

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” IAȘI**



**LUCRĂRI ȘTIINȚIFICE**

**Vol. 58**

**NR. 1**

**SERIA HORTICULTURĂ**

EDITURA “ION IONESCU DE LA BRAD”



**IAȘI 2015**



## COLECTIVUL DE COORDONARE AL REVISTEI „LUCRĂRI ȘTIINȚIFICE”

**Redactor șef:** Prof. dr. **Vasile VÎNTU** - USAMV Iași, Romania  
**Redactor adjunct:** Prof. dr. **Constantin LEONTE** - USAMV Iași, Romania  
**Membri:** Prof. dr. **Lucia DRAGHIA** - USAMV Iași, Romania  
Prof. dr. **Teodor ROBU** - USAMV Iași, Romania  
Prof. dr. **Liviu MIRON** - USAMV Iași, Romania  
Prof. dr. **Benone PĂȘĂRIN** - USAMV Iași, Romania

## COLEGIUL DE REDACȚIE AL SERIEI „HORTICULTURĂ”

**Redactor șef:** Prof. dr. **Lucia DRAGHIA** – USAMV Iași, Romania  
**Redactor adjunct:** Prof. dr. **Liliana ROTARU**– USAMV Iași, Romania  
**Membri:** Acad. **Valeriu D. COTEA** – USAMV Iași, Romania  
Prof. dr. **Ibrahim BAKTIR** - Akdeniz University, Faculty of Agriculture, Turkey  
Prof. dr. **Valerian BALAN** - UASM Chișinău, R. Moldova  
Prof. dr. **Gheorghe CIMPOIEȘ** - UASM Chișinău, R. Moldova  
Prof. dr. **Monika CHRISTMANN** - Forschungsanstalt Geisenheim University, Germany  
Prof. dr. **Valeriu V. COTEA** - USAMV Iași, România  
Prof. dr. **Carmelo DAZZI** - Università di Palermo, Italy  
Prof. dr. **Athanasios ECONOMOU** - Aristotle Univ. Thessaloniki, Greece  
Prof. dr. **Gheorghe GLĂMAN** - ASAS București, Romania  
Prof. dr. **Stefano GREGO** - Univ. Tuscia-Viterbo, Italia  
Prof. dr. **Gonca GÜNER DALKILIÇ** - Adnan Menderes University, Turkey  
Prof. dr. **Neculai MUNTEANU** - USAMV Iași, Romania  
Prof. dr. **Vicente SOTÉS RUIZ** - Universidad Politécnica de Madrid, ETSIA, Espagne  
Assist. Prof. dr. **Zeynel DALKILIÇ** - Adnan Menderes University, Turkey  
Conf. dr. **Gheorghe NICOLAESCU** - UASM Chișinău, R. Moldova  
Dr. **Hervé QUÉNOL** - CNRS - Université de Haute Bretagne - Rennes 2, France  
C.S. I dr. ing. **Silvia AMBĂRUȘ** - SCDL Bacău, Romania  
C.S. I dr. ing. **Eugen CÂRDEI** - SCDPP Iași, Romania  
C.S. I dr. ing. **Doina DAMIAN** - SCDVV Iași, Romania  
C.S. I dr. arh. **Mariana ȘLAPAC** - Institutul Patrimoniului Cultural al Academiei de Științe a R. Moldova

## COMISIA DE REFERENȚI ȘTIINȚIFICI

Prof. dr. Ilie <b>BURDUJAN</b> - USAMV Iași	Prof. dr. Stej. <b>BREZULEANU</b> -USAMV Iași
Prof. dr. Valeriu <b>V. COTEA</b> - USAMV Iași	Prof. dr. Culiță <b>SÎRBU</b> - USAMV Iași
Prof. dr. Lucia <b>DRAGHIA</b> - USAMV Iași	Conf. dr. Doina <b>DASCĂLU</b> USAMV Iași
Prof. dr. Mihai <b>ISTRATE</b> - USAMV Iași	Conf. dr. Feodor <b>FILIPPOV</b> - USAMV Iași
Prof. dr. Doina <b>JITĂREANU</b> - USAMV Iași	Conf. dr. Elena <b>GÎNDU</b> - USAMV Iași
Prof. dr. Valeriu <b>MOCA</b> - USAMV Iași	Conf. dr. Liviu <b>IRIMIA</b> - USAMV Iași
Prof. dr. Neculai <b>MUNTEANU</b> - USAMV Iași	Conf. dr. Mihai <b>MUSTEA</b> - USAMV Iași
Prof. dr. Servilia <b>OANCEA</b> - USAMV Iași	Conf. dr. Cornelia <b>PRISĂCARU</b> - USAMV Iași
Prof. dr. Teodor <b>ROBU</b> - USAMV Iași	Conf. dr. Lucia <b>TRINCĂ</b> - USAMV Iași
Prof. dr. Liliana <b>ROTARU</b> - USAMV Iași	Conf. dr. Mihai <b>STANCIU</b> - USAMV Iași
Prof. dr. Mihai <b>TĂLMACIU</b> - USAMV Iași	Șef lucr. dr. Antoanela <b>PATRAȘ</b> - USAMV Iași
Prof. dr. Ioan <b>ȚENU</b> - USAMV Iași	Șef lucr. dr. Tatiana <b>SANDU</b> - USAMV Iași
Prof. dr. Eugen <b>ULEA</b> - USAMV Iași	Șef lucr. dr. Alina <b>TROFIN</b> - USAMV Iași

© Editura “Ion Ionescu de la Brad” Iași  
ISSN–L=1454-7376  
(Print)-ISSN 1454-7376  
(Online)=ISSN 2069-8275  
(CD-ROM) = ISSN 2069 – 847X

## COORDINATOR OF JOURNAL „*LUCRĂRI ȘTIINȚIFICE*”

**Manager Editor:** Prof. PhD. **Vasile VÎNTU** - UASVM Iasi, Romania  
**Assistant Editor:** Prof. PhD. **Constantin LEONTE** - UASVM Iasi, Romania  
**Members:** Prof. PhD. **Lucia DRAGHIA** - UASVM Iasi, Romania  
Prof. PhD. **Teodor ROBU** - UASVM Iasi, Romania  
Prof. PhD. **Liviu MIRON** - UASVM Iasi, Romania  
Prof. PhD. **Benone PĂSĂRIN** - UASVM Iasi, Romania

## EDITORIAL BOARD OF „*HORTICULTURA*”

**Editor in chief** Prof. PhD. **Lucia DRAGHIA** – UASVM Iasi, Romania  
**Assistant Editor** Prof. PhD. **Liliana ROTARU** – UASVM Iasi, Romania  
**Editors:** Acad. **Valeriu D. COTEA** – USAMV Iași, Romania  
Prof. PhD. **Ibrahim BAKTIR** - Akdeniz University, Faculty of Agriculture, Turkey  
Prof. PhD. **Valerian BALAN** - UASM Chișinău, R. Moldova  
Prof. PhD. **Gheorghe CIMPOIEȘ** - UASM Chișinău, R. Moldova  
Prof. PhD. **Monika CHRISTMANN** - Forschungsanstalt Geisenheim University, Germany  
Prof. PhD. **Valeriu V. COTEA** - USAMV Iași, România  
Prof. PhD. **Carmelo DAZZI** - Università di Palermo, Italy  
Prof. PhD. **Athanasios ECONOMOU** - Aristotle Univ. Thessaloniki, Greece  
Prof. PhD. **Gheorghe GLĂMAN** - ASAS București, Romania  
Prof. PhD. **Stefano GREGO** - Univ. Tuscia-Viterbo, Italia  
Prof. PhD. **Gonca GÜNER DALKILIÇ** - Adnan Menderes University, Turkey  
Prof. dr. **Neculai MUNTEANU** - USAMV Iași, Romania  
Prof. PhD. **Vicente SOTÉS RUIZ** - Universidad Politécnica de Madrid, ETSIA, Espagne  
Assist. Prof. PhD. **Zeynel DALKILIÇ** - Adnan Menderes University, Turkey  
Assist. Prof. PhD. **Gheorghe NICOLAESCU** - UASM Chișinău, R. Moldova  
Dr. **Hervé QUÉNOL** - CNRS - Université de Haute Bretagne - Rennes 2, France  
C.S. I PhD. **Silvia AMBĂRUȘ** - SCDL Bacău, Romania  
C.S. I PhD. **Eugen CÂRDEI** - SCDPP Iași, Romania  
C.S. I PhD. **Doina DAMIAN** - SCDVV Iași, Romania  
C.S. I PhD. **Mariana ȘLAPAC** - Institutul Patrimoniului Cultural al Academiei de Științe a R. Moldova

## SCIENTIFIC REVIEWERS

Prof. PhD. Ilie <b>BURDUJAN</b> - USAMV Iași	Prof. PhD. Stej. <b>BREZULEANU</b> -USAMV Iași
Prof. PhD. Valeriu <b>V. COTEA</b> - USAMV Iași	Prof. PhD. Culiță <b>ȘIRBU</b> - USAMV Iași
Prof. PhD. Lucia <b>DRAGHIA</b> - USAMV Iași	Rd. PhD. Doina <b>DASCĂLU</b> USAMV Iași
Prof. PhD. Mihai <b>ISTRATE</b> - USAMV Iași	Rd. PhD. Feodor <b>FILIPPOV</b> - USAMV Iași
Prof. PhD. Doina <b>JITĂREANU</b> - USAMV Iași	Rd. PhD. Elena <b>GÎNDU</b> - USAMV Iași
Prof. PhD. Valeriu <b>MOCA</b> - USAMV Iași	Rd. PhD. Liviu <b>IRIMIA</b> - USAMV Iași
Prof. PhD. Neculai <b>MUNTEANU</b> - USAMV Iași	Rd. PhD. Mihai <b>MUSTEA</b> - USAMV Iași
Prof. PhD. Servilia <b>OANCEA</b> - USAMV Iași	Rd. PhD. Cornelia <b>PRISĂCARU</b> - USAMV Iași
Prof. PhD. Teodor <b>ROBU</b> - USAMV Iași	Rd. PhD. Lucia <b>TRINCĂ</b> - USAMV Iași
Prof. PhD. Liliana <b>ROTARU</b> - USAMV Iași	Rd. PhD. Mihai <b>STANCIU</b> - USAMV Iași
Prof. PhD. Mihai <b>TĂLMACIU</b> - USAMV Iași	Lect. PhD. Antoanela <b>PATRAȘ</b> - USAMV Iași
Prof. PhD. Ioan <b>ȚENU</b> - USAMV Iași	Lect. PhD. Tatiana <b>SANDU</b> - USAMV Iași
Prof. PhD. Eugen <b>ULEA</b> - USAMV Iași	Lect. PhD. Alina <b>TROFIN</b> - USAMV Iași

“Ion Ionescu de la Brad” Publishing House Iași  
ISSN-L=1454-7376  
(Print)-ISSN 1454-7376  
(Online)=ISSN 2069-8275  
(CD-ROM) = ISSN 2069 – 847X



## CONTENT

1.	<b>OANCEA Servilia, CAZACU Ana, PĂDUREANU Silvica</b> - The fractal dimension as a measure of the corn root change to the nickel action .....	11
2.	<b>TROFIN Alina, UNGUREANU Elena, TRINCĂ LUCIA Carmen, SANDU Tatiana, IACOB Cătălina Ioana</b> - The decrease in nitrate content of polluted waters by using waste wood as adsorbent material .....	17
3.	<b>UNGUREANU Elena, JIȚĂREANU Doina, TROFIN Alina, SANDU Tatiana, POPA V.I.</b> - Chemical and spectral characterizations for some lignin products .....	23
4.	<b>CHEDEA Veronica Sanda, PALADE L. M., ROTAR M. C., CĂLIN L. G., DRAGOMIR C.</b> - The anthocyanin composition of a red grape pomace in relation with the wine industry by-products valorization in animal feed .....	29
5.	<b>PĂDUREANU Silvica</b> - The palynological characterization and the pollen potential germination of <i>Rhododendron racemosum</i> French. ....	35
6.	<b>PĂDUREANU Silvica</b> - Peculiarities of pollen tube growth of <i>Rhododendron racemosum</i> French. ....	41
7.	<b>MELENCIUC M.</b> - Features of adaptation to drought in iso and anisohydric plants and effect of salicylic acid .....	47
8.	<b>MARTA Alina Elena, JIȚĂREANU Carmenica Doina, SLABU Cristina</b> - Effect of salt stress on seed germination and seedlings chlorophyll content of some tomatoes ( <i>Lycopersicon esculentum</i> Mill.) local landraces .....	53
9.	<b>SCHMIDT Brigitta, ȘUMĂLAN R., ȘUMĂLAN Renata, CĂLUȘERU Alina Lavinia, SAMFIRA I.</b> - The influence of arbuscular mycorrhizal fungi on tolerance to saline stress of tomatoes .....	61
10.	<b>SLABU Cristina, JIȚĂREANU Carmenica Doina, MARTA Alina Elena, BOLOGA (COVAȘĂ) Mihaela</b> - The behavior of some onion ( <i>Allium cepa</i> L.) local landraces under salt stress .....	67
11.	<b>STRATU Anișoara, LOBIUC A., COSTICĂ Naela</b> - The influence of copper on seed germination and growth in the first ontogenetic stages in the species <i>Brassica oleracea</i> L. and <i>Cucurbita pepo</i> L. ....	73

12.	<b>BOGOESCU M., DRAGOMIR Elena</b> - Grafting of vegetables in Romania .....	79
13.	<b>SIKAVELIS K., ROȘCA I.</b> - Trade market of vegetable seedlings, current and future perspective in Romania .....	87
14.	<b>HAMBURDĂ Silvia Brîndușa, MUNTEANU N., STOLERU V., TELIBAN G.C., GALEA (DELEANU) Florina-Maria, SAVAPAVĂL Simona, COJOCARU Al.</b> - The allelopathy relations' importance in developing technologies for intercropping cultivation system .....	93
15.	<b>GALEA (DELEANU) Florina-Maria, MUNTEANU N., HAMBURDĂ Silvia-Brîndușa</b> - Preliminary studies regarding the establishment of an ornamental garden in a geometrical style using vegetable plants in intercropping system .....	99
16.	<b>HAMBURDĂ Silvia Brîndușa, MUNTEANU N., STOLERU V., TELIBAN G.C., GALEA (DELEANU) Florina-Maria, SAVAPAVĂL Simona, COJOCARU Al.</b> - Results of runner bean ( <i>Phaseolus coccineus</i> L.) yield obtained in intercropping system ...	105
17.	<b>CORDUNEANU Oana, ȚENU I., STOLERU V., ROȘCA R., ȘOVĂIALĂ Gh., MATACHE Gabriela</b> - Researches regarding the fertigation through drip irrigation of solarium bell pepper crop .	111
18.	<b>TELIBAN G.C., MUNTEANU N., POPA Lorena-Diana, STOLERU V., HAMBURDĂ Silvia Brîndușa, STAN T., BUBURUZ Alexandra-Andreea, ONOFREI Vasilica, CIOBANU V.</b> - The behavior of a runner bean assortment for pods ( <i>Phaseolus coccineus</i> L.) in polyethylene tunnels, in a crop established by direct sowing .....	117
19.	<b>JAKAB-ILYEFALVI ZSOLT</b> - Preliminary results regarding natural tendency of feathering and growth habits of some apple scion-rootstock combinations, in the nursery .....	123
20.	<b>PESTEANU A.</b> - Effect of thinning "Idared" apple variety using NAD and Ethephon .....	129
21.	<b>VĂMĂȘESCU S.</b> - The influence of foliar fertilization on apple productivity .....	135
22.	<b>SFECLĂ Irina</b> - <i>Kniphofia</i> Moench. species of perspective for arrangement of green areas of Republic of Moldova .....	141
23.	<b>ALEXANDRU L.C., ROTARU Liliana, NECHITA Ancuța, DAMIAN Doina, COLIBABA Lucia Cintia</b> - The behavior of grape vine varieties Gelu and Paula at grafting .....	145

24.	<b>SAVIN GH., CORNEA V.</b> - Mobilization and exploration of genetic resources in development of sustainable viticulture in context of restrictive factors .....	151
25.	<b>DIACONU Andreea, ȚENU I., ROȘCA R, CHIRILĂ C.</b> - Research on improvement of spraying machine in vines plantations, in order to reduce degree of pollution soil .....	157
26.	<b>ISTRATE A., ROTARU Liliana, COLIBABA Lucia Cintia</b> - applications of the Principle Component Analysis (PCA) at grape varieties from the sortogroup Coarnă neagră for establishing phenotypical variability .....	163
27.	<b>NECHITA Ancuta, ZALDEA Gabi, DAMIAN Doina, ALEXANDRU L.C., MOROȘANU Ana Maria</b> - Testing and optimization methods and cultural practices of vines in the ecological system the period conversion .....	169
28.	<b>ODĂGERIU G., ZAMFIR C.I., LEFTER B., COLIBABA Lucia Cintia, DONICI Alina</b> - Aspects regarding the maturation dynamics of table grapes grown in Viile area of Bujoru wine centre .....	175
29.	<b>NICULAUA M., ODĂGERIU Gh., TUCALIUC Roxana, MOROȘANU Ana Maria, TELIBAN I., COTEA V.V.</b> - Cypermethrin assessment and possible persistence in grapes or wines .....	183
30.	<b>DUMITRIU (GABUR) Georgiana-Diana, LUCHIAN Camelia Elena, COTEA V.V., PEINADO R.A., LOPEZ DE LERMA Nieves, COLIBABA Lucia Cintia, NICULAUA M.</b> - Influence of new materials on the chemical composition of Muscat Ottonel wines .....	189
31.	<b>NECHITA C-tin. B., NICULAUA M., COTEA V.V.</b> - Assessment and adaptation of methods of extraction of grape seed polyphenol compounds .....	195
32.	<b>COLIBABA Lucia Cintia, COTEA V.V., KOKKINOFTA Rebecca, LUCHIAN Camelia Elena, CODREANU Maria</b> - Comparative study of some white romanian and cypriot wines .....	201
33.	<b>VERINGĂ Daniela</b> - Studies on the behavior of the cherries in the storage with modified atmosphere of carbon dioxide .....	207
34.	<b>BĂLĂNESCU (NEACȘU) Irina Ioana, ROȘCA I.</b> - Research on evolution of the main pests of platane in Romania .....	213
35.	<b>BUTNARIU Gianina, TĂLMACIU M., TĂLMACIU Nela, HEREA Monica</b> - Observation on the structure and ecological parameters of the population of invertebrates in plum orchards .....	221
36.	<b>TALMACIU M., ENEA C.I., BRUDEA V., TALMACIU Nela, HEREA Monica</b> - The prospect of molecular approaches in fundamental research and management of insects .....	227

37.	<b>BODESCU Maria-Mădălina, BODESCU Oana Maria, TĂLMACIU M., HEREA Monica</b> - Antioxidants buckthorn oil, adjuvant in mild cognitive dysfunction therapy .....	233
38.	<b>CEHAN Agata Mihaela, GHEORGHÎĂ Constanța Carmina</b> - Representations of the garden of eden in christian sacred architecture .....	239
39.	<b>DASCĂLU Doina Mira</b> - Ways of implementing in the landscaping education some rehabilitation studies for outdoor spaces of universities .....	245
40.	<b>NICA R.M., DUMITRAȘCU Aurora Irina, VÎNĂU Nely, CORDUBAN C.G.</b> - Sustainable community rehabilitation with forestry systems .....	251
41.	<b>ȘTEFAN Diana</b> - Matters regarding the identity of space between design and perception .....	257
42.	<b>SANDU Tatiana, TROFIN Alina-Elena, PANTAZI Viorica, UNGUREANU Elena</b> - Analysis of the green areas with unlimited access and their impact on the population from Iași city .....	263
43.	<b>NEGREA Roxana</b> - Studies regarding the behavior of some species of <i>Sedum</i> under the stress conditions induced by roof culture .....	269
44.	<b>BĂLAN Anca, LUCA M., AVRAM M.</b> - Influence of hydraulic connection of lake –sea system on flora and fauna of the coastal lakes, area "Slowinski National Park", Poland .....	275
45.	<b>BUHĂIANU Bianca, MUNTEANU N., HURA Carmen, GALEA (DELEANU) Florina-Maria, STOLERU V.</b> - Organophosphorus pesticide residues from soil and cucumber fruits .....	281
46.	<b>LUPU Iuliana Gabriela, GROSU M.C., HOGAȘ H.I.</b> - Nonwovens used in horticulture – optimization of needle punching process parameters .....	287
47.	<b>PRISĂCARU Cornelia, PRISĂCARU Anca-Irina</b> - Studies on the toxic effect of various pharmaceutical preparations obtained from <i>Hamamelis virginiana</i> .....	293
48.	<b>RĂILEANU Marcela, COJOCARU AL., IPĂȚIOAIEI C., MUNTEANU N., STOLERU V.</b> - The presence of heavy metals in rhubarb according by technology .....	299
49.	<b>Jose Ignacio MARÍN GUIRAO, Francisco Martín USERO, Victoria VELASCO ARROYO, José Manuel RODRÍGUEZ ZAFRA, Julio César TELLO MARQUINA</b> - Assessment of biological soil fatigue in horticultural monocultures .....	305

# CUPRINS

1. <b>OANCEA Servilia, CAZACU Ana, PĂDUREANU Silvica</b> - Dimensiunea fractală ca măsură a modificării rădăcinii de porumb la acțiunea nichelului .....	11
2. <b>TROFIN Alina, UNGUREANU Elena, TRINCĂ LUCIA Carmen, SANDU Tatiana, IACOB Cătălina Ioana</b> - Scăderea conținutului în nitrați a apelor poluate prin folosirea deșeurilor lemnoase ca material adsorbant .....	17
3. <b>UNGUREANU Elena, JITĂREANU Doina, TROFIN Alina, SANDU Tatiana, POPA V.I.</b> - Caracterizări chimice și spectrale ale unor produse ligninice .....	23
4. <b>CHEDEA Veronica Sanda, PALADE L. M., ROTAR M.C., CĂLIN L.G., DRAGOMIR C.</b> - Compoziția în antociani a tescovinei rezultate de la obținerea vinului roșu în vederea valorificării reziduurilor industriei vinicole în hrana animală .....	29
5. <b>PĂDUREANU Silvica</b> - Caracterizarea palinologică și a potențialului germinativ al polenului de <i>Rhododendron racemosum</i> French. ....	35
6. <b>PĂDUREANU Silvica</b> - Particularitățile de creștere ale tubului polinic de <i>Rhododendron racemosum</i> French. ....	41
7. <b>MELENCIUC M</b> - Particularități de adaptare la secetă a plantelor iso și anisohidrice și efectul acidului salicilic .....	47
8. <b>MARTA Alina Elena, JITĂREANU Carmenica Doina, SLABU Cristina</b> - Efectul stresului salin asupra germinăției semințelor și a conținutului de clorofilă a plantulelor la unele populații locale de tomate ( <i>Lycopersicon esculentum</i> Mill.) .....	53
9. <b>SCHMIDT Brigitta, ȘUMĂLAN R., ȘUMĂLAN Renata, CĂLUȘERU Alina Lavinia, SAMFIRA I.</b> - Influența fungilor arbuscular micorizali asupra toleranței la stresul salin la tomate .....	61
10. <b>SLABU Cristina, JITĂREANU Carmenica Doina, MARTA Alina Elena, BOLOGA (COVAȘĂ) Mihaela</b> - Comportamentul unor populații locale de ceapă ( <i>Allium cepa</i> L.) sub influența stresului salin .....	67
11. <b>STRATU Anișoara, LOBIUC A., COSTICĂ Naela</b> - Influența cuprului asupra germinăției semințelor și a creșterii în primele stadii ontogenetice la speciile <i>Brassica oleracea</i> L. și <i>Cucurbita pepo</i> L. ....	73
12. <b>BOGOESCU M., DRAGOMIR Elena</b> - Altoirea legumelor în România .....	79

13.	<b>SIKAVELIS K., ROȘCA I.</b> - Piața comercială de răsaduri de legume, perspectiva actuală și viitoare în România .....	87
14.	<b>HAMBURDĂ Silvia Brîndușa, MUNTEANU N., STOLERU V., TELIBAN G.C., GALEA (DELEANU) Florina-Maria, SAVA-PAVĂL Simona, COJOCARU Al.</b> - Importanța relațiilor alelopatiche în elaborarea tehnologiilor de cultivare în sistem intercropping .....	93
15.	<b>GALEA (DELEANU) Florina-Maria, MUNTEANU N., HAMBURDĂ Silvia-Brîndușa</b> - Studii privind realizarea unei grădini ornamentale în stil geometric utilizând plante legumicole în sistem de intercropping .....	99
16.	<b>HAMBURDĂ Silvia Brîndușa, MUNTEANU N., STOLERU V., TELIBAN G.C.<sup>1</sup>, GALEA (DELEANU) Florina-Maria<sup>1</sup>, SAVA-PAVĂL Simona<sup>1</sup>, COJOCARU Al.</b> - Rezultate de producție obținute în sistem intercropping la fasolea mare ( <i>Phaseolus coccineus</i> L.) .....	105
17.	<b>CORDUNEANU Oana, ȚENU I., STOLERU V., ROȘCA R.<sup>1</sup>, ȘOVĂIALĂ Gh. MATACHE Gabriela</b> - Cercetări privind fertilizarea prin picurare a unei culturi de ardei în solar .....	111
18.	<b>TELIBAN G.C., MUNTEANU N., POPA Lorena-Diana, STOLERU V., HAMBURDĂ Silvia Brîndușa, STAN T., BUBURUZ Alexandra-Andreea, ONOFREI Vasilica, CIOBANU V.</b> - Comportarea în spații protejate a unui sortiment de fasole mare ( <i>Phaseolus coccineus</i> L.) pentru păstăi, în cultură înființată prin semănat direct .....	117
19.	<b>JAKAB-ILYEFALVI ZSOLT</b> - Rezultate preliminare privind tendința naturală de emiteră a lăstarilor anticipați și particularitățile de creștere a unor combinații soi-portaltoi în pepinieră, la specia măr .....	123
20.	<b>PESTEANU A.</b> - Efectul răririi fructelor de soiul Idared utilizând produse pe bază de NAD și Ethephon .....	129
21.	<b>VĂMĂȘESCU S.</b> - Influența fertilizării foliare asupra productivității la măr .....	135
22.	<b>SFECLĂ Irina</b> - <i>Kniphofia</i> Moench. specii de perspectivă pentru amenajarea spațiilor verzi din Republica Moldova .....	141
23.	<b>ALEXANDRU L.C., ROTARU Liliana, NECHITA Ancuța, DAMIAN Doina, COLIBABA Lucia Cintia</b> - Comportarea la altoire a soiurilor de viță de vie Gelu și Paula .....	145
24.	<b>SAVIN GH., CORNEA V.</b> - Mobilizarea și explorarea resurselor genetice în dezvoltarea unei viticulturi sustenabile în contextul factorilor restrictivi .....	151

25.	<b>DIACONU Andreea, ȚENU I., ROȘCA R., CHIRILĂ C.</b> - Cercetări privind perfecționarea unei mașini de stropit în plantațiile de viță de vie, cu scopul de a reduce gradul de poluare al solului .....	157
26.	<b>ISTRATE A., ROTARU Liliana, COLIBABA Lucia Cintia</b> - Aplicarea Analizei în Componenti Principali (ACP) la soiurile din sortogrupul Coarnă neagră, în vederea stabilirii variabilității fenotipice a acestora .....	163
27.	<b>NECHITA Ancuta, ZALDEA Gabi, DAMIAN Doina, ALEXANDRU L.C., MOROȘANU Ana Maria</b> - experimentarea și optimizarea unor metode și practici de cultură a viței de vie în perioada de conversie la sistemul ecologic .....	169
28.	<b>ODĂGERIU G., ZAMFIR C.I., LEFTER B., COLIBABA Lucia Cintia</b> - Aspecte privind dinamica maturării unor struguri de masă cultivate pe plaiul Viile din centrul viticol Bujoru .....	175
29.	<b>NICULAUA M., ODĂGERIU Gh., TUCALIUC Roxana, MOROȘANU Ana Maria, TELIBAN I., COTEA V.V.</b> - Evaluarea cipermetrinului din struguri și posibila remanentă la vinuri .....	183
30.	<b>DUMITRIU (GABUR) Georgiana-Diana, LUCHIAN Camelia Elena, COTEA V.V., PEINADO R.A., LOPEZ DE LERMA Nieves, COLIBABA Lucia Cintia, NICULAUA M.</b> - Influența materialelor noi asupra compoziției chimice a vinului Muscat Ottonel .....	189
31.	<b>NECHITA C-tin. B., NICULAUA M., COTEA V.V.</b> - Evaluarea și adaptarea unor metode de extracție a compușilor polifenolici din semințele de struguri .....	195
32.	<b>COLIBABA Lucia Cintia, COTEA V.V., KOKKINOFTA Rebecca, LUCHIAN Camelia Elena, CODREANU Maria</b> - Studii comparative ale unor vinuri albe cipriote și românești .....	201
33.	<b>VERINGĂ Daniela</b> - Studii privind comportarea fructelor de vișin la păstrarea în atmosferă modificată în dioxid de carbon .....	207
34.	<b>BĂLĂNESCU (NEACȘU) Irina Ioana, ROȘCA I.</b> - Cercetări privind evoluția principalilor dăunători ai platanului în România ...	213
35.	<b>BUTNARIU Gianina, TĂLMACIU M., TĂLMACIU Nela, HEREA Monica</b> - Observații privind structura și parametrii ecologici ai populației de nevertebrate în plantațiile pomicole de prun .....	221
36.	<b>TALMACIU M., ENEA C.I., BRUDEA V., TALMACIU Nela, HEREA Monica</b> - Rolul volatilelor emise de plante în „pregătire” (priming) la atacul erbivorelor .....	227

37.	<b>BODESCU Maria-Mădălina, BODESCU Oana Maria, TĂLMACIU M., HEREA Monica</b> - Antioxidanții uleiului de cătină, adjuvanți în terapia disfuncției cognitive medii și ușoare .....	233
38.	<b>CEHAN Agata Mihaela, GHEORGHITĂ Constanța Carmina</b> - Reprezentări ale grădinii raiului în arhitectura spațiului sacru creștin .....	239
39.	<b>DASCĂLU Doina Mira</b> - Moduri de aplicare în învățământul peisagistic a unor studii de reabilitare a spațiilor exterioare aferente universităților .....	245
40.	<b>NICA R.M., DUMITRAȘCU Aurora Irina, VÎNĂU Nely, CORDUBAN C.G.</b> - Reabilitarea sustenabilă a comunităților prin sisteme de culturi forestiere în România .....	251
41.	<b>ȘTEFAN Diana</b> - Aprecieri privind identitatea spațiului între concepție și percepție .....	257
42.	<b>SANDU Tatiana, TROFIN Alina-Elena, PANTAZI Viorica, UNGUREANU Elena</b> - Analiza spațiilor verzi cu acces nelimitat și impactul acestora asupra populației din municipiul Iași .....	263
43.	<b>NEGREA Roxana</b> - Studii privind comportarea unor specii de <i>Sedum</i> la condițiile de stres induse de cultura pe acoperiș .....	269
44.	<b>BĂLAN Anca, LUCA M., AVRAM M.</b> - Influența conexiunii hidraulice lac –mare asupra faunei și florei din lacurile de coastă, zona „Parcului Național Słowiński”, Polonia .....	275
45.	<b>BUHĂIANU Bianca, MUNTEANU N., HURA Carmen, GALEA (DELEANU) Florina-Maria, STOLERU V.</b> - Reziduurile de pesticide organofosforice din sol și fructe la o cultură de castraveți .....	281
46.	<b>LUPU Iuliana Gabriela, GROSU M.C., HOGAȘ H.I.</b> - Nețesute folosite în horticultură – optimizarea parametrilor procesului de interțesere .....	287
47.	<b>PRISĂCARU Cornelia, PRISĂCARU Anca-Irina</b> - Studii privind efectele toxice ale unor preparate farmaceutice obținute din <i>Hamamelis virginiana</i> .....	293
48.	<b>RĂILEANU Marcela, COJOCARU Al., IPĂTIOAIEI C., MUNTEANU N., STOLERU V.</b> - Prezența metalelor grele în revent, în funcție de tehnologia aplicată .....	299
49.	<b>Jose Ignacio MARÍN GUIRAO, Francisco Martín USERO, Victoria VELASCO ARROYO, José Manuel RODRÍGUEZ ZAFRA, Julio César TELLO MARQUINA</b> - Evaluarea oboselii solului din punct de vedere biologic în monoculturile horticole .....	305



## THE FRACTAL DIMENSION AS A MEASURE OF THE CORN ROOT CHANGE TO THE NICKEL ACTION

### DIMENSIUNEA FRACTALĂ CA MĂSURĂ A MODIFICĂRII RĂDĂCINII DE PORUMB LA ACȚIUNEA NICHELULUI

**OANCEA Servilia<sup>1</sup>, CAZACU Ana<sup>1</sup>, PĂDUREANU Silvica<sup>1</sup>**  
e-mail: liaoancea@yahoo.com

**Abstract.** The main objective of this study was to evaluate the impact of the treatment with nickel on growth of corn roots (*Zea mays* L.), using fractal analysis. In order to evaluate the change on root plants we determined the fractal dimension for untreated and treated corn plant roots. Seeds of corn were put into Petri dishes on double filter paper together with two solutions of  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$  and they were kept here for four days. The germinated seeds were planted in soil where they continued to growth. After two weeks the root plants have been collected and the fractal analysis was performed. Our results demonstrated that the fractal structure of corn roots changed after the treatment with nickel. We confirm the fact that  $\text{Ni}^{2+}$  is a toxic ion that decreased the plant capacity to develop complex roots and the fractal analysis is a useful method to characterize the structure of plant roots.

**Key words:** root system, fractal analysis

**Rezumat.** Obiectivul principal al acestei lucrări este de a evalua impactul tratamentului cu nichel în creșterea rădăcinilor plantelor de porumb (*Zea mays* L.), folosind analiza fractală. Pentru a evalua modificările rădăcinilor plantelor, am determinat dimensiunea fractală a plantelor tratate și a celor netratate. Semințele de porumb au fost puse la germinat în sticle Petri pe hârtie de filtru dublă și cu două concentrații de  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$  unde au fost ținute patru zile. Semințele germinate au fost plantate apoi în sol unde au continuat să crească. După două săptămâni au fost colectate rădăcinile și s-a efectuat analiza fractală. Rezultatele arată că rădăcinile plantelor s-au modificat după tratamentul cu nichel prin comparație cu martorul. Noi confirmăm astfel că  $\text{Ni}^{2+}$  este un ion toxic care scade capacitatea plantelor de a dezvolta rădăcini complexe și că analiza fractală este o metodă utilă pentru caracterizarea structurii rădăcinii plantelor.

**Cuvinte cheie:** sistemul radicular, analiza fractală

## INTRODUCTION

Nickel is considered one of the most abundant heavy metal pollutants of the environment. On the other hand, nickel is one of essential micronutrients for plants growth, the concentration of nickel required by vegetal tissues is of maximum 0,1 mg/L.

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

The high concentrations of nickel (Ni) have deleterious effects on plant growth and metabolism, and produce visible effects of toxicity. The general effects associated with Ni toxicity in plants include inhibition of germination, reduced shoot and root growth, low development of branching system, deformation of the different plant segments, decrease of biomass, mitotic disturbances to root tip (Ahmad *et al.*, 2011).

The complexity of the plant roots has been studied by many researchers but there are few results due the influence of the rizosphere heterogeneity (Akasaka *et al.*, 1998; Berntson, 1994; Campbell, 1996; Eshel, 1998; Melniciuc Puică *et al.*, 2006; Nielsen *et al.*, 1997; Oancea, 2006; Puzon, 2005). Root growth is related to the consumption of water and nutrients of plants and it directly take the effect of environmental change. By exploring different spatial niches, plants with contrasting root architecture may reduce the extent of competition among neighboring root systems. Root complexity has been difficult to comprehend using simple Euclidean methods. The main objective of this study was to evaluate the impact of the treatment with nickel on growth of corn roots, using fractal analysis.

## MATERIAL AND METHOD

To study the effect of nickel on root growth, two concentrations of  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$  have been prepared and we sorted the following variants:

- 1 - control;
- 2 – solution of 0.25%  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$ ;
- 3 - solution of 0.5%  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$

Generally, regarding chemical compound toxicity, researchers used such a range of dose just a 2x difference in concentration (Masih J. and Bhadauria V., 2010).

The experiments were conducted in the Biophysics Department Laboratory of the University of Agricultural Sciences and Veterinary Medicine Iasi (Foca. *et al.*, 2004), (Oancea S. *et al.*, 2005). As a biological material we used corn (*Zea mays* L.), the most widely used cereal in our country. 100 seeds of corn were put into Petri dishes on double filter paper together with 20 mL treatment solution. Here the seeds were kept in dark at the optimal temperature ( $23^\circ\text{C}$ ) for 4 days. After that the germinated seed were planted in soil in the our laboratory (fig. 1), where they developed for two weeks. For fractal analysis the plants were extracted from the soil, washed and the root were cut from the shoot. Then we make many photos of the root with a Canon camera (fig. 2). There are many other methods to determine the fractal dimension (Box Counting method, Yardstick method, Mass-Dimension Method, Perimeter–Area Method, Slit-Island Method, Asymptotic fractal formulas etc.). The photos were prepared in order to use the HarFA soft to determine the fractal dimension. We prepared the black and white images of the painting using the Digital Image Processing with Matlab. In Thresholding procedure a grey scale image is turned into a binary (black and white) image by first choosing a grey level T in the original image, and then turning every pixel black or white according to whether its grey value is greater than or less than T. The grey images of the coloured paintings have been processed also in Matlab.

In HarFA is used a modification of traditional Box Counting method. By this modification on obtain three fractal dimensions, which characterise properties of black plane DB, black-white border of black object DBW (and this information is the most

interesting) and properties of white background DW. The fractal dimension is the slope of the straight line „Black&White” (Zmeškal et al., 2001).



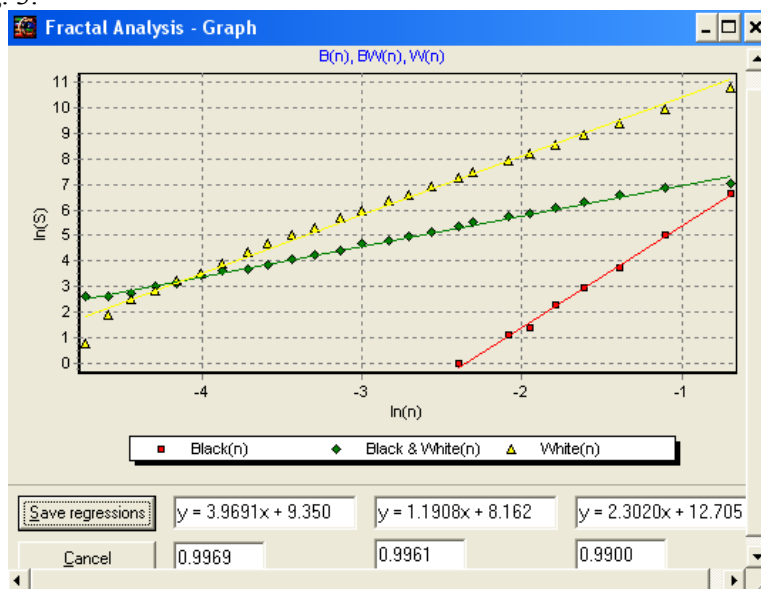
**Fig. 1** – The corn plant



**Fig. 2** – The corn roots

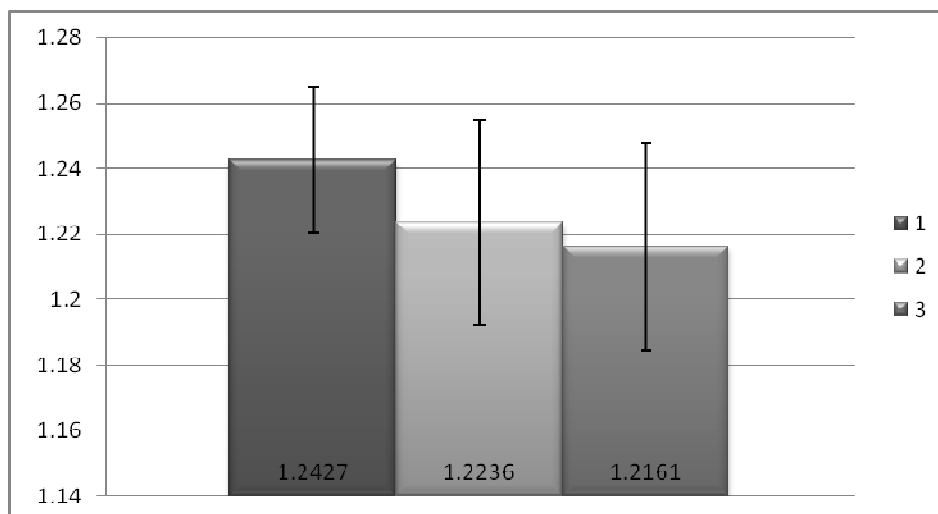
## RESULTS AND DISCUSSION

For the last root from the figure 2 we obtain the results given in the graph from fig. 3.



**Fig. 3** – The graph for fractal dimension of the treated plants with 0.25 concentration  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$

In figure 4 the mean values of the fractal dimension for plantlets are presented.



**Fig. 4** – The fractal dimension of the plantlet roots (Error bars are 95% confidence intervals for  $n=5$ , Oancea, 2007)

The fractal analysis of the roots showed that the mean fractal dimension decreases from 1.2427 (in the case of the control corn roots) to 1.2161 (in the case of the treatment with 0.5 concentration of  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$ ). These values sign up a straight line with a good correlation R- squared factor of 0.94. This result showed that the treatment of the corn plant with this chemical compound decreases the fractal dimension of the roots and the root complexity, contributing to a poor water transport in plant. Comparing these results with direct observations from figure 2 we can see that the fractal dimension is found to be correlated with root topology and root architecture.

## CONCLUSIONS

From biological point of view, our results prove that plantlets treated with  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$  show significant toxicity symptoms. However, the plantlets are able to cope with chemical stress due to a high capacity for change and adaptability.

In this work we pointed out the importance of concept of fractal structure in physiological characterization of root architecture. Our results demonstrated that the fractal structure of corn roots decreased after the treatment with nickel. Due the fact that the fractal dimension is a direct measure of the relative degree of complexity of the figure, we confirm the fact that  $\text{Ni}^{2+}$  is a toxic ion that decreased the plant capacity to develop complex roots.

We determined the planar fractal dimension of the roots from the above mentioned photos which can be connected with the other morphological changes in the root system.

## REFERENCES

1. **Ahmad M.S.A., Ashraf M., Hussain M., Ozturk M., 2011** - *Effect of nickel on seed germinability of some elite sunflower (*Helianthus annuus* L.) cultivar*, J. Hazard. Mat., 185, 1295 – 1303
2. **Akasaka Y., Mii M., Daimon H., 1998** - *Morphological alterations and root nodule formation in *Agrobacterium rhizogenes*-mediated transgenic hairy roots of peanut (*Arachis hypogaea* L.)*. Annals of Botany, 81 (2), 355-362
3. **Berntson G.M., 1994** - *Root systems and fractals: how reliable are calculations of fractal dimensions*. Annals of Botany, 73 (3), 281-284
4. **Campbell R.D., 1996** - *Describing the shape of fern leaves: a fractal geometrical approach*. Acta Biotheoretica 44, 119-126
5. **Eshel A., 1998** - *On the fractal dimensions of a root system*. Plant Cell and Environment, 21 (2), 247-251
6. **Foca N., Oancea S., Condurache D., 2004** - *Growth and photosynthetic activity for tomato plants treated with different cations*, Molecular crystals and Liquid crystals Journal, 418, 971-981
7. **Masih J., Bhadauria V., 2010** - *Studies on the effect of heavy metal (Cd and Ni) stress on the growth and physiology of *Allium cepa**, Annals of Biological Research, 1 (3) , 139-144
8. **Melniciuc Puică N., Oancea S., Dorohoi D.O., 2006** - *Fractal analysis as investigation method in old organic support*. Romanian Journal of Biophysics, 16(2), 135-140

9. **Nielsen K.L., Lynch J.P., Weiss H.N., 1997** - *Fractal geometry of bean root systems: Correlations between spatial and fractal dimension*, American Journal of Botany, 84 (1), 26-33
10. **Oancea S., Foca N., Airinei A., 2005** - *Effects of heavy metals on plant growth and photosynthetic activity*. Analele Univ. Al. I. Cuza, Tom I, s, Biofizica, Fizică medicală și Fizica mediului, 107-110
11. **Oancea S., 2006** - *Fractal analysis of the root architecture for tomato plants treated with cadmium*. Lucrari stiintifice, USAMV Iasi, seria Agronomie, vol. 49, 246-250
12. **Oancea S., 2007** - *Ghid de prelucrare rapidă a datelor experimentale*. Editura Performantica, Iasi
13. **Puzon K. A. M., 2005** - *Mathematical Analysis of Root Growth in Gamma-irradiated Cashew (Anacardium occidentale L.) and Mangosteen (Garcinia mangostana L.) Using Fractals*. Nature and Science, 3(1), 59-64
14. **Zmeškal O., Veselý M., Nežádal M., Buchníček M., 2001** - *Fractal Analysis of Image Structures*. HarFA - Harmonic and Fractal Image Analysis, 3 – 5

## THE DECREASE IN NITRATE CONTENT OF POLLUTED WATERS BY USING WASTE WOOD AS ADSORBENT MATERIAL

SCĂDEREA CONȚINUTULUI ÎN NITRAȚI A APELOR POLUATE PRIN  
FOLOSIREA DEȘEURILOR LEMNOASE CA MATERIAL ADSORBANT

**TROFIN Alina<sup>1</sup>, UNGUREANU Elena<sup>1</sup>, TRINCĂ LUCIA Carmen<sup>1</sup>,  
SANDU Tatiana<sup>1</sup>, IACOB Cătălina Ioana<sup>1</sup>**  
e-mail: atrofin@uaiasi.ro

**Abstract.** Large amounts of waste wood obtained from logging and wood processing have found various uses in the furniture industry, agriculture, entering into various compost recipes with applications in restoring soil's fertility or as absorbent material in numerous technologies, given the porous structure and composition of such materials. In the present study, we tested the capacity of grinded sawdust, simple and activated in acidic and alkaline solutions, to retain nitrates from aqueous solutions of different concentrations. Although all the variants have given very good results, sawdust activated in a solution of hydrochloric acid registered the retention of nitrate in an amount of 99.99% at concentrations up to 250 mg / l and 99.95% for the ones up to 400 mg / l. The method may be also considered for large scale applications due to the high adsorption speed.

**Key words:** sawdust, nitrate, pollution, water, adsorption

**Rezumat.** Cantitățile mari de deșeuri lemnoase obținute în urma tăierii și prelucrării lemnului și-au găsit diverse utilizări în industria mobilei, în agricultură, intrând în variate composturi cu aplicații în refacerea fertilității unor soluri sau în diverse tehnologii ca materiale absorbante, dată fiind structura poroasă și compoziția chimică a acestor deșeuri. În lucrarea de față, am testat capacitatea rumegușului fin măcinat, simplu și activat în soluții acide și bazice, de a reține nitrații din soluții apoase de diferite concentrații. Deși toate variantele au dat rezultate foarte bune, rumegușul activat în soluție de acid clorhidric a condus la reținerea cantității de nitrat în proporții de 99,99% la concentrații de până la 250 mg/l și 99,95% la cele de până la 400 mg/l. Metoda ar putea fi luată în considerare pentru aplicații la scară largă și datorită vitezei mari de adsorbție.

**Cuvinte cheie:** rumeguș, nitrat, poluare, apă, adsorbție

### INTRODUCTION

A source of vegetable waste, which in our country has not so far been considered valuable in many industrial branches, is the significant amount of wood chips and sawdust resulting from logging and wood processing.

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania



Capitalization of bark and sawdust by composting did not completely solve the problem of such large quantities of waste, but can become a source of income for a number of enterprises of forest economy.

Another way in which these wood waste materials are consumed is their use as absorbent material for different substances. For example, in a pilot study made by the department of U.S. Army Corps of Engineers, they tested a technology mainly consisting of kitchen waste passing through a large cylindrical vessel filled with wood chips. Grease is removed from the liquid by absorption and filtration, being attached to the surface of the chips. The effluent from this process then passes through a secondary absorbent filter tube. (Department of the army, U.S. Army Corps of Engineers, 2008)

Another study was conducted using wood chips as absorbent material for  $\text{SO}_2$  from air, base on the fact that this naturally porous material, especially when moistured, can adsorb and solve important amounts of sulphur dioxide. The conclusion was that moisture saturated wood chips retained about 90 times more sulphur dioxide than the dry material, for all the active concentrations used, between 1,12 - 4.60ppm. (Wang Uen-Ping, 1971)

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, 2008; Keng, 2013; O'Connell, 2008)

A study made in Turkey on straw, wood chips and corn stalks confirmed their capacity of removing a very wide used organophosphorus pesticide, 2,2'-dichlorovinyl-o,o'-dimethyl phosphate (DDVP, Dichlorvos) from waste waters, in a discontinued system. (Balkaya, 2002)

Sawdust and wood chips of various sizes were used in numerous studies as adsorbent materials for dyes, showing very good efficiency, especially concerning the terms of material costs. Residual wood, in natural or carbonized form, showed a higher affinity for basic dyes. (Crini, 2010)

The production of ethanol from sawdust is possible without relying on sources of edible ethanol. In "Well for Wheel", model created by Michael Wang of Argonne National Laboratories, cellulosic ethanol showed when used a reduction of green gas emissions by 80% (over gasoline) compared to corn ethanol, which showed reduction by 20 % -30%. (Nwakaire, 2013)

Sawdust bioreactors are a new option to reduce the amount of nitrate in drainage before reaching local surface waters. Sawdust bioreactors are also known as denitrifying bioreactors, a name that is more descriptive for the current process developing inside. Denitrification is the conversion of nitrate ( $\text{NO}_3^-$ ) into nitrogen gas ( $\text{N}_2$ ), which is carried out by bacteria that live in soil and also in bioreactors. (Christianson Laura, 2011)

In this paper, we used natural and activated grinded sawdust as adsorbent material for different concentrations of nitrate solutions, to see if the effluent falls



under the allowable concentration for drinking water of 10 mg  $\text{NO}_3^-/\text{l}$ , even 5 mg  $\text{NO}_3^-/\text{l}$ , when preparing baby food.

### MATERIAL AND METHOD

The adsorbent material was alder sawdust, grinded and sieved in order to keep only the smaller particles (fig. 1). For the control variant, the sawdust was boiled for half an hour in distilled water and then air-dried (fig. 2). The other two variants used the sawdust activated by half hour boiling in hydrochloric acid 0.1 n, respectively ammonia 0.1 n, followed by filtration and pH correction to 5.0 – 6.0 and then also air-drying (fig. 3, 4).

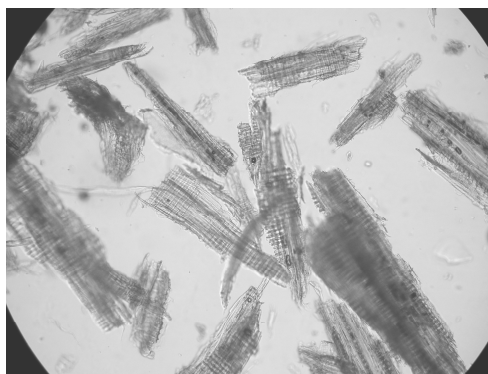


Fig. 1 -Natural sawdust



Fig. 2 - Adsorbent sawdust samples

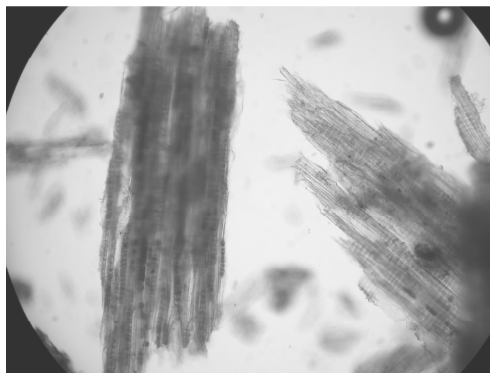


Fig. 3 - Acid activated sawdust

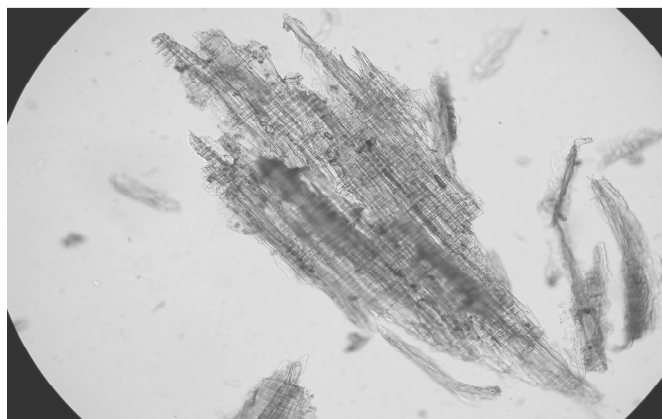


Fig. 4 - Alkali activated sawdust

The sodium nitrate solutions had initial concentrations from 50 to 400 mg  $\text{NO}_3^-$  /l. For each variant, we used 1 g of adsorbent material and 40 ml of nitrate solution, stirred continuously for 15 minutes, then filtered. We used Griess colorimetric method to determine the amount of nitrate in the filtrate of each variant.

## RESULTS AND DISCUSSIONS

The considered adsorbent material succeeded to remove almost the entire amount of nitrate from the samples, especially for the variants with activated forms. We tested solutions only up to 400 mg/l but given the obtained results, we estimate that even higher concentrations could be removed from natural or wastewaters by this simple procedure.

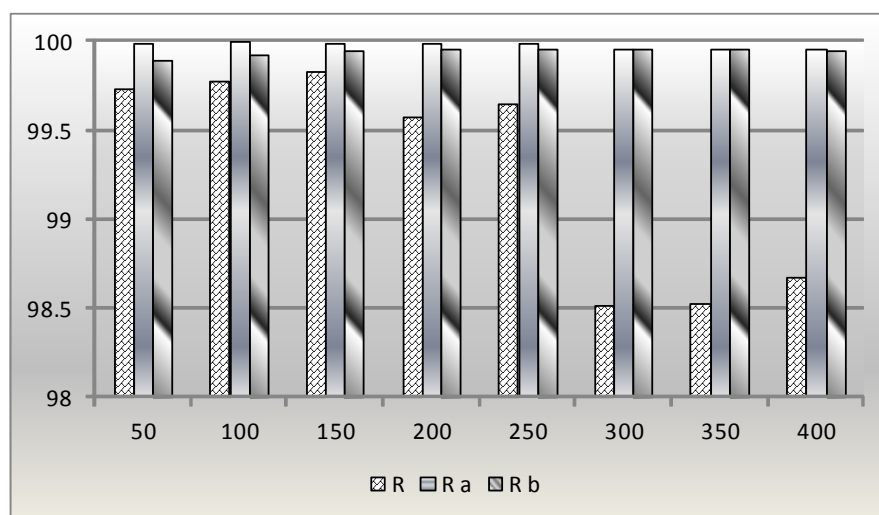
The data regarding the percentage of nitrate retained by one gram of sawdust from the initial amount and the realized adsorption coefficient are presented in tables 1 and 2 and comparatively, in figure 5.

Table 1

Conc. ion Sample	Percentages of removed nitrate amounts							
	50 mg/l	100 mg/l	150 mg/l	200 mg/l	250 mg/l	300 mg/l	350 mg/l	400 mg/l
Natural sawdust	99.7278	99.7776	99.8281	99.5748	99.6423	98.5187	98.5217	98.6697
Acid activated sawdust	99.9882	99.9932	99.99077	99.99195	99.9901	99.9585	99.9547	99.9556
Alkali activated sawdust	99.8882	99.9226	99.9432	99.9561	99.9566	99.9557	99.9595	99.9409

Table 2

Conc. Sample	Adsorption coefficient values							
	50 mg/l	100 mg/l	150 mg/l	200 mg/l	250 mg/l	300 mg/l	350 mg/l	400 mg/l
Natural sawdust	98.157	196.53	297.93	391.87	490.75	588.41	680.26	787.94
Acid activated sawdust	100.31	199.75	296.42	394.29	494.41	593.23	690.02	797.25
Alkali activated sawdust	98.626	198.653	299.77	393.29	496.407	591.92	688.426	793.34



**Fig. 5** - Percentages of nitrate amounts removed by simple (R), acid activated (Ra) and alkali activated (Rb) sawdust

Furthermore, the activation process, with acidic or alkaline 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 nitrate solution, but for larger water quantities the process can be conducted in cylindrical vessels in continuous flow, by gravitational means or under minor pressure or statically in large chambers filled with adsorbent sawdust.

## CONCLUSIONS

1. Sawdust obtained from alder wood proved excellent adsorption capacity for nitrate solutions in various concentrations, estimating that higher amounts of nitrate can be removed;

2. The activation procedure improved the material adsorption activity, especially when realized in acidic solution, in a very short period of time;
3. For all nitrate concentrations, the acid activated sawdust retained the considered ion over 99.9%;
4. The alkali activated sawdust retained almost as well at smaller concentrations and similar for the concentrations over 300 mg NO<sub>3</sub><sup>-</sup>/l;
5. The method proved effective for decreasing nitrate levels in wastewaters or in natural waters in order to be safe for consumption, even for small children.

## REFERENCES

1. Argun M.E., Dursun S., Ozdemir C., Karatas M., 2008 - *Heavy metal adsorption by modified oak sawdust*, Thermodynamics and kinetics. Journal of Hazardous Materials, 141: 77-85.
2. Balkaya N., 2002 - *Pesticide removal from wastewater*, Int. J. Water, 2 (2/3): 212,
3. Christianson Laura E., Helmers M. J., 2011 - *Woodchip bioreactors for nitrate in agricultural drainage*, Agriculture and Environment Extension Publications, Agriculture and Natural Resources, no. 10
4. Crini G., Badot P-M., 2010 – *Sorption processes and pollution – conventional and non-conventional sorbents for pollutant removal from wastewaters*, Presses universitaires de Franche-Comte, ISBN 978-2-84867-304-2
5. Department of the army, U.S. Army Corps of Engineers, 441 G Street, NW, Washington, DC 20314-1000, CEMP-CE, 2008 - *Pilot study using wood chips as an absorbent to treat wastewater from grease trap servicing*, Public Works Technical Bulletin, No. 200-1-59
6. Keng, P.S., S.L. Lee, S.T. Ha, Y.T. Hung and Ong S.T., 2013 - *Removal of hazardous heavy metals from aqueous environment by low-cost adsorption materials.*, Environmental Chemistry Letters (Article In Press).
7. Nwakaire J. N., S. L. Ezeoha, and B. O. Ugwuishiwu, 2013 - *Production of cellulosic ethanol from wood sawdust*. Agric Eng Int: CIGR Journal, 15(3): 136 – 140.
8. O'Connell, D.W., C. Birkinshaw and T.F. O'Dwyer, 2008 - *Heavy metal adsorbents prepared from the modification of cellulose: A review*, Bioresource Technology 99 : 6709-6724.
9. Wang Uen-Ping David, 1971 - *Adsorption of Sulphur Dioxide on Douglas Fir Woodchips*, Master of Science (M.S.) in Applied Science Thesis , Portland State University

## CHEMICAL AND SPECTRAL CHARACTERIZATIONS FOR SOME LIGNIN PRODUCTS

### CARACTERIZĂRI CHIMICE ȘI SPECTRALE ALE UNOR PRODUSE LIGNINICE

UNGUREANU Elena<sup>1</sup>, JITĂREANU Doina<sup>1</sup>, TROFIN Alina<sup>1</sup>,  
SANDU Tatiana<sup>1</sup>, POPA V.I.<sup>2</sup>  
e-mail: eungureanu@uaiasi.ro

**Abstract.** Lignin derivative (the commercial product - Protobind 1000) offered by the Granit Recherche Developement S.A. company, Lausanne-Schweitzerland was synthesized from annual plants. The present study's aim was to modify commercial lignins by the reaction of hydroxymethylation (produced in alkaline medium) and epoxydation (reaction with epichlorohydrin was performed in basic catalysis, aiming at increase the functionality) and to characterize the lignin derivatives chemical, spectral (<sup>1</sup>H NMR) and thermogravimetric analysis (TG). Studies have revealed some functional changes related to the difference in reactivity and reaction conditions.

**Key words:** Protobind 1000, lignin, hydroxymethylation, epoxydation, spectral and thermogravimetric analysis.

**Rezumat.** Lignina derivativă (produsul comercial Protobind 1000) oferită de firma Granit Recherche Developement S.A. Lausanne-Elveția a fost sintetizată din plante anuale. Scopul prezentului studiu este a de a modifica ligninele comerciale prin reacția de hidroximetilare (produsă în mediul alcalin) și epoxidare (reacție cu epiclorhidrina în cataliza bazică ce crește funcționalitatea) și de a caracteriza derivații ligninici prin analize chimice, spectrale (<sup>1</sup>H RMN) și termogravimetrice (TG). Studiile au relevat unele modificări funcționale legate de diferența de reactivitate și condițiile de reacție.

**Cuvinte cheie:** Protobind 1000, lignină, hidroximetilare, epoxidare, analize termogravimetrice și spectrale.

## INTRODUCTION

Globally, lignin is regarded as a raw material with high recovery potential, accessible from renewable sources, with low costs and a negligible pollution degree (Popa 1983; Ungureanu, 2011). The reactivity of lignin is determined by its particular structure in which specific functional groups are identified, as well as structural modifications induced by methods used for its separation from wood (Davison *et al*, 2006).

It is known that lignin has a very complex structure, which varies

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> "Gheorghe Asachi" Technical University of Iași, Romania

depending on the plant species, separation method and modification reactions that may induce particular characteristics. Regarding functional groups lignin presents at least three base functional groups in its structure: methoxylic, hydroxylic (alcoholic and phenolic) and the lateral propanic chain. Alongside these functional groups, in lesser amounts, there can be found carbonylic groups (approximate 1 group of CO at 5 C9 units), most of the times, fixated at the lateral chain. In some cases, the presence of carboxylic groups into the lignin can be noticed under the form of phenol carboxylic acids or of some small quantities of lactonic groups (Hoareau *et al*, 2004; Ungureanu *et al.*, 2009).

Lignin modification through hydroxymethylation and epoxydation offers possibilities of developing its functionality and this allows the extension of the application area for the synthesized derivatives (Căpraru *et al*, 2009).

Taking into account all these aspects, the objectives of this study are the modification of some lignin's from annual plants through a reaction of hydroxymethylation produced in an alkaline medium in the presence of formic aldehyde and of epoxydation effected out in an alkaline medium in the presence of epichlorhydrin and the characterization of lignin derivatives from a chemical, spectral ( $^1\text{H}$  NMR) and thermogravimetric methods.

## MATERIAL AND METHOD

The following materials have been used:

- Protobind 1000 (Pb1000), commercial lignin offered by Granit Recherché Développement Switzerland, with the following chemical characteristics presented in Table 1;

Table 1

The characteristics Protobind 1000

Characteristics	Protobind 1000
Solide, %	97.5-98.6
Ash, %	1.4-1.8
pH (10 % dispersion)	~ 3.5
Densitatea, g/mL	~ 0.3
Aromatic OH, mmole/g	1.8-1.9
COOH, mmole/g	2.1-2.3
T softening, °C	~ 200
Solubility in furfuryl alcohol, %	40.1
Solubility in aqueous alkali, %	94

- Formic aldehyde (37 %);
- Dimetil sulfoxid (DMSO);
- NaOH solution 0.1 N;
- Epichlorhydrin;

Work procedure:

### The hydroxymethylation reaction

The method used in the hydroxymethylation of the three lignin products was performed in a basic medium, in the presence of formic aldehyde (37 %), according to the technical literature (Ungureanu, 2011).

**Determination of total hydroxyl groups**

The total OH groups content was determined by chemical method with acetic anhydride in pyridine medium and from FT-IR spectral analysis. The Ar-OH group's content was determined by a UV-VIS method.

**The epoxidation reaction**

The epoxydation method achieved in a basic medium in the presence of epichlorhydrin through which the three types of lignin studied have been modified has been effected out according to the technical literature (Ungureanu, 2011).

**Epoxydation index**

Determination of the epoxy group was effected out by HCl addition on the epoxy group and titration of the acid excess with NaOH solution 0.1 N.

**Proton nuclear magnetic resonance spectroscopy ( $^1\text{H}$  NMR)**

Nuclear magnetic resonance (NMR) offers the richest and most complete information on the structure of organic compounds. For this purpose it was used a Bruker Avance DRX 400 MHz spectrometer.

**Process:** For investigation was necessary lignin acetylation and derivatives for a better dissolution in DMSO- $d_6$ . To obtain a "good" spectrum it is required to have concentrations of about 0.2 mmol/mL. Spectra processing was performed with a specialized program from SpectraManager series.

**Thermogravimetry**

The thermal analysis was performed using the METTLER TOLEDO derivatograph in  $\text{N}_2$  atmosphere with a flow of 20 mL/min and a heating rate of 15°C/min, in the temperature range 25-800 °C and sample mass of 4 ÷ 6 mg.

**RESULTS AND DISCUSSIONS**

During the reaction of hydroxymethylation performed for lignin, the reaction conditions have been varied (50°C temperature, 90°C respectively, reaction duration of three hours and pH 10.5, pH 12 respectively) in order to obtain highly functionals product.

The content of functional groups was determined according to the methods presented by different research groups. The other methods applied for chemical characterization were: the determination of carboxylic groups and of the metoxyl groups, the determination aromatic hydroxyl groups, the calculation of the fenolic groups/aliphatic groups' ratio, as well as, the determination siringyl/guaiacyl unit's ratio ( $\bar{S}/\bar{G}$ ).

Table 2

**The content of functional groups of modified and unmodified lignins**

Sample	T, °C	pH	OH total groups	Ar-OH groups	OCH <sub>3</sub> groups	Ak/Ar ratio	C=O groups	S/G ratio
Pb 1000	-	-	1.11	0.89	1.05	1.17	0.89	0.83
	90	12.0	1.23	0.98	1.15	1.27	0.95	0.96
	90	10.5	1.15	0.98	1.13	1.20	0.91	0.96
	50	10.5	1.14	0.98	1.12	1.22	0.95	0.96
	50	12	1.16	0.99	1.14	1.21	0.94	0.96

The information obtained has allowed the determination from this point of view of the optimal reaction conditions, namely: 90°C temperature, pH 10.5 and the reaction duration of three hours (Table 2).

The lignin obtained in optimal conditions there was characterized from the point of view spectral and thermogravimetric. As a consequence of the thermal analyses, it can be noticed that the modified product has a higher degradation temperature in the third stage, compared to the unmodified sample. (Table 3).

Table 3

**Characteristics of the thermal degradation process of the lignin derivatives**

Samples	Degradation stage	T <sub>i</sub> (°C)	T <sub>max</sub> (°C)	T <sub>f</sub> (°C)	Mass losses (%)
Pb1000	I	52	77	106	3.31
	II	229	267	330	17.05
	III	330	383	532	42.47
Pb1000H	I	64	83	122	4.71
	II	244	264	341	14.03
	III	341	379	523	34.28

(T<sub>i</sub> - initial temperature at which the degradation starts; T<sub>max</sub> – temperature corresponding to the maximum rate of degradation, T<sub>f</sub> – final temperature și W – mass losses %).

The characterization of the lignin has been achieved by monitoring the influence of temperature (50°C and 70°C respectively), the mass ratio between the lignin (L) and NaOH (L:NaOH = 1:3 and 1:6) and the reaction duration (3, 5 and 7 hours respectively). It can be noticed from table 4 that the best results can be obtained when the reaction is achieved at 70°C, for a L:NaOH=1:3 ratio and a three-hour reaction duration, appreciated as being *optimal reaction* conditions. The reaction yield was included in the 50-90 % interval, related to the mass of the reactants and it differs according to the type of sublayer and the purification degree after washing the derivatives. It can also be noticed that along with the temperature increase and the reaction duration, from 3 to 7 hours, appear a decrease of the epoxydation number.

