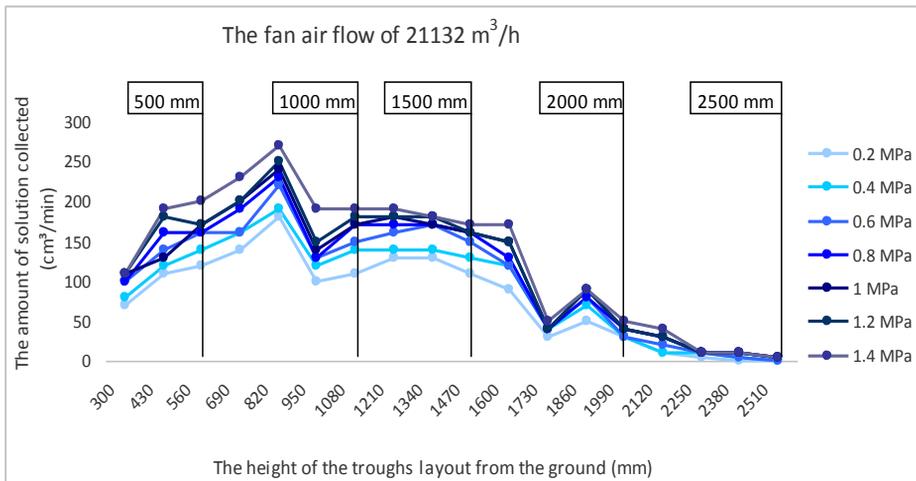
**Fig. 2** The amount of liquid collected at the level of each trough for the fan air flow of  $12204 \text{ m}^3/\text{h}$



**Fig. 3** The amount of liquid collected at the level of each trough for the fan air flow of  $17280 \text{ m}^3/\text{h}$

The amount of liquid collected by the troughs decreases from the height of the of  $1080 \text{ mm}$  from the ground to the upper part of the stand of  $1600 \text{ mm}$  and from  $560 \text{ mm}$  to the lower part of the stand at  $300 \text{ mm}$ . The decreasing of the amount of liquid collected in the  $1000\text{-}1500 \text{ mm}$  range is more pronounced in the air flow case of  $12204 \text{ m}^3/\text{h}$  than in the air flow case of  $17280$  and  $21132 \text{ m}^3/\text{h}$ . In

the upper part of the panel, from 1730 mm to 2510 mm, the amount of fluid collected is smaller and comes from the drift of the droplets (fig. 2, 3 and 4).



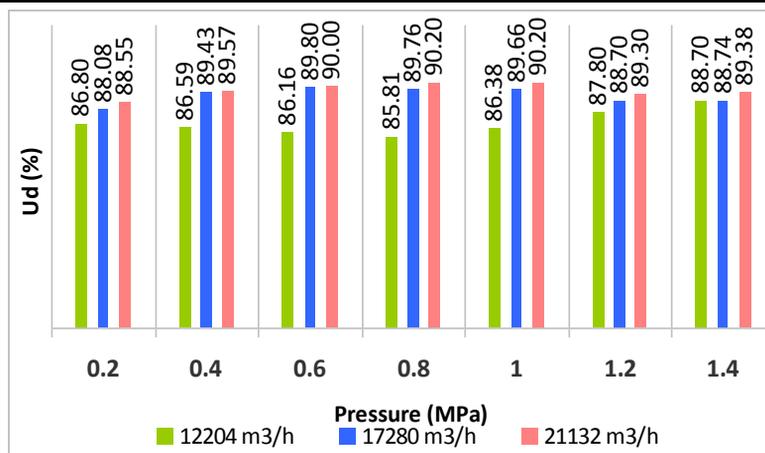
**Fig. 4** The amount of liquid collected at the level of each trough for the fan air flow of 21132 m<sup>3</sup>/h

By increasing the pressure it is noted that for all air flow rates generated by the spraying machine's fan the amount of liquid collected at the level of each trough increases, due to the larger amount given by the nozzles (fig. 2, 3 and 4).

To determine the uniformity of the liquid dispersion  $U_d$  (%) on the spraying machine working height, there have been removed from the calculation troughs from 15 to 18, i.e. those positioned above the height of 2000 mm, because above this quota, the vegetation of the plantation is reduced, and the amount of dispersed solution values are insignificant.

It is noted that the obtained values of the uniformity constant were greater than 85% in all the variants tested. The fan air flow of 21132 m<sup>3</sup>/h has achieved the best uniformity, obtaining the following values: 90.20% for pressures of 0.8 and 1.0 MPa, 90.00% for 0.6 MPa, 89.57% for 0.4 MPa and 89.55% for 0.2 MPa. For the fan air flow of 17280 m<sup>3</sup>/h there were obtained high values of uniformity to the following pressures: 89.80% for the pressure of 0.6 MPa, 89.76% for 0.8 MPa and 89.66% for 1.0 MPa. For the fan air flow of 12204 m<sup>3</sup>/h it was obtained a lower uniformity than in the other cases, the best uniformity being 88.70% obtained at the pressure of 1.4 MPa, followed by the pressure obtained at 1.2 MPa with 87.80% and then by 0.2 MPa with 86.80% (fig.5).

So, by increasing the air flow generated by the spraying machine fan, you can get a better treatment uniformity (fig.5).



**Fig. 5** Liquid dispersion uniformity on the working height of spraying machine for different axial fan air flow rates and working

## CONCLUSIONS

The fan air flow of the TARAL 200 PITON TURBO spraying machine affects the dispersion uniformity of the air absorption nozzles. The best uniformity was obtained at the flow rate of air 21132 m<sup>3</sup>/h, being of 90.20% for the 0.8 and 1.0 MPa pressures.

*Acknowledgments:* This paper was published under the frame of European Social Fund, Human Resources Development Operational Programme 2007-2013, project no. POSDRU/159/1.5/S/ 132765.

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## RESEARCHES ON OF GRAPE MOTH THE EVOLUTION (*LOBESIA BOTRANA* - DEN ET SCHIFF) OF DEALUL BUJORULUI THE VINEYARD, IN THE CONTEXT OF CURRENT CLIMATE CHANGE

CERCETĂRI PRIVIND EVOLUȚIA MOLIEI STRUGURILOR (*LOBESIA BOTRANA* – DEN ET SCHIFF) ÎN PLANTAȚILE VITICOLE DIN PODGORIA DEALUL BUJORULUI, ÎN CONTEXTEL ACTUALELOR SCHIMBĂRI CLIMATICE

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**Abstract.** *The paper presents analysis of research conducted at The Research and Development for Winegrowing and Winemaking in the period 2005-2015 Bujoru climate factors (temperature, precipitation, humidity, etc.) that influence the biology and evolution of grape moth (Lobesia botrana - Den et Schiff) in plantations vineyards south of Moldova. Lately there was a deviation of climatic factors on the annual average (high temperature, frequency absolute minimum temperature below the freezing of the vine, increased the frequency of droughts, desertification) that can cause changes in the biological and behavioral the principal harmful vine vineyard Dealul Bujorului (grape moth), with consequences difficult to assess the integrity of the ecosystem vineyard.*

**Key words:** grapevine, grape moth, pheromone traps

**Rezumat.** *Lucrarea prezintă analiza cercetărilor efectuate la Stațiunea de Cercetare și Dezvoltare pentru Viticultură și Vinificație Bujoru în perioada 2005-2015 a factorilor climatici (temperatură, precipitații, umiditate etc.) care influențează biologia și evoluția moliei strugurilor (Lobesia botrana – Den et Schiff) în plantațiile viticole sudul Moldovei. În ultima perioadă s-a observat o abatere a factorilor climatici de la media multianuală (temperaturi ridicate, frecvența temperaturilor minime absolute sub pragul de îngheț al viței de vie, accentuarea și frecvența secetelor, aridizarea) care pot provoca schimbări de ordin biologic și comportamental asupra principalului dăunător al viței de vie din podgoria Dealul Bujorului (molia strugurilor), cu consecințe greu de evaluat pentru integritatea ecosistemului viticol.*

**Cuvinte cheie:** viță de vie, molia strugurilor, capcane feromoni

### INTRODUCTION

Grape moth (*Lobesia botrana* Den et Schiff) is the main pest of vine vineyards of SE Moldova. Product losses in some years are between 30-50% of grape production. Lately there has been a significant reduction in grape moth attack produced from plantations with vines vineyard Dealul Bujorului.

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Knowledge of pest biology and ecology of particular importance to the warning and applying the treatments to combat the economic damage threshold is exceeded (PED) 100 butterflies males / trap / week (Mirică *et al.*, 1976; Simion Cristina, 2003; Tabaranu *et al.*, 2005; Tabaranu *et al.*, 2007; Teodorescu *et al.*, 2003).

## MATERIAL AND METHOD

The research was conducted during 2005-2015 in vineyards from SCDVV Bujoru. Over ten years have made observations varieties (Merlot, Black Feteasca, Aligoté) on the evolution of the main pest of vine (grape moth - *Lobesia botrana*) in the current climate changes and changes in microclimate conditions due to the greenhouse effect.

To establish the population of when treatment applications in plots with vines were installed for warning and monitoring by 3 traps / ha with synthetic sex attractant type Atrabot (fig. 1).



**Fig. 1** Sex pheromone trap type Atrabot

Registration butterfly capture was done weekly and based on the results was drawn curve flying pest. It materializes the flight, the maximum flight activity, the succession of generations, etc. data on which were established for the submission of clutch moments, the emergence of larvae, warning and appropriate treatments. Capsules with synthetic sex pheromone and valves adhesive were changed after 6 weeks of use.

## RESULTS AND DISCUSSIONS

The average air temperature (average for the last 10 years) is 11.5°C identical to normal temperature, with a maximum during the growing season of 24.2°C (July) and a minimum of -1.3°C (January). Temperatures remain low until late spring in April and sometimes May. There is a slight increase in air temperature in the range 2005-2015 (table 1). In the analyzed period in June and July recorded maximum temperatures frequently exceeding 32 - 35°C in most years in the study with an absolute maximum temperature of 41.5°C / 07.VII.2012.

The amount of monthly precipitation (average for the last 10 years) is 490.1mm, 455.9 mm normal girl (tab. 2). Rainfall distribution is heterogeneous, characterized by long periods legally deficient rainfall (droughts), falling short periods of precipitation. In the period analyzed the behavior and potential vine pest attack was highly influenced by climatic conditions (temperature, humidity etc.).

Temperature has a major influence on the development of grape moth, such as: high temperature > 34-36°C are lethal for the eggs and adults, eggs and larvae resistant to temperatures between 0°C and 10°C and to 23°C crisalidele.

Following research on the development of grape moth in the period 2005-2015 showed a slight downward trend in the pest population level from 2008 and 2009 (tab. 3). Adverse weather conditions have led to the development of pest braking biological activities (mating, laying and larval emergence). Flight dynamics of male butterflies had a high frequency, the average number of butterflies males / trap / week exceeding the PED 100 catches in 2005, 2006, 2007 and 2008. In subsequent years (2010, 2011, 2012, 2013, 2014 and 2015), due to high temperatures that exceeded repeatedly threshold  $O_t = 26.7^\circ\text{C}$  (optimum heat during the egg-laying and hatching deposition) more eggs were dehydrated (Ur% under 50%), no more and finally hatched the number of population decreased and the attack was weak.

In 2005, 2006, 2007 and 2008 there was a sharp rise in the average number of butterflies caught exceeding the economic damage threshold (PED) 100 butterfly males / trap / week all three generations, except a- generation II of 2008 (table 3). For integrated control grape moth they were recommended specific treatments with plant protection products and mass capture of male butterflies using synthetic sex pheromone traps ATRABOT 7-9 type traps / ha for every generation.

In the years 2010-2015 the climatic conditions of high temperatures and drought prevailing atmospheric, caused dehydration hatching eggs and larval mortality, significantly reducing the insect population Economic damage threshold (PED) 100 butterfly males / trap / week all three generations.

Table 1

The average air temperature in the period 2005-2015

No.	Year	Average monthly (°C)												Average
		I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	
1	2005	1.3	-0.6	4.6	11.7	18.2	20.8	24.3	23.3	19.1	11.7	5.4	2.5	11.9
2	2006	-4.3	-0.6	4.6	12.3	17.5	22.3	24.2	23.5	18.1	12.5	7.6	2.9	11.7
3	2007	4.3	3.1	8.3	12.1	21.2	25.5	28.1	25.4	17.4	12.1	4.1	0.4	13.5
4	2008	-0.7	3.4	9.1	13.0	17.9	23.1	24.0	25.8	15.6	11.3	5.5	2.2	12.5
5	2009	-0.6	1.6	4.4	11.1	16.5	21.0	23.8	21.9	17.1	11.6	6.2	2.1	11.4
6	2010	-4.6	-0.1	4.5	10.8	16.8	20.4	22.8	24.0	16.1	7.8	10.1	-1.7	10.6
7	2011	-2.9	-2.9	3.7	9.5	16.1	20.1	24.2	23.3	18.5	8.5	2.0	2.3	10.2
8	2012	-2.3	8.0	4.4	12.9	17.9	20.1	28.0	26.4	20.7	14.6	6.7	-1.9	11.6
9	2013	-2.2	1.8	3.6	12.7	18.5	20.9	21.5	22.2	15.0	10.7	8.3	0.1	11.1
10	2014	-1.3	-0.6	8.1	11.0	15.6	20.7	22.1	22.3	17.0	10.0	4.3	0.0	10.8
11	2015	-1.2	1.2	5.2	10.4	17.0	20.3	23.7	23.0	19.0	9.5	6.6	1.6	11.4
<b>Average</b>		<b>-1.3</b>	<b>-0.2</b>	<b>5.5</b>	<b>11.6</b>	<b>17.6</b>	<b>21.4</b>	<b>24.2</b>	<b>23.7</b>	<b>17.6</b>	<b>10.9</b>	<b>6.1</b>	<b>0.9</b>	<b>11.5</b>
<b>Normal</b>		<b>-1.2</b>	<b>0.1</b>	<b>5.2</b>	<b>11.7</b>	<b>18.1</b>	<b>21.8</b>	<b>24.1</b>	<b>23.1</b>	<b>17.5</b>	<b>11.4</b>	<b>5.2</b>	<b>0.4</b>	<b>11.5</b>

Table 2

The average yearly precipitates (mm) in 2005-2015

No.	Year	Precipitations (mm)												Amount
		I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	
1	2005	14.7	31.4	25.7	18.4	59.2	74.8	98.6	50.8	14.5	15.7	66.9	26.4	497.1
2	2006	27.1	10.8	40.6	54.2	40.0	77.7	31.8	92.1	34.4	7.7	4.7	9.4	430.5
3	2007	51.5	23.1	43.1	14.9	31.3	96.4	1.0	57.3	36.5	59.4	60.1	67.3	541.9
4	2008	14.1	1.3	38.9	37.7	60.6	38.5	55.5	9.8	49.8	21.6	15.4	57.4	401.1
5	2009	46.8	21.2	40.6	12.8	28.4	86.8	30.9	13.8	56.0	35.2	14.4	32.6	419.5
6	2010	12.8	41.0	11.0	26.6	103.2	127.6	101.8	40.2	20.2	76.4	21.8	51.6	634.2
7	2011	15.2	13.4	5.4	53.6	32.2	45.2	93.4	28.0	5.2	28.0	0.2	9.8	329.6
8	2012	39.3	25.4	9.2	18.6	115.8	13.8	27.1	23.1	24.6	42.0	6.8	102.3	448.0
9	2013	34.4	35.2	39.4	32.0	89.6	90.6	18.6	81.6	203.6	45.2	38.8	4.1	713.1
10	2014	16.8	4.8	29.2	72.4	4.1	38.5	84.6	55.0	3.4	44.4	53.4	43.6	450.2
11	2015	27.0	35.6	59.0	32.0	13.8	53.0	23.0	74.4	22.0	74.0	111.4	1.2	526.4
<b>Average</b>		<b>27.2</b>	<b>22.1</b>	<b>31.1</b>	<b>33.9</b>	<b>52.6</b>	<b>67.5</b>	<b>51.5</b>	<b>47.8</b>	<b>42.7</b>	<b>40.9</b>	<b>35.8</b>	<b>36.9</b>	<b>490.1</b>
<b>Normal</b>		<b>21.5</b>	<b>19.2</b>	<b>26.7</b>	<b>36.5</b>	<b>48.0</b>	<b>72.1</b>	<b>50.5</b>	<b>48.6</b>	<b>40.3</b>	<b>32.4</b>	<b>29.5</b>	<b>30.5</b>	<b>455.9</b>

Table 3

Evolution grape moth (*Lobesia botrana* - Den et Schiff) Peony Hill vineyard in the period 2005-2015

Number	Year	The maximum curve flight G a I-a		The maximum curve flight G a II-a		The maximum curve flight G a III-a		The degree of infection %	Observations
		Date	Number of exemplary	Date	Number of exemplary	Date	Number of exemplary		
1	2005	6.V	219	7.VII	243	28.VIII	302	10-20 %	medium attack
2	2006	19.V	206	14.VII	351	11.VIII	395	20-53 %	strong attack
3	2007	9.V	640	20.VI	585	10.VIII	300	20-50 %	strong attack
4	2008	7.V	215	25.VI	73	13.VIII	179	10-20 %	medium attack
5	2009	29.V	67	17.VI	208	20.VIII	177	10-20 %	medium attack
6	2010	4.V	87	22.VI	120	10.VIII	77	5-10 %	weak attack
7	2011	18.V	64	22.VI	47	10.VIII	7	5-10 %	weak attack
8	2012	11.V	16	15.VI	63	06.VIII	16	5 %	weak attack
9	2013	8.V	3	12.VI	4	17.VII	29	5 %	weak attack
10	2014	4.VI	8	9.VII	5	20.VIII	12	5 %	weak attack
11	2015	7.V	16	18.VI	10	14.VIII	42	5 %	weak attack

## CONCLUSIONS

1. Under water stress, high temperatures etc., changes in grape moth vine plantations is much affected as greatly diminished frequency (F) and the degree of attack (GA%);

2. The climatic conditions in the period 2010-2015 were generally unfavorable grape moth and visibly influenced generations attacks (G I , G-II and G-III);

3. Changes biologically determined conditions due to climate change are visible G II and G III when larvae overlap in most cases with pupated and adult and larval mortality causes temperatures above 32<sup>0</sup>C young dehydration eggs.

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## CONTRIBUTIONS TO KNOWLEDGE THE STRUCTURE, DYNAMICS AND ABUNDANCE OF THE COLLECTED ENTOMOFAUNA FROM WHEAT CROPS

### CONTRIBUȚII LA CUNOAȘTEREA STRUCTURII, DINAMICII ȘI ABUNDENȚEI ENTOMOFAUNEI COLECTATE DIN CULTURILE DE GRÂU

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**Abstract:** *In the present conditions, increased the production of cereals and especially of production wheat is possible only through an intensive culture, by applying the modern technology to work in accordance with the requirements of cultivated varieties, irrigation, prevention and integrated control of diseases and pest, weed through the mechanization, to ensure on time execution of work under optimal conditions. The research on the knowledge of beetles species, dynamics and their abundance were made using soil traps type Barber, in three experimental stationary with wheat. The observations were made in 2015, the material collecting was done from May to July. The harvesting of the material was made at intervals of 14 days, all being done in a number of four harvests. The species most frequently collected were: *Pentodom idiota*, *Epicometis hirta*, *Opatrum sabulosum*, *Phyllotreta atra*, *Phyllotreta nemorum* and *Tanymecus dilaticollis*.*

**Key words:** Carabids, plum, useful entomofauna

**Rezumat:** *În condițiile actuale, creșterea producției de cereale și mai ales a producției de grâu, este posibilă numai printr-un sistem de cultură intensivă, prin aplicarea tehnologiilor moderne de lucrare, în concordanță cu cerințele soiurilor cultivate, irigații, prevenirea și combaterea integrată a bolilor și dăunătorilor, a buruienilor, prin mecanizare care să asigure executarea lucrărilor la timp și în condiții optime. Cercetările cu privire la cunoașterea speciilor de coleoptere, a dinamicii și a abundenței acestora s-au realizat cu ajutorul capcanelor de sol tip Barber, în trei loturi experimentale cu grâu. Observațiile au fost efectuate în anul 2015, colectarea materialului s-a făcut din luna mai până în luna iulie. Recoltarea materialului s-a făcut la intervale 14 zile, în total efectuându-se un număr de 4 recoltări. Speciile cele mai frecvent colectate au fost: *Pentodom idiota*, *Epicometis hirta*, *Opatrum sabulosum*, *Phyllotreta atra*, *Phyllotreta nemorum* și *Tanymecus dilaticollis*.*

**Cuvinte cheie:** carabide, prun, entomofauna utilă.

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## INTRODUCTION

*Coleoptera* are the most numerous species, not only of insects, but throughout the world creatures. All are characterized by converting previous wings sheath that covers very well the rest abdomen and defend it. They are spread all terrestrial, aquatic and cave adopted in part to supply all regions. A large number are predatory entomophagous very useful to man, the vast majority are phytophagous, so harmful. Some have adapted to parasitism (Popov, 2003, Tălmăciu and Tălmăciu, 2005).

There are also species of predatory beetles that can populate ecosystems such as tree species of beetles *Calosoma*, *Pterostichus*, *Brachynus* etc (Tălmăciu *et al*, 2001; Tălmăciu *et al*, 2008). Ideally, some species (predatory and harmful) to find a balance so harmful species to cause damage (Panin, 1951). The present study is a comparative study of beetles found in four corn lots with four different types of plants preceding.

## MATERIAL AND METHOD

For gathering the material have been used Barber soil traps. They have been placed in wheat area Tișița Vrancea, each 6 traps in each variant (Tălmăciu *et al*, 2008), namely:

- Untreated wheat for consumption during the growing season, only to seed;
- Wheat for consumption on treatments that were applied during the growing season against the pathogens and pests;
- Wheat seed which has been applied technology to do so.

The traps have been placed in two rows, each row at distances 3 each between 6 and 8 m and between two rows distance of about 10 m. For capturing and killing species it has been gathered used a solution of concentration of 5.4% formalin into the item (Tălmăciu and Tălmăciu, 2005).

The traps have worked from May until late June. The harvesting of the material gathered has been made at intervals of between 10 and 20 days during 2015. Each harvest the species gathered from each of variant and traps have been put in the gauze, previously removing the the crop residues, soil particles or other contaminants. Such evidence is in this way was then tagged on the label specifying:

- the date of collection;
- variant;
- the trap of number.

The material was then brought into the laboratory after he was washed in a stream of water, they were selected and identified species of beetles. They counted the specimens gathered for each trap, the variants and species. The determination was made by using the German determinant (Reitter, 1908) or the Manual determinator (Rogojanu and Perju, 1979) with other colaborators and the internet to comparing the different pictures species.

## RESULTS AND DISCUSSIONS

In total, the 3 variants were gathered 2610 samples belonging to 105 species (taxons). On variants the situation is as follows (tab. 1, fig. 1):

- For consumption wheat untreated have been collected 845 beetles belonging to a total of 55 species;

- Wheat consumption treated 838 specimens of beetles have been collected in a total belonging to a 65 number of species (taxons);

- Wheat for seed of has been collected 927 beetles total, belonging to a number of 72 species (taxons):

A number of 29 species gathered were common to in the 3 experimental variants. These include: *Amara aenea*, *Anthicus antherinus*, *Anthicus floralis*, *Anthicus humeralis*, *Blaps mortisaga*, *Cartodere ruficollis*, *Coccinella 7 punctata*, *Colodera nigrata*, *Conosoma bipunctata*, *Dermestes lanarius*, *Drasterius bimaculatus*, *Emphilus glaber*, *Epicometis hirta*, *Formicomus pedestris*, *Gryllus campestris*, *Haplothrips tritici*, *Harpalus distinguendus*, *Hypnoidus pulchellus*, *Opatrum sabulosum*, *Oxyporus rufus*, *Pedinus femoralis*, *Pentodon idiota*, *Pleurophorus caesus*, *Pterostichus marginalis*, *Pteryngium crenatum*, *Pyrrhocoris apterus*, *Tachyporus ruficollis*, *Tanymecus palliatus* and *Tanymecus dilaticollis*.