Table 4

**Characteristics of the modified lignin's by epoxydation**

Sample	T, °C	L:NaOH (w/w)	t, h	CE, %		η, %	U, %	Ash %	Const. f.liq, %
				f.sol.	f.liq.				
Pb1000E	70	1:6	3	1.14	0.20	61	6.22	9.62	15.8
	70	1:3	3	1.62	0.61	62	4.36	0.98	17.4
	50	1:3	3	1.52	0.4	52	4.8	4.90	12.3
	70	1:3	5	1.32	0.52	60	5.12	3.21	11.32
	70	1:3	7	1.54	0.36	58	4.87	5.65	13.50

For characterization by <sup>1</sup>H-NMR spectroscopy the lignin was subjected to acetylation to aid dissolution in DMSO-d<sub>6</sub>. In figures 1 and 2 are shown the <sup>1</sup>H-



NMR spectra for Pb1000 lignin unmodified (Pb1000N) and hydroxymethylation (Pb1000H), and the results were interpreted using literature data. The spectrum recorded for the two lignins weak signals in the aromatic domain at 8.64 ppm and the methoxyl groups.

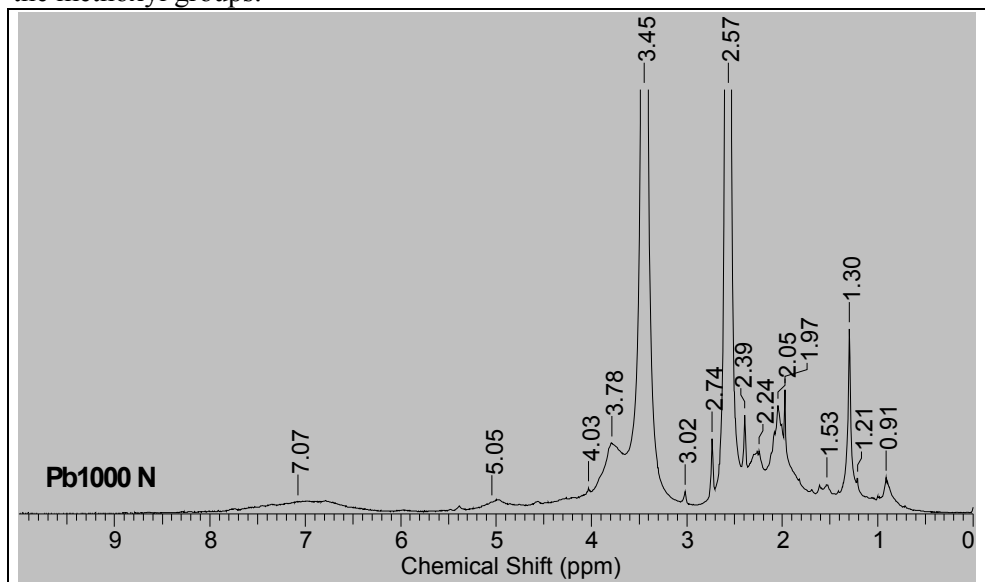


Fig. 1 -  $^1\text{H}$ -NMR spectra for unmodified lignin Pb1000

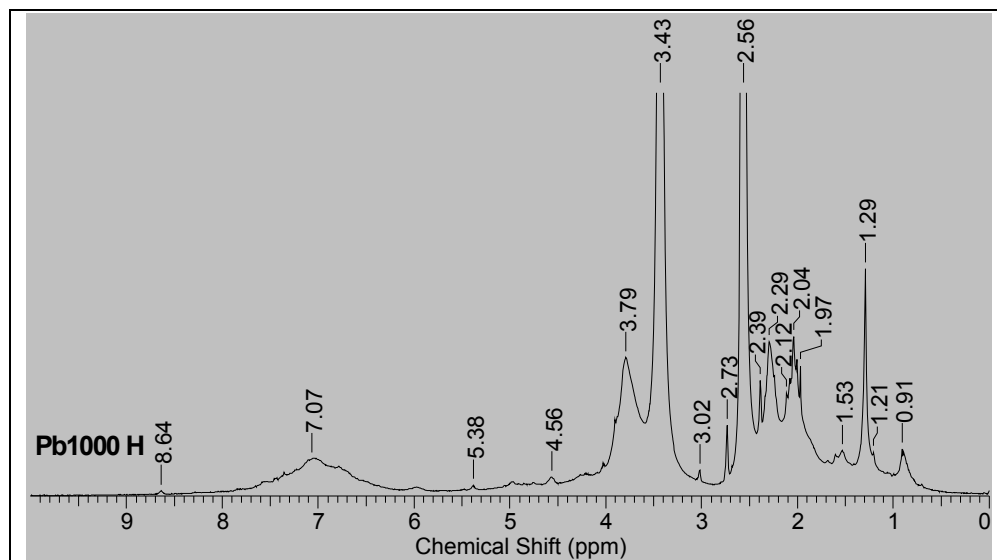
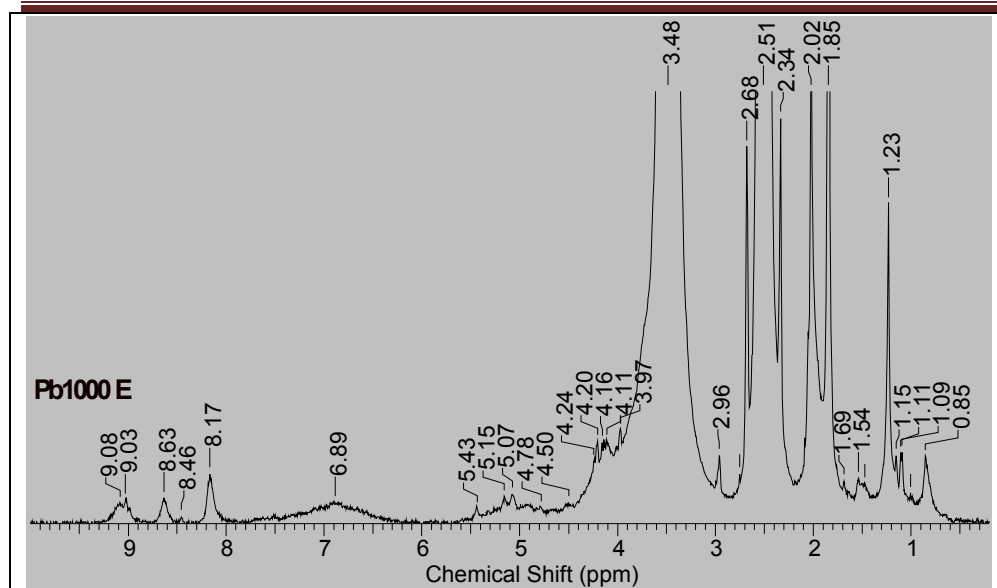


Fig. 2 -  $^1\text{H}$ -NMR spectra for modified lignin Pb1000H

Signals from 9.08-7 ppm confirms the presence of epoxy groups in lignin structure. Also stands out the signals of methoxyl and acetyl groups, more intense in the spectra of epoxidised lignin (Fig. 3).

Fig. 3.  $^1\text{H}$ -NMR spectra for modified lignin Pb1000E

## CONCLUSIONS

1. The  $^1\text{H}$ -NMR spectroscopy shows the change of functionality for lignin as a result of hydroxymethylation and epoxydation reaction.
2. The thermogravimetric analyses have proved that thermal degradation occurs in two and three stages respectively, according to the type and the degree of modification of the products tested but the hydroxymethylated /epoxydated derivatives have a higher thermostability, compared to the unmodified lignin.

## REFERENCES

1. Căpraru Adina-Mirela, Ungureanu Elena, Popa, V.I., 2009 - *Aspects concerning some biocides systems based on natural aromatic compounds aromatic compounds and their copper complexes*, Fibre and Pulping Chemistry, Oslo, Norway, p. 50-54.
2. Davison B.H., Drescher S.R., Tuskan G.A., Davis M. F., Nghiem N. P., 2006 – *Variation of S/G ratio and lignin content in a Populus family influences the release of xylose by dilute acid hydrolysis*, Appl. Biochem. Biotechnol., 42:129–132.
3. Hoareau W., Trindade W. G., Siegmund B., Castellan A., Frollini E., 2004 - *Sugar cane bagasse and curaua lignins oxidized by chlorine dioxide and reacted with furfuryl alcohol: characterization and stability*, Polym. Degrad. Stab., p. 86.
4. Popa V.I., 1983 - *Tehnologii de valorificare a ligninei*, Ed. Inst.Polith.Iași, p. 98.
5. Ungureanu Elena, 2011 - *Lignina, polimer natural aromatic cu ridicat potențial de valorificare*, Ed. PIM, Iași, p. 112.
6. Ungureanu Elena, Ungureanu O., Căpraru Adina-Mirela, Popa, V.I., 2009 - *The chemical modification and the characterization the straw lignin*, Cell. Chem. and Technol., 43 (7-8): 263-269.

## THE ANTHOCYANIN COMPOSITION OF A RED GRAPE POMACE IN RELATION WITH THE WINE INDUSTRY BY-PRODUCTS VALORIZATION IN ANIMAL FEED

### COMPOZIȚIA ÎN ANTOCIANI A TESCOVINEI REZULTATE DE LA OBȚINEREA VINULUI ROȘU ÎN VEDEREA VALORIFICĂRII REZIDUURILOR INDUSTRIEI VINICOLE ÎN HRANA ANIMALĂ

CHEDEA Veronica Sanda<sup>1</sup>, PALADE L. M.<sup>1</sup>,  
ROTAR M. C.<sup>1</sup>, CĂLIN L. G.<sup>1</sup>, DRAGOMIR C.<sup>1</sup>

e-mail: chedea.veronica@ibna.ro

**Abstract.** Grape pomace (GP), by-product of wine making, contains a large part of antioxidant polyphenols originating from the input material, the grapes. High production volumes, environmental impact and nutritional content of grape pomace makes it an important subject for careful valorisation. In order to valorize it, it is necessary to determine its chemical composition in bioactive molecules. In this study the anthocyanins were determined by LC-MS from a dried red grape pomace from Valea Călugărească, Romania. Based on their retention times, UV-VIS and MS spectra using standard compounds one anthocyan and three anthocyanidins were identified and quantified from GP acetone extract: peonidin 3-O-glucoside, delphinidin, cyanidin and malvidin. The most abundant anthocyanidin is delphinidin ( $23.93 \pm 0.12 \mu\text{g/mL}$  acetone extract) followed by malvidin ( $10.01 \pm 0.06 \mu\text{g/mL}$  acetone extract), the anthocyan peonidin 3-O-glucoside ( $4.78 \pm 0.04 \mu\text{g/mL}$  acetone extract) and cyanidin ( $2.63 \pm 0.06 \mu\text{g/mL}$  acetone extract). The results show that GP contains anthocyanins and anthocyanidins, which possess anti-inflammatory and anti-carcinogenic activity, cardiovascular disease prevention, obesity control, and diabetes alleviation properties, all of which are more or less associated with their potent antioxidant property.

**Key words:** grape pomace, anthocyanins, anthocyanidins, LC-MS

**Rezumat.** Tescovina, un reziduu al industriei vinicole conține o cantitate importantă de polifenoli provenind de la strugurii din care a fost obținută. Cantitățile mari rezultate, impactul asupra mediului precum și proprietățile nutritive ale tescovinei, fac din acest material rezidual subiectul unei valorificări atente. În vederea valorificării acesteia este necesară determinarea compoziției chimice. În acest studiu compoziția în antociani și antocianidine a unei tescovine rezultate de la obținerea vinului roșu din centru viticol Valea Călugărească, România, a fost determinată prin tehnica LC-MS. Pe baza timpilor de retenție, a spectrelor UV-VIS și a spectrelor de masă, folosind compușii standard corespunzători, au fost identificați și cuantificați un antocian și trei antocianidine: peonidina-3-O-glucozida, delfinidina, cianidina și malvidina. Delfinidina a fost găsită ca fiind cea mai abundentă antocianidină

---

<sup>1</sup> National Research Development Institute for Animal Biology and Nutrition, Balotești, Romania

( $23.93 \pm 0.12 \mu\text{g/mL}$  extract acetonă), urmat de malvidină ( $10.01 \pm 0.06 \mu\text{g/mL}$  extract acetonă), peonidină-3-O-glucozidă ( $4.78 \pm 0.04 \mu\text{g/mL}$  extract acetonă) și cianidină ( $2.63 \pm 0.06 \mu\text{g/mL}$  extract acetonă). Rezultatele indică faptul că tescovina conține antociani și antocianidine, compuși ce au activitate anti-inflamatorie și anticancerigenă, de prevenire a bolilor cardiovasculare, de control al obezității și a diabetului, care toate sunt mai mult sau mai puțin asociate cu activitatea lor antioxidantă.

**Cuvinte cheie:** tescovină, antociani, antocianidine, LC-MS

## INTRODUCTION

Europe produced in 2012, 16209965 tones of wine (Faostat, 2015). Romania ranks in 2012 on the 12<sup>th</sup> place in Europe and 21<sup>st</sup> place in the world in terms of wine production, with 123450 tones, resulting a large amount of waste that needs to be managed (Faostat, 2015).

Wine industry wastes account for almost 30% of the grapes used for wine production (Rondeau *et al.*, 2013). Grape pomace, a remnant of the winemaking process, is one of the most important residues of the wine industry. It consists of different amounts of grape, skin, pulp, seeds and stems if not previously removed (Fontana *et al.*, 2013; Yu and Ahmedna, 2013).

These waste materials contain biodegradable organic matter; however, their disposal generates huge amounts of industrial waste and creates serious environmental problems (Gonzalez-Paramas *et al.*, 2004). The waste loads at the processing plants could be significantly reduced through by-product usage (Bordiga *et al.*, 2015). Grape pomace, still contain a significant amount of phenolic compounds with beneficial health-related effects (Torres *et al.*, 2002; Laufenberg *et al.*, 2003; Sagdic *et al.*, 2011).

Within the grape pomace the grape skin brings the highest concentration of anthocyanins. The most abundant of these compounds in red grapes are anthocyanins, mainly 3-glycosides, 3-acetylglycosides and 3-p-coumaroylglycosides of malvidin (Mv), peonidin (Pn), delphinidin (Dp), petunidin (Pt) and cyanidin (Cy) (Wulf and Nagel, 1978).

Anthocyanins are specific compounds of red grapes, located mainly in the skin of the grapes. The flavylium cation from their structure includes two benzene rings, linked by an oxygenated cationic unsaturated heterocycle, derived from the 2-phenyl-benzopyrylium nucleus (Lorrain *et al.*, 2013). They are glycosylated derivatives of five aglycones or anthocyanidins: cyanidin, peonidin, petunidin, delphinidin and malvidin (Makris and Kefalas, 2013; Ky *et al.*, 2014).

The chemical composition of the grape marc has to be determined prior to its utilization as dietary feedstuff for farm animals. In our study, the anthocyanins composition of a red grape pomace from Valea Călugărească winery, Romania, was determined by High Performance Liquid Chromatography-Photo Diode Array coupled with Mass Spectroscopy (HPLC-PDA-MS).

## MATERIAL AND METHOD

### Anthocyan extraction

The red grape pomace was provided by a local producer and derived from Valea Calugareasca, a Romanian winery. The anthocyanins from the dried grape pomace were extracted in acetone 80% (ratio, sample: solvent being 1:7 w/v) for 20 hours at 37°C with continuous shaking. After the extraction the liquid phase was collected, representing the GP acetone extract.

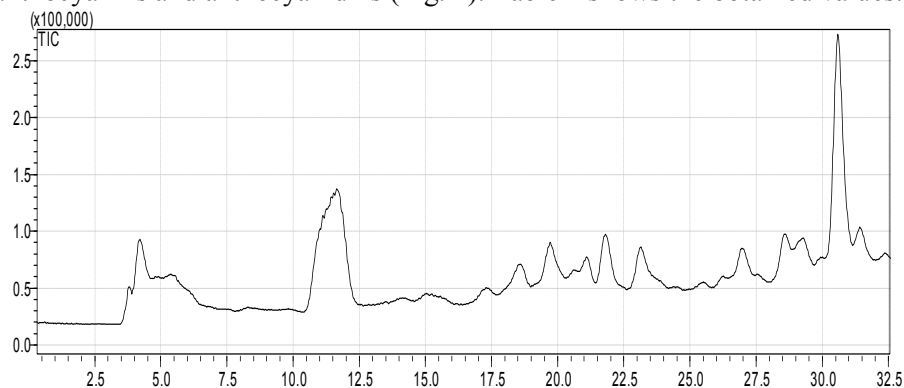
### High Performance Liquid Chromatography-Photo Diode Array coupled with Mass Spectroscopy (HPLC-PDA-MS) anthocyanin composition analysis for the grape pomace

The chromatographic measurements were performed using a complete HPLC SHIMADZU system using a C18 column. The HPLC system was coupled to a mass spectroscopy detector, LCMS-2010, using an electrospray ionization interface ESI. Due to the specificity of the studied components, we used the positive ionization mode. The analytical mobile phase consisted of 5% formic acid in water (solvent A) and 5% formic acid in methanol (solvent B). The compounds were separated using gradient elution with 0.15 mL/min flow. The experiments were conducted at 40°C constant column temperature; one analytical run took 47 minutes.

The calibration curve and the correlation coefficient were calculated, which allowed us to determine the range of response linearity. Calibration curves were produced for 7 different points, the measurements being done in triplicates.

## RESULTS AND DISCUSSIONS

HPLC-MS method was used for the qualitative and quantitative analysis of GP anthocyanins and anthocyanidins (Fig. 1). Table 1 shows the obtained values.



**Fig. 1** - HPLC-MS chromatogram of the dry grape pomace acetone extract (GP; delphinidin  $[M+H]^+=303$ , malvidin  $[M+H]^+=331$ , cyanidin  $[M+H]^+=287$ , peonidin 3-O-glucoside  $[M+H]^+=463$ ).

Table 1

Some performance characteristics of HPLC-MS method and the results concerning the anthocyanidins and anthocyanins

Analyte	t <sub>R</sub> minutes	[M+H] <sup>+</sup>	Equation of the standardization curve A:aria; C: conc. (µg/mL)	R	LoD (µg/mL)	Range of response linearity (µg/mL)	GP content (µg/mL)
Delphinidin	20±0.4	303	A=616931.5×C+349797.1	0.9996	0.04	0.5-50	23.93±0.12
Peonidin 3-O-glucoside	19.6±0.2	463	A=756370.8×C+516791.1	0.9996	0.07		4.78±0.04
Cyanidin	20.9±0.1	287	A=1651123×C+873662.6	0.9996	0.11		2.63±0.06
Malvidin	27.1±0.3	331	A=1109632×C+619282.7	0.9996	0.12		10.01±0.06

The equations of the calibration curves show a good linearity between the peak area and the concentration of the analyte, over a 1.5 ( $\mu\text{g/mL}$ ) range of concentrations. The performance characteristics of HPLC-MS method for 4 anthocyanins and the analytical results are shown in Table 1.

Based on the retention time, of the UV-VIS spectra and on the mass spectra, using the proper standard compounds, we identified and quantified 1 anthocyan, peonidin 3-O-glucoside and three anthocyanidins: delphinidin, cyanidin and malvidin. The most abundant anthocyanidin is delphinidin ( $23.93 \pm 0.12 \mu\text{g/mL}$  acetone extract), followed by malvidin ( $10.01 \pm 0.06 \mu\text{g/mL}$  acetone extract), peonidin 3-O-glucoside ( $4.78 \pm 0.04 \mu\text{g/mL}$  acetone extract) and cyanidin ( $2.63 \pm 0.06 \mu\text{g/mL}$  acetone extract).

These results show that the grape pomace is rich in anthocyanins and anthocyanidins, compounds with anti-inflammatory and anticarcinogenic effects, compounds which may prevent cardiovascular diseases, may control obesity and diabetes in human patients. These effects are more or less associated to the antioxidant activity of these anthocyanins.

## CONCLUSIONS

1. The experimental results show that the red grape pomace, by-product from Valea Călugărească winery, contains the following anthocyanins and anthocyanidins: delphinidin, malvidin, peonidin 3-O-glucoside and cyanidin.
2. Delphinidin is the most abundant anthocyanidin.

*Acknowledgments:* This work was supported by funds from the National Research Project PN-II-RU-TE-2012-3-0048 granted by the UEFISCDI of Romanian Ministry of Education.

## REFERENCES

1. **FAOSTAT, 2005** - Browse data. Retrieved 06 July 2015 from <http://faostat3.fao.org/browse/Q/QP/E>.
2. **Rondeau P., Gambier F., Jolibert F., Brosse N., 2013** – *Compositions and chemical variability of grape pomaces from French vineyard*. Industrial Crops and Products, 43: 251–254.
3. **Fontana A.R., Antonilli A., Bottini R. 2013** - *Grape pomace as a sustainable source of bioactive compounds: extraction, characterization, and biotechnological applications of phenolics*. Journal of Agricultural and Food Chemistry, 61: 8989–9003.
4. **Yu J., Ahmedna M. 2013** - *Functional components of grape pomace: their composition, biological properties and potential applications*. International Journal of Food Science and Technology, 48: 221–237.
5. **Gonzalez-Paramas A., Esteban-Ruano S., Santos-Buelga C., Pascual-Teresa S., Rivas-Gonzalo J., 2004** - *Flavanol content and antioxidant activity in winery byproducts*. Journal of Agricultural Food and Chemistry, 52: 234–238.
6. **Bordiga M., Travaglia F., Locatelli M., Arlorio M., Coisson J. D., 2015** - *Spent grape pomace as a still potential by-product*. International Journal of Food Science and Technology, 50: 2022–2031.

7. **Torres, J.L., Varela, B., García, M.T. Carilla J., Matito C., Centelles J. J., Cascante M., Sort X., Bobet R. 2002** - Valorization of grape (*Vitis vinifera*) by-products. Antioxidant and biological properties of polyphenolic fractions differing in procyanidin composition and flavonol content. *Journal of Agricultural and Food Chemistry*, 50: 7548–7555.
8. **Laufenberg G., Kunz B., Nystroem M., 2003** - Transformation of vegetable waste into value added products: (A) the upgrading concept, (B) practical implementations. *Bioresource Technology*, 87: 167–198.
9. **Sagdic O., Ozturk I., Ozkan G., Yatim H., Ekici L., Yilmaz M.T., 2011-** RP-HPLC-DAD analysis of phenolic compounds in pomace extracts from five grape cultivars: evaluation of their antioxidant, antiradical and antifungal activities in orange and apple juices. *Food Chemistry*, 126: 1749–1758.
10. **Wulf L.W., Nagel C.W., 1978-** High-pressure liquid chromatographic separation of anthocyanins of *Vitis vinifera*. *American Journal of Enology and Viticulture* 29(1):42–49.
11. **Lorrain B., Ky I., Pechamat L., Teissedre P.L., 2013** - Evolution of analysis of polyphenols from grapes, wines, and extracts, *Molecules* 18:1076-1100.
12. **Makris D., Kefalas P., 2013** - Characterization of polyphenolic phytochemicals in red grape pomace, *International Journal of Waste Resources*, 3 (2):1-4.
13. **Ky I., Crozier A., Cros G., Teissedre P.-L., 2014** – Polyphenols composition of wine and grape sub-products and potential effects on chronic diseases, *Nutrition and Aging*, 2:165-177



## THE PALYNOLOGICAL CHARACTERIZATION AND THE POLLEN POTENTIAL GERMINATION OF *RHODODENDRON RACEMOSUM* FRENCH.

### CARACTERIZAREA PALINOLOGICĂ ȘI A POTENȚIALULUI GERMINATIV AL POLENULUI DE *RHODODENDRON RACEMOSUM* FRENCH.

**PĂDUREANU Silvica<sup>1</sup>**

**e-mail:** silvyp27@yahoo.com

**Abstract.** The palinological characterization of *Rhododendron racemosum* highlights the organization of pollen in the pollen tetrad maintained by viscin threads. The set of determinations related to pollen morphology (the color of pollen grains, the ornamentation of exin, the pollen tetrad size, the number of pore/pollen grain) from *Rhododendron racemosum* complements the information in the literature regarding the characterization of from this taxon. To test the germination potential were used many variations of nutrient medium with different concentrations in sugar from 0% to 100%. The pollen germination of *Rhododendron racemosum* is expressed in the highest degree on the nutrient medium enriched with 25% sucrose, 168 hours after inoculation. Very high germination capacity, over 90% to *Rhododendron racemosum* pollen demonstrates that the pollen meiosis in this genotype is balanced and the male gametes are fertile. The data obtained have theoretical importance (in taxonomy) and in getting interspecific hybrids within the genus *Rhododendron*.

**Key words:** *Rhododendron racemosum*, pollen tetrad pollen, nutritive medium, germination capacity

**Rezumat** Caracterizarea palinologică la *Rhododendron racemosum* scoate în evidență modul de organizare a polenului în tetrade polinice, menținute prin fibre de viscină. Setul de determinări privitoare la morfologia polenului (culoarea granulelor, ornamentația exinei, dimensiunea tetradelor polinice, numărul porilor germinativi/granulă de polen) la *Rhododendron racemosum* completează informațiile din literatură referitoare la caracterizarea polenului acestui taxon. Pentru testarea potențialului germinativ s-au folosit mai multe variante de mediu nutritiv cu diferite concentrații glucidice, cuprinse între limitele 0% și 100%. Germinarea polenului de *Rhododendron racemosum* se exprimă în cel mai înalt grad pe substrat nutritiv îmbogățit cu 25% zaharoză, după 168 de ore de la inoculare. Capacitatea de germinare foarte ridicată, de peste 90% al polenului de *Rhododendron racemosum* demonstrează că meioza polinică la acest genotip este echilibrată, iar gameții masculini sunt fertili. Datele obținute au importanță teoretică (în taxonomie), dar și practică pentru lucrările de ameliorare pentru obținerea hibridilor interspecifice din cadrul genului *Rhododendron*.

**Cuvinte cheie:** *Rhododendron racemosum*, tetradă polinică, mediu nutritiv, capacitate de germinare

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine Iași, Romania

## INTRODUCTION

The genus *Rhododendron* L., one of the largest and diverse genera of *Ericaceae* comprises over 1000 species. The centre of diversity of the genus is in the Himalaya (Maurizio and Grafl 1969, Podbielkowski 1991, cited by Weryszko-Chmielewska and Chwil M., 2005).

These plants are represented by shrubs of great beauty and elegance with a large spread. The species and cultivars obtained by hybridization are in large numbers. Called "kings ornamental shrubs" around the rhododendrons has developed an entire horticultural specialized on production and marketing of ornamental woody plants.

In the works hybridization has great importance the pollen of parental plants. This study aimed to determine the morphological and germination capacity of pollen from *Rhododendron racemosum* French.

## MATERIAL AND METHOD

The biological material is represented by the fresh pollen of *Rhododendron racemosum* French. In order to define the pollen morphology, we determined shape of pollen grains, exine sculpturing, size of pollen tetrad and number of germinative pores/pollen grain. For determining the shape of pollen grains, the apertures (number and arrangement), the exine sculpturing, we have used the Hund Wetzlar microscope, at which we took microphotographs. For determining the size of pollen we did micromesurements at 1000 pollen tetrads (Oldfield, 1959). The values obtained were statistically processed, resulting the biostatistics indexes. For establishing the number of germinative pores/pollen grain, we have done determinations on 1000 pollen tetrads. The method consisted in introducing the pollens in a mixture of sulphuric acid and acetic acid. For study of the germination pollen, we have used the hanging drop method (Stanley and Linskens, 1985). The nutritive mediums necessary for the germination of pollens consisted in distilled water, agar 1% and sucrose at different concentrations: 0%, 5%, 15%, 25%, 40%, 50%, 70% and 100%. Thus, 8 experimental variants resulted. For each experimental variant, we have used 10 "wet rooms". The amount of inoculated pollen per each medium was the same in all cases. Readings at the Hund Wetzlar optic microscope were done at 4, 24, 48, 72, 96, 168 and 192 hours since the pollen inoculation in mediums, thus, being established the percent dynamics of the germination capacity for this genotype. The germination capacity was expressed as percentage, by reporting the number of germinated grains to total pollen grains.

## RESULTS AND DISCUSSIONS

### The palynological characterization of *Rhododendron racemosum*

The pollen of *Rhododendron racemosum* is spheroidal, white isabelline. The surface sculpturing is psilate.

The pollen grains of this taxa remain adherent in tetrahedral tetrads of four pollen grains. Each tetrad are provided on her outer surfaces with long viscin threads. Viscin threads occur among the pollen tetrads and presumably play a role

in pollen removal from the anthers and its adhesion to pollinators (Sarwar and Takahashi, 2013). Hesse M. *et al.* (2000) suggest that any pollen with viscin threads to the highly specialized pollination mode. According to Hesse M. *et al.* (2000) that viscin threads increase the efficiency of pollination, and their presence implies highly specific pollinators for accurate delivery of pollen to stigma. In our view, pollen in tetrad tetrahedral organization associated with viscina is minimum energy and maximum efficiency.

That pollen from *Ericaceae* and *Onagraceae* presents viscin threads (Hesse M., 1983). In addition, in *Boraginaceae* and *Scrophulariaceae* are reported (Hesse M *et al.*, 2000). For taxonomic purpose, the pollen of *Rhododendron* has also been studied by others (Bowers C, 1930; Tarnavschi I. *et al.*, 1987; Weryszko-Chmielewska and Chwil, 2005; Zhang *et al.*, 2009).

Each pollen grain/tetrad has three equally spaced apertures, having 3-colporate (fig. 1).

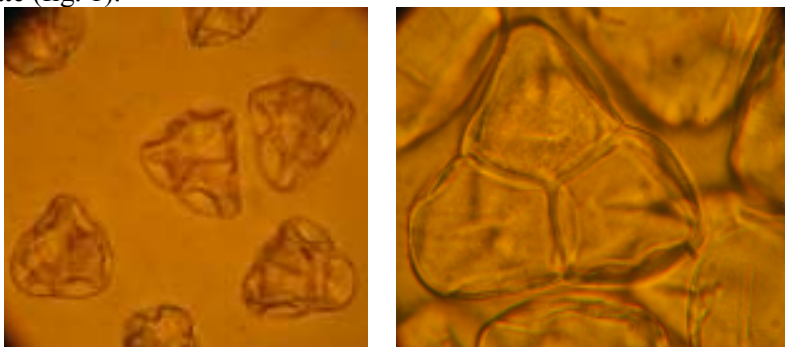


Fig. 1 - Pollen tetrads of *Rhododendron racemosum*: 400x (left); 1000x (right) (Original)

The measurements of pollen tetrads were made in conformance with the recommendations by Oldfield F. (1959), by determining only one side of the tetrad, since all other sides are perfectly equal. Side of tetrad has an average value of 62.79  $\mu\text{m}$  (tab. 1). The coefficient of variation (s%) indicates a low variability for pollen tetrad of *Rhododendron racemosum*. Each pollen grains/tetrad is a tricolporate grain, with the pore area of adjacent grains in contact.

The pollen grains present in the pollen tetrad of *R. racemosum* have more frequently in three pores, rarely two pores or one pore germinating (tab. 2, fig. 2). The data are consistent with the literature (Oldfield, 1959; Tarnavschi *et al.*, 1987).

Table 1

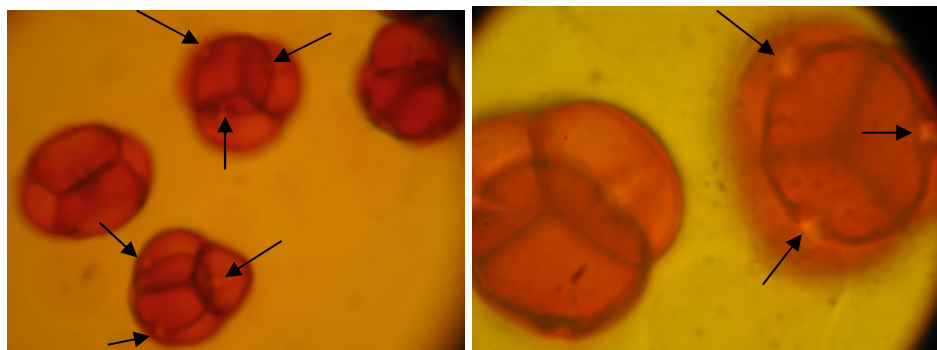
**Variability of pollen tetrad size in *Rhododendron racemosum***

Type of axis	Mean value ( $\mu\text{m}$ )	Minimum value ( $\mu\text{m}$ )	Maximum value ( $\mu\text{m}$ )	Variation height ( $\mu\text{m}$ )	S ( $\mu\text{m}$ )	S%	$\bar{S\chi}$ ( $\mu\text{m}$ )
side of pollen tetrad	62.79	55.20	79.35	24.15	4.83	7.69	0.76

Table 2

**Number of germinative pores/pollen grain in *Rhododendron racemosum***

mean value ( $\bar{X}$ )	% pollen grains with 1 or 2, or 3 germinative pores		
	1	2	3
2.84	3	10	87



**Fig. 2** - Germinative pores at pollen grain of *Rhododendron racemosum*: 400x (left); 1000x (right) (Original) (the arrows indicate the germinative pores)

The fact that the pollen not shows more than 3 germinative pores indicated that probably polyploidy not occurred in this taxa.

Germinating potential of pollen at *Rhododendron racemosum*

Results related to pollen germination of *R. racemosum* are summarized in table 3. Of the four pollen grains of the tetrad, only one germinate. After 4 hours since inoculation of the pollen on nutrient mediums, it was found that no germination were recorded on any of the eight types of medium. After 24 hours of inoculation, pollen germinated on six mediums. On the medium without sucrose and on that with 50% sucrose germinated only 4% and 5% respectively pollen tetrads. On mediums enriched with 25% and 40% sucrose, registered 73% and 72% respectively germinated pollen. After 48 hours, on medium deficiency in sucrose, pollen tubes were resorbed. On the medium with 100% sucrose not germinated pollen at all. Note that three mediums ensured the highest % of germinated pollen: 15%, 25% and 40% addition sucrose. After 72 hours, is found that the two mediums (pure water and 100% sucrose) are recalcitrant for pollen germination process. The level increased, maintains in three cases: with 15%, 25%, 40% sucrose, that supported the highest in percentage of germination capacity of the pollen. After 96 hours, the germination potential increases on mediums with 5-70% sucrose. Even after this time, the highest values of the germination capacity are provided from mediums with 15%, 25%, 40% sucrose. Of these, on medium with 25% sucrose, pollen recorded the highest values of germination.

After 168 hours it is noted that the pollen germination rate values remain almost constant with slight growth (for mediums with 25% to 50% sucrose) comparing to previous interval. On the medium enriched with 25% sucrose, pollen achieved the highest share of the germination (94%). After 192 hours, the germination remains the same as in the previous interval. Even after 8, days the pollen germination was not produced on the mediums with 0% and 100% sucrose.

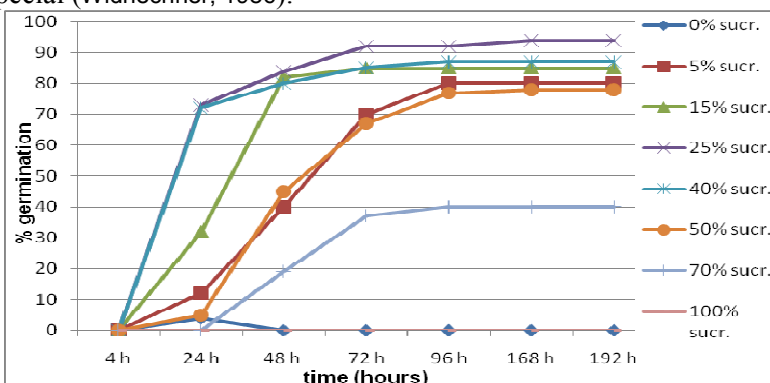
Table 3

**The pollen germination (%) of *Rhododendron racemosum***

time from inoculation (hours)	% sucrose in medium							
	0%	5%	15%	25%	40%	50%	70%	100%
after 4 h	0	0	0	0	0	0	0	0
after 24 h	4	12	32	73	72	5	0	0
after 48 h	0	40	82	84	80	45	19	0
after 72 h	0	70	85	92	85	67	37	0
after 96 h	0	80	85	92	87	77	40	0
after 168 h	0	80	85	94	87	78	40	0
after 192 h	0	80	85	94	87	78	40	0

The dynamic analysis of pollen germination rate of *Rhododendron racemosum* show that in the first 24 hours after inoculation, the pollen germinates explosive on certain mediums. The values of the germination capacity are very different depending on the sugar concentration in mediums. After 168 hours of inoculation reach the maximum values of the germination on medium fortified with 25% sucrose (fig. 3).

Investigations of the potential germination reveal that the viability of pollen of *R. racemosum* it is high. This is in agreement with the literature according to which *Rhododendron* breeders store the pollen up to 2 weeks under the conditions of the special (Widrechner, 1986).

**Fig. 3 - The germination dynamics of pollen in *Rhododendron racemosum***

## CONCLUSIONS

Pollen of *Rhododendron racemosum* is organized in tetrahedral tetrads, maintained by viscin threads. The pollen grains is tricolporate. The size of pollen tetrads is homogeneous, correlated with high fertility. The maximum values of potential germination achieves after 168 hours after inoculation of pollen on nutrient medium with 25%. The pollen of *R. racemosum* is viable for at least 8 days tested in this experiment, at room temperature.

## REFERENCES

1. Bowers C.G., 1930 - *The Development of Pollen and Viscin Strands in Rhododendron catawbiense*. Bulletin of the Torrey Botanical Club, vol. 57, no. 5, p. 285-313.
2. Hesse M., 1983 - *Dissimilar pollen tetrad development in Ericaceae and Onagraceae causes family-specific viscin thread configuration*. Plant Systematics and Evolution, vol. 143, no. 2, p. 163-165.
3. Hesse M., Vogel S., Halbritter H., 2000 - *Thread-forming structures in angiosperm anthers: Their diverse role in pollination ecology*. Plant Systematics and Evolution, vol. 222, no. 1, p. 281-292.
4. Oldfield F., 1959 - *The pollen morphology of some of the west European Ericales. In Preliminary descriptions and a tentative key to their identification*. Pollen et Spores Museum National d'Histoire Naturelle, vol. 1, p. 19-48.
5. Sarwar G.A.K.M., Takahashi H., 2013 - *Pollen morphology of Rhododendron L., and related genera and its taxonomic significance*. Bangladesh J. Plant Taxon, vol. 20 no. 2, p. 185-199.
6. Stanley R.G., Linskens H.F., 1985 - *Pollen biology, biochemie, gewinnung und verwendung urs freund verlag*. Greifenberg Ammersee, p. 349.
7. Tarnavski I.T., Șerbănescu-Jitariu G., Mitroiu-Rădulescu N., Rădulescu D., 1987 - *Monografia polenului florei din România*. Ed. Acad. R.S.R., vol. 2, p. 65-667.
8. Weryszko-Chmielewska E., Chwil M., 2005 - *Morphological features of the nectary and of the pollen grains and the foraging value of the flowers of yellow azalea (Rhododendron luteum Sweet)*. J. of Apicultural Science, vol. 49, no. 2, p. 5-12.
9. Widrechner M. P., 1986 - *Short term pollen storage of two Rhododendron Simsii cultivars*. J. American Rhododendron Society, vol. 40, no. 3.
10. Zhang Y.J., Jin X.-F., Ding B.-Y., Zhu J.P., 2009 - *Pollen morphology of Rhododendron subgen. Tsutsusi and its systematic implications*. Journal of Systematics and Evolution, vol. 47, no. 2, p.123-138.

## PECULIARITIES OF POLLEN TUBE GROWTH OF *RHODODENDRON RACEMOSUM* FRENCH.

### PARTICULARITĂȚILE DE CREȘTERE ALE TUBULUI POLINIC DE *RHODODENDRON RACEMOSUM* FRENCH.

**PĂDUREANU Silvica<sup>1</sup>**

e-mail: silvyp27@yahoo.com

**Abstract:** This paper is an extension of a previous study on the pollen morphology and germination potential of *Rhododendron racemosum* French. In this study are referred cytological aspects of process of pollen germination at this taxon. Was examined the growth of pollen tube length based on the concentration of sucrose in mediums, over a period of 192 hours after inoculation of pollen. The increase in length of the pollen tubes was in most cases directly proportional to level of the pollen germination capacity, in sense that the longest pollen tubes were formed on nutrient mediums which permit the biggest germination of the pollen tetrad. In this respect was demonstrated that the medium with 25% sucrose ensure the growth of the longest pollen tubes correlated with the style's length of flower from this genotype. Also highlights the formation stages of pollen tube, his longevity, anomalies in male gametophyte development to *Rhododendron racemosum*.

**Key words:** *Rhododendron racemosum*, nutritive medium, pollen, pollen tube

**Rezumat:** Lucrarea reprezintă o extensie a unui studiu anterior referitor la morfologia și potențialul germinativ al polenului de *Rhododendron racemosum* French. În prezentul studiu se face referire asupra aspectelor citologice ale procesului germinativ al polenului. Se analizează creșterea în lungime a tuburilor polinice în funcție de compoziția glucidică a mediilor nutritive în decursul a 192 ore de la inocularea polenului. Creșterea în lungime a tuburilor polinice a fost în majoritatea cazurilor, în relație de directă proporționalitate cu nivelul capacității de germinare a polenului, în sensul că cele mai lungi tuburi polinice s-au format pe mediile nutritive care permit germinarea a cât mai multor tetrade polinice. În acest sens se demonstrează că mediul cu 25% compoziție glucidică asigură creșterea celor mai lungi tuburi polinice în corelație cu lungimea stilului florii de la acest genotip. De asemenea se evidențiază particularitățile edificării tubului polinic, longevitatea și anomaliiile în dezvoltarea gametofitului mascul la *Rhododendron racemosum*.

**Cuvinte cheie:** *Rhododendron racemosum*, medii nutritive, polen, tub polinic

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine Iasi, Romania



## INTRODUCTION

Within the *Rhododendron* genus there are performed a variety of hybridization with the goal of wider biodiversity of this genus, in ornamental purpose. In addition, in *Rhododendron* genus were identified natural hybrids, as are those between *Rhododendron spiciferum* and *R. spinuliferum*, and populations of *R. × duclouxii* represent hybrid swarms from backcrossing or selfing of  $F_1$  plants (Yan *et al.*, 2013). This fact demonstrates for the first time the existence of a natural homoploid hybrid in this genus.

In hybridizations there is very important the quality of male parent pollen. In this regard there have been studies on pollen tube growth in rhododendrons. For example, in the cross *Rhododendron impeditum*  $\times$  *R. williamsianum*, the majority of the tubes do not reach an ovule because the tubes have often thickened tips or, when they continue growth, they start to coil without penetrating into an ovule (Kho and Baer J., 1970).

Hybrid sterility at rhododendrons occurs from other causes, such as the chromosome incompatibilities during the production of sex cells of  $F_1$  (Krebs and Leach, 1997).

This paper is an extension of the work on palinological characterization and pollen potential germination at *Rhododendron racemosum*. This paper brings in the foreground the behavior of pollen tube at this taxon.

## MATERIAL AND METHOD

The biological material is represented by pollen of *Rhododendron racemosum* French. Pollen was sampled at the anthesis phase.

The pollen germination process was determined with hanging drop (Erdman G., 1952). So, pollen grains have been inoculated on agar nutritive mediums (1%), at which sucrose was added at different concentrations: 0%, 5%, 15%, 25%, 40%, 50%, 70%, 100%. Eight experimental variants resulted. For each experimental variant, we have used 10 "wet rooms". The quantity of pollen inoculated on these mediums was the same in all the cases.

In parallel with the study of germination capacity, were made micromasurements for determining the dynamics for the extension of pollen tubes. In this respect, the readings at microscope were done at 4, 24, 48, 72, 96, 168 and 192 hours since the inoculation of the pollen grains on nutritive medium.

Micromasurements of pollen tube length were recorded directly by an ocular micrometer fitted to the eyepiece on microscope based on micrometer scale ( $\mu\text{m}$ ). The length of the pollen tube was expressed by micrometers ( $\mu\text{m}$ ). For pointing out the characteristics of pollen tubes from this genotype, photographs were taken at Hund Wetzlar optical microscope.

## RESULTS AND DISCUSSIONS

Results related to the average length of pollen tubes of *Rhododendron racemosum* depending on the sugar composition of the nutrient mediums and the time from inoculation pollen on mediums are summarized in table 1.



Table 1

**The average length of the pollen tube ( $\mu\text{m}$ ) at *Rhododendron racemosum***

time from inoculation (hours)	% sucrose in medium							
	0%	5%	15%	25%	40%	50%	70%	100%
after 4 h	0	0	0	0	0	0	0	0
after 24 h	345	658	688	855	242	9	0	0
after 48 h	0	1280	1309	1402	1112	756	138	0
after 72 h	0	2268	2295	2359	1123	1110	1106	0
after 96 h	0	2707	2950	3577	1803	1952	1638	0
after 168 h	0	3560	3910	4036	2175	2087	1999	0
after 192 h	0	4144	4200	4385	2995	2767	2520	0

After the first 4 hours after inoculation of the nutrient mediums, pollen tubes no were formed at all in experiment.

After 24 hours since inoculation of the pollen on artificial mediums, occurred the first pollen tubes on six mediums: pure water (0% sucrose), 5%, 15%, 25%, 40% and 50% sucrose. The length of pollen tubes formed first varied widely, depending on the sugar composition. So, the shortest were those edified on medium enriched with 50% sucrose, and the longest on medium with 25% sucrose. On the medium very enriched with sucrose, germination has not occurred and therefore have not formed any pollen tubes.

After the 48 hours of the inoculation, the pollen tubes have been stretched enough, were doubling the length in some cases. Pollen tubes formed on deficient medium in sucrose, 24 hours ago, were resorbed, completely degenerated. On the mediums with the addition of 70% sucrose, they were built first tubules with an average length of 138  $\mu\text{m}$ . The longer pollen tubes grew on mediums by 5%, 15% and 25% sucrose, the average values were 1280, 1309 and 1402  $\mu\text{m}$  respectively.

After 72 hours, reveals significant increases in pollen tubes already formed. The medium saturated in sucrose (100% sucrose), did not allow pollen tube formation. Also, lack of sugar in medium (0% sucrose) still remains averse for the development of the male gametophyte from this taxa.

After 96 hours of inoculation pollen on artificial mediums, were significant increases in the length of pollen tubes already formed. The longest tubes measured in average 3577  $\mu\text{m}$ , and the shortest 1638  $\mu\text{m}$ . The longest were formed on medium with 25% sucrose, and the shorter on medium with 70% sucrose.

After 168 hours of starting the experiment, pollen tubes already formed in all previous intervals had significant increases. The longest tubes were the conditions on medium with 25% sucrose, followed in descending order by pollen tubes formed in environments enriched with 15%, 5%, 40%, 50%, 70%.

After 192 hours, recorded insignificants increase of the pollen tubes.

Even after eight days after the inoculation, the pollen of *R. racemosum* did not germinate on medium absent in sucrose and on medium with 100% sucrose.

Dynamic analysis shows that the pollen tube growth during 8 days is generally upward. Growth rate is increasing after 48 hours, the maximum rate is on medium with 25% sucrose, when was the longest tubes formed after 8 days (fig. 1).

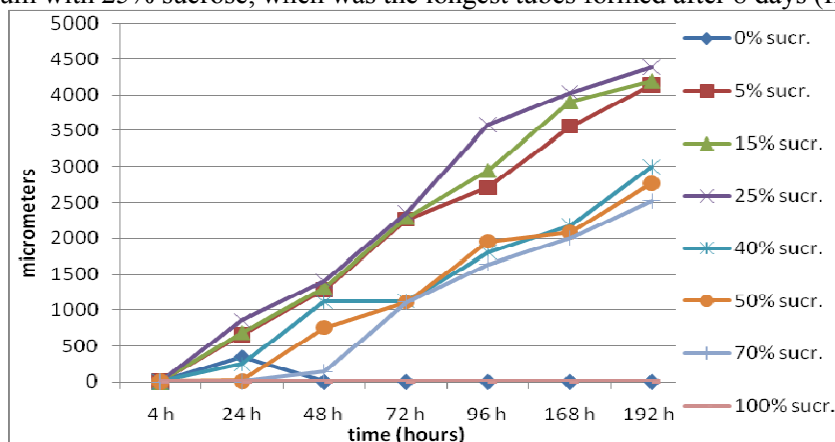


Fig. 1 - Dynamics of average length of the pollen tube at *Rhododendron racemosum*

Analysis of pollen tube growth in length highlights that to *R. racemosum*, the pollen germinated has high viability proven by the viability of pollen tubes that even after 8 days of their growth, they have not degenerated. These findings are consistent with data Palser's *et al.* (1992), according to which, tubes reached the ovary in 5-10 days depending on the species of *Rhododendron* and took several days after entering the upper ovary to reach the base of the placentae. To *R. macgregoriae*, fertilization occurs about 6–7 days after pollination (Williams *et al.*, 1991).

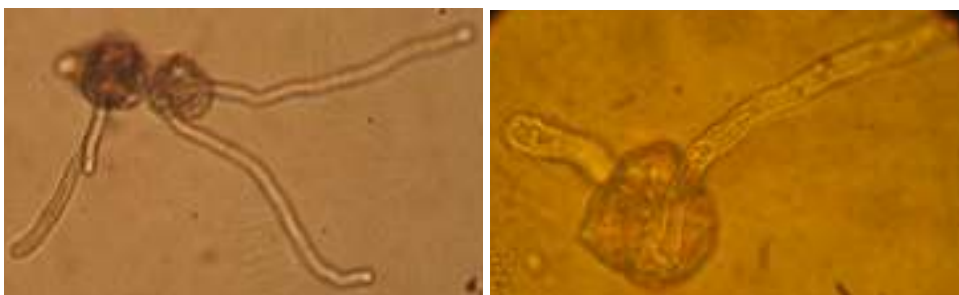
Regarding the pollen tube growth in length, should be noted that there is always a direct correlation between flower style length and pollen tube length. In this case, the styles of *R. racemosum* has an average length (obtained by measuring of 100 styles) of 4.75 mm. So, only the pollen tube of at least 4750  $\mu\text{m}$  length can reach to the embryo sac for to make the double fertilization. We estimate that this pollen tube length of *R. racemosum* shall be in natural conditions. Always, *in vitro* the pollen tube rate of growth is much slower (Erdman, 1952). According to Okonkwo. and Campbell (1990) the tubes of *R. racemosum* x *R. barbatum* and its reciprocal covered 100% of the style length in 96 hours, the same as of the intraspecific compatible cross *R. racemosum* x *R. racemosum*. According to Williams and Rouse. (1990), the pollen tube growth rates were generally greater for species of *Rhododendron* with longer pistils and larger pollen. The same correlation is accentuated by Fernández *et al.* (2009) showing that the relationship between pollen size and pistil length among species

suggests that the pollination system may be of less importance as a selective force than flower functionality.

The pollen germination process of *R. racemosum* was marked by anomalies that consisted in dilatations or ramifications at the top of the pollen tube (fig. 2) and in tetrads which 2-3 pollen tubes (fig. 2, 3). In this case the 2-3 tubes do not grow enough and so are not effective in fertilizing. These anomalies are not correlated with sucrose concentration in mediums.

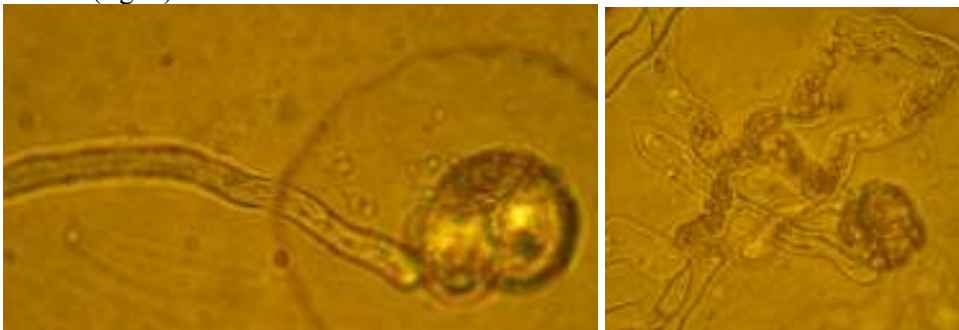


**Fig. 2** - Pollen tube with dilatations at tops (left) (100x); branched pollen tube at top (middle) (400x); tetrad with two pollen tubes (right) (100X) (Original)

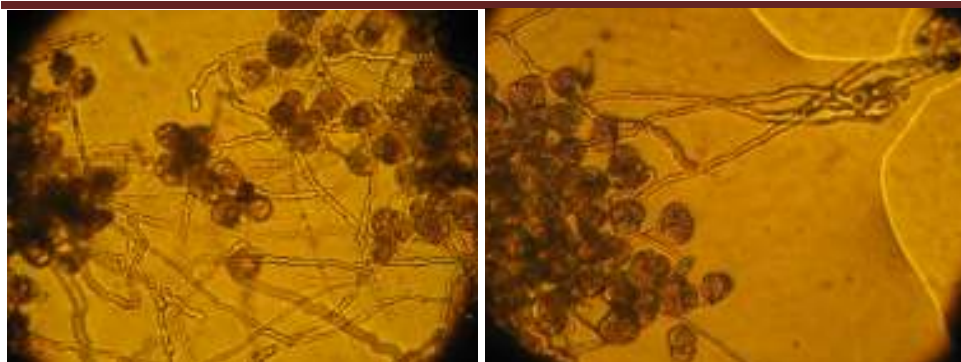


**Fig. 3** - Two tetrads with 3 and 2 pollen tubes (left) (400x); tetrad with two pollen tubes (1000x) (Original)

In 4 and 5 photomicrographs are presented aspects of the germination at *R. racemosum*. The germination process at this genotype not starts by forming a vesicle (fig. 4).



**Fig. 4** - The debut of edification of the pollen tube at *Rhododendron racemosum* (left) (1000x); pollen tube after 48 hours since inoculation on medium with 15% sucrose (right) (1000x) (Original)



**Fig. 5** - Pollen tubes formed after 24 hours (left) and after 96 hours (right) since inoculation on medium with 25% sucrose (400x) (Original)

## CONCLUSIONS

The longer pollen tubes more than 4000  $\mu\text{m}$  were developed after 8 days on medium with 25% sucrose. The increase in the length of pollen tubes is directly proportional to the ability of pollen germination. The increase in the length of pollen tubes is in close correlation with the length of floral style; in this case only long pollen tubes can cross over 4000  $\mu\text{m}$  stylar tissue. Extrapolating the *in vivo* situation, the pollen of *Rhododendron racemosum* germinates on a stigmatic liquid containing 25% carbohydrates, and pollen tubes cross the style in about 7 days to reach the ovules and fertilize them.

## REFERENCES

1. Erdman G., 1952 – *Pollen morphology and Plant taxonomy*. Angiosperms (An introduction to Palynology I). Almquist and Wiksele, Stockholm, pp. 53-71.
2. Fernández V.A., Galetto L., Astegiano J., 2009 - *Influence of flower functionality and pollination system on the pollen size-pistil length relationship*. *Organisms Diversity and Evolution*, vol. 9 (2), nr. 25, pp. 75-82
3. Kho Y.O., Baër J., 1970 - *A microscopical research on the incompatibility in the cross *Rhododendron impeditum* × *R. Williamsianum**. *Euphytica*, vol. 19, no. 3, pp. 303-309.
4. Krebs S.L., Leach D.G., 1997 - *A cytogenetic study of sterile *Rhododendron* hybrids*. *American Rhododendron Society Journal*, vol. 51, no. 2.
5. Okonkwo C.A.C., Campbell R.J., 1990 - *Pollen tube growth in pistils of *Rhododendron* following intra- and inter-specific pollinations*. *Genetika i Seleksiya*, vol. 23, no. 1, pp. 9-15.
6. Palser B.F., John L. Rouse J.L., Williams E.G., 1992 - *A scanning electron microscope study of the pollen tube pathway in pistils of *Rhododendron**. *Canadian Journal of Botany*, vol. 70, no. 5, pp. 1039-1060.
7. Williams E.G., Rouse J.L., 1990 - *Relationships of pollen size, pistil length and pollen tube growth rates in *Rhododendron* and their influence on hybridization*. *Sexual Plant Reproduction*, vol. 3, no. 1, pp. 7-17.
8. Williams E.G., Rouse J.L., Kaul V., Knox R.B., 1991 - *Reproductive timetable for the tropical *Vireya rhododendron*, *R. macgregoriae**. *Sexual Plant Reproduction*, vol. 4, no. 3, p.155-165.
9. Yan L.J., Gao L.M., Li D.Z., 2013 - *Molecular evidence for natural hybridization between *Rhododendron spiciferum* and *R. spinuliferum* (Ericaceae)*. *Journal of Systematics and Evolution*, vol. 51, no. 4, pp. 426-434.

## FEATURES OF ADAPTATION TO DROUGHT IN ISO AND ANISOHYDRIC PLANTS AND EFFECT OF SALICYLIC ACID

### PARTICULARITĂȚI DE ADAPTARE LA SECETĂ A PLANTELOR ISO ȘI ANISOHIDRICE ȘI EFECTUL ACIDULUI SALICILIC

**MELENCIUC M.<sup>1</sup>**

**email:** melenciuc79@mail.ru

**Abstract:** It has been studied the effect of salicylic acid derivatives of the cations  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{++}$  on the physiological processes of the isohydric plant of *Zea mays* L and anisohydric one of *Sorghum bicolor* L. Moench in different conditions of soil water content. It was established that the peculiarities of the status of the water, induced by drought, were associated with adaptive morphological characters: reducing surface evaporation by the lower leaf senescence (maize) and adventitious shoots (on sorghum), leaf rolling (maize), cutinization and exudation of silicon oxide (on sorghum). It was shown that salicylates participated in the induction mechanisms of drought tolerance of plants by modulating the absorption, hydraulic conductivity, stomatal resistance and transpiration intensity. Such adaptive changes are geared towards maximum efficiency of water use in the production process. Treatment of seeds and foliage of plants with relevant salicylates increased the organism's tolerance to subsequent bad weather, which in turn affected growth, productivity and plants crop.

**Key words:** isohydric and anisohydric plant, hydraulic conductivity, stomatal resistance, salicylates, growth, productivity, rezistance.

**Rezumat:** S-a studiat efectul acidului salicilic și a derivaților lui cu cationi de  $\text{NH}_4^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{++}$  asupra proceselor fiziologice a plantelor isohidrice de *Zea mays* L și anisohidrice de *Sorghum bicolor* L. Möench în diferite condiții de umiditate. S-a stabilit că, particularitățile de autoreglare a status-ului apei, induse de secetă, sunt asociate cu caractere morfologice adaptive: reducerea suprafeței de evaporare prin senescența frunzelor inferioare (la porumb) și lăstarilor adventivi (la sorg), frunze rulante (la porumb), cutinizare și exudatii de oxizi de siliciu (la sorg). S-a demonstrat că, salicilații participă în inducția mecanismelor de toleranță a plantelor la secetă prin modularea absorbției, conductibilității hidraulice, rezistenței stomatelor și intensității transpirației. Prin urmare modificările adaptive sunt orientate spre eficientizarea maximă a utilizării apei în procesul de producție. Tratarea semințelor înainte de semănat și a aparatului foliar al plantelor cu salicilați relevant majorează toleranța organismului la intemperii ulterioare, ceea ce se repercutează asupra creșterii, productivității și recoltei plantelor.

**Cuvinte-cheie:** plante isohidrice și anisohidrice, deficit de saturație, conductibilitate hidraulică, conductibilitatea stomatelor, salicilați, creștere, productivitate, rezistență.

---

<sup>1</sup> Institute of Genetics, Plant Physiology and Protection of Academy of Sciences, Chișinău, Republic of Moldova

## INTRODUCTION

Study of mechanisms of the formation adaptation and resistance to drought of plants, especially mesophytes, are one of the priority directions of contemporary biology. Production losses caused by unfavorable factors from the environment, entails the need to widen the investigations relating to the elucidation of the mechanisms underlying adaptation and tolerance of plants to environmental conditions, such as suboptimal and search for ways of minimising the negative impact. Property to maintain water plant in the organs under conditions of moderate moisture deficit is at the cellular level and tissue it is codependency of changing water retention capacity (WRC), intensity of osmotic water absorption, coupled with additional energy consumption (Gupta *et al.*, 2000; Jones, 2007). At the organism maintain fluid balance is conditioned by dimming uptake, translocation through plant transpiration and water consumption in the process (Chavies *et al.*, 2003). Proceeding from the fact that the regulation and coordination of growth, development and productivity of plants under the control of plant hormones, more research in phytophysiology the is oriented to elucidating mechanisms and explore ways of exogenous adjusting plant functional status in moderate drought conditions. Salicylic acid is referred to phytohormones with properties regulating the growth and development of plants ((Gudvin and Mercer, 1983), its effect is associated with the maintenance of the balance of the AIA, ABA and CK (Gupta *et al.*, 2000). It is supposed that AS participates in regulating hydraulic conductivity (Lp) plant (Moreschet *et al.*, 1990). In the literature few investigations are subordinate to elucidate the mechanisms of action of AS on components of the water status, growth, development and crop productivity in drought conditions.

## MATERIAL AND METHOD

As subjects served isohidric plants of *Zea mays* L. cultivar (cv.) P459, and anisohidric plants of *Sorghum bicolor* (L.) Möench, cv. Moldovenesc 40. The experiments were conducted in the greenhouse complex under controlled hydric regime of the IGFP with plants grown in containers Mitcerlih with 30 kg dry soil. The hydric stress was created by the gradual decline in soil water content range 70 - 60 - 50 to 40 - 30% of total water capacity (TWC) of the soil. Drought stress was modeled on the sixth stage - she organogenesis by reducing water supply plant and maintain the moisture level of 30% TWC. Parallel variants include plants treated with AS. The parameters of the water status in the segment "root-stalk" and "strain - leaves" have been determined by conventional methods (Vasseu and Sharkey, 1989). Hydraulic conductivity plant was determined by inhibiting water channels with  $\text{HgCl}_2$  (Zhongin and Newmann, 1999) and calculated according to the equation:  $F = L_p \cdot \Delta \Psi_w / \Delta x$ ; where F - transpiration rate, or volumetric flow of water;  $L_p$  hydraulic -conductibilitatea;  $\Delta x$  - distance between metamerele organ;  $\Psi_w$  - value of the water potential (Jones, 2007). The results were statistically analyzed using the software package "Statistica 7" for computers. Authenticity is true differences in the probability  $P \leq 0.05$  and  $P \leq 0.01$ .



## RESULTS AND DISCUSSIONS

Salicylic acid is referred to phytohormones with properties regulating the growth and development of plants (Gudvin and Mercer, 1983), its effect is associated with maintaining the balance of the AIA, ABA and CK (Gupta *et al.*, 2000). Pretreatment with salicylates seed conditioned acceleration of plant growth in the initial stages of ontogenesis (Tab. 1). Salicylates effect is especially evident in plants exposed "physiological drought". Drought physiological root system inhibited the growth of seedlings from seeds treated with 8,7 to 10,00%, while the root system of seedlings treated values did not differ significantly from those of plantlets "martor" unexposed stress. The diurnal rate of accumulation of biomass plantlets "martor" within 5 days after germination amounted to  $123,78 \text{ mg} \cdot \text{pl}^{-1}$ ; plantlets treated with AS –  $131,8 \text{ mg} \cdot \text{pl}^{-1}$ . Inaccessibility of to water plantlets from the background "physiological drought" has conditioned a reduction of 147.12 mg biomass accumulation in 24 hours from plantlets treated with 60,54 mg - in those treated (Tab. 1).

Table 1.

**Influence salicylates on the growth of the plantlets *Zea mays* L., cv. P459 the a background „ physiological drought ” at the initial stage of ontogenesis.**

Variant	Length, mm		Biomass, mg		
	Root	Coleoptile	Roots	Coleoptile	Plantlets
Control	132,1± 0,69	84,52±1,4	373,33±7,2	145,5±1,73	618,92±4,5
Salicylat	156,0± 0,85	98,85±0,4	381,00±5,2	277,9±2,37	658,94±3,8
Martor*Drought	121,56±0,58	77,69±1,2	282,63±3,4	189,1±5,29	471,80±4,3
Salicylat*Droug ht	132,06±1,67	86,83±0,3	358,05±6,2	200,3±4,87	558,36±5,5

As a result of the effect of SA and increase linearly radiclei salicylate was two times the mean length of the control plants radiclei. At the same time coleoptile length was 17,3 percent higher than coleoptile length seedlings from seeds treated with water. It follows that at the early stages of of ontogenesis AS favors the growth of the plantlets in general, but especially to increase the rate of growth of the root system. The dates obtained by us argue indirectly ideas raised by Raskin I. (Raskin, 1992) as the salicylates are able to maintain the balance between IBA, ABA and CK. (Ниловская *et al.*, 1997; Пустовойтова *et al.*, 2000). In all probability AS stimulates biosynthesis because following the data tab. 1 rate of biomass accumulation of plantlets treated with AS exceeded by 1,4 to 1,5 times control plantlets biomass accumulation. A consequence of differences in growth rate of plant organs at the initial stage of of ontogenesis was the change in mass ratio shoot / root mass ( $m_1 / m_r$ ). On the background of accelerating growth processes seedling entirely AS the initial stages of of ontogenesis accelerates especially root system formation. Effect distinctive of the AS on the growth of roots and shoots the correlation change allowed to assume that the seed priming with AS could serve as a modulator of growth, development and plant resistance

in wet conditions suboptimal. Just know that one of the causes drought tolerance of plants is to develop a strong root system, deeply penetrating and high absorption activity. Moreover, one of the main factors that determine the size road in conditions in the water supply, is the state of the roots. It was established that plants well supplied with water under the influence salicylate in connection majorarează  $L_p$  true "root - leaf" and maintaining its value to a significantly higher level in terms of inadequate water availability.

Table 2.

**Influence salicylates on the parameters water status plantlets *Zea mays* L., cv. P459 the a background „ physiological drought ” at the initial stage of ontogenesis.**

Variant	Water content , $g \cdot 100 g^{-1}$ f. m.		Intensity of transpiration, $g \cdot dm^{-2} \cdot h^{-1}$	$L_p$ mg.cm.h
	Root	Coleoptile		
Control	89,67± 0,53	65,37± 0,87	0,68±0,012	4,4375±0,07
Salicylat	90,05± 0,63	96,55± 0,72	0,78±0,009	4,7737±0,13
Martor*Drought	84,53± 0,47	56,67± 1,10	2,87±0,007	1,0249±0,04
Salicylat*Drought	85,65± 0,03	60,40± 0,75	0,89±0,004	2,9541±0,05

$L_p$  under optimum conditions is increased by 9,11% compared to the control. After 24 h of stress ("physiological drought")  $L_p$  to the latter decreased 4,9 times, while the plantlets from seeds treated with salicylates - only 1,4 times (tab. 2) and beyond  $L_p$  plantlets the treated seed 3,44 times. Arguments about the peculiarities of homeostatare the status of the water by *Zea mays* L. and plants *Sorghum bicolor* (L.) Moench were obtained in the experiences and vegetation moisture controlled fond (Table 3).