Table 1

The structure and abundace of the collected entomofauna from the wheat crops

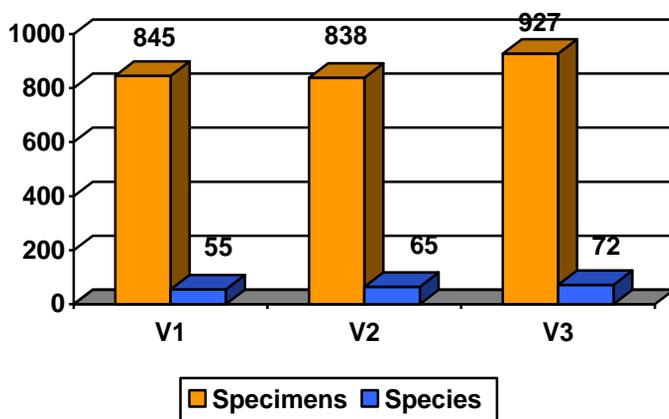
No.	The scientific name	Variants			Total
		1	2	3	
1	<i>Agriotes lineatus</i>	-	9	23	32
2	<i>Amara aenea</i>	3	3	1	7
3	<i>Amara eurynota</i>	-	2	1	3
4	<i>Anisodactylus binotatus</i>	-	2	1	3
5	<i>Anisoplia segetum</i>	1			1
6	<i>Anobium punctatum</i>	-	4	1	5
7	<i>Anthicus antherinus</i>	17	48	18	83
8	<i>Anthicus floralis</i>	5	4	32	41
9	<i>Anthicus gracilis</i>	10	1		11
10	<i>Anthicus humeralis</i>	11	2	4	17
11	<i>Anthicus humilis</i>	5	-	1	6
12	<i>Anthicus quadriguttatus</i>	-	-	2	2
13	<i>Aphodius fimetarius</i>	-	2		2
14	<i>Aphthona euphorbia</i>	10	-	2	12

15	<i>Armadilludium vulgare</i>		18	26	44
16	<i>Astenus filiformis</i>	-	1		1
17	<i>Atomaria fuscicollis</i>	-	-	1	1
18	<i>Bidessus geminus</i>	1	-	-	1
19	<i>Blaps mortisaga</i>	1	2	1	4
20	<i>Brachynus explodens</i>	-	2	4	6
21	<i>Broscus cephalotes</i>	1			1
22	<i>Calathus fuscipes</i>	-	-	3	3
23	<i>Callistus lunatus</i>		1		1
24	<i>Calosoma inquisitor</i>		1		1
25	<i>Cantharis fusca</i>	-	-	2	2
26	<i>Cartodere ruficollis</i>	4	1	1	6
27	<i>Cassida nobilis</i>	2	-		2
28	<i>Cephus pygmaeus</i>	-	1		1
29	<i>Cercyon lateralis</i>	3	-	4	7
30	<i>Cerylon ferrugineum</i>	-	-	1	1
31	<i>Cetonia aurata</i>	-	2	1	3
32	<i>Ceutorhynchus punctiger</i>	1	1	-	2
33	<i>Chrysopa perla</i>	-	-	2	2
34	<i>Coccinella 5 punctata</i>	1			1
35	<i>Coccinella 7 punctata</i>	12	5	1	18
36	<i>Coccinulla quatuordecimpustulata sinensis</i>	2			2
37	<i>Colodera nigrata</i>	17	29	7	53
38	<i>Conosoma bipunctata</i>	41	38	200	279
39	<i>Corticaria longicornis</i>	4	-	2	6
40	<i>Cryptophagus dentatus</i>	20	-	2	22
41	<i>Cryptophagus dorsalis</i>	-	-	1	1
42	<i>Cyrticus quisquilius</i>	1			1
43	<i>Dermestes lanarius</i>	60	22	25	107
44	<i>Drasterius bimaculatus</i>	10	102	34	146
45	<i>Emphilus glaber</i>	1	1	2	4
46	<i>Epicometis hirta</i>	67	91	29	187
47	<i>Eurygaster integriceps</i>	2			2
48	<i>Forficula auricularia</i>	-	-	1	1
49	<i>Formicomus pedestris</i>	35	24	41	100
50	<i>Gryllus campestris</i>	23	5	15	43
51	<i>Haplothrips tritici</i>	144	15	6	165
52	<i>Harpalus cupreus</i>	-	-	1	1
53	<i>Harpalus distinguendus</i>	18	9	6	33
54	<i>Harpalus smaragninus</i>	-	-	3	3
55	<i>Harpalus spp.</i>	-	-	1	1

56	<i>Harpalus tardus</i> Panzer	4	2		6
57	<i>Hister quadrimaculatus</i>	-	-	1	1
58	<i>Hypnoidus pulchellus</i>	4	5	1	10
59	<i>Idiochroma dorsalis</i>	1	-	8	9
60	<i>Ityocara rubens</i>	1	-	12	13
61	<i>Melanotus brunnipes</i>	-	-	1	1
62	<i>Metabletus foveatus</i>	-	-	1	1
63	<i>Metabletus truncatulus</i>	-	5	5	10
64	<i>Microletes maurus</i>	-	9	3	12
65	<i>Mycetophagus populii</i>	-	1		1
66	<i>Necrophorus antennatus</i>		1		1
67	<i>Onthophagus taurus</i>	-	1		1
68	<i>Opatrum sabulosum</i>	68	65	46	179
69	<i>Ophonus azureus</i>	-	-	1	1
70	<i>Ophonus sabulicola</i>	-	-	4	4
71	<i>Orchestes fagi</i>	-	-	7	7
72	<i>Otiorrhynchus laevigatus</i>	6	3	-	9
73	<i>Otiorrhynchus singularis</i>	2	-		2
74	<i>Oulema melanopa</i>	-	1		1
75	<i>Oxyporus rufus</i>	5	3	1	9
76	<i>Paederus limnophilus</i>	-	-	1	1
77	<i>Paradons quadrisignatus</i>	-	1	1	2
78	<i>Paramecosoma melanocephalum</i>	-	3	-	3
79	<i>Pedinus femoralis</i>	21	2	6	29
80	<i>Pentodon idiota</i>	39	34	18	91
81	<i>Phyllotreta atra</i>	6	11	5	22
82	<i>Phyllotreta nemorum</i>	37	98	-	135
83	<i>Phyllotreta nodicornis</i>	-	17	5	22
84	<i>Pleurophorus caesus</i>	4	14	8	26
85	<i>Psammobius porcicollis</i>	-	1		1
86	<i>Pseudocleonus cinereus</i>	2	3	-	5
87	<i>Pseudophonus rufipes</i>	-	-	6	6
88	<i>Pterostichus aterrimus</i> var. <i>niger</i>	1	1		2
89	<i>Pterostichus cupreus</i>	-	2		2
90	<i>Pterostichus lepidus</i>	3	-		3
91	<i>Pterostichus marginalis</i>	9	33	220	262
92	<i>Pteryngium crenatum</i>	62	9	8	79
93	<i>Pyrrhocoris apterus</i>	8	42	9	59
94	<i>Scirtes hemisphaericus</i>	-	1		1
95	<i>Selatosomus latus</i>	-	-	1	1

96	<i>Silpha obscura</i>	-	2	11	13
97	<i>Sipalis circularis</i>	-	-	2	2
98	<i>Staphylinus caesareus</i>	-	1		1
99	<i>Stomodes gyrosicollis</i>	-	3		3
100	<i>Tachyporus ruficollis</i>	3	5	10	18
101	<i>Tachyusa constricta</i>	-	1		1
102	<i>Tanymecus dilaticollis</i>	24	8	12	44
103	<i>Tanymecus palliatus</i>	1	2	2	5
104	<i>Zabrus blapoides</i>	1		1	2
105	<i>Zabrus tenebrioides</i>	-	-	1	1
<b>Total species</b>		<b>845</b>	<b>838</b>	<b>927</b>	<b>2610</b>

The highest number of beetles gathered from the 3 variants, have had a total of 34 species. These were (tab. 2):



**Fig.1** The number of individuals and species collected at the 3 variants

With a total of 297 specimens the *Conosoma bipunctata* species, representing 10.69% of the total; *Pterostichus marginalis*, with a total of 262 specimens, representing 10.04% of the total; *Epicometis hirta*, with a total of 187 specimens, representing 7.14% of the total; *Opatrum sabulosum* L., with a total of 179 specimens, representing 6.86% of the total; *Haplothrips tritici*, with a total of 165 specimens, representing 6.32% of the total; *Drasterius bimaculatus*, with a total of 146 specimens, representing 5.6% of the total; *Phyllotreta nemorum.*, with a total of 135 specimens, representing 5.17% of the total; *Dermestes lanarius* L., with a total of 107 specimens, representing 4.1% of the total.

Table 2

The structure of species (taxa) with the largest number of specimens collected

No.	The scientific name	Total	%
1	<i>Conosoma bipunctata</i>	279	10.69
2	<i>Pterostichus marginalis</i>	262	10.04
3	<i>Epicometis hirta</i>	187	7.14
4	<i>Opatrum sabulosum</i>	179	6.86
5	<i>Haplothrips tritici</i>	165	6.32
6	<i>Drasterius bimaculatus</i>	146	5.6
7	<i>Phyllotreta nemorum</i>	135	5.17
8	<i>Dermestes lanarius</i>	107	4.1
9	<i>Formicomus pedestris</i>	100	3.83
10	<i>Pentodon idiota</i>	91	3.48
11	<i>Anthicus antherinus</i>	83	3.18
12	<i>Pteryngium crenatum</i>	79	3.02
13	<i>Pyrrhocoris apterus</i>	59	2.26
14	<i>Colodera nigrita</i>	53	2.03
15	<i>Tanymecus dilaticollis</i> and <i>Armadilludium vulgare</i>	44+44	3.37
17	<i>Gryllus campestris</i>	43	1.64
18	<i>Anthicus floralis</i>	41	1.57
19	<i>Harpalus distinguendus</i>	33	1.26
20	<i>Agriotes lineatus</i>	32	1.22
21	<i>Pedinus femoralis</i>	29	1.11
22	<i>Pleurophorus caesus</i>	26	0.99
23	<i>Phyllotreta nodicornis</i> , <i>Cryptophagus dentatus</i> and <i>Phyllotreta atra</i>	22+22+22	2.52
26	<i>Coccinella 7 punctata</i> and <i>Tachyporus ruficollis</i> <i>Gravenhorst</i>	18+18	1.37
28	<i>Anthicus humeralis</i>	17	0.65
29	<i>Silpha obscura</i> L. and <i>Ityocara rubens</i>	13+13	0.99
31	<i>Microletes maurus</i> and <i>Aphthona euphorbia</i>	12+12	0.92
33	<i>Anthicus gracilis</i>	11	0.42
34	<i>Metabletus truncatulus</i> and <i>Hypnoidus pulchellus</i>	10+10	0.75
35	Other species < 10 samples each	193	7.4
<b>TOTAL</b>		<b>2610</b>	<b>100</b>

## CONCLUSIONS

1. In the 3 samples has been collected a number of 2610 specimens belonging to a total of 105 species of beetles in wheat. On variants, the situation is as follows:

- In the the untreated wheat variant for consumption has been collected 845 beetles belonging to a number of 55 species (taxons);
- the variant number 2, the treated wheat, 830 beetles has been collected from a number belonging to 65 species (taxons);
- The variant number three, wheat for seed the beetles has been collected 927 belonging to 72 species (taxons).

2. A number of 10 species had more than 100 specinens. The species with the greatest number of samples were: *Conosoma bipunctata* with 279 samples representing 10.69% of the total; *Pterostuchus marginalis* with a total of 262 samples, representing 10.04% of the total; *Epicometis hirta* with a total of 187 samples, representing 7.14% of the total *Opatrum sabulosum* with a total of 179 samples, representing 6.86% of the total; *Haplothrips tritici* with a total of 165 samples, representing 6.32% of the total; *Drasterius bimaculatus* with a total of 146 samples, representing 5.6 % of the total; *Phyllotreta nemorum* with a total of 135 samples, representing 5.17% of the total; *Dermestes lanarius* with a total of 107 samples, representing 4.10% of the total and *Formicomus pedestris* with a total of 100 samples, representing 3.83% of the total.

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## THE STUDY IN THE SEEDLING STAGE OF TOMATO VARIETIES CULTIVATED IN ORGANIC AGRICULTURE

### STUDIUL ÎN FAZA DE RĂSAD A UNOR SOIURI DE TOMATE CULTIVATE ÎN AGRICULTURĂ ECOLOGICĂ

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**Abstract.** The study was performed in Vegetable Research-Development Station Bacău - Romania, during 2015 – 2016 in order to evaluate the tomato varieties resistance at soil-borne diseases attack in seedling stage. The following tomatoes cultivars were cultivated in protected area - variants V2 – Ghittia, V3 – Monymaker, V4 – Ruxandra, V6 - Inima de bou, V8 – Bobalna, V10 – LMV, V12 – TFC had a degree attack (GA%) below 1.5 %. Other tomatoes cultivars V1 - Brandywine black, V2 - Brandywine black real, V3 - Brandywine black red, V5 – Delicios, V6 - Delicios de Podis, V8 - Indigo ‘Sun’, V9 – Omar’s Lebanese, V10 - Pantene Romanesco, V11 – Thesaloniki, V14 – Vilma were tolerant at the soil borne diseases.

**Key words:** tomato, soil-borne diseases, seedling

**Rezumat.** În perioada 2015 – 2016, la SCDL Bacău a fost studiată rezistența/toleranța la atacul patogenilor de sol, la diferite cultivari de tomate în faza de răsad, cultivat în agricultură ecologică în solar: V2 – Ghittia, V3 – Monymaker, V4 – Ruxandra, V6 - Inima de bou, V8 – Bobalna, V10 – LMV, V12 – TFC au avut un grad de atac (GA%) sub 1,5%. Alte soiuri de tomate: V1 - Brandywine black, V2 - Brandywine black real, V3 - Brandywine black red, V5 – Delicios, V6 - Delicios de Podis, V8 - Indigo ‘Sun’, V9 – Omar’s Lebanese, V10 - Pantene Romanesco, V11 – Thesaloniki, V14 – Vilma au fost tolerante la atacul patogenilor de sol.

**Cuvinte cheie:** tomate, patogeni de sol, răsad

## INTRODUCTION

Important diseases that have occurred in seedling stage of tomato include: *Pythium debaryanum* (Hesse), *Rhizoctonia solani* (Kühn), *Fusarium* spp., *Phytophthora parasitica* (Dast.), Calin, 2015.

The plants wilt and die suddenly, sometimes before emerging from the soil (preemergence damping-off) and sometimes after emerging from the soil (post

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emergence damping-off). Symptoms can include root rot, stem lesions, and general seedling wilt.

Soilborne pathogens often survive for long periods on host plant debris, soil organic matter, or as free-living organisms.

Each vegetable crop may be susceptible to several pathogens. Many soil factors including soil type, texture, pH, moisture, temperature, nutrient levels and ecology affect the activity of soilborne pathogens. Soil-borne pathogens such as *Pythium* and *Phytophthora*, often called water molds, can be particularly destructive if the soil is kept too wet for a long period of time. Although temperature is important, different species of these so-called water molds can infect at different temperatures. *Pythium* is the most common water mold pathogen found on diseased vegetable seedlings and is often associated with excessive nutrition or ammonium toxicity. Damping-off caused by the water molds is less likely to occur during warm dry springs.

The best practice for minimizing the incidence of disease in organic vegetable crops is planting high quality disease-resistant cultivars (Calin, 2015) and other disease management control practices (Baysal *et al.*, 2008; Cao *et al.*, 2010; Diab *et al.*, 2003; Jeanine Davis *et al.*, 2007; Klein *et al.*, 2011; McKellar and Nelson, 2003). This is even more important, because the number of resistant varieties available to grower increases and the fungicide resistance continues to challenge effective control with permitted fungicide in organic gardening.

The purpose of researches in this paper is the establishment the differences in susceptibility of tomato cultivars at soil-borne diseases for reducing the number of treatments and getting a quality seedling tomatoes grown in organic farming.

## MATERIAL AND METHOD

During 2015 – 2016 years, greenhouse experiments were performed in Vegetable Research-Development Station Bacau - Romania, in order to evaluate the behavior of tomato plant cultivars in the seedling stage, to the soil-borne diseases attack.

The trial in tomato seedling were performed at the following cultivars:

- tomatoes in protected area: V1 - Saint Pierre, V2 - Ghittia, V3 - Monymaker, V4 - Ruxandra, V5 - Buzau 1600, V6 - Inima de bou, V7 – Siberian, V8 – Bobalna, V9 – Roman, V10 – LMV, V11 – TN, V12 – TFC, V13 - Prekos F2, V14 - Cherry Bacau;

- tomatoes in protected area – world cultivars: V1 - Brandywine black , V2 - Brandywine black real, V3 - Brandywine black red, V4 - Carbon Neamt, V5 – Delicios, V6 - Delicios de Podis, V7 - Indigo Apple tomato, V8 - Indigo ‘Sun’, V9 – Omar’s Lebanese, V10 - Pantene Romanesco, V11 – Thesaloniki, V12 - Timny Tim, V13 – LM, V14 – Vilma.

The cultivars were sown in the greenhouse on March. At 3 – 6 days after emergence, the young plants were transplanted in cell plastic trays. No preventive chemicals treatments were applied, in order to encourage the development of the natural diseases.

Ratings were based on Pathogenically Rating Scale 0-5 (0 is no disease, 5 is terminally infected). The plants were visually evaluated. The following scale was used:

1 No spots and lesions,

- 2 1 - 3 spots or lesions present on stem or leaves,  
 3 4 - 8 spots present on stem, leaves and bracts,  
 4 Lesions and spots present on stem, bracts, leaves, flowers and stems,  
 5 Collapse of plant.

The observations were accomplished every 10 days, during a 30 days period, after plantation of young vegetable plants in cell plastic trays.

The attack estimation was determined using the following indicators: frequency of attack (F%), intensity of attack (I%), degree of attack (DA%).

The results obtained will be use in organic agriculture in order to decrease the number of diseases treatments in the organic agriculture practices of vegetable.

## RESULTS AND DISCUSSIONS

The suitability of tomato cultivars in organic farming is presented in table 1.

Table 1

**The behavior of some cultivars of tomato cultivated in organic farming at seedling stage**

Cultivar	Attack			Health plants (%)	Comments
	Frequency (F%)	Intensity (I%)	Degree of attack (DA%)		
1	2	3	4	5	6
Tomatoes in protected area					
Saint Pierre	8.2	100	8.2*	91.8	<i>Ability for organic agriculture</i>
Ghittia	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Monymaker	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Ruxandra	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Buzau 1600	4.5	100	4.5**	95.5	<i>Good ability for organic agriculture</i>
Inima de bou	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Siberian	5.3	100	5.3**	94.7	<i>Good ability for organic agriculture</i>
Bobalna	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Roman	2.2	100	2.2**	97.8	<i>Good ability for organic agriculture</i>
LMV	0	0	0***	100	<i>Very good ability for organic agriculture</i>
TN	11.1	100	11.1	88.9	-

1	2	3	4	5	6
TFC	1.1	100	1.1***	98.9	<i>Very good ability for organic agriculture</i>
Prekos F2	1.8	100	1.8**	98.2	<i>Good ability for organic agriculture</i>
Cherry Bacau	2.7	100	2.7**	97.3	<i>Very good ability for organic agriculture</i>
Tomatoes in protected area – world cultivars					
Brandywine black	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Brandywine black real	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Brandywine black red	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Carbon Neamt	20	100	20	80	-
Delicios	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Delicios de Podis	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Indigo Apple tomato	25	0	25	75	-
Indigo 'Sun'	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Omar's Lebanese	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Pantene Romanoesco	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Thesaloniki	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Timny Tim	25	100	25	75	-
LM	33.3	100	33.3	66.7	-
Vilma	0	0	0***	100	<i>Very good ability for organic agriculture</i>
Control					

LSD 5% - 1.5

LSD 1% - 7.2

LSD 0.1% - 9.4

### Tomatoes in protected area

The results regarding the degree of attack (DA%) was:

- Under 1.5% at: Ghittia, Monymaker, Ruxandra, Inima de bou, Bobalna, LMV, TFC. They are distinguished by a very good suitability in organic farming in the seedling stage (fig. 1).
- Under 5.5% at: V5 Buzau 1600, V7 Siberian, V13 Prekos F2, V14 Cherry Bacau with good suitability in organic agriculture.
- Cultivars: Saint Pierre and TN were sensible at pathogen attack. The recommendations of cultivation methods for these cultivars are that disease control practices in seedling stage should be used.

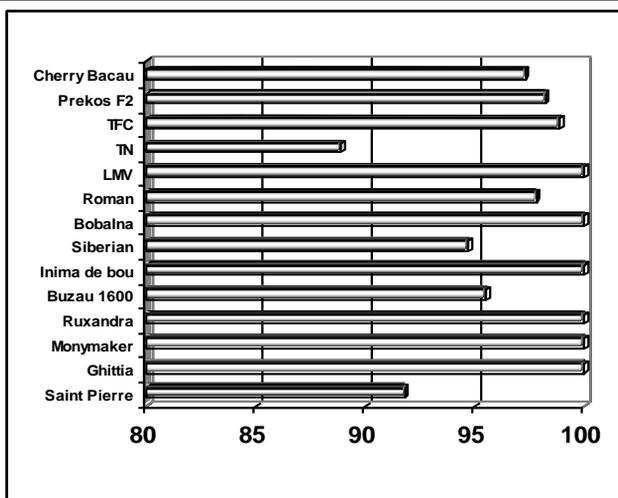


Fig. 1 The suitability of tomato cultivars in protected area at organic farming

### Tomatoes in protected area – world cultivars

The data obtained show a very good suitability at organic agriculture in seedling stage for: Brandywine black, Brandywine black real, Brandywine black red, Delicios, Delicios de Podis, Indigo ‘Sun’, Omar’s Lebanese, Pantene Romanesco, Thesaloniki, Vilma (fig. 2). The cultivars: Carbon Neamt (Degree attack – DA% – 20%), Indigo Apple tomato (DA - 25%), Timny Tim (DA – 25%), LM (DA – 33.3%) were sensible at soil borne diseases.

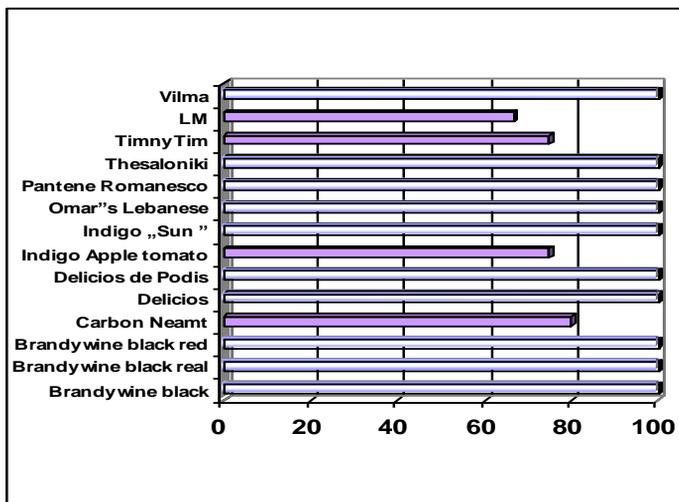


Fig. 2 The suitability of tomato cultivars in protected area - world cultivars at organic farming

## CONCLUSIONS

### Tomatoes in protected area

The attack of soil borne diseases degree was:

- Under 1.5% at: Ghittia, Monymaker, Ruxandra, Inima de bou, Bobalna, LMV, TFC. They are distinguished by a very good suitability in organic farming in the seedling stage.
- Under 5.5% at: V5 Buzau 1600, V7 Siberian, V13 Prekos F2, V14 Cherry Bacau with good suitability in organic agriculture.

Cultivars: Saint Pierre and TN were sensible at pathogen attack.

The recommendations of cultivation methods for these cultivars are that disease control practices in seedling stage should be used.