Table 3

**Influence salicylates on the parameters water status plantlets *Zea mays* L., cv. P459 and *Sorghum bicolor* (L.) Möench cv. Moldovenesc 40 in conditions of insufficiency humidity.**

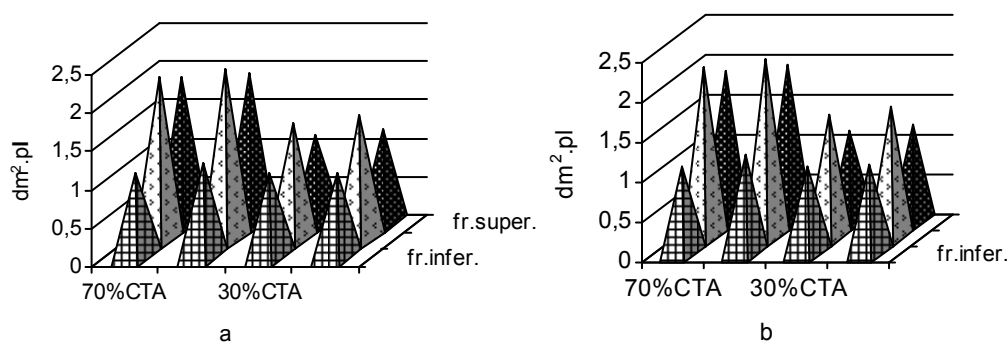
Cultivar	Varianta	Saturation deficiency, % of full saturation	Stomatal conductance, $g \cdot dm^{-2}$	Intensity of transpiration, $mg \cdot dm^{-2} \cdot h^{-1}$	$L_p$ , mg.cm.h
P459	Martor	1,42±0,04	44,15 ± 0,86	508,7 ± 14,9	0,475±0,012
		18,70±0,23	16,34 ± 0,68	201,7 ± 8,45	0,235±0,007
	Salmet	1,32±0,03	47,54 ± 0,81	535,2 ± 15,6	0,56±0,009
		14,01±0,25	24,17 ± 0,71	225,6 ± 10,7	0,275±0,004
Moldove nesc 40	Martor	0,95 ± 0,04	59,20 ± 1,13	844,85 ± 18,4	0,855±0,011
		2,72 ± 0,07	47,42 ± 1,12	676,77 ± 16,3	0,55±0,008
	Salmet	0,76 ± 0,02	63,85 ± 1,83	822,74 ± 9,5	0,91±0,006
		1,71 ± 0,05	53,00 ± 1,49	719,36 ± 12,9	0,64±0,010

The stabilizing effect of the status of the water produced by the compounds is due to their property AS control hydraulic conductivity and the state of the stomata. Salicylates increase the capacity of the attraction of the water, in particular to the leaves from the top of the plant, inducing redistribution of water bodies. Responding to drought plant sorghum is characterized not so much by reducing water consumption in the process of sweating, as the property



to obtain water due to the increase surface root system and force absorbing water from the soil and preserving conductivity hydraulic plant. Increasing intensity of water delivery stomata allow the plant to keep ajar for longer, facilitating the assimilation of  $\text{CO}_2$  and biomass accumulation. Therefore, the drought conditioned prompt changes in the intensity of both the water of perspiration, coupled with the change in conductance of stomata and the potential value of the hydrostatic pressure of the water and foliage of plants. The reduction of the water flow to the leaves (IT) are associated with reduced hydraulic conductivity of the tissues - most significant in sensitive plants as opposed to the typical anisohidrice plants tolerant to a long-lasting drought. The stabilizing effect of the status of the water produced by the compounds of salicylic acid is a consequence of the maintenance of the hydraulic conductivity and the state of the stomates.

Identifying differences of *Zea mays* plant morphophysiological L., cv. P459 and *Sorghum bicolor* (L.) Moench, cv. Pișcevoi 1 (Fig. 1) under controlled humidity, revealed the reaction of different plant maize and sorghum to water



**Fig. 1** - The influence of the salicylic acid on formation of the leaf area to different levels insertion,  $\text{dm}^2$  plants *Sorghum bicolor*, L. Moench cv. Pișcevoi (a) and *Zea mays* L. cv. P459 (b) under varying humidity.

in the soil and changes in compensatory induced AS-level body, achieved by maintaining growth, the accumulation of biomass, training area, assimilation, etc. Drought leaves its mark on training and assimilation machine by reducing the leaf area metamerelor lower leaves, as a result of drying them, and through halting the growth of leaves and higher levels of employability. Already after 5 days of drought (30% TWC) total leaf area was reduced to *Zea mays* L. plants, cv P459 by 33,0% and those of *Sorghum bicolor* (L.) Moench, cv. Pișcevoi 1-19% compared with control plants exposed to water stress us. The use of AS reduced the negative effect of drought by 6,2 and 5,1 percent respectively maize and sorghum plants. Note that the leaf surface and higher levels decreased under severe moisture 28,1 and 39,4% compared to control plants of sorghum and 30,9 and 40,0% - in the maize. The latter, along with the trend growth inhibition upper leaves, there has been a genuine reduction in lower leaves most significant surface (Fig. 1). A distinctive feature of sorghum plant growth type indeterminate reduce surface evaporation is not so much the main shoot leaves account as partial drying

as a result of adventitious shoots of leaves. AS effect was manifested by mitigating the impact of drought, especially on the leaf area at the bottom and the environment. Therefore, plants treated with foliar AS seminal and easier exceed ancestry stressogenic factors. Its distinctive effect is achieved through better training device assimilate what biomass accumulation undoubtedly reflects on the production process.

## CONCLUSIONS

1. Property homeostatare of the water status in the plant induced AS is achieved by enhancing / amplification water uptake by the root system, hydraulic conductivity increases, the degree of hydration of tissues, decrease the saturation deficit and increasing turgidity organs.

2. Salicylates conditioned activation of vital processes already at the initial stages of individual development of plant root and shoot growth stimulation system, the formation of more vigorous plants in hydric suboptimal conditions, lessening the impact on productivity.

3. The phenotype of the plant, comprising the administration of AS is distinguished by improved tolerance to weather during the growing season.

*Acknowledgement:* Investigations were performed with the financial support of young researchers project 15.819.05.08F.

## REFERENCES

1. Chavies M. M., Maroco J. P., Pereira J. S. 2003 - *Understating plant response to drought: from genes to the whole plant*. In: Functional Plant Biology, 30: 239 – 264.
2. Gudvin T. W., Mercer E. I. 1983 - *Introduction to Biochemistry. Second Edition*. Oxford. New York. Toronto. Sydney. Paris. Frankfurt: Pegamon Press. pp. 123 – 126.
3. Gupta N. K., Gupta S., Kumar A. 2000 - *Exogenous cytokinin application increases cell membrane and chlorophyll stability in wheat (Triticum aestivum L.)*. In: Cereal Research Communication, 28(3): 287-291.
4. Jones H. G. 2007 - *Monitoring plant and soil water status: established and novel methods revisited and their relevance to studies of drought tolerance*. In: Journal of Experimental Botany, 58: 119 – 130.
5. Raskin I. 1992 - *Role of Salicylic Acid in Plants*. In: Annu. Rev. Plant Physiol. Plant Mol. Biol., 43: 439-463.
6. Vasseu T.L., Sharkey T.D., 1989 - *Mild water stress of Phaseolus vulgaris plants leads to reduced starch synthesis and extractable sucrose phosphate synthase activity*. In: Plant Physiol. V. 89. P. 1066-1070.
7. Zhongin Lu., Newmann P. 1999. - *Water stress inhibits Hydraulic Conductance and Leaf Growth in Pice Seedlings but Not the Transport of Water via Mercury Sensitive Water Channels in the Root*. In: Plant Physiology, 120(1): 143-151.
8. Ниловская Н., Осипова Л., Морозова Э. 1997 - *Влияние кинетина на жизнедеятельность апексов яровой пшеницы при действии водного стресса*. В: сб. Регуляторы роста и развития растений. Москва: Наука, С. 220.
9. Пустовойтова Т.Н., Баврина Т.В., Жданова Н.Е. 2000, *Особенности засухоустойчивости трансгенных растений табака с генами iaaM и iaaH биосинтеза ауксина*. В: Физиология растений. Т.47, №3, С.431-436.

## EFFECT OF SALT STRESS ON SEED GERMINATION AND SEEDLINGS CHLOROPHYLL CONTENT OF SOME TOMATOES (*LYCOPERSICON ESCULENTUM* MILL.) LOCAL LANDRACES

### EFFECTUL STRESULUI SALIN ASUPRA GERMINAȚIEI SEMINTELOR ȘI A CONȚINUTULUI DE CLOROFILĂ A PLANTULELOR LA UNELE POPULAȚII LOCALE DE TOMATE (*LYCOPERSICON ESCULENTUM* MILL.)

MARTA Alina Elena<sup>1</sup>, JIȚĂREANU Carmenica Doina<sup>1</sup>, SLABU Cristina<sup>1</sup>  
e-mail: alinamarta\_fiziologie@yahoo.com

**Abstract.** Soil salinity is one of the most important abiotic stress factors affecting the global food security. The impact of salt-affected soils on plant productivity is sometimes disastrously. This requires the identifying and creating new plants genotypes tolerant to osmotic stress. These activities are difficult because the physiological, biochemical and molecular mechanisms responsible for the growth reduction of crops is not completely understood. In the effort to increase plant resistance to salinity, special attention should be given to local landraces as important genetic resources. The aims of the present study were to contribute to a better understanding of the physiological mechanisms involved in salt stress tolerance of *Lycopersicon esculentum* Mill. plants, especially during seed germination and seedlings growth and to identify the local landraces tolerant of salts excess. The results show that all from the tested cultivars represent a valuable germplasm source useful for improvement of the salinity resistance in the tomato plants.

**Key words:** salinity, tolerance, *Lycopersicon esculentum* Mill.

**Rezumat.** Salinitatea solurilor este unul dintre cei mai importanți factori de stres abiotic care afectează securitatea alimentară a omenirii. Impactul solurilor cu exces salin asupra productivității diferitelor culturi este uneori dezastruos, fapt care determină identificarea și crearea de noi genotipuri de plante tolerante la stresul osmotic. Aceste activități sunt dificile deoarece mecanismele fiziologice, biochimice și moleculare răspunzătoare de reducerea creșterii nu sunt pe deplin elucidate. În efortul de a crește rezistența plantelor la salinitate, o atenție deosebită ar trebui acordată populațiilor locale ca importante resurse genetice. Obiectivele cercetărilor au fost acelea de a contribui la o mai bună înțelegere a mecanismelor fiziologice implicate în toleranța la stres salin a speciei *Lycopersicum esculentum* Mill., în special în timpul proceselor de germinare și creștere a plantulelor și de identificare a populațiilor locale tolerante la excesul de săruri. Rezultatele demonstrează că toate cultivarele testate reprezintă surse valoroase de germoplasmă pentru ameliorarea rezistenței tomatelor la stres salin.

**Cuvinte cheie:** salinitate, toleranță, *Lycopersicon esculentum* Mill.

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

## INTRODUCTION

Plant response to salinity has been much studied, since a good knowledge of the effect it has on plants to NaCl, it is a critical factor of cultivation in saline areas. The salinity may inhibit plant growth by a low water absorption potential from the outside, by toxicity and by reducing the uptake of nutrient ions, in particular potassium.

During the development, the tomato seeds are the first to face with the stress conditions, particularly affect the salinity of the active growth of the embryo. Throughout the growing season, the salinity affects various physiological and biochemical processes, the main effect being in the hydric relations, but is also manifested by ion toxicity (Jităreanu *et al.*, 2014).

Following investigations, Dayme Camejo and W. Torres (2000) suggested that in this stage of development, the germination, is strongly influenced by the environment salinity. Thus the saline concentrations may change in time the moment of maximum speed of the germination process, due to a low power seed to absorb water and / or a toxic effect of the ions, that cause problems in some of the enzymes and the hormones of the seed, the consequences can be seen by delay or inhibit the germination process.

On the other hand, high concentrations of NaCl causes the physico-chemical changes in the seeds, with the effect of delay or reduction in germination capacity value. The effect of external environment salinity on seed germination may be partially osmotic or toxic ionic can affect physiological processes and enzymatic activities (Jogendra Singh *et al.*, 2012).

Chlorophyll, one of the main components of the chloroplast involved in photosynthesis and chlorophyll content is correlated positively with the degree of photosynthesis. Water deficit and water transport inability to the leaf, can decline the photosynthesis (Marta *et al.*, 2014).

The total chlorophyll concentration is significantly reduced in tomato leaves under salt stress. This is a common phenomenon reported in many researches, due to its adverse effects on membrane stability, and a low concentrations of photosynthetic pigments can cause a direct reduction in production (Marta *et al.*, 2014).

## MATERIAL AND METHOD

For testing we used local tomato populations collected in households of Iasi and Botosani. The populations analyzed were: Moșna 2, Moșna 3, Șcheia 3, Copalau, 3, 4 and 5, Ursula F1 and Jebel.

Seed germination under stress was tested to saline 75 mM NaCl concentrations, having a control salinity resistant population Jebel and Ursula F1, a resistant hybrid, created in Israel. The testing was made in growth chamber at 22°C and 80% humidity. The comments were recorded for 7 days from the establishment of experiences.

After the completion of germination and growth of plants from the cotyledon leaves were extracted and studied the chlorophyll pigments by the spectrophotometric method.

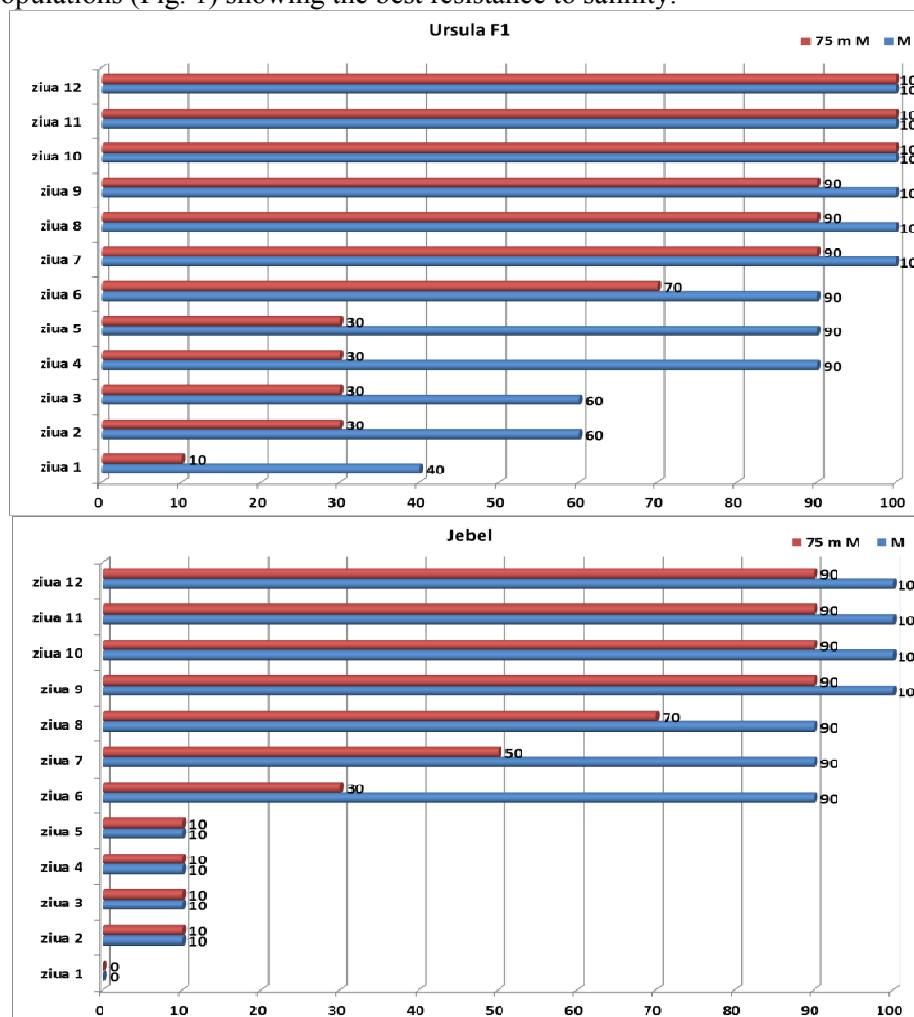
## RESULTS AND DISCUSSION

### Analysis of tomato seed germination capacity in saline

Among the physiological traits, state standards require only suitable for germination, showing the number as a percentage of seeds capable of producing germs during normal laboratory conditions of temperature and humidity and light set as optimal for each species.

The comments were recorded for 7 days from the establishment of experiences and germination was analyzed for 12 days.

Analyzing all the tomato populations, a behavior close to control Ursula F1 and Jebel observed the local population Copalau 3, the germination at the variant treated with saline began in the 2nd day of observations compared to other local populations (Fig. 1) showing the best resistance to salinity.



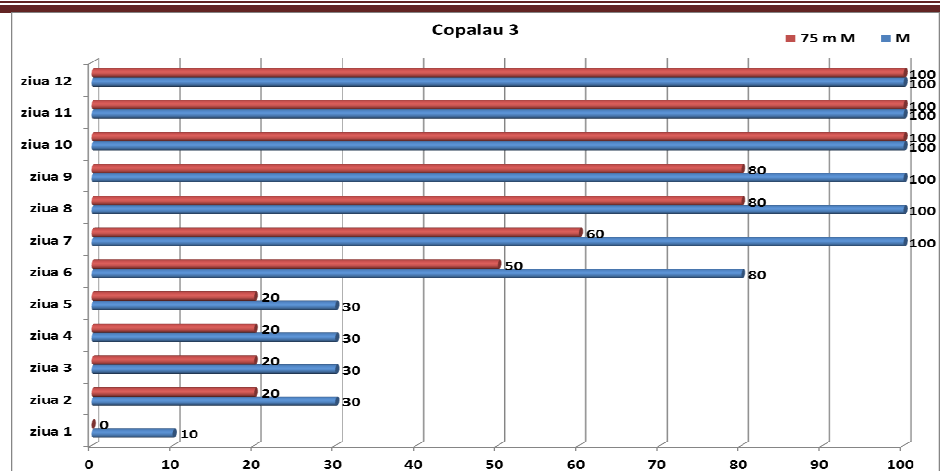
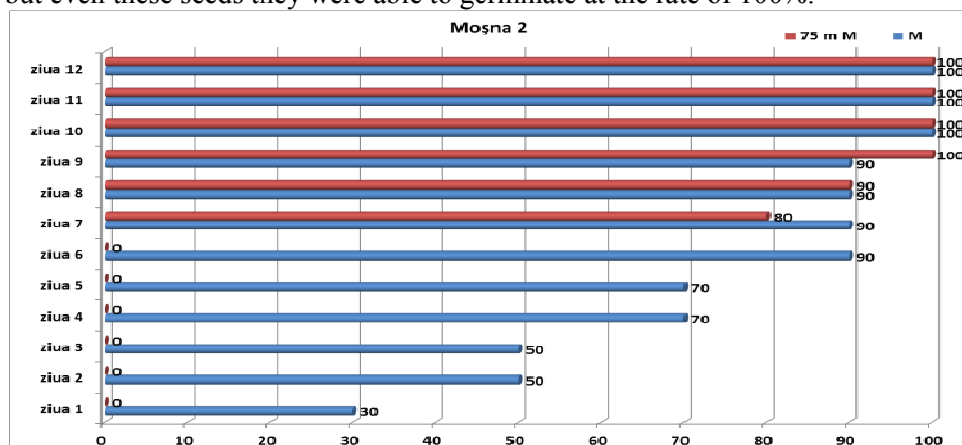


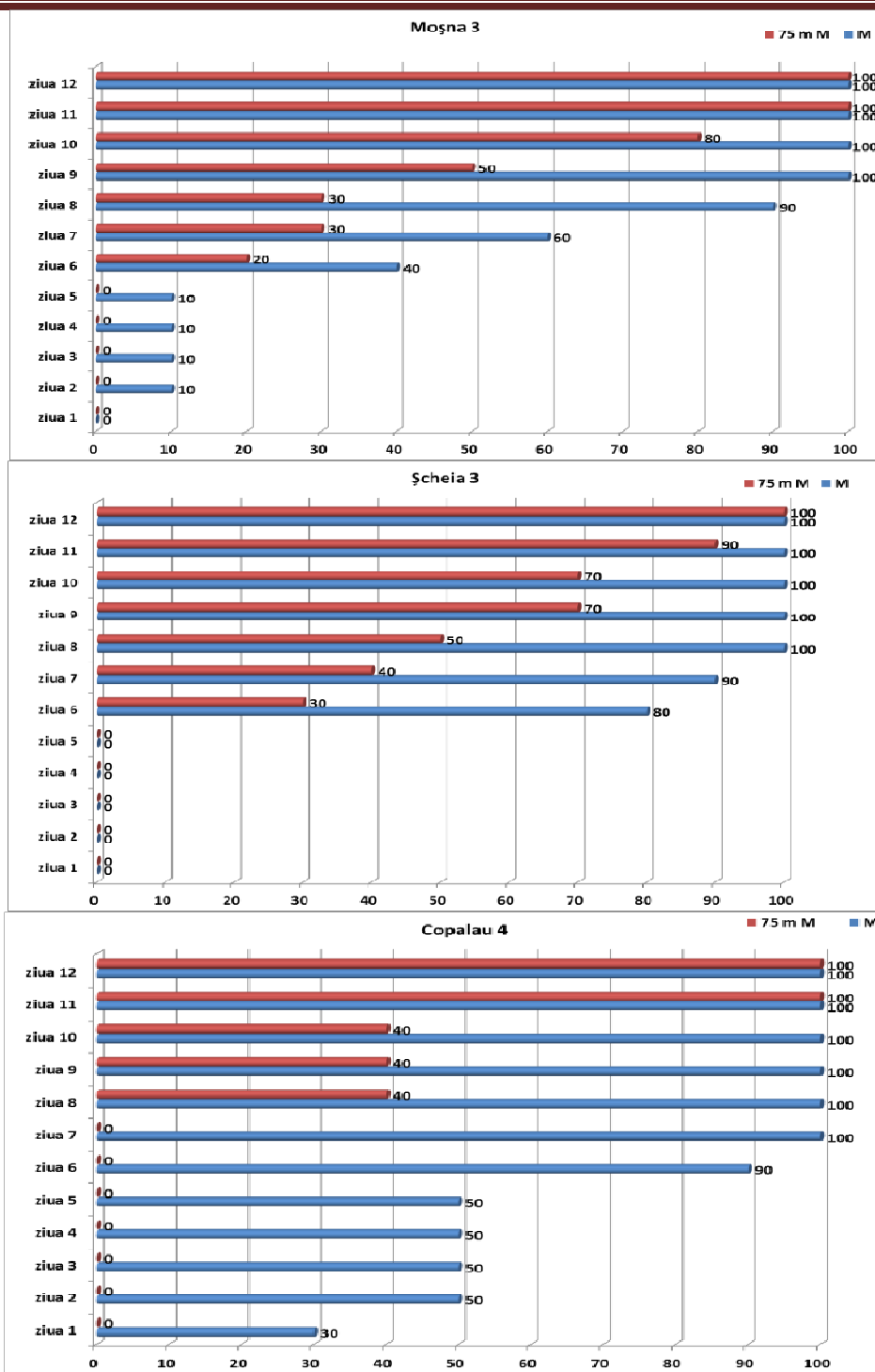
Fig. 1 - Germination of Copalau 3 population, compared to controls resistant to salinity

In most tomato populations concentration 75 mM NaCl had resulted a delay of 5-6 days in the germination capacity, however, finally reaching a maximum of 100% germinabilitate (Fig. 1 and 2).

At populations Moșna 2 and 3 and Copal 4 the treatments with saline solutions have resulted a delay germination, while at Șcheia 3 was a slight inhibition, and at Copălău 5 the germination was affected by saline treatments, resulting in the end only 70% germinated seeds (Fig. 2).

We recommend the tests carried out Copălău 3 population as the most resilient, followed by Moșna 2 and 3 and Copalau 4 that could be kept and used as tomato germplasm to improve resistance to salt stress. In the case of plants, the salt stress adaptation strategies are undergo biochemical, physiological and molecular processes. Even if were observed negative effects of salinity on seed germination, most obvious differences were observed in populations Copălău Moșna 2 and 4 with a delay of 7-8 days compared to other populations analyzed, but even these seeds they were able to germinate at the rate of 100%.





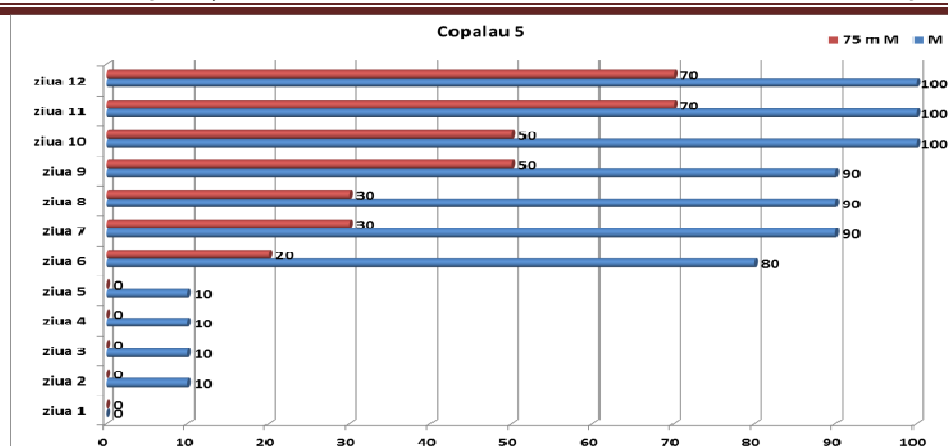


Fig. 2- Inhibition of germination capacity by treatment with saline in the first 5-7 days of starting observations

### Analysis of chlorophyll pigments in cotyledon leaves of tomato in spectrophotometer

By analyzing in spectrometer it was determined the chlorophyll pigments in cotyledon leaf from seedlings grown from seed germination testing in saline.

Of the types of chlorophyll pigments analyzed, the highest values were recorded by **431-432 nm** chlorophyll, the main component of the absorption center in photosynthetic systems. This type of chlorophyll was determined to 6 local populations of tomato and two controls, a population (Jebel) and a hybrid (F1 Ursula) both resistant to salinity variations with 75 m M NaCl treated and controls with distilled water.

From figure 3 it notes three types of behavior: at control Jebel nearly equal values.882 absorbent). At Ursula F1, Moșna 2 Moșna 3; scaffolding three values are observed maximum absorbance at variants control (1113 to Moșna 3, 1082 at Moșna 2, 1051 at Șcheia 3 and 0.808 Ursula F1) and lower in those treated with saline (are observed both in control and treated with 75 mM solution in saline (0.881 resp. Moșna 0.943 to 3, 0.810 to Moșna 2, 3 and 0.778 to 0.681 Ursula Șcheia F1), saline treatment with inhibiting effect on the photosynthetic capacity of these populations.

At variants of Copalau (Copălău 3 Copălău 4 Copălău 5) there is a completely different reaction: maximum values in those treated with saline (1,529 to Copal 4, 1041 at Copal 5 and 0.918 at Copal 3) and lower in controls (1.011 at copal 5; 0.953 to 0.686 in copal copal 4 and 3), being stimulated photosynthetic capacity of these populations as a response to salt stress.

The high values obtained in Copalau recommends variants to be the most resistant to saline (Fig. 3).



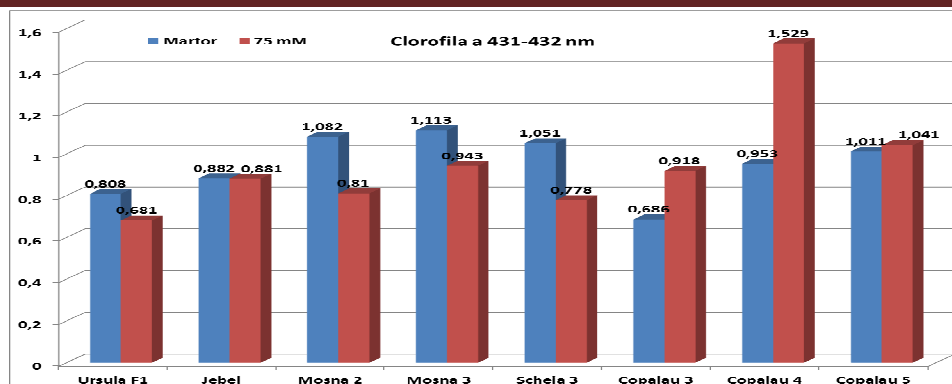


Fig. 3 - 431-432 nm chlorophyll content in tomato cotyledon leaf

Chlorophyll content of 662-663 nm, the main component of the photosynthetic reaction center systems showed two different behaviors: Ursula F1, Moșna 2, 3 and Șcheia3 notes higher values in controls (0.593 to Moșna 2; 0.568 Șcheia3 to Moșna 3 and 3; 0.400 to Ursula F1), the chlorophyll content of cotyledonar leaves being subjected to salt stress inhibited variants (fig. 4).

Another type of behavior is observed in populations from Copalau, with maximum in variants treated with saline (0.831 at Copalau 4, 0.578 at Copalau 5 and 0.496 at Copalau 3) and much lower in controls (0.497 at Copalau 4, 0.530 to Copalau 5 and 0.353 in Copalau 3), the photosynthetic activity of variants subject to salt stress was highest in this case being registered a stimulating effect on the chlorophyll content of 662-663 (Fig. 4).

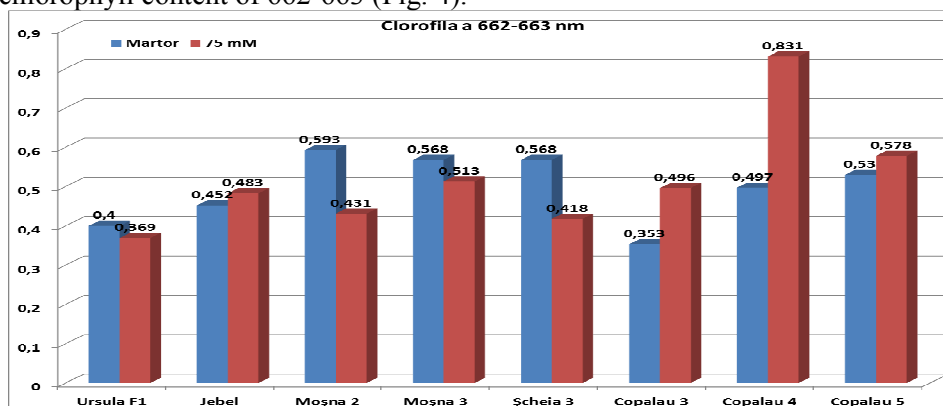


Fig. 4 - 662-663 nm chlorophyll content in tomato cotyledon leaf

## CONCLUSIONS

1. In terms of germination capacity the most resistant local population can be considered Copălău 3 while populations Moșna 2, 3 and 4 Copălău saline treatments have resulted in a delay of 5, 6 days of germination.

2. The tomato plants were reacted to salt stress according to the biphasic model proposed by Mann, with the first phase, osmotic, characterized by reducing the growth and chlorophyll content of leaf and the manifestation of a phase of toxicity caused by excess sodium ions, and chlorine, characterized by the appearance of foliar chlorosis and necrosis.

3. Of the types of chlorophyll pigments analyzed in cotyledon leaves, the highest values were recorded by 431-432 nm chlorophyll, the main component of the absorption center of photosynthetic systems, and chlorophyll 662-663 nm. At variants of Copalau (Copălău 3, 4 copal, copal 5) it was observing a particular behavior: maximum values at variants treated with saline and lower in control, plantlets in salt stress response is increased photosynthetic activity.

### RECOMMENDATIONS

► The analyzes and observations show that some populations of tomato analyzed react to salt stress after biphasic pattern proposed by Manns, so we recommend Copălău 3 as the most resilient, followed by Moșna 2 and 3 and Copălău 4 could be kept and used as germplasm.

► The high values of germination capacity and chlorophyll pigments content obtained in variants from Copalau, recommends as the most resistant to saline solutions, given that in this area there are about 100 ha of soils strongly and moderately saline

***Acknowledgements:** 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.*

### REFERENCES

1. **Camejo D., Torres W., 2000** - *La salinidad y su efecto en los estadios iniciales del desarrollo de dos cultivares de tomate (Lycopersicon esculentum, Mill)*. Cultivos Tropicales, 21 (2): 23-26.
2. **Jităreanu Carmenica Doina, Slabu Cristina, Marta Alina Elena, Bologa (Covașă) Mihaela, 2014** - *Dynamics of the flavonoids content in some tomato cultivars from Nord - East Romania*. Lucrări științifice USAMV Iași, seria Horticultură, Iași, 57(1): 57 – 62.
3. **Marta Alina Elena, Jităreanu Carmenica Doina, Slabu Cristina, 2014** - *Chlorophyll content index of some NE-Romania Phaseolus vulgaris L. local cultivars, under salt stress*. Lucrări științifice USAMV Iasi, seria Horticultură, Iași, 57(1): 63-68.
4. **Singh Jogendra, Divakar E.V., Singh Vijayata, 2012** - *Effect of salinity on tomato (Lycopersicon esculentum Mill) during seed germination stage*. Physiol. Mol. Biol. Plants (January-March), 18 (1): 45-50.

## THE INFLUENCE OF ARBUSCULAR MYCORRHIZAL FUNGI ON TOLERANCE TO SALINE STRESS OF TOMATOES

### INFLUENȚA FUNGILOR ARBUSCULAR MICORIZALI ASUPRA TOLERANȚEI LA STRESUL SALIN LA TOMATE

**SCHMIDT Brigitta<sup>1</sup>, ȘUMĂLAN R. <sup>1</sup>, ȘUMĂLAN Renata<sup>1</sup>,  
CĂLUȘERU Alina Lavinia<sup>1</sup>, SAMFIRA I. <sup>1</sup>**  
e-mail: brigischmidt@yahoo.com

**Abstract.** *A Romanian tomato landrace was tested for tolerance to saline stress in pot culture. To improve the tolerance of plants to osmotic stress, we tested the effect of inoculation with arbuscular mycorrhizal fungi. Plants were assessed before and after application of saline solution for chlorophyll and dry matter content. To test the level of tolerance to salinity, we extracted the proline from plants. Results show that arbuscular mycorrhiza determined a significant reduction of chlorophyll content in normal growing conditions, but after the stress was induced, the differences between mycorrhized and non-mycorrhized plants reduced and differences were not statistically different. Dry matter content was not significantly influenced by any factor. After application of NaCl solution, non-mycorrhized plants had the highest proline content, meanwhile the mycorrhiza alleviated the level of osmotic stress.*

**Key words:** tomato, saline stress, tolerance, proline

**Rezumat.** *O populație locală de tomate din România a fost testată la toleranța la stresul salin. Pentru a îmbunătăți toleranța la stresul osmotic la plante, am testat efectul inoculării cu fungi arbuscular micorizali. Plantele au fost evaluate înainte și după aplicarea soluției salină, măsurând conținutul de clorofilă și de substanță uscată ale acestora. Pentru a testa nivelul toleranței la salinitate, a fost extrasă și cuantificată prolina din plante. Rezultatele arată că micoriza a determinat o reducere semnificativă a conținutului de clorofilă în condiții normale de creștere, însă după aplicarea stresului, diferențele între plantele micorizate și nemicorizate a scăzut, acestea fiind nesemnificative. Conținutul de substanță uscată din frunze nu a fost influențat semnificativ de niciunul dintre factorii experimentali testați. Conținutul de prolină a crescut datorită aplicării stresului salin. După aplicarea stresului plantele nemicorizate au prezentat cel mai ridicat conținut de prolină, micorizarea ducând la o reducere ale efectelor salinității.*

**Cuvinte cheie:** tomate, stres salin, toleranță, prolină

## INTRODUCTION

Agricultural production is facing several obstacles due to the influences of

---

<sup>1</sup> Banat's University of Agricultural Sciences and Veterinary Medicine "King Michael I<sup>st</sup> of Romania" from Timișoara, Romania

biotic and abiotic stress factors. Drought, floods, lack of nutrients, various pests and diseases can compromise yield and thus conduct to bankruptcy of farmers or even to famine in parts of Earth where food is insufficient. Our century started with frightening statistics: the summers are drier and hotter than ever, extreme temperatures are more frequent, the average global temperatures are increasing and the polar ice already started to melt. The human population is alarmingly increasing and agriculture is constantly looking for ways to assure the massive quantities of food necessary for almost 7 billion people. Intensive agriculture produced soil erosion, salinization of agricultural areas and desertification. Salinization of soils is not common only in coastal areas. Due to intensive agriculture, especially because of irrigation of agricultural lands, a high amount of salts accumulated in the soil. Worldwide, 800 millions of hectares of land are affected by soil salinization, meanwhile in EU 1 million hectares are affected (FAO, 2008).

Most of cultivated species, including tomatoes (*Lycopersicon esculentum* L.) are sensitive to saline stress (Agong *et al.*, 2003; Flowers and Yeo, 1995).

High concentration of salts in soil causes osmotic stress to plants. More,  $\text{Na}^+$  accumulates in shoots (Pardo and Quintero, 2002) and produces metabolic toxicity by disturbing cytoplasmic enzymatic processes.

Plants use a series of different mechanisms to alleviate the negative effect of osmotic stress: inhibition of growing in above-ground organs, with the growing of radicular system (Creelman *et al.*, 1990), osmotic regulation, modifications of metabolic flux, lignifications of cell walls reduction of growing rate, modifications of biomass phenology, leaf senescence, and finally, plant death (Munns, 2005).

It was also discovered that growth-promoting bacteria confer salt tolerance to plants, including tomatoes too (Mayak *et al.*, 2004). Cortés-Jiménez *et al.* (2014) suggested that microorganisms associated to tomato seedlings growing in saline culture act as osmoprotectant.

Once discovered, scientists are constantly looking for ways to fight the effects of salinity on cultivated plants. Developing tolerant cultivars is a promising but difficult solution, as genes for tolerance to osmotic stress are not naturally present in most of the cultivated species and most of the old cultivars, tolerant to many stress factors, are already forgotten and lost because modern hybrids which are now available on the market proved to be much more productive.

A new approach in agriculture is sustainability through biological solutions. More and more scientists are searching for symbiotic microorganisms which could play a role in protecting their host plants against the adverse conditions of the environment. As naturally plants and edaphic microorganisms evolved together, it proved that these microbes have a more important role in the physiology of plants that we presumed in the past.

Our study is only a step of an extensive research program that aims to collect, assess and preserve local landraces of vegetables in order to discover salt tolerant genotypes.

On the other hand, we aim to isolate salinity tolerant microbes from naturally saline areas and assess them for improving saline tolerance of cultivated plants when inoculated with these microbes.

In the present study we assessed the effect of inoculation with arbuscular mycorrhizal (AM) fungi on salt stress tolerance of a tomato landrace, simulating the cultivation conditions of normal farms. In most studies substrate is sterilized in order to show the effect of AM fungi on the physiology of host plants, but in farms real production conditions are different, as soil and peat are used in natural form, only sometimes fungicides are used in order to control the soil-borne diseases. Thus in this experiment we wanted to simulate farm conditions and to highlight the effects of AM fungi in “out of laboratory” conditions.

## MATERIAL AND METHOD

In the experiment we utilized tomato seeds from local landrace Tarnova 673, selected from our germplasm collection due to its characteristic large fruits. The name of the landrace derives from the village where it originates from.

20 seeds of each landrace were surface sterilized with sodium hypochlorite and germinated in Petri dishes on paper disks impregnated with distilled water. After germination, plantlets were transferred into 5 L pots with 2:1 peat:sandy soil mixture and half of the plants were inoculated with arbuscular mycorrhizal fungi (INOQ, Germany).

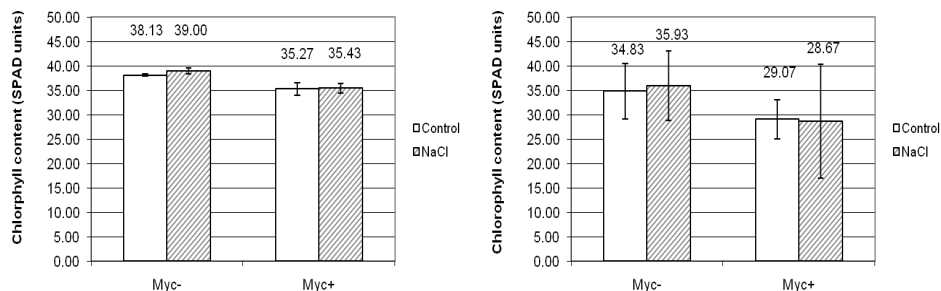
Plants were grown until the beginning of generative phenophase then watered with 100mM NaCl solution for 4 weeks. Before and after the saline stress the following parameters were measured: dry matter content of leaves using a KERN electronic moisture analyzer, chlorophyll content of leaves with a SPAD-502 chlorophyll meter (KONICA MINOLTA) and proline content of plants.

## RESULTS AND DISCUSSIONS

It was observed that before saline stress commenced, the mycorrhization produced a considering reduction of chlorophyll content of leaves. Non-mycorrhized plants showed values around 38-39 SPAD units, meanwhile inoculated plants had around 35 SPAD units, representing only 91.65% of the values of non-mycorrhized plants (Fig. 1). Also standard deviation shows that values are uniform, meaning that plants are quite homogenous in means of chlorophyll content of their leaves. A slightly higher diversity of values were recorded in case of mycorrhized plants, meaning that individual plants reacted differently to inoculation and symbiosis had various effects on their physiological processes probably due to the success of infection, the response of the plant to infection and the potential of fungi to behave parasitic (Johnson *et al.*, 1997).

Measurements after the application of stress factor showed that differences became more pronounced after flowering. Even control plants, which did not

suffer by osmotic stress, had generally lower values but with a higher variation between individual measurements. The mean chlorophyll content was higher at non-inoculated plants (34.83 SPAD units at control plants and 35.93 SPAD units at plants watered with NaCl solution) than at mycorrhized plants (29.07 SPAD units at control plants and 28.67 SPAD units at stressed variant). In each case, after flowering the chlorophyll content of leaves decreased compared to plants in vegetative stage.



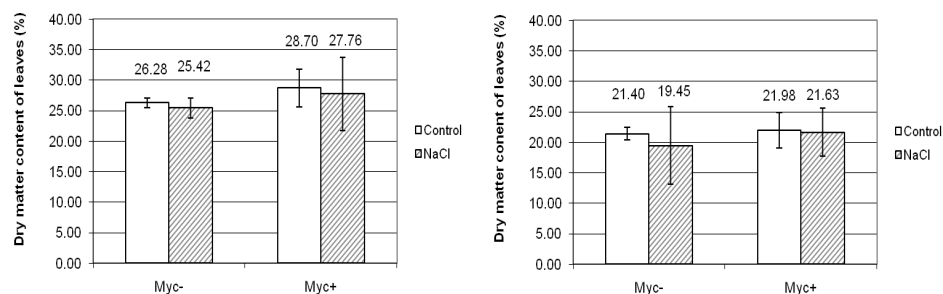
**Fig. 1** - Chlorophyll content of leaves before (left) and after (right) saline stress.

Dry matter content of leaves was slightly lower at non-mycorrhized plants before the application of saline stress. I

f compared to chlorophyll content, values show an opposite behaviour. Thus dry matter content is also higher at mycorrhized plants in vegetative stage (an average of 25.85% at non-inoculated plants and 28.23% at mycorrhized plants), meanwhile in generative stage, after stress application, the mycorrhized plants still show slightly higher values (non-mycorrhized plants: 21.40% at control plants and 19.45% at NaCl variant; mycorrhized plants: 21.98% at control plants and 21.63% at NaCl variant) (fig. 2).

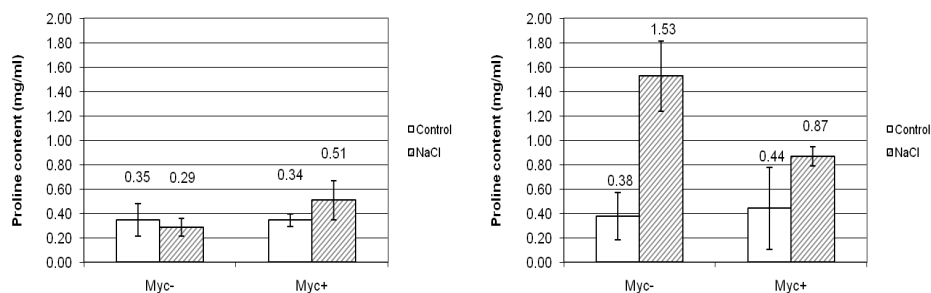
As in case of chlorophyll content, in vegetative stage, standard deviation of values recorded for mycorrhized plants were higher, but in case of dry matter content, these deviations were comparable to those in generative stage, meaning they were very high.

If in vegetative stage values for mycorrhized plants were higher, in vegetative stage the mean values are very close, only the non-mycorrhized and NaCl variants had a lower mean value for dry matter content, but also with very high variation of individual values.



**Fig. 2** - Dry matter content of leaves before (left) and after (right) saline stress.

Proline content of leaves indicates the level of a stress factor on plant physiology and also the level of plant response to stress (fig. 3). In case of non-mycorrhized plants, the values varied quite as expected: control plants did not increase the level of proline content meanwhile the application of NaCl in high concentrations for a certain time determined a drastical increase of proline synthesis in plant tissues. In our experiment saline stress determined a 4 fold increase of proline content in leaves of non-inoculated plants. In contrast, an interesting effect of mycorrhization was that in leaves the concentration of proline was significantly lower than in non-mycorrhized plants after salines stress. This proves that mycorrhization alleviates the negative effects of saline stress on plant metabolism by supplemental pathways compared to non-inoculated plants.



**Fig. 3** - Proline content of leaves before (left) and after (right) saline stress.

## CONCLUSIONS

1. Chlorophyll and dry matter content of leaves are two physiological indexes, which were the less affected by experimental factors. Their variations appeared both as a response to inoculation and also as an effect of osmotic stress.
2. A broader insight to the changes produced by mycorrhizal fungi on salt tolerance of host plants are given by the assessment of proline content in plants.

Our results showed that at molecular level the AM fungi produced the reduction of negative effects of saline stress on plant physiology.

*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.

## REFERENCES

1. Agong S.G., Kingetsu M., Yoshida Y., Yazawa S., Masuda M. 2003 - *Response of tomato genotypes in induced salt stress*. African Crop Science Journal, 11(2): 133-142.
2. Cortés-Jiménez D., Gómez-Guzmán A., Iturriaga G., Suárez R., Montero Alpírez G., Escalante F.M.E. 2014 - *Microorganisms associated to tomato seedlings growing in saline culture act as osmoprotectant*. Brazilian Journal of Microbiology, 45(2): 613-620.
3. Creelman R., Manson H.S., Bensen R.J., Boyer J.S., Mullet J.E. 1990 - *Water deficit and abscisic acid cause differential inhibition on shoot versus root growth in soybean seedlings*. Plant Physiology, 92: 205-214.
4. FAO. 2008 - *FAO Land and Plant Nutrition Service Management*. <http://www.fao.org/ag/agl/agll/spush>
5. Flowers T.J., Yeo A.R. 1995 - *Breeding for salinity resistance in crop plants – where next?* Australian Journal of Plant Physiology, 22: 875-884.
6. Johnson N.C., Graham J-H, Smith F.A. 1997 - *Functioning of mycorrhizal associations along the mutualism-parasitism continuum*. New Phytologist 135(4): 575-585.
7. Mayak S., Tirosh Tsipora, Glick B.R. 2004 - *Plant growth-promoting bacteria confer resistance in tomato plants to salt stress*. Plant Physiology and Biochemistry, 42(6): 565-572.



## THE BEHAVIOR OF SOME ONION (*ALLIUM CEPA* L.) LOCAL LANDRACES UNDER SALT STRESS

### COMPORTAMENTUL UNOR POPULAȚII LOCALE DE CEAPĂ (*ALLIUM CEPA* L.) SUB INFLUENȚA STRESULUI SALIN

**SLABU Cristina<sup>1</sup>, JIȚĂREANU Carmenica Doina<sup>1</sup>,  
MARTA Alina Elena<sup>1</sup>, BOLOGA (COVAȘĂ) Mihaela<sup>1</sup>**  
e-mail: cslabu@uaiasi.ro

**Abstract.** Soil salinity is a very important ecological factor that affects the growth and the yield of cultivated plants, especially in arid and semi-arid regions. This decrease in land productivity due to salinization, come in contradiction with increased food need. In the effort to create plants resistant to salinity, exploitation of diverse sources of variability as local landraces is required. Onion (*Allium cepa* L.) is a very important vegetable crop considered salt sensitive. The aim of this work was to determinate the effect of salt stress of 11 landraces collected from NE Romania, in Iasi and Botosani districts in areas with saline excess. The results of research show that only some of the studied cultivars reacted to salt stress according to the biphasic model Munns. Only one of this local landraces presented a better salt resistance expressed by high chlorophyll content and a yield close to the control variant.

**Key words:** salt stress, *Allium cepa*, chlorophyll content

**Rezumat.** Salinitatea solului este un factor ecologic foarte important, care afectează creșterea și productivitatea plantelor cultivate, în special în regiunile aride și semi-aride. Aceasta reducere a productivității terenurilor salinizate vine în contradicție cu nevoia crescândă de hrană. În efortul de a crea plante rezistente la stres salin, se impune exploatarea tuturor surselor de variabilitate, de exemplu populațiile locale. Ceapa (*Allium cepa* L.) este o plantă legumicolă foarte importantă, considerată a fi sensibilă la salinitate. Scopul lucrării a fost de a determina efectul stresului salin asupra a 11 de populații locale de ceapă, colectate din județele Iasi și Botosani, din zone cu exces de salinitate. Rezultatele cercetării arată că doar o parte dintre cultivarele studiate a reacționat la stres salin conform modelului bifazic Munns. Numai unul dintre acestea a prezentat o rezistență mai bună la salinitate, exprimată printr-un conținut ridicat de clorofilă și o producție apropiată de cea a variantei martor.

**Cuvinte cheie:** stres salin, *Allium cepa*, conținut de clorofilă

## INTRODUCTION

Soil salinization is a worldwide environmental problem with severe economic and social consequences (Sidike *et al.*, 2014). Salinity limits the plant growth and productivity around the world, especially in arid and semi-arid

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

regions. Currently, over 6 % of land and 20 % of irrigated areas are salt-affected (Munns, 2011). The decrease in land productivity due to salinization, come in contradiction with increased food need. Exponential growth in the ensuing 200 years resulted in the global population exceeding 7 billion, placing enormous demands on modern agriculture (McClung, 2014). The total size of the world population is likely to increase from its current 7 billion to 8–10 billion by 2050 (Lutz and Samir, 2010). In this context, a special attention should be paid to food security. It is necessary not only the quantitative but also the qualitative aspects of food to be considered.

Onion (*Allium cepa* L.) is a very important vegetable crop. Nowadays are cultivated almost worldwide at latitudes between 5-60° in both hemispheres (Sumalan et al., 2014). From 1995 to 2013, worldwide, both harvested area and production has registered a continuous increase, so that in 2013 the dry onions yield was estimated at around 85,78 Million tonnes, and from onions shallots green, at 4,2 Million tonnes (FAO, 2014).

The accumulation of salts in soil is harmful to plants because it increases the concentration of the soil solution, inhibits seeds germination, plants root system morphogenesis, disturb the root absorption and due to the toxic effect of excess ions accumulate in plants. During the last decades, the development of salt-resistant crops, has made very limited progress despite tremendous efforts (Schubert et al., 2009). A reason could be that, the possible physiological, biochemical and molecular mechanisms behind the growth reduction of crops has not yet been adequately understood (Läuchli and Grattan, 2007).

On the other hand, in the effort to increase plant resistance to salinity, local plant landraces should not be overlooked. A first step in plant breeding for the improvement of salt tolerance of cultivated species should be the capitalization of salt tolerance variability in local landraces. In comparison with introduced cultivars, the onion landraces are well adapted to local edaphic and climatic conditions, resulting in high yields and better post-harvest storage ability (Sumalan et al., 2014).

The aim of this work was to determinate the effect of salt stress of 11 landraces collected from NE Romania, in Iasi and Botosani districts in areas with saline excess to capitalize the variability of salt tolerance in local landraces.

## MATERIAL AND METHOD

A pot experiment with factorial arrangements on the bases of randomized complete block with three replications was conducted in 2015 to investigate the salinity tolerance at 11 onion local landraces. These local landraces have been selected from 20 collected landraces after a previous investigation of sets germination capacity under salt stress. The cultivars were collected from NE Romania Iasi and Botosani districts from areas with saline excess. Three sets of each cultivar with a diameter of 15 mm and a mean weight of 3,2 g were planted in garden soil, in 10 kg pots. The pots were placed in the greenhouse at a temperature between 17- 25°C. Two weeks after emergence, the plants were exposed to salt stress. Experimental

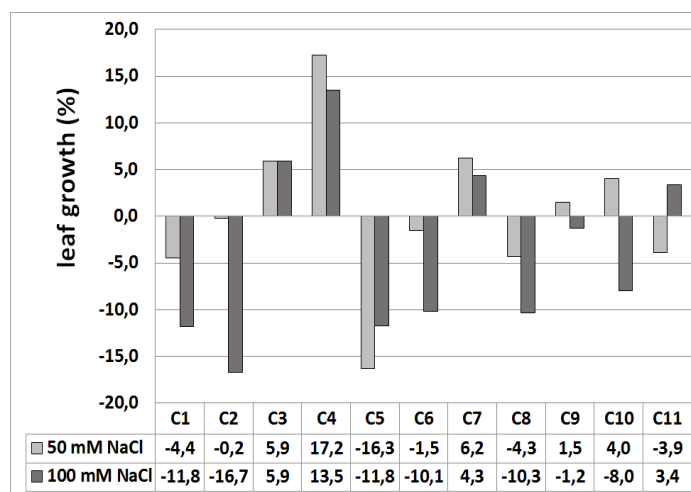
variants: V1 – control, watered with water only; V2 - watered with 1 liter 50 mM NaCl solution for each watering; V3 - watered with 1 liter 100 mM NaCl solution for each watering. Plants were grown to full maturity. Measured were: leaf growth, onion bulb weight and chlorophyll content as chlorophyll content index. The chlorophyll content index was measured in each plot by using a portable chlorophyll content meter (CCM-200, Opti-Sciences Inc., NH, USA).

## RESULTS AND DISCUSSIONS

The growth of crop plants suffering from salt stress can be described by the biphasic model, originally proposed by Munns in 1993 and modified by Munns and Tester in 2008 and Schubert in 2011 (Hütsch *et al.*, 2014). According to the two-phase model of growth response to salt stress by Munns, two major physiological problems may limit crop performance under saline conditions. In a first phase, osmotic problems reduce extension growth so that plants show stunted growth and look dark-green in colour. In a second phase, ion toxicity develops and growth of sensitive plants is severely inhibited (Schubert *et al.*, 2009).

The behaviour of the studied landraces was according to this model evaluated. After 15 days salt treatment only two of the studied landraces (C1 and C8) have responded according to this model, by a reducing of foliar growth and increasing of chlorophyll content (Fig. 1 and 2).

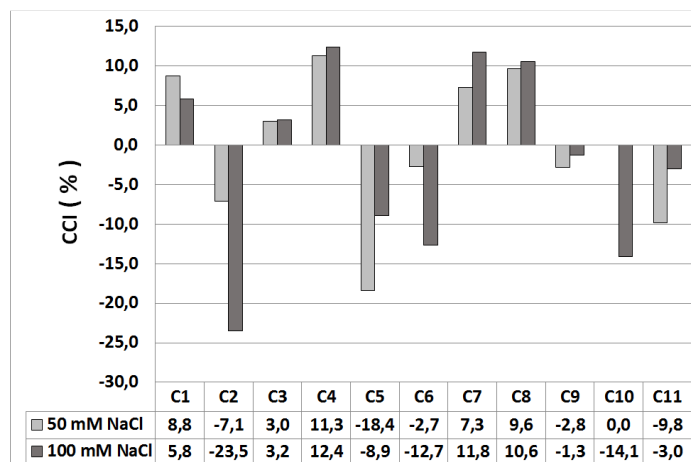
Any other three landraces (C3, C4 and C7) showed a significantly better growth (fig. 1) and a higher chlorophyll content compared with the control variant (Fig. 2).



**Fig. 1** - Effect of 15 days salt stress on the leaves growth (percentage difference to the control variants)

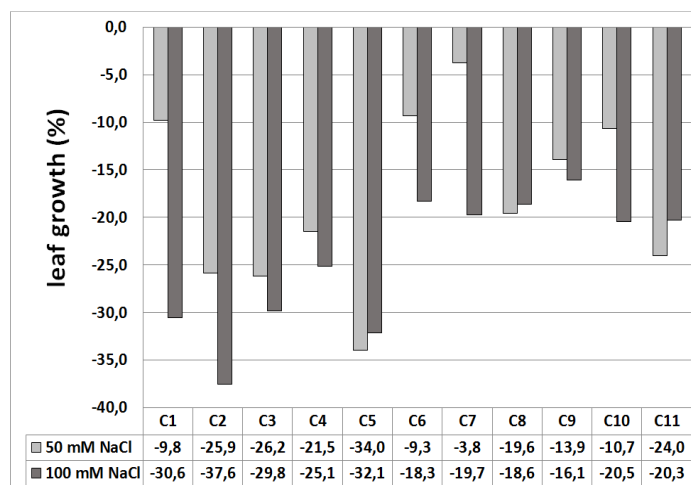
In this case, NaCl promoted the growth. This behaviour can be explained by the fact that onions have a high requirement for Cl, The reason is related to stomates, which regulate movement of gases in and out of plant leaves (Randle, 2004).

On the other hand, onion is considered to be salt sensitive more to sulphate and excluders of both Na and Cl (Shannon et Grieve, 1999).



**Fig. 2** - Effect of 15 days salt stress on the chlorophyll content (percentage difference to the control variants)

After 30 days salt stress, leaf growth content were reduced in the both variants (Fig. 3): after the 50 mM NaCl treatment between 3,8 % (C7) and 34,0 (C5), and after 100 mM treatment between 16,1% (C9) and 37.6% (C2).



**Fig. 3** - Effect of 30 days salt stress on the leaves growth (percentage difference to the control variants)

Also, after 30 days of exposure to salt stress, the leaves chlorophyll content, has registered a significant decrease in case of 9 local landraces compared with untreated control variants. An increase in chlorophyll content was measured at the

landraces 7 and 8. At the first, the increase was at 14% after the 100 mM NaCl treatment and 19% after 50 mM treatment and at the second, the increase was between 28.5 and 65.7% (Fig. 4). Several studies have shown that salinity decreased bulb diameter, bulb weight, root growth, plant height, and number of leaves per plant (Shannon et Grieve, 1999).

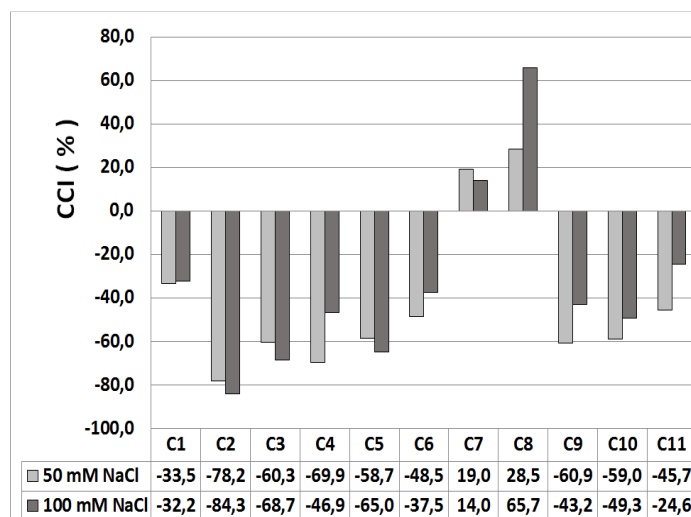


Fig. 4. Effect of 30 days salt stress on the chlorophyll content (percentage difference to the control variants)

Also in this study the onion yield was reduced at 8 until 58% of the control variant under 50 mM NaCl treatment and at 4.0% until 40.7% under 100 mM NaCl treatment (Fig. 5). A best behaviour under NaCl excess manifested landrace 7 with a loss of production from 23% in the 50 mM NaCl variant and from 35 % in 100 mM NaCl variant, which proves a better salinity resistance compared to all of the studied cultivars.

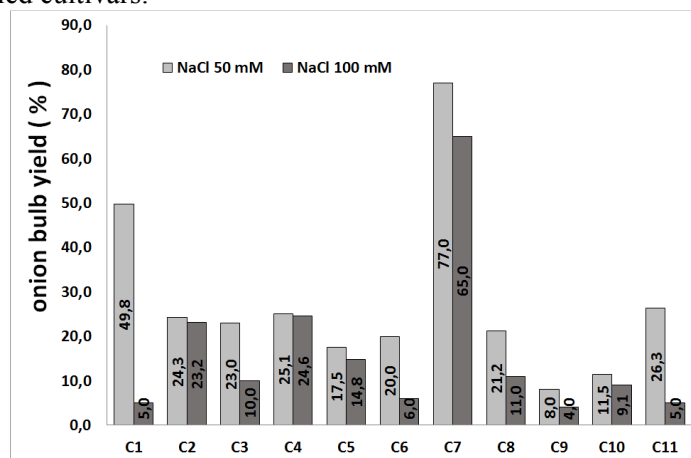


Fig. 5 - Effect of salt stress on bulb yield (percentage to the control variants)

## CONCLUSIONS

1. The analysis of physiological reactions of local landraces of onions to salt stress, according to the biphasic model of Munns, has clearly indicated that not all cultivars follow this model.

2. After 30 days of exposure, salt stress caused a decrease in plant height in all of the cultivars analysed. A decrease in chlorophyll content was observed for nine cultivars.

3. Compared to the control variants under salt stress, the bulb yield was reduced in all of the cultivars analysed.

4. Best tolerance to salt stress, compared to all of the other local landraces studied, was observed for landrace 7, which has been collected in Iasi County. Notably, the lowest decrease in bulb yield was recorded for this particular cultivar, compared to the control.

**Acknowledgements:** *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.*

## REFERENCES

1. Hütsch B. W., Jung S., Schubert S., 2015 . – *Comparison of Salt and Drought-Stress Effects on Maize Growth and Yield Formation with Regard to Acid Invertase Activity in the Kernels*. Journal of Agronomy and Crop Science. Journal of Agronomy and Crop Science Vol. 201(5): 353–367.
2. Läuchli A, Grattan S.R., 2007- *Plant growth and development under salinity stress*. In: Advances in Molecular Breeding Toward Drought and Salt Tolerant Crops (eds. Jenks MA, Hasegawa PM, Jain SM), Springer, Dordrecht, Netherlands, p. 285-315.
3. Lutz W., Samir. K.C., 2010 - *Dimensions of global population projections: What do we know about future population trends and structures?* Phil. Trans. R. Soc. B 365.
4. McClung C. R., 2014 - *Making Hunger Yield*. Science, 344( 6185): 699-700.
5. Munns R., Tester M., 2008 - *Mechanisms of Salinity Tolerance*. Annual Review of Plant Biology, 59: 651-681.
6. Munns R., 2011 - *Plant adaptations to salt and water stress: differences and commonalities*. Adv. Bot. Res., 57: 1–32.
7. Randle W. M., 2004 - *Chloride Requirements in onion: clarifying a widespread misunderstanding*. Better Crops Plant Food, 88(4):10-11.
8. Shannon M.C., Grieve C.M., 1999 - *Tolerance of vegetable crops to salinity*. Scientia Horticulturae 78: 5-38.
9. Schubert S., Anja Neubert, Antje Schierholt, Ali Sümer, Christian Zörb, 2009 - *Development of salt-resistant maize hybrids: The combination of physiological strategies using conventional breeding methods*. Plant Science 177:196–202.
10. Sidike A., Zhao S., Wen Y., 2014 - *Estimating soil salinity in Pingluo County of China using QuickBird data and soil reflectance spectra*. International Journal of Applied Earth Observation and Geoinformation, 26: 156-175.
11. Sumalan R., Ion D., Popescu I., Schmidt B., Sumalan R., Camen D., Ciulca S., 2014 - *Assessment of phenotypic diversity for some red onion landraces from Timis county*. Annals of the University of Craiova-Agriculture, Montanology, Cadastre Vol. 44(1):262-267.
12. \*\*\*FAO, 2014, <http://faostat3.fao.org/compare/E>,

## THE INFLUENCE OF COPPER ON SEED GERMINATION AND GROWTH IN THE FIRST ONTOGENETIC STAGES IN THE SPECIES *BRASSICA OLERACEA* L. AND *CUCURBITA PEPO* L.

### INFLUENȚA CUPRULUI ASUPRA GERMINAȚIEI SEMINTELOR ȘI A CREȘTERII ÎN PRIMELE STADII ONTOGENETICE LA SPECIILE *BRASSICA OLERACEA* L. ȘI *CUCURBITA PEPO* L.

**STRATU Anișoara<sup>1</sup>, LOBIUC A.<sup>1</sup>, COSTICĂ Naela<sup>1</sup>**  
e-mail: anisoara\_stratu@yahoo.com

**Abstract.** This paper presents the results of a study regarding the influence of treatment with copper in different concentrations (60mg/l, 100mg/l and 150mg/l) on germination and incipient growth on two crop species. The following indicators were analyzed: the percentage of germinated seeds, the mean time of germination, the length of the root and of the hypocotyl, the tolerance index, the chlorophyll fluorescence. In both species, the following effects were recorded: a statistically not significant increase in germination percentage; a reduction of the germination mean time, a negative influence on root and hypocotyle elongation; a reduction of the tolerance index, a reduction of the chlorophyll fluorescence  $\Phi$ PSII parameter. *Brassica oleracea* was more sensible compared to *Cucurbita pepo* towards the applied treatments.

**Key words:** copper, chlorophyll fluorescence, germination indices

**Rezumat.** Lucrarea prezintă rezultatele unui studiu referitor la influența cuprului în concentrații diferite (60mg/l, 100mg/l și 150mg/l) asupra germinației și creșterii incipiente la două specii cultivate. S-au luat în studiu următorii indicatori: procentajul de germinație; timpul mediu de germinație; lungimea rădăcinii și lungimea hipocotilului; indicele de toleranță și fluorescența clorofilei. S-au constatat următoarele efecte la ambele specii test: creștere valorică nesemnificativă a procentajului de germinație; reducerea timpului mediu de germinație, influență negativă asupra elongației rădăcinii și a hipocotilului; scăderea indicelui de toleranță, reducere valorică a parametrului fluorescenței clorofilei  $\Phi$ PSII. *Brassica oleracea* comparativ cu specia *Cucurbita pepo* a fost mai sensibilă la concentrațiile aplicate.

**Cuvinte cheie:** cupru, fluorescența clorofilei, indici de germinație

## INTRODUCTION

Copper is a microelement with multiple physiological roles: it is a component of the plastocyanine and numerous enzymes (cytochrome c oxidase, superoxide dismutase etc.); it activates certain enzymes; it has a role in the processes of oxide reduction, respiration, photosynthesis, in the synthesis of the chlorophyll, on organogenesis, in the synthesis of the proteins, carbohydrates

---

<sup>1</sup> "Alexandru Ioan Cuza" University of Iasi, Romania



(Davidescu *et al.*, 1988; Maksimiec, 1997; Rusu *et al.*, 2005).

In natural state, copper is found in form of oxygenated compounds or sulfides (Davidescu *et al.*, 1988). It is absorbed by plants in bivalent ionic form. The copper solubility in the soil, its absorption and translocation in plants are influenced by several factors (Rusu *et al.*, 2005). Industrial emissions, applying in excess plant products, fertilisers, mining activities, depositing waste (Śnieg and Nowak, 2005), processing waste (Luo *et al.*, 2011) contribute to the increase in the content of copper in the soil and the appearance of phenomena of toxicity in plants. According to Kabata- Pendias and Pendias (1993) quoted by Śnieg and Nowak (2005), the maximum content tolerable of copper in the soil is 100mg/kg.

Even though copper is essential for the life of plants, in high concentrations it becomes toxic (Wolhouse, 1983). As manifestations of copper excess, are quoted: reduction/inhibition of the germination and the growth of the root/growth of seedlings/plants (Kumar *et al.*, 2009; Asharge *et al.*, 2013); chlorosis, it is affected the permeability of the membranes, chlorophyll synthesis, photosynthesis (Wolhouse, 1983; Maksimiec, 1997); the activity of some enzymes is modified (Maksimiec, 1997; Yurekli and Banu Porgali, 2006).