### • Tomatoes in protected area – world cultivars

The cultivars: Brandywine black, Brandywine black real, Brandywine black red, Delicios, Delicios de Podis, Indigo ‘Sun’, Omar’s Lebanese, Pantene Romanesco, Thesaloniki, Vilma had very good suitability at organic agriculture in seedling stage. The cultivars: Carbon Neamt (DA% – 20%), Indigo Apple tomato (DA - 25%), Timny Tim (DA – 25%), LM (DA – 33.3%) were sensible at soil borne diseases.

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## TRENDS IN SUSTAINABLE LANDSCAPING OVER THE LAST DECADES

### TENDINȚE PEISAGISTICE SUSTENABILE DIN ULTIMELE DECENII

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**Abstract.** *Currently, in the sustainable landscaping domain, the focus is on creating unpolluted environmental areas, which can offer facilities for collective forms of contact, recreation and leisure. These new creations of ambient will be able to effectively solve the “4 basic requirements” of human settlements: healthy life, balanced work, leisure and communication. The accelerated rhythm of work and overwork tendencies have changed the structure of human leisure time, landscaping process being forced to adapt to this phenomenon, offering a multifunctional and original design of various creations. The implementation of these sustainable projects, in last decades, was able to effectively combat the urban and territorial congestion, overcrowding and pollution.*

**Key words:** sustainable, landscaping, trends, design

**Rezumat.** *În prezent, în domeniul sustenabilității peisagistice, accentul se pune pe crearea unui mediu urban nepoluat, care să dispună de amenajări și facilități pentru forme colective de contact și odihnă. Aceste ambientări noi vor putea să rezolve eficient cele patru cerințe de bază ale așezărilor omenești: viață sănătoasă, muncă echilibrată, recreere și comunicare. Ritmul accelerat de muncă și munca în exces au modificat structura timpului liber al omului, peisagistica fiind obligată să se adapteze acestui fenomen prin oferte multifuncționale din ce mai variate și mai originale. Concretizarea în ultimele decenii a tendințelor sustenabile peisagistice a reușit să combată eficient congestia, aglomerarea și poluarea urbană și teritorială.*

**Cuvinte cheie:** sustenabil, peisagistică, tendințe, design

### INTRODUCTION

Over time the man allowed himself to be mesmerized by the material power, by the search for new techniques and sciences, to such an extent that led to the destruction of most of the planet on which he lives. Many of the contemporary troubles are obviously the result of the rush for wealth and power. The aggressive human intervention in the natural environment, especially in the last two centuries, shows an accelerated involution of understanding and respect for nature. Currently, it seems extremely doubtful that in future, the man can still survive in an atmosphere totally artificial and polluted, on a planet where natural

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elements are deeply affected. Most current problems arise from the fact that pragmatism, managed by human greed, try to destroy much of the health and balance of the Earth.

"Toward what are we heading?" is the question which, nowadays, has become extremely stressful for humanity, considering the rapid degradation of human awareness, on multiple levels, about the state of natural environment and human relationships. The actual urban stress is emphasized by the multiple forms of pollution, also by the overwork and the lack of free time needed for relaxation. Added to this is the lack of adequate public spaces designed to foster the needed human contact so healthy for both, communities and residents (Krier, 2006). The consequences of all these phenomena, longer-acting in time, are visible to the younger generations, dramatically alienated by the artificial universe in which they live.

## **MATERIAL AND METHOD**

Comparative analysis of the latest relevant landscaping achievements has a particular importance, enriching the knowledge with the most efficient approaches and scientific researches (Dascălu, 2016). Currently, in the landscaping field, the focus is on creating unpolluted urban and territorial environments, equipped with facilities for collective forms of relaxation, comfort and repose. These new types of proposals could give a positive response to the "4 basic requirements" of human settlements: life, work, leisure and communication. The accelerated rhythm of work and overwork has changed the structure of human leisure time - landscape architecture is forced to an adaptation process to this phenomenon, increasing the offer of varied and original sustainable design. By this way, urban congestion, overcrowding, pollution and many other actual problems could be effectively countered.

Analyzing landscaping experiments and achievements, starting with the postmodern era until now, we can see the gradual emergence of new issues and sustainable trends, in contradiction with some future trends that are trying to create an artificial universe. Nowadays human awareness of huge pollution has led to the increased collective concern for the environmental health and for the development of policies to protect natural values. Multipurpose research of sustainable solutions it was generated by the growing need for urban natural spaces, facing the lack of urban space where landscape projects can be created. The accelerated degradation and degeneration of urban public spaces created also the need to find new ways to increase the search for curative and preventive solutions of urban rehabilitation and regeneration (Dascălu and Cojocariu, 2016).

## **RESULTS AND DISCUSSIONS**

A brief overview of the most important landscape trends from the second half of the twentieth century and early twenty-first century would be very useful to understand toward what moves the landscaping today.

Major programs to achieve regional and urban parks, created in the early twentieth century, were expanded, upgraded and functional diversified in the second half of the century.

The tendency to use in a rational way the urban land came into contradiction with the explosive demand of urban entertainment facilities consuming large surfaces.

In this context, in the last decades of twentieth century were created *huge entertainment parks and not least the great multifunctional urban parks*. All these far surpassed as surface and functional diversity the first early twentieth century public parks.

An important branch of the great parks programs is represented by the *technological parks, created to promote technical and scientific innovations or research* in a more attractive manner of landscaping, for tourism and economic development. Their names vary from Techno-Polis to Scientific-Technical Parks, depending on the areas occupied, because some areas are very large compared to other parks, covering surfaces equivalent to cities. Design effort for the parks is considerable, either in urban-architectural terms or in landscaping ones. The results are spectacular, attracting many visitors, contributing to economic growth.

Major *sports parks and Olympic parks* constitute also a theme offering ample opportunities for the development of landscape design.

Another theme that offers the possibility to develop on extensive surfaces and very diverse presentation facilities is represented by *the parks for annual landscaping exhibitions or landscaping shows*, increasingly visited by nature lovers from all around the world.

One of the new trends approached both by the major entertainment landscape programs and also by the general green spaces programs, is the *spectacular aquatic design* created in special aquatic parks or in public spaces. These programs involve the presence of aquatic show facilities, which occupy large surfaces, both indoor and outdoor.

A unique combination of dynamic and interactive landscaping manner, currently offering multiple major attraction areas, is the theme of the *new modern botanical gardens* created for multipurpose activities. To generate a new way of communicating information, in order to attract more visitors to the regions where they are located, the new botanical gardens manage to combine scientific research and education with fun and relaxation, giving up old thematic programs imposed by the history.

The trend of *storied systems of huge landscaped tiles* located above areas stations, crossings, car parks, commercial galleries, watercourses, etc., generated new types of urban public spaces for relaxation and healthy natural recreation. In the same context, *landscape conversion of old overhanging industrial railways* created new multifunctional famous promenades.

The sustainable issue of *green roofs* is linked to the fight against urban pollution, either visually or by noxes or by overheating. Studies started with experiments on horizontal small roofs, continuing with gardens designed on whole floors, open or closed.

*Green walls or green facades trend* have been conceived in the last decades to cover various degraded buildings and provide research subjects for urban and regional landscape rehabilitation domain. Scientific research of plants capable of covering external surfaces, being resistant to pollution and extreme weather conditions, currently still offers a large field for scientist's studies (Dascălu and Negrea, 2016).

*Sustainable landscaping design could be an instrument helping the city to become a homogenous whole, unifying its fragments.* In this context, we can understand the utility of the ideas that support the use of landscaping to correct, rehabilitate and unify urban areas and damaged or broken tissues. Feature green spaces to serve as a bridge, a link for interstitial transition between different urban areas, has inspired new directions for research and implementation of new projects. In this context, there is a new type of urban landscape that favors the entering in crowded urban textures of the green multifunctional spaces prevailing environmental protection.

Landscaping solutions to solve urban traffic problems propose a particular design, especially in residential areas. The routes are called "*streets for all*" or "*woonerf*" and require slowing traffic to 15 km/h using landscaping instruments. In this way on these streets can circulate peacefully together pedestrians, cars and cyclists, without any inconvenience.

Another issue to be considered is represented by the trend of *urban sprawl trough university campuses* areas that have promoted the multifunctional and sustainable landscaping.

*The tendencies of planting the new emerging neighborhoods on the periphery of big urban centers*, improving their microenvironment, constitute another sustainable development direction of the late twentieth and early twenty-first century (Krier, 2006).

*Large landscaped areas with symbolic and monumental meanings* illustrate another important trend of the end of the twentieth century. We can find in the great capitals of the world the exemplified idea of symbolic cities axis linking major urban points in order to restore town's personality through a comprehensive and spectacular landscaping.

But the most important trend of all, the one that can really save the Earth, is the domain where the landscaping was deeper involved lately: *the rehabilitation and conversion of landscape areas damaged by industrial activities or natural processes and phenomena* (Rottle and Yocom, 2011).

This theme bestows real hope to the humankind through many creations, important architectural and landscape rehabilitation and conversions, saving and regenerating polluted and degraded areas. Interdisciplinary scientific research and studies are currently looking for effective curative and preventive landscaping solutions. Designers teams from all over the world are connected for saving large degraded areas, their projects being an example of living systems, where green infrastructure provides multiple services created both for society and for nature.

Another priority issue for efficient sustainable landscaping is the *environmental rehabilitation of areas damaged by the presence of old technical equipment or by car or rail services and circulation* (Bendtsen, 2010). These interventions consist not only in masking landscape design, but especially in the functional conversion proposals that provide multifunctional creations using landscaping elements and special plantations to combat multiple pollution forms.

The problems raised by *the safe circulation of wild animals on their migration routes generated environmental projects called "green bridges" crossing the highways*: these are bridges with widths ranging from 10-50 m, covered with vegetation.

Landfill created in beautiful natural areas with high tourism potential represent another important problem of the current period. *The pollution caused by the leakage of waste into soil and water required significant regeneration and conversion solution for huge surfaces*. Concretization required scientific ingenious researches: soils and water infested with pollutants were cleaned and treated in successive stages. The beauty and health rebirth of the landscapes gave a new life to the many polluted areas (Krauel, 2006).

Currently, digital design and computational techniques have turned into an important part of the act of landscaping creation, so that contemporary design is no longer just a simple software-aided process. In this context, the role of many experts, as architects, urban planners, landscape designers, should be redefined.

*Uptake of digital technologies in architecture, urbanism and landscaping design, increasingly differentiate many contemporary creations of previous achievements*. The explosion of "blobitectural" current with futuristic computerized forms has inspired sustainable landscaping creations, either organic or geometrical, many of which being already implemented (Dascălu and Cojocariu, 2016). *The wave of futuristic green buildings with abstract forms and the green cities with buildings imitating planted landforms* is increasingly appreciated and approached by many designers.

*The idea of extending on water some sustainable urban and territorial landscaping projects attracted numerous designers*. Inspired by structures of marine oil platforms, *floating green parks were designed in the shape of giant trees with many levels*, each floor hosting varied natural habitats with plants, birds, animals and insects. Another important sustainable trend consists in *floating green cities projects* for approx. 50 000 inhabitants, designed to be placed either near the shore or on the high seas.

## CONCLUSIONS

Generally, landscaping concepts involves complex spatial projects and arrangements, which are to be perceived in relation to the environment – natural, artificial, social and historical.

Sustainable landscape design requires a work that cannot be reduced to a simple assembly of components. It comes to proposals of functional solutions,

stemming from the real needs of cities and inhabitants, according to their characteristics and particularities, helping to ensure their physical and psychological comfort, a healthy life and, not least, to restore their identity, self-esteem and confidence in future.

In tandem with the architecture and urbanism creations, the landscape design researches undertakes many difficult tasks vital for humanity, in order to regenerate Mother Earth, to offer curative and aesthetic experiences, to materialize fascinating interactions between causes and effects, to embody for real seemingly utopian projects.

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## EVOLUTION OF EARTH CONSTRUCTION METHODS

### EVOLUȚIA METODELOR DE CONSTRUCȚIE CU PĂMÂNT

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**Abstract.** Human kind history bring testimonies about using earth or clay as building materials on territories of ancient civilisations. According to some populations tradition of earth building, architects and construction engineers of our days have used this common material for modern solutions of buildings or in organic composition of new materials with improved properties. The paper illustrate evolution of earth building methods and also highlighting modern constructive and architectural solutions involving this material.

**Key words:** earth building evolution, earth building methods

**Rezumat.** Istoria umanității aduce mărturii despre utilizarea pământului sau argilei drept materiale de construcție pe teritoriile unor civilizații străvechi. În conformitate cu tradiția construcțiilor de pământ prezentă la multe popoare, arhitecții și inginerii constructori din zilele noastre au utilizat acest material comun în soluții moderne de construire a clădirilor sau în alcătuirea de materiale noi ecologice cu proprietăți îmbunătățite. În lucrarea de față se relatează evoluția modalităților de a construi cu pământ, subliniind totodată soluțiile constructive și arhitecturale moderne care implică acest material.

**Cuvinte cheie:** construcții de pământ, evoluție, metode de construire cu pământ

## INTRODUCTION

Since ancient times human settlements were built using local materials, largely available in those areas, following cultural tradition and according to historical stage. The use of simple earth or in various combinations with other materials (wood and stone), is the commonly used method in houses built for thousands of years in various geographical and climatic areas. Anthropological research but also the recent years evolutions in the construction industry have highlighted the ecological potential, the sanogenous quality, but also the effectiveness in energy savings of earth as a building material, which is now being reinvented in combinations with other materials but also in terms of implementing technologies.

## MATERIAL AND METHOD

Earth architecture is one of the most powerful and expressive of human creations based on materials from the surrounding environment. This includes the most varied manifestations, both in terms of architecture and urban planning: from simple housing to stately palaces, from granaries to religious buildings, the historic

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cities centers to ancient archaeological sites. UNESCO started a program - WHEAP (World Heritage Earthen Architecture Programme), which was made an inventory of historic remains, earth buildings and architecture worldwide. In 2012 a document was drafted totaled 150 such sites involving earth architecture (fig. 1, fig. 2, fig. 3, fig. 4, fig. 5, fig. 6) (World Heritage, 2012)

Building techniques for earth edifices are known for over 9000 years, houses dating from 8000-6000 BC land Turkistanull being discovered in Russian (Pumpelly, 1908) (Minke, 2008). The earth was used in all ancient cultures, not just for building houses but alsow religious and representation buildings (fig.1).



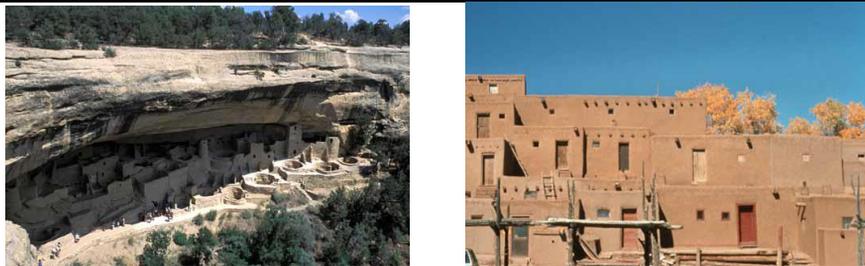
**Fig. 1** The images of Funerary temple of Ramesses II, 1200 BC. Thebes Necropolis, Egipt (left), Old city of Ashur, Iraq, the ziggurat and the great temple of the god Ashur, III milenium-II BC, (right) (World Heritage, 2012)



**Fig. 2** Old Towns of Djenné, Mali, Africa, III century BC (World Heritage, 2012)



**Fig. 3** Fortified medieval town of Provence, France, IX-XXI century (World Heritage, 2012)



**Fig. 4** Mesa Verde National Park, USA, X-XXI century (left), Pueblo de Taos, Arizona and New Mexico, USA, XI-XV century (right) (World Heritage, 2012)



**Fig. 5** Old Walled City of Shibam, Yemen, XVI century (left), The Mausoleum of Oljaytu, Iran, XIII-XVII century (right) (World Heritage, 2012)



**Fig. 6** Himeji-Jo Castel, Japan, XVII century (left). Fujian Tulou, China, XV-XX century (right) (World Heritage, 2012)

Earth as a building material can be scientifically called "clay" and is actually a mix of clay, fine sand (sludge), sand and possibly coarser components such as pebbles or stones. It can be used in several forms: "clay bricks" or "adobe" for unburned earth bricks, compressed earth uses the term "earth blocks" and when the earth is compacted in a mold, it is called "rammed earth". Also used, earth lumps mixed with straw, can be used as such or can be built on a trellis, between two such networks of twigs or on slats of wood, with a clay plaster on the outside.

Compared to standard industrialized building materials, clay has both advantages and disadvantages (World Heritage, 2012).

Among the disadvantages we can mention:

- clay material is not standardized (based on place of origin it can have different compositions and properties);
- mixture of clay shrinks when it dries, causing cracking of this construction;
- clay is not waterproof, it should be protected from rain and frost especially if it is wet (by covering the walls with roofs with wide eaves, or a layer of moisture-resistant masonry, or other layers of protection).

Instead, clay has many advantages that industrial materials do not have:

- clay balances humidity (can absorb and release moisture faster and to a greater extent than any other building material);
- clay keep the heat due to large thermal inertia balancing indoor climate where there are large temperature differences between day and night;
- clay saves energy and reduces pollution of the environment;
- clay is reusable by soaking with water;
- clay reduces costs for materials and transportation;
- ideal for constructions made in with local labor force;
- helps preserve the wood and other organic material embedded in it;
- clay absorbs pollutants.

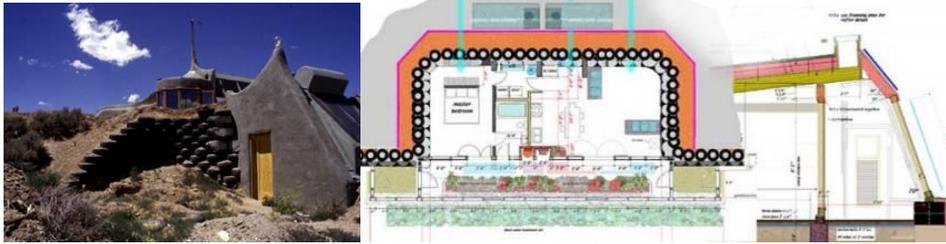
## RESULTS AND DISCUSSIONS

Coming from Canada, in 2004, romanian architect Ileana Mavrodin, built a experimental clay lodge in Sasca village, Caras Severin, subsequently, moved permanently, and today is often visited by tourists curious to see a bio house, perfectly integrated into the landscape (<http://agointel.ro>).



**Fig.7** Bio House, Sasca, Caras Severin, Romania (<http://agointel.ro>)

Michael Reynolds is one of the American architects who implemented the concept of Biotope, building especially in New Mexico, USA, sustainable and energy independent houses, incorporating recycled materials (tires, aluminum cans, plastic bottles, colored bottles, etc.), all filled or embedded in compacted earth. These homes accumulate sun's heat in summer within the thick walls and use it in winter. Rainwater is also collected and integrated it into 4 cycles of use, household waste being processed into fertilizer for greenhouse plants grown in winter (fig. 8) (<http://agointel.ro>, <https://earthshipstore.com>, <http://www.greenhomebuilding.com>).

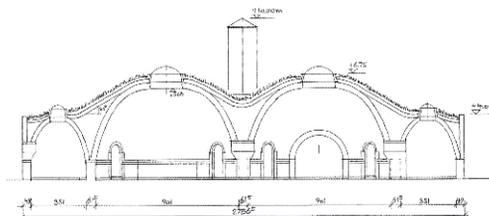


**Fig.8.** Images of Michael Reynolds houses  
 (<https://earthshipstore.com>, <http://www.greenhomebuilding.com>,  
<http://www.tboake.com/earth.html>)

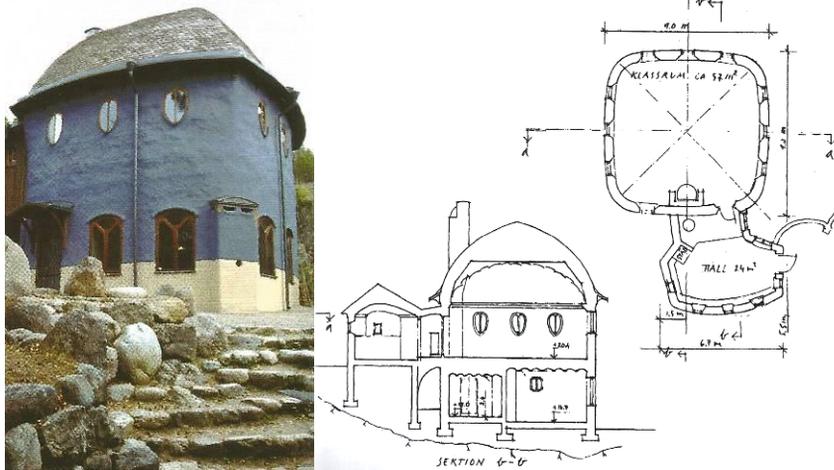
Another way of building with earth is filling plastic bags or jute bags with soil or sandy loam and then building that into a wall covered with earth plaster or whitewash (for jute preservation). Californian architect Nadir Khalili took further the idea, using continuous tubes made of jute bags filled with earth and built a house in Brazil in this constructive system (fig. 9) (World Heritage, 2012).



**Fig. 9** House built from continue raffia tubes filled with earth, Brazil  
 (World Heritage, 2012)



**Fig. 10** Mosque with two domes, built entirely of unfired clay blocks covered with earth and grass SII, arch. Gernot Minke, Wabern, Germany, 2007 (World Heritage, 2012)



**Fig. 11** Waldorf School built with blocks of solid clay, arch. Mats Wedberg, Jarna, Sweden, 1993 (World Heritage, 2012)

Although earth construction are rather suitable for warm or temperate climates, in the world were built even in cold areas (fig. 10, fig. 11). Lately appeared more and more buildings using beaten earth or unburned clay bricks, in combination with wood or straw, as recognition of the respect for nature. The high energy consumption and huge transport costs, construction and maintenance emplied by modern materials, generates an awernes regarding pollution and natural resources consumption.

## CONCLUSIONS

Earth buildings where always a handy and easy way to put into practice a building sistem. Nowadays, the clay advantages, that other industrial materials do not have, encompass a higher consciousness on the environment.

Earth construction methods, put into practice and combined with other materials, generate today many sustainable buildings and high energy efficiency.

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## LANDSCAPE PLANNING OF HOSPITAL CONSTRUCTION RELATED GREEN AREAS - CURRENT TRENDS

### AMENAJAREA PEISAGERĂ A SPAȚIILOR VERZI AFERENTE CONSTRUCȚIILOR SPITALICEȘTI – TENDINȚE ACTUALE

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**Abstract.** Nature has been venerated since ancient times for its beneficial effects on human health from a holistic point of view. With the progress of science and technology, man has forgotten to look for healing in nature, sanitary facilities evolving in to a real industrial plant for health, the rigour of medical procedures and specialty of pharmaceutical synthesis products exclude almost brutal in most cases anything can we called healing effect of nature through what she gives us. Studies on the subject have demonstrated curative role of landscape and gave a new direction on designing medical centers. According with that, recently appeared so-called „therapeutic gardens” and „healing gardens”. This paper illustrates this tendency of returning to the therapeutic effects of the nature and compares the achievements of international medical centers with the first steps taken in the same direction in Romania.

**Key words:** hospital, healing landscaping, therapeutic gardens

**Rezumat.** Natura a fost din cele mai vechi timpuri venerată pentru efectele sale benefice asupra sănătății ființei umane din punct de vedere holistic. Odată cu progresul științei și tehnologiei, omul a uitat să mai caute vindecarea în mijlocul naturii, spațiile sanitare evoluând spre adevărate uzine medicale, în care rigorile procedurilor terapeutice și specializarea produselor farmaceutice de sinteză exclud, aproape brutal și în cele mai multe cazuri, orice poate fi numit efectul vindecător al naturii prin tot ce ne oferă ea. Studii realizate pe această temă au demonstrat rolul curativ al peisajelor și au dat o nouă direcție proiectării centrelor medicale. Au apărut astfel, în ultimii ani, așa numitele grădini vindecătoare precum și grădinile terapeutice. Lucrarea de față aduce în lumină această tendință de revenire la efectele terapeutice ale naturii și compară realizările unor centre medicale internaționale cu primii pașii pe aceeași direcție realizați în România.