*Brassica oleracea* L. var. *capitata* is a valuable food plant, with therapeutic properties and average economic and apicultural share (Pârvu C., 2005). This species can accumulate heavy metals (copper, zinc, lead etc.) in the root and aerial organs (Luo *et al.*, 2011; Rădulescu *et al.*, 2013). *Cucurbita pepo* presents alimentary and fodder value, medicinal properties and average economic and apicultural share (Pârvu, 2003); it can accumulate organic compounds and heavy metals in root and in leaves (Mattina *et al.*, 2003). The species that accumulate large quantities of heavy metals could be used to remediate the polluted soils.

This paper has as purpose the investigation of the copper influence on the germination and growth of seedlings in the species *oleracea* L. var. *capitata* and *Cucurbita pepo* L. var. *oblonga* Wild.

## MATERIAL AND METHOD

The biological material, was represented by seeds of *Brassica oleracea* L. var. *capitata* - "Dittmarcher" and *Cucurbita pepo* L. var. *oblonga* Wild - "Crișan" obtained from S. C. Unisem S. A. Iași. The experimental variants consisted in: control variant (with distilled water) and three variants of treatment with solutions of copper sulphate (copper concentrations: 60 mg/l; 100 mg/l; 150 mg/l). In selecting the concentrations used for the experiment we started from the critical concentration of copper in soil (60 mg/l) (according to Alloway, 1990; Beckett and Davis, 1979, quoted by www.cprm.gov.br /). The seeds were disinfected for 12 minutes at the pumpkin and 8 minutes at the white cabbage, with solutions of oxygenated water 2 % and afterward washed several times with distilled water. The seeds were placed to germinate in Petri dishes, on a filter paper humidified with distilled water (a control variant) and copper sulphate solutions (treatment variants). The plates were kept at room temperature (20 - 23 °C), a photoperiod corresponding to the month of May, 2015. The total duration of the experiment was of 9 day after mounting. For each variant we used four replications, each replication with ten seeds at pumpkin and 20 seeds at white cabbage. Germinated seeds were counted each 24 hours.



The following indicators were analyzed: the percentage of germinated seeds (at 8 days for the pumpkin and 9 days for the white cabbage); the mean time of germination; the length of the root, the length of the hypocotyl; the tolerance index; the chlorophyll fluorescence. The mean time of germination was calculated by the formula described by Ellis and Roberts, (1981) (Moradi *et al.*, 2008). The tolerance index of heavy metals (TI) was calculated by the formula described by Iqbal and Rahmati, (1992) (Ahmad *et al.*, 2012). The fluorescence of the chlorophyll was measured at cotyledon leaves at pumpkin (three cotyledon leaves from each variant of the experiment), by using a portable fluorimeter for chlorophyll of type Hansatech Instruments FMS2 (England). The fluorescence parameters measured were: Fs (state of fluorescence balance) measured by applying on the surface of the leaf of a luminous modulator flux of low intensity, in an interval of 1.8 seconds; Fm' (maximum intensity of the fluorescence emitted by the tissue adapted to light), by applying for 0.7 seconds of a saturating pulse with an intensity  $> 3000 \mu\text{mol}/\text{m}^2/\text{s}$ ; the quantum efficiency of the photo system II ( $\Phi\text{PSII}$ ) as being the proportion  $((Fm' - Fs) / Fm')$ .

The results presented in tables are expressed as mean value  $\pm$  standard error (for the germination percentage and the mean germination time  $n = 4$ ; for the morphological indicators  $n = 20$ ). The data obtained from the germination indices, and the morphological indicators and the fluorescence parameter  $\Phi\text{PSII}$  were statistically interpreted. The unifactorial Anova test followed by the Tukey test ( $\alpha = 0.05$ ) was used in order to test the differences between means (Zamfirescu and Zamfirescu, 2008).

## RESULTS AND DISCUSSIONS

In the treatment variants, comparing with the control, ***the final percentage of germination*** recorded a slight increase in value, but insignificant statistically ( $p < 0.05$ ) (Table 1). An exception to the current situation was registered at the pumpkin: at the concentration of 60 mg/l, the percentage of germination was reduced insignificantly from the statistic point of view (by 10.72%) compared to the control.

***The mean time of germination*** was reduced in all the treatment variants (with the exception of the variant V3 - at the pumpkin), comparing with the control, the reduction being statistically significant ( $p < 0.05$ ) only at the white cabbage, at the concentration of 150 mg/l (Table 1).

The effects of germination stimulation in the case of exposure to various concentrations of copper were reported at: *Phaseolus mungo* (in concentration of 1 mM) (Kumar *et al.*, 2009); psyllium (copper sulphate: 40 mg/l and 60 mg/l) (Mohammadi *et al.*, 2013). At other cultivated species, the exposure to copper determined the reduction of the percentage of germination: chickpea (Kumar *et al.*, 2009); tomatoes, cultivar Roma VF, (in concentration of 100 ppm and 200 ppm) (Asharge *et al.*, 2013); wheat, variant GW-366 (in concentration of 50 ppm - 500 ppm) (Gang *et al.*, 2013). It is considered that the tegument of the seed can represent a barrier between the embryo and the environment in the immediate neighbourhood (Araújo and Monteiro, 2005). The morphological and structural particularities of the seminal tegument in the two species can influence the penetration of the ions of copper. At the pumpkin, the seed tegument is sclerified and at cabbage it is thin.

Table 1

## The percentage of germination and the mean time of germination

Species	The variant	The analysed indicator			
		Germination percentage (%)	+/- (%)	Mean time of germination (day)	+/- (%)
<i>Brassica oleracea</i>	Control	82,5±3,22	0.00	3,65±0,27	0.00
	V1(60mg/l)	91,25±1,25	10,6	3,16±0,10	-13,43
	V2 (100mg/l)	90±3,53	9,09	3,19±1,14	-12,61
	V3 (150mg/l)	91,25±2,39	10,6	3,05±0,12*	-16,44
<i>Cucurbita pepo</i>	Control	70±4,08	0.00	3,78±0,07	0.00
	V1(60mg/l)	62,5±4,78	-10,72	3,40±0,10	-10,06
	V2 (100mg/l)	85±5	21,42	3,68±0,15	-2,65
	V3 (150mg/l)	72,5±2,5	3,75	3,81±0,25	0,79

Note: \* indicates significant differences (Tukey test,  $p < 0.05$ ); +/- represents the percentage of reduction or increase compared to the control

**The length of the root (LR)** and of the **hypocotyls (LH)** recorded a significant reduction in value ( $p < 0.05$ ) comparing with the control in both species (with the exception of the variant V1 – white cabbage, hypocotyl). This fact indicates a negative influence on the elongation of the root and hypocotyl (Table 2). The percentage of the reduction of the root elongation comparing with the control increases with the increase of the concentration of the solution (negative, strong correlation;  $r = -0.906$  for the length of the root at white cabbage and  $r = -0.986$  for the length of the root at pumpkin), fact that indicated a high toxicity of the copper on the root of the two test species. This fact was also presented by Kumar *et al.*, (2009) at chickpea and *Phaseolus mungo*.

Table 2

## The morphological indicators and the tolerance index

Species	The variant	The analysed indicator				
		LR (mm)	+/- (%)	LH (mm)	+/- (%)	TI (%)
<i>Brassica oleracea</i>	Control	31,3±1,62	0.00	23,15±1,44	0,00	-
	V1(60mg/l)	12,8±0,87*	- 59,11	18,6±0,89	-19,66	40,89
	V2 (100mg/l)	9,4±0,74*	- 69,97	15,25±0,91*	-34,13	30,03
	V3 (150mg/l)	7,8±0,81*	- 75,08	14,4±0,93*	- 37,8	24,92
<i>Cucurbita pepo</i>	Control	48,1±3,31	0,00	64,1±4,15	0,00	-
	V1(60mg/l)	34,05±2,58*	- 29,21	45,2±3,19*	- 29,49	70,79
	V2 (100mg/l)	21,75±2,57*	- 54,79	33±2,46*	- 48,52	45,21
	V3 (150mg/l)	16,1±2,15*	- 66,53	26,75±2,25*	- 58,27	33,47

Note: \* indicates significant differences (Tukey test,  $p < 0.05$ ); +/- represents the percentage of reduction or increase compared to the control

The reduction of the length growth of the root, and respectively of the root and hypocotyl/seedlings was reported by other authors, too: Yurekli and Banu Porgali, (2006), at the bean root; Kumar *et al.*, (2009) at the chickpea and bean; Ashagre *et al.*, (2013) at tomatoes, cultivar Roma VF; Gang *et al.*, (2013) at the wheat root, variety GW-366. According to Maksimiec (1997), the copper ions in excess

inhibit to a large extent the elongation, inducing the formation of free radicals that cause oxidative stress and affect the permeability of the membranes. The stress caused by heavy metals reduces the vitality of the root and affects the process of growth (Cheng, 2003).

**The tolerance index (TI)** presented in both species a gradual decrease in value, with the increase of the metal concentration. The pumpkin, comparing with the white cabbage, presented a higher tolerance in the concentrations of copper used (Table 2). A low tolerance to high concentrations of copper was reported at: chickpea and *Phaseolus mungo* (Kumar *et al.*, 2009); tomatoes (Asharge *et al.*, 2013). According to Cheng (2003), plants have their own mechanisms of detoxification of the heavy metals; these mechanisms are integrated and protect the plants against the negative effects caused by heavy metals.

**The chlorophyll fluorescence.** At the concentration of 150 mg/l were recorded the highest values of the parameters  $F_s$  and  $F_m'$ , fact that suggest an unfavourable influence of the copper on the functioning of the photosynthetic apparatus. It was noticed a decrease in value of the parameter  $\Phi PSII$ , insignificant at concentrations of 60 mg/l and 100 mg/l and significant at the concentration of 150 mg/l (Table 3). This fact indicates a sensitivity of the activity of the photo system II in the chloroplasts of the cotyledon leaves at the treatment applied. It is considered that the parameter  $\Phi PSII$  is very sensitive to the excess of heavy metals (Vasiliev and Manolov, 1999).

Table 3

The fluorescence parametres at pumpkin

Species	The variant	The analised indicator		
		$F_s$ (bits)	$F_m'$ (bits)	$\Phi PSII$
<i>Cucurbita pepo</i>	Control	636,66 (100)	3416 (100)	0,813 (100)
	V1(60mg/l)	638 (100,21)	3222,33 (94,33)	0,801 (98,52)
	V2 (100mg/l)	586 (92,04)	3075,66 (90,03)	0,809 (99,50)
	V3 (150mg/l)	785,66 (123,40)	3694,33 (108,14)	0,787* (96,80)

Note:\* indicates significant differences (Tukey test,  $p < 0.05$ ); the number in brackets represents the percentage relative to control

## CONCLUSIONS

1. The copper in the concentrations used did not affect the germination, but it influenced unfavourably the growth of seedlings and the parameters of the chlorophyll fluorescence.

2. The two test species presented a similar evolution of the sensitivity comparing with the increase in the metal concentration. *Cucurbita pepo* comparing with *Brassica oleracea* was less sensitive at the applied concentrations.

**Acknowledgements:** The work received support from the CERNESIM project (SMIS/CSNR 3984/901) by using the portable fluorimeter for chlorophyll.

## REFERENCES

1. Ahmad I., Akhtar M., J., Zahir Z. A., Jamil A., 2012 - Effect of cadmium on seed germination and seedling growth of four wheat (*Triticum aestivum* L.) cultivars. *Pak. J. Bot.*, 44 (5):1569-1574.
2. Araújo A.S.F., Monteiro R.T.R., 2005 - Plant bioassays to assess toxicity of textile sludge compost. *Sci. Agric. Piracicaba Brazil*, 62: 286-290.
3. Ashagre H., Almaw D., Feyisa T., 2013 - Effect of copper and zinc on seed germination, phytotoxicity, tolerance and seedling vigor of tomato (*Lycopersicon esculentum* L. cultivar Roma VF). *International Journal of Agricultural Science Research*, 2, (11):312-317.
4. Cheng S., 2003 - Effects of heavy metals on plants and resistance mechanisms. *Environ. Sci & Pollut Res.*, 10 (4):256-264.
5. Davidescu D., Davidescu Velicica, Lăcătușu L., 1988 - Microelementele în agricultură. Ed. Acad. R. S. R., București, p. 80-82,132-136.
7. Gang A., Vyas A., Vyas H., 2013 - Toxic effect of heavy metals on germination and seedling growth of wheat. *Journal of Environmental Research And Development*, 8 (2) :206-213.
8. Luo C., Liu C., Wang Y., Liu X., Li F., Zhang G., Li X., 2011 - Heavy metal contamination in soils and vegetables near e-waste processing site, south China. *Journal of Hazardous Materials*, 186:481-490.
9. Kumar N., Baudh K., Singh R., Ananad K., Barman S. C., Singh D. P., 2009 - Phytotoxicity of trace metals (Cu & Cd) to gram (*Cicer arietinum*) and mung (*Phaseolus mungo*). *J. Ecophysiol. Occup. Hlth.* 9:59-65.
10. Maksymiec W., 1997 - Effect of copper in higher plants. *Photosynthetica*, 34: 321-342.
11. Mattina M.J.I., Lannucci-Berger W., Musante C., White, J.C., 2003 - Concurrent plant uptake of heavy metals and persistent organic pollutants from soil. *Environ. Pollut.* 124:375-378.
12. Mohammadi, Z., Nabavi Kalat, S. M., Sadrabadi Haghaghi, R., 2013 - Effect of copper sulfate and salt stress on seed germination and proline content of psyllium (*Plantago psyllium*). *American-Eurasian Journal of Agricultural & Environmental Sciences*, 13 (2) :148-152.
13. Moradi D., Sharifzadeh P.F., Janmohammadi M., 2008 - Influence of priming techniques on seed germination behavior of maize inbred lines (*Zea mays* L.). *ARPJ. J. Agric. Biol. Sci.*, 3:22-25.
14. Pârnu C., 2003/2005 - *Enciclopedia plantelor. Plante din flora României*. vol. II, IV. Ed. Tehnică, București, p. 47-62, 817-843.
15. Rădulescu C., Stihl C., Popescu I.V., Dulamă I. D., Chelărescu E.D., Chilian A., 2013 - Heavy metal accumulation and translocation in different parts of *Brassica oleracea* L. *Rom. Journ. Phys.*, 58 (9-10) :1337-1354.
16. Rusu M., Mărghiș M., Oroian I., Mihăiescu T., Dumitraș Adelina, 2005 - *Tratat de agrochimie*. Ed. Ceres, București, p.399 - 407.
17. Śniek B., Nowak J., 2005 - Urease activity and ATP content in soil and plant related to copper concentration. *Polish Journal of Ecology*, 53(1) :105-111.
18. Vasiliev A., Manolov P., 1999 - Chlorophyll fluorescence of barley (*H. vulgare*) seedlings grown in excess of Cd. *Bulg. J. Plant Physiol.*, 25(3-4): 67-76.
19. Woolhouse H. W., 1983 - Toxicity and tolerance in the responses of plants to metals, in: Lange O. L., Nobel P. S., Osmond C. B., Ziegler H., (Eds.). *Encyclopedia of Plant Physiology*. New series, 12 C, Springer- Verlag, Berlin, Heidelberg, New York, 275-282.
20. Zamfirescu Ș., Zamfirescu Oana, 2008 - *Elemente de statistică aplicate în ecologie*. Ed. Univ. „Al. I. Cuza”, Iași, p. 108-116.
21. Yurekli F., Banu Porgali Z., 2006 - The effects of excessive exposure to copper in bean plants. *Acta Biologica Cracoviensi, Series Botanica*, 48(2):7-13.
22. [www.cprm.gov.br/pgagem/Manuscripts/jeliazkova.htm](http://www.cprm.gov.br/pgagem/Manuscripts/jeliazkova.htm)

## GRAFTING OF VEGETABLES IN ROMANIA

### ALTOIREA LEGUMELOR ÎN ROMÂNIA

**BOGOESCU M.<sup>1</sup>, DRAGOMIR Elena<sup>1</sup>**

**e-mail:** bogoescumarian@gmail.com

**Abstract.** Soil diseases and nematodes cause great damages to vegetable crops and represent some of the most limiting factors for farmers' income. Their global management was based on the soil's fumigation with methyl bromide before planting, a compound whose elimination procedure was initiated by the Protocol from Montreal (1992), due to its dangerous effects on the environment. Romanian Government decided to eliminate gradually the use of methyl bromide since 2005. In the recent years, the research has focused on investigating the alternatives to methyl bromide for prevention and combating the soil diseases and nematodes, particularly in protected vegetable crops. These alternatives were based both on the use of other chemical compounds and some non-chemical methods (soil disinfection with steam, use of green houses, culture practices, improving the resistance of plants). The purpose of this paper is to present the actual achievements to in grafting of vegetables in Romania.

**Key words:** scion, rootstock, marketable production, commercial quality, nutritional value, soil diseases

**Rezumat.** Bolile de sol și nematozii produc mari pagube în culturile de legume și reprezintă unii dintre cei mai limitativi factori ai veniturilor fermierilor. Gestionarea lor la nivel mondial a fost bazată pe fumigația solului înaintea plantării cu bromură de metil, un compus a cărui procedură de eliminare a fost inițiat prin Protocolul de la Montreal (1992), datorită efectelor sale periculoase asupra mediului. Guvernul României a decis să elimine treptat utilizarea bromurii de metil începând cu anul 2005. În ultimii ani, cercetarea s-a concentrat pe investigarea alternativelor la bromura de metil pentru prevenirea și combaterea bolilor de sol și a nematozilor, în special în culturile protejate de legume. Aceste alternative s-au bazat atât pe utilizarea altor compuși chimici cât și pe unele metode nechimice (dezinfectia solului cu abur, solarizarea, practicile culturale, îmbunătățirea rezistenței plantelor). Scopul acestei lucrări este de a prezenta realizările la zi în domeniul altoirii legumelor din România.

**Cuvinte cheie:** altoi, port-altoi, producția vandabilă, calitatea comercială, valoarea nutritivă, boli de sol

### INTRODUCTION

Abiotic environmental factors from protected areas offer favorable conditions for many species of pathogens and pests which cause high production damages. Soil diseases and nematodes have a particularly destructive action in

---

<sup>1</sup> Research and Development Institute for Industrialization and Marketing of Horticultural Product, București, Romania

vegetable crops and represent one of the major factors of limiting farmers' income, making necessary to adopt soil disinfection practices, or other technological methods (Gullino *et al.*, 2003). Before 2002, the most common methods of soil disinfection in Romanian greenhouses were soil sterilization by steam and use of methyl bromide (107.72 tones of methyl bromide in 2003), (Bogoescu *et al.*, 2005). Traditionally, the greenhouses were built around the for thermal power plants from main cities, using the hot water produced by them for heating.

Methyl bromide was until now and remained perhaps the only fumigant that is efficient against pathogens, nematodes, weeds, insects and rodents. But, according the Montreal Protocol (1997) use the methyl bromide (a compound which is applied to soil by fumigation for plant protection) leads to ozone's layer reduction in the stratosphere and is forbidden in Romania since 1 January 2005. In this situation, beginning with 2002 there was performed research that aimed to establish alternatives to the use of methyl bromide in Romanian horticulture. Grafting the high quality and productivity of varieties on rootstocks that are resistant to diseases and pests from the soil, it is a method known for a long time, but has been improved and has spread rapidly in the recent years (Bausher and Chellemi, 2001). Grafting of a grafted plant on the wild type of rootstock which has a a root robust system and strong resistance to pests and diseases ,a cultivar with remarkable technological and sensorial qualities, all that lead to a superior tolerance of the grafted plant to the less favorable conditions of soil and environmental (Edelstein, 2004; Oda, 1993).

However, the most important result of the grafting process of vegetables is given by conferring the high resistance to soil diseases (such as *Fusarium*, *Verticillium*) or nematodes (Bogoescu *et al.*, 2005).

The first grafting of vegetables were performed in Romania in the 70s of last century. Further, until 2001 this activity has never took place. Beginning with 2002 were started researches in this area of horticulture, under a project funded by UNIDO. The research aimed to establish alternatives to the use of methyl bromide in Romania; the biological alternatives for the vegetable's grafting were recorded similar results to those found in many chemical alternatives.

By this reason, the research have been developed as far as in present it has been established the production technologies and cultivation of grafted seedlings for tomatoes, eggplants, melons and water melons, cucumbers. Since 2015 have been initiated the necessary research for specification the technology for obtaining the grafted seedlings and cultivation of vegetables from pepper's group.

As result of the developed researches and dissemination of obtained results, the grafted seedlings technology extended to many private manufacturers and watermelon's culture it is realizing in a proportion of 80% by using grafted seedlings. Only in the village Dăbuleni, Dolj county—with tradition in melon's cultivation - were purchased in 2014 over two million of grafted seedlings of melons from import.



The aim of this paper is to present some results obtained during development the researches in the field of vegetable's grafting, in Romania.

## MATERIAL AND METHOD

The researches were organized during 2008 – 2013 on tomatoes, cucumbers and watermelons cultivated in demonstration station of HORTING Institute, in order to identify new alternatives to use the methyl bromide in Romania.

The efficiency of the chemical alternative with methamsodium (100 ml/m<sup>2</sup>), respectively the non-chemical alternative, vegetable's grafting, were tested and compared with the results obtained after soil disinfection with methyl bromide (75 g/m<sup>2</sup>), used as control variant.

V1= methamsodium

V2= grafted plants

V3=methyl bromide

The variants, with an average surface of 900 m<sup>2</sup>, were organized by the method of random blocks, each experimental variant having three replicates.

The soil disinfectants were applied through a dripping irrigation system. Soil disinfection was done according to the specific climate conditions, respectively to a temperature over 15°C at a 10 cm depth in the soil. The soil has been covered with polyethylene film (4 m width; 0,11 mm thickness; gas-proof). The waiting time was 8 days for methyl bromide and 21 days for methamsodium.

The Mondial F1(Enza Zaden) tomatoes hybrid was grafted on the Beaufort (De Ruiter Seeds) resistant rootstocks; the Mathilde F1 (Royal Sluis, Holland) cucumber hybrid was grafted on the Shintoza (*Curcubita maxima* x *Cucurbita moschata*) resistant rootstock and the Cicerio F1 (Ergon Seed) watermelon hybrid was grafted on the ES30900 (Ergon Seed) resistant rootstock. The grafted plant culture had the following densities:

Grafted tomatoes = 18000 plants/ha, realized by 2 stems

Grafted cucumber = 32.000 plants/ha

Grafted watermelons = 3000plants/ha

There were done observations and determinations regarding the influence of soil disinfections treatments and the use of grafted plants, about:

- marketable production
- appearance of first crop
- quality of fruits
- nutritional value of fruits
- frequency of soil diseases and nematodes:
  - corky root induced by *Pyrenochaeta lycopersici* - tomatoes
  - *Fusarium oxysporum* f. sp. *cucumerinum* – cucumbers and watermelons
  - incidence of nematode's attack on roots

The presence of galls induced by nematodes from the *Meloidogyne* genus was visually assessed at the end of deforestation of plots, on 15% of the plants harvested from the middle of the plots. The following indexing scale was used (Lamberti and Di Vito):

- 0 = no galls
- 1 = very slight infection, not widespread galls, presence of 1-5 galls located only on few roots

- 2 = slight infection, rare widespread galls, presence of no more than 20 galls spread on roots system
  - 3 = medium infection with widespread galls, more than 20 revealed galls and well spread on all roots system;
  - 4 = strong infection, roots system integral affected and deformed due the presence of big galls on the main roots;
  - 5 = very strong infection, roots system completely affected and totally deformed due to the presence of big galls, absence of capillary roots.
- The root's index (0-5) was calculated as follows:

$$\frac{\sum \text{nematode's index of all plants}}{\text{Number of plants}}$$

For the pathogens determined there were calculated the frequency, by the following formula:

$F\% = N \times 100 / N_t$ , where:

N = number of attacked plants

$N_t$  = total number of analyzed plants

Data's analysis was performed on the average data recorded for the examined species of vegetables (tomatoes, cucumbers, watermelon).

Statistical analysis of results was performed by Duncan's test.

## RESULTS AND DISCUSSION

In order to reveal the influence of different alternatives regarding the marketable production, data were collected and presented in Table 1. The use of grafting plants conducted to a significant higher production comparing with soil disinfection with methamsodium, with over 8,5% at tomatoes and cucumbers, respectively 95,1% at water melons. The obtained results showed a significant difference at the variant with grafted plants comparing with the method by soil disinfection with sodiummetham (over 18,70%). At the same time, there were no registered significant differences between the grafted plants production and the soil disinfection variant, using methyl bromide (witness variant), just in watermelons culture where the production was with 54% higher.

Table 1

**Influence of vegetable's grafting and some chemical methods of soil disinfection on marketable production (t/ha)\***

Culture	Sodium metham	Grafted plants	Methyl bromide
Tomatoes	114 b	124a	128a
Cucumbers	47a	51a	55a
Water melons	48b	82a	53b

\*Into a row, the values written with the same letter do not present significant differences after Duncan test, for  $p=5\%$ .

Data presented in Table 2 refers at experiments which followed the influence of variants which were studied on the earliness grade of vegetables. The observations and determinations followed the registration of the crop's earliness,



expressed by first harvesting day, respectively the number of days between plantation and harvesting. Recorded data reveals a delay for first harvesting in grafted plants variant with 5-7 days, comparatively with first harvesting day at non-grafted plants situation, which were planted in disinfected soil with soil fumigants. The differences obtained between the disinfection method by soil fumigation and grafted plants method was statistical ensured for  $p=5\%$ , by Duncan test.

Table 2

**Influence of vegetable's grafting and some chemical methods of soil disinfection on earliness (nr.days)**

Culture	Methamsodium	Grafted plants	Methyl bromide
Tomatoes	81a	88b	80a
Cucumbers	32a	37b	32a
Water melons	78a	85 b	77a

Into a row, the values written with the same letter do not present significant differences after Duncan test, for  $p=5\%$ .

In Table 3 are presented data which refer to the influence of alternative methods comparing with methyl bromide, on the percentage of first class fruits. Fruit's quality was assessed according to the quality standards for fresh fruits and vegetables: SR 1421/2001 for tomatoes, SR 1416/2003 for cucumbers and SR 3654/2003 for watermelons. The analyzed data showed an improvement of commercial quality at grafted plants, respectively at first class fruits, in percentage of over 84,4% at tomatoes and cucumbers and over 93% at water melons, comparatively with non-grafted plants, where depending of variety and treatment method, the percentage of first class fruits never exceeding values between 69,9–81,1%; the differences between the disinfection methods by fumigation and use of grafted plants are statistical ensured for  $p=5\%$ , by Duncan test.

Table 3

**Influence of vegetable's grafting and some chemical methods of soil disinfection on quality (%)**

Culture	Sodium metham	Grafted plants	Methyl bromide
Tomatoes	80,7b	84,6a	81,1ab
Cucumbers	78,4b	84,4a	78,3b
Water melons	69,9c	93,1a	78,8b

Into a row, the values written with the same letter do not present significant differences after Duncan test, for  $p=5\%$ .

Regarding the nutritional value's evaluation there were made determinations of soluble dry substance and soluble carbohydrates content. Determinations were made on standard samples of 6 melons /sample and 3 kilograms/sample of tomatoes and cucumbers. Samples were taken from the mass of product during two harvests. The biochemical analysis were realized by refract

metric method for soluble dry substance (STAS 5956/1991) and respectively, by Bertrand method for soluble carbohydrates content.

The results shown represent the average of both harvests (Table 4).

Table 4

**Influence of vegetable's grafting and some chemical methods of soil disinfection on nutritional value (%)**

Culture	Sodium metham		Grafted plants		Methyl bromide	
	Soluble substance	Soluble carbohydrates	Soluble substance	Soluble carbohydrates	Soluble substance	Soluble carbohydrates
Tomatoes	5,6a	4,18m	5,4a	3,99m	5,6a	4,09m
Cucumbers	4,6a	3,81m	4,4a	3,72m	4,7a	3,94m
Water melons	6,5a	5,84m	6,2a	5,75m	6,4a	5,78m

Into a row, the values written with the same letter for soluble substance (a..b) and soluble carbohydrates(m...n) do not present significant differences after Duncan test, for p=5%.

The obtained results showed a slight decrease regarding the value of the two indicators of nutritional value at the variant in which were used grafted plants of any variety. The differences registered, between the soil disinfection by fumigation variant and grafted plants cultivation, are not relevant statistically.

Based on observation made "in vitro" culture there were identified soil pathogens as *Pyrenochaeta lycopersici* at tomatoes and *Fusarium oxysporum f. sp. cucumerinum* at cucumbers and water melons. The data revealed the efficiency of soil disinfection treatments with methyl bromide and methamsodium and in the same time the tolerance and resistance of grafted plants to soil diseases (table 5). The frequency of the attack of *Pyrenochaeta lycopersici* fluctuated between 1,39% at tomatoes planted in disinfected soil with methyl bromide and 1,49% at grafted plants. At soil pathogen *Fusarium oxysporum f. sp. cucumerinum* was registered a frequency of attack of 2,29% to cucumbers planted in disinfected soil with methyl bromide and 2,43% at grafted cucumbers; a similar curve was registered at the water melons culture, too.

Table 5

**Influence of vegetable's grafting and some chemical methods of soil disinfection on frequency of soil disease (%)\*\***

Culture	Sodium metham	Grafted plants	Methyl bromide
Tomatoes	1,41a	1,49a	1,39a
Cucumbers	2,34m	2,43m	2,29m
Water melons	0,91m	0,87m	0,81m

\*Into a row, the values written with the same letter ("a" for *Pyrenochaeta lycopersici*) and ("m" for *Fusarium oxysporum*) do not present significant differences after Duncan test, for p=5%.

\*\* *Pyrenochaeta lycopersici* to tomatoes

*Fusarium oxysporum f. sp. cucumerinum* to cucumbers and water melons

Although was registered a higher frequency of the soil diseases on grafted plants – indifferently of variety- the differences are statistical found in experimental error, not being registered significant differences after Duncan test, for  $p=5\%$ .

Regarding the alternatives effect to nematode attack (*Meloidogyne* spp.), the results at the moment of removing the experimental variants are shown in Table 6.

Tabel6

**Influence of vegetable's grafting and some chemical methods of soil disinfection on the incidence of nemathodes attack (%)\*\***

Culture	Sodium metham		Grafted plants		Methyl bromide	
	Root index <sup>2</sup>	Frecvency <sup>1</sup>	Root index <sup>2</sup>	Frecvency <sup>1</sup>	Root index <sup>2</sup>	Frecvency <sup>1</sup>
Tomatoes	1,8	3,1b	1,9	3,1b	0,9	1,1a
Cucumbers	1,7	2,9b	1,8	3,3b	0,8	0,6a
Water melons	1,2	0,9a	1,2	1,1a	1,1	0,8a

Into a row, the values written with the same letter do not present significant differences after Duncan test, for  $p=5\%$ .

<sup>1</sup>Average of percentage of infected plants evaluated with all plants, collapsed, but still alive at the end of cycle.

<sup>2</sup> Galles Index of nemathodes on the rooths, after Lamberti and DI Vito: variation interval 0:5

It is observed a decrease of frequency of nematode'sgalls in case of variants where the soil was disinfected with methyl bromide (0,6 – 1,1%) and methamsodium (0,9 – 3,1%). The frequency of nematode's attack registered to grafted plants was (according to varieties) between (1,1 - 3,1%).

In the same time, the index regarding the galls on roots registries a variation from 0,8% in the situation of treatment with methyl bromide (water melons) until 1,90 at grafted tomatoes. The analysis of data confirms the tolerance effect of grafted plants on nematode's attack *Meloydogine* spp. The obtained results do not shows significant statistical differences between chemical alternatives (sodium metham) and use of grafted plants.

## CONCLUSION

1. Results revealed that in Romanian conditions can be successfully used both chemical method (methamsodium) and non-chemical method of grafted plants, for preventing diseases of plants caused by soil pathogens and pests

2. Grafting of vegetables leads to increasing of marketable production, of commercial quality and do not affects the nutritional value of plants

3. Obtained results show as appropriate the following alternatives at methyl bromide:

- Chemical method: fumigation with methamsodium
- Non-chemical method: grafted plants method

4. Between the non-chemical methods the benefits of grafted plants in vegetable's cultivation overcome the possible risks.

#### REFERENCES

1. **Bausher M.G., Chellemi D.O., 2001** - *Performance of grafted tomatoes in open field trials at two locations in Florida*. USDA ARS.
2. **Bogoescu M., Gullino M.L., Minuto A., Amadio A., 2005** - *Alternatives to methyl bromide Romanian protected crops*. Acta Horticulturae, 698: 315-320.
3. **Di Vito M., 1979** - *Status of research on biology and control of the root-knot nematodes in Italy*. Proc. II Planning Conference on Root-Knot Nematodes, *Meloidogyne* ssp., Athens (Greece), 26-30: 135-137
4. **Edelstein M., 2004** - *Grafting vegetable crop, plants: pros and cons*. Acta Hort. (ISHS) 659:235-239
5. **Gullino M. L., Camponogara A., Gasparrini G., Rizzov-Clini C., Garibaldi A. 2003** – *Replacing methyl bromide for soil disinfestations: the italian experience and the implication for other countries*. Plant Disease, 87: 1012-1021
6. **Oda M., 1993** - *Present state of vegetable production using grafted plants in Japan*. Plant Disease, 87:442- 446.

## TRADE MARKET OF VEGETABLE SEEDLINGS, CURRENT AND FUTURE PERSPECTIVE IN ROMANIA

### PIAȚA COMERCIALĂ DE RĂSADURI DE LEGUME, PERSPECTIVA ACTUALĂ ȘI VIITOARE ÎN ROMÂNIA

**SIKAVELIS K.<sup>1</sup>, ROȘCA I.<sup>1</sup>**

**e-mail:** ioanrosca\_usamv@yahoo.com

**Abstract.** It is presented current situation in Romania of vegetables cultivation, disease and pest keys in protected and field vegetable crops, using of pesticides and adjacent national and European regulations at farm and national level. In Romania, vegetable production technology based on grafted vegetable seedlings was introduced in 2009. The market today has grafted seedlings of eggplant, tomatoes, peppers, cucumbers, watermelons and melons, but tomato seedlings grafted on two arms are the most common and popular. It is discussed trade market of vegetable seedlings, current and future perspective in Romania with a special reference on the market addressed to farmers which are using grafted vegetables from Greece, Turkey and Hungary. It is discussed the advantages of using grafted vegetable plants in conditions when over 80% of the farmers do not receive subsidies on product support even on the surface (EU standards 0.3 hectares), grafting can be regarded as an important link in obtaining biological or ecological vegetables. It is discussed trade case for vegetable seedlings of the firm Agris S.A from Salonic, the experience with most well-known Romanian greenhouses.

**Key words:** evolution of vegetable seedlings trade market, Romania

**Rezumat.** Este prezentată situația actuală în România privind cultivarea legumelor, bolile și dăunătorii cheile în culturile de legume în spații protejate și câmp, folosirea pesticidelor și reglementările naționale și europene adiacente la nivel de fermă și la nivel național. În România, tehnologia de producție de legume pe bază de răsaduri altoite a fost introdusă în 2009. Pe piață astăzi există răsaduri altoite de vinete, roșii, ardei, castraveți, pepeni verzi și galbeni, dar răsadurile de roșii altoite pe două brațe sunt cele mai comune și populare. Este discutată piața comercială de material săditor de legume, perspectiva actuală și viitoare în România, cu o referire specială pieței adresată fermierilor care utilizează legume altoite din Grecia, Turcia și Ungaria. Sunt discutate avantajele utilizării legumelor altoite în condițiile în care peste 80% dintre agricultorii nu primesc subvenții pe produs sau pe suprafață (standardele UE 0,3 hectare), altoirea poate fi privită ca o verigă importantă în obținerea legumelor biologice sau ecologice. Se discută cazul comerțului pentru răsaduri de legume ale firmei Agris SA din Salonic, experiența sa cu cele mai cunoscute sere românești.

**Cuvinte cheie:** Evoluția pieței comerciale de răsaduri de legume, România

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Bucharest, Romania

## INTRODUCTION

In China, the earliest literature about vegetable grafting was recorded in an ancient "Book of Fan Shengzhi shu jing shi" in 32-37 B.C., an agriculturist book of China written by Fan Shengzhi translated and commented upon by Shi Shenghan (1959). Due to intensive horticultural practice and limited land, grafting of vegetable seedlings is more and more spreaded as new technology, even this was applied from late 20<sup>th</sup> century in Eastern Asia, being most suitable for trade market of vegetable seedlings. The grafted varieties on to a rootstock brings qualities to the plant that the vegetable does not possess, avoid the situation where the soil is infested by diseases and pests, for this reason this method was spreaded and applied with success. In Eastern Asia watermelons were grafted to prevent root rot and now is possible to sell grafted vegetable of rootstocks. After Kubota (2008), the first record of interspecific, herbaceous grafting as a yield increase and pest/disease control strategy was for watermelon *Citrullus lanatus* (Thunb.) Matsum. & Nakai, using a squash rootstock (*Cucurbita moschata* Duch.), reportedly developed by a watermelon farmer in Japan (Tateishi, 1927). Grow vegetables in the same soil year after determined increasing of frequency of soil-borne diseases which decrease quantity and quality of vegetable, in this respect rootstock most often used for grafted vegetables is resistant to some soil-borne diseases. Grafted plants have earlier and higher crop, seems to be resistant to cultivation in the same place year after year, at lower temperatures of soil and field overwatering. However, commercial application began in 1970<sup>s</sup> and increased with the rapid development of protected cultivation. The main purposes of grafted vegetable production are to overcome soil borne diseases and increase resistance against abiotic stress (Williamson, 1998; Cohen *et al.*, 2000; Igarashi *et al.*, 2001; Ioannou, 2001; Cao *et al.*, 2005; Sigüenza *et al.*, 2005). Due to resistance through grafting to the pathogens and even some pests, part from very expensive chemical treatments difficult to apply were reduced.

## MATERIAL AND METHOD

Based on own experience it is presented results of Agris S.A. in selling seedlings in Romania. It is discussed trade case for vegetable seedlings of the firm, the experience with most well-known Romanian greenhouses [Pipera, IVAs (Constanta), Leoser (Popesti Leordeni and Constanta), Chirana (Slobozia), Interagro (Zimnicea), Rovina (Deva), Topsemconsult (Iasi), Aroneanu (Iasi), Agromir (Hateg), Agros Braila, Cernavoda] regarding influence of grafting on soil diseases and nematode's attack, where the observation were done twice on month after seedling planting. Based on local experience were synthesized using of pesticide in vegetable cultivation.

## RESULTS AND DISCUSSIONS

Disease and arthropod pests are a continual problem for field and greenhouse vegetable production. These problems range from minor infestations to major disease

or arthropod pest outbreaks that can destroy an entire crop. In Romania, in the past, the major management strategy was pesticide control. Pesticides registered in Romania for watermelons and melons with active ingredient clorotalonil; fosetil + propamocarb; triadimenol + folpet; sulphur; thiophanate-methyl. For tomatoes, fungicides, *seed treatment*, metalaxil-M; mefenoxam, *treatment in vegetation*, dimethomorph 9% + mancozeb; copper; metallic copper; metallic cooper from copperoxychloride; propineb; metalaxyl + copper oxychloride with metallic cooper; clorotalonil; piraclostrobin + metiram; mancozeb; cymoxanil + famoxadone; folpet; iprovalicarb + copper oxychloride; cymoxanil + copper; tebuconazole; azoxystrobin; mandipropamid + mancozeb; metiram; fosetil + propamocarb; mefenoxam + mancozeb; mefenoxam + metallic cooper; difenoconazole difenoconazole; triadimenol + folpet; boscalid + pyraclostrobin; fludioxonil + cyprodinil; fenhexamid; thiophanate-methyl; fosetyl aluminum + fenamidone, insecticides, emamectin benzoate; thiamethoxam; alfa-cipermetrin; cipermetrin; pyriproxyfen; spinosad; lufenuron; tau-fluvalinate; spirotetramat; dimetoat; imidacloprid (in field after flowering); cipermetrin + clorpirifos; gamma cihalotrin; abamectin abamectin; abamectin + chlorantraniliprole; oxamyl, molluscocides and nematocides, methiocarb; fostiazat, soil disinfectants, dazomet; metam-sodiu. For pepper, fungicides, *treatment in vegetation*, copper from copper oxychloride + fosetyl aluminum + myclobutanil; azoxystrobin; sulfur; penconazole; thiophanate-methyl; insecticides, emamectin benzoate; alfa-cipermetrin alfa-cipermetrin; cipermetrin; pyriproxyfen; acetamiprid; flonicamid; oxamyl acaricides, hexythiazox; abamectin; soil disinfectants, metam-sodiu. For cucumber, fungicides, *treatment in vegetation*, dimethomorph + mancozeb; metallic copper; fosetyl aluminum; propineb; clorotalonil; popiconazole; metallic cooper from copperoxychloride; copper hydroxide + metallic cooper; mancozeb; folpet; fluopicolide + propamocarb; meptyldinocap; cooper from copperoxychloride + fosetyl aluminum; mancozeb + myclobutanil + fosetyl aluminum; improvalicarb + cooper as copperoxychloride; tebuconazole; mandipropamid + mancozeb; metiram; fosetyl + propamocarb; mefenoxam + mancozeb; clorotalonil; triadimenol + folpet; sulfur; myclobutanil; fenhexamid; sulfur; penconazole; thiophanate-methyl; fosetyl aluminum + fenamidone, insecticides, thiamethoxam; alfa-cipermetrin; cipermetrin; pyriproxyfen; spinosad; tau-fluvalinate; acetamiprid; imidacloprid; oxamyl, acaricides, milbemectin; hexythiazox; abamectin, nematicides, fostiazat, soil disinfectants, metam-sodiu. For eggplant, fungicides, *treatment in vegetation*, thiophanate-methyl, insecticides, thiamethoxam, deltametrin, alfa-cipermetrin; pyriproxyfen; lambda-cihalotrin; oxamyl, acaricides, abamectin; soil disinfectants, metam-sodiu.

However, many plant pathogen, insect and mite pests are resistant to registered pesticides and few new pesticides are being developed. Nowadays, there is a global attempt to minimize the use of harmful substances, particularly pesticides in agriculture, in order to reduce the actual levels of pollution in the environment. In addition, customers worldwide demand produce for consumption that is free of



pesticide residues. It is for this reason that non-chemical methods are welcome in the context of the agricultural production as one of the ways to reduce the use of substances of different levels of toxicity. Alternative control strategies exist or are being developed for most major pests and diseases.

Introducing grafted vegetables into usual cultivation system determined increasing sales in the next years with 5-10% per year at tomatoes, cucumbers, eggplant and other vegetables, especially where the farmers have problems with difficult conditions connected with infestation of soils by pesticides and pests and diseases. It is the case of Agris S.A. company as is presented (Table 1). It is the large number of seedlings demand on market for large-scale greenhouses and open-fields, of course part of this involve grafted seedling, for it is necessary a lot of hand work or semi or fully automated grafting robots. In case of Agris S.A. company the main market for seedlings is for eggplant (increase with 1731% in 2015 versus 2014), melon (increase with 162% in 2015 versus 2014), watermelon (increase with 26% in 2015 versus 2014), (Table 2).

Table 1

Evolution of Agris S.A. company's sales of in Romania

PRODUCT GROUP SEEDLINGS	2015	2014	2013	2012	2011
watermelon	1.558.374	1.222.050	989.645	436.246	362.314
greenhouse tomato	482.408	459.041	449.858	40.467	39.665
melon	59.668	37.947	28.106	33.803	18.357
eggplant	18.165	152.478	7.700	8.741	2.998
pepper	41.558	161.717	24.416	10.088	1.686
open field tomato	19.993	12.725	6.243	1.914	0
short cucumber	999	1.529	392	546	588
lettuce		5.580	4.215	1.881	1.080
broccoli		0	3.069	3.447	0
long cucumber	276	482	1.809	38.299	0
cauliflower		1.245	12.045	9.465	1.125
cabbage		0	53.760	123.231	486
TOTAL	2.181.441	2.054.794	1.581.258	708.128	428.299

Many seedling nursery companies, institutes and universities carry out studies on the seedling production, rootstock breeding, facilities development, plant physiology and molecular biology in relation to vegetable grafting. At least 40 cultivars of rootstock for the vegetable grafting have been bred and released, a series of grafting robots have been developed, and some scientific results have been published in international journals. Grafting may increase resistance to stress factors such as temperature and water and pathogen attack (key diseases *Pythium*, *Rhizoctonia*, *Phytophthora*, *Fusarium*, etc.), grafted crops yield and the use of organic chemical fertilizers as well as water is higher than the check culture. Price of grafted seedlings for fresh market is higher than classical seedling (excluding seed costs), it is the result of intensive labour input for propagation, a longer production period, and



the additional costs of the rootstock. Those expenses often discourage potential users of grafted seedlings. Not often acknowledged is that growers may be compensated for the greater initial cost of buying grafted seedlings by additional benefits of increased yield and reduced cost of control measures for soil borne pests.

Table 2

## Sales of Agris S.A. company in Romania

general category	product group	category	Value sales in 2015 versus targets value %	Values sales in 2015 versus sales in 2014 %
grafted plants	watermelon	grafted	-2%	26%
	greenhouse tomato	grodan	-21%	-6%
		grafted	-98%	-98%
	melon	grafted	102%	162%
	eggplant	grafted	1331%	1731%
	long cucumber	grafted		
<b>total - grafted plants</b>			<b>-11%</b>	<b>12%</b>
regular plants	greenhouse tomato	ungrafted	-96%	5%
		grodan	-100%	-100%
	pepper	ungrafted	-95%	-94%
	open field tomato	ungrafted	-45%	-29%
	eggplant	ungrafted	-99%	-99%
	melon	ungrafted	-78%	-72%
	short cucumber	ungrafted		66%
	watermelon	ungrafted	-100%	-100%
	long cucumber	ungrafted		
<b>total - regular plants</b>			<b>-96%</b>	<b>-90%</b>
vegetable seeds	rootstock	seeds	-59%	27%
	various vegetables	seeds	-31%	142%
	greenhouse tomato	seeds	-52%	4%
	squash	seeds		
	carrot	seeds	-88%	-36%
	open field tomato	seeds	-93%	-58%
	onion	seeds		
	processing tomato	seeds	-100%	
	pepper	seeds		
	melon	seeds		
<b>total - vegetable</b>			<b>-63%</b>	<b>35%</b>
<b>general total</b>			<b>-29%</b>	<b>0%</b>

In generally it was an decreasing of nemathodes and soil born diseases from 0-5,5%, in average 3.7%, for all Romanian greenhouses were firm's seedling were sold.

## CONCLUSIONS

1. In Romania, in the past, the major management strategy was pesticide control, however, many plant pathogen, insect and mite pests are resistant to registered pesticides and few new pesticides are being developed.

2. Introducing grafted vegetables into usual cultivation system determined increasing sales, in case of Agris S.A. company the main market for seedlings is for eggplant (increase with 1731% in 2015 versus 2014), melon (increase with 162% in 2015 versus 2014), watermelon (increase with 26% in 2015 versus 2014).

3. In generally it was an decreasing of nemathodes and soil born diseases from 0-5,5%, in average 3.7%, for all Romanian greenhouses were firm's seedling were sold.

**Acknowledgments:** Thanks, in this way, to the company Agris S.A. Seeds – SeedlingS Klidi, 59032 Imathia Greece, for its support in realising experiments and permission to use the data obtained. This paper is a part from PN II project No. 121/2012 „SIOPTEF”.

## REFERENCES

1. Cao Z.P., Chen G.K., Chen Y.F., Yang H., Han L.F., Dawson R., 2005 - Comparative performance of nematode resistant rootstock and non-resistant tomato cultivars on soil biota. *Allelopathy Journal*, 15: 85–94;
2. Cohen R., Pivonia S., Berger Y., Edelstein M., Gamliel A., Katan J., 2000 - Toward integrated management of *Monosporascus* wilt of melons in Israel. *Plant Dis.* 84: 496–505;
3. Igarashi I., Kano T., Kawabe T., 1987 - Disease and pest resistance of wild *Cucumis* species and their compatibility as rootstock for melon, cucumber and watermelon. *Bull. Natl. Res. Inst. Veg. Ornam. Plants and Tea Japan*. A1, pp. 173–185;
4. Ioannou N., 2001 - Integrating soil solarization with grafting on resistant rootstocks for management of soil-borne pathogens of eggplant. *J. Hort. Sci. Biotech.* 76: 396–401.
5. Kubota Ch., McClure M. A., Kokalis-Burelle Nancy, Bausher M. G., Roskopf E. N., 2008 - Vegetable Grafting: History, Use, and Current Technology Status in North America. *HortScience*, 43(6): 1664-1669;
6. Tateishi K., 1927 - Grafting watermelon on squash. *Japan. J. Hort.*, 39: 5–8;
7. Sigüenza C., Schochow M., Turini T., Ploeg A., 2005 - Use of *Cucumis metuliferus* as a rootstock for melon to manage *Meloidogyne incognita*. *J. Nematol.* 37: 276–280;
8. Shi Shenghan, 1959 - *Book of Fan Shengzhi shu jing shi*, Peking Science Press;
9. Williamson V.M., 1998 -Root-knot nematode resistance genes in tomato and their potential for future use. *Annu. Rev. Phytopathol.* 36: 277–293.

## THE ALLELOPATHY RELATIONS' IMPORTANCE IN DEVELOPING TECHNOLOGIES FOR INTERCROPPING CULTIVATION SYSTEM

### IMPORTANȚA RELAȚIILOR ALELOPATICE ÎN ELABORAREA TEHNOLOGIILOR DE CULTIVARE ÎN SISTEM INTERCROPPING

**HAMBURDĂ Silvia Brîndușa<sup>1</sup>, MUNTEANU N.<sup>1</sup>, STOLERU V.<sup>1</sup>, TELIBAN G.C.<sup>1</sup>, GALEA (DELEANU) Florina-Maria<sup>1</sup>, SAVA-PAVĂL Simona<sup>1</sup>, COJOCARU AL.<sup>1</sup>**

**e-mail:** silvia\_hamburda@yahoo.com

**Abstract.** *This paper presents an overview of the knowledge of allelopathy relations based on a large Romanian and international bibliography. The species to which particular reference has been made is the runner bean (Phaseolus coccineus L.). There were highlighted both the companion plants as well as non-companion plants with runner bean species. Also, a range of plants that have allelopathy effects was presented. Allelopathy relations have a very important role in developing cultivation technologies, because the combination of improper species leads to crop compromise, respectively considerable financial losses.*

**Key words:** *allelopathy, companion plants, runner bean.*

**Rezumat.** *Lucrarea prezintă o sinteză asupra cunoștințelor referitoare la relațiile alelopatice, pe baza unei ample bibliografii din țară și străinătate. Specia la care au fost făcute referiri în special a fost fasolea mare (Phaseolus coccineus L.). Au fost puse în evidență atât plantele companion cât și plante non-companion cu această specie. De asemenea, au fost prezentate o serie de plante care prezintă proprietăți alelopatice. Relațiile alelopatice au un rol foarte important în elaborarea tehnologiilor, deoarece asocierea necorespunzătoare a unor specii duce la compromiterea culturilor, respectiv pierderi financiare considerabile.*

**Cuvinte cheie:** *alelopatie, plante companion, fasole mare.*

## INTRODUCTION

Allelopathy is a natural phenomenon that belongs to a field of organic biochemistry and studying interrelationships between plants via chemical messages (signals) (Neamțu, 1983). Between different plant species, and between individuals within the same species, allelopathy effects occur.

Chemical compounds involved in the biochemical interactions between plants are generically called allelopathy substances. They are secondary products of plants, with low molecular weight, present in different organs of many plant

---

<sup>1</sup>University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

species such as leaves, flowers, fruits and seeds, but also in stems and roots, especially those which are perennial (Miró *et al.*, 1998; Delachiave, 1999). These allelochemical products have no role in primary metabolic processes, essential for plant survival, and are produced as consequences of primary metabolic pathways. In contrast to primary metabolism, which comprises several hundred molecular compounds, with low weight, tens of thousands of secondary products of metabolism are known, characteristic to plant species, but only a limited number of them are involved in allelochemical processes (Rice, 1984), at least at the current level of knowledge.

According to the literature review, there are reported both favourable associations among crops by intercalation or succession (Indrea, 1967; Aubert, 1975; Dumitrescu and Conea, 1986; Renaud and Dudouet, 1988; Stein, 1995; Toncea, 2002) and unfavourable associations (incompatibility) between related species (Stein, 1995; Toncea, 2002; Furnea, 2010).

The paper presents allelopathy relations' role in developing technologies for the intercropping cultivation system. The need for such research stems from the fact that allelopathy relations have a particular influence on crops and an improper association can ultimately lead to compromising crops or financial losses.

## MATERIAL AND METHOD

The biological material to which assessments are made on allelopathy relations is runner bean (*Phaseolus coccineus* L.) species. Runner bean is an annual herbaceous species that usually multiplies by seeds, but in some cases by tuberized roots (Munteanu, 1985). In this study we propose to consider companion and non-companion plants with bean, respectively a number of plants with allelopathy properties.

Reaching the goal and targets is carried out based on a literature review. As basic research methods we have used observation, case studies and statistics group.

## RESULTS AND DISCUSSIONS

Allelopathy compounds affect cell division of competing plants, phytohormones' activity, operating efficiency of the chloroplasts and mitochondria, biomarkers, the functions of biomembranes, the plant-water relationship and various other plant processes (Einhellig, 2002; Blum *et al.*, 1999; Macias *et al.*, 1999). Other important aspects of allelopathy, in addition to the production and release into the environment of phytotoxic compounds to certain crop species, aim at the absorption and translocation of allelochemical substances in the recipient organism (Ferreira and Aquila, 2000).

Intensive scientific research for recognition and understanding of allelopathy has only been done in the last few decades, although the allelopathy phenomenon existed for thousands of years. In 1832, the idea of an influence exerted by certain chemical substances released into the environment by certain organisms to other neighboring bodies was issued by De Candolle (Harborne,

1977). De Candolle concluded that all the plants secrete, through the roots, certain substances that stimulate or inhibit the growth of other plants.

In 1937, a step forward was made by Molisch, when he published his research on ethylene action on some superior plants, a phenomenon that he called "*allelopathy*". He is the one who has defined the term allelopathy for the first time (Molisch, 1937). Allelopathy meant, at that time, "biochemical relations established between all the plants", the same concept including both harmful interactions and those which are stimulative. The term "allelopathy" derives from the Greek words "*allelon*" = mutual and "*pathos*" = suffering. It rooted in literature, becoming one of the branches of the modern eco-physiological biology.

A real breakthrough in allelopathy was produced in 1974, with the publication of "Allelopathy" by Rice. It defined allelopathy as "detrimental effect exerted by a plant on another plant by producing chemicals that are released into the environment." Advances in chemistry, anatomy and physiology, in plant biotechnology and digital technology, as well as a better understanding of ecological processes, helped allelopathy research develop lately.

Contemporary researchers have tended to broaden the alelopathy context and also include in this phenomenon interactions between plants and animals (Rizvi and Rizvi, 1992), and suggested that allelopathy is a part of a whole network of chemical communication between plants, between plants and other organisms and that such communication may contribute to plant defense.

The commonly accepted definition of alelopathy is the production of biomolecules by plants; these molecules consist mainly of secondary metabolites that may beneficially affect another plant, or vice versa (Rizvi and Rizvi, 1992).

The over time accumulated data showed that allelopathy substances can have both an inhibitor or a stimulating role. Allelopathy substances with inhibitor character are widely distributed in nature, as highlighted in the desert plants, in the wetlands and forest ecosystems plants in temperate regions. Long studied allelopathy substances with a stimulating role are the vitamins.

The presence of weeds and debris into the soil in the early stages of germination and growth of vegetable plants may hamper crop through allelopathy phenomena, caused by the presence of toxic substances released from underground organs in the soil that come through volatilization and secretion or debris due to decomposition of residues thereof. These phenomena, poorly understood until now, of mutual influence can be destructive to crop plants, against some weed or to weeds among each other. Currently, allelopathy phenomena of species *Poa* (bluegrass) on crop of tomatoes, *Portulaca oleracea* (watergrass) on peas, *Artemisia vulgaris* (mugwort) on cucumbers are known (Indre *et al.*, 2009).

Also, there are other plant species with allelopathy properties, such as sorghum, poppy, mugwort, nut, etc.

The genus *Sorghum* (*sorghum*) includes sorghum plants, whose roots eliminate a poisonous substance that blocks respiration and photosynthesis of plants it comes in contact with.

For cereals, it is good to know that the seeds of poppy (*Papaver* sp.) do not germinate without the presence of wheat, while rye (*Secale cereale*) prevents certain types of weed germination.

Wormwood (*Artemisia absinthium* L.) inhibits the growth of plants such as lovage, cumin, basil, lemon balm and sage.

Black walnut (*Juglans nigra* L.) is one of the most famous plants with allelopathy properties. It secretes juglone, a substance that inhibits respiration in plants located in its vicinity. Since juglone is also present in the black walnut tree and roots, which can handle a generous area, the garden area affected by allelopathy could be quite large (<http://gradina.acasa.ro/boli-si-daunatori-116/alelopatia-in-gradina-164583.html#ixzz2YiRTtP3T>).

Given the above, in order to organize a garden, farm or agricultural land, association with optimal plants, in order to obtain maximum yield with minimum costs, if possible, and without the use of petrochemicals, with as little mechanization as possible should be considered (<http://forum.softpedia.com/lofiversion/index.php/t752500.html>).

The concept of "companion plants" is especially common in small and organic gardens, relying on the idea that plants can benefit from each others' company. This consists in the operation of interconnection, in order to ensure the growth and development of plants and to replace the use of chemical herbicides and insecticides, organically control and prevention of pests, attracting birds, insects and other useful animals (<http://www.vegetalshapes.com/Romana/PlanteCompanionI.html>) (Fig. 1).



**Fig. 1** - Associated crops

(<https://gradinadeacasa2.wordpress.com/2013/08/29/cultura-asociata/>)

Among the companion plants for runner bean we include: anise, asparagus, basil, potatoes, cucumbers, strawberries, zinnias, thyme, yarrow, coriander,



cosmos, chrysanthemums, bay leaves, pumpkin, squash, corn, spinach, rosemary, dill, carrots, celery, borage, lettuce, lovage, radishes, etc. (<http://deepgreenpermaculture.com/companion-planting/companion-planting-table/>) (Fig. 2).



**Fig. 2** - Runner bean in an associated crop  
(<http://www.vegetable-gardening-online.com/growing-runner-beans.html>)

Among non-companion plants for runner bean we mention: peppers, chives, onions, cauliflower, garlic, leeks, tomatoes, chili peppers, fennel, gladiolas, beets etc. (<http://deepgreenpermaculture.com/companion-planting/companion-planting-table/>).

## CONCLUSIONS

1. Allelopathy relations have a very important role in developing technologies for cultivation in intercropping system, as the combination of improper species leads to crop compromise, respectively financial losses.

2. Runner bean is a vegetable species that can be successfully associated with many species, but presents a number of non-companion plants, including chili peppers, onion, garlic etc.

***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.*

## REFERENCES

1. Aubert Cl., 1975 - *Le jardin potager biologique*. Le courrier de livre, Paris.
2. Blum R., Pfeiffer, F., Feick, P., Nastainczyk, W., Kohler, B., Schäfer, K.H. and Schulz, I., 1999 - *Intracellular localization and in vivo trafficking of p 24A and p 23*. J. Cell Sci. 112, 537-548.
3. Delachiave M.E.A., Rodrigues J.D., Ono E.O., 1999 - *Efeitos alelopático de Losna (Artemisia absinthium L.) na germinação de sementes de pepino, milho, feijão e tomate*. Revista Brasileira de Sementes, 21 (2):265-269.

4. Dumitrescu M., Conea Al., 1986 - *Aspecte tehnico-economice ale folosirii intensive a terenurilor irigate cultivate cu legume*. Horticultura, 7.
5. Einhellig F.A., 2002 - *The Physiology of Allelochemical Action: Clues and Views*. In: Allelopathy, From Molecules to Ecosystems. Ed. Reigosa, M., Pedrol, N., Departamento de Biología Vegetal e Ciencia del Suelo, Universidad de Vigo, Vigo, Spain, p. 1-17.
6. Furnea (Condor) Cornelia Cristina, 2010 - *Contribuții la studiul comportării unor specii de legume rădăcinoase în culturi succesive și asociate*. Teză de doctorat. USAMV Cluj-Napoca.
7. Harborne J.B., 1977 - *Introduction to ecological biochemistry*. Edit. Acad. Press, London, New York.
8. Indrea D., 1967 - *Cercetări privind agrobiologia salatei și fasolei de grădină în vederea sporirii eficienței lor în culturi succesive*. Teză de doctorat. I.A.N.B., București.
9. Indrea D., Apahidean S. Al., Apahidean Maria, Măniuțiu D.N., Sima Rodica, 2009 - *Cultura legumelor*. Editura Ceres, București.
10. Macias F.A., Galindo J.C.G., Molinillo J.M.G., Castellano D., Velasco R.F., Chinchilla D., 1999 - *Developing new herbicide models from allelochemicals*. Pesticide Science. 55: 633-675.
11. Miró C.P., Ferreira A.G., Aquila M.E.A., 1998 - *Alelopatia de frutos de erva-mate (Ilex paraquariensis) no desenvolvimento do milho*. Pesquisa Agropecuária Brasileira 33 (8): 1261-1270.
12. Molisch H., 1937 - *Der Einfluss einer Pflanze auf die andere – Allelopathie*. În: Introduction to Ecological Biochemistry. Ed. Harborne, J.B., 9, Acad. Press, London – New York – San Francisco, 1977, p. 178-179.
13. Munteanu N., 1985 - *Phaseolus coccineus L. – o specie legumicolă care merită mai multă atenție*. Producția vegetală. Horticultura, nr. 4: 17-19.
14. Neamțu G., 1983 - *Interacțiuni biochimice între plantele superioare*. Biochimie ecologică. Editura Dacia, Cluj-Napoca, p. 146-160.
15. Renaud V., Dudouet Chr., 1988 - *Le potager par les methodes naturelles*. Ed. Dudouet, Magny-Cours, France.
16. Rice E.L., 1984 - *Allelopathy 2<sup>nd</sup>*. Editura Academic Press, New York, p. 421.
17. Rizvi S.J.H., Rizvi V., 1992 - *Exploitation of allelochemicals in improving crop productivity*. Rizvi, S.J.H. & Rizvi, V. (Eds.). Allelopathy: basic and applied aspects. London, Chapman & Hall. pp. 443-472.
18. Stein S., 1995 - *Gemüse*. B. L. V. München, Wien, Zürich, p. 21.
19. Toncea I., 2002 - *Ghid practic de agricultura ecologica*. Editura Academicpres, Cluj – Napoca. 170 p.
20. <http://deepgreenperma culture.com/companion-planting/companion-planting-table/>
21. <http://forum.softpedia.com/lofiversion/index.php/t752500.html>
22. <http://gradina.acasa.ro/boli-si-daunatori-116/alelopatia-in-gradina-164583.html#ixzz2YiRTtP3T>
23. <http://www.vegetalshapes.com/Romana/PlanteCompanionI.html>.
24. <http://www.vegetable-gardening-online.com/growing-runner-beans.html>.



## PRELIMINARY STUDIES REGARDING THE ESTABLISHMENT OF AN ORNAMENTAL GARDEN IN A GEOMETRICAL STYLE USING VEGETABLE PLANTS IN INTERCROPPING SYSTEM

### STUDII PRIVIND REALIZAREA UNEI GRĂDINI ORNAMENTALE ÎN STIL GEOMETRIC UTILIZÂND PLANTE LEGUMICOLE ÎN SISTEM DE INTERCROPPING

**GALEA (DELEANU) Florina-Maria<sup>1</sup>,  
MUNTEANU N.<sup>1</sup>, HAMBURDĂ Silvia-Brîndușa<sup>1</sup>**  
e-mail: florinagalea@gmail.com

**Abstract** This paper presents a literature review of the species *Lactuca sativa* L. regarding its decorative characters and productive potential, in association with other vegetables, based on morphological, physiological and technological characteristics. Based on these characteristics, the geometric display of the species and the possibility of growing them in intercropping system was elaborated. The studied species are two tips of lettuce, Lollo Rossa and May King, in intercropping systems with spinach, red orache, onions and lavender. The obtained results were congruent with the ones that specialized literature offered, the needed ornamental valences of an aesthetic family vegetable garden are offered by the decorative characters of the chosen species.

**Key words:** Classic garden style, Vegetables, Production.

**Rezumat.** Lucrarea prezintă o sinteză a literaturii de specialitate referitoare la caracterele decorative și cele ale potențialului productiv ale speciei *Lactuca sativa* L. în asociere cu alte plante legumicole, pe baza principalelor caracteristici morfologice, fiziologice și tehnologice ale acestei specii. Pe baza acestora a fost elaborată dispunerea în stil geometric a speciilor alese și posibilitatea cultivării lor în sistem de intercropping. Au fost luate în studiu două soiuri de salată, Lollo Rossa și May King, asociate cu spanac, lobodă roșie, ceapă și levănțică. Rezultatele obținute au fost în concordanță cu cele întâlnite în literatura de specialitate, caracterele decoartive ale speciilor alese oferind valențele ornamentale necesare unei grădini legumicole estetice în cadrul familial.

**Cuvinte cheie:** asociere, grădină decorativă, producție.

## INTRODUCTION

Humans began practicing agriculture 15-20.000 years ago, close to the end of the Neolithic period, until then they were hunters and gatherers. The first step in becoming a farmer was to tame and grow plants; historically speaking this

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine Iași, Romania

represents the start of the first agrarian revolution. Through out time, as agriculture evolved, man has continued to enhance, and to perfect, the way that plants are cultivated, so that in our days the evolution of scientific and technical knowledge allows us to see the prominence in the agricultural systems.

According to their degree of development, we can observe systems that range from simple to complex and modern, with a great degree of intensification and usage of resources, such as precise agriculture and smart agriculture.

In the practice of vegetable growing, in these circumstances, from the beginning of time, plants were grown blended, associated or in intercropping system. A modern intercropping system, much used in our days, must be defined from a biological and ecological point of view, in the same time we must enhance as many crop valances as we can, including ornamental ones. Thus we reached the practice and concept of vegetable gardens.

The ornamental character of vegetable gardens aroused at first in the court of noble palaces and monasteries. Among the most noted and beautiful vegetable gardens, that passed the test of time, can be found in France. The court of Villandry Castle is renowned for its aesthetic and playful design, in which vegetables are presented in different stages of their development (Kluckert, 2007).

The ornamental vegetable garden can be found within the family yard, and it must carry out many conditions, from the financial one, to the nutritive and aesthetic ones. Vegetable gardens were a part of individual yards from ancient time. The decorative character of this landscape projects emerged through time, with different titles. The concept of edible landscaping belongs to Robert Kourik, known ecologist and american writer, this concept was then adopted by the renowned architect Rosalind Creasy. Nowadays the concept has suffered a widening of its significance, and it is used both in family gardens and in public establishments. The widening of the concept has permitted its ramification, from growing ornamental species with some vegetables, to ornamental designs only with vegetables (Sima, 2009).

Regarding the possibility of exploitation of the accumulated knowledge of an ornamental vegetable garden in a family setting, we consider of interest to synthesize this knowledge and to create an ecological and biological database which will allow to create this type of gardens in a scientific manner. In our research we also have in sight their alimentary and therapeutic importance of the chosen species.

Due to its alimentary importance and its therapeutic effects *Lactuca sativa* L. is a necessary species that can't be absent from human alimentation. Lettuce is a rich species in vitamins (vit. A, vit. C, vit. PP), mineral salts (Ca, Fe, P, K) and nutritious substances (Stan *et al.*, 2003). From the traditional point of view, lettuce is known for its therapeutic goods, regarding ache treatments, digestive and inflammatory problems of the stomach, and in the same time for its antidepressant and antioxidant properties (Hammad and Bushra, 2015).