**Cuvinte cheie:** spitale, amenajări vindecătoare, grădini terapeutice

## INTRODUCTION

Man and nature in its wholliness are in perfect cohesion for millennia, but this link is in a fragile balance. In last decades, the human being forgoted to

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maintain this balance, with the accelerated development of modern consumist society based on industrialization, robotics and computerization.

Human health is also given by a good balance between states of physical, psycho-emotional and spiritual, but also the harmonious integration into the natural environment. Although the concern for therapeutic gardens existed since the Middle Ages, by arranging monasteries courtyards where they took care of the sick people, nowadays, very few green areas related to hospitals or health care facilities are designed so as to be used for the benefit of patients, healthcare professionals or visitors. Many of them are taken with the historic buildings of the nineteenth century, pavilionar type constructions, which includes landscaped gardens already. Numerous studies have highlighted both the need for a return to the concept of holistic medicine (healing concomitant soul, psyche and then the physical body), but rather the restoration of the millennial traditions on healing methods by harmonizing between man and nature but also with our inner self. Thus appeared the so-called "healing gardens" or "therapeutic gardens" near the medical centers of the world where both sick people and the healthy ones, the elderly or young people with physical disabilities or mental illnesses, children and parents can regain their balance, self-confidence, life force and even its own healing resources.

## MATERIAL AND METHOD

A study conducted in 1995, by Clare Cooper Marcus, MA, MCP and Marni Barnes, MLA, LCSW, in University of California, Berkeley (Cooper-Marcus and Barnes, 1995), highlighted the history of medical centers gardens, a number of typologies of green spaces of hospitals, also several case studies of such gardens, after giving them some recommendations for the design and development of therapeutic garden.

This creates describes some types of green areas related to medical facilities, the authors found in this study (Cooper-Marcus and Barnes, 1995).

- Rarely seen, generally on the outskirts of cities, "**Landscape grounds**" or "**park**" or "**campus**" are surrounds and connects buildings of the same medical center through walking alleys and rest places, such as the medical center Kaiser Permanente in Walnut Creek, California, or St. Mary's Hospital, Newport, Isle of Wight, England (fig.1).



**Fig. 1** The images of St. Mary's Hospital Park, Newport, Isle of Wight, England (Cooper-Marcus and Barnes, 1995)

- **Landscape setback**, located between the principal façade and the property alignment to the main street building access. Being the only green space outside, this setback can become frustrating for patients and staff due to lack of rest places or paths for walking that are not found in most such gardens (eg.: main entrance of the Alta Bates Medical Center, Herrick Campus, Berkeley, California).
- **The front porch** at the main entrance of the building is often found to hospitals or care centers and holds both a representative role by giving visibility to entry, but also functional, to protect exterior stairs or ramps of the buildings, visitor reception with pedestrian and auto access protected from rain, with the possibility of placing some banks for waiting or telephone booths, and small places for smokers (eg.: Main Entry, John Muir Medical Center, Walnut Creek, California).
- **The entrance garden** is one that is designed from the start to be used by patients and staff as green space for relaxation and rest outdoors, being in danger of overexposure to noxes of the car traffic or parking lots adjacent to the main street (eg.: Main Entrance, Marin General Hospital, Greenbrae, California).
- **The patio** is usually surrounded by buildings or pavilions of the same medical complex and should be visible and accessible to visitors from entering in the main hall of the building. It can be a space planted with trees small and medium, flowers or decorative herbs, with even water games and can serve as rest area or outdoor terrace for a coffee shop or a buffet adjacent to the court (eg.: Cafeteria Courtyard, Novato Community Hospital, Novato); It can also be equipped with games for children (eg.: Children's Courtyard, Kaiser Permanente Medical Center, Vallejo, California).
- **The plaza** is an outside area of buildings that can be mostly paved, with little vegetation inserts and furnished so that it can be used for rest and relaxation by the beneficiaries of medical services but also by visitors or medical staff (eg.: Seating Plaza, Alta Bates Medical Center, Herrick Campus, Berkeley, California).
- **The roof terrace** is usually open space, bordered on one side of the building that can be used to enjoy views of the surrounding (eg.: Perimeter Terrace, Davies Medical Center, San Francisco, California).
- **Roof Gardens** of medical centers are specifically designed to benefit all users and can be too exposed to strong sunlight or wind, noise or air conditioning equipment usually positioned on the roof (ex.: Alta Bates Medical Center) (fig.2).
- **Healing gardens**, indoors or outdoors, are designed specifically for this purpose in hospitals and through the planning austerity, fosters retrieving self-consciousness or activities that require confinement - reading or meditation (eg.: Healing garden of Oncology Department, Marin General Hospital, Outpatient Medical Building, Greenbrae, California; The healing garden of Kaiser Permanente Medical Center, Roseville, California).



**Fig. 2** Roof garden of Alta Bates Medical Center, Berkeley, California  
(Cooper-Marcus and Barnes, 1995)

- **Viewing garden** is the kind of landscaped green space that can be viewed but can not be accessed within it, with all sensitive perceptions, what becomes frustrating some times (ex.: Central Atrium, John Muir Medical Center, Walnut Creek, California).
- **Viewing walking garden**, related with the aterior garden, is a space that can be accessed but very few people at the same time (ex: Internal Gardens, St. Mary's Hospital, Newport, England; Flower Gardens, Stanford University Medical Center, Stanford, California)

According to Roger Ulrich's theory, taken by Marcus C. Cooper and M. Barnes (Cooper-Marcus and Barnes, 1995, Cooper-Marcus, 2007) in order to draw up a guide to achieve a healing gardens, the researchers have start with the premises of meeting several requirements observed during the studies as essential necessary for such a space:

- to create opportunities for movement and exercise;
- to give the opportunity of choosing between spaces, to ensure discretion and create a feeling of self-control;
- to provide meeting space and socializing for all those who would like to use such spaces;
- to ensure access to nature and other fun activities and beneficial.

In addition to these basic requirements, to achieve the full potential of such gardens, the researchers stressed the importance of fulfilling several other features:

- the visibility garden space since its entry into the building or signs of proper medical;
- the easy access to this space;
- the familiarity garden;
- the peace and comfort of the garden;
- the presence of objects of art with positive message, unambiguous.

## RESULTS AND DISCUSSIONS

Compared with early foreign achievements in the field of therapeutic gardens, in Romania, not early than June 2014 was inaugurated the first

therapeutic garden in Mocrea courtyard of Psychiatric Hospital in Arad (fig. 3) (<http://www.healinglandscapes.org>). The garden covers an area of 1500 square meters and consists of three subareas:

- the seedlings product area where patients work on tables, planting seeds and sprouts in pots;
- the area for planting in wooden boxes
- the planting area on earth where patients can create floral arrangements.

The plants used are chosen based on their role, being distinguished three categories:

a) the plants that stimulate tactile sensory (Sedum, Miscanthus, Salvia argentea);

b) the herbs that besides the olfactory role they have also therapeutic and medicinal effects by domestic consumption (Mentha piperita, Ocimum basilicum, Thymus, Melissa);

c) the plants which relaxes by color and scent (Lavender, Jasmine, Impatiens, Dianthus, Euonymus, Clematis, Rudbeckia).

Each of the 40 patients of the hospital are empowered and helped to socialize, to join the group by attending various easy tasks, from preparing the soil, planting seeds and taking care of plants in their various stages of growth.



**Fig. 3** Therapeutic garden at the Psychiatric Hospital in Mocrea Arad, Romania (<http://www.healinglandscapes.org>)

The second event followed the previous one at short time, in November 2014, by opening the garden for horticultural therapy, at Căpâlnaș Psychiatric Hospital, Arad, under the supervision of horticultural engineer Marcela Necșanu (<http://www.romania-insider.com>).

Also in October 2016, the event Ecoweek (<http://ecoweek.org.wixsite.com>) presented a project to redevelop the green space for the Victor Babeș Hospital in Bucharest, so as to turn it into a useful space for patients, medical staff and visitors of the hospital, respectively a therapeutic garden.

Susan Erickson points out in his paper (Erickson, 2012) that every garden should be designed for the specific needs of its users: patients with various physical or mental disabilities, patients recovering from strokes or other injuries debilitating, patients suffering from terminal illnesses or other patient populations including children who want to regain vitality and joy of life. Also refers to the medical staff of that medical institution or hospital visitors who can spend relaxing moments in a revitalizing environment specially designed for them. This can only be achieved through close collaboration between landscape architect and specialized medical personnel to different therapies that can be done in an outer natural frame (eg.: occupational therapy including activities horticultural chineto and physio-therapy, therapy recreation or therapies complementary such as chromotherapy, music therapy, aromatherapy, cristaloterapy, etc).

## CONCLUSIONS

Studies have shown that green spaces related to hospital facilities are beneficial to both patients and medical staff or visitors, whether under these arrangements are taken into account the needs of each category of persons who benefit from them. The first part of this paper reviews the many types of design of green spaces, related to hospitals abroad and refers to some requirements and characteristics necessary for the successful implementation of such a design. In the second part refers to therapeutic gardens benefits of the two psychiatric hospitals mentioned, in western of Romania. This paper emphasize the importance of involving in the process of designing a healing garden of all those who are creating the garden but also the specialists in charge of treating and caring for patients.

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## THE GREEN ROOF - AN AESTHETIC SOLUTION FOR BUILDING REHABILITATION

### ACOPERIȘUL VERDE – SOLUȚIE ESTETICĂ DE REABILITARE A CLĂDIRILOR

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**Abstract.** *In recent decades, the green roof is a modern system and environmentally friendly of coating buildings, partially or completely, with soil and vegetation. This particular type of landscaping allows vegetation to grow harmoniously upon considerable areas which is why architects, builders and urban planners around the world have begun to turn to green roofs not for aesthetic reasons, a concern rather secondary, but for their practical nature and the ability to alleviate environmental extremes specific to conventional roofs. Therefore this paper aims to present some of the most suitable solutions presenting the rehabilitation of a building in the area of Podu Ros-Iasi, Romania.*

**Key words:** rehabilitation, substrate, sustainability, roof culture

**Rezumat.** *În ultimele decenii, acoperișul înverzit este un sistem modern și ecologic, de acoperire, parțială sau completă, cu sol și vegetație a clădirilor. Acest tip special de amenajare, permite vegetației să crească armonios pe suprafațe considerabile motiv pentru care, arhitecți, constructori și proiectanți urbani din toată lumea au început să apeleze la acoperișurile verzi nu din considerente estetice, o preocupare mai degrabă secundară, ci pentru caracterul lor practic și pentru capacitatea de a atenua extremele de mediu specifice acoperișurilor convenționale. Drept urmare lucrarea de față își propune să prezinte câteva din cele mai pretabile soluții prezentând o propunere de reabilitare a unei clădiri din Podu Roș-Iași, Romania.*

**Cuvinte cheie:** reabilitare, substrat, sustenabilitate, cultură pe acoperiș

### INTRODUCTION

With direct relation to ensuring the ecological balance of the environment, landscape architecture is concerned with the preservation and development of landscapes and their associated values for the benefit of current and future generations.

Landscapes contribute to improving the quality of human life and through their social function they provide the framework and a favorable environment for public or private outdoors recreating (Kristin *et al.*, 2007), they also grace the cities, places of life and work, some of which have cultural importance (gardens - museum, gardens exhibition, historic gardens) or scientific (botanical gardens,

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rosaries, reserves, national parks, etc.). Thus, the green roof - garden is a modern system, environmentally friendly, coating partial or complete, with soil that enables these types of gardens, providing a favorable environment for the growth of vegetation (Nagase and Dunnett, 2011). Buildings with low slope roofs are favorable for applying the green roofs - garden technology, in composition of which vegetation is comprised of plants that require minimal care in a multilayer system (Palla *et al.*, 2010) which is actually a continuation outwards of the building's roof. It thus offers the possibility of a zone of vegetation on new or rehabilitated rooftops.

But living roofs are not a new concept. They were common in land homes in the American prairie, and the grassed ones can still be seen in houses and wooden barns in Northern Europe.

The term "green roof" is new though in landscape architecture (Compagnone, 2009) and amounts to more than a simple "greening the skyline".

It is a term that highlights changes made to thousands of neglected municipal roofs, to turn them into a genuine wealthy real estate, which benefits not only the owners of apartments in the area, but rather more the owners of buildings, who can enjoy hanging gardens, living spaces with enhanced comfort, offering quality leisure in nature.

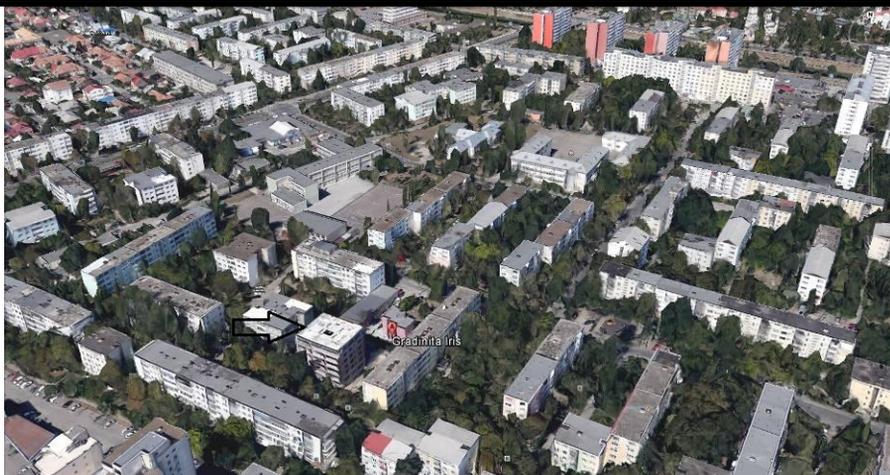
At the same time, increasingly more researchers such as Maureen Connelly - who heads a laboratory for green roofs at Institute of Technology in Columbia - studying the practical benefits of this solution (Rowe, 2009) helping to quantify performance and accurate measurement of the ability to reduce leakage rainwater, increase energy efficiency and improve urban acoustic environment. Thus more and more green roofs started appearing in the world, each one being an experiment.

## MATERIAL AND METHOD

This paper aims to revitalize green areas in Podu Ros, by adding a new green space located on a surface that is not being valued while it is in a very circulated and used by residents and visitors as a venue, as well as relaxation and rest area.

Design principles apply also in vegetable compositions carried out on the roof garden design, choosing of species and planting in general. These principles were essential in developing color schemes when we took into account the succession of flowering for drafting the rehabilitation of the roof of an apartment building in Iasi County, Podu Ros neighborhood, Rozelor Alley.

In order to achieve the arrangement a wide range for of flower species (*Begonia*, *Schizostachyum*, *Lavandula*, *Pelargonium*), decorative herbs (*Poa*, *Lolium*) and dendrological species (*Prunus*, *Acer*, *Cotoneaster*, *Rosa*, *Salix*, *Spiraea*, *Cornus* etc ) was used (Haggas, 2006).



**Fig. 1** Framing the proposed space for rehabilitation

The neighborhood area in which the building stands on whose roof the rehabilitation project is going to be carried out has an area of about 1973 square meters, with the limits in the west Str. Nicolina (fig. 2. a, b), in North Blvd. Socola (fig. 2 d), and in the south the Iris kindergarten.



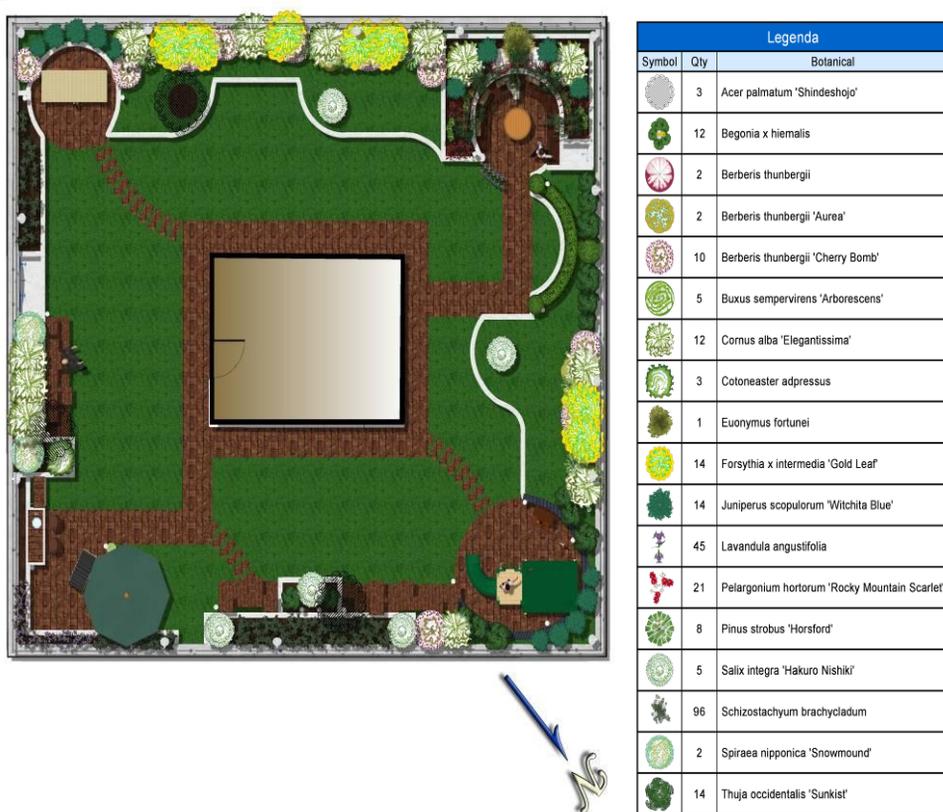
**Fig. 2** The roof before rehabilitation

A large part of area of this street is paved and the current vegetation does not have a unified composition, which is why the introduction in this area of a new green surface (Negrea *et al.*, 2014) is meant not only to revitalize the area, but also to provide the tenants of the building possibility to benefit from a green space plus the comfort of parking.

The overall objective of research pursued in this paper was to develop a formula for designing this enjoyable order called composition by using universally valid principles such as sequencing, unity, balance and emphasis for the drafting the rehabilitation of a roof of 289 sq.m. (fig. 2) belonging to a block of flats on the Roses Alley.

## RESULTS AND DISCUSSIONS

Investigations addressed in this paper aimed to identify and the sustainable use of disused spaces in cities in order to turn them into green areas, taking into account, in particular demographic developments. The opportunity of the study is in direct relation to ensuring the ecological balance of the environment and has as main objective the preservation and development of landscapes and their associated values for the benefit of current and future generations.



**Fig. 3** The systematization proposal of the space on the roof of the building

Each plant in this arrangement has a different way of growth, forming a unique mass and volume (fig. 4.a), which develop, intervening changes as the plant matures. These forms of species used, whether pyramidal, spherical,

weeping, columnar or displayed (fig. 4.d) define and subdivided the space of the roof. Some forms give a much more dramatic effect than others and have been used in order to attract attention (fig. 4.b).



**Fig. 4** Details on compositional units that form the landscape design

Selected plant shape and location fitting was essential to create dynamic, attractive and comfortable spaces resulting in uniform distinct compositional units, but unitary. Also, their texture creates a wide range of emotions, as a visual stimulus in these gardens. This is perceived as a mass, devoid of foliage, bark, leaves or flowers and changes according to the light of the day and season. Up close, the size and shape of the leaves and branches become predominant textural elements of each group. From a distance, appears light and shadow effect on the entire arrangement, different intensities of light and dark translate as texture in this garden. Rough textures of specimens of roses and hawthorn tend to create a relaxed mood and are visually dominant, while fine and smooth textures of species of *Prunus* L. and *Malus* Mill. are associated with official, elegant, discreet attitudes and visually are more passive.

Powerful texturized contrasts between the species used add intensity and volume to the arrangement and create interest for the both spaces. The bark of *Cornus* L. and *Crataegus* L. and leaves of the *Prunus* L. and *Corylus* L. are ways textural interest for the space was added, complemented by herbs and spring flowers, embellishing both the texture and the color.

Fragrance of the flowers in this arrangement has been rigorously taken into account by use of specimens of roses with scented flowers that add extra charm to the garden, creating a point of interest by expanding sensory awareness.

## CONCLUSIONS

These types of gardens have an important role in the greening of cities affected by serious pollution in recent decades. Green gardens contribute to lowering energy consumption of the building, ecological and sustainable solves the problem of rainwater and improve the aesthetic appearance of the building and area. Finding ways to bring nature in cities - even as nature itself is urbanized - will make them more livable, and not just for people.

The main objective of this paper was to present a way of designing a vegetal composition by using universally valid principles of sustainable landscaping.

**Acknowledgements:** *The research has been carried out in the POSDRU project "Programe doctorale și postdoctorale pentru promovarea excelenței în cercetare, dezvoltare și inovare în domeniile prioritare – agronomic și medical veterinar, ale societății bazate pe cunoaștere" coordinated by USAMV Cluj-Napoca, ID: 132765*

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## RESEARCH ON SOME PARAMETERS OF SEED PRODUCTION OF CUCUMBER VARIETY 'CALIBRI'

### CERCETĂRI PRIVIND UNII PARAMETRI AI PRODUCȚIEI DE SEMINȚE LA SOIULUI DE CASTRAVEȚI 'CALIBRI'

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**Abstract.** *Calibri cucumber variety with semi-long fruit intended for fresh consumption and for canning has been approved in 2015. During the period 2013 - 2014, determinations were performed on coefficients of variation for the main characters of the variety Calibri. Thus, the character of the fruit shape index registered a low variability in both technological maturity (5.4% - 7.0%) and physiological maturity (6.2% - 9.0%), which represents a great uniformity of variety on the shape of the fruit. Concerning the characteristics of quality, variety Calibri achieved an overall good organoleptic assessment (82.1%) compared to Rodnicioc F1 (69.2%), showing a pleasant commercial aspect, fine texture and good flavor - without bitterness. From the synthesis of data on the behavior of Calibri variety results that the main characters studied fall within the limits of variation of the analyzed variety, which shows a good conservative routing selection.*

**Key words:** cucumber, quantitative characters, qualitative characters, correlations, Calibri variety, Rodnicioc F1 variety.

**Rezumat.** *Soiul de castraveți Calibri cu fructe semilungi, destinate pentru consum în stare proaspătă și pentru conserve, a fost omologat în anul 2015. În perioada anilor 2013 – 2014, s-au efectuat determinări cu privire la coeficienții de variabilitate la principalele caractere ale soiului Calibri. Astfel, la caracterul indicelui de formă a fructului s-a înregistrat o variabilitate mică, atât la maturitatea tehnologică (5,4% - 7,0%), cât și la maturitatea fiziologică (6,2%-9,0% ), ceea ce reprezintă o mare uniformitate a soiului cu privire la forma fructului. În privința caracteristicilor de calitate, soiul Calibri realizează o notă generală de apreciere organoleptică bună (82,1%) față de Rodnicioc F1 (69,2% ), prezentând aspect comercial plăcut, textură fină și gust bun – fără amăreală. Din sinteza datelor cu privire la comportarea soiului Calibri rezultă că principalele caractere luate în studiu se încadrează în limitele de variație ale soiului analizat, ceea ce denotă și o bună dirijare a selecției conservative.*

**Cuvinte cheie:** castraveți, caractere cantitative, caractere calitative, relații, soiul Calibri, soiul Rodnicioc F1.

## INTRODUCTION

Economic and social impact of the product to the olericulture is the introduction and use new modern methods and techniques that have the effect of getting new genotypes with benefits resulting from the implementation in production activity and is well received by users. Characteristics of the economic impact are very well known,

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they are specific to new products the modern in general, and its impact on society and on the processes of research and production are sufficiently publicized, so the debate of the positive effects and benefits attracted by introducing synthetic varieties and of new commercial hybrids F1 in vegetable production. I consider that is absolutely necessary the production of new varieties and hybrids, at the base of which are re-found the autochthonous genetic resources, and which will produce the revision of research activity and olerioculture production.