In this study we want to do a display of the steps that must be followed in order to realize an ornamental vegetable garden in a classic style, in an ecological system, by applying intercropping technology. The main objects of this study are to identify how to associate lettuce with other species, based on its biological and ecological requirements, and to increase its ornamental impact.

## MATERIAL AND METHOD

For the fulfillment of the established purpose and objectives, an experience was organized in the experimental field of the Vegetable growing department, at the USAMV farm V. Adamachi, in the year 2015.

The biological material used in the experience was: two varieties of lettuce (*Lactuca sativa* L.), May King and Lollo Rossa, two varieties of spinach (*Spinacea oleracea* L.), Matador and Modi F1 hybrid, a local population of red orache (*Atriplex hortense* L.), white communal onion (*Allium cepa* L.) and Common English lavender (*Lavandula angustifolia* Mill.).

The experience was realized on a 20 m<sup>2</sup> area, in a geometric style based on symmetry, balance and rhythm. The ornamental vegetable garden includes two rectangles in a mirror design. The rhythm of the establishment is given by the flowers made from lettuce, in a total of six, based on a game of chromatics and shape, with a total of 204 seedlings, from which 112 were May King variety and 92 Lollo Rossa. The distance between the rows of lettuce was of 20 cm and between plants 30 cm. The spinach was sown at the distance of 20 cm between rows, which were in number of three, between the flowers made of lettuce, with a total of twelve rows. In the outer corners of the garden design red orache seedlings were planted, while in the inner corners lavender was planted. The landscape design was outlined by an onion headboard, from white chive. The garden was established between 28.04-01.05

The experience was conducted according to crop technology found in literature review (Stan *et al.*, 2003; Sima, 2009; Ciofu *et al.*, 2003). The crop was established on leveled ground, well stocked with nutrients and organic matter, in an ecological system.

The applied research methods are bibliographic study, observation and experiment, based on which the performances of *Lactuca sativa* L., regarding ornamental and agricultural productivity characters, were evaluated.

## RESULTS AND DISCUSSIONS

Lettuce is grouped in three varieties, head lettuce, marula and leaf lettuce. *Lactuca sativa* L. is an annual plant, with a short vegetation period. It has a taproot system, with numerous side ramifications, but with a superficial developing in seedling. Due to the different characters that the varieties of lettuce present, size, form and color of leafs, vary from one kind of lettuce to another. The short period of vegetation gives us the freedom to establish this crop by using seedlings or seeds, thus lettuce is suitable in successive crops or intercropping system (Ciofu *et al.*, 2003).

May King lettuce is a part of the head lettuce variety, suitable for an open ground crop. This variety of lettuce formed medium sized head, which can be observed in Fig. 1.

Compared to May King lettuce, Lollo Rosso is a variety of leaf lettuce, with italian origin, that has a rich, well developed and compact rosette, with numerous ornamental characters. Leaves are green at the base and burgundy at the top, with an undulated lamina, that can be seen in Fig. 2.



**Fig. 1** - Lollo Rosso lettuce



**Fig. 2** - May King lettuce

Based on the ornamental characters of the two chosen varieties of lettuce and due to the positive effects that this species has on the human body, it represents an essential element in a vegetable garden at the beginning of the year.

Being a part of the vegetable greens group, lettuce was suitable in a successive and associated crop. The intercropping system presented numerous advantages, among them were, the increase of agro ecosystem diversity, reduced soil erosion and reduced degree of weeds. Beside this advantages, in the vegetable garden, there were also less pests or disease attacks and reduced quantity of water used in irrigation.

The ornamental vegetable garden presented in Fig. 3, was designed in an ecological system, by applying the technology of intercropping systems. It has a classic design, representing an attraction point and a spot of color in the same time, in a family garden.



**Fig. 3** - Ornamental vegetable garden

The varieties of lettuce, May King and Lollo Rossa developed well, in the metrological conditions during spring time, and presented specific variety characteristics. The biometric indicators represented by head and rosette diameter, height, weight and number of leafs, that can be seen in table 1, were compared with the result obtained by Hoza Gheorghita in his study regarding edible ornamental carpets, in which chemical fertilization NPK was used (Hoza, 2012). As oppose to that, in the case of intercropping system, with no fertilizer being applied, May King lettuce had an average diameter of 33,81 cm compared to 34,50. Lollo Rossa variety had an average diameter of 26,33 cm as opposed to 28,8. The average weight of the two varieties of lettuce was 242,55 g for Lollo Rossa and 444,36 g for May King.

Table 1

Biometric aspects of the edible part			
Type	Average diameter (cm)	Average weight (g)	Average height (cm)
Lollo Rossa	26,33	242,55	19,77
May King	33,81	444,36	16,36

The number of leafs for the two varieties was between 21-30 for Lollo Rossa and 35-48 for May King head lettuce.

The establishment of an ornamental vegetable garden has the purpose to decorate, but also the purpose to heal (hortitherapy garden) and to nourish. The association of vegetables with flowers creates color, shape and volume effects, integrating the vegetable garden in the family garden frame.

## CONCLUSIONS

1. By taking into consideration the biological and ecological needs of the species *Lactuca sativa* L., the plant was well developed.
2. The variation of form and color, given by the associated species in intercropping system, has increased the ornamental effect of the vegetable garden.
3. Growing ecologic lettuce in intercropping system has results in an increase of biodiversity and a decrease of weeds, in the vegetable garden.
4. By associating vegetables with flowers benefic insects were attracted.
5. The geometric style of the design has allowed the usage of a large number of plants on a relative small area.

## REFERENCES

1. Ciofu Ruxandra, Stan N., Popescu V., Chilom Pelaghia, Apahidean S., Horgoș A., Berar V., Lauer K.F., Atanasiu N., 2003 - *Tratat de legumicultură*. Ed. Ceres, București, pp. 900-908.
2. Hammad I., Bushra M., 2015 - *Evaluation of analgesic, anti-inflammatory, anti-depressant and anti-coagulant properties of Lactuca sativa (CV. Grand Rapids) plant tissues and cell suspension in rats*. Journal BMC Complementary and Alternative Medicine, 15:199.

3. **Hoza Gheorghița, 2012** - *Research regarding the exploitation of the ornamental potential of some lettuce varieties, by forming edible ornamental carpets*. Journal of Horticulture, Forestry and Biotechnology, Volume 16(2):90-95.
4. **Kluckert E., 2007** - *European garden design. From classical antiquity to the present day*. Ed. H.f. ullmann, Oxford, pp. 22, 119-123.
5. **Mousavi S.R., Eskandari E., 2011** - *A general overview on intercropping and its advantages in sustainable agriculture*. Journal of Applied Environmental and Biological Sciences, 1: 482-484.
6. **Sima Rodica, 2009** - *Legumicultura. Sursă de hrană și potențial ornamental*. Ed. AcademicPres, Cluj-Napoca, 21-22: 280-282.
7. **Stan N.T., Stan T.N., 1999**- *Legumicultură*. Ed. „Ion Ionescu de la Brad”, Iași, vol.I, pp. 7-9.
8. **Stan N.T., Munteanu N., Stan T.N., 2003** - *Legumicultură*, Ed. „Ion Ionescu de la Brad”, Iași, vol. III, pp.156-162, 171-173, 176-177.

## RESULTS OF RUNNER BEAN (*PHASEOLUS COCCINEUS* L.) YIELD OBTAINED IN INTERCROPPING SYSTEM

### REZULTATE DE PRODUCȚIE OBȚINUTE ÎN SISTEM INTERCROPPING LA FASOLEA MARE (*PHASEOLUS COCCINEUS* L.)

**HAMBURDĂ Silvia Brîndușa<sup>1</sup>, MUNTEANU N.<sup>1</sup>, STOLERU V.<sup>1</sup>, TELIBAN  
G.C.<sup>1</sup>, GALEA (DELEANU) Florina-Maria<sup>1</sup>, SAVA-PAVĂL Simona<sup>1</sup>,  
COJOCARU AL.<sup>1</sup>**

**e-mail:** silvia\_hamburda@yahoo.com

**Abstract.** *This paper presents the influence of the calendaristic establishment date on the yield (dry seeds) of runner bean (*Phaseolus coccineus* L.) cultivated in intercropping system, respectively intercropping with common maize, intercropping with sunflower and intercropping with Jerusalem artichoke, in three distinct calendaristic dates of establishment: 1.05, 15.05, 30.05. In order to achieve this goal, we intended to find out in which date the highest yield is obtained, by comparing the variants with each other. The crop establishment was performed by direct sowing in the field. The results revealed significant differences between the studied variants. The date in which the highest yields were achieved was 15.05. The variant which obtained the highest yield was the one where runner bean was interleaved with sunflower.*

**Key words:** *associated plants, setting up time, common maize, sunflower, Jerusalem artichoke*

**Rezumat.** *Lucrarea prezintă influența epocii de înființare a culturii asupra cantității de recoltă (boabe uscate) obținută la fasolea mare (*Phaseolus coccineus* L.) cultivată în sistem intercropping, respectiv intercropping cu porumb comun, intercropping cu floarea soarelui și intercropping cu topinambur legumicol, în trei epoci distincte de înființare a culturii: datele calendaristice 1.05, 15.05 și 30.05. Pentru realizarea scopului propus, ne-am stabilit să aflăm în care epocă și la care variantă se obține cantitatea cea mai mare de producție, prin compararea variantelor între ele. Înființarea culturii a fost realizată prin semănat direct, în câmp. Rezultatele au pus în evidență diferențe semnificative între variantele studiate. Epoca de înființare în care au fost obținute cele mai mari rezultate de producție a fost epoca 15.05. Varianta la care au fost obținute cele mai mari cantități de recoltă a fost varianta în care fasolea mare a fost intercalată cu floarea soarelui.*

**Cuvinte cheie:** *plante asociate, epoca, porumb comun, floarea soarelui, topinambur legumicol*

## INTRODUCTION

Runner bean is a tropical species adapted to the humid highlands. Compared with common bean, it is a thermophilic species that tolerates well

---

<sup>1</sup>University of Agricultural Sciences and Veterinary Medicine of Iași, Romania



relatively low temperatures. Starting at 8-12<sup>0</sup>C, germination occurs, the optimum temperature for growth and development being 16-18<sup>0</sup>C (Popa, 2010).

Establishment of runner bean crop is one of the most important stages in a culture technology. In principle, it includes operations like: selection, procurement and preparation of biological material, setting the era, the choice of crop system (rows, nests, mixture, plant association), scheme and densities.

Setting up time is conditioned by the cultivation system, runner bean biological peculiarities and the period when the yield should be harvested. The main factors to be taken into account in determining sowing time are climatic conditions, particularly thermal conditions and soil type. As in common bean, setting up scheme is extremely variable, according to tradition and managerial, technical or material possibilities. The main technical element that determines the crop establishment scheme is the trellising method.

Trellising system is a key issue for the success of the crop. In vegetable growing and also in other areas, such as wine growing, trellising system has a very important role on growth, development and crop production. Any trellising system aims to provide a support for the plants so that the goal of yield can be achieved (Branas, 1974; Champagnol, 1984).

The pure culture is simpler, compared to the intercropping/associated system, but fails to clearly modify the environmental conditions of the crop. Instead, intercropping system, although it seems complicated, provides some improvement in microclimate and, also, by ensuring the runner bean support system by associated / intercropping crops.

Intercropping has been known for thousands of years (Kass, 1978) and it is the system in which, on the same area of the field and at the same time, two or more species are grown (Andrews and Kassam, 1976; Anil *et al.*, 1998; Ofori and Stern, 1987).

The purpose of this paper is to establish the setting up time influence on the amount of crop (dry seeds) obtained from runner bean, grown in intercropping system. To achieve this goal, we set targets to find the best setting up time and the intercropping variant where the highest yields are obtained.

## MATERIAL AND METHOD

The research took place in the "Vasile Adamachi" farm of the University of Agricultural Sciences and Veterinary Medicine, in an experimental polygon of the Vegetable Growing department.

The biological material was represented by the local population Coccineus 3 of runner bean, hybrid cultivar Flato F1 of common maize, hybrid cultivar Tristan F1 of sunflower and clone cultivar Topstar of Jerusalem artichoke.

The studied experimental factor was the crop setting up time, with three graduations: dates of 1.05., 15.05. and 30.05, available in three plants arrangements, namely intercropping with common maize (*Zea mays* L.) (V<sub>1</sub>), intercropping with sunflower (*Helianthus annuus* L.) (V<sub>2</sub>) and intercropping with Jerusalem artichoke (*Helianthus tuberosus* L.) (V<sub>3</sub>).

The placement of the experimental factor, as well as its graduations, was carried out in sub divided triplicate plots.



On each repetition plot, two rows were placed, spaced at 1.0 m, and between the runner bean plants, on row, there was a distance of 0.4 m. Between associated plants (common maize, sunflower, Jerusalem artichokes ) there was a distance of 0.8 m.

Common maize and sunflower sowing, respectively planting Jerusalem artichoke, were made about two weeks before sowing the runner bean. Crop establishment was performed by direct sowing, with three runner bean seeds / nest and two seeds of common maize, respectively two seeds of sunflower / nest. In the time of emergence, two runner bean plants and one plant of common maize or sunflower have been left in each nest. For Jerusalem artichoke two tubers / nest were planted and at emergence only two stems / nest were left.

The experiment was conducted according to technological norms arising from the literature review (Munteanu *et al.*, 1989; Stan *et al.*, 2003; Ruști, 2007; Popa, 2010; Axinte *et al.*, 2006). Basic research methods were observation and experiment.

Yield data were processed by the algorithm for ANOVA (analysis of variance) using Fisher's exact test, Student's t test, the least significant difference (LSD) for three levels of confidence: LSD 5% (P = 95%), LSD 1% (P = 99 %) and LSD 0.1% (P = 99.9%) (Săulescu și Săulescu, 1967; Jităreanu, 1999).

## RESULTS AND DISCUSSIONS

### Runner bean yield results obtained in intercropping with common maize (V<sub>I</sub>)

Runner bean yield ranged from 1383 kg/ha to 2498 kg/ha. The highest yield was obtained in 15.05 setting up time, with a value of 2498 kg/ha, registering very significant positive differences compared to the average experience (1966 kg/ha), while the lowest yield was obtained in 01.05 setting up time, highlighted the very significant negative differences from the average. In 30.05 setting up time, yield was within the variation of the experimental mean, respectively 2017 kg/ha (Tab. 1; Fig. 1).

Table 1

Runner bean yield results obtained in intercropping with common maize

No.	Setting up time	Yield (dry seeds)		Yield differences between setting up time and their significance <sup>w</sup>			
		kg/ha	% of the $\bar{x}$	01.05 setting up time	15.05 setting up time	30.05 setting up time	mean ( $\bar{x}$ )
1.	01.05.2013	1383	70	-	-1115 <sup>ooo</sup>	-634 <sup>ooo</sup>	-583 <sup>ooo</sup>
2.	15.05.2013	2498	127	+1115 <sup>***</sup>	-	+481 <sup>***</sup>	+532 <sup>***</sup>
3.	30.05.2013	2017	103	+634 <sup>***</sup>	-481 <sup>ooo</sup>	-	+51 <sup>NS</sup>
4.	Mean ( $\bar{x}$ )	1966	100	+583 <sup>***</sup>	-532 <sup>ooo</sup>	-51 <sup>NS</sup>	-

<sup>w</sup>Significance of differences made by ANOVA (analysis of variance) for experimental factors and interaction of them; NS, \*, \*\*, \*\*\* - indicate nonsignificant and positive significant at p ≤ 0.05, 0.01, 0.001, respectively;

ooo - negative significant at p ≤ 0.05, 0.01, 0.001, respectively

LSD 5% = 99,1 (kg/ha); LSD 1% = 164,3 (kg/ha); LSD 0,1% = 306,9 (kg/ha).

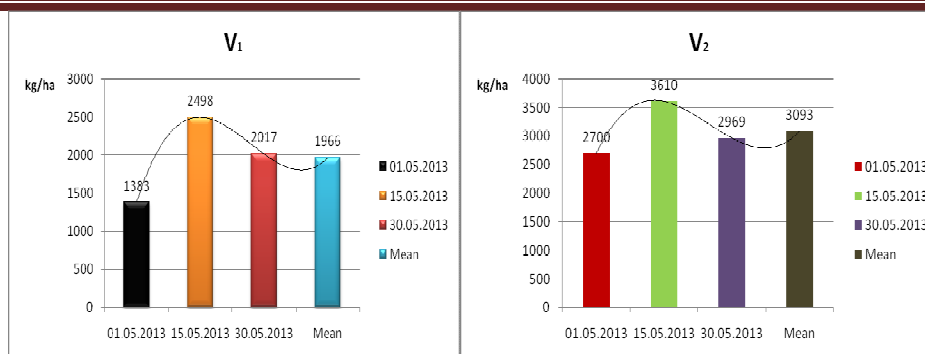


Fig. 1 - Yield graphic representation (V<sub>1</sub>)      Fig. 2 - Yield graphic representation (V<sub>2</sub>)

### Runner bean yield results obtained in intercropping with sunflower (V<sub>2</sub>)

From the conducted researches, it appears that runner bean yield ranged from 2700 kg/ha to 3610 kg/ha. The highest yield was obtained in 15.05 setting up time, with a value of 3610 kg/ha, registering positive differences significantly distinct from the average experience (3093 kg/ha), while the lowest yield was obtained in 01.05 setting up time, highlighted distinct significant negative differences from the average. In 30.05 setting up time yield was 2969 kg/ha, being within the average experimental variation limits (Tab. 2; Fig. 2).

Table 2

Runner bean yield results obtained in intercropping with sunflower

No.	Setting up time	Yield (dry seeds)		Yield differences between setting up time and their significance <sup>w</sup>			
		kg/ha	% of the $\bar{x}$	01.05 setting up time	15.05 setting up time	30.05 setting up time	mean ( $\bar{x}$ )
1.	01.05.2013	2700	87	-	-910 <sup>ooo</sup>	-269 <sup>o</sup>	-393 <sup>oo</sup>
2.	15.05.2013	3610	117	+910 <sup>***</sup>	-	+641 <sup>**</sup>	+517 <sup>**</sup>
3.	30.05.2013	2969	96	+269 <sup>*</sup>	-641 <sup>oo</sup>	-	-124 <sup>NS</sup>
4.	Mean ( $\bar{x}$ )	3093	100	+393 <sup>**</sup>	-517 <sup>oo</sup>	+124 <sup>NS</sup>	-

<sup>w</sup>Significance of differences made by ANOVA (analysis of variance) for experimental factors and interaction of them; NS, \*, \*\*, \*\*\* - indicate nonsignificant and positive significant at  $p \leq 0.05$ , 0.01, 0.001, respectively;

o,oo,ooo - negative significant at  $p \leq 0.05$ , 0.01, 0.001, respectively

LSD 5%= 221,8 (kg/ha); LSD 1%= 367,8 (kg/ha); LSD 0,1%= 686,9 (kg/ha).

### Runner bean yield results obtained in intercropping with Jerusalem artichoke (V<sub>3</sub>)

From the conducted researches, it appears that runner bean yield ranged from 545 kg/ha to 1065 kg /ha. The highest yield was obtained in 15.05 setting up time, with a value of 1065 kg/ha, registering significantly positive differences from the average experience (789 kg / ha), while the lowest yield was obtained in 01.05 setting up time, being within the average experimental variation limits. In 30.05 setting up time, the yield was 757 kg/ha, within the average experimental variation limits (Tab. 3; Fig. 3).

Table 3

## Runner bean yield results obtained in intercropping with Jerusalem artichoke

No.	Setting up time	Yield (dry seeds)		Yield differences between setting up time and their significance <sup>w</sup>			
		kg/ha	% of the $\bar{x}$	01.05 setting up time	15.05 setting up time	30.05 setting up time	mean ( $\bar{x}$ )
1.	01.05.2013	545	69	-	-520 <sup>oo</sup>	-212 <sup>NS</sup>	-244 <sup>NS</sup>
2.	15.05.2013	1065	135	+520 <sup>**</sup>	-	+308 <sup>*</sup>	+276 <sup>*</sup>
3.	30.05.2013	757	96	+212 <sup>NS</sup>	-308 <sup>o</sup>	-	-32 <sup>NS</sup>
4.	Mean ( $\bar{x}$ )	789	100	+244 <sup>NS</sup>	-276 <sup>o</sup>	+32 <sup>NS</sup>	-

<sup>w</sup>Significance of differences made by ANOVA (analysis of variance) for experimental factors and interaction of them; NS, \*, \*\*, \*\*\* - indicate nonsignificant and positive significant at  $p \leq 0.05$ , 0.01, 0.001, respectively;

o,oo,ooo - negative significant at  $p \leq 0.05$ , 0.01, 0.001, respectively

LSD 5%= 246,3 (kg/ha); LSD 1%= 408,5 (kg/ha); LSD 0,1%= 762,9 (kg/ha).

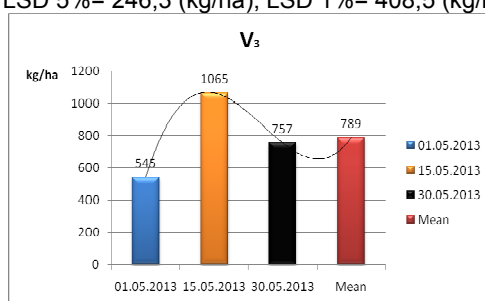
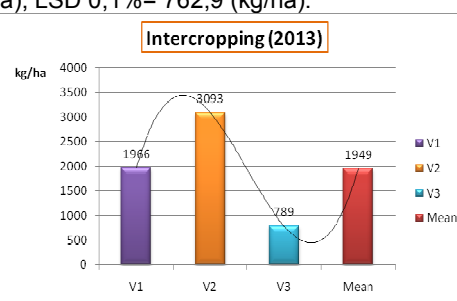
Fig. 3 - Yield graphic representation ( $V_3$ )

Fig. 4 - Yield graphic representation (intercropping, 2013)

## Total experiment

Table 4

## Runner bean yield obtained in intercropping (2013)

Variant			Runner bean yield (dry seeds)		Differences from the mean (kg/ha)	Signification <sup>w</sup>
No.	specification		kg/ha	% of the mean		
1	intercropping	V <sub>1</sub>	1966	101	+17	NS
2		V <sub>2</sub>	3093	159	+144	**
3		V <sub>3</sub>	789	40	-1160	oo
Mean ( $\bar{x}$ )			1949	100	-	-

<sup>w</sup>Significance of differences made by ANOVA (analysis of variance) for experimental factors and interaction of them; NS, \*, \*\*, \*\*\* - indicate nonsignificant and positive significant at  $p \leq 0.05$ , 0.01, 0.001, respectively;

o,oo,ooo - negative significant at  $p \leq 0.05$ , 0.01, 0.001, respectively

LSD 5%= 383,8 (kg/ha); LSD 1%= 636,4 (kg/ha); LSD 0,1%= 1188,6 (kg/ha).

Runner bean yield varied between 789 kg/ha and 3093 kg/ha. The highest yield was obtained in intercropping with sunflower (3093 kg / ha), which recorded positive differences distinct significantly from the average experience (1949 kg/ha), while the lowest yield was obtained in intercropping with Jerusalem

artichoke (789 kg/ha), being highlighted distinct significantly differences from the average negative experience. V<sub>2</sub> variant, intercropping with common maize, registered a yield of 1966 kg/ha, being within the average experimental variation limits (Tab. 4; Fig. 4).

### CONCLUSIONS

1. The setting up time in which the highest runner bean yield was obtained was 15.05 in all three variants of intercropping.

2. Comparing the variants to one another, the highest yield was obtained for variant V<sub>2</sub> (intercropping with sunflower), followed by variant V<sub>1</sub> (intercropping with common maize) and V<sub>3</sub> (intercropping with Jerusalem artichoke).

*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.

### REFERENCES

1. Andrews D.J., Kassam A.H., 1976 - *The importance of multiple cropping in increasing world food supplies*. R. I. Papendick, P. A. Sanchez, and G. B. Triplett (eds.). Multiple cropping Amer Soc Agron, Madison, Wis Spec Publ 27:1-10.
2. Anil L., Park J., Phillips R.H., Miller F.A., 1998 - *Temperate intercropping of cereals for forage: A review of the potential for growth and utilization with particular reference to the UK*. Grass Forage Sci., 53:301-317.
3. Axinte M., Roman Gh.V., Borcean I., Muntean L.S., 2006 - *Fitotehnie*. Editura „Ion Ionescu de la Brad”, Iași, ISBN 973-7921-82-8, 662 p.
4. Branas J., 1974 - *Viticulture*. Di:han, Montpellier, France.
5. Kass D.C.L., 1978 - *Polyculture cropping systems: A review and analysis*. Cornell Univ, Ithaca, NY Cornell Intl. Agr Bul., 32.
6. Champagnol F., 1984 - *Elements de physiologie de la vigne et de viticulture generale*. B.P. 13 Prades-le-Lez, 34980 Saint-Gely-du-Fesc, France.
7. Jităreanu G., 1999. *Tehnică experimentală agricolă*. Editura „Ion Ionescu de la Brad”, Iași, ISBN 973-98979-3-2. 256 p.
8. Munteanu, N., Timofte Valentina, Timofte E., 1989 - *Variante tehnologice pentru cultura fasolei urcătoare*. Cercetări agronomice în Moldova, vol.4/1989, Iași. 4(88): 105-113.
9. Ofori F., Stern W.R., 1987 - *Cereal-legume intercropping system*. Advance in Agronomy 41:41-90.
10. Popa Lorena Diana, 2010 - *Cercetări privind agrobiologia speciei Phaseolus coccineus L. în vederea optimizării cultivării*. Teză de doctorat. USAMV Iași. 232 p.
11. Ruști Gr., 2007 - *Cercetări privind îmbunătățirea tehnologiei de cultură a fasolei de grădină urcătoare (Phaseolus vulgaris L. var. communis L.)*. Teză de doctorat. UȘAMV Iași.
12. Stan N., Munteanu N., Stan T, 2003 - *Legumicultură, vol. III*. Editura „Ion Ionescu de la Brad” Iași, ISBN 973-8014-91-3, 315 p.

## RESEARCHES REGARDING THE FERTIGATION THROUGH DRIP IRRIGATION OF SOLARIUM BELL PEPPER CROP

### CERCETĂRI PRIVIND FERTIRIGAREA PRIN PICURARE A UNEI CULTURI DE ARDEI ÎN SOLAR

**CORDUNEANU Oana<sup>1</sup>, ȚENU I.<sup>1</sup>, STOLERU V.<sup>1</sup>,  
ROȘCA R.<sup>1</sup>, ȘOVĂIALĂ Gh.<sup>2</sup>, MATACHE Gabriela<sup>2</sup>**

**e-mail:** itenu@uaiasi.ro

***Summary.** The paper presents some aspects concerning the effect of fertilization factors and drip irrigation over the production level of the bell pepper crop placed in a protected environment. The experiments took place in a solarium from "V. Adamachi" Didactic Farm. The aim of the research was to assess the influence of the fertilization method, when using classic fertilizers and microorganism based fertilizers, simultaneously with drip irrigation. Significant differences regarding the production levels were noticed when the crop was fertilized using the three methods and drip irrigation. The highest production level compared with the control variant (43.885 kg / ha) was obtained for the drip irrigation fertilization method.*

***Key words:** fertigation, bell pepper, harvest, solarium.*

***Rezumat.** În lucrare sunt prezentate aspecte privind influența unor factori de fertilizare și irigarea prin picurare asupra producției obținute la o cultură de ardei gras amplasată în spațiu protejat. Cercetările experimentale au fost organizate într-un solar aparținând disciplinei de Legumicultură, situat în Ferma Didactică "V. Adamachi" din județul Iași. Scopul cercetărilor este de a determina influența metodei de fertilizare, folosind îngrășăminte clasice și pe bază de microorganisme, concomitent cu irigarea prin picurare, comparativ cu varianta nefertilizată. În acest sens, cultura a fost supusă fertilizării prin cele trei metode și irigării prin picurare, constatându-se diferențe semnificative în ceea ce privește producția. Producția cea mai mare, comparativ cu martorul, a fost obținută în cazul fertilizării concomitent cu irigarea prin picurare, obținându-se 43.885 kg/ha.*

***Cuvinte cheie:** fertirigare, ardei, recoltă, solar.*

## INTRODUCTION

Fertigation achieves fertilization in the same time with irrigation by injecting exact quantities of fertilizers into water; through this method the plants receive the appropriate quantities of fertilizer, depending on the growing stage. A fertigation system contains the aspersion or drip irrigation installation and the devices for injecting the liquid fertilizer into water, including tanks for the concentrated liquid

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> I.H.P. – Research Institute for Hydraulics and Pneumatics, Bucharest, Romania

fertilizer, dosing pumps or automatic programmers and an ejector or Venturi type homogenizing device (Țenu, 2004).

In order for the vegetable plants to absorb the necessary soil minerals, they consume an amount of water about 10,000 times greater than the amount of these substances (Grumeza and Drăgănescu, 1983).

Drip irrigation is characterized by the slow, drop wise distribution of water through a network of valves, pipes, tubes and emitters, in the root area of plants, thus wetting only a part of the soil surface and of the root system (Țenu 2004; Hoble, 2010). Drip irrigation was used in ancient times by the burial in soil of clay pots filled with water, allowing its gradual infiltration into the soil.

Modern drip irrigation has its early development in the 1860s Germany, when researchers began experimenting using clay pipes in order to create combined irrigation and drainage systems (Reinders, 2007).

Bell pepper (*Capsicum annuum* L.) is a nutritious plant, mainly due to the high content of vitamin C (150-300 mg/100g) and carotenoids (1.8 to 4.5 mg/100g) (Hoble, 2010).

Pepper has high demands for water during the growing season due to a poorly developed root system and to the abundant biomass, strongly exposed to perspiration (Tanaskovik, 2013). In the case of bell pepper the amount of fertilizer has a distinct significant influence over the differences in the total yield (Stan *et al.*, 2006).

Another key factor for a successful fertigation is the choice of the cultivar, which has to be suitable for the salt stress conditions (Stoleru *et al.*, 2012; Ciobanu *et al.*, 2009).

## MATERIAL AND METHOD

The researches were conducted in a semicircular type solarium farm with an area of 270 m<sup>2</sup>, located within the "V. Adamachi" Didactic Farm of USAMV. Two experiments were organized within the solarium area, for two different vegetable species, respectively tomatoes and pepper, alternatively positioned in the same number of variants (Fig. 1).

The studied plants belong to the *Brillant F1* bell pepper cultivar, grouped into four experimental variants (Table 1), on 80 cm spaced bands, with a distance of 60 cm between the rows per band and a distance of 45 cm between the plants in a row, resulting in a density of 31,740 plants/ ha. The same hybrid, but unfertilized, was used for the protective band of the experiment.

Table 1

Experimental variants (2015)

Experimental variant	Fertilization method
V <sub>1</sub>	Drip fertigation with soluble fertilizers
V <sub>2</sub>	Classic fertilization, through soil spreading
V <sub>3</sub>	Microorganism fertilizer spread on the soil surface
V <sub>4</sub>	Unfertilized (M)

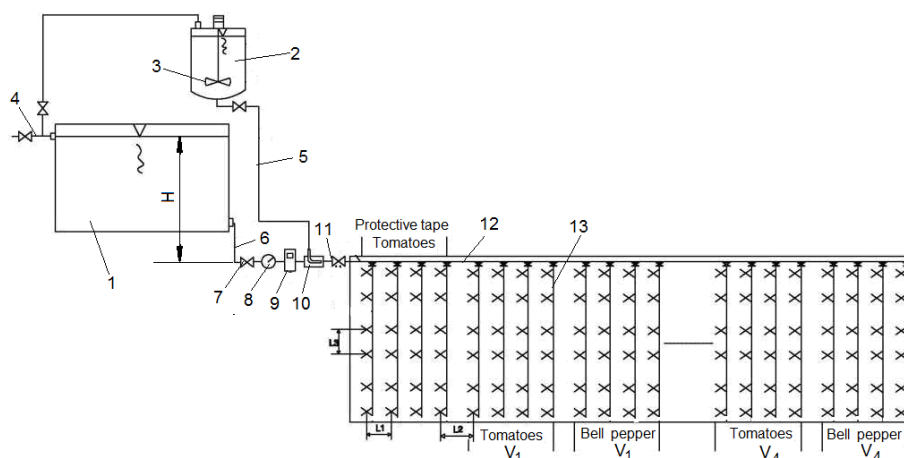
A dripping line was used for plant irrigation; the watering bands had a diameter of 16 mm and a distance of 10 cm between the individual drippers. An equivalent quantity of water of 5200m<sup>3</sup>/ha was administered during the entire vegetation period; the irrigation was performed every two days, 2 hours/day, between 8 am and 10 am or between 7 am and 9 am, depending on the temperature.

The fertigation system consists of a constant level water tank (20 tons), a tank for the concentrated fertilizer solution, an automatic programming system and watering lines (Fig. 1).

The plants in variant 1 were fertilized simultaneously with the drip irrigation, twice a week, the fertigation being carried out in the morning. The fertilizer was: Nutrispore® - NPK (MgO) 10.30.10 (2), boron (B), iron (Fe), manganese (Mn), zinc (Zn), (PGPR) - 300 kg / ha; Nutrispore® NPK (MgO) 30.10.15 (2), boron (B), iron (Fe), manganese (Mn), zinc (Zn), (PGPR) - 425 kg / ha; NPK Nutrispore® 12-48 -8 (2) with boron (B), iron (Fe), manganese (Mn), zinc (Zn), (PGPR) - 400 kg / ha.

In order to obtain the best fertigation possible plant nutrition was achieved twice a week, between two consecutive irrigations.

For variant V<sub>2</sub> the chemical fertilizer was spread around each plant as follows: 200 kg/ha Cristaland® NPK 20-20-20 for the base fertilization; 250 kg/ha Cristaland® NP 15-50 + 2MgO in the phase of floral button (first inflorescence); 200 kg/ha Cristaland® NPK 9-18-27+ 2 MgO in the forming of the first fruits of the inflorescence phenophase (Ø 3 cm).



**Fig 1 - Experimental setup**

1- water pool; 2- graduated cylinder; 3- electric motor; 4 - rotating mixing paddle; 5, 7, 10 - valves; 6 - hose with fertilizing solution; 8 - filter; 9 - hose; 11 - water meter; 12 - programmer; 13 - dripping watering band; L1 – distance between rows within the band; (60 cm); L2 – distance between the bands (80 cm); L3 – distance between the plants in a row (45 cm).

The plants in variant 3 were fertilized with the microorganism type fertilizer Micoseed® MB, in quantity of 60 kg/ha applied during tillage, 2-3 days before planting the peppers. According to the literature data Micoseed MB is a fertilizer based on *Glomus sp.*, *Beauveria sp.*, *Metarhizium sp.* and *Trichoderma sp.* (Stoleru et al., 2014). For this variant



two fertilizations with Nutryaction® (5 L/ha) were applied during the growing season, aiming to boost the biological activity).

Drip irrigation was applied to the control variant  $V_4$ , in the same conditions as the ones for variants  $V_1$ ,  $V_2$  and  $V_3$ .

Biometric measurements (plant height and the number of flowers/fruits) were performed weekly, aiming to evaluate the plant growth dynamics for each variant.

The pepper plants were cared for in accordance with the literature data (Ciofu *et al.*, 2004; Indrea *et al.*, 2003). Experimental data processing was performed using the analysis of variance (ANOVA), in order to calculate the limit differences (Săulescu and Săulescu 1967).

## RESULTS AND DISCUSSION

The dynamics of the bell pepper plants height in the year 2015 is presented in Figure 2. Seven biometric measurements were performed during the experiments, every 7 ... 10 days, beginning with the third week after planting. The *F1 Brilliant* bell pepper cultivar displayed an indeterminate increase, being used for two crop cycles. The average height obtained from the measurements was 43.15 cm.

Figure 2 shows that the height of the *F1 Brilliant* hybrid increased until the beginning June, after which the plant growth ceased. At the beginning of the growing season (26.05), the highest values for the plant height - 35.6 cm - were obtained for variant  $V_3$  (microorganisms).

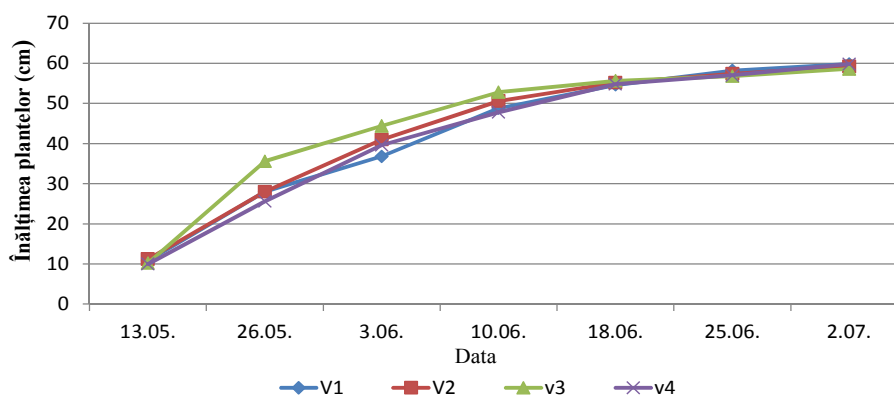


Fig. 2 - Plant height dynamics

The data presented in Figure 2 show that the control variant  $V_4$  recorded lower heights of the plants at the beginning of the growing season compared with the fertilized variants. In the second half of the growing season the highest increases in the height of the pepper plants (59.8 cm) were recorded for  $V_1$  (fertilization) and  $V_4$  (unfertilized).



The results concerning the dynamics of the pepper plants are shown in Table 2. The average number of fruits per plant ranged from 10.06 in the control variant ( $V_4$ ) to 13.31 in the fertigation variant ( $V_1$ ). Average values close to the one recorded for  $V_1$  (12.60) were achieved by the microorganism variant,  $V_3$

Table 2

**Results regarding the dynamics of the sweet pepper plants**

Experimental variant	Average number of fruits per plant	Average fruit weight (g)
$V_1$	13,31	104,30
$V_2$	11,77	95,05
$V_3$	12,60	118,85
$V_4$	10,06	105,55

The average weight of the *Brillant F1* bell pepper fruits ranged from 95.05 g (for  $V_2$ , classic fertilization) to 118.85 g ( $V_3$ , fertilized with microorganisms).

The results concerning the pepper production in 2015 are shown in Table 3. The production of pepper in the experiment varied within wide limits, from 29.048 kg/ha in the control variant ( $V_4$ ), to 43.885 kg/ha in the fertigation variant ( $V_1$ ).

Table 3

**Results regarding the production of sweet peppers (2015)**

Experimental variant	Total production, kg/ha	Relative production, %	Difference from the control variant (kg/ha)	Differential significance
V1	43,885	151,07	14,837	***
V2	32,785	112,86	3,737	*
V3	29,164	102,39	116	ns
V4 (M)	29,048	100	0	ns

DL 5 % = 3719 t/ha; DL 1 % = 5831 t/ha; DL 0.1 % = 9321 t/ha

The difference of 14.837 kg/ha between variant V1 and the control variant was considered to be very significant. A significant positive difference (3737 kg/ha) was also achieved in the classically fertilized variant. The difference for the microorganism fertilized variant  $V_3$  (116 kg / ha) was considered to be insignificant.

## CONCLUSIONS

1. At the beginning of the growing season, the highest values of the plant height were obtained for the  $V_3$  version (microorganisms), respectively of 35.6 cm; in the second half of the growing season, the greatest increases of the height of the pepper plants (59.8 cm) were recorded variants  $V_1$  (fertigation) and  $V_4$  (unfertilized).

2. The average number of fruits per plant ranged from 10.06 in the control variant,  $V_4$ , to 13.31 in the fertigation variant,  $V_1$ . Average values close to the ones for variant 1 were achieved by the microorganism fertilized variant,  $V_3$  (12.60).

3. The average mass of the *Brilliant F1* pepper fruits ranged from 95.05 g in variant  $V_2$  (classically fertilized) to 118.85 g in variant  $V_3$  (fertilized with microorganisms).

4. The pepper production was ranged between 29.048 kg/ha (control variant,  $V_4$ ) and 43.885 kg/ha (fertigation variant,  $V_1$ ); this indicates that the variant using fertigation achieved a better distribution, in time and space, of the fertilizer towards the root system and that the fertilizer was uniformly distributed 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 Program, Financial Agreement no. 158/2014.

## REFERENCES

1. Ceașescu I., Bălașa M., Savițchi P., Voican V., Radu Gr., Stan N., 1984 – *Legumicultură generală și specială*. Editura Didactică și Pedagogică, București.
2. Ciofu Ruxandra., Stan N., Popescu V., Chilom Pelaghia., Apahidean S. Al., Horgoș A., Berar V., Lauer K. F., Atanasiu N., 2004 – *Tratat de legumicultură*. Editura Ceres, București.
3. Grumeza N., Drăgănescu O., 1983 - *Irigații prin picurare*. Editura Ceres, București.
4. Hoble Adela, Luca E., Dirja M., Laura Luca, Sălăgean T., 2010 - *Suitability of micro-irrigation- drip irrigation - used for pepper growth, in conditions of field cultivation*. Revista Agricultura nr. 1-2: 73-74.
5. Metin S., Attila S., Yazar S., Eker., 2006 - *Effect of drip irrigation regimes on yield and quality of field grown bell pepper*. Agricultural Water Management 81: 115-131.
6. Săulescu N. A., Săulescu N.N., 1967 – *Câmpul de experiență*. Editura Agro-Silvică, București.
7. Stan N., Munteanu N., Stoleru V., Stan T., Stoleru Carmen Maria, Păduraru E., 2006 - *Organic fertilizing effect on the sweet pepper crop in poly-tunnels, in NE area of Romania, during the conversion period*. Lucrări științifice vol.49, seria, Horticultură, UȘAMV Iași, pp. 869-874.
8. Stoleru V., Munteanu N., Sellito V.M., 2014 – *New approach of organic vegetable systems*. Editura ARACNE, Italia.
9. Stoleru V., Munteanu N., Stoleru Carmen Maria, Rotaru Liliana., 2012 – *Cultivar selection and pest control techniques on organic white cabbage yield*. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, Volume 40 (2), pp. 190-196.
10. Tanaskovik V., Cukaliev O., Spalevic V., Markoski M., Nechkovski S., 2013 - *The effect of irrigation and fertilization techniques on specific water consumption of green pepper*. Proceedings of the 1<sup>st</sup> International Congress On Soil Science, XIII National Congress In Soil Science "Soil – Water – Plant", 23-26 September, Belgrade, Serbia.
11. Țenu I., 2004 – *Echipamente pentru irigații*. Editura Cermi, Iași.
12. \*\*\* Reinders, 2007

## THE BEHAVIOR OF A RUNNER BEAN ASSORTMENT FOR PODS (*PHASEOLUS COCCINEUS* L.) IN POLYETHYLENE TUNNELS, IN A CROP ESTABLISHED BY DIRECT SOWING

COMPORTAREA ÎN SPAȚII PROTEJATE A UNUI SORTIMENT DE FASOLE MARE (*PHASEOLUS COCCINEUS* L.) PENTRU PĂSTĂI, ÎN CULTURĂ ÎNFIINȚATĂ PRIN SEMĂNAT DIRECT

TELIBAN G.C.<sup>1</sup>, MUNTEANU N.<sup>1</sup>, POPA Lorena-Diana<sup>2</sup>, STOLERU V.<sup>1</sup>,  
HAMBURDĂ Silvia Brîndușa<sup>1</sup>, STAN T.<sup>1</sup>, BUBURUZ Alexandra-Andreea<sup>2</sup>,  
ONOFREI Vasilica<sup>1</sup>, CIOBANU V.<sup>3</sup>  
e-mail: teliban\_gabriel\_ciprian@yahoo.com

**Abstract.** The paper is focused on the study of agroproductive behavior of a runner bean for pods assortment, in polyethylene tunnel conditions. The trial was conducted in the experimental polygon of the Vegetable Growing discipline, belonging to the Faculty of Horticulture from Iasi. The culture was established in 2014, by direct sowing, in nests, two plants in nest, being established three different densities through the experimental protocol: 20, 25 and 33 thousand nests/ha. The research sought to determine the total yield at four indeterminate cultivars of runner bean (Lady Di, Desiree, Polestar and White Apollo), the control variant being represented by the Desiree cultivar, considered classic in the home country (UK), established at a density of 25 thousands nests/ha. Within the bifactorial experience, the highest total production was obtained by the Polestar cultivar, sown at the distance of 50 cm between nests per row (density of 20 thousands nests/ha).

**Key words:** runner bean for pods, planting distances, polyethylene tunnel.

**Rezumat.** Lucrarea se axează pe studiul comportamentului agroproductiv al unui sortiment de fasole mare pentru păstăi, în condiții de solar. Experiența a fost derulată în poligonul experimental al disciplinei de Legumicultură, aparținând Facultății de Horticultură din Iași. Cultura a fost înființată în anul 2014, prin semănat direct, în cuiburi, câte două plante la cuib, fiind stabilite prin protocolul experimental trei densități diferite: 20, 25 și 33 mii cuiburi/ha. Cercetările au urmărit determinarea producției totale la patru cultivare urcătoare de fasole mare (Lady Di, Desiree, Polestar și White Apollo), varianta martor fiind reprezentată de cultivarul Desiree, considerat clasic în țara de origine (Marea Britanie), înființat la o densitate de 25 mii cuiburi/ha. În cadrul experienței bifactoriale, producția totală cea mai ridicată a fost obținută de cultivarul Polestar, semănat la distanța de 50 cm între cuiburi pe rând (densitatea de 20 mii cuiburi/ha).

**Cuvinte cheie:** fasole mare pentru păstăi, distanțe de plantare, solar.

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> Agricultural Research Development Station Secuieni, Romania

<sup>3</sup> Institute of Genetics, Physiology and Plant Protection of Academy of Sciences of Moldova, Republic of Moldova

## INTRODUCTION

The runner bean (*Phaseolus coccineus* L.) is a well-known species in Romania, with food and ornamental utilities, but occupies relatively small areas, being especially common in the gardens of the rural population (Munteanu, 1985). This species is grown especially for its dry and green beans, are less known the forms from which pods are used (Munteanu, 2005; Munteanu *et al.*, 2007; Popa *et al.*, 2008; Popa, 2010; Hamburdă *et al.*, 2014). The growth of runner bean for pods in protected areas is not known in Romania, although the results from other countries recommend this culture system (Klaus, 2013). Researches focused on this theme were held worldwide, in UK, Netherlands, France etc., but in Romania there were no concerns in this regard (Phillips, 1993).

The assortment for pods, used at European level, comprising, in general, improved climbing cultivars, but also improved cultivars or local populations with determined growth, for these being required a support system (<http://www.marshalls-seeds.co.uk/>, <http://www.thompson-morgan.com/>, <http://www.tozerseeds.com/eu/en/>).

Knowing the need to diversify the vegetables assortment, as well as some technological characteristics of this culture (Munteanu, 1985; Hidalgo *et al.*, 1986), our research aimed assessing the cultivation possibilities of the runner bean for pods in polyethylene tunnels, in Romania conditions, establishing as main objectives the study of cultivar and density influence on the crop and yield.

## MATERIAL AND METHOD

The researches were organized in the experimental conditions of Didactic Station of USAMV Iasi, Horticultural Farm, in 2014. The experience was established in a covered polyethylene tunnel, with the dimensions of 40 x 10 x 3.5 m, on a chernozem cambic soil type, medium stocked in nutrients, with 3% organic matter and pH = 6.5.

The biological material used was represented by four cultivars of runner bean for pods, stringless, from UK: Lady Di, Desiree, Polestar and White Apollo (table 1).

Table 1

Runner bean assortment used in the experience

No.	Cultivar	Provenance	Flowers color	Seeds color	Presence/absence of thread in pods
1.	Lady Di	U.K.	red	purple with black arabesque	stringless
2.	Desiree	U.K.	white	white	stringless
3.	Polestar	U.K.	red	purple with black arabesque	stringless
4.	White Apollo	U.K.	white	white	stringless

The experience was of bifactorial type, organized in a device of subdivided plots, with three repetitions. The experimental factors were:

- A factor – the used assortment, with four graduations:  $a_1$  = Lady Di;  $a_2$  = Desiree;  $a_3$  = Polestar;  $a_4$  = White Apollo; as the control graduation was considered  $a_2$  (Desiree cultivar).

- B factor – culture density, expressed by three graduations:  $b_1 = 33000$  nests/ha (100 x 30 cm);  $b_2 = 25000$  nests/ha (100 x 40 cm);  $b_3 = 20000$  nests/ha (100 x 50 cm); as control density was the 25000 nests/ha.

The culture was established on 20 May, in nests of three seeds/nest, after emergence were kept only two plants/nest. The sowing was done in equidistant rows, spaced at 100 cm, the distances between nests per row being of 30, 40, and 50 cm. The support system consisted of a plastic net, with a mesh of 15 x 15 cm and the width of 1.7 m, placed at 30 cm from the ground. The net has been stretched over the rows, immediately after plant emergence (Fig. 1).



**Fig. 1** – Aspects from the runner bean for pods experience, in polyethylene tunnel, from USAMV Iasi  
a – aspect from polytunnel culture, b – ento pollination with bumblebees (original photo)

During the vegetation season, the maintenance work consisted in manual and mechanical weeding, phytosanitary treatments, root and foliar fertilizations, drip irrigation, trellising and pinching plants (Popa, 2010; Stan *et al.*, 2003). Also, in the polyethylene tunnel was placed a hive of bumblebees for flower pollination, the *Phaseolus coccineus* L. species being allogam (Kendall and Smith, 1976; Munteanu, 1985; Munteanu, 2005). The pods harvesting in order to determine yield was performed at 7-8 days intervals between harvests.

During the experience, were made determinations of the total harvest quantity, the data being processed by appropriate statistical – mathematical methods (Săulescu and Săulescu, 1967; Jităreanu, 1999).

## RESULTS AND DISCUSSION

The total yield of pods, within the study assortment, has ranged, in 2014, between 6591 kg/ha (Desiree x 33000 nests/ha) and 20726 kg/ha (Polestar x 20000 nests/ha), while the control variant (Desiree x 25000 nests/ha) has achieved 7533 kg/ha (Table 2).

The statistical analysis (Table 3) for the total yield shows that the yield differences or the yield variance between cultivars is due to their characteristics, as demonstrated by the fact that the value of F ratio ( $sv^2/se^2$ ) calculated is 167.17 compared with  $F_{\text{theoretical}}$ , of 4.76, at a degree of confidence of 95%. Also, the Fischer test shows that the total yield is determined significantly distinct and by

the variation of the cultivation densities,  $F_{\text{calculated}}=28.09$  compared with  $F_{\text{theoretical}}=3.63$ .

Table 2

## Results of runner bean assortment yields for the studied densities (kg/ha)

No.	Cultivar	33000 nests/ha	25000 nests/ha	20000 nests/ha	Average
1.	Lady Di	14043	14381	17964	15463
2.	Desiree	6591	7533	11272	8465
3.	Polestar	16694	17763	20726	18394
4.	White Apollo	7422	7533	10314	8423
	Average	11187	11803	15069	12686

The Fischer test, for the cultivar x density factors interaction, demonstrates that the yield differences compared to the control haven't any significance and is due, mainly, to the experimental error ( $F_{\text{calculated}} = 0.26 < F_{\text{theoretical}} = 2.74$ ).

Table 3

## The variance analysis for the bifactorial experience

Variance source	SP	GL	S <sup>2</sup>	F <sub>calculated</sub>	F <sub>theoretical</sub>
Large plots	695630900	11			
Repetitions	873472	2			
A factor	686543900	3	228848000,0	167,17	4,76
Error (a)	8213504	6	1368917,0		
Small plots	832771100	35			
B factor	104455200	2	52227590,0	28,09	3,63
A x B	2936320	6	489386,7	0,26	2,74
Error	29748740	16	1859296,0		

SP = sum of squared deviations; GL = degrees of freedom; S<sup>2</sup> = variance; F = factor

In the studied cultivars, the total yield ranged between 8423 kg/ha (White Apollo) and 18394 kg/ha (Polestar), the value recorded by the control cultivar (Desiree) being of 8465 kg/ha (Table 4).

Table 4

## The comparative analysis of the total yield determined by the studied cultivars

No.	Cultivar	Yield			Difference significances
		kg/ha	% to C	Differences to the C	
1.	Lady Di	15463	182.67	6998	xxx
2.	Desiree (C)	8465	100.00	0	
3.	Polestar	18394	217.30	9929	xxx
4.	White Apollo	8423	99.51	- 42	-

DL 5% = 1351.3 kg/ha, DL 1% = 2046.2 kg/ha, DL 0.1% = 3287.2 kg/ha

The yield differences obtained when comparing to the control (Desiree cultivar – 8465 kg/ha) have recorded very significant degree of confidence in the case of Lady Di (15463 kg/ha) and Polestar (18394 kg/ha) cultivars.

Although the studied assortment was reduced, however there was a very high variability of yield, the lowest yield being extremely close to the control (White Apollo cultivar – 99.51%), the highest, however, representing 217.3% (Polestar cultivar). This aspect presents a particular importance in the improvement

field of this species, and could use some technological links with impact on improving the productive capacity, such as distances, establishing epochs etc.

Regarding the analysis of the yield results, on average for each of the three cultivation densities, compared with the control variant (the density of 25 thousands nests/ha), the 20 thousands nests/ha density has achieved a total yield of 15069 kg/ha, representing a yield increase of 27.68%, that can be recommended in culture, because the plants thus have a space of light and nutrition efficiently distributed (Table 5).

Table 5

**The average of yield results determined by the three studied densities**

No	Density (nests/ha)	Yield			Difference significances
		kg/ha	% to C	Differences to the C	
1.	33000	11187	94.79	-616	-
2.	25000 (C)	11803	100.00	0	
3.	20000	15069	127.68	3266	xxx

DL 5% = 1180.1 kg/ha, DL 1% = 1625.5 kg/ha, DL 0.1% = 2237.8 kg/ha

The yield differences recorded at a density of 20 thousands nests/ha (3266 kg/ha) were ensured at a very significant level when compared to the control of 25 thousands nests/ha (11803 kg/ha).

The 33 thousands nests/ha density presented yield values beneath control variant, the negative differences were not statistically ensured (-616 kg/ha).

Table 6

**Comparative results between the cultivar x distance between rows combinations (A x B)**

No.	A x B	Production			Difference significances
		kg/ha	% to C	Differences to the C	
1.	a1b1	14043	186.42	6510	xxx
2.	a1b2	14381	190.91	6848	xxx
3.	a1b3	17964	238.47	10431	xxx
4.	a2b1	6590	87.49	-943	-
5.	a2b2 (C)	7533	100.00	0	
6.	a2b3	11271	149.63	3738	xx
7.	a3b1	16694	221.61	9161	xxx
8.	a3b2	17763	235.80	10230	xxx
9.	a3b3	20726	275.14	13193	xxx
10.	a4b1	7422	98.53	-111	-
11.	a4b2	7533	100	0	-
12.	a4b3	10314	136.92	2781	x

DL 5% = 2348.6 kg/ha, DL 1% = 3330.8 kg/ha, DL 0.1% = 4829.4 kg/ha

Combination between cultivar x density has generated production results which ranged from 6590 kg/ha (Desiree x 33 thousands nests/ha) to 20726 kg/ha (Polestar x 25 thousands nests/ha), under a control of 7533 kg/ha (Desiree x 25 thousands nests/ha), the differences being very significant for six combinations compared to the control (table 6).



## CONCLUSIONS

1. Analyzing the results obtained due to the cultivar influence, it may be noted that the highest yields, compared to the control (Desiree – 8465 kg/ha), were achieved at the Polestar and Lady Di cultivars (18394 kg/ha, respectively 15463 kg/ha), the obtained increases were statistically ensured as significant.

2. The comparative analysis of the three experimented densities showed that the highest yield increase (27.68%), when comparing to the control (25 thousands nests/ha) was recorded at the density of 20 thousands nests/ha, this may be recommended for growing runner bean for pods in protected areas, in order to improve the light and nutrition space.

*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

## REFERENCES

1. **Hamburdă Silvia Brândușa, Munteanu N., Popa Lorena Diana, 2014** –*Evaluation of a runner bean assortment (Phaseolus coccineus L.) in Iasi county conditions.* Știința Agricolă, nr. 1, Chișinău.
  2. **Hidalgo R. et al., 1986** –*The cultivated species of Phaseolus.* Centro Internacional de Agricultura Tropical CIAT, Cali, Columbia.
  3. **Jităreanu G., 1999** –*Agricultural experimental technique.* Editura "Ion Ionescu de la Brad", Iași.
  4. **Kendall D.A., Smith B.D., 1976** – *The Pollinating Efficiency of Honeybee and Bumblebee Visits to Flowers of the Runner Bean Phaseolus coccineus L.* Journal of Applied Ecology, Vol. 13, No. 3.
  5. **Klaus L., 2013** –*Vegetables and herbs for the greenhouse and polytunnel.* Constable and Robinson Ltd., London.
  6. **Munteanu N., 1985** –*Phaseolus coccineus L. –A vegetable species that deserve more attention.* Vegetable production, Horticulture, no.4.
  7. **Munteanu N., 2005** –*Preliminary studies regarding the biodiversity of runner bean (Phaseolus coccineus L.) species.* Lucrări științifice, USAMV Iași, seria Horticultură.
  8. **Munteanu N., Popa Lorena-Diana, Teliban G.C., 2007** –*Suitability of Phaseolus coccineus L. species for cultivation in sustainable agriculture systems.* Lucrări științifice, seria Horticultură, anul L, USAMV Iași.
  9. **Phillips R., 1993** –*Vegetables (Runner bean).* Martin Rix.
  10. **Popa Lorena-Diana et al., 2008** –*Yield comparative study of some local populations of runner bean (Phaseolus coccineus L.).* Lucrări științifice, seria Horticultură, anul LI, USAMV Iași.
  11. **Popa Lorena-Diana, 2010** – *Research on agrobiological of Phaseolus coccineus L. in order to optimize the cultivation.* Doctoral thesis, USAMV Iași.
  12. **Săulescu N.A., Săulescu N.N., 1967** – *Field experience-second edition.* Editura Agrosilvică, București, 1967.
- Stan N., Munteanu N., Stan T., 2003** –*Vegetable growing, vol III.* Editura "Ion Ionescu de la Brad", Iași, 2003.
- <http://www.marshalls-seeds.co.uk/>.  
<http://www.thompson-morgan.com/>.  
<http://www.tozerseeds.com/eu/en/>.



## PRELIMINARY RESULTS REGARDING NATURAL TENDENCY OF FEATHERING AND GROWTH HABITS OF SOME APPLE SCION-ROOTSTOCK COMBINATIONS, IN THE NURSERY

### REZULTATE PRELIMINARII PRIVIND TENDINȚA NATURALĂ DE EMITERE A LĂSTARILOR ANTICIPATI ȘI PARTICULARITĂȚILE DE CREȘTERE A UNOR COMBINAȚII SOI-PORTALTOI ÎN PEPINIERĂ, LA SPECIA MĂR

**JAKAB-ILYEFALVI ZSOLT<sup>1</sup>**

e-mail: zsolt.jakab@yahoo.com

**Abstract.** *There were studied several scion cultivars in the nursery regarding natural tendency of feathering at Fruit Research and Development Station Bistrita in 2015. The goal of the study was to assess the natural behavior of forming preformed shoots of several scion-rootstock combinations in the first year after grafting. Several researches showed that not only grafted scion has a direct effect on the sylleptic branching of fruit trees and feathering but also the rootstock has an important influence. The studied scion-rootstock combination were: Florina/M106, Florina/M26, Jonathan/M106, Jonathan/M26. Results indicate that the hypothesis that not only the cultivar but also the rootstock influences the emergence of the feathers is confirmed, medium to strong vigor rootstock M106 induces feathering on Florina cultivar naturally (average number of 3.13 shoots), probable the more complex and dense root system provides sufficient nutrients in order to promote lateral branching. Medium feathering was observed also at Starkprim cultivar on M106 rootstock (2,8 shoots), not included in the statistical analysis but observed in singular combination, showed high sylleptic shoot formation. Even more sylleptic shoots were formed in the vigorous combination of Auriu de Bistrita cultivar on M106 (3,8 shoots) also not included in the analysis in this experimental phase. Based on these results in further researches using agro-technical methods like pinching in optimum time intervals, foliar fertilizing and other techniques we will study and develop good feathered trees with more lateral shoots and strong root system in order to provide quality planting material for farmers.*

**Key words:** scion, rootstock, grafting, feathers, shoots, lateral branching, insertion angle, planting material

**Rezumat.** *Au fost efectuate studii privind emiterea naturală de lăstări anticipați în pepinieră, la Stațiunea de Cercetare-Dezvoltare pentru Pomicultură Bistrița în anul 2015. Obiectivul studiului a fost de a evalua tendința naturală de formare a lăstarilor anticipați la unele combinații soi-portaltoi în primul an după altoire. Combinațiile soi-portaltoi au fost Florina/M106, Florina/M26, Jonathan/M106, Jonathan/M26. Rezultatele indică faptul că, se confirmă ipoteza ca nu numai soiul dar și portaltioiul influențează emiterea de lăstari*

---

<sup>1</sup> Fruit Research and Development Station Bistrita, Romania

*anticipați, portaltoiul mediu-viguros M106 a influențat emiterea de lăstari anticipați la soiul Florina în mod natural (număr mediu de lăstari 3.13), cel mai probabil datorită sistemului radicular mai complex și mai dens care furnizează suficienți nutrienți din sol pentru favorizarea emiterii lăstarilor anticipați. O lăstărire laterală medie a fost observat și la soiul Starkprim pe portaltoiul M106 (2.8 lăstari), acesta nu a fost inclus în studiul statistic dar s-au efectuat observații în combinații individuale, portaltoiul favorizând apariția de lăstari anticipați la acest soi. S-a observat o mai pronunțată emiterie de lăstari anticipați la soiul Auriu de Bistrița altoit pe portaltoiul M106 (3.8 lăstari anticipați) acesta de asemenea nu s-a inclus în analiza statistică în această fază experimentală. Bazat pe aceste observații utilizând metode agrotehnice ca ciupitul frunzelor efectuat în intervale de timp optime, fertilizări foliare și alte tehnici, vom studia și produce pomi fructiferi cu mai mulți lăstari anticipați preformați cu un sistem radicular puternic pentru a furniza material de plantare de calitate fermierilor.*

**Cuvinte cheie:** soi, portaltoi, altoire, lăstari anticipați, lăstărire laterală, unghi de inserție, material de plantare

## INTRODUCTION

Fruit tree planting material quality depends primarily on good developed root system, trunk and a number of well developed preformed shoots with wide insertion angles (Wertheim and Webster, 2003). Nursery planting material can be found in two categories, trees without preformed shoots and trees with lateral branches with 3-4 rarely 5 shoots, called feathers. The importance of the feathers is crucial, a reduced number of preformed shoots or the inexistence of them has the influence of lately bearing of the new orchards (Robinson, 2007). Extensive research was carried out in several countries to investigate the methodology to induce feathers, using plant hormones like cytokinins (BA-benzyladenine) and gibberellins GA4+7 (giberellic acid) or agro-technical methods like pinching and foliar fertilizations to brake the apical dominance and to induce feathering (Hrotko *et al.* 1996; Magyar *et al.*, 2008; Gudarowska, 2004; Dragan Radivojevic *et al.*, 2015; Csiszar and Buban, 2004) but there are few articles regarding the natural behavior of cultivars and rootstocks in the nursery regarding feathering. The number of shoots and the length of shoots influences the yield per tree (Pietranek *et al.*, 2006) together with the insertion angles of branches along with the ecological factors like temperature of soil, atmospheric humidity, air temperature, the existent macro and micro elements in the soil thus the ecological factors of the orchards (Tromp, 1996). The apple cultivars have different capacity to promote sylleptic lateral branching. First shoots of the trees will provide future elements for flowering, developing branching system for vegetative system. Research on scion-rootstock interaction showed that rootstock influences the growth rate of shoots. Also there were observed that rootstocks have not only the size controlling effect but also the effect on scion shoot growth. The goal of the present experiment was to observe the natural behavior of some scion cultivars and the influence of rootstocks regarding the growth habit, number of shoots, insertion angles of shoots in the first growth year after grafting. The hypothesis of the

research was that both of the two factors have a direct influence on feathering, cultivar and rootstock. Research will provide information which part of the scion-rootstock combination influences the natural feathering of trees.

## MATERIAL AND METHODS

Researches were effectuated at the Nursery of Fruit Research and Development Station Bistrita located in Bata village, Bistrita-Nasaud county, Romania, near Dej city. The nursery is located near Somes river, soil conditions were optimum, establishment was on a sandy-loam site, well drained, with optimum thermal and rainfall conditions in 2015. The experiment followed a completely randomized block design with 3 repetitions per variant. Factor A was represented by cultivar type (Florina, Jonathan) and factor B was represented by rootstock type (M106, M26). Florina and Jonathan apple cultivars were grafted in 2014 on M106 medium to strong vigor rootstock and M26 medium vigor rootstock.

At the final period of the growth period in 2015 there were counted number of shoots, length of shoots and measured the shoot growing angle. The registered data was statistically analyzed by the analysis of variance test using Excel 2003 add on Daniel's Toolbox statistical software.

## RESULTS AND DISCUSSIONS

### Research results regarding number of shoots in natural feathering

The observations in the research plot regarding the natural feathering of the cultivars due to factor cultivar alone showed no particular distinctive differences, it seems that the few number of the preformed shoots was not influenced by the two cultivars Florina and Idared. Average values of the counted shoots showed that in both cultivars 2-3 shoots were formed at M106 rootstock combination (Fig. 1), while in M26 grafting combination just 1-2 shoots were formed (Table 1). The values regarding number of shoots in both factors were close each other. On the other hand the factor B –rootstock influenced very significantly ( p value 0.0003) the scion-rootstock combination (Table 2). The interaction between the rootstocks and cultivars showed also significance ( p value 0.0174).

Table 1

**Average number of shoots of the studied scion-rootstock combinations at FRDS Bistrita in 2015**

Cultivar	Rootstock	
	M106	M26
Florina	3.13	1.67
Jonathan	2.27	1.93

Table 2

**Analysis of variance regarding number of shoots in natural feathering**

Computation	SS	DF	MS	F	P
Total	63.3	59	1.1		
Factor A	1.4	1	1.4	1.7	0.1999
Factor B	12.2	1	12.2	15.1	<b>0.0003***</b>
Interaction	4.8	1	4.8	6.0	<b>0.0174*</b>
Res. Error	44.9	56	0.8		



**Fig.1** - Natural tendency of feathering in the fruit nursery, Idared cultivar/M106 rootstock

### Research results regarding length of shoots in natural feathering

Studying the average length of shoots it appears that in both of the cultivars at the M106 more vigorous shoots were formed (23.90-24.62 m) than on the M26 rootstock (13.28-19.3 cm) (Table 3).

*Table 3*  
**Average length of shoots (cm) of the studied scion-rootstock combinations at FRDS Bistrita in 2015**

Cultivar	Rootstock	
	M106	M26
Florina	24.62	13.28
Jonathan	23.90	19.3

Length of shoots it is influenced not only by cultivar but also by rootstock it appears in the following research ( table 4). The M106 and M26 rootstocks had a strong influence on the length of shoots, factor B being very significant (p-value 0.0002).

*Table 4*  
**Analysis of variance regarding length of shoots in natural feathering**

Computation	SS	DF	MS	F	P
Total	4591.4	59	77.8		
Factor A	105.2	1	105.2	1.8	0.1911
Factor B	953.3	1	953.3	15.9	<b>0.0002***</b>
Interaction	169.9	1	169.9	2.8	0.0982
Res. Error	3363.1	56	60.1		

### Observations regarding insertion angles of shoots

*Table 5*  
**Insertion angles of shoots (°) of the studied scion-rootstock combinations at FRDS Bistrita in 2015**

Cultivar	Rootstock	
	M106	M26
Florina	55	45
Jonathan	50	48

The insertion angle of the cultivars were between 50-55 ° at M106 rootstock combinations (fig.2) and slightly sharp angles at M26 rootstock combinations, general observation being that in the first year after grafting sharp and narrow angles of feathers are formed.