Lately, two qualities have acquired a great importance: resistance and tolerance. In this paper, Resistance: is used to describe varieties with genes that prevent or reduce the rate of multiplication of pests and diseases, and tolerance: is used to describe varieties of which fruit production on a land infected by diseases and pests is completely reduced from their attack.

In European countries, the cucumber improvement is divided into two stages: first, to entrust the research institute or some highly specialized groups who seek to gain parental material. This is taught to licensed companies or resorts of practical improvement to be implemented in the improvement, applicative multiplication.

In recent years, the first stage has grown significantly, both in volume of performed biological material, and through theoretical problems scientifically based. Parent material has a great advantage that shows resistance to diseases, pests and aphids are relatively known heredity, giving a high proportion of offspring with valuable qualities. This priority given to creating scientifically parental lines is reflected in the major percentage that represents them in the origin of cultivated varieties of cucumber.

## MATERIAL AND METHOD

The investigations were conducted on a culture material from last year, in experiments conducted at the Scientific and Practical Institute of Horticulture and Food Technologies from Republic of Moldova.

In setting up the culture at the first stage in experimental plots was used the hybrid Rodnicioc F1 and the used biological material was the variety of cucumber 'Calibri' created at the Practical- Scientific Institute of Horticulture and Food Technology and approved in 2015, in solarium cucumbers were grown by seedling, preventive sown and grown in plastic trays that were sown on date 05/10/2015 aged 35 days, the planting on experimental plots was held on 14/06/2015.

During were made the vegetation the observations on completing the main phenological phases (sow - east, east - flourished, east - technological maturity, sunrise - physiological maturity).

At the technological and physiological maturity were performed biometric measurements on the following characters: the index of the fruit form, seed production, seed number, seed number in fruit and number of harvested fruits per plant.

For each character were calculated the following statistical indices: arithmetic mean ( $\bar{x}$ ), the variation amplitude ( $a$ ), variation ( $s^2$ ), standard deviation ( $s$ ), dispersion ( $k = x + -s$ ) (Dumitrescu, 1977; Pena, 1986; Săulescu and Bălașa, 1968).

Also, it was monitored the correlation degree of these characters. In assessing the quality of the fruit, the data taken in experimental plots were correlated with the results of lab chemical analysis.

## RESULTS AND DISCUSSIONS

Variability of the studied main characters of the Calibri variety during 2014-2015 is presented in table 1.

**The shape index of the fruit (cm).** In the reviewed period, this character has varied between 3.4 – 4.3 cm, with an average of 3.8 cm. The variability coefficient for the two years has an average of 7.5% (tab. 1). Coefficient value of variability was low and in the case of determinations made at technological maturity (5.4 to 7.0%), which demonstrates the fruit uniformity of this variety.

**Production of seeds (g).** The Rodnicioc F1 hybrid and the Calibri cucumber variety had a high variability coefficient of between 29.0 to 37.1% during the years 2014 to 2015. The average production of seeds was 9.5 g seed / plant (in terms of limiting the number of fruits per plant).

Table 1

**Variability of characters variety of cucumbers  
'Calibri' during 2013-2014**

Characters	Variation Limits	Average (x)	Deviation Standard (s)	The coefficient of variation (s%)
Index form				
2013	3.4 – 4.3	3.8	0.5	9.0
2014	3.4 – 4.1	3.7	0.4	7.4
Production of seed ( g / ul)				
2013	11.3 - 20.6	16.0	4.6	29.0
2014	4.0 – 3.8	3.9	0.1	37.1
The number of seeds in the fruits				
2013	180.3 – 250.3	217.3	35.0	25.1
2014	129.2 – 202.0	165.6	36.4	21.9
The number of harvested fruits per plant				
2013	2.3 – 4.1	3.2	0.9	29.3
2014	1.3 – 2.1	1.7	0.4	24.2

**3. The number of seeds in the fruit.** At this character have been registered values between 180.3 to 250.3 seeds, with an average of 215.3 seeds / fruit. The coefficient of variation had an average of 25.1%, indicating a high variability (tab.2.) Between the number of seeds per plant and seed production there is a strong direct correlation (fig.1).

**4. The number of harvested fruits per plant.** Under conditions of limiting the number of fruit at physiological maturity was collected between 1 and 4 fruits per plant, fruit with an average of 2.4. The coefficient of variation had high values every year (24.2 - 29.3%).

Calibri variety reach the technological maturity after 51-66 days after emergence (tab.3, fig.2) recording a peak of production at 73 days after emergence (18% of total) (tab.4). Until physiological maturity of this variety are needed in average 117 days.

The analysis indicates that between the number of fruits per plant and fruit production is no direct correlation.

Table 2

Summary of results obtained in the period 2013-2014

Character analyzed	Average (x)	The coefficient of variation (s%)	Standard deviation (s)
Index form	3.88	7.56	0.29
Prod. of seed (g/ pl)	9.53	31.61	3.20
Nr. Fruit seeds	70.86	28.16	20.20
Nr. harvested fruit / plant	2.40	27.06	0.65

The amount of seeds per fruit (g) Seed / fruit - 20.6

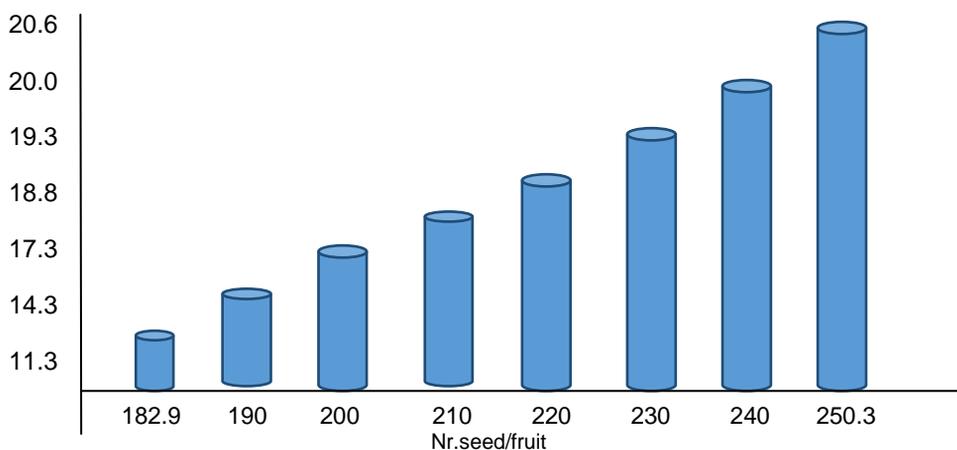


Fig. 1 The relationship between the number of seeds / fruit and seed production on the plant for the Calibri cucumber variety

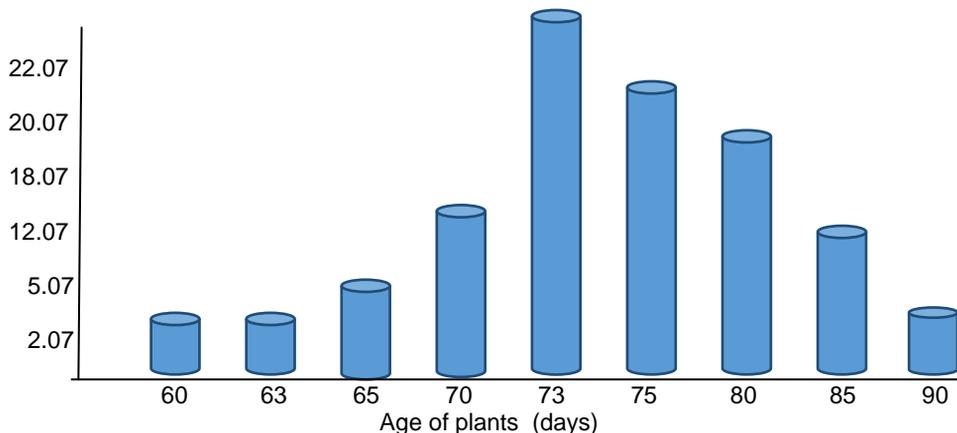
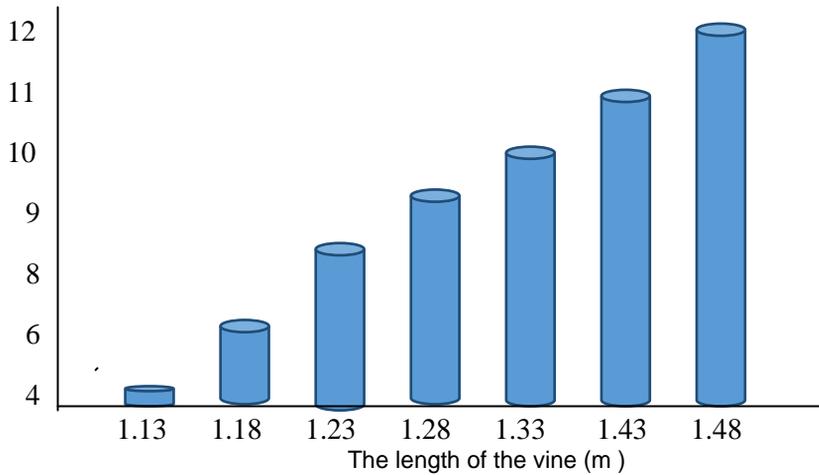


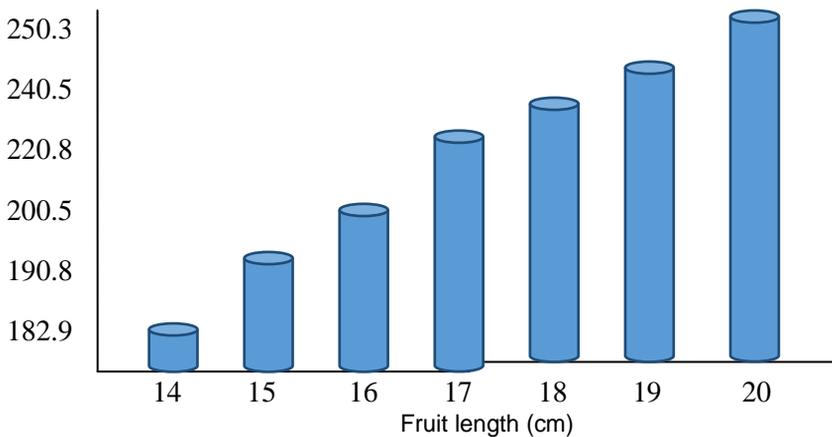
Fig. 2 Harvest dynamics for the Calibri cucumber variety

The number of fruit / plant



**Fig.3** Correlation between the length of vine and the number of harvested fruit per plant

The number of fruit



**Fig. 4** The correlation between fruit length and number of seeds in the fruit

Regarding the relationship between vine length and number of harvested fruits at technological maturity, was proved the existence of a moderate correlation.

Also, it has been established that there is a significant and moderate correlation between the length of fruit at physiological maturity, and number of seeds in fruit. Analyzing the relationship between fruit diameter at the physiological maturity and the number of seeds in the fruit, it has resulted in a weak correlation, statistically uninsured.

Table 3

## Phenophase's duration of Calibri variety

Phenophase	Number of days 2013	Number of days 2014	Average (2013 – 2014)
Seeding – sunrise	7	6	6
Sunrise – flowering	48	40	44
Sunrise tech. mat.	60	57	58
Sunrise physic. mat.	124	111	117

Table 4

## Harvesting dynamic for the Calibri variety

Date of harvest	Ageplant (days)	No. Plants	Production / kg	% of total
02.07	60	415	31.5	5.1
05.07	63	415	33.8	5.5
09.07	67	415	41.8	6.8
12.07	70	415	72.2	11.8
15.07	73	415	110.3	18.0
18.07	77	415	93.1	15.2
22.07	80	415	80.9	13.2
26.07	83	415	63.0	10.3
02.08	87	415	48.1	7.8
06.08	90	415	39.0	6.3

## CONCLUSIONS

The uniformity of fruits of this variety is proven by the shape index, which has a low variability for both fruits that reached technological maturity (5.4 to 7.0%) and physiological maturity (6.2 to 9.0%).

It has been shown that there is a significant correlation between the number of fruit and fruit production per plant, as well as between the number of seeds and seed production per plant.

There have been established high levels of coefficients of variability in seed production per plant (29.0 to 37.1%) and the number of seeds in the fruit (21.9 - 25.1%).

The data presented show that in the period 2013 - 2014, by directing the selection process, the variety Calibri has maintained within the limits of normal of variability.

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## THE MAIN QUANTITATIVE AND QUALITATIVE TRAITS OF EARLY WHITE CABBAGE VARIETY 'DE BUZĂU'

### PRINCIPALELE ÎNSUȘIRI CANTITATIVE ȘI CALITATIVE LA VARZA ALBĂ TIMPURIE SOIUL 'DE BUZĂU'

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**Abstract.** *It was found that the half-seed method by setting the control field in spring and then the wintering field in rosette stage (15-20 well-developed leaves) can maintain the variety in the allowable variability. Starting from the wintering plants in the rosette stage, the average seed quantity was 50.7 g/plant, while plants that overwinter in the head stage was only 28.8 g/plant. In the first case, floral stems are more vigorous, showing an average height of 127.6 cm to 118cm average height of floral stem in the second case. In 2014 was achieved an average production/ha of 1 574.5 kg conditional seed, to 1008 kg in 2015. Under the program for the conservative selection, by applying rigorous selection methods, this variety has been maintained within the allowable variability.*

**Key words:** selection, seed, production, variety De Buzau

**Rezumat.** *S-a observat că metoda jumătății de sămânță prin înființarea câmpului de control în primăvară și apoi a câmpului care ierneză în stadiul de rozetă (15-20 de frunze bine dezvoltate) poate menține soiul în limitele de variabilitate admisibile. Pornind de la plante care ierneză în stadiul de rozetă, cantitatea medie de sămânță a fost de 50.7 g/plantă, pe când la plantele care ierneză în stadiul de căpătână a fost numai de 28.8 g/plantă. În primul caz, tijele florale sunt mai viguroase, prezentând o înălțime medie de 127.6 cm față de 118 cm media, înălțimii tijeii florale în al doilea caz. În anul 2014 s-a obținut o producție medie /ha de 1 574.5 kg sămânță condiționată, față de 1 008 kg în anul 2015. În cadrul programului de selecție conservativă, prin aplicarea riguroasă a metodelor de selecție, acest soi a fost menținut în limitele de variabilitate admisibile.*

**Cuvinte cheie:** selecție, sămânță, producție, soiul De Buzău

## INTRODUCTION

Scientific community, based on an extensive analysis of events in recent years, notes that the economic and socio-political situation of the country was aggravated and could be appreciated as an extremely difficult one.

Only under conditions in which science and innovation will enjoy support from the state and society can be ensured the development of economy, creation of advanced technologies, production of new multifunctional materials, including a Center for olericulture, developing new biotechnology for production with a increased efficiency, developing new pure ecological products and methods of treatment, enhancement of alternative vegetable production, ultimately

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overcoming the economic crisis, management of the olericultural sphere of research and innovation, technological transfers, highly appreciated by international scientific bodies and solving pressing socio-economic problems of the country (Bălașa, 1973; Ceapoiu, 1976; Dumitrescu, 1988).

## MATERIAL AND METHOD

Researchers of this paper we propose to present the recorded results at the Scientific - Practical Institute of Horticulture and Food Technologies on method of producing of elite seeds of early white cabbage variety De Buzau, in the years 2014 and 2015.

Cabbage is one of the most common vegetable species, is widespread in all regions of our country.

Early cabbage occupies 20-30% of the vegetable cultivated area (cabbage, red cabbage, kale High content of minerals, carbohydrates, vitamins (A, B, C) as well as obtaining high revenues from production valorisation are elements that define economic and food value.

White cabbages for head (*Brassica oleracea* L - variety *capitata* L - white form, Cruciferae family) is a biennial plant, allogamous used both fresh in a wide range of culinary products and as raw material for canning industry, ensuring consumption throughout the year.

It is one of vegetable species that has a high biological plasticity, what really gives a peculiarity to adapt to various environmental conditions, which is why it can be cultivated on a very large geographical area.

Early cabbage varieties grown in our country presents some difficulties in seed production caused by biological traits and applied technology.

To maintain production at a higher level both quantitatively and qualitatively, a first and essential condition is to start from an elite planting of high biological value.

To obtain a good quality seeds with high biological purity of seeds that do not pass through the stage of loaf is necessary to establish a control field. The experience was conducted in the years 2014 - 2015 at the Scientific - Practical Institute of Horticulture and Food Technologies, in field research, using the seeds and plants from biological mother of early white cabbage variety De Buzau.

Typable plants percentage increases by applying the technology of seed production plant wintering in the open field in the rosette stage (for verification typicality parent plant).

The removal was done by applying the method of "half-seed". So, from the first half of seeds of the elite and of the families at this stage were set up a control field during optimal culture for early cabbage (5.III - 5.IV).

During the growing season have been made observations and measurements that are used to establish typicality of the variety.

In the other half of the elites and families arrested during analysis of variance for some character were established in 2015 in a field where plants wintering rosette stage (15-20 well-developed leaves) and also in that field, where plants spent the winter phase physiologically matured head.

In order to obtain elite seed it has been done according to selection scheme specific to the species, using the individual method of selection in C. A. and Selection method on families in C. S. mother plants and C.S.E. mother plants.

Through the analysis of variance was calculated arithmetic mean ( $\bar{x}$ ), standard deviation  $s$ , coefficient of variability % and the limits of variability ( $\bar{x} + s$ ) for each character included in the study.

Experience has been placed on the moldsoil with a neutral reaction ( $\text{pH} = 7.0 - 7.1$ ), well stocked nitrogen (N -  $\text{NO}_3$ : 70.0 to 94.2 ppm), phosphorus (PAL: 107 - 189 ppm) and the environment supplied with potassium (KAL 246 ppm).

## RESULTS AND DISCUSSIONS

Results achieved through the statistical processing of the data recorded in 2014 and 2015 for certain quantitative characters of early cabbage seeds which were wintering in the open field or in the rosette stage (15-20 well-developed leaves) or in the stage of physiologically matured head, they are shown in table 2. Quantitative variability of the main characters of early cabbage seeds variety De Buzau, seeding phase.

*Table 1*

**Quantitative variability of the main characters early cabbage variety De Buzau**

Phase	Specification	Height rod Floral (cm)	The average amount of seed per plant (g)
Wintering plants in rosette stage	X	127.6	50.7
	s	14.8	9.8
	s%	11.6	19.36
	x+s	142.4-112.8	60.5-40.9
Plants wintering in Headstage	X	118.0	28.8
	s	13	8.27
	s%	11.0	26.15
	x+s	131.0-105.0	37.07-20.53

x = arithmetic average

s = standard deviation

s%= coefficient of variation

x+s= range of variability

In both cases, by setting the control field during the optimal period of early cabbage culture can be certainly set compliance with the limits of allowable variability of this variety.

From this table it follows that cabbage seeds are more vigorous in the case of the plants that are in the case of plants that spent the winter in the stage of heads matured physiologically issued floral stems are less vigorous ( $\bar{x} = 118\text{cm}$ ) presenting also a middle variability.

Regarding the average amount of seed per plant, there is a difference of 56.8% between the average obtained on plants that have wintered the rosette stage ( $\bar{x} = 50.7\text{g}$ ) and plants spent the winter in the stage of physiologically mature head (average 28.8g).



**Fig.1** Seed harvesting was done manually and separately for each elite and family apart

## CONCLUSIONS

After the study performed on methods for the production of elite seed of early cabbage variety De Buzau, we can say that through the half-seed method by setting the control field in the spring and then the field that is wintering in the rosette stage (15-20 leaves well developed) can be determined the classification of the variety within the allowable variation.

Starting from the plants that winters in the rosette stage average amount of was 50.7 g seeds and plants that winters in the stage of physiologically matured head of 28.8 g which are less vigorous, reaching an average height of 118cm.

Under the program of conservative selection, applying rigorous selection methods, this variety has been maintained within the allowable variability.

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## THE DETECTION AND QUANTIFICATION OF MYCOTOXINS DIFURANICE IN MEDICINAL SPECIES

### DECELAREA ȘI CUANTIFICAREA MICOTOXINELOR DIFURANICE ÎN SPECII MEDICINALE

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**Abstract.** *Species (medicinal species) is a pharmaceutical form made up of mixtures of different plant organs (plant products), dried, which is used in therapy in the form of infusions, decoctions, macerates, syrups, tinctures, glycerine extracts, oil extracts, wine etc. In temperate continental area in which our country stands, saprophyte fungi live on the medicinal herbs so the medicinal species are often seen as different mycotoxins, metabolites of fungi. The highest incidence in this type of phyto is represented by mycotoxinsdifuranice (aflatoxins and sterigmatocistins), ochratoxin and patulin. The objective of this work consists in the qualitative and quantitative determination of mycotoxins in samples of medicinal species from pharmacies, freelance producers deprived of knowledge of toxicokinetics and toxicodynamics active principles of plants and herbal shops. The experiment was performed on 36 samples of vegetable and medicinal species, samples that were tested by first screening test in LUV and samples which showed fluorescence were studied further by high pressure chromatography. Most plant products from manufacturers freelancers do not correspond with the organoleptic rules and shows mycotoxins load.*

**Key words:** medicinal species, active principles, vegetal products, mycotoxin, quantitative and qualitative mycological test, mycotoxicological exam, *Tiliaeflos*, *Hypericicherba*, *Maydis stigmata*, ochratoxin, sterigmatocistin

**Rezumat.** *Specii (specii medicinale) reprezintă o formă farmaceutică alcătuită din diferite amestecuri de organe de plante (produse vegetale) uscate, care se utilizează în terapeutică sub formă de infuzii, decocturi, macerate, siropuri, tincturi, extracte glicerate, extracte uleioase, vinuri etc. În zona temperat continentală în care se situează țara noastră muceșii trăiesc saprofit pe plantele medicinale, astfel încât în speciile medicinale se decelează adesea diferite micotoxice, produși metabolici ai muceșilor. Incidența cea mai crescută în acest tip de fitopreparate o au micotoxinele difuranice (aflatoxinele și sterigmatocistinele), ochratoxina și patulina. Obiectivul acestei lucrări constă în determinarea calitativă și cantitativă a micotoxinelor în probe de specii medicinale provenite din farmacii, producători liber profesioniști lipsiți de cunoștințe legate de toxicocinetica și toxicodinamia principiilor active din plante și de la magazine naturiste. Experimentul s-a efectuat pe 30 probe de produse vegetale și specii medicinale, probe care au fost testate mai întâi prin testul screening în LUV, iar probele ce au prezentat fluorescență au fost studiate în continuare prin cromatografie de înaltă presiune. Majoritatea produselor vegetale provenite de la producători liber profesioniști nu corespund organoleptic normelor în vigoare și prezintă încărcătura micotoxinică.*

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**Cuvinte-cheie:** specii medicinale, principii active, produse vegetale, micotoxină, test micologic calitativ și cantitativ, examen micotoxicologic, *Tiliaeflos*, *Hypericiberba*, *Maydis stigmata*, ochratoxina, sterigmatocistina

## INTRODUCTION

Medicinal species (*Species, FR X*) represent an old pharmaceutical formula obtained from the mixture of vegetal products (plant organs with a certain pharmacological action used in therapy). Depending of the therapeutic and physicochemical features of their active principles, medicinal species are administered as aqueous, alcoholic, hydro-alcoholic, glycerinate, hydro-glycerinated extract solutions, oils, aromatic waters, syrups, tinctures, wines, dry extracts etc. Due to its high bioavailability and good tolerance, phytomedicines occupy an important place among the modern therapeutic methods. Medicinal and aromatic plants can be the ideal substrate for numerous mycetes that can parasite the plant in both vegetative and storage phase, especially in continental climate areas where the environmental factors ease their proliferation and the biosynthesis of mycotoxins. Mycetes possess a remarkable capacity to synthesise certain secondary metabolites: pigments, antibiotics, chemotherapics, phytotoxins and mycotoxins. (Butler, 1974, Coman *et al.*, 1985). Equally stunning is their capacity to adapt to the most different environments (Davis, 1987; Feng, 1998). Their hyphal structure facilitates their access and their development on solid life-hostile surfaces and the synthesis of mycotoxins (Vining, 1992; Moss, 1996). Mycotoxins are metabolic products with different chemical structures and highly stabile physicochemical features which make the detoxification of contaminated food products by means of chemical, physical and biological procedures accessible to the food industry to be impossible. Mycotoxins manifest the so called “relay toxicity” for humans and all the inconvenients of expressing toxicity through  $DL_{50}$ , an indicator that excludes the “cumulative toxicity” which includes chronic toxicity, carcinogenesis, teratogenesis and immunosuppression. The real dimension of mycotoxin pathogenicity is not yet considered to be clearly established, as the presence of mycetes and their metabolites in diseases with occult pathologies is suspected (Reye syndrome, acute respiratory distress syndrome, congenital malformations etc.) (Prisăcaru, 1998; Moretti, 2010).