**Fig. 2** - Insertion angles (50-55 ° ) of the new feathers at Idared cultivar / M106 rootstock

Analyzing the registered data it appears that the rootstocks M106 and M26 influences strongly the rootstock-scion combinations, higher number of roots in M106 and a diverse and strong root system influences more nutrient uptake thus the easier braking of apical dominance and the forming of new lateral shoots depends also on the type of rootstock. Rootstock M26 has a medium root system, the mineral uptake possibility is reduced in comparison with M106 rootstock which has a strong root system, sylleptic shoot formation being more pronounced in M106 combinations.

## CONCLUSIONS

1. The effectuated observations showed that natural feathering of apple cultivars is influenced not only by scion but also definitely by rootstocks also. In the experiment no significant influence of the cultivars was observed regarding the number of shoots, it seems that the natural habit of feathers formation of the two studied cultivars in 2015 was quite the same.

2. The use of medium to strong rootstock M106 and medium vigor rootstock M26 induced the natural appearance of 2-3 shoots. The length of the feathers were 23-24 cm at M106 rootstock and 13-19 cm at M26, rootstocks influencing the scion-rootstock combination.

3. Further researches will be carried out in order to assess and understand other cultivars natural tendency of feathering in different scion-rootstock combinations and there will be applied agro-technical methods to improve the natural feathering of nursery fruit trees and produce healthy, quality planting material.

**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.*

## REFERENCES

1. **Csiszar Laszlo, Buban Tamas, 2004** - *Improving of the feathering of young apple trees in environment friendly way by modified benzyladenine application*, Journal of Fruit and Ornamental Plant Research, Orchard Management in sustainable fruit production, 12: 31-39.
2. **Dragan Radivojevic, Ivan, S. Momirovic, Jasminka M. Milivojevic, Milovan M. Velickovic, Cedo D. Oparnica, Milan M. Lukic., 2015** - *The influence of BA and BA+GA4+7 on formation of sylleptic shoots on one year old apple nursery trees*, Journal of Agricultural Sciences, 60(1): 89-95.
3. **Gudarowska Ewelina, Adam Szewczuk, 2004** - *The influence of agro-technical methods used in the nursery on quality of planting material and precocity of bearing in young apple trees in the orchard*, Journal of Fruit and Ornamental Plant Research, Orchard Management in sustainable fruit production, 12: 91-96.
4. **Hrotko K., Magyar L., Buban T., 1996** - *Effect of Benzyladenine Application on One year old 'Idared' Apple Trees in Nursery*, Horticult. Science 28 (3-4):49-53.
5. **Magyar L., Brancsi Z., Hrotko K., 2008** - *Improved feathering by combined application of benzyladenin (BA) and biostimulators*, Scientific papers of the Research Institute for fruit growing Pitesti, Romania, ISSN 1584-2231, Vol.XXIV: 67-70.
6. **Pietranek A., Jadzuk E., 2006** - *Growth and bearing of Jonagold apple trees as affected by rootstock and the type of nursery trees used for planting*, Latvian Journal of Agronomy, No. 9: 103-108.
7. **Robinson T.L., 2007** - *Recent advances and future directions in orchard planting Systems*, Acta Horticulturae, 732: 367-381
8. **Wertheim S.J., Webster A.D., 2003** - *Propagation and nursery tree quality* In. D.C.Feree and I.J.Warrington. Eds. Apples: Botany, Production and Uses.
9. **Tromp J., 1996** - *Sylleptic shoot formation in young apple trees exposed to various soil temperature and air humidity regimes in three successive periods of the growing season*, Annals of Botany, 77: 63-70.



## EFFECT OF THINNING "IDARED" APPLE VARIETY USING NAD AND ETHEPHON

### EFFECTUL RĂRIRII FRUCTELOR DE SOIUL IDARED UTILIZÂND PRODUSE PE BAZĂ DE NAD ȘI ETHEPHON

**PESTEANU A.<sup>1</sup>**

**e-mail:** a.pesteanu@uasmd.md

**Abstract.** *The experimental plot is placed in the orchard "Codru-ST" Ltd. founded in 2009 with trees of a „knip boom” canopy type. The study subject of the experience was Idared apple variety grafted on M 9 rootstock. The distance of plantation is 3.5 x 1.2 m. The research was conducted during the period of 2012 year. The tested agents were NAD (Geramid-New) and Ethiphon (Cerone 480SL), which was sprayed in different thinning period. During the research, it was studied the yield, mean fruit weight, average fruit diameter, the number of first class fruits. It was established that, the spray with Geramid-New in dose 1.5 l/ha when 80% of the petals have fallen + 2-3 days, or Cerone 480SL in dose 0.3 l/ha at 10 - 20 days after full flowering have a significant effect on yield and fruits quality.*

**Keywords:** *apple, thinning, ANA, Ethephon, yield.*

**Rezumat.** *Cercetările s-au efectuat în livada întreprinderii „Codru-ST” fondată în anul 2009 cu pomi cu coroană formată după metoda „knip boom”. Obiectul cercetărilor a fost soiul Idared altoit pe portaltoiul M9. Distanța de plantare 3,5 x 1,2 m. Cercetările s-au efectuat pe parcursul anului 2012. Regulatorii de creștere utilizați au fost pe bază de NAD (Geramid-New) și Ethephon (Cerone 480SL), care au fost testați în diferite perioade de rărire. În perioada cercetărilor s-au analizat recolta, greutatea și diametrul mediu a fructelor, precum și ponderea fructelor de calitate I. S-a stabilit, că tratarea cu Geramid-New în doza 1,5 l/ha când 80% din petale au căzut + 2-3 zile, ori Cerone 480SL în doza 0,3 l/ha la 10 – 20 zile după înflorirea completă au influențat asupra producției și calității fructelor.*

**Cuvinte cheie:** *măr, rărire, ANA, Ethephon, producție.*

## INTRODUCTION

Achieving high quantities of qualitative fruits can only be done by using appropriate modern technologies applied in accordance with natural conditions, economic, land sector-specific basis (Babuc V. *et al.*, 2013; Cimpoieș Gh., 2012).

Determining the fruit load is the technological operation that specifically determines the cargo of fruits to get a big production as constant year after year and high quality (Babuc *et al.*, 2013; Cimpoieș, 2012; Pesteanu, 2013, Stopar, 2000; Robinson *et al.*, 1998).

---

<sup>1</sup>State Agrarian University of Moldova, Chișinău, Republic of Moldova

Some apple varieties are linking abundant large amounts of fruits that are small and of poor quality. As a result, it gains widespread phenomenon of alternating the fruiting trees (Babuc *et al.*, 2013). To adjust the load with fruit trees is necessary to act on fruiting branches, fruit buds, flowers and fruit (Cimpoieș, 2012; Pesteanu, 2014.).

Using fruit chemical thinning with various active ingredients based on NAD, ANA, BA and Ethephon allow developing broader strategy for standardization of fruit load in different varieties (Basak, 2004; Greene, 1993; Robinson *et al.*, 1998; Stopar, 2000).

Ethephon is a harmless regulator, it has a wider range of use, and it increases the average weight of the apples by 10% and improves fruit bud differentiation (Stopar, 2000; Pfammatter *et al.*, 2000; Widmer *et al.*, 2008).

The optimal treatment period starts from the opening of 20% of flowers until the ovaries have a diameter of 22 mm (Pfammatter *et al.*, 2000).

NAD induces early mild thinning, which starts the differentiation process among the flower clusters. This paves the way for a second later treatment, with NAA and BA, which normally hits the weakened fruits, leaving 1 or 2 undisturbed fruitlets per cluster (Greene, 2002; Pesteanu, 2013; Tromp, 2000).

## MATERIAL AND METHOD

The researches were made during the period of 2012 year in the apple orchard founded near the village Radeni from district Strășeni in the autumn of 2009 at the "Codru-ST" Ltd., with trees of a „knip boom” canopy type.

The study object of experience was Idared apple tree variety grafted on M9 rootstock. The trees were trained as slender spindles. Distance of plantation is 3.5 x 1.2 m.

The chemical growth regulators used was Geramid – New, (44.8 g/l NAD), the preparation by the „L. Gobbi Ltd.”, Italy and Cerone 480SL (480 g/l Ethephon), the preparation by the „BAYER Crops Science AG”, Germany.

To optimize the fruit load of the apple trees were experimented the following variants:

- V<sub>1</sub> - Control variant- without chemical treatments;
- V<sub>2</sub> - Geramid-New – 1.5 l/ha;
- V<sub>3</sub> - Cerone 480SL – 0.3 l/ha;
- V<sub>4</sub> - Cerone 480SL – 0.4 l/ha.

In variant two, the treatment with Geramid-New was done when 80% of petals were fallen + 2-3 days (05/05/12) and the third and fourth variants were spray with Cerone 480SL 10 to 20 days after full flowering (15/05/1215).

The effects on the fruit set, the yield, fruit quality at harvest and subsequent blooming were recorded and evaluated according to the following measurements: the number of inflorescences and the number of fruitlets on each tree; the fruit yield produced by each tree, and, for 1 ha orchard of apples; weight of 1 apple; the share of fruits (in %) in size classes based on their diameter from 50 mm to 85 mm with the intervals of 5 mm and relative effectiveness of treatments.



## RESULTS AND DISCUSSIONS

Our investigation demonstrates that the growth regulators as product for chemical thinning of fruit have an essential influence.

If, in the control, where it has not been practiced fruit thinning, the difference between the total number of inflorescences and productive ones was not registered properly, constituted 115 and 111 pcs/tree (tab. 1).

In order not to overload the trees of Idared variety with fruits, it is necessary to carry the standardization on load of fruit through various methods of thinning, either manually or with chemical growth regulators.

Table 1

**The number of total inflorescences (TIN), formed (FIN) in the crown of the variety Idared apple trees and fruit weight in a inflorescence**

Experience variants	TIN, pcs/tree	FIN, pcs/tree	The share of fruits in an inflorescence, %			
			1 pc.	2 pcs.	3 pcs.	>4 pcs.
Control, without treatment	115	111	73.0	19.8	3.6	3.6
Geramid - New, 1.5 l/ha	120	59	81.2	15.4	3.4	-
Cerone 480SL, 0.3 l/ha	116	53	78.6	21.4	-	-
Cerone 480SL, 0.4 l/ha	113	23	60.9	30.4	8.7	-

The research demonstrates that treatment with growth regulators Geramid - New and Cerone 480SL linked to fewer inflorescences in the trees crown (tab. 1).

When treating with growth regulator Geramid - New dose 1.5 l/ha, the number of inflorescences was 59 pcs/tree and decreased by 88.1% compared to the control variant.

When using Cerone 480SL dose 0.3 l/ha, the number of inflorescences was 53 pcs/tree, and when the dose increased up to 0.4 l/ha the key index in the study declined to about 23 pcs/tree, or a decrease of 382.6% compared to the control variant.

The study on fruit weight in an inflorescence shows that in the variant without treatment on 3.6% of inflorescences there were four fruits, three fruits on 3.6%, on 19.8% two fruits and on 73.0% one fruit.

A more rational location of fruit in inflorescence was recorded when treating with Geramid - New in dose of 1.5 l/h and Cerone 480SL in dose of 0.3 l/ha, where 78.6-81.2% of fruits have one blossom, 15.4-21.4% formed by 2 fruits and in 3.4% of inflorescences was obtained 3 fruits.

When treating with the growth regulator Cerone 480SL in dose of 0.4 l/ha, we noticed that the share of inflorescences with one fruit decreases, but those with two or three fruits increases.

After the studied index, it was determined that treatments with Geramid - New in dose 1.5 l/ha and Cerone 480SL in dose of 0.3 l/ha had a positive effect on the number of related inflorescences and the placement of fruits in the trees crown.

The conducted research demonstrates that treating the trees with the growth regulators influences on the number of the fruits in the trees crown which is different according to used dosage.

The largest amount of fruit (tab. 2) was obtained in the control variant - 153 pcs/tree, then in decreasing order is placed version Geramid - New in dose 1,5 l/ha with 71 pcs/tree. When using growth regulator Cerone 480SL dose 0.3 l/ha, the amount of fruits was 57 pcs/tree, and when the dose of Cerone 480SL was 0,4 l/ha the given index decreased from to 34 pcs/tree .

The study on the number of fruit per 100 inflorescences shows regularity similar to that described above properly constituted 133, 59, 49 and 30 pcs.

Table 2

**The influence of growth regulators on yield and fruit quality parameters in the trees crown on Idared apple variety**

Experience variants	Number of fruits, pcs.		Quantity of fruits		Average weight, g	Average diameter, mm
	tree	100 infl.	kg/tree	t/ha		
Control, without treatment	153	133	13.20	31.41	86.3	60.9
Geramid - New, 1.5 l/ha	71	59	12.90	30.70	181.7	77.6
Cerone 480SL, 0.3 l/ha	57	49	11.57	27.53	206.6	79.8
Cerone 480SL, 0.4 l/ha	34	30	7.83	18.63	230.1	85.1
LSD 5%	3,08	-	0.58	1.38	2.61	-

Since the largest number of fruits per tree and on 100 inflorescences was recorded in the control variant, respectively and higher production from a tree and a surface unit was obtained in the given variant. The productivity of a tree was 13.20 kg and 31.43 t/ha per unit surface. An insignificant reduction, demonstrated statistically, was registered in the variant Geramid - New in dose 1.5 l/ha, where fruit production was respectively 12.90 kg/tree and 30.70 t/ha.

When treating the trees with regulator Cerone 480SL in dose 0.3 l/ha, the production declined to 11.57 kg fruit/tree, or a decrease of 3.17 t/ha compared to variant Geramid - New in dose 1.5 l/ha and 3.88 t/ha compared to the control.

The lowest production of fruits was recorded when using the growth regulator Cerone 480SL use in dose 0.4 l/ha, the harvest on a tree representing 7.83 kg and at a unit area 18.63 t.

The quality of the fruit to a great extent depends on the weight and the average diameter of the fruit. Our investigation shows that growth regulators Geramid - New and Cerone 480SL had an essential influence on quality of production.

If in the control variant, the average weight of a Idared fruit was 86.3 g, then in the variants where was made treatment with growth regulator, it was respectively Geramid - New in dose 1.5 l/ha - 181.7 g, with Cerone 480SL dose 0.3 l/ha - 206.6 g, and in the variant Cerone 480SL dose 0.4 l/ha - 230.1 g. The

average weight of the fruit is in direct correlation with the number of fruit remaining in the trees crown.

The average diameter of the fruits is an indicator that directly touches the weight average output obtained. The lowest average diameter was recorded in control variant - 60.9 mm, where trees were without thinning. Further, in increasing order where placed variants where we used of growth regulators such as, Geramid - New in dose 1.5 l/ha - 77.6 mm, Cerone 480SL dose version 0.3 l/ha - 79.8 mm and Cerone 480SL version dose 0.4 l/ha - 85.1 mm.

The obtained results demonstrate that the highest fruit production was recorded when we used chemical thinning with Geramid - New in dose 1.5 l/ha. In version Cerone 480SL dose treatment with 0.3 l/ha, the harvest of fruits decreased insignificantly, but the fruit quality is the highest.

The study was focused not only on knowing the diameter of fruits, but also the fruit division in diameter from 5 to 5mm in correlation with the experience variants (Tab. 3).

Table 3

**The influence of growth regulators on fruit redistribution depending on their diameter on Idared apple variety**

Experience variants	Fruit Share (%) depending on their diameter (mm)						
	55	56-60	61-65	66-70	71-75	76-80	81-85
Control, without treatment	12.3	34.7	42.3	10.7	-	-	-
Geramid - New, 1.5 l/ha	-	-	-	7.0	18.8	51.0	23.2
Cerone 480SL, 0.3 l/ha	-	-	-	1.3	11.8	36.7	50.2
Cerone 480SL, 0.4 l/ha	-	-	-	1.8	2.0	5.2	91.0

The obtained results demonstrate that inferior quality production was obtained in control variant. The largest share of fruits (12.3%) are less than 55 mm diameter, 34.7% with the diameter 56-60 mm, 42.3% with the diameter 61-65 mm and only 10.7% have a diameter of 65-70 mm. Therefore, in variant without thinning, the fruits of the first category were 10.7%.

The used of growth regulators influence on fruit quality. Practically, in all variants exhibited chemical thinning the obtained production assigns to category I and category of extra quality.

When treating with Geramid - New, the share of fruits with the diameter of 66-70 mm is 7.0%, 18.8% with the diameter of 71-75 mm, 51.0% with the diameter of 76-80 mm and 23.2% of fruits with the diameter of 81 mm or higher.

When treating with growth regulator Cerone 480SL in dose of 0.3 l/ha, we record an increase in fruit weight with 81-85 mm diameter (50.2%) and a decrease in the quantity of fruit which is attributable to 66-80 mm diameter (49.8%). When treating with Cerone 480SL in doses of 0.4 l/ha, the share of fruits with 66-70 mm diameter was 1.8%, with 71-80 mm diameter - 7.2%, and having a diameter

greater than 81 mm – 91.0%.

Therefore, convincing results about the fruit diameter were recorded in variants Geramid - New in dose 1.5 l/ha and Cerone 480SL dose 0.3 l/ha.

## CONCLUSIONS

1. The effectiveness of growth regulators Geramid - New and Cerone 480SL in regulating the fruit cargo on Idared variety through chemical thinning influenced on productivity and fruits quality.

2. When using chemical thinning on Idared variety, higher fruit production and a higher quality were recorded when using the growth regulator Geramid-New in dose of 1.5 l/ha to 1000 l/ha solution when 80% of the petals have fallen further 2-3 days.

3. In case of unfavorable weather conditions during treatment with growth regulator Geramid-New, consider using Cerone 480SL dose 0.3 l/ha to 1500 l/ha solution at 10 to 20 days after full flowering.

## REFERENCES

1. Babuc V., Peșteanu A., Gudumac E., Cumanici A., 2013 - *Producerea merelor*. Ed. Bons Offices, Chișinău, 240 p.
2. Cimpoeș Gh., 2012 - *Cultura mărului*. Ed. Bons Offices, Chișinău, 380 p.
3. Basak A., 2004 - *Fruit thinning by using benzyladenine (BA) with ethephon, ATS, NAA, urea and carbaryl in some apple cultivars*. Acta Horticulturae, 653: 99–106.
4. Greene D., 1993 - *A review of the use of benzyladenine (BA) as a chemical thinner for apples*. Acta Horticulturae, 329: 231-236.
5. Greene D., 2002 - *Chemicals, timing, and environmental factors involved in thinner efficacy on apple*. Hortscience 37: 477-480.
6. Peșteanu A., 2013 – *Efficiency of fruitlet thinning apple "Golden Reinders" by use naphthylacetamide acid (NAD)*. Bulletin of University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Horticulture, 70(1): 180-186.
7. Peșteanu A., 2014 – *The influence of thinning agent on base of 6-BA and NAA on productivity and fruit quality of „Gala Must” variety*. Bulletin of University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Horticulture, 72(1): 151-156.
8. Pfammatter W., Bertschinger L., Stadler W., Krebs C., 2000. - *Efficacité et fiabilité de produits éclaircissage en agriculture. Essais avec l'ethephon*. Revue suisse Vitic. Arboric. Hortic. 32(2): 77-79.
9. Robinson T., Lakso A., Stover E., Hoying S., 1998 - *Practical apple thinning programs for New York*. New York fruit quarterly 6: 14-18.
10. Stopar M., 2000. - *Comparison of the most frequently used apple thinning compounds for the thinning of, 'Elstar' and 'Golden delicious' apples*. Res. rep. biot. fac. Agriculture. 75: 89-94.
11. Tromp J., 2000 - *Flower-bud formation in pome fruits as affected by fruit thinning*. Plant Growth Regulation. 31(1–2): 27–34.
12. Widmer A., Golles M., Kockerols W., Stadler W., Christen D., 2008 – *Possibilités et stratégies éclaircissage du pommier à l'ethephon*. Revue suisse Vitic. Arboric. Hortic. 40(2): 87-93.

## THE INFLUENCE OF FOLIAR FERTILIZATION ON APPLE PRODUCTIVITY

### INFLUENȚA FERTILIZĂRII FOLIARE ASUPRA PRODUCTIVITĂȚII LA MĂR

VĂMĂȘESCU S.<sup>1</sup>

e-mail: sergiuvamasescu@mail.ru

**Abstract.** Investigations of the development system foliar fertilization in apple was conducted through research methods and field stationary. As study material served apple trees aged 8 years old Golden Delicious and Idared varieties, grafted on rootstock M26. Distance planting 4 x 2 m. As a foliar fertilizer was used to Urea 46% in a concentration of from 0.4% to 1.2% in the various stages of fruit development, Poly-feed (N<sub>19</sub> P<sub>19</sub> K<sub>19</sub>) at a concentration of 0.1% calcium chloride CaCl<sub>2</sub> at a dose of from 0.5% to 0.7%. In average years of research on fertilized with urea 46% N stages of fruit development: the petals fall of 75%, 20 mm and 30 mm central fruit of the central fruit, fruit number was 172 units in version control and 211 units in variant V4.

**Key words:** apple tree, rootstock, urea, yield buds, variety.

**Rezumat.** Ca material biologic s-au folosit soiurile Golden Delicious și Idared, altoite pe portaltoiul M26, vârsta pomilor fiind de 8 ani.. Distanța de plantare 4 x 2 m. Ca fertilizant foliar s-a utilizat Uree 46 %<sub>s.a.</sub> în concentrație de la 0,4 % până la 1,2% în diferite faze de dezvoltare a fructelor, Poly-feed (N<sub>19</sub> P<sub>19</sub> K<sub>19</sub>) în concentrație de 0,1% și clorură de calciu CaCl<sub>2</sub> în doze de la 0,5% la 0,7 procente. În anii de cercetare, în urma fertilizării cu uree 46%N, în fazele de dezvoltare a fructelor: de la căderea a 75% din petale, la 20 mm a fructului central și la 30 mm a fructului central, numărul mediu de fructe a fost de 172 buc/pom în varianta martor și 211 buc/pom în varianta V4.

**Cuvinte cheie:** măr, mugur floral, portaltoi, soi, uree.

## INTRODUCTION

Foliar fertilization in horticulture is particularly effective because it concentrates are used which use high purity technical elements and the nitrogen, phosphorus, potassium and other elements are combined into a desirable balance in the medium (Cimpoieș, 2013). By this method give a balanced fertilizer, which not only ensure the NPK but all trace elements and hormones of growth and development (Ghena *et al.*, 2004, Balan, 2009).

As a result, foliar fertilizers can influence the growth of shoots, fruit bud differentiation, flowering and fruit tying, low productivity and other

---

<sup>1</sup> State Agrarian University of Moldova, Chișinău, Republic of Moldova

characteristics of plant (Robinson and Lopez, 2012).

At the same time fruit crops need different amounts of nutrients in the vegetation phenophases (Balan and Vămășescu, 2012). Therefore, the present time of application, equipment usage and environmental factors to be taken into account when applying foliar nutrients (De Angelis *et al.*, 2011; Klein, 2002).

## MATERIAL AND METHOD

Investigations were effectuated in 2011- 2014 in the orchard of apple company "Zubrești" SA, planted with trees grafted aged 1 year Zubrești near the village, Strasenii. Plantation was conducted in spring 2003 with Golden Delicious and Idared varieties grafted on rootstock M26. Distance 4 x 2 m planting trees. Trees were led by slim spindle-shaped crown.

Research is carried out by general methods of tree species experiences. Nitrogen is used in the form of urea to 46% (NH<sub>2</sub>)<sub>2</sub>CO consuming solution to each 1000 liters per hectare in the respective concentration. Spray solution pH was neutral weak acid. Poly-Feed is a high-quality NPK fertilizer, totally soluble in water, pH 5-6, free chlorine, heavy metals and other harmful elements.

Trace elements in the form of chelați: Mn, Cu, Zn, Fe, Mo Calcium is used in the form of calcium chloride CaCl<sub>2</sub>. Alternatively witness trees served sprinkled with water (Tab. 1). Spraying was done in the morning when the wind is minimal and lower temperature on both sides of the leaves.

Table 1

Scheme applying apple foliar fertilizers

No.	The period of effect foliar fertilization	Foliage fertilizer concentration			
		V1	V2	V3	V4
Urea 46 % active substance					
1	after bloom (when the 75% where in bloom)	water	0.4	0.5	0.6
2	When the fruit is size one nuts(fruit have 10-12 mm in diameter)	water	0.7	0.8	0.9
3	When the fruit are in size one walnuts (fruit have 25-30mm in diameter)	water	1.0	1.1	1.2
Polyfeed (N19 : P19 : K19)					
4	When fruits are in the ripen stages(20-30 July)	water	0.1	0.1	0.1
Calcium chloride(CaCl <sub>2</sub> )					
5	With 20-30 days before harvest	water	0.5	0.6	0.7

The number of fruits, weight and production as a tree were determined 10 days before harvesting the fruit from the calculations 3 each variant.

## RESULTS AND DISCUSSIONS

The number of fruit research years (2011-2014) by variety and concentration of fertilizer applied, Golden Delicious variety was from 100 pcs 223 pcs. In version control their number tends to increase by the year 2012 to 170

units, and again in 2014 to reach 100 pcs. In foliar fertilization variants fruit number varies depending on the applied concentration of urea 46% N.

In the variant V3 where the concentration of the fertilizer was 0.5%; 0.8%; 1.1%. The fruit number was from 169 units in 2011 to 204 units in 2014. The average for the years of research at the Golden Delicious variety fruit number was from 129 units in the control variant V1 to V4 variants 195 units applying a urea concentration of 46% N 0.6%; 0.9%; 1.2%.

The number of fruits at Idared variety in 2011 amounted to 155 units in the control variant V1 to V4 variants 195 units applied where the highest concentration of urea 46% N. In variant V2 where the concentration of urea 46% N was the lowest number was 170 units of fruit. In 2012 the number of fruits increased from the year 2011 and amounted to 184 units from 233 units in control variant in 2014. This increase in the number of fruit in fertilizer variants compared to the control is from 9.7% in variant V2 to 26.6% for the variant V4 (Tab. 2).

In the years 2013 -2014 fruit number was lower in 2013 due to deposit one small number of fruit buds in 2012 and amounted to 148 units in variant control or 20% less than in 2012-187 units or 22% less than in 2012. In 2014 the number of fruit in all variants investigated increased compared to 2013 and amounted to 200 units in the control variant V1 to V4 variant 227 units.

Table 2

**Number of fruit trees apple on the application of foliar fertilization  
with mineral fertilizers, pcs.**

*(M26 rootstock, 4x2m planting distance, trees aged 8-11 years, SA "Zubrești")*

Variant	Years				Average (2011 – 2014)
	2011	2012	2013	2014	
Golden Delicious variety					
V <sub>1</sub>	100	170	146	100	129
V <sub>2</sub>	152	190	165	190	174
V <sub>3</sub>	169	200	174	204	187
V <sub>4</sub>	171	223	186	200	195
DL	2,55	5,41	1,93	3,17	-
Idared variety					
V <sub>1</sub>	155	184	148	200	172
V <sub>2</sub>	170	202	167	204	186
V <sub>3</sub>	186	219	176	225	202
V <sub>4</sub>	195	233	187	227	211
DL	3,28	4,19	3,30	2,27	-

In average years of research on fertilized with urea 46% N stages of fruit development: the petals fall of 75%, 20 mm and 30 mm central fruit of the central fruit, fruit number was 172 units in version control and 211 units in variant V4.



Another indicator studied is the weight of the fruit, which Golden Delicious variety in 2011 was from 114 g to 134 g. In 2012 the average weight of fruit increased in all variants (Tab. 3).

The largest fruit weight was recorded in the V4 variant with 171 g, and the lowest in control variant with 139 g of fruits such weight gain of control variant and fertilization variants amounted to 23%. In 2014 fruit weight remained at the same high level of control variant but fruit weight was greatly reduced at 70 g reaching a fruit. In average years of research at Golden Delicious variety of foliar fertilization using urea 46% based on N has increased the weight of the fruit depending on the concentration of fertilizer applied up to 31 percent.

Idared variety of research years (2011-2014), fruit weight recorded values of 99 g in the control, but in variants with foliar fertilization, this index made up of 108 g to 119 g. In the years 2012-2013 this increasing trend of average fruit weight was maintained indicating the largest fruit in the V4 version where foliar fertilizer dose based on urea 46% N was in various stages of fruit development from 0.6% to 1.2 %.

In 2014 and Golden Delicious variety fruit witness variant recorded average fruit weight of 63 g or less than 174% fruit weight in V4 version.

In average years of research (2011- 2014) 46% urea N applied in different phenophase of fruit development has made a positive impact on recorded values of fruit weight by 40% compared to the control.

Table 3

**The weight of the fruit trees with foliar fertilization  
with mineral fertilizers application, g.**

(M26 rootstock, 4x2m planting distance, trees aged 8-11 years, SA "Zubrești")

Variant	Years				Average (2011 – 2014)
	2011	2012	2013	2014	
Golden Delicious variety					
V <sub>1</sub>	134	139	120	70	116
V <sub>2</sub>	114	154	129	164	140
V <sub>3</sub>	133	167	136	167	151
V <sub>4</sub>	134	171	144	160	152
DL	1,91	3,74	2,52	3,76	-
Idared variety					
V <sub>1</sub>	99	130	127	63	105
V <sub>2</sub>	108	152	130	140	133
V <sub>3</sub>	108	142	129	169	137
V <sub>4</sub>	119	158	136	173	147
DL	2,81	2,19	2,75	4,07	-

One of the most important indicators is studied fruit production.

Golden Delicious variety fruit harvest per hectare in 2011 was 28.8 t / ha in the control, but crop fertilization variants ranged from 32.8 t / ha in V2 and 33.3 t / ha in V3. In 2012 harvest in all variants has increased since 2011, reaching 47.7 t / ha in the variant with the highest concentration of urea 46% N applied (V4).



In 2013 fruit harvest decreased in all variants but just as in 2012 the largest harvest was in V4 with 32.0 t / ha (Tab. 4).

Table 4

**Fruit production based application foliar fertilization with mineral fertilizers t / ha.**  
(M26 rootstock, 4x2m planting distance, trees aged 8-11 years, SA "Zubrești")

Variant	Years				Average (2011 – 2014)
	2011	2012	2013	2014	
Golden Delicious variety					
V <sub>1</sub>	28,8	46,6	29,4	8,7	28,3
V <sub>2</sub>	32,8	36,6	29,5	42,6	35,3
V <sub>3</sub>	33,3	40,0	30,1	39,9	35,8
V <sub>4</sub>	32,9	47,7	32,0	40,0	38,2
DL	1,59	1,95	1,35	2,96	-
Idared variety					
V <sub>1</sub>	19,2	29,9	23,4	15,8	22,0
V <sub>2</sub>	22,9	38,4	27,3	39,3	31,9
V <sub>3</sub>	25,1	38,9	28,3	43,1	33,8
V <sub>4</sub>	29,0	46,0	32,2	49,1	39,0
DL	3,17	2,26	1,05	3,13	-

In 2014 fruit production in foliar fertilization variants constituted around 40 t / ha. In version control fruit harvest was only 8.7 t / ha this is due to a very weak deposits of fruit buds in 2013 against a backdrop of large harvests of fruit inhibited the process.

In average years of research (2011 - 2014) application of foliar fertilization Golden Delicious variety has a beneficial impact on a constant crop.

Idared variety during research (2011-2014) recorded productivity per hectare from 15.8 t / ha to 49.1 t / ha. In 2011 harvest per hectare was to 19.2 t / ha in control variant, and fertilization variants harvest is over 23 tons. As Golden Delicious variety in harvest of fruit in all variants was investigated over 38 tones. In the years 2014 harvest V3 and V4 variants, which received the largest amount of foliar fertilizer, fruit production per hectare was over 40 tones. In average years of research (2011 - 2014) highlights that we harvest the fruit is responsive Idared variety of applications such Urea 46% N harvest V4 version where the concentration was highest at 0.6%; 0.9%; 1.2% productivity was 39.0 tons per hectare.

## CONCLUSIONS

1. In the variant V3 where the concentration of the fertilizer was 0.5%; 0.8%; 1.1%, fruit number was from 169 units in 2011 to 204 units in 2014. The average for the years of research at the Golden Delicious variety fruit number was from 129 units in the control variant V1 to V4 variant 195 units applying a urea concentration of 46% N 0.6%; 0.9%; 1.2%.

2. In average years of research (2011- 2014) 46% urea N applied in different phenological phases of fruit development has made a positive impact on recorded values of fruit weight by 40% compared to the control.

3. In average years of research (2011 - 2014) highlights that we harvest the fruit is responsive Idared variety of applications such Urea 46% N harvest V4 version where the concentration was highest at 0.6%; 0.9%; A 1.2% productivity was 39.0 tons per hectare.

## REFERENCES

1. Angelis V., De Sánchez E., Tognetti J., 2011 - *Timing of nitrogen fertilization influences color and anthocyanin content of apple (Malus domestica Borkh. cv 'Royal Gala') fruits*. Journal International Journal of Fruit Science, 11(4): 364-375
2. Balan V., 2009 - *Sisteme de cultură în pomicultură. Randamentul producției de fructe*. Academos, Chișinău, nr. 4 (15): 82-90.
3. Balan V., Vămășescu S., 2012 - *Apple foliar surfaces în function of foliar fertilizer application*. Lucrări științifice USAMV București, p. 342- 346.
4. Babuc V., 2013 - *Pomicultura*. Chișinău: Tipografia Centrală, 662 p.
5. Cimpoeș Gh., 2013 - *Cultura mărului*. Chișinău: Bonus Offices, 382 p.
6. Ghena N., Braniște N., Stănică F., 2004 - *Pomicultura generală*. Buc/pomurești: Matrix Rom., 562 p.
7. Klein I., 2012 - *Nitrogen pool enrichment in fruit trees for specific target requirement*. Journal Acta Horticulturae 2002(594): 131-137
8. Robinson T., Lopez S., 2012 - *Crop load affects 'Honeycrisp' fruit quality more than nitrogen, potassium, or irrigation*. Journal Acta Horticulturae, 940: 529-537
9. Stampar F., Hudina M., Usenik V., Sturm K., Veberic R., Veber G., 2002 - *Experience with foliar nutrition in apple orchard*. Journal Acta Horticulturae 594: 547-552
10. Wargo J. M., Merwin I. A., Watkins C. B., 2003- *Fruit size, yield, and market value of 'GoldRush' apple are affected by amount, timing and method of nitrogen fertilization*. Journal Hort. Technology, 13(1 : 153-161.

## **KNIPHOFIA MOENCH. SPECIES OF PERSPECTIVE FOR ARRANGEMENT OF GREEN AREAS OF REPUBLIC OF MOLDOVA**

### **KNIPHOFIA MOENCH. SPECII DE PERSPECTIVĂ PENTRU AMENAJAREA SPAȚIILOR VERZI DIN REPUBLICA MOLDOVA**

**SFECLĂ Irina<sup>1</sup>**

**e-mail:** irinasfecla@gmail.com

**Abstract.** *There is no doubt that the main element of amenity planting is flowers, their species are numerous and their diversity is enormous. Therefore, it is important to select plants that will meet both aesthetic requirements and environmental conditions. Kniphofia genus belongs to the family Asphodelaceae Juss. and includes about 70 species. A variety of colors and the original structure of flowers, beautifully situated on a high spike, decorative foliage, the continuing until frost, heat - and drought tolerance, simplicity of habitat conditions and relatively few diseases and pests, do species of the genus Kniphofia priority when choosing the assortment for gardening. To assess decorative qualities of certain kniphofia types we followed the methodology developed in the "Floriculture" laboratory, the Botanic Garden A.N.M. (1991). Kniphofia uvaria Hook was used as a control group. The evaluation was carried out on the basis of the five-point scale, regarding every indicator and was multiplied by the corresponding coefficient. As a result, they summed up all the points, the maximum index number was 100 points. This study was repeated for three times for each species separately.*

**Key words:** *Kniphofia, ornamental characters, evaluation decorative qualities.*

**Rezumat.** *Principalul element care aduce culoare în cadrul unei amenajări peisagere sunt plantele floricole, a căror diversitate specifică este enormă. În acest caz, este importantă alegerea corectă a sortimentului utilizat, care trebuie să corespundă atât cerințelor estetice, cât și condițiilor staționale. Genul Kniphofia face parte din familia Asphodelaceae Juss. și include aproximativ 70 de specii. Varietatea mare de culori și aspectul original al florilor dispuse în inflorescențe spiciforme, frunzișul decorativ care se menține până la îngheț, toleranța la căldură și secetă, simplitatea condițiilor de mediu și numărul mic de boli și dăunători, determină alegerea cu prioritate a speciilor genului Kniphofia pentru grădini. Pentru o evaluare a calităților decorative ale unor specii de kniphofii, am utilizat metodologia dezvoltată în laboratorul de "Floricultură" al Grădinii Botanice a AȘM (1991). Kniphofia uvaria Hook a fost folosită ca grup de control. Evaluarea a fost efectuată pe baza unei grilei de cinci puncte la diverși indicatori și înmulțită cu coeficientul corespunzător. Ca urmare s-au însumat punctele, maximul fiind 100 puncte. Acest studiu a fost repetat de trei ori pentru fiecare specie separat.*

**Cuvinte cheie:** *Kniphofia, caractere ornamentale, evaluare calități decorative.*

---

<sup>1</sup> Botanical Garden (Institute) of the Academy of Sciences of Moldova, Chișinău, Republic of Moldova

## INTRODUCTION

There is no doubt that the main element of amenity planting is flowers, their species are numerous and their diversity is enormous. Therefore, it is important to select plants that will meet both aesthetic requirements and environmental conditions (Академия Наук Молдавской ССР, 1986).

The usage of new and naturalized species is a basic principle when selecting plants for landscape gardening. It in its turn requires a careful study of biological and agronomic features of flowering plants in specific climate conditions.

Herbaceous perennials, such as kniphofias are multi-functional, ornamental plants, which, if properly cared for, can bring you joy by their beauty and elegance for years.

## MATERIAL AND METHOD

To assess decorative qualities of certain kniphofia types (*Kniphofia uvaria* Hook; *K. nelsonii* Mast.; *K. Tukii* Baker.; *K. citrina* Baker.; *K. ensifolia* Baker.) we followed the methodology developed in the "Floriculture" laboratory, the Botanic Garden (I) A.N.M. (1991). *Kniphofia uvaria* Hook was used as a control group.

## RESULTS AND DISCUSSION

*Kniphofia* Moench. (kniphofia, torch-lily) is genus with perennial, rhizomatous, herbaceous plants of the *Asphodelaceae* family (Bailey, 1947; Tony, 2003). There are about 70 species of the genus that grow in Southern and Central Africa and on Madagascar (Bailey, 1947; Preda, 1989; Tony, 2003). Plants that are from 60 to 120 cm high have short, thick rhizomes. Kniphofia's leaves are ensiform, thick and leathery, collected in a radical and thick rosettes. The flowers are small (1.5 - 2.0 cm), campanulate, bent, of the red, coral, orange or yellow colour. The thick, spicate inflorescences are placed at a big, leafless, thick, rounded flower-bearing stem. The fruit is a capsule (Bailey, 1947; Tony Lord, 2003).

If you grow plants on the field you should follow some natural biological processes and terms of the genus development (Академия Наук Молдавской ССР, 1986). Since kniphofias are plants of an average winter hardiness, they may be cultivated on the field in the Republic of Moldova, taking into consideration its climate conditions.

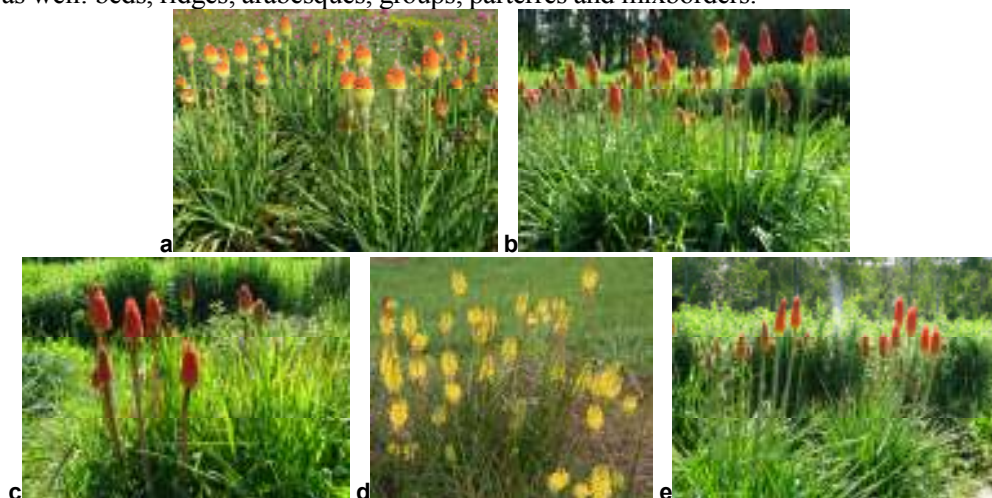
This plant can be bred both in a generative and vegetative ways. Generative breeding is a longer method, considering the fact that the plants appeared from seeds will bloom in the second or the third year of their life. The vegetative breeding is primary. In this case plants bloom in the same year (the spring division of a bush), or the next year (the autumn division of a bush) (Preda, 1989; Selaru, 2007).

A variety of colours and flowers of the original structure, beautifully placed at a big flower-bearing stem, decorative foliage that is preserved until the frost, drought resistance, easy management in local environmental conditions, and the fact that they are relatively rarely affected by diseases and pests, make species of the *Kniphofia* genus to be the best when selecting an assortment (Fig. 1).

From the architectural point of view, the above mentioned plants can be used in different styles, both in regular forms and in free planning projects. The kniphofias may be used as single plants as well as in various floral associations.

To highlight their decorative qualities in single beddings kniphofias are used against a bright green background of a lawn that pleasantly contrasts with the red color of flowers. Their decorativeness in single planting is flawless in every respect: the decorativeness and originality of the foliage, a beautiful habit, multiple and continuous blooming.

This type can be attributed to architectural plants. British landscape designers suggested using the term to refer to the plants with an interesting habit. They are planted as single plants and can play the role of small architectural forms in the garden space ([http://articles.m-strou.ru/article\\_1215.html](http://articles.m-strou.ru/article_1215.html)). Torch-lilies integrate well in floral compositions as well: beds, ridges, arabesques, groups, parterres and mixborders.



**Fig.1** - The studied species (blossoming period)

a. *Kniphofia uvaria* Hook; b. *K. nelsonii* Mast.; c. *K. Tukii* Baker.; d. *K. citrina* Baker.; e. *K. ensifolia* Baker.

The colour of the plant and its compatibility are not less important. It is the colour which can affect a person's emotional state and mood. The inflorescence of the genus *Kniphofia* may have different shades of yellow, orange and red. These colours are described as warm and energetic at the same time which attracts our attention. Warm colours are generally used in amenity planting. Due to the fact that the yellow-red colour attracts our attention, kniphofias should be better used to emphasize interesting places or significant landscape elements. On the other hand the red colour is mixed well with almost the entire palette of colours ([http://articles.m-strou.ru/article\\_1215.html](http://articles.m-strou.ru/article_1215.html)).

To assess decorative qualities of certain kniphofia types we followed the methodology developed in the "Floriculture" laboratory, the Botanic Garden (I) A.N.M. (1991). *Kniphofia uvaria* Hook was used as a control group. The evaluation was carried out on the basis of the five-point scale, regarding every indicator and was

multiplied by the corresponding coefficient. As a result, they summed up all the points, the maximum index number was 100 points. This study was repeated for three times for each species separately.

Taking into consideration the results received (Table 1), we can say that these species have proved to be very promising and decorative in the climate conditions of the Republic of Moldova.

Table 1

**The evaluation of decorative qualities of some *Kniphofia* genus species**

The species	The decorativeness evaluation on the basis of the 5-point scale - 1; Coefficient -2																				The total number of points (out of 100)		
	The colour of the plant (3)		The size of the flower (2)		The form of the flower (1)		Th flower texture (1)		The number of flowers at a flower-bearing stem (2)		The abundance of blossom (2)		The resistance of the flower-bearing stem (2)		The decorative ness of the vegetative part (2)		The uniformity of plants (2)		The originality of plants (2)			The condition of plants (1)	
	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	
<i>K. uvaria</i> Hook.	4	12	4	8	5	5	5	5	5	10	5	10	5	10	5	10	5	10	5	10	5	5	98
<i>K. tukii</i> Baker.	4	12	4	8	5	5	5	5	5	10	5	10	5	10	5	10	5	10	5	10	5	5	98
<i>K. ensifolia</i> Baker.	5	15	4	8	5	5	5	5	5	10	5	10	5	10	5	10	5	10	5	10	5	5	99
<i>K. nelsonii</i> Mast.	5	15	4	8	5	5	5	5	5	10	5	10	5	10	5	10	5	10	5	10	5	5	99
<i>K. citrine</i> Baker.	5	15	5	8	5	5	5	5	5	10	5	10	5	10	5	10	5	10	5	10	5	5	99

## CONCLUSIONS

1. After evaluating, the studied species have accumulated 98 (*K. uvaria* and *K. tukii*) and 99 (*K. ensifolia*, *K. nelsonii* and *K. citrine*) points out of 100. This allows us to find that kniphofias represents species of perspective for Moldova's climatic conditions.

2. Red-orange-yellow of kniphofia inflorescences can be combined easily with almost all color palette used in arrangement of green areas.

3. For plant multiplication, more effective is using of vegetative propagation. This method offers us flowering plants in the next season of vegetation. If we using generative multiplication, the plant reaches the generative phases (flowering, fructification) within 3-4 years of the life cycle.

## REFERENCES

1. Bailey L., 1947 - *The standard cyclopedia of horticulture* Vol. II. Ed. The Macmillan Company, New York., p. 1752-1755.
2. Preda M., 1989 - *Dicționar dendrofloral*. Ed. Științifică și Enciclopedică, București, p. 305-306.
3. Șelaru E., 2007 - *Cultura florilor de grădină*. Ed. Ceres, București, 645 p.
4. Tony Lord, 2003 - *Flora The gardener's bible* Vol. I. Ed. „Cassell” London, p. 780-781.
5. Академия Наук Молдавской ССР, 1986 - *Интродукция, отдаленная гибридизация растений, озеленение*, Кишинев «Штиинца», с. 104-114.
6. [http://articles.m-strou.ru/article\\_1215.html](http://articles.m-strou.ru/article_1215.html)



## THE BEHAVIOR OF GRAPE VINE VARIETIES GELU AND PAULA AT GRAFTING

### COMPORTAREA LA ALTOIRE A SOIURILOR DE VIȚĂ DE VIE GELU ȘI PAULA

ALEXANDRU L.C.<sup>1</sup>, ROTARU Liliana<sup>1</sup>, NECHITA Ancuța<sup>2</sup>,  
DAMIAN Doina<sup>2</sup>, COLIBABA Lucia Cintia<sup>1</sup>  
e-mail: lulu75cata@yahoo.com

**Abstract.** Table grapes present some specific biological properties that distinguish them from wine grapes. The high production potential, the production being influenced in particular by choosing the best combinations of scion - rootstock, knowing that rootstock can affect both production and quality, is very important. High and very high vigor of the vines requires the use of certain varieties of rootstock with the same vigor that can support high production potential. In this work, two new creations of grape vines obtained at SCDVV Iasi, respectively Gelu and Paula were studied, in terms of their behavior after grafting, compared to a reference variety, Aromat de Iași. The reaction to grafting and the behavior during nursery period emphasized that the two varieties had a good behavior, between 73 and 88% of grafted and forced vines presented circular callus and new shoots, lower values than the control variety (92%), and the STAS obtained vines had values between 44-47% at Gelu, 46-55% at Paula, also lower than control variety Aromat de Iași (49 - 54%).

**Keywords:** grapevine varieties, rootstock, grafting, nursery

**Rezumat.** Soiurile de struguri pentru masă prezintă unele însușiri biologice specifice, care le diferențiază de cele pentru struguri de vin. Potențial de producție ridicat, nivelul de producție al acestora fiind influențat în special, de alegerea celei mai bune combinații altoi-portaltoi, știut fiind faptul că portaltoiul influențează atât producția cât și calitatea acesteia. Vigoarea mare și foarte mare de creștere a butucilor, impune folosirea unor soiuri de portaltoi cu aceeași vigoare, care să susțină potențialul ridicat de producție. În lucrare au fost luate în studiu două creații noi de viță de vie obținute la SCDVV Iași Gelu și Paula și studiate sub aspectul comportării lor la altoire, față de soiul martor Aromat de Iași. Comportarea la altoire și în școala de vițe, evidențiază faptul că cele două soiuri au avut un comportament bun, între 73 și 88% din vițele altoite și forțate având calus circular și lăstari porniți, valori inferioare soiului martor (92%), iar randamentul în vițe STAS a avut valori cuprinse între 44 - 47% la soiul Gelu, 46 - 55% la soiul Paula, de asemenea, mai mici decât soiul martor Aromat de Iași (49 - 54%).

**Cuvinte cheie:** soi de viță de vie, portaltoi, altoire, școală de viță de vie

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> Vine and Wine Research and Development Station of Iași, Romania

## INTRODUCTION

Table grape varieties have some specific biological characteristics, that differentiate them from grape varieties used for wine-making. Knowing these characteristics has a major importance being the basic line for elaborating specific cultural technologies capable of insuring high and constant yields, qualitative and efficient from an economical point of view (Calistru și Damian, 1986).

The different aspects of grafting were studied, over the years, by many researchers. Juncu, in 1958, underlines a good grafting affinity between rootstock Riparia gloire and the very good rooting capacity of rootstock Kober 5BB. Baltagi (1960-1968) underlines the influence of the maturation degree of the wood on calus formation, roots development, growth and maturation of vine shoots. These conclusions were confirmed by the studies made by Văleanu et al., 1973. Oșlobeanu et al., 1975, in Murfatlar conditios, proving the superiority of rootstock Riparia Grand Glabre, 44 and 53 Malegue and Riparia 106.8 MG concerning the percentages of STAS vines obtained (Dobrei și colab., 2005).

Grecu (1990) focuses in the superiority of the rootstock Ru 140 that insured a high yield of STAS vines. In SCDVV Bujoru, Simion underlines the excellent grafting behaviour of Ru 140 as well as rootstock SO<sub>4-4</sub>. Calistru et al., 1994 recommend the use of rootstock SO<sub>4</sub> for Aromat de Iași grape variety in Iași vineyard. Profir (1997) recommends for viticultural center Copou Iași the use of SO<sub>4</sub> and Ru 140, with better grafting behaviour and yields.

## MATERIAL AND METHOD

The studied table grapes varieties, respectively Gelu, Paula and control sample Aromat de Iași have been grafted on the same rootstock, namely Berlandieri x Riparia selection Oppenheim 4, clone SO<sub>4-4</sub> and were forced in the same conditions. Temperatures of 32°C and 85% humidity for the first 3 days were used, while beginning with the fourth days, temperatures were brought down to 28°C, thus the average period for forcing was of 15 days.

After forcing, the grafted vines (fig. 1) were acclimatised for 24 – 48 ore at temperatures of 8 – 10°C, following quality classification that took into account the existing shoot and the existing circular calus (fig. 2).



**Fig. 1** - Grafted cuttings bins at the end of forcing





**Fig. 2** - The aspects concerning caluss and shoots formation on grafted cuttings after forcing process is finished

## RESULTS AND DISCUSSIONS

The classification of grafted vines showed that Gelu and Paula grape varieties had a good behaviour, nevertheless inferior to that of control variety, Aromat de Iași.

Table 1

**Behavior to forcing of the varieties concerned**

Studied elements	Paula			Gelu			Aromat de Iași		
	2012	2013	2014	2012	2013	2014	2012	2013	2014
Total vines, %	100	100	100	100	100	100	100	100	100
Vines with circular calus and shoot, %	84	80	73	86	88	78	90	92	91
Vines with circular calus without shoot, %	12	10	15	7	6	10	6	4	5
Vines with partial calus without shoot, %	7	6	6	4	2	6	1	1	1
Rejects, %	7	4	6	3	4	6	3	3	3

The analysis has shown that, in the three years of research, Paula grape variety had an average of 79% grafted vines with circular calus and shoot, Gelu grape variety presented 83% grafted vines with circular calus and shoot, both under the average of Aromat de Iași with 92% quality grafted vines.

**Vegetative development in the nursery.** The analysis made during intense growth period showed that the grafted vines had good growth potential, higher in 2013 than in 2014. The measurements regarding the average length of shoots in the nursery show that Gelu grape variety, grafted on rootstock Berlandieri x Riparia selection Oppenheim 4, clone SO<sub>4-4</sub>, had the biggest growths, 63,6 cm in

2012, 70,3 cm in 2013 and 59,5 cm in 2014, the average lengths of a shoot being superior to that of control sample Aromat de Iași with 57,3 respectively 51,0 cm (fig. 3).

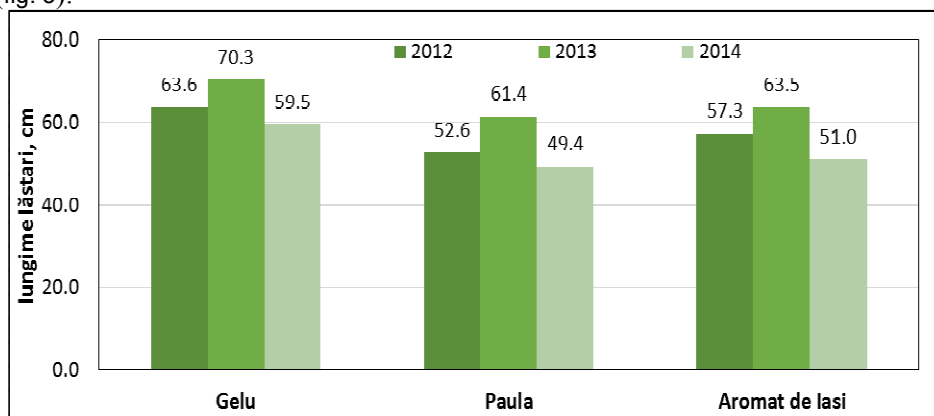


Fig. 3 - The average length of shoots in the wine-growing nursery

Paula grape variety, grafted on the same rootstock, had inferior values of the shoots' length (49,4 and 61,4 cm) compared to the control sample. These values show that the analysed varieties, including the control sample, had favourable climatic conditions and proper agrophytotechnical measures: phytosanitary measures, irrigation, soil tillage.

**Standard vines yield.** At the end of the vegetation period, after the grafted vines are dug out of the nursery and classified, biometrical measurements are made regarding the total number of roots, roots with a diameter over 2 mm, length of the matured shoot and width of shoot at the second internode.

Table 2

Quality of the vines produced in the wine-growing nursery in the year 2012

Grape variety	Repetition	STAS vines, %	No. Roots/ vine		Length of matured shoot, cm	Width of shoot at the 2nd internode, mm
			total	Ø > 2 mm		
Gelu	1	44%	7,51	2,80	26,30	6,15
	2		7,30	2,63	28,25	6,30
	3		8,40	2,54	27,8	5,75
	Average		7,74	2,66	27,45	6,06
Paula	1	46%	8,35	3,15	25,30	5,18
	2		8,80	3,00	22,45	6,00
	3		9,05	2,70	23,35	5,95
	Average		8,73	2,95	23,70	5,71
Aromat de Iași (control)	1	49%	8,16	2,45	23,50	6,15
	2		9,85	2,10	24,60	5,90
	3		10,66	2,15	24,85	6,00
	Average		9,55	2,23	24,32	6,01

Based on all measurements for the studied grafted varieties, the yield of STAS vines was obtained: 44 - 47 % for Gelu variety, 46 - 55% for Paula variety, both lower than the results obtained for the control sample Aromat de Iași with a yield of 49 - 56% (tab. 2, 3 și 4).

The studies regarding the rooting capacity of the used rootstock Berlandieri x Riparia selection Oppenheim 4, clone SO<sub>4-4</sub> and its influence on the graoe variety, length of the matured shoot and diameter of the shoot at the 2nd internode, showed that the grafted vines have at least 7 roots/vine, at least 2 with over 2 mm diameter, the matured length of the shoot being over 20 cm, the diameter of the second internode being between 5 and 6 mm. These results express a good compatibility between the two grafting partners, aspect which recomand their use in the grafting process.

Table3

Quality of the vines produced in the nursery in the year 2013

Grape variety	Repetition	STAS vines, %	No. Roots/ vine		Length of matured shoot, cm	Width of shoot at the 2nd internode, mm
			total	Ø> 2 mm		
Gelu	1	46%	8,30	3,33	28,92	6,10
	2		8,00	2,84	30,88	5,95
	3		9,15	2,75	28,75	6,12
	Average		8,48	2,97	229,52	6,05
Paula	1	51%	9,25	2,75	23,60	6,10
	2		8,40	3,05	24,05	5,20
	3		9,10	2,80	22,75	5,30
	Average		8,92	2,87	23,47	5,53
Aromat de Iași (control)	1	54%	10,15	2,30	23,40	5,30
	2		9,30	1,85	25,25	6,00
	3		11,25	2,25	24,35	5,25
	Average		10,23	2,13	24,33	5,52

Table 4

Quality of the vines produced in the nursery in the year 2014

Grape variety	Repetition	STAS vines, %	No. Roots/ vine		Length of matured shoot, cm	Width of shoot at the 2nd internode, mm
			total	Ø> 2 mm		
Gelu	1	47%	8.48	2.92	28.90	6.47
	2		7.48	2.48	30.48	6.46
	3		9.28	2.96	29.32	5.72
	Average		8.41	2.79	29.57	6.22
Paula	1	55%	9.12	3.00	22.48	5.08
	2		9.00	3.16	24.92	4.73
	3		8.28	2.64	24.44	4.69
	Average		8.80	2.93	23.95	4.83
Aromat de Iași (control)	1	56%	8.28	2.40	23.12	6.28
	2		12.12	1.52	25.36	4.45
	3		9.68	2.04	24.98	5.01
	Average		10.03	1.99	25.17	5.25

## CONCLUSIONS

1. Nursery behaviour during 2012-2014 shows that the two grape varieties had between 73 and 88% grafted vines with circular calus and shoots, nevertheless inferior to that of the control sample - 92%.

2. STAS vines yield was between 44 - 47% at Gelu grape variety, 46 - 55% at Paula grape variety, lower than the control sample, Aromat de Iași, with values of 49 - 54%.

3. Therefore, the two table grape varieties Gelu and Paula, created at SCDVV Iași, are considered well adapted to the specific conditions of the ecosystem in which they have been created. They are also recommended for culture on wide surfaces in the North-East of Moldova or other regions with similar conditions.

4. Using these varieties insures fresh table grapes in a period when the varietal deficit is high.

*Acknowledgement: This work was secured from the European Social Fund, through Sectorial Operational Program Human Resources Development 2007 - 2013, the draft POSDRU/ 159/ 1.5/S/132765 "Doctorale and postdoctorale programs to promote excellence in research, development and innovation in the priority areas - agronomic and veterinary medical, of the knowledge-based society".*

## BIBLIOGRAPHY

1. **Calistru Gheorghe, Damian Doina, Crăcană Alexandru, 1994** - *Afinitatea de producție a soiului Aromat de Iași cu cei mai buni parteneri de viță portaltoi*, Cercetări agronomice în Moldova, Vol. 3-4, Iași.
2. **Calistru Gheorghe, Doina Damian, 1986** – *Potențialul biologic al unor soiuri de viță de vie pentru struguri de masă, folosită ca sursă de germoplasmă*. Cercet. agron. în Moldova, vol. 1, Iași.
3. **Dobrei Alin, Rotaru Liliana, Mustea Mihai, 2005** - *Cultura viței de vie*. Editura "Solness", Timișoara.
4. **Grecu Virgil, 1990** - *Cercetări privind afinitatea de altoire a unor soiuri noi de viță de vie pentru struguri de masă*. Anale I.C.V.V., Vol. XIII.
5. **Profir Constantin, 1997** - *Studiul optimizării combinațiilor portaltoi - soi vinifera în podgoria Iași*. Teza de doctorat U.A.M.V. Iași.
6. **Simion Cristina, 1998** – *Studiul biosistemului soi vinifera-portaltoi, în vederea stabilirii celui mai corespunzător portaltoi pentru podgoria Dealurile Bujorului (județul Galați)*. Teză de doctorat, U.A.M.V. Iași.

## MOBILIZATION AND EXPLORATION OF GENETIC RESOURCES IN DEVELOPMENT OF SUSTAINABLE VITICULTURE IN CONTEXT OF RESTRICTIVE FACTORS

### MOBILIZAREA ȘI EXPLORAREA RESURSELOR GENETICE ÎN DEZVOLTAREA UNEI VITICULTURI SUSTENABILE ÎN CONTEXTUL FACTORILOR RESTRICTIVI

SAVIN GH.<sup>1</sup>, CORNEA V.<sup>1</sup>

e-mail: ghsavin@yahoo.com

**Abstract.** *The viticulture of the Republic of Moldova, situated at the northern limit of industrial viticulture, during the millennia supports climate risk factors. Grapevine assortment evolved to varieties with advanced resistance to winter conditions and an increased adaptability. The phenomenon of „Climate Change”, documented in recent decades, already manifested by increased frequency and pronounced intensity of adverse conditions (extreme temperatures, drought, etc.), requires us to use, in this context, the presented genetic resources, as well as researching and introducing of existing in the world genetic sources with a potential for broadening the base of resistance to unfavorable factors, preserving and enhancing quality and productivity, competitiveness of viticulture. Biological material created until now, existing genetic resources were found to be promising, and the assortment need diversification, focusing on increasing adaptability and biological resistance.*

**Key words:** *grapevine, genetic resources, restrictive factors, climate change, adaptability*

**Rezumat.** *Viticultura Republicii Moldova, situată la frontiera de nord a viticulturii industriale, suportă de milenii factorii de risc climatic. Sortimentul viticol a evoluat spre soiuri cu rezistență avansată la condițiile de iernare și cu adaptabilitate sporită. Fenomenul „Modificarea climei”, atestat în ultimele decenii, ce se manifestă deja prin frecvența sporită și intensitatea mai pronunțată a condițiilor nefavorabile (temperaturi extreme, secete prelungite etc.), ne obligă să valorificăm, în acest context, resursele genetice care le avem la dispoziție, documentându-ne totodată și introducând surse genetice care există în lume, cu un potențial de lărgire a bazei de rezistență la factorii nefavorabili, păstrând și amplificând calitatea și productivitatea, competitivitatea viticulturii. Materialul biologic creat până în prezent, resursele genetice deja existente s-au constatat a fi de perspectivă, iar sortimentul necesită diversificare, accentul fiind pus pe sporirea adaptabilității și a rezistenței biologice.*

**Cuvinte cheie:** *vița de vie, resurse genetice, factori restrictivi, modificări climatice, adaptabilitate*

## INTRODUCTION

Climatic particularities of our geographical area create risks for the industrial cultivation of the grapevine, with significant economic impact (Constantinov *et al.*,

---

<sup>1</sup> Research and Practical Institute for Horticulture and Food Technology, Republic of Moldova

2008; Vronskih, 2011): winter thermal regime is diverse and fluctuate significantly, even within 24 hours, the temperature can reach absolute minimum of  $-27...-34^{\circ}\text{C}$ , and the intervention of air masses from the Mediterranean provoke thaws up to 45-60 days; irregular distribution of rainfall causing prolonged drought or flooding. Fluctuations of these parameters vary different: annual averages may differ by up to 3,1 times; the means for one and the same season - up to 18 times, and for one and the same month - up to 500 times. During the past decades are manifested more strongly the effects of the phenomenon of climate change.

These phenomena influence to a greater extent the main ecological factors on which depends the development of viticulture, which is attested worldwide (Jones and Davis, 2000), and confirmed by research in border regions of R. Moldova (Donici *et al.*, 2007; Rotaru and Colibaba, 2011; Bucur and Dejeu, 2014). In those circumstances, in order for reducing the negative economic impact, proved to be effective some agrotechnical processes (Enache and Donici, 2014), planting of some grapevine varieties with economic importance in their untraditional areas (Mustea *et al.*, 2011) and old autochthonous varieties Frâncușa, Fetească albă and Fetească regală showed a high potential of adaptability to climatic stresses under three vines areas in the hills of Moldova (Rotaru and Colibaba, 2011).

We consider the improving of assortment with new varieties with advanced biological resistance, adaptability and increased ecological plasticity, which are based on the potential of the diversity of grapevine genetic resources, one of the main ways for diminishing of consequences of these phenomena. Strategy of accumulation and study, for use in breeding programs, of genetic resources is designed, in part, and through these fluctuations and instability, the objective being to create a new generation of varieties for diverse use with competitive quality and productivity, including the material for an organic vitiviniculture, but with increased resistance (Savin 2012, 2014).

## MATERIAL AND METHOD

General characteristic of weather conditions in Republic of Moldova over the last decade was based on data State Hidrometeorological Service (<http://meteo.md/>). Information on multiannual developments and forecasts of climatic parameter values and nature of impacts of climate change phenomenon were presented on the sources of literature.

Genetic resources have been evaluated over the years in the grapevine gene pool of Scientific and Practical Institute of Horticulture and Food Technologies, located in the south of the mun. Chișinău ( $46^{\circ}58'39.65''\text{N}$  and  $28^{\circ}46'21.68''\text{E}$ ). Description of genotypes was performed according the OIV descriptor list (1983).

## RESULTS AND DISCUSSIONS

Analysis of annual averages values of main meteorological parameters indicates an increasing, over the last decade, of multiannual climate average of air temperatures with  $+0,4...+2,6^{\circ}\text{C}$ , while in 2007 and 2012 was achieved the record of maximum temperature in the history of instrumental observations in R.Moldova ( $+41,5^{\circ}\text{C}$  and  $+42,4^{\circ}\text{C}$ ) (<http://meteo.md/>). It also noted the increased duration (3-15

times higher than the norm) of manifestation of extreme weather (very low or very high temperatures). In 2012, and especially 2015 was maintained throughout the soil and atmospheric drought (attributed to very strong drought), and taking into account the affected area, the phenomenon is classified as catastrophic drought.

The analysis evolution of climatic parameters in Moldova for longer periods of time confirms the general trend in changing their (Constantinov *et al.*, 2008; Vronskih, 2011): during the last 30-40 years an increase in average air temperature estimated at + 1,41 ° C, and within each season there is most pronounced fluctuation during the winter, namely increasing of average winter temperatures during the years 1955-1985 (from -3,32°C to -1,56°C); while the amount of active temperature rose from +3790°C to + 3810°C (by +320°C or +4,92° C per year on average).

Evaluation of various climate change scenarios for R.Moldova, for various periods of time, made under climate models CSIRO-Mk2, HadCM2 and ECHAM4 and aimed to evaluate the increase of the mean annual temperature (Nicolenco, 2000), duration of periods without precipitation (Petreanu and Mironov, 2000) shows the same trend. Overall, for all models, is forecast that annual average temperature increases from 1,4-2,1 ° C over the period 2010-2039 to 3,3-4,6°C by the end of XXI century. According to the used models the duration of periods without precipitation is forecast to grow, compared with the current statistical norm of 10-103 days, with 7-35 days during the next 15 years and by 16-77 days by the end of XXI century.

The consequences of these changes, at global and local level, are multiple and complex, reducing, ultimately, to the socio-economic impact. The results of a study on the impacts of climate change on the economy of Washington State (Impacts of Climate Change on Washington's Economy, 2006) had highlighted different aspects of this phenomenon: the climatic (glacier melting, reducing precipitation as snowfalls, frequent floods and vegetation fires, rising of sea level); direct economic (increased costs and expenses to extinguish the vegetation fire and to conserve water sources, reducing incomes from tourism, redevelopment planning in relation to sea level rise); social (will be affected the quantity and quality of sources of raw material and first of drinking water; harm to human health - premature mortality, health expenditure, epidemics fluctuations, decreased quality of life, etc.); biological and agricultural (higher temperatures affect the physiology of plants, animals and humans; migration of pests and diseases, the impact to the forest resources, etc.). The economic impact seems to be more pronounced not because of the change in mean values, but because of the frequent manifestation of extreme events and the cumulative effect will increase due to the interaction between the various sectors of industry and economy. In the same study was found a mixed impact of climate change on viticulture, a branch of significant socio-economic importance (by revenue, by the number of involved people etc.) for the Washington state: some traditional areas of some varieties will be abandoned due to exceeding temperature tolerance limits, other, colder will be populated.