## MATERIAL AND METHOD

The experimental model (tab. 1) consisted in the mycological and mycotoxicological examination performed on 36 samples of *Tiliaeflos* (linden flowers with stigmata), *Hypericiberba* (the upper part of Saint John's wort in adult plants) and *Maydis stigmata* (corn silk). The samples were collected from the Copou – Iasi, Pietrarie – Iasi, Comanesti-Bacau areas and from herbal shops of Iasi city and afterwards dried.

**A) The mycological examination** began with the *quantitative mycological exam* that was intended to establish the total number of mycetes per gram of vegetal product (NTM/g). The method used was the serial dilution method (Prior, 1981). The mycological exam continued with the *qualitative mycological exam* that was intended to identify the genders and eventually the species they belong to.

**B) The mycotoxicological examination** consisted of three phases, the results of each of them determining the passing to the next one. The three phases were: (I) the LUV exposure of 3 extracts from each sample, (II) analysis of the extracts with positive results in phase I using the HPLC test and (III) the TLC confirmation (thin layer chromatography) for the samples that had positive results in the previous phases (Badria, 1994; Prisăcaru, 1998; Radulović, 2013).

The Experimental Model				
Crt. no.	Vegetal product	No. of samples	Provenience areas	Sample abbreviation
1	<i>Tiliaeflos</i>	2	Iasi: Pharmacy I, Pharmacy II	F <sub>I</sub> , F <sub>II</sub>
		2	Copou area gardens	C <sub>I</sub> , C <sub>II</sub>
		3	Comănești (Lăloaia) locality	L <sub>I</sub> , L <sub>II</sub> , L <sub>III</sub>
		3	Comănești (Leorda) locality	L <sub>1</sub> , L <sub>2</sub> , L <sub>3</sub>
		1	Pietrărie: monastery	P <sub>1</sub>
		1	Pietrărie: orchard	P <sub>2</sub>
		1	Pietrărie: center	P <sub>3</sub>
		5	Iași: health stores	N <sub>1</sub> , N <sub>2</sub> , N <sub>3</sub> , N <sub>4</sub> , N <sub>5</sub>
2	<i>Hypericiberba</i>	2	Iasi: Pharmacy I, Pharmacy II	F <sub>I</sub> , F <sub>II</sub>
		2	Copou area gardens	C <sub>I</sub> , C <sub>II</sub>
		3	Comănești (Lăloaia) locality	L <sub>I</sub> , L <sub>II</sub> , L <sub>III</sub>
		3	Comănești (Leorda) locality	L <sub>1</sub> , L <sub>2</sub> , L <sub>3</sub>
		1	Pietrărie: monastery	P <sub>1</sub>
		1	Pietrărie: orchard	P <sub>2</sub>
		1	Pietrărie: center	P <sub>3</sub>
		5	Iași: health stores	N <sub>1</sub> , N <sub>2</sub> , N <sub>3</sub> , N <sub>4</sub> , N <sub>5</sub>
3	<i>Maydis stigmata</i>	1	Comănești (Leorda) locality	CL <sub>1</sub>
		1	Comănești (Lăloaia) locality	CL <sub>2</sub>
		2	Pietrărie: monastery	PM <sub>1</sub> , PM <sub>2</sub>
		2	Pietrărie: orchard	PL <sub>1</sub> , PL <sub>2</sub>

## RESULTS AND DISCUSSIONS

The results obtained from the mycological and mycotoxicological analysis of the 36 samples were recorded in table 2. After studying the data it was noticed the presence of the *Fusarium* gender in one of the samples coming from a herbal shop. This gender includes highly toxicogenic mycetes that produce ochratoxins, fumonisins and trichothecenes. The corresponding extract, subjected to the mycotoxicological test indicated a relatively high mycotoxin load (Ochratoxin A, mycotoxin involved in the apparition of the acute respiratory distress syndrome in pigs NEF, Balkan endemic nephropathy). All the 15 samples of *Hypericiberba* are clean from both mycological and mycotoxicological point of view. Two of the six samples of *Maydis stigmata*, although collected and preserved according to the FR rules, 10<sup>th</sup> edition, presented a mycological load (CL<sub>1</sub> includes mycetes from the *Penicilium* gender, and CL<sub>2</sub> from the *Aspergillus* gender). As it results from table 2, from these two samples contaminated with toxigenic mycetes, only the CL<sub>1</sub> sample was characterised by the presence of furo-furanicmetabolite, sterigmatocystin.

Table 2

Results of the mycological and mycotoxicological study of the samples				
Crt. no.	Vegetal product	Sample	Mycological examination	Mycotoxicological examination
1	<i>Tiliaeflos</i>	F <sub>I</sub> , F <sub>II</sub>	-	-
		C <sub>I</sub> , C <sub>II</sub>	-	-
		L <sub>I</sub> , L <sub>II</sub> , L <sub>III</sub>	-	-
		L <sub>1</sub> , L <sub>2</sub> , L <sub>3</sub>	-	-
		P <sub>1</sub>	-	-
		P <sub>2</sub>	-	-
		P <sub>3</sub>	-	-
		N <sub>1</sub> , N <sub>2</sub> , N <sub>3</sub> , N <sub>4</sub> , N <sub>5</sub>	N <sub>4</sub> - <i>Fusarium</i>	N <sub>4</sub> - ochratoxin A

2	<i>Hypericiberba</i>	F* <sub>I</sub> , F* <sub>II</sub>	-	-
		C* <sub>I</sub> , C* <sub>II</sub>	-	-
		L* <sub>I</sub> , L* <sub>II</sub> , L* <sub>III</sub>	-	-
		L* <sub>1</sub> , L* <sub>2</sub> , L* <sub>3</sub>	-	-
		P* <sub>1</sub>	-	-
		P* <sub>2</sub>	-	-
		P* <sub>3</sub>	-	-
		N* <sub>1</sub> , N* <sub>2</sub> , N* <sub>3</sub> , N* <sub>4</sub> , N* <sub>5</sub>	-	-
3	<i>Maydis stigmata</i>	CL <sub>1</sub>	<i>Penicillium</i>	Sterigmatocistin
		CL <sub>2</sub>	<i>Asperillus</i>	-
		PM <sub>1</sub> , PM <sub>2</sub>	-	-
		PL <sub>1</sub> , PL <sub>2</sub>	-	-

## CONCLUSIONS

1. From the 15 samples of *Tiliaeflos* (linen flowers) only one sample presented the existence of mycological load (the presence of *Fusarium* gender). The sample came from anherbal shop.

2. In the extract of sample N<sub>4</sub> where a mycetic load was identified there was also Ochratoxin A, a mycotoxin whose target are the kidneys and the lung.

3. The 15 samples of *Hypericiberba* presented no mycological load, and no mycotoxins.

4. Two of the 6 analysed samples, belonging to the vegetal product *Maydisstigmata*, presented a mycetic load (the presence of *Penicillium* and *Aspergillus* genders in CL<sub>1</sub> and CL<sub>2</sub> and the presence of mycotoxin, chemically related to aflatoxin (sterigmatocystin) was discovered only in sample CL<sub>2</sub>.

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## RESEARCH ON INCIDENCE OF ACRYLAMIDE IN THERMALLY PROCESSED FOODS

### CERCETĂRI PRIVIND INCIDENTA ACRILAMIDEI IN PRODUSE ALIMENTARE PRELUCRATE TERMIC

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**Abstract.** Acrylamide is a substance well known, industrial synthesized and used since the late nineteenth century. Toxicological profile include toxic effects to the reproductive system, neurotoxicity, genotoxicity and significant carcinogenic potential. This substance has sparked the interest of ecotoxicologists and the whole medical world in 2002 when it was detected in foods eaten by students from the University of Stockholm. The amide of acrylic acid is formed in various chemical ways from carbohydrates and amino acids from plant-based foods by frying, baking, gratin, processes involving exposure to high temperatures. The experiment detailed in this article attempts to assess the content of acrylamide in foods excessive consumed by children and adolescents. We analyzed 25 samples of potato chips, popcorn, biscuits, cookies, cocoa, coffee, cakes, all in various assortments. For statistical correlations between acrylamide and components of the nutritional declaration, it was used Spearman rank coefficient calculation, retaining as significant correlation coefficients with significance level of  $p < 0.05$ .

**Key words:** acrylamide (ACR), asparagine, acrolein, nutrition declaration, fatty acids

**Rezumat.** Acrilamida este o substanță cunoscută, sintetizată și utilizată industrial încă de la sfârșitul secolului al XIX-lea. Profilul său toxicologic include efecte toxice la nivelul aparatului reproducător, neurotoxicitate, genotoxicitate și un semnificativ potențial carcinogenic. Această substanță a stârnit interesul ecotoxicologilor și întregii lumi medicale, în 2002, când a fost decelată în produse alimentare consumate de studenții de la Universitatea din Stockholm. Amida acidului acrilic se formează pe diferite căi chimice din glucide și aminoacizi din alimente de origine vegetală prin prăjire, coacere, gratinare, procedee ce presupun expunere la temperaturi ridicate. Experimentul detaliat în acest articol încearcă să evalueze conținutul în acrilamidă al unor produse alimentare consumate excesiv de copii și adolescenți. S-au analizat 25 probe de chipsuri, popcorn, biscuiți, fursecuri, cacao, cafea, prăjituri, toate în diverse sortimente. Pentru stabilirea unor corelații statistice între valorile acrilamidei și a componentelor din declarația nutrițională, s-a utilizat calcularea coeficienților de rang Spearman, reținând ca fiind semnificativi coeficienții de corelație cu prag de semnificație  $p < 0,05$ .

**Cuvinte cheie:** acrilamidă (ACR), asparagină, acroleină, declarație nutrițională, acizi grași

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## INTRODUCTION

Acrylic acid amide is a chemical substance of low molecular weight, the structure of which can be found in the two centers of unsaturation (Pelluchi, 2007). Although its toxicity is well known, acrylamide is synthesized from the nineteenth century and continue to find more and more uses in industrial scale, mostly by polymer his polyacrylamide, considering that not a danger to the environment and, especially for humans (Bergmark, 1992; Calleman C.J., 1994; Mencinicopschi, 2005, FAO/WHO, 2002). Only in 2002 a study conducted at the University of Stockholm highlights acrilamidei presence in the French fries in hazardous concentrations for life, drawing attention to the real risk to public health (IARC, 1994, WHO, 2006). Soon, it proved that acrylamide is formed in foods vegetable thermally processed. These vegetable products occurs acrylamide containing amino acids (asparagine, in particular), ozide reducing or poliglucide (starch).

The main way of training of acrilamidei in foodstuffs is considered Maillard reaction, starting from amino acids and carbohydrate, but cannot be ignored nor the possibilities of obtaining from the acrylic acid, acroleina resulting from the hydrolysis of the fatty acids, even the glycogen synthesis of meat products etc. (Burlacu, 2009).

Since the range of foods, especially the so-called category of "substitution foods, foods that have replaced a table (biscuits, popcorn, chips, etc.), a "food to relax" (which is bored or watching an action movie, concert, parties, etc.) or "light food" (convenient to purchase food) is very wide and varied and is likely to be task acrylamide risk, the work of this aim to draw the attention of the relevant mode on the danger to which they are exposed especially the age classes: vulnerable children and young people who have not ensured be wholesome food before evening.

In this respect it had in view, detection acrilamidei, the substance of which the toxicological profile includes neurotoxicity, carcinogenicity, mutagenicity, reproductive toxic effects etc. in as many samples of snacks, crackers, chips, snaksuri, popcorn, pastries, cake, salted and roasted seeds etc.

## MATERIAL AND METHOD

The experimental model has included 20 samples of food products obtained through a thermal processing, food category of the most known and used because of the convenience of buying and excessive advertising.

The samples from which have been analyzed acrylamide have been collected from various kinds of chips (Lays, Extra DEEP Chio, Chio Chips salt etc.), popcorn (Star Popcorn salt), biscuits (East Harmony, Alexia, Sandwich crackers taste lemon), coffee (Tchibo, Espresso instant coffee decaffeinated Gourmet Selection), cocoa (Dr Oetker), sweets (cakes confectionery).

The content of acrylamide was analyzed by gas chromatography using the system of extraction of acrylamide, fats, proteins, and electrolytes to observe correlations between the values of acrylamide and the values of fats, proteins and electrolytes.

**RESULTS AND DISCUSSIONS**

Quantification of acrylamide of thermally processed food samples are presented in table 1.

Table 1

**Acrylamide concentrations in samples**

Sample	Product name	Acrylamide [µg%]
P1	Lay's	0.017
P2	Chio Exxtra DEEP	0.099
P3	Popcorn Star	0.001
P4	Chio Sare Chips	0.11
P5	Krax Grill Paprika	0.012
P6	Tortilla Chips	0.75
P7	Lay's Maxx DEEP RIDGED	0.058
P8	Krax Original Bacon	0.012
P9	Biscuiți Armonia Estului	0.076
P10	Biscuiți Sandwich with lemon flavor	0.009
P11	Alexia – Cocoa flavor	0.058
P12	Biscoff	0.090
P13	Petit Beurre	0.0089
P14	Cocoa Oetker	0.09981
P15	Coffee Tchibo	0.007813
P16	Instant coffee decaffeinated "Gourmet Selection"	0.0091
P17	Instant coffee Espresso	0.0121
P18	Coffee Dallmayr Prodomo	0.02601
P19	Turmeric powder Kotanyi	-
P20	Ground cinnamon Kotanyi	-

From studying these data shows that the highest concentrations of acrylamide were recorded in chips of sorts Tortilla (sample 7 - 0.75 mg%), Chio Salt (sample 4 - 0.11 mg%), Chio Exxtra DEEP (sample 2-0099 mg %) and cocoa powder Oetker (sample 14 - 0.0998 mg%).

To establish correlations between concentrations of acrylamide and components of carbohydrate, protein, electrolyte and the presence of fibers or use systematic methods were used value-processing, nonparametric, namely calculating the Spearman correlation coefficients for significance threshold  $p < 0.05$ .

Increasing the concentration of acrylamide is associated with high content of fatty acids and other fats.

Revealed a direct correlation significant between the amount of acrylamide and energy value of foods expressed both in kJ and in kcal ( $R = 0.8289$ ,  $p = 0.02$ ) increase in concentration in acrylamide being associated significantly with increased energy value of products food rations 30g.

There was also a significant correlation directly between acrylamide concentration and lipid content in the portion of 30g ( $R = 0.8289$ ,  $p = 0.02$ ) and reference consumption of fat percentage ( $R = 0.8074$ ,  $p = 0.03$ ).

It also found a correlation significant inverse between the concentration of acrylamide and consumption of fatty acids reference% / 30g portion ( $R = -0.7667$ ,  $p = 0.04$ ). Another significant inverse correlation has been established between the concentration of acrylamide in carbohydrate content and portion size of 30g ( $R = -0.8289$ ,  $p = 0.02$ ) (Bohosevici, 2016).

## CONCLUSIONS

1. In the process of formation of acrylamide there is a significant correlation between its concentration and increased direct energy value of foods studied.

2. It finds a correlation between the concentration of acrylamide reverse significant and fatty acid content.

3. A significant inverse correlation was also between acrylamide concentration and carbohydrate content.

4. The results show that potato chips and French fries contain acrylamide most abundant followed by biscuit, popcorn and coffee.

5. Variations in concentrations of acrylamide on the categories of products are due to both the concentrations of different acrylamide precursors (amino acids and reducing sugars) which depend on the variety of potato and cereal used as raw materials, as well as the technology parameters (temperature, the duration of the heat treatment, pH, the type of oil used etc.)

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**STUDIES REGARDING THE BIOREGULATORS  
INFLUENCE OVER THE MORPHOLOGIC AND  
ORNAMENTAL CHARACTERS OF  
*ALLIUM* 'PURPLE RAIN'**

**STUDII PRIVIND INFLUENȚA SUBSTANȚELOR BIOREGULATOARE  
ASUPRA CARACTERELOR MORFOLOGICE ȘI DECORATIVE LA  
*ALLIUM* 'PURPLE RAIN'**

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**Abstract.** *The experiment was carried out to study the effect of the bioregulators on the Allium 'Purple Rain' ornamental onion cultivar morphologic and decorative traits. There were used two bioregulators, represented by a growth retardant, Cycocel (CCC), and a growth stimulant, gibberellic acid (GA<sub>3</sub>), applied by foliar spraying, in three concentrations (250, 500 and 1000 ppm). The plants were studied during the vegetation season. They were evaluated regarding the main morpho-decorative characters, like leaves length, leaves number/plant, flowering stem height, inflorescence diameter, bulbs weight/plant and bulbs number/plant. The aim of this study was to indentify the product and concentration witch favors the studied characters. The most favorable influence was registered in case of the GA<sub>3</sub> treatment, in the 500 ppm doses for leaves length, flower stem length, inflorescence diameter and bulb yield and CCC, in 500 ppm doses for bulbs number.*

**Key words:** *Allium 'Purple Rain', bioregulators, morpho - decorative characters*

**Rezumat.** *Experimentul a fost realizat pentru a studia efectul substanțelor bioregulatoroare asupra caracterelor morfo-decorative la cultivarul de ceapă ornamentală Allium 'Purple Rain'. Substanțele bioregulatoroare utilizate au fost reprezentate de un retardant de creștere, cycocel (CCC), și un stimulator de creștere, acidul giberelic (GA<sub>3</sub>). Acestea au fost aplicate prin pulverizare foliară, în trei concentrații (250, 500 și 1000 ppm). Plantele au fost studiate pe durata sezonului de vegetație și au fost evaluate din punct de vedere al caracterelor morfologice și ornamentale: lungimea frunzelor, numărul de frunze, înălțimea tijeii florifere, diametrul inflorescențelor, masa totală și numărul de bulbi/plantă. Scopul lucrării a urmărit identificarea celei mai favorabile substanțe bioregulatoroare și a concentrației aferente, pentru caracterele cultivarului studiat. Tratamentele cu GA<sub>3</sub> 500 ppm au favorizat creșterea în lungime a frunzelor și a tijeii florifere, diametrul inflorescenței și producția de bulbi, iar tratamentele cu CCC 500 ppm au favorizat numărul de bulbi/plantă.*

**Cuvinte cheie:** *Allium 'Purple Rain', substanțe bioregulatoroare, caractere morfo - decorative*

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## INTRODUCTION

The *Allium* genus includes about 750-860 species of perennial, monocots plants. Extensively studied, *Allium* genus is one of the most diverse, including not only plant food and medicinal qualities, but also with ornamental characters (Stearn, 1992; Gregory *et al.*, 1998, cited by Harding, 2004).

The *Alliums* earned in the latest decades, an important economic trait, for the ornamental species which belong to this genus. Many studies from the ornamental plants area shows a very significant increase of the interest on the ornamental species and cultivars of this genus, not only for gardeners but also for florists, due to the colours ranges, shapes and resistance as cut flowers (Harding, 2004; Szot *et al.*, 2009).

The growth regulators substances have a wide practicability in the horticulture area and in particular for the floriculture area. The Cycocel (CCC) and the gibberellic acid (GA<sub>3</sub>) are two much studied growth regulators in the last years, due to their effects over the vegetative growing and generative processes (Zheng *et al.*, 2012).

Many studies shown that the exogenous gibberellins, can induce the flowering for lots of long day flowering plants. But outstanding results can be obtained at the *Liliaceae* plant family (Harding, 2004).

Regarding the Cycocel growth retardant, Nidhish *et al.* (2014) shows that the leaves number of plants can be increased at the *Allium* genus (*Allium sativum*), by applying treatments with CCC, in 1000 ppm concentration.

## MATERIAL AND METHOD

The experiment was conducted in the period 2014–2016, during two experimental years, in the field of Floriculture discipline, from the University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania.

As biological material, was used the ornamental onion cultivar, *Allium* 'Purple Rain'. This is a cultivar obtained from *Allium* 'Purple Sensation' and *Allium afflatunense*, witch have very decorative deep purple, star shaped flowers, grouped in simple spherical umbels. Its long leaves are grown around the stem base. The stem can grow to 60-80 cm height. Every year, the plants generate new bulbs which can be separated and replanted. The flowering period occur between April and June. The *Allium* 'Purple Rain' bulbs were planted in open field, every year in the autumn, in a well drained loose garden soil. The experiment was organized in randomized blocs design, with three replications. A plot has a surface of 3 m<sup>2</sup> and it was planted with 30 bulbs. Before planting, the bulbs were disinfected with Kaptan (1%) and Topsin (0,7%) solutions and during the frosty season the plots were mulched with dried vegetal material (straw). The experimental factors were represented by two growth regulators, a growth stimulant (gibberellic acid - GA<sub>3</sub>) and a growth retardant (cycocel CCC) in 250, 500 and 1000 ppm concentrations (tab. 1).

The treatments were made by foliar application, in the vegetative growth period, twice per season, repeated at two weeks. Thus, in 2014-2015, the first treatment was made on 24<sup>th</sup> March and the second on 7<sup>th</sup> April and in the year 2015-2016, the first treatment was made on 22<sup>nd</sup> March and the second on 5<sup>th</sup> April.

Table 1

**Experimental variants**

Experimental factors	Specification	Variant/Graduations
Control	Untreated	V <sub>1</sub>
Growth Stimulant	Gibberellic Acid - GA <sub>3</sub>	V <sub>2</sub> - 250 ppm
		V <sub>3</sub> - 500 ppm
		V <sub>4</sub> - 1000 ppm
		V <sub>5</sub> - 250 ppm
Growth Retardant	Cycocel - CCC	V <sub>6</sub> - 500 ppm
		V <sub>7</sub> - 1000 ppm

The plants were studied thru the biometric measurements and determinations, regarding the main morpho-decorative characters, like leaves length, leaves number, flower stem height, umbel diameter and new bulbs yield (number/plant and weight/plant). The experimental data was processed using analysis of variance, which established limits of probability for each planting times, compared with the control (untreated variant). The significance of the differences was assessed by taking into account the LSD test (Săulescu and Săulescu, 1967).

**RESULTS AND DISCUSSIONS**

Even if the ornamental *Alliums* are known to be decorating thru the inflorescences lot of them have very aesthetic foliage, regarding the colour or shape. In the last years, many studies were conducted for establish the influence of the bioregulators over the morphologic and decorative traits of the *Alliums* foliage. Maji *et al.* (2015) highlight the vegetative growing increase (plant height, leaves number, leaves length) by applying GA<sub>3</sub> in different concentrations.

For 'Purple Rain' cultivar, the treatment with GA<sub>3</sub> and CCC had different influences over the foliage. In this article, were studies characters like leaves length and number/plants.

From the results synthesis, the leaves length was increased, as compared with the control (untreated variant), in case of the GA<sub>3</sub> treatments. Regardless the concentration, the differences were very significant positive. But the CCC treated variant, did not registered significant differences as compared with the control (tab. 2). Regarding the leaves number, the differences were not significant, in case of the two bioregulators used. But it can be observed that the plants treated with GA<sub>3</sub> obtained a small increase, as compared with the control (tab. 2).

At the 'Purple Rain' cultivar, the GA<sub>3</sub> treatment determined the flower stem increase proportionally with the concentration, up to 500 ppm (54.23 cm and 55.03 cm), the differences being very significant positive, as compared with the control (49.57 cm). At 1000 ppm concentration, they decrease slowly (53.00 cm), with a distinct positive difference (tab. 3).

Table 2

The bioregulators influence of over the *Allium`Purple Rain`* foliage characters

Variant	Leaves length (cm)	±d (cm)	Leaves number/plant	±d (no.)
V <sub>1</sub> –control	44.53	-	5.40	-
V <sub>2</sub> – GA <sub>3</sub> 250 ppm	50.17 <sup>***</sup>	5,63	5.57 <sup>ns</sup>	0,17
V <sub>3</sub> – GA <sub>3</sub> 500 ppm	52.50 <sup>***</sup>	7,97	5.93 <sup>ns</sup>	0,53
V <sub>4</sub> –GA <sub>3</sub> 1000 ppm	51.17 <sup>***</sup>	6,63	5.77 <sup>ns</sup>	0,37
V <sub>5</sub> –CCC 250 ppm	45.00 <sup>ns</sup>	0,47	5.30 <sup>ns</sup>	-0,01
V <sub>6</sub> –CCC 500 ppm	44.00 <sup>ns</sup>	-0,53	5.23 <sup>ns</sup>	-0,17
V <sub>7</sub> –CCC 1000 ppm	43.80 <sup>ns</sup>	-0,73	5.17 <sup>ns</sup>	-0,23
	LSD 5%	1.29		0.74
	LSD 1%	1.81		1.03
	LSD 0.1%	2.56		1.76

Table 3

The bioregulators influence over the *Allium`Purple Rain`* flower stems and inflorescences

Variant	Flower stem length (cm)	±d (cm)	Inflorescence diameter (cm)	±d (cm)
V <sub>1</sub> –control	49.57	-	23.00	-
V <sub>2</sub> – GA <sub>3</sub> 250 ppm	54.23 <sup>***</sup>	4,67	24.67 <sup>**</sup>	1,67
V <sub>3</sub> – GA <sub>3</sub> 500 ppm	55.03 <sup>***</sup>	5,47	25.67 <sup>***</sup>	2,67
V <sub>4</sub> –GA <sub>3</sub> 1000 ppm	53.00 <sup>**</sup>	3,43	25.50 <sup>***</sup>	2,50
V <sub>5</sub> –CCC 250 ppm	50.00 <sup>ns</sup>	0,43	23.10 <sup>ns</sup>	0,10
V <sub>6</sub> –CCC 500 ppm	49.80 <sup>ns</sup>	0,23	22.67 <sup>ns</sup>	-0,33
V <sub>7</sub> –CCC 1000 ppm	48.63 <sup>ns</sup>	-0,93	22.33 <sup>ns</sup>	-0,67
	LSD 5%	0.74		1.16
	LSD 1%	1.03		1.63
	LSD 0.1%	1.46		2.29

The CCC treatments did not influence the stem length, the differences being statistically insignificant.

With respect at the inflorescence diameter, the increases follow the same trend as the flower stem length. The diameter increase was more favored more by the gibberellic acid, particularly in 500 and 1000 ppm concentrations, which have determined increases with 10.9-11.9% against the control, the differences being very significant. Positive effect has been noticed also at the low doses (250 ppm), but more reduced (with 7.2% above the untreated variant).

The plants treated with CCC had the tendency of reduction the inflorescence diameter, excepting the lowest dose (250 ppm), which led to increase with 0.4% toward the control (untreated variant), but the differences were statistically insignificant.

Regarding the bulb yield, Pogroszevska *et al.* (2007) discovered that the GA<sub>3</sub> treatment increased the new bulbs number at *Allium moly*.

The bulbs weight/plant ranged from 140.7 g (CCC 1000 ppm) up to 190.3 g (GA<sub>3</sub> 500 ppm). The only type of treatment which led to a significant difference was GA<sub>3</sub> 500 ppm (tab. 4).

The bulbs number/plant was influenced different by the treatments. The plant had positive answer for the both treatments. In case of GA<sub>3</sub>, the most favorable result was registered at 500 ppm dose, were the plants formed 12.33 bulbs/plant, the difference being very significant positive.

Table 4

The bioregulators influence over the *Allium* 'Purple Rain' bulbs yield

Variant	Bulbs weight/plant (g)	±d (g)	Bulbs number/plant	±d (no.)
V <sub>1</sub> –control	154.30	-	8.17	-
V <sub>2</sub> – GA <sub>3</sub> 250 ppm	166.17 <sup>ns</sup>	12,13	10.90*	2,17
V <sub>3</sub> – GA <sub>3</sub> 500 ppm	190.33***	36,30	12.33***	4,17
V <sub>4</sub> –GA <sub>3</sub> 1000 ppm	162.67 <sup>ns</sup>	8,63	9.00 <sup>ns</sup>	0,83
V <sub>5</sub> –CCC 250 ppm	155.67 <sup>ns</sup>	1,63	10.73*	2,57
V <sub>6</sub> –CCC 500 ppm	164.67 <sup>ns</sup>	10,63	12.83***	4,67
V <sub>7</sub> –CCC 1000 ppm	140.70 <sup>ns</sup>	-13,27	8.20 <sup>ns</sup>	0,03
	LSD 5%	15.35		2.03
	LSD 1%	21.56		2.86
	LSD 0,1%	30.41		4.03

In case of CCC treatments, the highest number of bulbs were obtained at the 500 ppm dose, too (12.83 bulbs/plant) and the difference was very significant positive (tab. 4).

## CONCLUSIONS

1. The bioregulators have different influences over the morphologic and decorative traits at the ornamental onion 'Purple Rain' cultivar.

2. The GA<sub>3</sub> treatments have positive influence regarding the leaves length, the flower stem length inflorescence diameter and bulbs yield like number and weight/plant.

3. The CCC treatments lead to favorable results only regarding the bulbs number/plant. Excepting this character, it has insignificant influences over the morpho-decorative characters of 'Purple Rain' cultivar.

4. Based on the results obtained in this study, it can be recommended for the 'Purple Rain' cultivar, the GA<sub>3</sub> application, in 500 ppm, for decorative traits improvement and CCC in 500 ppm doses for the bulbs number improvement.

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## EFFECT OF BIOREGULATORS TREATMENTS ON *ALLIUM SAXATILE* M. Bieb.

### EFFECTUL BIOREGULATORILOR LA *ALLIUM SAXATILE* M. Bieb.

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**Abstract.** This research was conducted to investigate the influence of the bioregulators on the *Allium saxatile* M.Bieb. vegetative growth and flowering period. There were used two bioregulators, represented by a growth retardant, Cycocel (CCC) and a growth stimulant, Gibberellic Acid (GA<sub>3</sub>), applied by foliar spraying, in three concentrations (250, 500 and 1000 ppm). The plants were evaluated with reference to the vegetative growth, like leaves length, leaves number, flowering stem height, inflorescence diameter and with reference to the flowering period. The analysed results, highlighted that application of GA<sub>3</sub> promote the vegetatives growth. The treatments with CCC had a noticeable influence in the flowering period delay. Therefore, treated with CCC, the plants bloomed ealier with about 10-20 days than the other variants.

**Key words:** *Allium saxatile* M. Bieb., plant bioregulators, vegetative growth

**Rezumat.** Acest studiu a fost realizat în scopul investigării influenței substanțelor bioregulate asupra creșterii vegetative și perioadei de înflorire la *Allium saxatile* M.Bieb. Au fost utilizate două sunstanțe regulate de creștere, un retardant de creștere (CCC) și un stimulator de creștere (GA<sub>3</sub>), aplicate prin pulverizare foliară, în trei concentrații (250, 500 și 1000 ppm). Plantele au fost evaluate din punct de vedere al creșterilor vegetative, la nivelul lungimii frunzelor, numărului de frunze, lungimea tijei florifere, diametrul inflorescenței și din punct de vedere al perioadei de înflorire. Rezultatele analizate, au evidențiat faptul că administrarea de GA<sub>3</sub> îmbunătățește creșterile vegetative. Tratamentele cu CCC au avut o influență notabilă asupra extinderii perioadei de înflorire. Prin urmare, tratate cu CCC, plantele au înflorit mai devreme cu aproximativ 10-20 de zile decât celelalte variante.

**Cuvinte cheie:** *Allium saxatile* M. Bieb., substanțe bioregulate, creștere vegetativă

## INTRODUCTION

*Allium saxatile* M. Bieb. (syn. *Allium globosum* M. Bieb.) is one of the plentiful number of the *Allium* genus species (about 750-860 species of perennial, monocots plants), wildy growing in Europe, from Italy to NW China. In the spontaneous flora of Romania it has the position of a rare species, found on

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craggy highlands, calcareous alpine areas (Oprea, 2005; Zahariadi, 1966; cited by Draghia *et al.*, 2013; Harding, 2004).

In the last years, the ornamental plants of the genus *Allium* became more and more popular, getting in the same time an important economical place like ornamental crop. These species can be used worldwide like garden plant, due to their disease and climate resistance and to the great variety of species and cultivars. They also entered to the cut flowers market because of their resistance as cut flowers, the hardness of flower stems and the wide range of colours and shapes (Harding, 2004; Szot *et al.*, 2009).

The bioregulators substances are widely used in floriculture area. The gibberellic acid ( $GA_3$ ) and cycocel (CCC) were extensively studied in the last years, due to their effects over the vegetative growing and generative processes (Zheng *et al.*, 2012).

Nidhish *et al.* (2014) discovered that the CCC treatment in 1000 ppm concentration, can increase the leaves number of plants.

The exogenic gibberellins can promote flowering for a very wide range of long day flowering plants. In case of *Allium karataviense* 'Ivory Queen' it was observed the inflorescence shoot elongation and the increased number of flowers in inflorescence (Pogroszewska, 2007).

## MATERIAL AND METHOD

The research was carried out in the period 2014–2016, during two experimental years, in the field of Floriculture discipline, from the University of Agricultural Sciences and Veterinary Medicine of Iași, Romania.

The biological material was represented by the *Allium saxatile* M. Bieb. species. This *Allium* species has a great ornamental potential. The flowers are grouped in spherical umbels, light purplish-pink, the leaves are simple, thin, light green. It grows in the areas with plenty of sun, in well drained, sandy or craggy soils. It blooms in the summer months, from June to August. The plants persist at the crop place all over the year, in the winter they stay in a latent vegetation rest.

The experiment was organized in randomized blocs design, with three replications. A plot has a surface of 3.00 m<sup>2</sup> and it was planted with 45 plants. Before planting, the bulbs were disinfected with Kaptan (1%) and Topsin (0.7%) solutions and during the frosty season the plots were mulched with dried vegetal material (straw). The experimental factors were represented by two growth regulators, a growth stimulant (gibberellic acid -  $GA_3$ ) and a growth retardant (cycocel CCC) in 250, 500 and 1000 ppm concentrations (table 1).

The treatments were made by foliar application, in the vegetative growth period, twice per season, repeated at two weeks. Thus, in 2014-2015, the first treatment was made on 6<sup>th</sup> May and the second on 20<sup>th</sup> May and in the year 2015-2016, the first treatment was made on 4<sup>th</sup> May and the second on 18<sup>th</sup> May.

The plants were investigated thru the biometric measurements and determinations, for the vegetative growth, like leaves length, leaves number, flower stem height, umbel diameter and regarding the flowering period. The experimental data was processed using analysis of variance, which established limits of probability for each planting times, compared with the control (untreated variant). The

significance of the differences was assessed by taking into account the LSD test (Săulescu and Săulescu, 1967).

Table 1

Experimental variants		
Experimental factors	Specification	Variant/Graduations
Control	Untreated	V <sub>1</sub>
Growth Stimulant	Gibberellic Acid - GA <sub>3</sub>	V <sub>2</sub> - 250 ppm
		V <sub>3</sub> - 500 ppm
		V <sub>4</sub> - 1000 ppm
Growth Retardant	Cycocel - CCC	V <sub>5</sub> - 250 ppm
		V <sub>6</sub> - 500 ppm
		V <sub>7</sub> - 1000 ppm

## RESULTS AND DISCUSSIONS

For many species of the genus *Allium*, the foliage is very perishable, getting dry before the plant are getting flowers. This character has a bad influence over the aesthetic aspect of the *Alliums*, for many species and cultivars, being necessarily a combination with a leaves resistant plant. So, the studies shown that the bioregulators can have good influence over the *Alliums* foliage, regarding the vegetative growing (leaves number, leaves length, plant height) (Maji et al., 2015).

The analysis of the *Allium saxatile* foliage shows that the plants answered most for the leaves length and less for the leaves number, under the bioregulators influence. The leaves length varied between 18.3 and 26.5 cm, the untreated variant registering around 18.6 cm (tab. 2).

Table 2

The bioregulators influence of over the *Allium saxatile* foliage characters

Variant	Leaves length (cm)	±d (cm)	Leaves number/plant	±d (no.)
V <sub>1</sub> —control	18,60	-	5.77	-
V <sub>2</sub> — GA <sub>3</sub> 250 ppm	26,30***	-	5.77 <sup>ns</sup>	0,0
V <sub>3</sub> — GA <sub>3</sub> 500 ppm	26,50***	7,70	5.83 <sup>ns</sup>	0,07
V <sub>4</sub> —GA <sub>3</sub> 1000 ppm	21,50*	7,90	5.80 <sup>ns</sup>	0,03
V <sub>5</sub> —CCC 250 ppm	19,50 <sup>ns</sup>	2,90	5.50 <sup>ns</sup>	-0,27
V <sub>6</sub> —CCC 500 ppm	18,87 <sup>ns</sup>	0,90	5.83 <sup>ns</sup>	0,07
V <sub>7</sub> —CCC 1000 ppm	18,33 <sup>ns</sup>	0,27	5.50 <sup>ns</sup>	-0,27
	LSD 5%	2.33		0.55
	LSD 1%	3.27		0.77
	LSD 0.1%	4.61		1.09

The GA<sub>3</sub> treatment have led to increase the leaves length, in the higher proportion for the 250 and 500 ppm doses, with around 41-42% toward the control and very significant positive differences. The influence was less for the 1000 ppm, with 15% toward the control. The CCC treatment determined the inhibition of the leaves growing for *Allium saxatile* (tab. 2).

The number of leaves/plant was relatively closed between the variants and had not been influenced by any of the two products used into the experiment. This is indicated by the insignificant differences between the treated variants and control (tab. 2).

For *Allium saxatile*, it has been demonstrated that the bioregulators may have a much higher influence over the flower stem height. From the presented data, in the table 3, it should be noted that the flower stem vary between 18.5 and 43.3 cm.

Table 3

The bioregulators influence over the *Allium saxatile* flower stem and inflorescence

Variant	Flower stem length (cm)	±d (cm)	Inflorescence diameter (cm)	±d (cm)
V <sub>1</sub> –control	23.00***	-	2.77	-
V <sub>2</sub> – GA <sub>3</sub> 250 ppm	36.07***	13,7	2.80 <sup>ns</sup>	0,03
V <sub>3</sub> – GA <sub>3</sub> 500 ppm	40.10***	17,10	3.00 <sup>ns</sup>	0,23
V <sub>4</sub> –GA <sub>3</sub> 1000 ppm	43.33***	20,33	3.00 <sup>ns</sup>	0,23
V <sub>5</sub> –CCC 250 ppm	22.80 <sup>ns</sup>	-0,20	2.57 <sup>ns</sup>	-0,20
V <sub>6</sub> –CCC 500 ppm	20.40 <sup>ns</sup>	-2,60	3.10 <sup>ns</sup>	0,33
V <sub>7</sub> –CCC 1000 ppm	18.50 <sup>o</sup>	-4,50	3.27 <sup>ns</sup>	0,50
	LSD 5%	4.37		0.69
	LSD 1%	6.13		0.97
	L SD 0.1%	8.66		1.37

The gibberellic acid has the most powerful action at 1000 ppm doses, causing the stem increase with about 88% towards the control (untreated plants). For all the three GA<sub>3</sub> doses, the differences 13.7 and 20.3 cm were very significant.

Under the cycocel influence the flower stem length has been reduced from 22.8 cm at the 250 ppm to 18.5 cm at the 1000 ppm doses.

As regards the diameter of the inflorescences, the bioregulators treatments did not indicated notable differences. The very small differences are insignificant as compared with the control, but it can be noticed a positive effect under the GA<sub>3</sub> treatments and also under CCC, on the inflorescences sizes compared to control, excepting the 250 ppm dose of CCC (tab. 3).

From the phenological diagram (fig.1) regarding on the treatment influence over the *Allium saxatile* are represented the monthly period of the phenophases.

As it can be seen, for the CCC treated plants, the full flowering got earlier with around 20 days than the control and with around 10 days earlier than the GA<sub>2</sub> treated plants (GA<sub>3</sub> also extended the flowering period, but less than CCC).

Treatment	Month/Decade																							
	II			III			IV			V			VI			VII			VIII					
	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3	1	2	3			
Control																								
GA <sub>3</sub> 250																								
GA <sub>3</sub> 500																								
GA <sub>3</sub> 1000																								
CCC 250																								
CCC 500																								
CCC 1000																								

**Fig.1** The phenological diagram regarding the influence of treatments at *Allium saxatile*

**Legend:**



Vegetative stage



Flowering

The flowering period ended, for all the variant in the same time, around the middle of the august month (fig. 1).

## CONCLUSIONS

1. The bioregulators can improve the quality of *Allium saxatile* specie, having different influence. The gibberellic acid stimulates the vegetative growing and CCC over delay flowering period.

2. By applying GA<sub>3</sub> in 250 and 500 ppm doses, the *Allium saxatile* foliage can be improved, regarding the leaves length. For the flower stem and inflorescence diameter increasing, there were necessary higher doses (500 and 1000 ppm).

3. The CCC treatment have influence more over the flowering period, by extended it, than over the morphologic characters. Thus, the plants which were treated with CCC, flourished earlier with around 20 days than the control and 10 days than the GA<sub>3</sub> treated plants.

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## A STUDY ON THE SEED GERMINATION AND PLANTLETS SPRING AT RHUBARB SPECIES (*RHEUM RHABARBARUM* L.)

### STUDII ASUPRA GERMINAȚIEI SEMINȚELOR ȘI RĂSĂRIII PLANTELOR DE REVENT (*RHEUM RHABARBARUM* L.)

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**Abstract.** Seed germination and plantlets spring at rhubarb species were studied in the standard conditions on a peat substrate. Results revealed that the germination rate was of 90%, and spring rate was of 82%. Germination period had 12 days, starting in the 7-th day of experiment, and the spring was done in 10 days.

**Key words:** rhubarb, cultivar, seeds germination

**Rezumat.** Germinația semințelor și răsării plantelor de revent au fost studiate în condiții standard de lucru, pe un strat de turbă. Rezultatele arată că rata de germinare a fost de 90%, iar cea de răsărire de 82%. Germinația a durat 12 zile, începând cu a șaptea zi de la semănat, iar răsărirea a avut loc în 10 zile.

**Cuvinte cheie:** rubarbă, cultivar, germinarea semințelor

## INTRODUCTION

The culture of rhubarb in Romania has a tradition less developed and less known (Ciofu *et al.*, 2004). The establishment of this culture is the key to promoting the rhubarb, with the knowledge of how to use this herb in the human nutrition (Stan *et al.*, 2003; Henriksen and Bjorn, 2004).

The knowledge regarding germination helps us define and calculate seed cultural value and the knowledge regarding the emergence process provides information on seed strength, as well as on germ ability to evolve into seedlings (Munteanu and Falticeanu, 2008).

As biological material for the establishment of a culture, there can be used seedlings or cutting roots (Ivanova, 1975; Frenz and Lechl, 1977). The process of getting seedlings requires some knowledge on the processes of rhubarb seed germination and the way in which plant emergence arises. Responding to this need, in this paper we proposed as objective the study of rhubarb seed germination.

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## MATERIAL AND METHOD

The biological material in this experiment is the seeds of rhubarb.

The substrate germination was red composted peat, with the adjusted pH to 6.5 and eutrophized according to the standards of a manufacturing company.

The study of seed germination – the determination of seed germination and plant emergence was necessary to have enough information to assess seeds quality.

The germination temperature varied between 20-22°C, the humidity was approximately 70% (68-71%), and the light had an intensity of 3000 lux (Stan and Stan, 2010). During experimentation they have not been made any additional watering. The ventilation was automated for the regulation and equalise of ambient temperature germination.

The evaluation of the germination and emergence of seeds and plants was carried out using the following indicators: the rate of germination / emergence, growth rate of germination / emergence, the velocity coefficient and the velocity of germination and germination / emergence.

From the start of plant emergence observations and determinations have been made about the dynamics of emergence until the moment when 2-3 consecutive measurements showed the same number or percentage of emerged plants (fig. 1).



**Fig. 1** Plant emergence (original photo)

The germination rate is the percentage of emerged plants every day in part or percentage of plants emerged from one day to another.

The velocity of germination (the germination velocity or speed of germination) represents the percentage of germinated plants per unit of time (day) and is calculated using the following formula:

$$V_G = \frac{G_i}{n}, \text{ where:}$$

$G_i$  = germination at a certain date;

$n$  = number of days in which germination was achieved  $G_i$

The coefficient of germination velocity represents the germination velocity relative to the final germination of seeds and it is calculated using the following formula:

$$CV_G = \frac{G_i}{G_f \cdot n} \times 100, \text{ where:}$$

$G_i$  = germination at a certain date;

$n$  = number of days in which germination was achieved  $G_i$

$G_f$  = the final germination.

## RESULTS AND DISCUSSIONS

Results of seed germination and emergence of rhubarb seedlings.

The study of rhubarb seed germination and emergence of seedlings has highlighted a number of elements relevant for the planting rhubarb production technology.

The results obtained are presented analytically in table 1.

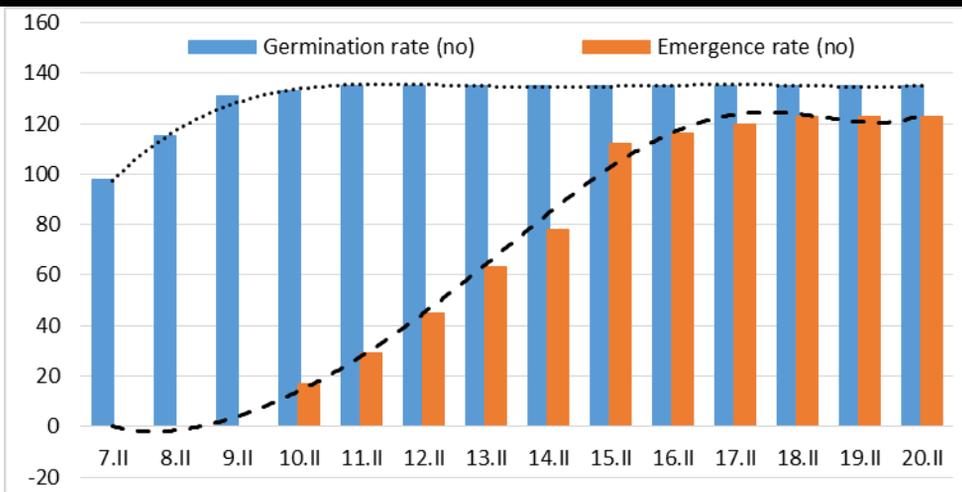
The seeds germination began after seven days from sowing (from 07/02/2013) and lasted four days, up to 11.02. From a total of 150 seeds the germination process by that date began only for 135 seeds, so a rate of 90%. The rate of 50% was achieved in the first observation germination (7.02), when, in fact, the percentage of germinated seeds was 65%.