It is estimated that the effect of climate change will have on viticulture economic impact, caused primarily by the change in the quality, typicality of final



traditional product (Jones et al., 2000) as a result of displacement of optimal climate values for forming the production (an increase in average temperatures by  $2,1^{\circ}\text{C}$  is predicted to advance the deployment of phenological phases 10 to 20 days). On analyzing of a period of 28 years (1979-2006), under vine growing zone Dealurile Bujorului (Donici, 2007), it appears, since 2000, average temperature increasing by  $1,5-2,0^{\circ}\text{C}$  and as a result reducing the difference between the starting date of the phenological phases for varieties Fetească regală, Babească neagră and Merlot, and in the years when water deficit occurs grape production is reduced. The same phenomenon is noted in condition of R.Moldova (Vronskikh, 2011) - reduced production of grapes at the average by 17,4% in years with extreme temperatures in summer. At the same time, being performed a comparative analysis of the average production in Moldova for four crops (winter wheat, maize, sunflower and sugar beet) in the most favorable and most dry years (1961-1990) their difference is not found significant (Pali, 2000). The impact of unfavorable weather conditions was diminished by the implementation, during this period, of new varieties, hybrids and compliance and improve of used phytotechnical processes. In this context the resolution OIV-VITI 517-2015 provides recommendations on adaptation to new climatic conditions of planting material (varieties, clones, rootstocks etc.), of care techniques etc. Therefore, reducing the impact of these destabilizing factors should be based on judicious use in breeding programs of diversity of grapevine genetic resources, taking into consideration the presence of a high potential for adaptability to climatic stresses of old autochthonous varieties. Given the complex nature of the forecasts of climate change is difficult to formulate univocal requirements for the future assortment, however, essentially they are related to the genetic limits of variety, so the heritage of genetic diversity, which would allow anticipated decrease of possible negative phenomena.

Efficient solving of the problem of advanced resistance of grapevine to restrictive factors can be solved by creating new varieties, where is ensured by the genetic constitution of plant. The evolution of this process, initiated in the early century XIX, resulting during the years by development and implementation, including in R.Moldova, of some hybrid, varieties having resistance to environmental stress factors, achieving in recent decades an advanced quality of production, including seedlessness and productivity (Savin, 2012). Thus, this approach has confirmed its effectiveness, possess an significant potential, inclusive in solving the future problems of viticulture. Therefore, the success of creating new varieties that would ensure a sustainable, ecological vitiviniculture, therefore competitive, depends on the diversity of available genetic resources.

Given the stated objectives, over the years has increased the share of created genotypes that possess complex qualities: early maturation, seedless, biological resistance. In Institute's grapevine gene pool were introduced from the Central Asia genotypes with early maturation, large berry, seedless; from viticulture centers of Europe – elite and varieties with increased or advanced resistance, including ennobled hybrids with advanced resistant; from the collections of University Davis, USA –



resistant seedless varieties and *V. vinifera* L. seedless elite with large berry. Thus, in the gene pool has been considerably expanded the diversity of origin of sources of character with strategic importance for the development of viticulture (Savin, 2012) (Table 1).

Table 1

**Diversity of sources of characters with strategic importance for grapevine breeding**

Ecological-Geographic origin	Genotypic origin	Example of genotypes
<i>Advanced or increased resistance to stressful factors</i>		
North America	<i>V. labrusca</i> , <i>V. riparia</i> , <i>V. rupestris</i> , <i>V. lincecumii</i>	Isabela, Campbell, Delaware, Extra
The Far East	<i>V. amurensis</i>	No 1, No 10
Complex interspecific hybrids	Interspecific	Villard blanc, Pierrelli
<i>V. vinifera</i> varieties from Euro-Asiatic area	<i>V. vinifera</i>	Riesling de Rhin, Rkațiteli, Coarnă neagră, Fetească neagră,
New sources, with complex characteristics, from over the world	Interspecific	Prezentabil, Bianca, Cunleany, Cristal, Hibernai, Regent, Moldova, Urojainai, Decabrischii, Apiren negru de Grozești, Apiren roz Basarabean, Apiren roz extratimpuriu
<i>Seedlessness</i>		
Middle Asia, Caucasus, Middle East	<i>V. vinifera</i>	Kiș-miș krasnâi turkmenskii, Kiș-miș mramornâi, Sultanina, Corinth
USA	<i>V. vinifera</i> , <i>V. labrusca</i> , Interspecific	Centennial seedless, Flaim seedless, Romulus, Perlette
New diversified sources from over the world	<i>V. vinifera</i> , Interspecific	Kiș-miș lucistâi, Kiș-miș moldovenesc, Călina, Apiren alb, Apiren roz, Mecita, Kiș-miș ciornâi zimostoikii
<i>Early ripening, productivity</i>		
Old <i>V. vinifera</i> varieties	<i>V. vinifera</i> ,	Madelein Angevine
New diversified sources	<i>V. vinifera</i> , Interspecific	Aromat de Iași, Favorit, Prezentabil, Apiren roz extratimpuriu, Himrood

Old autochthonous varieties have in genotype adaptability to abiotic environment, including drought, winter conditions and some pathogens (Constantinescu *et al.*, 1959-1971) and also have a valuable potential in creation of sustainable viticulture.

## CONCLUSIONS

1. The phenomenon of climate change is manifested more frequently over the past decades by increasing the mean annual temperature, more pronounced intensity of extreme weather events, the same trend is expected for the future, and their actions will have a significant economic impact on viticulture.

2. Presented grapevine genetic resources that hold in different combinations important characteristics: quality, including seedlessness, productivity, early

maturation, resistance to stress factors, presents a biological material with strategic potential available for breeding programs, with important meanings in the context of diminishing of consequences of climate disturbances.

## REFERENCES

1. **Bucur G.M., Dejeu L., 2014** – *Influence of climate variability on growth, yield and quality of grapes in south part of Romania*. Scientific Papers, Series B., Horticulture. Vol. LVIII, p. 133-138.
2. **Constantinescu Gh. et al., 1959-1971** - *Ampelografia Republicii Populare Române*. Editura Academiei RPR, București, Vol. 1-8.
3. **Constantinov T. et al., 2008** - *Evaluarea spațio-temporală a factorilor climatici de risc*. Diminuarea impactului factorilor pedoclimatici extremali asupra plantelor de cultură. Tipografia AȘM, Chisinau, p. 14-40.
4. **Donici A., Enache V., Simion C., 2007** – *Aspects concerning climatic changes influence on vine vegetative phenological phase in "Dealul Bujorului" vineyard*. Lucrări științifice UASMV, Seria Horticultură, vol. 50, p. 427-432.
5. **Enache V., Donici A., 2014** – *Research on minimizing the disruptive effect of climate change on viticulture applying adapted technologies*. Lucrări științifice UASMV, Seria Horticultură, vol. 57, p. 169-174.
6. **Jones G., Davis R., 2000** - *Climate Influences on Grapevine Phenology, Grape Composition and Wine Production and Quality for Boerdeaux, France*. American Journal of Enology and Viticulture. Vol. 51, nr. 3, p. 249-261.
7. **Mustea M. et al., 2011** – *Behaviour of Fetească neagră, Cabernet Sauvignon and Merlot vine varieties in the viticol center Bohotin of Iași vineyard*. Lucrări științifice UASMV, Seria Horticultură, vol.54, nr. 2, p. 389-392.
8. **Nicolenco A., 2000** – *Development of Moldova climate change scenarios for different time horizons*. Climate change. Researches, studies, solutions. Articles collection. Chisinau, p. 20-23.
9. **Palii A., 2000** – *Starea actuală și estimarea impactului schimbărilor climatice asupra agrofitocenozelor principale din Republica Moldova*. Climate change. Researches, studies, solutions. Articles collection. Chisinau, p. 86-89.
10. **Petreanu V., Mironov T., 2000** – *Proiecțiile schimbării duratei perioadelor fără precipitații în Moldova ca reflectare a încălzirii globale la nivel regional*. Climate change. Researches, studies, solutions. Articles collection. Chisinau, p. 24-27.
11. **Rotaru L., Colibaba C., 2011** - *The influence of climatic changes on the behavior of some grape varieties for white wines in moldavian vineyards*. Lucrări științifice UASMV, Seria agronomie. vol. 54, nr. 1, p. 174-179.
12. **Savin Gh., 2012** - *Ameliorarea sortimentului viticol al Republicii Moldova*. Ch. : S.n., (Tipogr. AȘM). 260.
13. **Savin Gh., 2014** - *Fondul genetic al viței de vie – precondiție strategică a vitiviniculturii sustenabile*. Akademos, 1(32), p. 74-82
14. **Vronskih M.D., 2011** – *Izmenenie klimata i riski seliskohozyaistvennogo proizvodstva Moldovy*. Grafema Libris, Kisinev, 560 s.
15. **\*\*\*, 2006** - *Impacts of Climate Change on Washington's Economy - A Preliminary Assessment of Risks and Opportunities*. <https://fortress.wa.gov/ecy/publications/documents/0701010.pdf>
16. **\*\*\*, 2015** – *OIV guidelines for studying climate variability on viticulture in the context of climate change and its evolution*. <http://www.oiv.int/oiv/info/enresolution>
17. **\*\*\*, 1983** – *OIV Descriptor List for Grapevine Varieties and Vitis species*. Office International de la Vigne et du Vin, Paris.

## RESEARCH ON IMPROVEMENT OF SPRAYING MACHINE IN VINES PLANTATIONS, IN ORDER TO REDUCE DEGREE OF POLLUTION SOIL

### CERCETĂRI PRIVIND PERFEȚIONAREA UNEI MAȘINI DE STROPIT ÎN PLANTAȚIILE DE VIȚĂ DE VIE, CU SCOPUL DE A REDUCE GRADUL DE POLUARE AL SOLULUI

**DIACONU Andreea<sup>1</sup>, ȚENU I.<sup>1</sup>, ROȘCA R.<sup>1</sup>, CHIRILĂ C.<sup>1</sup>**  
e-mail: andreea\_diaconu\_a@yahoo.com

**Abstract.** *With all the important advantages in the use of pesticides to combat diseases and pests, their widespread use in high doses and repeated cause many ecological inconveniences, especially on soil, which is an important environmental factor, it is a fundamental support for the existence of life. These effects may be of ecological demo that is those affecting populations and especially their density and nature biocenotic - those causing ruptures biocenotic balances. To limit the effect of pollution treatment plant vine plantations, the USAMV Iasi, equipment was designed and developed to recover some material dispersed plant by spraying machine TARAL 200 PITON TURBO, which was not retained by foliar system of plants. Also spraying machine in intensive orchards and vineyards was equipped with air suction nozzle from LECHLER IDK 120-02, which can reduce drift droplets dispersed by up to 90%.*

**Key words:** *nozzle, pollution, recovery, spraying machine, soil.*

**Rezumat.** *Cu toate avantajele importante pe care le prezintă folosirea pesticidelor în combaterea bolilor și dăunătorilor, utilizarea lor pe scară largă, în doze mari și repetate provoacă numeroase inconveniențe de ordin ecologic, în special asupra solului, care este un factor important al mediului înconjurător, acesta fiind un suport fundamental pentru existența vieții. Aceste efecte pot fi de natură demoecologică, adică cele care afectează populațiile și în special densitatea acestora și de natură biocenotică – cele care provoacă rupturi ale echilibrelor biocenotice. Pentru a limita efectul de poluare a solului la USAMV Iași, s-a proiectat și realizat un echipament pentru a recupera o parte din substanța fitosanitară dispersată de către mașina de stropit TARAL 200 PITON TURBO, care nu a fost reținută de sistemul foliar al plantelor. Totodată, mașina de stropit în vii și livezi intensive a fost echipată cu duze cu absorbție de aer IDK 120-02 de la LECHLER, acestea putând reduce deriva picăturilor dispersate cu până la 90%.*

**Cuvinte cheie:** *duză, mașină de stropi, poluare, recuperare, sol.*

## INTRODUCTION

Applying repeated treatments with pesticides to combat diseases and pests in vineyards and orchards, the number of 7-10 treatments and even more during a

---

<sup>1</sup>University of Agricultural Sciences and Veterinary Medicine Iasi, Romania

production cycle has a negative impact on the environment, particularly soil by its pollution (Arias-Estévez *et al.*, 2008).

Soil is the most important environmental factor because it is a "living organism". Soil is the heart of terrestrial ecosystems as a fundamental support for the existence of life on Earth Solul (Munteanu, 2005; Dumitru *et al.*, 2011). Soil is the main source of food supply for the world population, the main means of production vegetation, the main source of nutrients for plants and renewable energy resource main (Răuță and Cârstea, 1983).

The pollution, the soil changes the physical properties, chemical and biological repercussions of unwanted fertility (Munteanu, 2011). It is estimated that for the formation of a layer of soil 3 cm need 300-1000 years to 20 cm, 2000-7000 years, and 40 cm, 3000-12000 years. Therefore, once destroyed, the ground can be brought into the initial state after a long time (Cârstea, 2003). Pesticides in soil can be transferred without undergoing alterations plants, and fruit, animals and people (Jităreanu *et al.*, 2007; Țenu *et al.*, 2014).

Droplet size is confirmed as the most important factor of effective dispersal of pesticides. Evaluating the coverage has been of interest to plant more than 75 years (Ginsburg, 1928; Panneton. and Lacasse, 2003). Small droplets achieved better coverage of plants, but are subject to drift phenomenon. Chemical drift at the time of treatment is regarded as one of the contributors to environmental pollution and soil (Farooq *et al.*, 2001). It is accepted that more than 10% of the substance is lost through plant derived dispersed.

In the context of the above research and this paper sign, which studied the recovery of the substance dispersed sprayer În contextul cercetarilor TARAL 200 PITON TURBO using equipment designed for this purpose and using air suction nozzle to prevent drift.

## MATERIAL AND METHOD

In order to reduce pollution of soil with pesticides while carrying out treatments of diseases and pests in vineyards, has designed and built a device that has mounted TARAL 200 PITON TURBO. This equipment recovered substance was not retained by vine leaves and vine would come via the ground polluting it.

This equipment consists of racks folded and curved polycarbonate panels fitted with gutters collecting plant substance (Fig.1). Thanks to this support with panels that straddles the one side of the rows of vines as a tunnel, these devices are called tunnel ramps.

The supports are adjustable hinged panels using a hydraulic cylinder in two positions work when they are placed on both sides of the rows of vines, and transportation or return the unit to their heads. The substance is recovered by panels plant and drained into gutters where it is absorbed pump spray machine returned to the tank.

The panels can be mounted on different distances from the axis of the spraying machine and at different heights from the ground with a tie.



**Fig. 1** - Sprayer unit with equipment for recovering sprayed solution: 1 - car splashes; 2 - the metal frame; 3 - graduated cylinders; 4 - hydraulic cylinder; 5 - thrusts; 6 - collapsible support; 7 - parallelogram mechanism; 8 - rods; 9:12 - panels; 10 - pumps; 11 - gutters.

To determine the volume dispensed in a minute by the two spraying ramps, two flow meters were installed. To determine the volume recovered from the dispersed substance has been pumped into the second pot used for measuring by a measuring cylinder.

Experimental tests have been carried out under laboratory conditions, the tank is filled only with water. It was determined the recovery for one minute at different pressure (0.2; 0.4; 0.6; 0.8; 1.0; 1.2 and 1.4 MPa), distance from the axis of the panels spraying machine (1500, 1700, 1900 and 2100 mm) and their height above the ground (300, 500 and 700 mm). Axial fan speed was controlled to 1400 rev/min.

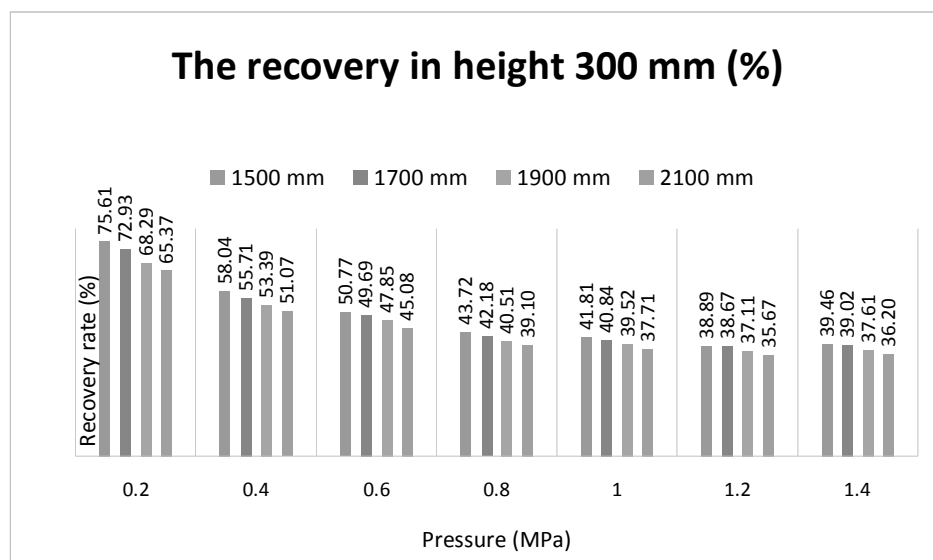
In order to reduce drift dispersed droplets have been mounted on the two ramps spraying the flat spray and air suction nozzle, IDK 120-02 from LECHLER. They carried large drops filled with air bubbles which are less influenced by drifting and performing an effective treatment because it is divided into fine droplets on the leaf.

## RESULTS AND DISCUSSIONS

It appears that with increasing operating pressure the recovery of the substance (water) by two panels decreases the progression of recovery, the best values being obtained at a pressure of 0.2 MPa for all distances and heights for mounting the panels.

For a height of 300 mm from the ground recovering panels there is the greatest degree of recovery. This was achieved at a pressure of 0.2 MPa for panel mounting distance from the axis spraying machine 1500mm, being 75.61%. The recovery decreases with increasing distance from the mounting

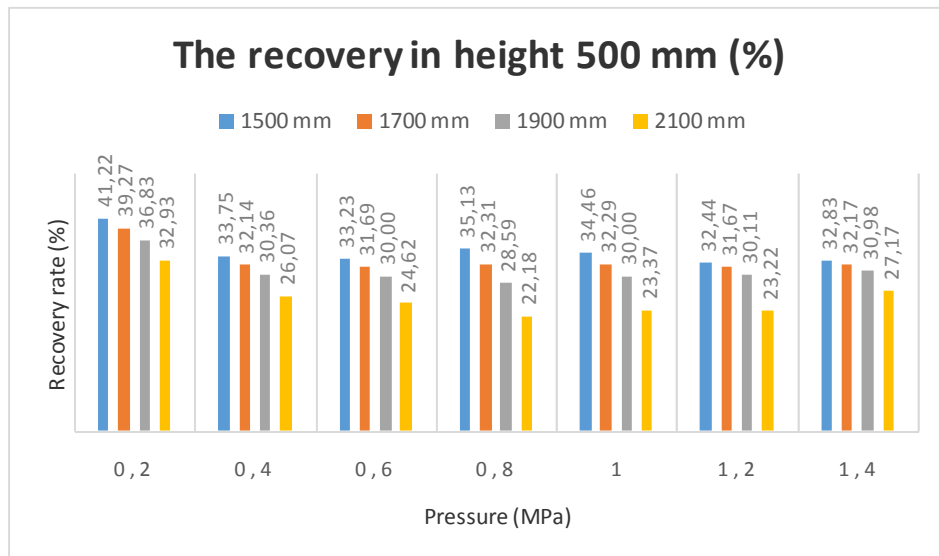
of the panels to 72.93% from 65.37% to 1700 mm 2100 mm. Also for the other operating pressures, the recovery rate decreased with increasing distance. Under pressure from 1.2 to 1.4 MPa notes that the recovery is not reduced and stabilized, the pressure of 1.2 MPa to 38.89% being 1500 mm and 2100 mm 36.20% for and to 1.4 MPa being 39.46% to 36.20% 1500 mm and 2100 mm (Fig. 2).



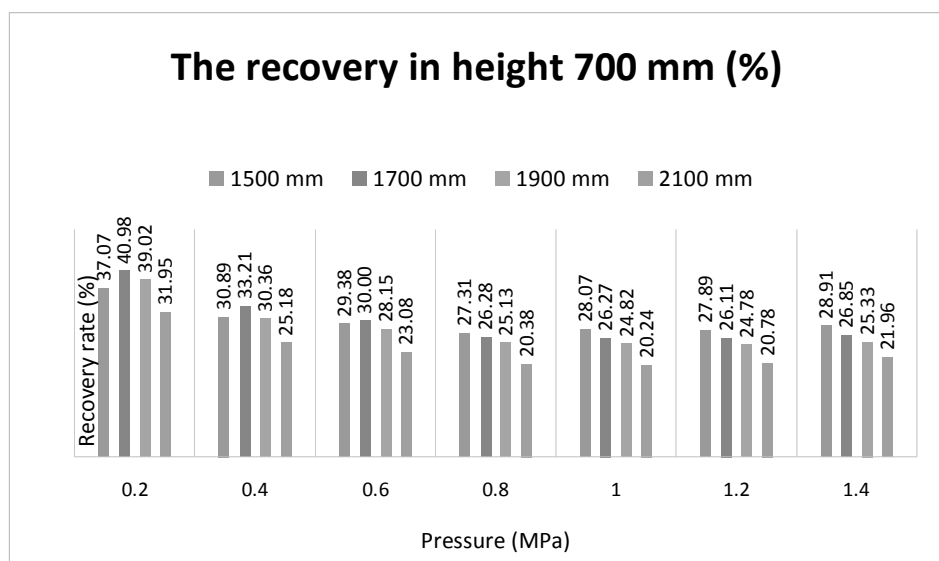
**Fig. 2** - The recovery height of 300 mm from the ground recovering panels for their different distances from the axis of the spraying machine and different working pressures

And height of 500 mm is a decrease in the degree of recovery with increasing work pressure and distance arrangement of panels between 0.4 and 1.4 MPa pressure not great differences, the recovery is more stable. There is a decrease in recovery gradulul to a height of 300 mm, pressure of 0.2 MPa from 75.61% to 41.22% of the distance of 1500 mm from 72.93 to 39,27% for 1700 mm, from 68.29 to 36.83% 1900 mm and from 65.37 to 32.93% 2100 mm (Fig. 3).

By increasing the height of the arrangement of the panels 700 mm is a decrease greater degree of recovery for the same operating parameters. Between 0.6 and 1.4 MPa pressure the recovery is also more stable. 0.2 MPa pressure is observed that the highest recovery rate is achieved at distances 1700 and 1900 mm, and 39.02% from 40.98 subtracting the distance of 1500 mm and 2100 to 37.07% mm to 31.95%. 0.4 and 0.6 MPa pressure is observed at higher recovery rate at a distance of 1700 mm, being 33.21 30% respectively, falling from distance to 1900 and then 1500 to 2100 mm (Fig. 4).



**Fig. 3** - The recovery height of 500 mm from the ground recovering panels for their different distances from the axis of the spraying machine and different working pressures



**Fig. 4** - The recovery height of 700 mm from the ground recovering panels for their different distances from the axis of the spraying machine and different working pressures



## CONCLUSIONS

1. The height of the layout of the panels affect the recovery rate, the more effective is the 300 mm, with a recovery rate of 75.61% for a pressure of 0.2 MPa at a distance of arrangement of the panel of 1500 mm.
2. Distance arrangement of the panels to the axis spraying machine also influences the recovery by increasing its droplet must travel a greater distance, with dwindling chances to get on the boards.
3. The pressure increase is observed that a recovery is obtained larger droplets influenced by derivatives.
4. Due to the air suction nozzle, the recovery rate is stable between 0.6 and 1.4 MPa pressure.
5. Equipment ramps tunnel to prevent soil pollution by recovering recycling plant substance dispersed.

*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.

## REFERENCES

1. Arias-Estévez M., López-Periago E., Martínez-Carballo E., Simal-Gándara J., Mejuto J.C., García-Río L., 2008 – *The mobility and degradation of pesticides in soils and the pollution of groundwater resources. Review.* Agriculture, Ecosystems and Environment, 123: 247-260.
2. Cârstea S., 2003 – *Soil and Food Quality.* Știința solului, vol. XXXVII, 1-2: 3-16.
3. Dumitru M., Dumitru S., Tănase V., Mocanu V., Manea A., Vrinceanu N., Preda M., Eftene M., Ciobanu C., Calciu I., Rîșnoveanu I., 2011 – *Monitoring the health of soils in Romania, National Research Institute for Soil development, agrochemical and environmental protection, ICPA Bucurest.* Publisher, Sitech, Craiova.
4. Farooq M., Balachandar R., Wulfsohn D., Wolf T.M., 2001 – *Agricultural sprays in cross-flow and drift.* Journal Agricultural Enging. Research, 78(4): 347-358.
5. Ginsburg J.M., 1928 – *An apparatus for obtaining measuredbareas of sprayed foliage for chemical analyses.* J. Agr. Res., 36: 1007-1009.
6. Jităreanu G., Țenu I., Cojocariu P., Bria N., Cojocaru I., 2007 – *Technologies and machines for mechanization of ground to practice sustainable agriculture concept,* Publisher Ion Ionescu de la Brad, Iași.
7. Munteanu I., 2005 – *Historical and contemporary challenges aspects.* Soil Science, XXXIX, 1-2: 22-44.
8. Munteanu C., Dumitrescu Mioara, Iliuta A., 2011 – *Ecology and environmental quality,* Publisher Balneară, Bucurest.
9. Panneton B., Lacasse B., 2003 – *Effect of air-assistance configuration on spray recovery and target coverage for a vineyard sprayer.* Can. Biosystems Eng., 46: 13-18.
10. Răuță C., Cârstea S., 1983 – *Preventing and combating soil pollution.* Publisher Ceres, Bucurest.
11. Țenu I., Cojocariu P., Neagu T., Suditu P., 2014 – *Horticultural machines,* Publisher "Ion Ionescu de la Brad", Iasi.

## APPLICATIONS OF THE PRINCIPLE COMPONENT ANALYSIS (PCA) AT GRAPE VARIETIES FROM THE SORTOGROUP COARNĂ NEAGRĂ FOR ESTABLISHING PHENOTYPICAL VARIABILITY

### APLICAREA ANALIZEI ÎN COMPONENTE PRINCIPALI (ACP) LA SOIURILE DIN SORTOGRUPUL COARNĂ NEAGRĂ, ÎN VEDEREA STABILIRII VARIABILITĂȚII FENOTIPICE A ACESTORA

**ISTRATE A.<sup>1</sup>, ROTARU Liliana<sup>1</sup>, COLIBABA Lucia Cintia<sup>1</sup>**

**e-mail:** aistratae@uaiasi.ro

**Abstract:** Multivariate analysis allows the analysis of variables of different individuals, data measured together. Therefore, a complete description of the interdependence relations that exist between groups of individuals and the measured variable is obtained. The multivariate analysis method deals with reducing the number of initial variables by substitution with others resulting from their combination. This can be represented in a graph by points in a two dimensional or three dimensional space, while not losing an excessive amount of information. The PCA technique can be seen from more points of view. For classical statistics, PCA is the determination of main axes of an ellipsoid, indicator of a normal multivariate distribution, these axes being estimated as random samples. PCA is a graphic representation of these, having as optimal character according to some algebraic and geometrical criteria that does not presume the emission of a initial hypothesis of statistical nature on the data that is to be analyzed. PCA allows the extraction of the maximum information, in a simple and coherent form, as a data ensemble, by underlining the interrelations between variables and individuals, either by similarity or opposition.

**Key words:** multivariate analysis, PCA technique, phenotypical variability, Coarnă neagră sortogroup

**Rezumat:** Analiza multivariatională permite analizarea variabilelor măsurate la indivizi diferiți, dar analizate împreună. Astfel se obține o descriere completă a relațiilor de interdependență care există între grupurile de indivizi și variabilele măsurate. Metoda de analiză multivariatională constă în reducerea numărului de variabile inițiale substituindu-se cu altele rezultate din combinația lor, astfel încât acestea pot fi reprezentate ca "roiuri" de puncte într-un spațiu cu două sau trei dimensiuni, fără ca acest lucru să ducă la pierderea excesivă a informației. Tehnica ACP poate fi privită din mai multe puncte de vedere. Pentru statistica clasică ACP constituie determinarea axelor principale ale unui elipsoid, indicator al unei distribuții normale multidimensionale, aceste axe fiind estimate plecând de la un eșantion de probe aleatoriu. ACP constituie o reprezentare a acestora, având un caracter optimal conform anumitor criterii algebrice și geometrice, și care nu presupune emiterea

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

*nici unei ipoteze inițiale de natura statistică asupra datelor de prelucrat. ACP permite extragerea maximei informații, sub formă simplă și coerentă dintr-un ansamblu de date, prin reliefarea interrelațiilor dintre variabile și indivizi, prin asemănarea sau contrarietatea lor.*

**Cuvinte cheie:** analiză multivariațională, tehnica ACP, variabilitate fenotipică, sortogrul Coarnă neagră

## INTRODUCTION

Grapevine genetic variability that is characteristic of plants occurs in response to the amendment of internal and external factors and is manifested both phenotypic and genotypic (Boursiquot and This, 1997).

Variability may be appropriate to the conditions that caused it, adequacy representing response to the action of the moment a factor, or adaptive, adaptation, written in the history of the species that is reflected in the ontology individuals, giving an orientation of individual variability, orientation which is the source of intraspecific diversification (Indreas *et al.*, 2003).

The purpose of the analysis in principal components is to present the information contained in ampelometrics matrices graphical form (circle of correlations, the plane defined by the main components) to be able indicate that two individuals or two variables are more like the more close each other on the charts. To do this, proceed by first calculating the correlation matrix of variables, based on the Pearson correlation coefficient (Rotaru and Țârdea, 1999).

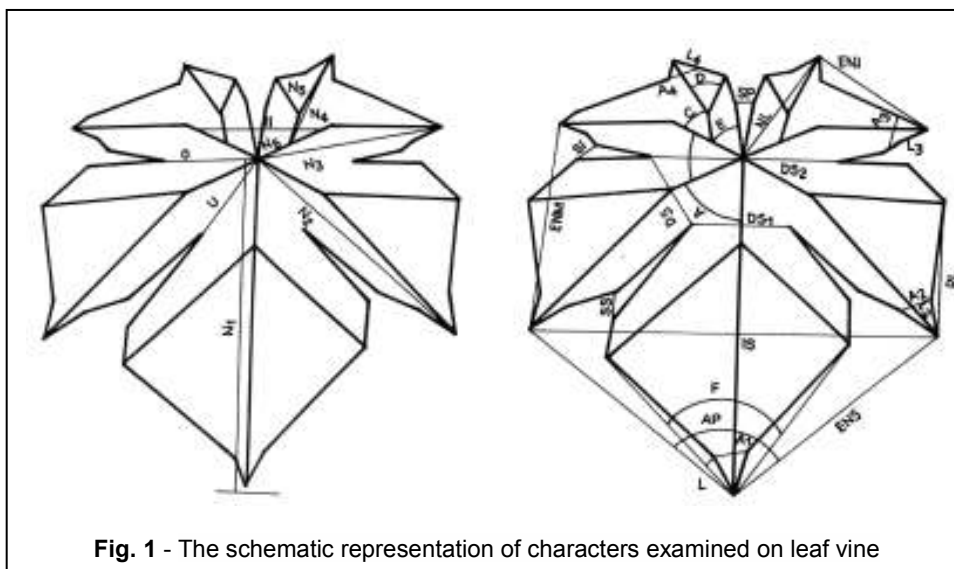
Pearson correlation coefficient corresponds classic linear correlation. Its value ranges from -1 to 1, and it expresses the degree of linear correlation between two variables. The square of the correlation coefficient Pearson, gives an idea of how much variability character is influenced by another character. P values are calculated for each coefficient, permit null hypothesis testing - coefficients are not significantly different from 0 (Rotaru and Țârdea, 2002).

## MATERIAL ȘI METHOD

The principle component analysis was performed on 7 native vine varieties: Coarnă neagră, Coarnă neagră selecționată, Azur, Mara, Ozana, Milcov and Gelu.

For these varieties were performed ampelometric measurements for a total of 30 variables analyzed in adult leaves: the length of the main ribs (N1, N2, N3, N4); the distance between the base of the lateral sinus and the petiole point (U,O); the opening of the lateral sinuses (SS,SI) and the petiole sinus (SP); the length (ALT) and the width (AN) of the limbus; the outer contour of the leaf (ENS, ENM, ENI, NL); the inner contour of the leaf (DS1, DS2, DS); the angle between the main ribs (A, B, C); the angles that define the shape of the median lobe (F, AP); the angle between the median rib and the extremity of the inferior lateral lobe (ABE); the ratio between the length of the ribs (21a, 31a, 41a); the ratio between the base of the lateral sinuses and the rib that support the sinuses (UN2, ON3); the ratio between the length and the width of the limbus (L-A), the ratio of the angles between the main ribs and the depth of the sinuses (AU, BO, ABUO) (Fig. 1).

For mathematical processing of the ampelometric data, it was used the XL-STAT software, with MICROSOFT license.



## RESULTS AND DISCUSSIONS

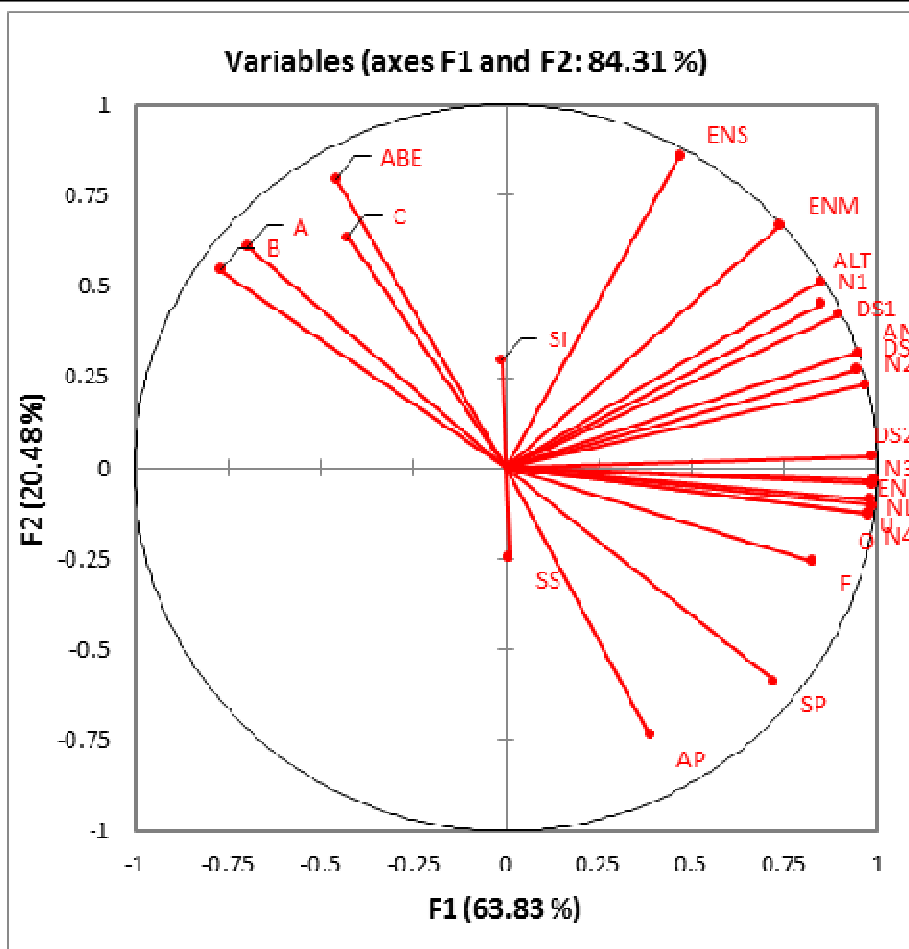
A first stage in the principle component analysis is represented by the calculation of the correlation matrix between variables, based on the Pearson coefficient (simple correlation coefficient) between two variables  $x$  and  $y$ .

It is calculated as the arithmetic average of the normalized deviations product of the two variables and which reflects the intensity of the linear connections between the variables. The values can be between +1 and -1 and the closer the coefficient value is to these limits, the stronger the correlation between the two variables will be.

The next stage represents the determination of the variables and the eigenvectors of them in the created space by the first two main components. (Fig. 2).

The inertia percentage of the first two main components, in the case of the analyzed varieties, was 84,31%, of which 63.83% the first principal component (Axis 1) and 20.48% the second principal component (Axis 2).

Thus it gives away the multidimensional space of the 30 variables analyzed initially, at the bidimensional one, created by the first two principal components, while maintaining an 84,31% of the total inertia (variation) of the individuals.



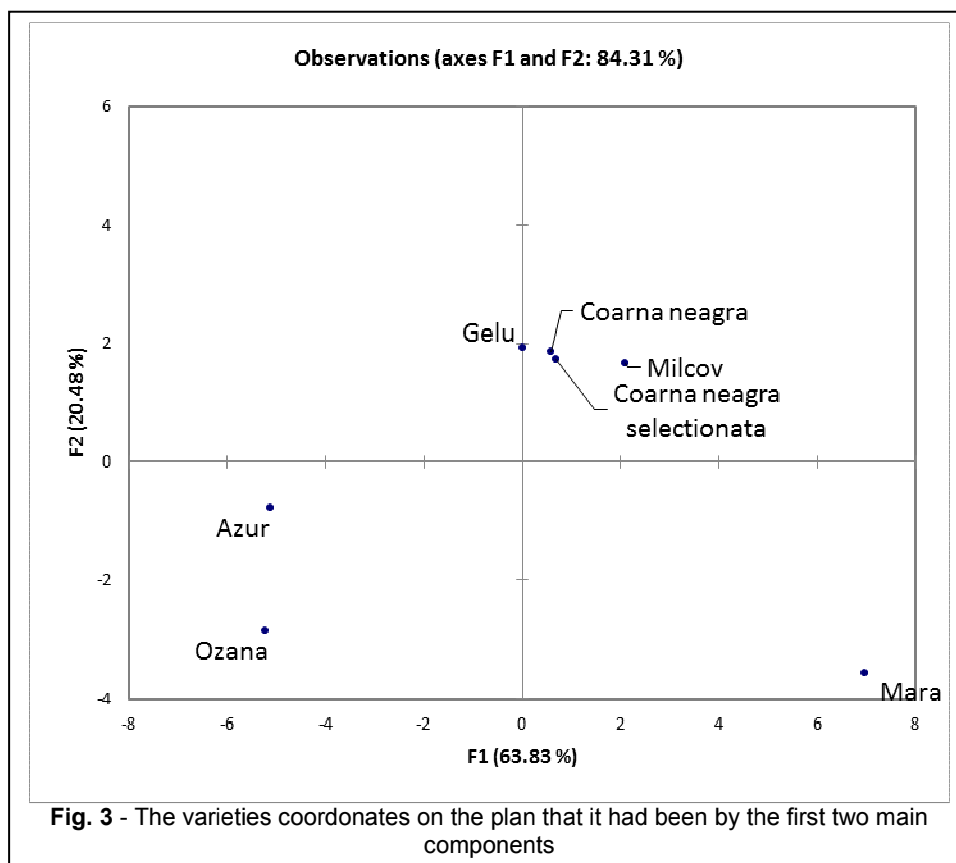
**Fig. 2** –The correlation circle determined by the first two main components

The analysis of the correlation circle reveals that the factor 1 (the main component 1) is determined by the N3 (0,9850), ENI (0,9756), U (0,9743) and DS2 (0,9719) variables. At the opposite side there are the Si and SS variables with 0,0001 each, AP (0,1499) and C (0,1829). All these variables are situated to the extremity of the correlation circle on the axis of the factor 1

The second main component is determined by the ENS (0,7392), ABE (0,6364) and AP (0,5421) variables. The smallest values were found at the N3 (0,0009), DS2 (0,0011), ENI (0,0019) and NL (0,0073) variables.

The 90° angles between the variables shows that they are not correlated with each other, each of them having an important contribution to the architecture of the leaf, unable to be replaced with correlations of these values.

The analysis of the individuals (varieties) coordinates on the main axes are interested in those individuals who had the highest contribution in defining the main components and which, by default, are placed eccentrically on the direction of the main axes (fig. 3). It also states the densest areas that can be considered future groups of individuals, but for their determination is necessary to make a cluster analysis.



**Fig. 3** - The varieties coordinates on the plan that it had been by the first two main components

In defining the main component 1, the biggest contribution had the Mara (6,9550) and Milcov (2,1038) varieties, varieties with medium leaves, trilobite, and in negative way, big contributions were from Ozana (-5,2330) and Azur (-5,1216) varieties, varieties with big leaves, pent lobate.

The main component 2 was determined mainly by the Gelu (1,9330) variety, variety with big leaf, slightly elongate, and in negative way by the Mara (-3,5619) variety.

It should be pointed the position of the Coarnă neagră and Coarnă neagră selecționată varieties which had almost the same contribution in defining the main

components which indicates the high similarity of the leaf architecture of those varieties

## CONCLUSIONS

1. The principle component analysis (PCA) represents a multidimensional statistical and mathematical method which can be applied in ampelography to establish in a first stage the characters that determines the differentiation of a set of varieties with similar phenotypic characteristics.

2. The inertia percentage of the first two main components was 84,31% from which 63,83% the first one and 20,48% the second one, which indicates the fact that bidimensional representation of the plan determined by them highlights the existing linear connections between the 30 analyzed variables at the 7 local vine varieties.

3. The first main component was determined by the N3, ENI, U and DS2 variables with a maximum value of 0,9850 and the main component 2 was given by the ENS, ABE and AP variables with a maximum value of 0,7392.

4. The factor 1 was characterized by Mara and Milcov varieties and the factor 2 was represented mostly by Gelu and Coarnă neagră varieties.

5. From the analysis of the factors 1 and 2 it can be observed a very big phenotypic resemblance of the Coarnă neagră and Coarnă neagră selecționată varieties fact confirmed by the cluster analysis previously established.

## REFERENCES

1. Boursiquot J.M., This P., 1997 – *Les nouvelles techniques utilisées en ampelographie: informatique et marquage*. Journal International des Sciences de la Vigne et du Vin, vol 40(1): 13-23.
2. Indreăș Adriana, Stroe Marilena, Rotaru Liliana, Mărcuță Alina, 2003 - *The establishment of the origin for some autochthonous vine varieties by multi-variable statistical methods*. Lucrări Științifice U.S.A.M.V.București, seria B, vol. XLVI.
3. Rotaru Liliana, Țârdea C., 1999 – *Contribuții la studiul ampelometric al soiurilor de viță de vie, prin prelucrarea datelor pe calculator-programul Microsoft EXCEL-97*. Lucr. Șt. ale UAMV Iași, seria Hortic. vol. 1 (42).
4. Rotaru Liliana, Țârdea C., 2002 – *Contribuții la prelucrarea datelor ampelometrice prin analiza în componenți principali*. Analele Universității din Craiova, vol. VII (XLIII).



## TESTING AND OPTIMIZATION METHODS AND CULTURAL PRACTICES OF VINES IN THE ECOLOGICAL SYSTEM THE PERIOD CONVERSION

### EXPERIMENTAREA ȘI OPTIMIZAREA UNOR METODE ȘI PRACTICI DE CULTURĂ A VIȚEI DE VIE ÎN PERIOADA DE CONVERSIE LA SISTEMUL ECOLOGIC

NECHITA Ancuta<sup>1</sup>, ZALDEA Gabi<sup>1</sup>, DAMIAN Doina<sup>1</sup>, ALEXANDRU L.C.<sup>1</sup>,  
MOROȘANU Ana Maria<sup>2</sup>  
e-mail: ancuta.vasile@yahoo.com

**Abstract.** Technologies currently used in viticulture are based on a brutal human intervention in natural ecosystems causing, exhaustion in short time of natural fertility of the soil, environmental pollution and increased vulnerability of ecosystems vineyards. Both reducing energy consumption and preserving the ecosystems are representing the aim of remodeling and method optimisation during the vine conversion period. In this regard, the wine center Copou Iasi were established specific technology area, differentiated from Feteasca albă and Feteasca regala. The suitability of the viticultural area Copou-Iași as organic culture of the vine is confirmed by the results obtained from observations and measurements of climatic factors, deployment phenophases of vegetation, soil moisture dynamics, weed spectrum of experimental plots, the degree of attack of pests and diseases as well as quantitative and qualitative assessment.

**Key words:** grapes, organic, conventional, climatic factors, quality

**Rezumat.** Tehnologiile de cultură promovate în prezent în viticultură reprezintă, în mare măsură o intervenție brutală a omului în ecosistemele naturale determinând epuizarea, în scurt timp a fertilității naturale a solului, poluarea mediului ambiant și creșterea vulnerabilității ecosistemelor viticole. Remodelarea și optimizarea unor metode și practici de cultură a viței de vie în perioada de conversie vizează atât reducerea consumurilor energetice, cât și păstrarea sau refacerea ecosistemelor. În acest sens, în centrul viticol Copou Iași au fost stabilite tehnologii specifice zonei, diferențiate pe soiurile Fetească albă și Fetească regală. Rezultatele obținute în urma observațiilor și determinărilor privind evoluția factorilor climatici, desfășurarea fenofazelor de vegetație, dinamica umidității solului, spectrul de buruieni din parcelele experimentale, gradul de atac al bolilor și dăunătorilor precum și evaluarea producției cantitative și calitative, confirmă preabilitatea arealului viticol Copou Iași la cultura în sistem ecologic a viței de vie.

**Cuvinte cheie:** struguri, ecologic, conventional, factori climatici

## INTRODUCTION

In organic viticulture the main goal is to capitalize at maximum the capacity of the potential of the ecopedoclimatic area, the local traditions exploitation of the vine in order to increase the quality wine production (Ionescu *et al.*, 1986; Glăman, 2000;

---

<sup>1</sup> Vine and Wine Research and Development Station of Iași, Romania

<sup>2</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

Roraru *et al.*, 2004).

In Europe the main winemaking countries presents a ascendant trend concerning the passage to organic culture system due to market demand and in particular the increasing of the consumption of vine products - organic wine (Stoleru *et al.*, 2008; Toncea, 2002; Jităreanu *et al.* 2003). Due the existence in Romania of small areas planted with organically vines, it was decided to investigate the variability of the main characters productivity for the quality varieties of the vines that are representative for viticultural center Copou Iasi and the pretability of the ecological technologies culture.

## MATERIAL AND METHOD

For the experimentation and optimization of some methods and practices concerning the culture of the vine in the ecological system in the Iasi Copou viticultural center there were realized some experimental models (ecological variant - VE and conventional versions - VC) for Feteasca alba and Feteasca regala varieties. The agrotechnical measures, that are specific to the area and differentiated for the two varieties studied, were correlated with the climatic factors and consisted of pruning of the vine, burning shoot and spring plowing for the cultivated intervals; the control of weeds was done manually by hoeing the row; mechanical mowing strips with lasting grassing; the control of the conventional diseases and pests was done in the ecological way; the operation during the anual vegetative period (weeding, tying the rods; pinching back). There were conducted observations and measurements on the vegetation phenophases, the soil moisture dynamics; the spectrum of weeds and the degree of infestation of the plots; the degree of attack of pests and diseases; the quantitative and qualitative assessment expressed by the average number of grapes on the vine; the average weight of a grape; the production off the hub; the content of the grape must in sugars and acidity.

## RESULTS AND DISCUSSIONS

The analysis of the main climatic elements of the year 2014 was compared to the multianuale averages (1981 - 2010) and showed an increase of the annual average temperature from 9,8°C to 10,3°C and also modified values of the thermal balance. Therefore, the global thermal balance was 3219.0°C compared to 3168.4°C, the active thermal balance 3076,7°C against 3048,9°C and the useful heat balance was 1426.7°C compared to 1386.0°C (Table 1).

The absolute minimum air temperatures recorded in the end of January of 20.6 °C and in the first day of February of 17.3°C affected the normal primary buds in the grapevine. The absolute maximum temperature in air recorded in the august was 34,2°C. The amount of precipitations recorded was very irregular distributed, totaling 377.1 mm during the vegetation period, under the normal 398.1 mm.

Among the synthetic character climate indicators there are included: the hydrothermal coefficient with a value of 1.2, indicating that the moisture was sufficient and correlated with the temperature, ensuring in this way the conditions for large and quality productions; the real heliothermic index with a value of 2.0 indicating an increase in the thermal resources and being optimal for the ripening of the late varieties; the bioclimatic index vineyard (7.0) shows that in the viticultural

center Copou Iasi, the thermal resources have increased thanks the background of optimal water resources; the oenoclimatic aptitude index (IAOe) with a value of 4354,8 indicates a middle favorability for the culture of vine varieties for table and wine grapes; the annual of aridity index Martonne (28,05) indicates a semi-humid steppe climate for the viticultural center Copou Iasi.

Table 1

The values of climatic elements of 2014 compared with multi annual averages.

Climatic elements studied	Multiannual average (1981-2010)	2014
Global thermal balance, $\sum t^{\circ}g, ^{\circ}C$	3168,4	3219,0
Active thermal balance, $\sum t^{\circ}a, ^{\circ}C$	3048,9	3076,7
Useful thermal balance, $\sum t^{\circ}u, ^{\circ}C$	1386,0	1426,7
Mean temperature of July, $^{\circ}C$	21,0	21,5
Mean temperature of august, $^{\circ}C$	20,3	21,6
Mean temperature of september, $^{\circ}C$	15,6	16,9
Annual average temperature, $^{\circ}C$	9,8	10,3
Absolute minimum temperature in air, $^{\circ}C$	-27,2/ 28.12.1996	-20,6/31.01
Absolute maximum temperature in air, $^{\circ}C$	42,3/ 20.07.2007	34,2/14.08
Number of days with maximum temperatures $> 30^{\circ}C$	17,3	22
$\sum$ annual real heatstroke, (hours)	2044,4	1971,1
$\sum$ real heatstroke during vegetation season, hours	1448,2	1405,2
$\sum$ annual rainfall, mm	579,6	618,0
$\sum$ season vegetation rainfall, mm	398,1	377,1
Hydrothermal coefficient, (CH)	1,3	1,2
Real heliothermic index, (IHr)	2,0	2,0
Bioclimatic index vineyard (Ibcv)	7,1	7,0
Oenoclimatic aptitude index (IAOe)	4106,1	4354,8
Annual aridity index Martonne ( $I_{ar-DM}$ )	30,3	28,05

The determinations conducted during the growing season from the soil concerning the humidity on both variantes: those worked on the ranges maintained as a field and those with long-term natural grassing have highlighted the optimal growth and development of the vine stocks. Concerning the month of august it was registered amomentary reduction of water reserves in the soil and an increase of the deficit.

From the observations concerning the succession and the physiological phenophases realized for each variety studied, it can be highlighted the fact that the onset of vegetation occurred by budbursting for the variety Fetească alba on April 20<sup>th</sup>, followed by Fetească regală on 22<sup>th</sup> April, under a heat balance situated between 18 6 and 27,3 $^{\circ}C$ . The flowering, a phenophase which defines the fructification and the influence on the grapes production, started on 5<sup>th</sup> June, the duration andthe end being influenced by the climatic with cumulative action of the genetic and agrotechnical factors. In these circumstances the earliest blooming succed in the case of Feteasca alba (05<sup>th</sup> june), followed closely by the variety Fetească regala (6<sup>th</sup> june). The useful thermal balance that also conditioned the phenophase had values of 255,7 $^{\circ}C$  respectively 257,0 $^{\circ}C$ . The grapes have reached full ripening after 22<sup>th</sup> September for the Feteasca alba variety and ended on 26<sup>th</sup> September with the variety Feteasca regala. During the vegetation period, from budburst to the the fall leaves the two varieties had values of 184 (Feteasca alba) and 182 (Feteasca regala).

The determinations realised on the composition of the vegetation on the intervals between the rows of plots and the proximity areas have highlighted the importance of the work of mechanical cultivators and mowers. On grass strips, the annual species (terofite) had a low frequency, best results being registered by the perennial species in particular hemicriptophytes (resistant to compression) and the meadowland vegetation was characterised by the same phenomenon. High values of the frequency were recorded for the following species: *Agropyron repens*, *Lolium perenne*, *Poa angustifolia*, *Trifolium repens*, *Trifolium pratense* and *Achillea millefolium*. The predominant species in the intervals maintained as a field were those with a very deep root system as: *Amaranthus retroflexus*, *Chenopodium album*, *Solanum nigrum* etc (annual species), *Convolvulus arvensis*.

The specific climatic conditions of the 2014 were favorable for the development of the main pathogen: vine mildew, powdery mildew and downy mildew of grapes. In those circumstances there were observed consequences on both the conventional variant and the ecological one on which were applied seven treatments (Table 2 and 3). Because of the hail that occurred in early July and effected the growth in phenological stage of the grains, there were applied two treatments with Bouille bordelaise. In august, because of low rainfall registered (34,8 mm), the degree of attack of pathogens was reduced and there weren't needed no treatments.

Table 2

**Phytosanitary treatments carried out the year 2014 - variant conventional**

Date	Target organism	The phytoprotection product use	Dose/ha
29.04	<i>Uncinula necator</i> + <i>Eriophies</i> sp.	Mixture sulfocalcic	12 L
18.05	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Antracol + Kumulus	3,0 kg, 3,0 kg
31.05	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Mikal + Topas	3,0 kg, 0,25 L
12.06	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Mikal + Topas	3,0 kg, 0,25 L
22.06	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Folpan + Kumulus	1,5 kg, 3,0 kg
03.07	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Bouille bordelaise + Kumulus	5,0 kg, 3,0 kg
21.07	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Bouille bordelaise + Kumulus	5,0 kg, 3,0 kg

Table 3

**Phytosanitary treatments carried out the year 2014 - variant ecological**

Date	Target organism	The phytoprotection product use	Dose/ha
29.04	<i>Uncinula necator</i> + <i>Eriophies</i> sp.	Mixture sulfocalcic	12 L
18.05	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Bouille bordelaise + Kumulus	3,0 kg, 3,0 kg
31.05	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Funguran + Thiovit	2,0 kg, 3,0 L
12.06	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Kocide + Kumulus	1,5 kg, 3,0 kg
22.06	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Kocide + Kumulus	1,5 kg, 3,0 Kg
03.07	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Bouille bordelaise + Kumulus	5,0 kg, 3,0 kg
21.07	<i>Plasmopara viticola</i> + <i>Uncinula necator</i>	Bouille bordelaise + Kumulus	5,0 kg, 3,0 kg

In assessing the aggressiveness main vine pathogens were made observations and values were calculated on the intensity, frequency and degree of attack for each variety. The effectiveness of treatments varied between 60 and 98% from ecological version and between 66 and 100% conventionally cultivated variant (Table 4 and 5).

Table 4

## The evolution of pathogens in experimental plot cultivated with Fetească albă

Pathogenic agents	Phenologi- cal stage BBCH	Organs analy- zed	Elements determined						Efficacy treatments %	
			Intensity %		Frequency %		Degree of attack %			
			VE	VC	VE	VC	VE	VC	VE	VC
<i>Plasmopara viticola</i>	15	leaf	0	0	0	0	0	0	-	-
	60	leaf	4,65	2,71	5,10	3,0	0,23	0,12	95	97
	65	leaf	4,75	11,0	5,62	3,64	0,26	0,40	97	96
	71	leaf	3,74	6,5	14,99	3,0	0,56	0,20	96	99
		grape	2,60	4,5	6,57	3,0	0,22	0,14	93	96
	81	leaf	8,54	22,0	32,61	5,23	2,83	1,1	71	89
		grape	7,0	14,50	33,27	13,58	2,33	1,97	76	80
	85 - 87	leaf	9,32	32,74	56,01	13,76	5,3	4,50	69	74
grape		8,12	10,5	16,35	10,52	1,42	1,10	75	81	
<i>Uncinula necator</i>	15	leaf	0	0	0	0	0	0	-	-
	60	leaf	0	0	0	0	0	0	-	-
	65	leaf	0	0	0	0	0	0	-	-
	71	leaf	7,50	13,65	55,10	24,43	4,19	3,33	87	90
		grape	1,04	0	1,41	0	0,05	0	98	100
	81	leaf	20,00	25,12	54,86	42,30	10,70	10,62	80	80
		grape	9,55	4,47	24,31	45,75	2,34	2,04	82	84
	87	leaf	22,38	24,92	56,36	50,42	12,61	12,17	82	83
grape		14,02	10,66	56,83	53,16	8,20	5,69	63	74	
<i>Botrytis cinerea</i>	87	grape	0	0	0	0	0	0	-	-

Table 5

## The evolution of pathogens in experimental plot cultivated with Fetească regală

Pathogenic agents	Phenologi- cal stage BBCH	Organs analyzed	Elements determined						Efficacy treatments %	
			Intensity %		Frequency %		Degree of attack %			
			VE	VC	VE	VC	VE	VC	VE	VC
<i>Plasmopara viticola</i>	15	leaf	0	0	0	0	0	0	-	-
	60	leaf	4,75	2,32	5,62	3,0	0,26	0,06	93	98
	65	leaf	9,0	3,0	11,00	25,0	0,99	0,75	92	94
	71	leaf	10,18	3,21	13,50	33,0	1,37	1,06	92	94
		grape	8,90	2,5	5,47	4,4	0,48	0,11	90	98
	81	leaf	7,39	7,25	18,55	17,85	1,37	1,29	88	88
		grape	11,30	6,79	11,50	13,65	1,30	0,92	88	91
	85 - 87	leaf	8,31	10,11	83,0	57,65	6,90	5,82	60	66
grape		4,47	5,06	45,75	35,77	2,17	1,80	75	80	
<i>Uncinula necator</i>	15	leaf	0	0	0	0	0	0	-	-
	60	leaf	0	0	0	0	0	0	-	-
	65	leaf	0	0	0	0	0	0	-	-
	71	leaf	6,41	7,88	63,0	41,42	4,04	3,34	90	92
		grape	4,4	0	2,5	0	0,11	0	97	-
	81	leaf	20,15	24,36	77,60	58,72	15,52	14,30	78	80
		grape	6,86	5,60	33,99	24,84	2,33	1,39	87	92
	87	leaf	15,12	11,81	32,41	34,19	4,90	4,03	94	95
grape		2,9	3,0	31,0	29,0	0,90	0,87	96	96	
<i>Botrytis cinerea</i>	87	grape	0	0	0	0	0	0	-	-

Note: VE - ecological variant; VC - conventional variant

The climatic conditions and plant protection that were mentioned above showed that the harvests varieties studied have realized their biological potential (Table 6) with no significant differences between ecological and conventional variant. The grape production was variable from one variety to another the most productive proved to be Fetească albă with 8.94 to 9.62 t/ha.

Table 6

**Grape production and quality varieties achieved by studied in year 2014**

Parameters determined	Fetească albă		Fetească regală	
	ecological variant	conventional variant	ecological variant	conventional variant
Production, kg/vine	2,36	2,54	2,10	2,18
Production, t/ha	8,94	9,62	7,00	7,26
Weight of a grape, g	174	168	180	172
Weight 100 grains, g	184	182	196	195
Sugars, g/l	206	213	196	199
Total acidity, g/H <sub>2</sub> SO <sub>4</sub>	6,4	6,5	5,7	5,6

## CONCLUSIONS

The ecopedoclimatic conditions for viticultural center Copou of Iasi make it possible the adoption and the gradual implementation of the system of ecological culture of the vine.

The results reaction of the attack of the main pathogens and the main characteristics of productivity and quality, attests the pretability of the vine varieties Fetească albă and Fetească regală for a ecological culture.

*Acknowledgments: The paper was developed in the project within the Sectoral Plan ADER 2020 PS 1.1.14 entitled "Developing a set of methods and techniques for growing vines organically, focusing on main and secondary sale of products resulting from the vineyards, to reduce energy consumption in the semi-subsistence"*

## REFERENCES

1. Ionescu A., Plotoagă G., 1986 – *Ecologie și protecția ecosistemelor*. Editura Bucureștii Noi, Constanța.
2. Jităreanu G., Samuil C., 2003 – *Tehnologii de agricultură organică*, Editura Pim, Iași.
3. Rotaru Liliana, Voiculescu I., 2004 - *Tehnici culturale de creștere a calității în viticultură*, Editura "Prahova" Ploiești.
4. Stoleru V., Grădinariu G., Munteanu N., Jităreanu G., Istrate M., Rotaru Liliana, Vrabie I., Senic I., 2008 - *Ghid de bune practici în producția agricolă ecologică*. Editura "Stef", Iași
5. Toncea I., 2002 - *Ghid practic de agricultură ecologică*. Editura Academic Press, Cluj-Napoca.
6. Glăman Gh., 2000 - *Producția horticola integrată sau biologică*. Hortinform 3: 91.

## ASPECTS REGARDING THE MATURATION DYNAMICS OF TABLE GRAPES GROWN IN VIILE AREA OF BUJORU WINE CENTRE

### ASPECTE PRIVIND DINAMICA MATURĂRII UNOR STRUGURI DE MASĂ CULTIVATE PE PLAIUL VIILE DIN CENTRUL VITICOL BUJORU

**ODĂGERIU G.<sup>1</sup>, ZAMFIR C.I.<sup>1</sup>, LEFTER B.<sup>2</sup>,  
COLIBABA Lucia Cintia<sup>3</sup>, DONICI Alina<sup>4</sup>**  
e-mail:odageriu@yahoo.com

**Abstract:** This article presents data regarding the oenological potential of some table grape varieties cultivated in Viile viticultural area of Bujoru Wine Centre from Dealu Bujorului vineyard by analysing the evolution of the maturation process. The main purpose of this article is to present data regarding the maturation dynamics of six table grape varieties (Victoria, Italia, Chasselas doré, Muscat of Hamburg, Tamina, Alphonse Lavallée), in the new climatic conditions of the last 5-10 years, as a result of global warming, specifically concerning the raise of the sum of active temperatures during the vegetation period, especially in the grape maturation period. The results obtained emphasise the beginning of the optimum time of grape consumption, when the grape harvest should start, the grape full maturity, and the post-maturation period, respectively. The data reflect the influence of climatic conditions of 2014, generally favourable.

**Key words:** table grapes, maturation, total sugars, titratable acidity.

**Rezumat:** Lucrarea de față prezintă date referitoare la potențialul oenologic al unor soiuri de struguri de masă cultivate în plaiul viticol Viile din centrul viticol Bujoru al podgoriei Dealu Bujorului prin urmărirea în principal, a evoluției procesului de maturare a acestora. Scopul principal al lucrării constă în prezentarea unor date referitoare la dinamica maturării a 6 soiuri de struguri de masă (Victoria, Italia, Chasseleas doré, Muscat de Hamburg, Tamina, Alphonse Lavallée), în noile condiții climatice ale ultimilor 5-10 ani, ca urmare a încălzirii globale, respectiv a creșterii sumei temperaturilor utile în perioada de vegetație, în special în cea de maturare a strugurilor. Rezultatele obținute reliefează începutul momentului optim de consum al strugurilor, maturitatea deplină a acestora, respectiv perioada postmaturare. Acestea reflectă în general influențele condițiilor climatice, în general favorabile, ale anului 2014.

**Cuvinte cheie:** struguri, maturare, zaharuri totale, aciditate titrabilă.

---

<sup>1</sup> Research Centre for Oenology- Iași branch of Romanian Academy, Romania

<sup>2</sup> S.C. EUROFRUCT S.R.L., Romania

<sup>3</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>4</sup> Vine and Wine Research and Development Station of Bujoru, Romania



## INTRODUCTION

The rich content of sugars, mineral salts, organic acids and vitamins in grapes and wine, as well as the assimilable form in which they can be found, constitute a valuable characteristic, adding also to people's food intake (Cotea, 1985). Table grapes are harvested at commercial maturity, *i.e.* at the moment when they fulfil the characteristics of composition and quality, which make them good for consumption. The evaluation of the harvesting time is made based on physical-chemical and organoleptic tests. Consequently, grapes are considered to reach the consumption maturity when they have a pleasant, balanced taste, the ratio between sugars and acidity (the glucose-acidic indicator) being higher than 10, and when the grape berries have the colour specific to the variety, with intact skins. In general, the total sugar content is between 130-180 g/L, and the total acidity is 5-8 g/L  $C_4H_6O_6$  (Cotea, 1985; Țârdea and Rotaru, 2003; Rotaru *et al.*, 2011). In order to establish the optimum time of grape consumption, we followed the evolution of the maturation process, from the beginning of ripening to their full maturity, and part of post-maturation period, respectively.

## MATERIAL AND METHOD

For the experiments, we studied six varieties cultivated in Viile viticulturale area, originated from the plantations of S.C. EUROFRUCT S.R.L., of which three white ones (Victoria, Italia, Chasselas doré), and three red ones (Muscat of Hamburg, Tamina, Alphonse Lavallée). The grape samples (1-2 kg) were harvested periodically, at intervals of 5 days, between the 5<sup>th</sup> of August and the 24<sup>th</sup> of October, 2014. After determining the mass of 100 berries, the grapes were crushed, and the juice obtained was tested from a physical and a chemical point of view. The main characteristics of grape composition concerning total content of sugars, titratable and real acidity (pH), tartaric acid, malic acid and conductivity (X) were determined on the day of sample harvesting at S.C. EUROFRUCT S.R.L., and at the Oenology Laboratory of the University of Agricultural Sciences and Veterinary Medicine of Iași. Along with the absolute values obtained, we also present the relative deviations ( $\delta r$ ) in (%), which modified the mass of 100 berries, total sugars, alcohol, titratable acidity, real acidity (pH), tartaric and malic acids of the grape juice samples analysed. The physical and chemical tests were performed using the methods indicated in the national and international standards in force (\*\*\*2012) or in the literature (Țârdea, 2007; Cotea *et al.*, 2009).

## RESULTS AND DISCUSSIONS

The data referring to the grape production of the varieties studied are presented in Table 1. Consequently, on the 24<sup>th</sup> of October, which was the last day of the study, the values were between 3.55 ÷ 5.69 kg/vine, and 11.7 ÷ 20.8 tons per hectare, respectively. The grape health was very good.

Table 1

## Data referring to the grape production of the tested varieties

No.	Variety	Number of vines/ha	Production	
			Kg/vine	tons/ha
1.	Victoria	3300	5.69	18.8
2.	Italia	3300	3.81	12.6
3.	Chasselasdoré	3780	5.49	20.8
4.	Muscat of Hamburg	3780	3.56	13.5
5.	Tamina	3300	4.70	15.5
6.	Alphonse Lavallée	3300	3.55	11.7

The results of the tests performed on the 2014 grape crop are presented in tables 2÷4.

The total sugar content (g/L) increased from values between 56 (Tamina) and 96 (Muscat of Hamburg) on the 5<sup>th</sup> of August, to values between 145 (Alphonse Lavallée) and 216 (Muscat of Hamburg) on the 24<sup>th</sup> of October. At full maturity, its values were the following: 170 on the 29<sup>th</sup> of September for Chasselas doré variety; 210 on the 9<sup>th</sup> of October for Muscat of Hamburg variety; 193 on the 14<sup>th</sup> of October for Italia variety; 144, 154, and 172 on the 19<sup>th</sup> of October for Alphonse Lavallée, Tamina, and Victoria varieties.

Titrateable acidity (g/L  $C_4H_6O_6$ ) decreased from values between 8.51 (Alphonse Lavallée) and 13.81 (Victoria) on the 5<sup>th</sup> of August, to values between 3.42 (Victoria and Alphonse Lavallée), and 4.67 (Tamina) on the 24<sup>th</sup> of October. At full maturity, it had the following values: 3.98 on the 29<sup>th</sup> of September for Chasselas doré variety; 4.48 on the 9<sup>th</sup> of October for Muscat of Hamburg variety; 4.37 on the 14<sup>th</sup> of October for Italia variety; 3.40, 3.41, and 4.65 on the 19<sup>th</sup> of October for Victoria, Alphonse Lavallée, and Tamina varieties.