Table 1

Results regarding the germination and plant emergence on a peat substrate  
(150 seeds, sown on 1.02.2013)

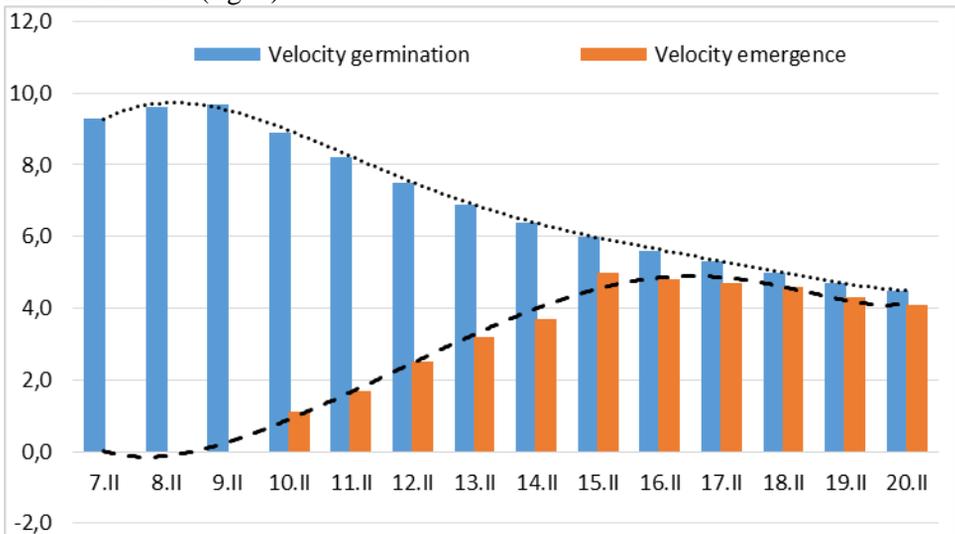
Indicators		Date of observation and calculations calendar													
		7.II	8.II	9.II	10.II	11.II	12.II	13.II	14.II	15.II	16.II	17.II	18.II	19.II	20.II
Germination rate	number	98	115	131	133	135	135	135	135	135	135	135	135	135	135
	%	65	77	87	89	90	90	90	90	90	90	90	90	90	90
Emergence rate	number	-	-	-	17	29	45	63	78	112	116	120	123	123	123
	%	-	-	-	11	19	30	42	52	75	77	80	82	82	82
Velocity germination		9.3	9.6	9.7	8.9	8.2	7.5	6.9	6.4	6.0	5.6	5.3	5.0	4.7	4.5
Velocity emergence		-	-	-	1.1	1.7	2.5	3.2	3.7	5.0	4.8	4.7	4.6	4.3	4.1
Germination velocity coefficient (%)		10.3	10.7	10.7	9.9	9.1	8.3	7.7	7.1	6.7	6.3	5.9	5.6	5.3	5.0
Emergence velocity coefficient (%)		-	-	-	1.34	2.11	3.05	3.94	4.53	6.10	5.87	5.74	5.56	5.26	5.0

The germination dynamics, for entire period of observations, from 1.02 to 20.02 is graphically represented in figure 2, from which we may observe an increase in the germination rate from the first observation made (7.02) until the maximum germination rate of 90%, registered on 11.02. The data from the table, as a graphic representation, after three consecutive observations, starting from 11.02, demonstrates that the germination stopped and therefore the rhubarb seeds from the Local population had a germination of 90%.



**Fig. 2** The graphic representation of the germination and plant emergence rate

The analysis of germination speed, expressed by the velocity of germination, shows that it increases from 9.3 from the first observation (7.02), up to 9.7 at the third observation, then it decreases to 8.2 when germination has the maximum value (fig. 3).

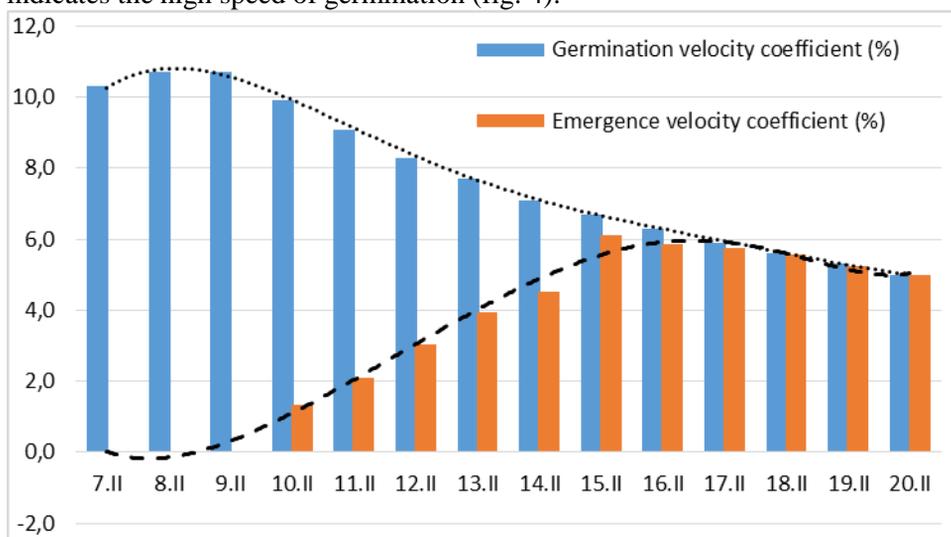


**Fig. 3** The graphic representation of the germination and plant emergence velocity

So, under these circumstances rhubarb germination begins after seven days and lasts for 10-11 days, which allows us to state that rhubarb is a plant with rapid germination, for example, like species from the class of cabbages.

The coefficient of velocity of germination has values ranging between 9.1% germination of 10.7%. These figures confirm a high rate of germination,

sprouting like species with a relatively early germination. For example, these values are comparable (or even superior) to tomatoes, to which Stan (2010) reports a maximum coefficient of 8.96% for peat substrate. For the values on germination rate from the first day, the ratio is 10.3% and in the next two days reaches the maximum value of 10.7%, but in the day of 11.02 while recording the final germination 90%, the velocity ratio has a value of 9.1%, a value which indicates the high speed of germination (fig. 4).



**Fig. 4** The graphic representation of the germination and plant emergence velocity Coefficient

The characterization of plant emergence on a peat substrate results analytically from table 1.

The plant emergence starts from the tenth day from sowing, respectively from the fourth day from germination. I have considered the plant to be emerged if it already has the two primary leaves above the soil surface.

The plant emergence lasts quite some time, that is eight days, and starting from the ninth day, a maximum number of emerged plants is registered, meaning 82%. The plant emergence has an ascending rhythm, starting from 11% until 82%, compared with the total number of seeds placed for germination. Compared with the germination, which has a 90% rate, the plant emergence has a rate of 82%, which means that 8% of germs did not have the capacity to reach emergence.

The speed of emergence is relatively small at the beginning of the emergence period and it reaches values of over 50%, after the fifth day from emergence, respectively eight days at the beginning of germination and 14 days from sowing. The velocity grows from 1.1, in the first day, to up to 5.0, after 15 days from the moment when the seeds have been placed for germination.

The velocity coefficient, which offers us an image taking also into account the final percentage of emerged plants, has quite small values during the first days, which results into a small emergence, starting from values of 1.34% up to 6.10%, in the 15th day from sowing and 6th day from the onset of emergence, respectively.

The results presented demonstrate a fast germination (4-5 days), but the emergence is slower – 8-9 days, which shows that the germ, as well as the seed, has a more reduced emergence force, exposing the seedlings for more time to possible adverse factors.

## CONCLUSIONS

1. The seed germination started from 7 days from sowing (on 7.02.2013) and lasted 4 days, until 11.02, which shows that the rhubarb is a plant with fast germination.

2. The plant emergence starts from the 10th day from sowing, respectively the 4th day from germination. I have considered the plant to be emerged if it already has both cotyledons above the soil surface.

3. The results presented demonstrate a fast germination (4-5 days) but the emergence is slower – 8-9 days, which shows that the germ, as well as the seed, has a more reduced emergence force, exposing the seedlings for more time to possible adverse factors.

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## STUDIES ON GROWTH AND FLOWERING OF *LILIUM REGALE* SPECIES IN CONDITIONS OF FERTILIZATION WITH OSMOCOTE

### STUDII PRIVIND CREȘTEREA ȘI ÎNFLORIREA SPECIEI *LILIUM REGALE* ÎN CONDIȚIILE FERTILIZĂRII CU OSMOCOTE

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**Abstract.** *The aim of the experiment was to evaluate the effect of Osmocote Pro fertilizer on the growth and flowering of the species Lilium regale. The experience has been organized in four variants with three repetitions: V1- without fertilizer, V2-4g Osmocote/plant; V3-6g Osmocote/plant and V4-8 g Osmocote/plant. The plants' growth in height and their ability of flowering have been stimulated by the fertilizer regardless of the dose used. Compared with the control variant, the increase in concentration caused an increase in the height of the plants, the number of flowers, the diameter and length of the cups.*

**Key words:** *Lilium regale*, Osmocote, assimilating pigments

**Rezumat.** *Scopul experimentului a fost de a evalua efectul aplicării îngrășământului Osmocote Pro asupra creșterii și înfloririi speciei Lilium regale. Experiența a fost organizată, în patru variante cu trei repetiții : V<sub>1</sub>- fără îngrășământ, V<sub>2</sub>- 4g Osmocote/plantă; V<sub>3</sub>- 6g Osmocote/plantă și V<sub>4</sub>- 8g Osmocote/plantă. Creșterea în înălțime și capacitatea de înflorire a plantelor au fost stimulate prin aplicarea îngrășământului indiferent de doza utilizată. Comparativ cu varianta martor, mărirea concentrației a indus un spor de creștere privind înălțimea plantelor, numărul de flori, diametrul și lungimea cupelor.*

**Cuvinte cheie:** *Lilium regale*, Osmocote, pigmenți asimilatori

## INTRODUCTION

The genus *Lilium* contains many species originating from different climatic zones, which explain the differences in behaviour of the species (Băla, 2007; Draghia and Chelariu, 2011). Lilies have a long history as ornamental plants and currently they are cultivated in large numbers Woodcock *et al.* (1950). Lately, there appeared on the market different hybrids that are very impressive through such qualities as the multitude of colours and their scent, while the different methods of culture turned the lily into one of the most beloved flowers in the contemporary society. The multitude of species and varieties make this plant to be one of the most valuable species of flowering bulbs used in landscape design and determine their name as everlasting flowers due to their life-

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length (Siljak-Yakovlev *et al.*, 2003). However, in the commercial horticultural industry, native species have preserved their place among the new hybrids due to their potential in creating new crops meant for landscaping design and those intended for the production of cut flowers (Pelkonen *et al.*, 2012).

## MATERIALS AND METHODS

Researches were conducted during the period October 2013 - November 2015. The experiment was organized using bulbs of *Lilium regale* as biological material acquired from a firm that is specialized in the production of flowering seedling material. The experiment was organized in randomized blocks with 4 variations and 3 repetitions each, each repetition consisting of 10 plants. In order to establish the plants' capacity of growth under conditions of fertilization, there has been tested the Osmocote®Pro fertilizer with controlled release, a high content of NPK (N11:P10:K19:2MgO + ME) and longevity for 5-6 months, being specially designed for the culture of flowering plants. In order to determine the influence of controlled-release fertilizer on the development of plants there have been administered different concentrations of fertilizer on the experimental variants while the control variant was left unfertilized.

The doses of fertilizer administered for each plant were: variant  $V_1$  was not fertilized, being the control variant; variant  $V_2$  was fertilized with Osmocote®Pro 4 g/each plant; variant  $V_3$  was administered Pro®Osmocote fertilizer 6 g/each plant and the variant  $V_4$  was administered Osmocote®Pro 8 g/each plant. It has been studied the influence of fertilization with Osmocote®Pro on the performance of vegetation phenophases while the phenological determinations focused on vegetation, the emergence of floral buds, the beginning and ending of flowering. Also, to highlight the influence of Osmocote®Pro fertilizer on the ornamental characteristics of the plants, there have been carried out biometric measurements throughout the period of vegetation, including measurements related to plants' height, diameter and number of flowers per plant.

## RESULTS AND DISCUSSION

The observations conducted in order to determine the influence of fertilization with Osmocote®Pro on the development of vegetation phenophases in the species *Lilium regale* showed an early start with the increase of the concentration of fertilizer (fig.1). The earliest vegetation was observed in the group of plants from the variants  $V_3$  and  $V_4$ , which were fertilized with the largest doses of fertilizer - they started to germinate with up to 12 days earlier as compared to the control group ( $V_1$ ). In  $V_2$ , the plants started to germinate 6 days earlier as compared to the control group. By comparing the results obtained in the four experimental variants, it is established that the application of Osmocote fertilizer had a significant influence on the beginning of germination. The plants of the variant  $V_1$ , which did not benefitted from fertilization, knew a delay in the germination process.

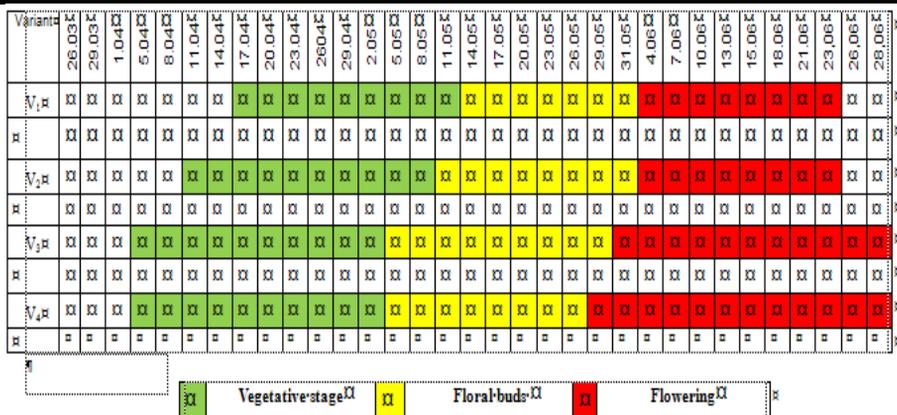


Fig. 1 Phenophases in the species *Lilium regale*

The emergence of floral buds was observed during the period 14-31 May in the group of plants from the unfertilized variant (V<sub>1</sub>), while in the case of V<sub>2</sub>, their occurrence was three days earlier as compared to the control group. In the case of the variants fertilized with the largest doses (V<sub>3</sub>, V<sub>4</sub>), the buds have started to form up to 9 days earlier compared with the plants in the unfertilized group. In the group of plants benefitting of 4 g of Osmocote on each plant, the occurrence of floral buds happened three days earlier as compared to the control group (V<sub>1</sub>) and six days later than in the group of the plants from V<sub>3</sub> and V<sub>4</sub> (table 1). In terms of flowering, it started on June 4th in the control group, while significant differences were observed only in the case of variants V<sub>3</sub> and V<sub>4</sub>. Variant V<sub>3</sub>, benefitting of 6g of fertilizer on each plant bloomed 3 days earlier than the plants from variant V<sub>1</sub>, while the plants from variant V<sub>4</sub>, fertilized with 8 g, and bloomed 6 days earlier. By comparing the results obtained in the group of plants from V<sub>3</sub> and V<sub>4</sub> with the results obtained in the control variant (V<sub>1</sub>), we can observe a delay of the flowering period by up to 12 days.

The influence of the concentrations of fertilizer on plants growth and development was highlighted through biometric measurements that focussed on the height of the floral stem, the length of the buds, the number of flowers in the inflorescence and the diameter of the cup. By comparing the results obtained in the experimental variants, there has been highlighted the positive influence of fertilization on the growth of floral rods in the species *Lilium regale*.

Determinations regarding the average height of the plants, reflects the influence of the concentration of fertilizer on the growth and development of plants (fig. 2). Within the four variants, it can be noted that the greatest increase in the floral rod was obtained in variant V<sub>4</sub> (118 cm), while the lowest increase was obtained in the group of plants from the control variant (82 cm).

Compared to the control variant, the variations in height in all the experimental groups were directly proportional to the increase of the fertilizer

dose, the largest increases being obtained in the group of plants from variant  $V_4$ , in which the dose of fertilizer was 8 g/each plant.

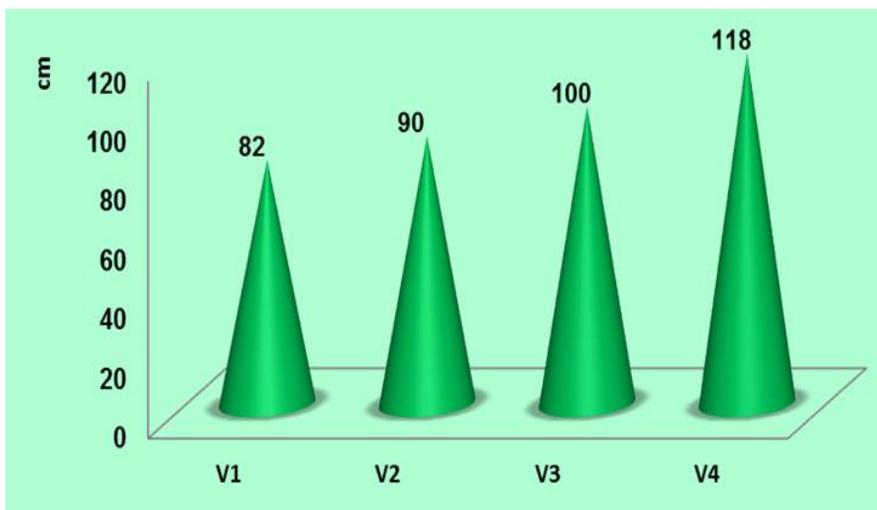


Fig. 2 The average height (cm)

The average plant height in  $V_2$  was 90 cm while in  $V_3$  was 100 cm. By analysing the results there was recorded a variation in the height of the floral stem from the 82 cm in the control group up to 118 cm in  $V_4$ , resulting a difference of 36 cm. Therefore, we can state that fertilization with Osmocote<sup>®</sup>Pro has a significant influence on the increase in height of the floral stem at the species *Lilium regale*.

Observations regarding the influence of fertilization with Osmocote on the formation of flowers at species *Lilium regale* revealed an increase in the number of flowers per plant corresponding to higher dosages of fertilizer administered to each experimental variant (tab. 1). Compared to the control group, the average number of flowers in the inflorescence was 8.2 flowers on each plant and there has not been observed any significant differences in the plants which benefitted of 4 g of Osmocote on each plant and registered an average number of flowers per plant of approximately 10.3.

The highest average number of flowers was obtained in the group of plants from  $V_3$  (with 12.4 flowers on the plant) and  $V_4$  (with 18.5 flowers on the plant). The increase in concentration of Osmocote<sup>®</sup>Pro in the substrate causes an increase of the average number of flowers, each variant recording positive difference as compared to the control group. Observing the tendency of increase in the number of flowers on each plant, we can observe that the variants fertilized with the highest doses recorded a significant increase in their number as compared to the control group.

Thus, the increase of the dosage of Osmocote<sup>®</sup>Pro in substrate caused an increase in the number of flowers as compared to the control sample with 2.1

flowers in the plants from variant V<sub>2</sub>, 4.2 in the plants from variant V<sub>3</sub> and 10.3 flowers in the plants of variant V<sub>4</sub> (tab. 1).

Table 1

The average number of flowers per plant				
Variant	No. flowers (PCs.)	% compared with control	Difference	The significance
V <sub>1</sub>	8.2	100.0	-	16.33
V <sub>2</sub>	10.3	125.6	2.1	xx
V <sub>3</sub>	12.4	151.22	4.2	xxx
V <sub>4</sub>	18.5	225.6	10.3	xxx
LSD 5% 1.2    LSD 1% 1.8    LSD 0.1% 2.9				

± d = difference compared to the control sample

At first sight we can say that fertilizer Osmocote stimulates the formation of flowers on the plants of *Lilium regale* which further increases their ornamental value. On a thorough analysis and comparing our results with those from different studies, the increased number of flowers was commonly attributed to the increase in the concentration of nutrients in the substrate of culture. Through the statistical interpretation of the results, there was observed a very significant positive difference in plants from V<sub>3</sub> and V<sub>4</sub>, while in the group of plants of V<sub>2</sub> there was noted a significant positive difference as compared with the control variant.

In the culture of flowers, a special importance is represented by the morphological side of the plants that gives them their decorative value. In the case of the species *Lilium regale*, the decorative value is largely determined by its flower and less by its conduct. As a result of the research it was found that the Osmocote® Pro fertilizer influences the dimensions of the corolla only in the group of plants fertilized with the largest doses of fertilizer, respectively 6 g and 8 g/each plant (tab.2). It can be observed that in the case of the control variant (V<sub>1</sub>), the average diameter of the corolla is 6.7 cm. As the concentration of fertilizer was increased there could be observed a slight increase in diameter, reaching up to 7.1 cm in V<sub>2</sub>, which resulted in a difference of 0.4 cm as compared to the control group. Compared to the control sample, the plants from variants V<sub>3</sub> and V<sub>4</sub> presented more significant differences, respectively 1.5 cm and 1.8 cm.

Table 2

The average diameter of the corolla (cm)				
Variant	Ø flowers' cup (cm.)	% compared with control	Difference	The significance
V <sub>1</sub>	6.7	100.0	-	c
V <sub>2</sub>	7.1	125.6	0.4	-
V <sub>3</sub>	8.2	151.22	1.5	xx
V <sub>4</sub>	8.5	225.6	1.8	xx
LSD 5% 0.9    LSD 1% 1.4    LSD 0.1% 2.2				

± d = difference compared to the control sample

Through the statistical interpretation of the results obtained as compared to the control group, in the group of plants from  $V_2$  there has been obtained an insignificant statistical difference, but in the case of the plants from variants  $V_3$  and  $V_4$  positive differences have been recorded (table 3). The fertilizer Osmocote®Pro has not influenced very significantly the diameter of the cup in any of the 3 experimental variants. registered a slight elongation of the flower as compared to the uncontaminated group.

### CONCLUSIONS

1. The administration of Osmocote®Pro fertilizer in the culture substrate leads to a significant earlier germination than in the case of the control group that did not benefit from the fertilizer Osmocote®Pro;

2. Fertilization with Osmocote®Pro induces earlier emergence of floral buds with up to 9 days.

3. The flowers belonging to the plants fertilized with the largest doses of Osmocote®Pro fertilizer, respectively  $V_4$  fertilized with 8 g/plant and  $V_3$  fertilized with 6 g/plant, will last up to 12 more days in comparison with the control sample.

4. Application of Osmocote®Pro fertilizer in the species of *Lilium regale* determined an increase of the plants' height, with the best results being highlighted in the group of plants benefitting of the highest dosage of fertilizer, that is  $V_4$ ;

5. Regarding the number of flowers/plant, in the group of plants from the variants fertilized with the largest doses of Osmocote®Pro ( $V_3$  and  $V_4$ ) there were recorded highly significant positive differences as compared to the control group, and significant positive differences in  $V_2$ ;

6. The diameter of the flowers' cup grew in all fertilized variants and positive significant differences were noted in the variants  $V_3$  and  $V_4$  as compared to the control group.

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**Consilier editorial:**

**Vasile VÎNTU**

**Tehnoredactori:**

**Elena Liliana CHELARIU**

**Corectori:**

**Lucia DRAGHIA  
Elena Liliana CHELARIU**

**Bun de tipar:**

**15.12.2016**

**Apărut:**

**2016**

**Format:**

**61x86/16**

**Editura:**

**„Ion Ionescu de la Brad” Iași  
Aleea M. Sadoveanu, 3  
Tel.: 0232-407471  
e-mail: [editura@uaiasi.ro](mailto:editura@uaiasi.ro)**

ISSN–L=1454-7376

(Print)-ISSN 1454-7376

(Online)=ISSN 2069-8275

(CD-ROM) = ISSN 2069 – 847X

**PRINTED IN ROMANIA**

**Editorial Consultant:**

**Vasile VÎNTU**

**Technical Editors:**

**Elena Liliana CHELARIU**

**Readers:**

**Lucia DRAGHIA  
Elena Liliana CHELARIU**

**Imprimatur:**

**15.12.2016**

**Published:**

**2016**

**Format:**

**61x86/16**

**Publishing House:**

**„Ion Ionescu de la Brad” Iași  
Aleea M. Sadoveanu, 3  
Tel.: 0232-407471  
e-mail: [editura@uaiasi.ro](mailto:editura@uaiasi.ro)**

ISSN–L=1454-7376

(Print)-ISSN 1454-7376

(Online)=ISSN 2069-8275

(CD-ROM) = ISSN 2069 – 847X

**PRINTED IN ROMANIA**