Real acidity (pH) increased from values between 2.864 (Tamina) and 3.135 (Chasselas doré) on the 5<sup>th</sup> of August, to values between 3.476 (Tamina) and 3.695 (Victoria) on the 24<sup>th</sup> of October.

Tartaric acid (g/L) decreased from values between 5.16 (Alphonse Lavallée) and 8.38 (Victoria) on the 5<sup>th</sup> of August, to values between 2.28 (Victoria) and 4.32 (Italia) on the 24<sup>th</sup> of October.

Malic acid (in g/L) decreased from values between 4.10 (Tamina) and 7.85 (Victoria) on the 5<sup>th</sup> of August, to values between 1.49 (Victoria and Alphonse Lavallée) and 2.90 (Tamina) on the 24<sup>th</sup> of October.

The glucose-acidic or maturation indicator (IM) increased from values between 4.4 (Tamina) and 9.3 (Chasselas doré) on the 5<sup>th</sup> of August, to values between 33.8 (Tamina) and 50.9 (Victoria) on the 24<sup>th</sup> of October. At full maturity this indicator had the following values: 42.7 on the 29<sup>th</sup> of September for Chasselas doré variety; 46.9 on the 9<sup>th</sup> of October for Muscat of Hamburg variety; 44.2 on the 14<sup>th</sup> of October for Italia variety; 33.1, 42.2, and 50.6 on the 19<sup>th</sup> of October for Tamina, Alphonse Lavallée and Victoria varieties.

Table 2

Grape maturation dynamics at *Victoria* and *Italia* varieties from Viile area of Bujoru Wine Centre, from Dealu Bujorului vineyard, in the harvesting year 2014 (age of plantation = 7 years)

No.	Test date	Mass of 100 berries		Total sugars		Titratable acidity		Real Acidity (pH)		Tartaric acid		Malic acid		Gluco- -acidic indic.  (I <sub>M</sub> )	Conduc tivity (X) (mS/ cm)
		g/L	δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)	g/L C <sub>4</sub> H <sub>6</sub> O <sub>6</sub>	δ <sub>r</sub> (%)		δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)		
Victoria															
1.	Aug. 5 <sup>th</sup>	271	0.0	66	0.0	13.81	0.0	2.994	0.00	8.38	0.0	7.85	0.0	4.8	7.5
2.	Aug. 15 <sup>th</sup>	335	23.6	76	15.2	12.32	-10.8	2.963	-1.04	8.38	0.0	7.56	-3.7	6.2	7.1
3.	Aug. 25 <sup>th</sup>	398	46.9	86	30.3	9.65	-30.1	3.099	3.51	6.79	-19.0	6.01	-23.4	8.9	6.7
4.	Sep. 4 <sup>th</sup>	527	94.5	115	74.2	6.98	-49.5	3.244	8.35	4.95	-40.9	4.12	-47.5	16.5	6.3
5.	Sep. 14 <sup>th</sup>	574	111.8	128	93.9	5.10	-63.1	3.394	13.36	3.59	-57.2	2.65	-66.2	25.1	5.9
6.	Sep. 24 <sup>th</sup>	626	131.0	145	119.7	4.25	-69.2	3.441	14.93	2.82	-66.3	1.92	-75.5	34.1	5.5
7.	Oct. 4 <sup>th</sup>	673	148.3	160	142.4	3.63	-73.7	3.522	17.64	2.41	-71.2	1.62	-79.4	44.1	5.1
8.	Oct. 14 <sup>th</sup>	695	156.5	169	156.1	3.42	-75.2	3.647	21.81	2.31	-72.4	1.51	-80.8	49.4	4.6
9.	Oct. 24 <sup>th</sup>	702	159.0	174	163.6	3.42	-75.2	3.695	23.41	2.28	-72.8	1.49	-81.0	50.9	4.2
Italia															
1.	Aug. 5 <sup>th</sup>	274	0.0	62	0.0	13.76	0.0	2.954	0.0	7.00	0.0	7.58	0.0	4.5	7.5
2.	Aug. 15 <sup>th</sup>	329	20.1	80	29.0	12.56	-8.7	2.986	1.1	6.78	-3.1	6.99	-7.8	6.4	6.5
3.	Aug. 25 <sup>th</sup>	374	36.5	112	80.6	9.53	-30.7	3.074	4.1	5.96	-14.9	5.22	-31.1	11.8	5.7
4.	Sep. 4 <sup>th</sup>	458	67.2	138	122.6	6.72	-51.2	3.141	6.3	5.11	-27.0	3.88	-48.8	20.5	5.1
5.	Sep. 14 <sup>th</sup>	484	76.6	158	154.8	5.23	-62.0	3.342	13.1	4.65	-33.6	3.06	-59.6	30.2	4.5
6.	Sep. 24 <sup>th</sup>	562	105.1	175	182.3	5.00	-63.7	3.410	15.4	4.55	-35.0	2.75	-63.7	35.0	4.2
7.	Oct. 4 <sup>th</sup>	602	119.7	185	198.4	4.54	-67.0	3.459	17.1	4.35	-37.9	2.63	-65.3	40.7	3.9
8.	Oct. 14 <sup>th</sup>	620	126.3	193	211.3	4.37	-68.2	3.504	18.6	4.27	-39.0	2.56	-66.2	44.2	3.8
9.	Oct. 24 <sup>th</sup>	615	124.5	197	217.7	4.42	-67.9	3.610	22.2	4.32	-38.3	2.60	-65.7	44.6	3.7

Table 3

**Grape maturation dynamics at *Chasselas doré* and *Muscat de Hamburg* varieties from Viile area of Bujoru Wine Centre, from Dealu Bujorului vineyard, in the harvesting year 2014 (age of plantation = 8years)**

No.	Test date	Mass of 100 berries		Total sugars		Titratableacidity		Real Acidity (pH)		Tartaric acid		Malic acid		Gluco-acidic indic. (I <sub>M</sub> )	Condu conductivity (X) (mS/cm)
		g/L	δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)	g/L C <sub>4</sub> H <sub>6</sub> O <sub>6</sub>	δ <sub>r</sub> (%)		δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)		
Chasselasdoré															
1.	Aug. 5 <sup>th</sup>	188	0.0	88	0.0	9.44	0.0	3.135	0.0	7.31	0.0	7.04	0.0	9.3	6.7
2.	Aug. 15 <sup>th</sup>	195	3.7	98	11.4	8.34	-11.7	3.166	1.0	6.59	-9.8	6.12	-13.1	11.8	6.4
3.	Aug. 25 <sup>th</sup>	206	9.6	112	27.3	7.21	-23.6	3.222	2.8	5.83	-20.2	4.63	-34.2	15.5	6.2
4.	Sep. 4 <sup>th</sup>	220	17.0	131	48.9	5.92	-37.3	3.294	5.1	4.78	-34.6	3.35	-52.4	22.1	5.9
5.	Sep. 14 <sup>th</sup>	228	21.3	151	71.6	4.67	-50.5	3.379	7.8	4.01	-45.1	2.32	-67.0	32.3	5.7
6.	Sep. 24 <sup>th</sup>	232	23.4	164	86.4	4.02	-57.4	3.433	9.5	3.62	-50.5	2.02	-71.3	40.8	5.5
7.	Oct. 4 <sup>th</sup>	232	23.4	176	100.0	3.94	-58.3	3.490	11.3	3.50	-52.1	1.96	-72.2	44.7	5.4
8.	Oct. 14 <sup>th</sup>	228	21.3	182	106.8	3.94	-58.3	3.550	13.2	3.54	-51.6	1.96	-72.2	46.2	5.2
9.	Oct. 24 <sup>th</sup>	222	15.6	186	100.0	3.98	-54.9	3.622	15.1	3.57	-48.2	1.99	-69.1	46.7	5.2
Muscat de Hamburg															
1.	Aug. 5 <sup>th</sup>	256	0.0	96	0.0	11.02	0.0	3.057	0.0	6.01	0.0	5.48	0.0	8.7	8.0
2.	Aug. 15 <sup>th</sup>	267	4.3	113	17.7	10.53	-4.4	3.097	1.3	5.93	-1.3	5.22	-4.7	10.7	8.9
3.	Aug. 25 <sup>th</sup>	304	18.8	132	37.5	9.39	-14.8	3.148	3.0	5.75	-4.3	4.45	-18.8	14.1	8.1
4.	Sep. 4 <sup>th</sup>	335	30.9	160	66.7	7.34	-33.4	3.321	8.6	5.24	-12.8	3.53	-35.6	21.8	7.4
5.	Sep. 14 <sup>th</sup>	340	32.8	186	93.8	6.02	-45.4	3.396	11.1	4.52	-24.8	2.78	-49.3	30.9	6.8
6.	Sep. 24 <sup>th</sup>	346	35.2	202	110.4	4.91	-55.4	3.492	14.2	3.93	-34.6	2.30	-58.0	41.1	6.1
7.	Oct. 4 <sup>th</sup>	363	41.8	208	116.7	4.45	-59.6	3.547	16.0	3.63	-39.6	2.15	-60.8	46.7	5.6
8.	Oct. 14 <sup>th</sup>	365	42.6	212	120.8	4.46	-59.5	3.568	16.7	3.52	-41.4	2.12	-61.3	47.5	5.0
9.	Oct. 24 <sup>th</sup>	362	40.3	216	111.8	4.50	-58.7	3.581	16.5	3.60	-39.5	2.15	-59.9	48.0	4.7

Table 4

Grape maturation dynamics at *Tamina* and *Alphonse Lavallée* varieties from Viile area of Bujoru Wine Centre, from Dealu Bujorului vineyard, in the harvesting year 2014 (age of plantation = 7 years)

No.	Test date	Mass of 100 berries		Total sugars		Titratableacidity		Real Acidity (pH)		Tartaric acid		Malic acid		Gluco- -acidic indic.  (l <sub>M</sub> )	Condu ctivity (X) (mS/ cm)
		g/L	δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)	g/L C <sub>4</sub> H <sub>6</sub> O <sub>6</sub>	δ <sub>r</sub> (%)		δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)	g/L	δ <sub>r</sub> (%)		
Tamina															
1.	Aug. 5 <sup>th</sup>	273	0.0	56	0.0	12.76	0.0	2.864	0.0	6.68	0.0	4.10	0.0	4.4	7.7
2.	Aug. 15 <sup>th</sup>	306	12.1	62	10.7	12.84	0.6	2.891	0.9	6.73	0.7	4.16	1.5	4.8	7.2
3.	Aug. 25 <sup>th</sup>	359	31.5	70	25.0	11.96	-6.3	2.883	0.7	6.18	-7.5	3.94	-3.9	5.9	6.7
4.	Sep. 4 <sup>th</sup>	405	48.4	93	66.1	9.78	-23.4	3.049	6.5	5.25	-21.4	3.61	-12.0	9.5	6.3
5.	Sep. 14 <sup>th</sup>	439	60.8	114	103.6	6.83	-46.5	3.180	11.0	4.21	-37.0	3.22	-21.5	16.7	5.9
6.	Sep. 24 <sup>th</sup>	487	78.4	128	128.6	5.66	-55.6	3.239	13.1	3.83	-42.7	3.13	-23.7	22.6	5.7
7.	Oct. 4 <sup>th</sup>	537	96.7	139	148.2	5.05	-60.4	3.294	15.0	3.53	-47.2	3.00	-26.8	27.5	5.4
8.	Oct. 14 <sup>th</sup>	551	101.8	149	166.1	4.69	-63.2	3.425	19.6	3.42	-48.8	2.90	-29.3	31.8	5.2
9.	Oct. 24 <sup>th</sup>	550	101.5	158	182.1	4.67	-63.4	3.476	21.4	3.45	-48.4	2.90	-29.3	33.8	5.1
Alphonse Lavallée															
1.	Aug. 5 <sup>th</sup>	535	0.0	65	0.0	8.51	0.0	3.044	0.0	5.16	0.0	4.85	0.0	7.6	7.5
2.	Aug. 15 <sup>th</sup>	573	7.1	77	18.5	8.05	-5.4	3.076	1.1	5.01	-2.9	4.61	-4.9	9.6	7.2
3.	Aug. 25 <sup>th</sup>	612	14.4	98	50.8	6.83	-19.7	3.132	2.9	4.61	-10.7	3.79	-21.9	14.3	7.0
4.	Sep. 4 <sup>th</sup>	672	25.6	109	67.7	5.14	-39.6	3.332	9.5	3.85	-25.4	2.72	-43.9	21.2	6.8
5.	Sep. 14 <sup>th</sup>	752	40.6	115	76.9	4.12	-51.6	3.552	16.7	3.24	-37.2	2.09	-56.9	27.9	6.7
6.	Sep. 24 <sup>th</sup>	815	52.3	124	90.8	3.80	-55.3	3.597	18.2	3.04	-41.1	1.70	-64.9	32.6	6.7
7.	Oct. 4 <sup>th</sup>	831	55.3	136	109.2	3.60	-57.7	3.612	18.7	2.90	-43.8	1.58	-67.4	37.8	6.7
8.	Oct. 14 <sup>th</sup>	840	57.0	142	118.5	3.45	-59.5	3.632	19.3	2.83	-45.2	1.50	-69.1	41.2	6.8
9.	Oct. 24 <sup>th</sup>	843	57.8	145	123.1	3.42	-59.8	3.652	20.0	2.82	-45.3	1.49	-69.3	42.4	6.9

Electrical conductivity (X) expressed in mS/cm decreased from values between 6.7 (Chasselas doré) and 8.8 (Muscat of Hamburg) on the 5<sup>th</sup> of August, to values between 3.7 (Italia) and 6.9 (Alphonse Lavallée) on the 24<sup>th</sup> of October.

Based on the data presented in Tables 2-4, Fig. 1 illustrates the graphical evolution of the mass of 100 berries during grape maturation. We can see that the grape varieties studied reached full maturity on the following days: 29<sup>th</sup> of September (Chasselas doré); 9<sup>th</sup> of October, (Muscat of Hamburg); 14<sup>th</sup> of October (Italia); 19<sup>th</sup> of October (Victoria, Tamina; Alphonse Lavallée).

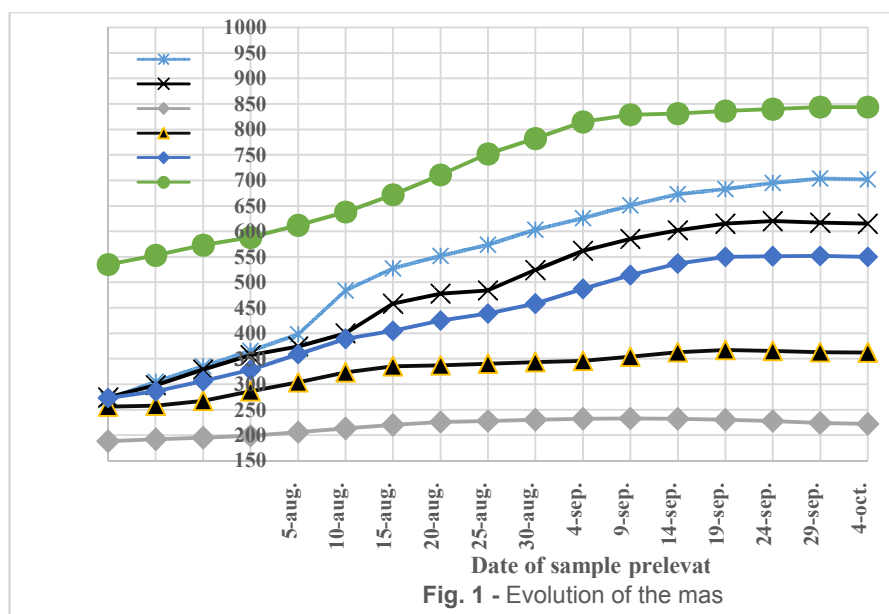


Fig. 1 - Evolution of the mas

The data referring to the beginning of consumption time for fresh grapes from the varieties studied are presented in Table 5. Consequently, the grapes were good to be harvested for consumption as follows: those from Muscat of Hamburg variety on the 25<sup>th</sup> of August, when the total sugar content was 132 g/L, the titratable acidity was 9.39 g/L  $C_4H_6O_6$ , and the value of glucose-acidic indicator was 14.1; those from Chasselas doré and Italia varieties on the 4<sup>th</sup> of September, when the total sugar content was 131 g/L, and 138 g/L, respectively, the titratable acidities were 5.92 and 6.72 g/L  $C_4H_6O_6$ , and the values of glucose-acidic indicator were 22.1 and 20.5; those from Victoria variety on the 19<sup>th</sup> of September, when the total sugar content was 132 g/L, the titratable acidity was 4.57 g/L  $C_4H_6O_6$ , and the value of glucose-acidic indicator was 25.1; those from Alphonse Lavallée and Tamina varieties on the 29<sup>th</sup> of September, when the total sugar content was 130 g/L and 133 g/L, respectively, the titratable acidities were 3.73 and 5.32 g/L  $C_4H_6O_6$ , and the values of glucose-acidic indicator were 34.9 and 25.0.

Most of the data presented above do not agree completely with the literature (Țârdea and Rotaru, 2003, Rotaru *et al.*, 2011), which defines the maturation time (II-VI)

as follows: II (August 1<sup>st</sup>–15<sup>th</sup> for Victoria variety); III (August 16<sup>th</sup>–31<sup>st</sup> for Chasselas doré variety); IV (September 1<sup>st</sup>–15<sup>th</sup> for Muscat of Hamburg and Alphonse Lavallée varieties); V (September 16<sup>th</sup>–31<sup>st</sup> for Tamina variety); VI (October 1<sup>st</sup>–15<sup>th</sup> for Italia variety).

Table 5

## Data referring to the beginning of consumption time at the varieties studied

No	Variety	Test date	Total sugars (g/L)	Titratable acidity (g/L C <sub>4</sub> H <sub>6</sub> O <sub>6</sub> )	Glucose acidific. (I <sub>M</sub> )	Maturation time	
						fore-seen	real
1.	Victoria	Sep. 19 <sup>th</sup>	132	4.57	25.1	II	V
2.	Italia	Sep. 4 <sup>th</sup>	138	6.72	20.5	VI	IV
3.	Chasselasdoré	Sep. 4 <sup>th</sup>	131	5.92	22.1	III	IV
4.	Muscat of Hamburg	Aug. 25 <sup>th</sup>	132	9.39	14.1	IV	III
5.	Tamina	Sep. 29 <sup>th</sup>	133	5.32	25.0	V	V
6.	Alphonse Lavallée	Sep. 29 <sup>th</sup>	130	3.73	34.9	IV	V

This disagreement can be mainly due to greater productions of grapes for the varieties analysed, and also to the climatic conditions of the year we studied.

## CONCLUSIONS

The results obtained, both from the quantitative aspect of grape production, and from the qualitative aspect of their composition characteristics, reflect the influences of the climatic conditions, generally favourable, of the year we studied.

The temperature increase during both vine vegetation period and grape maturation period was favourable to the accumulation of sugars for fresh consumption.

The aspects mentioned above, corroborated with the technological ones, proved insufficient for making most of the varieties studied fit to the maturation time specified in the literature.

This imperiously requires to conduct new studies regarding the evaluation of the maturation time, and to mark the limits of the areas with table grape varieties.

## REFERENCES

1. Cotea V. D., 1985 - *Tratat de Oenologie*, vol. 1. Ceres, Bucharest, 624 p.
2. Cotea V. D., Zănoagă V. C., Cotea V. V., 2009 - *Tratat de oenochimie*, vol. I. Academia Română, Bucharest, ISBN 978-973-27-1756-1, 684 p.
3. Rotaru Liliana, Vasile Ancuța, Nechita C. B., Niculaua M., Colibaba Cintia, 2011 - *Modernizarea tehnologiei de obținere și valorificare a strugurilor de masă prin implementarea Sistemului European de Calitate EUREPGAP*. "Ion Ionescu de la Brad", Iași, ISBN 978-973-147-090-0, 277 p.
4. Țârdea C., Rotaru Liliana, 2003 - *Ampelografie, volumul II*, "Ion Ionescu de la Brad", Iași, ISBN 978-973-147-030-6, 246 p.
5. Țârdea C., 2007 - *Chimia și analiza vinului*. "Ion Ionescu de la Brad", Iași, ISBN 978-973-147-004-7, 1398 p.
6. \*\*\* 2012 - *Recueil des méthodes internationales d'analyse des vins et de moûts*. Office International de la Vigne et du Vin, Officielle, June, Paris.



## CYPERMETHRIN ASSESSMENT AND POSSIBLE PERSISTENCE IN GRAPES OR WINES

### EVALUAREA CIPERMETRINULUI DIN STRUGURI ȘI POSIBILA REMANENȚĂ LA VINURI

NICULAUA M.<sup>1</sup>, ODĂGERIU Gh.<sup>2</sup>, TUCALIUC Roxana<sup>1</sup>,  
MOROȘANU Ana Maria<sup>1</sup>, TELIBAN I.<sup>1</sup>, COTEA V. V.<sup>2</sup>  
e-mail: niculaua@gmail.com

**Abstract.** *The use of insecticides in crops is needed to provide an increase of production at relatively low cost. Lately increased chemisation to crop led to problems in the food, but also directly with animal health or in directly to human consumption. In the present work we propose to study such a substance, namely cypermethrin, which is used widely by human society. The method used is sensitive (SBSE-GC-MS) with a limit of ppb-level for identification and quantification relatively wide interval. We try to eliminate the intermediate steps in order to reduce other influences. Results showed the presence of these substances in grapes, but in relatively low concentrations and lack of it to wine, below the quantification limit of 1 ppb.*

**Key words:** cypermethrin, grapes, wines

**Rezumat.** *Folosirea insecticidelor în culturile de plante este o necesitate care asigură un spor de producție la costuri relativ scăzute. În ultima perioadă chimizarea accentuată a culturilor a dus la apariția de probleme atât cu produsul alimentar, dar și în mod direct cu sănătatea persoanelor sau animalelor consumatori direcți. Prin lucrearea de față ne propunem să studiem o astfel de substanță și anume cipermetrin care este folosit pe scară largă de către societatea umană. Metoda utilizată este sensibilă (SBSE-GC-MS) având o limită de identificare la nivel de ppb și de cantificare relativ largă. S-a încercat eliminarea pașilor intermediari astfel încât să nu mai existe contaminații de altă natură. Rezultate au arătat prezența acestei substanțe la struguri, dar în concentrații relativ mici și lipsa ei la vin sub limita de cantificare de 1 ppb.*

**Cuvinte cheie:** cipermetrin, struguri, vin

## INTRODUCTION

Pyrethroids are a class of synthetically produced insecticides that are mainly used for domestic purposes to control insects such as mosquitoes. They behave very similarly to natural pyrethrins, which are derived from chrysanthemum flowers and are extremely toxic to fish and aquatic organisms, but have low toxicity towards humans. However, repeated exposure increases the risk of anaphylaxis and allergic

---

<sup>1</sup>University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

<sup>2</sup>Romanian Academy – Iasi branch (Research Centre for Oenology), Romania

reaction at very low concentrations and should be monitored.

Pesticides are substances or mixtures of substances intended for preventing, destroying, repelling or mitigating pests. A pesticide may be a chemical substance having antimicrobial or disinfectant properties, a biological agent (such as a virus or bacteria), or a device used against pests. Pests include insects, plant pathogens, weeds, molluscs, birds, mammals, fish, nematodes, and microbes that destroy property, spread disease, act as vectors for diseases or cause a nuisance. The word "pesticide" is an umbrella term for all insecticides, herbicides, fungicides, rodenticides, wood preservatives, garden chemicals and household disinfectants that may be used to kill some pests. Since pesticides varies in identity, physical and chemical properties, it's therefore logical to have them classified and their properties studied under their respective groups. Synthetic pesticides are classified based on various ways depending on the needs (Debbab *et. al.*, 2014).

Cypermethrin ( $C_{22}H_{19}Cl_2NO_3$ ) is a synthetic pyrethroid class of insecticide. It is commonly used to control various pests, including moth pests of cotton, fruit, and vegetable crops. It is also used for crack, crevice, and spot treatment to control insect pests in stores, warehouses, industrial buildings, houses and apartments, greenhouses, laboratories, ships, rail-cars, buses, trucks, and aircrafts. It may also be used in non-food areas in schools, nursing homes, hospitals, restaurants, hotels, and food processing plants (\*\*\*, 1989). Consumers expect a product to be free of pesticides (or low concentrations) and other contaminants (Thomas and Nagel, 2011).

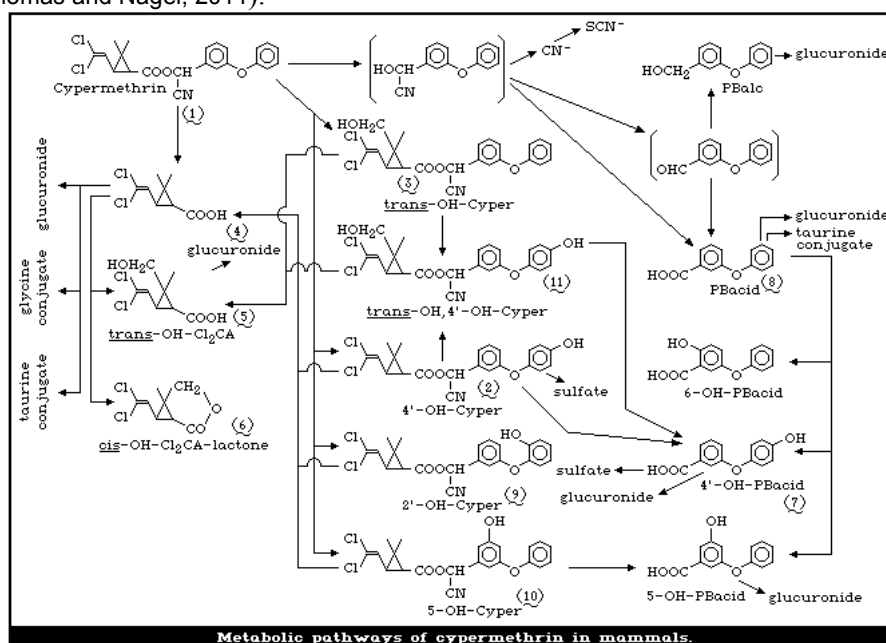


Fig. 1 - Metabolic pathways of cypermethrin in mammals (\*\*\*, internet)

It behaves as a fast-acting neurotoxin in insects. For mammals and this substance is broken in different metabolites (fig. 1) but mainly through 3-phenoxybenzoic acid with a LD50 about 200-300 mg/Kg.

Studies have been carried out on rats on the metabolism of the glucoside conjugate of 3-phenoxybenzoic acid, which occurs occasionally as a metabolite in plants. The results indicated that the rat hydrolyses the glucoside and then metabolizes the 3-phenoxybenzoic acid in virtually the same way as it would metabolize PBA liberated during the metabolism of cypermethrin.

## MATERIAL AND METHOD

Grapes samples and the wines are obtained through vinification by the traditional fermentation method for white wines technology. After gravity settler was done seeding with selected yeasts *Saccharomyces cerevisiae*. After alcoholic fermentation the samples of wine and the frozen grapes (at  $-18^{\circ}\text{C}$ ) were analysed. The samples studied from the following grape sort: Fetească albă; Fetească gală and Frâncușă harvested from Didactic Station "Vasile Adamachi" part of University of Agricultural Sciences and Veterinary Medicine "Ion Ionescu de la Brad" Iași.

For the analysis of the substance a gas chromatographic system is used that has a preconcentration unit SBSE (Stir Bar Sorptive Extraction). The system is an Agilent 7890B GC×GC (fig. 2) coupled to an ion trap mass spectrometer 420MS (ITMS) and also has the possibility to do ballistic GC to an electron capture detector ECD or to MS.

The Column Phenomenex WAX MS 30 m × 0.25 mm × 0.25 mm i.d. from Agilent 7890B GC oven. Injection is done in Split/Splitless inlet at splitless mode with a pulse pressure injection to 40 psi for 0.8 minutes. The injector purge flow of 100 mL/min for 0.75 minutes. The inlet temperature is  $250^{\circ}\text{C}$  and a constant flow mode 1 mL/min is maintained. Carrier gas used is Helium 6.0 purity.

The elution in the oven starts at  $80^{\circ}\text{C}$ ,  $15^{\circ}\text{C}/\text{min}$  to  $250^{\circ}\text{C}$  and then  $5^{\circ}\text{C}/\text{min}$  to  $300^{\circ}\text{C}$  and hold for 9 minutes. The total run time is 30 minutes.

The condition in the Agilent 240 Quadrupole Ion Trap MS are set to Auto-tune with +300 V to multiplier, and 45  $\mu\text{A}$  filament current. The acquisition is done with electron ionization (EI) and for better identification the MS<sup>3</sup> (MS/MS/MS) mode is used. The damping gas is 3.0 mL/min with a solvent delay for 17 minutes from the beginning. The source temperature is  $270^{\circ}\text{C}$ , the trap is set to  $150^{\circ}\text{C}$ , manifold to  $60^{\circ}\text{C}$  and the transfer line to  $300^{\circ}\text{C}$ .

For the ion extraction in the table 1 MS conditions are presented.

Table 1

The MS<sup>3</sup> ionization condition

Precursor	Excitation voltage	Product ion range	Emission current
MS1-181.3	2.2 V	151–153	45
MS2-152.2	3.4 V	74–124	45

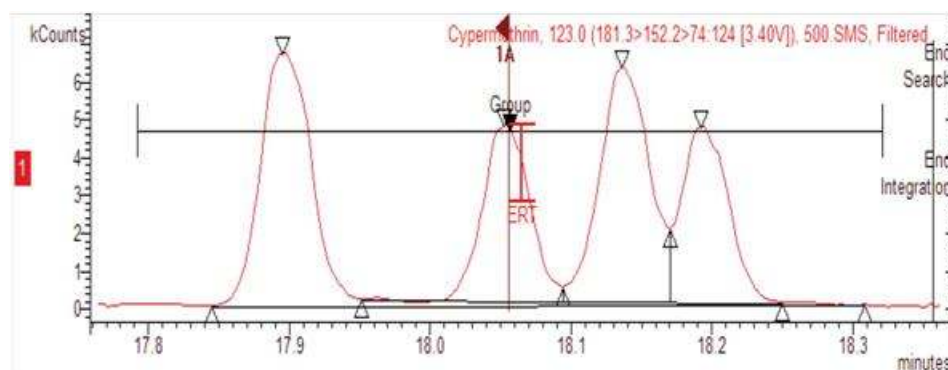
For the analysis the sample is directly placed in contact with the magnetic bar for adsorption of the compound of interest. The calibration of the instrument and the fine tuning was done by adding 5 ppm of 20% ethanoic solution to the magnetic bar and extraction for 60 minutes. The same procedure is done for spiking and recovery of the cypermethrin.



**Fig. 2 - SBSE-GC-ITMS**

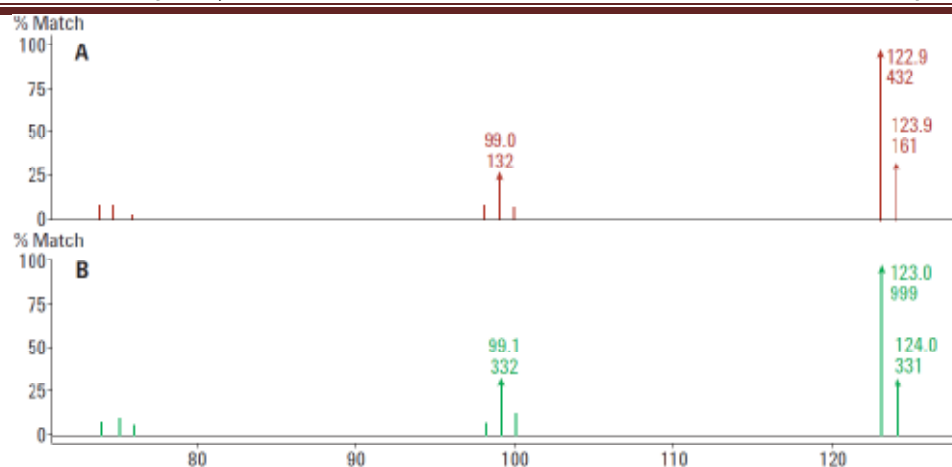
## RESULTS AND DISCUSSIONS

A series of five injection were made using the same methodology of extraction. Different concentration in triplicate were also injected and a range from 0.01-100 ppm. By using different specific transition a 10 ppb limit is detectible. As is presented in fig 3 there is a certain amount of interference close to the elution time of the substance, but the resolution between signals is better than 1.



**Fig. 3 - Cypermethrin specific pick of elution from a sample of must**

Corresponding to the standard injected in green (fig. 4) from the samples in red we have obtained a similarity that varies from 75-98% to the Nist 11 spectres database, but the spectral specific transition (fig. 3) are more specific then the deconvolution software.



**Fig. 4** - Cypermethrin specific pick of elution from a sample of must

The recovery rate is relatively good varying between 72.8 - 101.14 % for the test that we made.

Analysing the grapes observed that there is a quantity of pesticide to Feteascăregală ranged between 0.33 and 0.41 ppm. In the case of Feteascăalbă it was obtained 0.52 to 0.64. Somewhat higher values are in the case of grapes at Frâncușă 0.8 to 0.92. Low concentration determined do not certify that the grape vines were sprayed with this substance before harvest. A different story is the analysis of wine samples were the sample by MS<sup>3</sup> don't show this compound, so the limit that it's possible at 1 ppb it's not present in wine.

*Table 2*

**Results determine of oenological products**

Grape Variety	Grape ppm	Wine ppm
Fetească albă	0.58±0.06	< 0.001
Fetească regală	0.37±0.04	< 0.001
Frâncușă	0.86±0.06	< 0.001

The results are confirming once again that the wine is eliminates much of the reacting compound during the fermentation period.

## CONCLUSIONS

1. The analysis method for cypermethrin is efficient, convenient and can detect low concentrations of this insecticide.
2. This product residual wine is at ppb and not endanger the health of the consumer.
3. Low concentration determined do not certify that the grape vines were sprayed with this substance before harvest.

*Acknowledgments: This research was published under the frame of European Social Fund, Human Resources Development Operational Programme 2007-2013, project no. POSDRU/159/1.5/S/132765.*

#### REFERENCES

1. DebbabM., El HajjajiS., Amal H. Aly, DahchourA., El AzzouziM., ZrinehA., 2014 - *Cypermethrin Residues in Fresh Vegetables: Detection by HPLC and LC-ESIMS and their Effect on Antioxidant Activity*, Mater. Environ. Sci. 5 (S1): 2257-2266.
2. Thomas G. Nagel, 2011 - *Journal of agroalimentary processes and technologies*, 17 (2): 113 – 115.
3. \*\*\*, 1989 - “US Environmental Protection Agency” *Pesticide Fact Sheet Number 199, Cypermethrin*, US PA, Office of Pesticide Programs, Registration Div., Washington, DC, USA., Barlow S.
4. \*\*\*, <http://www.inchem.org/documents/ehc/ehc/ehc82.htm>

## INFLUENCE OF NEW MATERIALS ON THE CHEMICAL COMPOSITION OF MUSCAT OTTONEL WINES

### INFLUENȚA MATERIALELOR NOI ASUPRA COMPOZIȚIEI CHIMICE A VINULUI MUSCAT OTTONEL

**DUMITRIU (GABUR) Georgiana-Diana<sup>1</sup>, LUCHIAN Camelia Elena<sup>1</sup>,  
COTEA V.V.<sup>1</sup>, PEINADO R.A.<sup>2</sup>, LOPEZ DE LERMA Nieves<sup>2</sup>,  
COLIBABA Lucia Cintia<sup>1</sup>, NICULAUA M.<sup>1</sup>**  
e-mail: dumitriu.diana22@yahoo.com

**Abstract.** Nanotechnology is a rapidly evolving field of research with many potentially applications, such as advanced chemistry, medicine, energy production, as well as the food and wine making industry. The aim of this paper is to study the influence of mesoporous materials (MCM-41, SBA-15 and KIT-6) on the white wines physical-chemical parameters, turbidity, phenolic content and color. The used experimental wine was Muscat Ottonel from Bucium vineyard, harvested in 2014. Phenolic compounds were characterized by measuring the absorbance at 280 nm and browning index at 420 nm. The color parameters was analyzed spectrophotometrically with CIE Lch space method. The analyses showed that nanomaterials reduce the nephelometric turbidity units (NTU), which are related with protein content. Also, the results indicate that the total polyphenol index (TPI) of treated wine samples decreased.

**Key words:** nanomaterials, Muscat Ottonel, phenolic compounds, color, turbidity.

**Rezumat.** Nanotehnologia este un domeniu de cercetare cu o evoluție rapidă cu multe aplicații potențiale, cum ar fi în chimia avansată, medicina, producția de energie, precum și în industria alimentară și a vinului. Scopul acestei lucrări este de a studia influența materialelor mezoporoase (MCM-41, SBA-15 și KIT-6) asupra parametrilor fizico-chimici, turbidității, conținutului fenolic și culorii vinurilor albe. Probele de vin utilizate au fost Muscat Ottonel din arealul viticol Bucium, iar anul de vinificație 2014. Compușii fenolici au fost caracterizați prin măsurarea absorbantei la 280 nm și a indicelui de oxidare la 420 nm. Parametrii de culoare au fost analizați spectrofotometric prin metoda CIE Lch. Analizele au arătat că nanomaterialele reduc turbiditatea vinului exprimată prin unități nefelometrice de turbiditate (NTU), care sunt corelate cu conținutul proteic. De asemenea, rezultatele indică faptul că indicele total de polifenoli (TPI) din probele de vin tratate a scăzut.

**Cuvinte cheie:** nanomateriale, Muscat Ottonel, compuși fenolici, culoare, turbiditate.

## INTRODUCTION

During the last two decades, nanostructured materials offer exciting opportunities for a large number of applications (Geszske-Moritz and Moritz, 2013; Ren et

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

<sup>2</sup> University of Cordoba, Spain



*et al.*, 2011). Among these nanomaterials, mesostructured materials have been intensively investigated. Mesoporous materials are defined by IUPAC as the materials with pore sizes between 2 and 50 nm. Mesoporous structures are presented by a wide class of substances including silica, metal oxides, metal hydroxides, metal salts, carbon structures, hybrid materials, organic structures and many others. Mesoporous materials are highly attractive for a wide variety of applications in technical sciences such as catalysis (Perego and Millini, 2013), electronics (Rauda *et al.*, 2013), photocatalytic hydrogen production (Onsuratoom *et al.*, 2011), solar cells (Zhu *et al.*, 2012) and others.

A family of ordered mesoporous silica molecular sieves known as M41S assures a bright future due to their properties: a well-defined pore size between 2-20 nm, an extraordinary high specific surface up to 1000 m<sup>2</sup>/g and distinct adsorption properties due to their pore volume around 0.9 cm<sup>3</sup>/g (Kresge *et al.*, 1992).

SBA-15 is by far the largest pore-size mesoporous material. It has highly ordered hexagonally arranged mesochannels, with thick walls, adjustable pore size from 3 to 30 nm, high hydrothermal and thermal stability. Also, they are ideal for size and shape exclusion separation of polyphenols, proteins and other small biological molecules (Zhao *et al.*, 1998).

In literature, it was reported that the synthesis of other mesoporous materials with larger pores, KIT-6, with Ia3d cubic type structure and a network of interconnected channels. Siliceous material KIT-6, has numerous applications in adsorption and catalysis, thanks to an unique 3-D structures (Xiaoying *et al.*, 2002).

Wine proteins may become insoluble and precipitate which is an important problem in wine stability. This fact is known as protein haze and it generally occurs when the wine is stored at a high temperature or when it is enriched with tannins from cork or wood. Higher protein levels are typical in overripe grapes, grapes sourced from warmer regions and grapes harvest mechanically. Skin contact will typically increase protein concentration. Protein haze is not common in red wines because the proteins flocculate with tannins during alcoholic fermentation. In white wines the protein are eliminated with a bentonite treatment (Sarmientoa *et al.*, 2000).

The aim of this paper is to study the influence of mesoporous materials (MCM-41, SBA-15 and KIT-6) on the white wines physical-chemical parameters, turbidity, phenolic content and color.

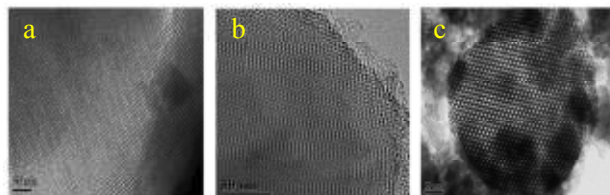
## MATERIAL AND METHOD

Muscat Ottonel grape variety (*V. vinifera*) harvested in 2014 from the Bucium vineyard (Iași, Romania) was crushed and the must obtained was fermented up to 14.2 alcohol degree.

2 g/L of three types of nanomaterials (Fig. 1) were added at three samples of wine. The solutions were stirred 30 minutes in hermetic glass flask. Also, a control was carried out without nanomaterials. The resulting samples were centrifuged at 5000 rpm and 4 °C for 10 minutes.

Turbidity was measured with a nephelometer (HANNA instruments, HI 93703 C), browning index was determined by measuring the absorbance at 420. Wine color was defined by the CIE Lch space in which there are three axes: h\* (hue), L\*

(lightness) and  $c^*$  (chroma or saturation). Total phenolic content was determined by measuring the absorbance at 280 nm.

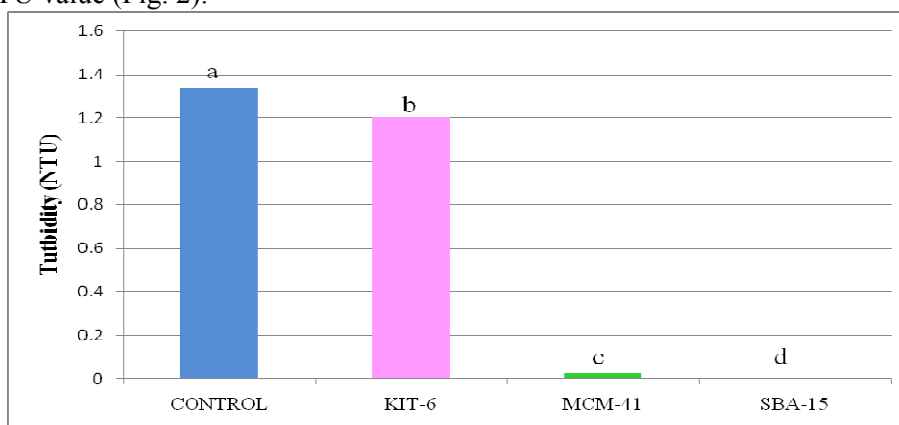


**Fig. 1** - Transmission electron microscopy images of nanomaterials: KIT-6(a), MCM-41(b) and SBA-15(c)

All analyses were made for triplicate and a homogeneous group's analysis was made by means of the statistical package Statgraphics Centurion XVI from StatPoint Technologies, Inc. (Warrenton, VI, USA) to study if there were significant differences among the samples in the determined parameters.

## RESULTS AND DISCUSSIONS

Protein instability in white wines causes clarity problems known as protein casse. The white wine is considered to be stable if the resulting turbidity, or haze, is less than 1.1 nephelometric turbidity units (NTU) (Moreno and Peinado, 2012). Results showed that all nanomaterials increased the limpidity of wines although only the treated wines with SBA-15 and MCM-41 shown an admissible NTU value (Fig. 2).



**Fig. 2** - Turbidity of wines after treatment, expressed as nephelometric turbidity units (NTU). Different letters indicate significant differences at 95% of confidence level

The analytical parameters of control wine were habitual of white wines (Table 2). Enological parameters of treated wines showed slight decreases respect to control wine. This may be due to the interaction of nanomaterials with the components of wine or to the displacement of the chemical balances by the change of pH. Anyway, nanomaterials contribute to reduce both the volatile acidity and the browning index, parameters which are related to analytical quality of wine.

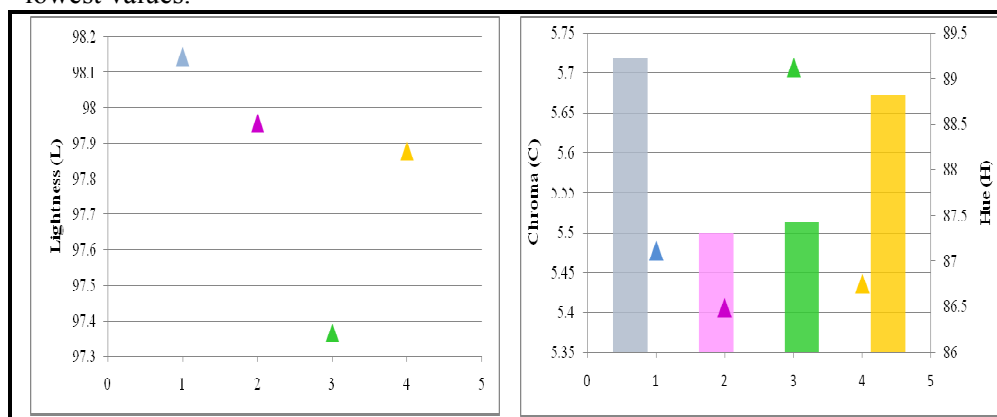
Table 1

**Enological parameters of wines after treatment. TA: titratable acidity (g tartaric acid/L), VA: volatile acidity (g acetic acid/L), BI: browning index, HGA: homogenous groups analysis, different letters indicate significant differences at 95% of confidence level**

	pH	TA	VA	BI
<b>Control</b>	3.38±0.01	4.05±0.02	0.5±0.0	0.091±0.003
<b>KIT-6</b>	3.37±0.00	4.00±0.09	0.4±0.0	0.071±0.005
<b>MCM-41</b>	3.37±0.01	4.05±0.04	0.4±0.0	0.079±0.007
<b>SBA-15</b>	3.37±0.01	4.05±0.01	0.4±0.0	0.074±0.006
<b>HGA</b>	a b b b	a b a a	a b b b	a d b c

Figure 3 shows the color properties  $h^*$  (hue),  $c^*$  (chroma) and  $L^*$  (lightness) for the wine samples. All wines had a yellowish hue –the hue values can range from 0° (red), through 90° (yellow), 180° (green), 270° (blue) and back to 360° or 0°– but the treated KIT-6 and MCM-41 wines showed a more orange-yellow hue that treated wines with SBA-15 and Control.

The property chroma or saturation is related with purity color (bandwidth) and it can range from 0, which is completely unsaturated (very broad set of wavelengths), to 100 for pure color (very limited set of wavelengths). All wine samples showed very little saturated colors, being the treated wines those with the lowest values.

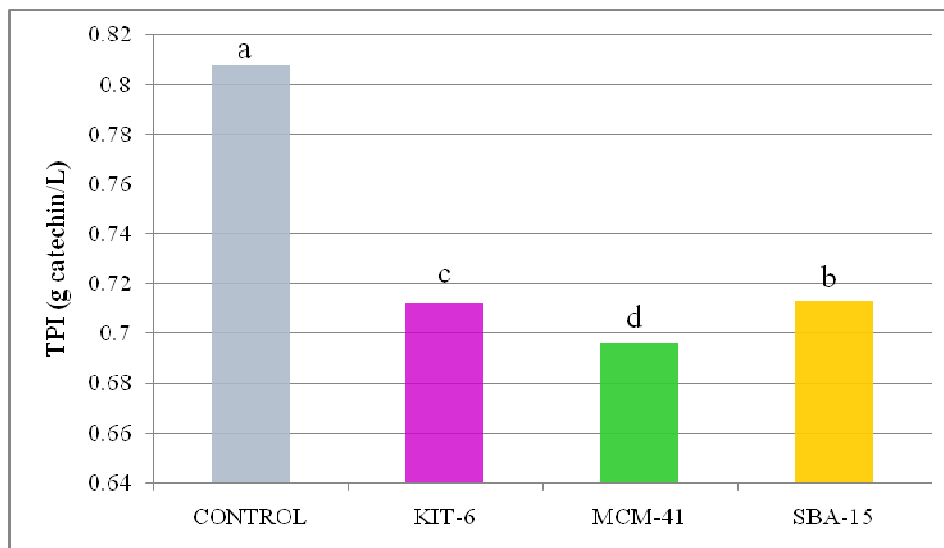


**Fig. 3 - Color properties lightness, chroma (triangle) and hue (full bars) of control wine (blue) and wines treated with KIT-6 (pink), MCM-41 (green) and SBA-15 (yellow)**

Finally, the property lightness shows clarity or darkness color (pigment concentration). It can range from 0, which has no lightness (pigment concentration very high, i.e. absolute black), to 100, which is maximum lightness (pigment concentration very low, i.e. absolute white). All wine samples had colors very lightness, their  $L$  values were above 97. Moreover, the treated wines

showed lower pigment concentration than control wine, which may be due to the interaction of nanomaterials with the pigments of wine.

Relative to total polyphenols index (Fig. 4), the nanomaterials modify significantly the phenolic content respect to control wine. It can be observed that between three types of nanomaterials, MCM-41 retained the highest amount of phenols.



**Fig. 4** - Total polyphenols index (TPI), expressed as grams of catechin per liter (full bars) of wines after treatment. Different small letters indicate significant differences at 95% of confidence level in TPI values.

## CONCLUSIONS

1. Results had showed that nanomaterials reduce the NTU value, which are related with protein concentration, without adversely modifying the parameters measured.
2. Enological parameters of treated wines showed slight decreases respect to control wine. Anyway, nanomaterials contribute to reduce both the volatile acidity and the browning index, parameters which are related to analytical quality of wine.
3. The nanomaterials modify significantly the phenolic content respect to control wine. The results indicate that the total polyphenol index (TPI) of treated wine samples decreased.

**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. We are grateful for the OIV Research Grant and to Vitenol research Group, University of Córdoba.*

## REFERENCES

1. Geszke-Moritz M., Moritz M., 2013 - *Quantum dots as versatile probes in medical sciences: synthesis, modification and properties*. Mater. Sci. Eng. C, 33: 1008–1021.
2. Kresge C. T., Leonowicz M. E., Roth W. J., Vartuli J. C., Beck. J. S., 1992 - *Ordered mesoporous molecular sieves synthesized by a liquid-crystal template mechanism*. Nature, 359: 710–712.
3. Moreno J., Peinado R., 2012 - *Enological Chemistry*. Academic Press. Elsevier.
4. Onsuratoom S., Chavadej S., Sreethawong T., 2011 - *Hydrogen production from water splitting under UV light irradiation over Ag-loaded mesoporous-assembled TiO<sub>2</sub>-ZrO<sub>2</sub> mixed oxide nanocrystal photocatalysts*. Int. J. Hydrogen Energy, 36: 5246–5261.
5. Perego C., Millini R., 2013 - *Porous materials in catalysis: challenges for mesoporous materials*. Chem. Soc. Rev., 42: 3956–3976.
6. Rauda I. E., Saldarriaga-Lopez L. C., Helms B. A., Schelhas L. T., Membreno D., Milliron D. J., Tolbert S. H., 2013 - *Nanoporous semiconductors synthesized through polymer templating of ligand-stripped CdSe nanocrystals*, Adv. Mater., 25: 1315–1322.
7. Ren X., Chen C., Nagatsu M., Wang, X., 2011 - *Carbon nanotubes as adsorbents in environmental pollution management: a review*. Chem. Eng. J., 170: 395–410.
8. Xiaoying, L., Bozhi, T., Chengzhong, Y., Feng, G., Songhai, X., Bo, T., Renchao, C., Lian-Miao, P., Dongyuan, Z., 2002 - *Room-Temperature Synthesis in Acidic Media of Large-Pore Three-Dimensional Bicontinuous Mesoporous Silica with Ia3d Symmetry*. Angew. Chem., 114: 4032–4034.
9. Zhao D. Y., Feng J. L., Huo Q. S., Melosh N., Fredrikson G. H., Chmelka Stucky B. F., 1998 - *Triblock copolymer syntheses of mesoporous silica with periodic 50 to 300 angstrom pores*. Science, 279: 548–552.
10. Zhu P., Reddy M. V., Wu Y., Peng S., Yang S., Nair A. S., Loh K .P., Chowdari B. V. R., Ramakrishna S., 2012 - *Mesoporous SnO<sub>2</sub> agglomerates with hierarchical structures as an efficient dual-functional material for dye-sensitized solar cells*. Chem. Commun., 48: 10865–10867.

## ASSESSMENT AND ADAPTATION OF METHODS OF EXTRACTION OF GRAPE SEED POLYPHENOL COMPOUNDS

### EVALUAREA ȘI ADAPTAREA UNOR METODE DE EXTRACȚIE A COMPUȘILOR POLIFENOLICI DIN SEMINȚELE DE STRUGURI

NECHITA C-tin. B.<sup>1</sup>, NICULAU M.<sup>1</sup>, COTEA V.V.<sup>1</sup>,  
e-mail: bnechita@gmail.com

**Abstract.** The extraction of polyphenolic compounds in the plant material is influenced by the chemical and structural characteristics and the particle size of the plant material, as well as the presence of interfering substances. The solubility of the phenolic compounds depends on the solvent used, the degree of polymerization of the compounds, as well as interactions with other plant compounds. The paper presents several methods used for extracting polyphenolic compounds from grape seed: Soxhlet continuous extraction method; extraction with supercritical fluids (liquid CO<sub>2</sub>); extraction under reduced pressure (3 bar) and high pressure (15 bar) with water and 75% alcohol concentration. Characterization of polyphenolic extracts carried out by HPLC allowed the recommendation of two methods: with supercritical fluid and Soxhlet method. In the same time it was also observed a higher extraction yields for the phenolic acids, hydrolysable tannins, flavones of stilbene using alcohol as a solvent.

**Key words:** seed, extraction, Soxhlet, supercritical fluids, pressure

**Rezumat.** Extracția compușilor polifenolici din materialele vegetale este influențată din caracteristicile chimice structurale, metoda de extracție utilizată, dimensiunea particulelor materialului vegetal, precum și de prezența substanțelor interferente. Solubilitatea compușilor polifenolici depinde de solventul utilizat, gradul de polimerizare al compușilor, precum și de interacțiunile cu alți compuși vegetali. În lucrare sunt prezentate rezultatele mai multor metode de extracție a compușilor polifenolici din semințele de struguri selectate din tescovină, respectiv: metoda extractivă continuă Soxhlet; extracția cu fluide supercritice (CO<sub>2</sub> lichid); extracția la presiune scăzută (3 bari) și ridicată (15 bari) cu apă și alcool etilic de concentrație 75%. Caracterizarea extractelor polifenolice realizată prin analiza HPLC a permis recomandarea metodelor de extracție cu fluide supercritice și metoda Soxhlet. De asemenea, s-a constatat o extracție superioară a acizilor fenolici, a taninurilor nehidrolizabile, a stilbenilor și a flavonelor în cazul extractelor la care s-a utilizat ca solvent alcoolul etilic.

**Cuvinte cheie:** semințe, extracție, Soxhlet, fluide supercritice, presiune

## INTRODUCTION

Obtaining polyphenolic extracts raises several technological problems of which the most important one is to ensure maximum extraction efficiency, this

---

<sup>1</sup>Enology Research Center of the Romanian Academy, Iași Branch, Romania

being particularly dependent on the factors involved in physical and chemical extraction processes. The extraction procedures used, namely the extraction solid – liquid in continuous and discontinuous system provides an advanced recovery of the crumbling degree, of the nature of the extraction solvent and a refresh of it, as well as adequate contact time or extraction (Maria Teresa Escribano – Bailon *et al.*, 2003).

In the case of grape seeds, the vegetable extracts obtained by Soxhlet continuous extraction method are richer in polyphenols, anthocyanins and tannin materials, but the extraction time is higher, varying according to the plant material used (Jackson *et al.*, 1996; Luque de Castro *et al.*, 1998; Lin *et al.*, 1999; Lapornik *et al.*, 2005; Yilmaz *et al.*, 2005; Downey *et al.*, 2010; Schieber *et al.*, 2001).

Supercritical fluid extraction method is a technique for non-destructive separation, belonging to the processes occurring at high pressure (SPE) based on the power of leaching fluids at appropriate temperatures and pressures above the critical point. The solvent that is most commonly used is the carbon dioxide, due to its properties: low toxicity, non-polluting and the final extract does not contain other substances, allowing at the same time to achieve a selective extraction (Lin *et al.*, 1995; Palma *et al.*, 1999).

The extraction of the polyphenolic compounds at high / low pressure is a technique recently introduced among the methods used for the extraction of polyphenolic compounds. This method offers the possibility to conduct the extraction under an inert atmosphere and the stability of the polyphenolic compounds during this process is extremely high (Palma *et al.*, 2001).

At the moment an optimal procedure to ensure the total or fractional extraction of polyphenolic compounds isn't available. Considering this aspect, in the present paper in order to obtain the polyphenolic extracts from the grape seeds selected from pomace, there were tested: the continuous Soxhlet method, the extraction with supercritical fluids method and the extraction at high / low pressure method.

## MATERIAL AND METHOD

In order to remove the substances with lipophilic character it has been used a static method which consisted in keeping in contact for one hour the plant material with ethyl ether. Soxhlet extraction was carried out continuously with 96% ethyl alcohol, in relation to 1/10 (plant material (g) / solvent (ml), at a constant temperature of 78°C for 48 hours.

In the case of supercritical fluid method (liquid CO<sub>2</sub>) there were performed five extractions with seven g of plant material each, using liquid CO<sub>2</sub> at a rate of 1 mL / minute and ethyl alcohol at a rate of 0.5 mL/min. The extraction time was 32 minutes for a total volume of 48 ml extraction fluid.

The extractions under reduced pressure (3 bar) and high pressure (15 bar) were carried out using as solvents water and ethyl alcohol with a concentration of 75% and an amount of 7.5 g vegetable material. The extraction time was 2 minutes and in the final stage it was collected a volume of 215 mL of extract.

Based on the nature of the solvent used, the concentration of the extracts was done differently. So, for the samples extracted with the ethyl alcohol, the concentration was carried out at a pressure of 120 mbar, at a temperature of 40 ° C



and a rotational speed of 32 rpm. Instead for the samples extracted with water, the conditions of concentration were: a pressure of 40 mbar, a temperature of 40°C and a rotational speed of 80 rpm. In order to characterize the polyphenolic extracts, there were determined the total polyphenols using the method of Folin-Ciocalteu, the index of tanoide materials (IMT) through the method established by Bourzex.

In addition, through HPLC analysis there were identified and quantified a series of phenolic acids, stilbenes, tannins that are not hydrolysable and several flavones using the method presented by M. Castellar (Castellar *et al.*, 2002).

## RESULTS AND DISCUSSIONS

The data obtained during the characterization of the raw polyphenol extracts are presented in figures 1 and 2. Analysing the results, the vegetal extract from grape seeds obtained by supercritical fluid extraction, with a concentration in polyphenols 3.02 g GAE/L was noted (Figure 1).

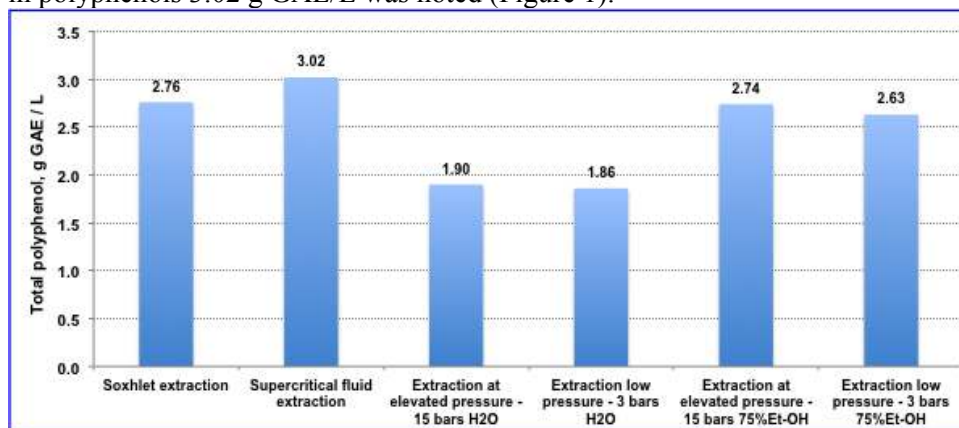


Fig. 1 - The total polyphenol content of the raw extracts obtained from the seeds of grapes

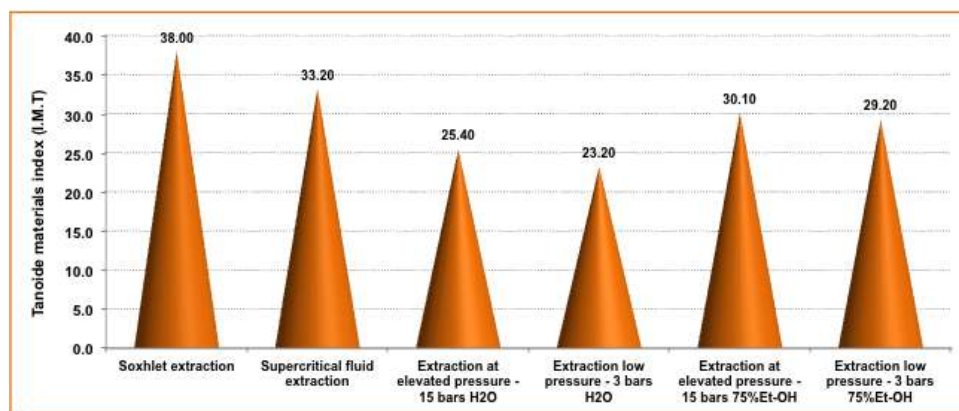


Fig. 2 – The tanoide materials index (IMT) of the raw polyphenolic extracts obtained by various methods of extraction

In what concerns the index of tanoide materials, the highest values were

determined at the extracts obtained using the Soxhlet method (38.0) and the supercritical fluid method (33.2). Lower values for both tanoide materials index (25.4 and 23.2) and for total polyphenol concentration (1.86 to 1.90 g GAE / L) are observed in the case of the extracts on which the water was used as a solvent (Figure 2).

Through the HPLC analysis of the concentrated extracts several phenolic acids were identified such as hydroxybenzoic acids and hydroxycinnamic acids (tables 1 and 2). Analyzing the data presented in Table 1 is particularly significant the extract obtained by supercritical fluid method, by showing the highest content of gallic acid, protocatehic acid, syringic acid, vanilic acid and m-hydroxybenzoic acid.

Table 1

**The hydroxybenzoic acids identified in the polyphenolic extracts concentrated**

Hydroxybenzoic acids, mg	Soxhlet extraction	Supercritical fluid extraction	Extraction at elevated pressure		Extraction low pressure	
			H <sub>2</sub> O	75%Et-OH	H <sub>2</sub> O	75%Et-OH
gallic acid	10,08	317,42	40,12	54,73	20,16	67,07
protocatehic acid	-	366,87	35,81	41,78	11,00	51,56
p-hydroxybenzoic acid	0,95	2,79	1,40	1,18	0,96	1,79
gentisic acid	-	0,31	0,91	2,27	0,14	0,68
syringic acid	114,05	126,51	1,53	13,92	6,31	0,30
vanillic acid	0,45	34,77	5,51	3,83	1,07	4,61
m-hydroxybenzoic acid	0,40	29,86	5,32	2,50	0,06	0,25
salicilic acid	92,86	2,26	0,53	6,17	-	4,83

The most significant hydroxybenzoic acid, namely the galic acid showed values between 10.08 mg / L of the extract obtained by Soxhlet extraction method and 317.42 mg / L of the extract obtained by method of supercritical fluids.

Although in the literature is remarked that salicylic acid is formed only during the alcoholic fermentation process, it was identified in the largest quantity in the extract obtained by Soxhlet method (96.64 mg/L) and in amounts more small, between 0.53 and 6.17 mg / L in the extracts obtained by other methods.

It has also been identified syringic acid, with very large variation limits between 0.30 mg/L (the extract obtained at low pressure with 75% alcohol) and 126.51 mg/L (the extract obtained by extraction with supercritical fluids ). In lower amounts other hydroxybenzoic acids were identified, such as: protocatechinic acid, p-hydroxybenzoic acid, gentisic acid, m-hydroxybenzoic acid and vanillic acid.

In the concentrated polyphenol extracts there were determined a number of hydroxycinnamic acids respectively, caffeic acid, , chlorogenic acid, p-coumaric, ferulic and synapic acids (table 2). It can be observed, that in the case of extracts obtained at low pressure using water the amount of the hydroxycinnamic acids was below the detection limit.

Table 2

**The hydroxycinnamic acids identified in the concentrated polyphenol extracts**

Hydroxycinnamic acids, mg/L	Soxhlet extraction	Supercritical fluid extraction	Extraction at elevated pressure		Extraction low pressure	
			H <sub>2</sub> O	75%Et-OH	H <sub>2</sub> O	75%Et-OH
cafeic acid	0,53	1,27	1,54	2,32	-	0,15
clorogenic acid	2.29	15,06	2,62	1,90	-	2,78
p-cumaric acid	4,58	0,25	0,31	0,68	-	0,71
ferulic acid	0,98	0,38	0,33	1,94	-	3,44
sinapic acid	0,36	0,20	-	-	-	0,21

Through HPLC analysis of the polyphenols extracts it was revealed the presence of some non- hydrolysable tannins (condensed), namely, the catechin and epicatechin (table 3). In what concerns the content of catechin, it can be observed a high variation limit, larger amounts being extracted with the method that used ethyl alcohol as a solvent. In the case of epicatechin, significantly higher amounts were found in the extracts obtained by extraction with supercritical fluids (157.91 mg/L).

Table 3

**The non-hydrolysable tannins identified in the concentrated polyphenolic extracts**

Method of extraction	catechin, mg/L	epicatechin, mg/L
Soxhlet extraction	6,05	4,58
Supercritical fluid extraction	4,18	157,91
Extraction at elevated pressure - 15 bars H <sub>2</sub> O	2,33	0,14
Extraction low pressure - 3 bars H <sub>2</sub> O	0,97	-
Extraction at elevated pressure - 15 bars 75%Et-OH	3,76	1,64
Extraction low pressure - 3 bars 75%Et-OH	0,40	3,97

In addition to phenolic acids, in the extract samples it was also identified the trans-resveratrol (table 4). As for the phenolic acids, the higher amount was found in the extracts obtained by extraction with the Soxhlet method and the supercritical fluid method, respectively, 2.40 and 1.38 mg/L.

Table 4

**The content in stilbene and flavones from the concentrated polyphenol extracts**

Method of extraction	trans-resveratrol, mg/L	rutine mg/L	quercetine, mg/L
Soxhlet extraction	2,40	0,55	1,54
Supercritical fluid extraction	1,38	0,75	0,37
Extraction at elevated pressure - 15 bars H <sub>2</sub> O	-	0,72	-
Extraction low pressure - 3 bars H <sub>2</sub> O	-	-	-
Extraction at elevated pressure - 15 bars 75%Et-OH	0,02	0,29	0,11
Extraction low pressure - 3 bars 75%Et-OH	0,21	20,60	1,15

Among the flavones were identified routine and quercetin, standing out especially the extract obtained by Soxhlet extraction method with 1.54 mg/L

quercetin and the extract obtained at low pressure using water 20.6 mg/L rutine.

## CONCLUSIONS

1. The characterization of polyphenolic extracts carried out by HPLC (high performance liquid chromatography) allows the recommendation of the supercritical fluid extracting method for and the Soxhlet method.
2. In the case of the extracts that used ethyl alcohol as a solvent, it could be remarked a superior extraction of the phenolic acids, of non- hydrolyzed tannins, of the stilbene and the flavonols.

*Acknowledgments: This work was carried out by the Partnership in priority areas - PN II, developed with the support of MEN -UEFISCDI, project no. 183/2014 (PN-II-PT-PCCA-2013-4-0333) "Technology of capitalization of the bioactive elements from the grape seed waste with usefulness in the food and pharmaceutical industry, plant and environmental protection (Acronym: PROVITIS)".*

## REFERENCES

1. **Castellari M., Sartini E., Fabiani A., Arfelli G., Amati A., 2002** - Analysis of wine phenolics by high-performance liquid chromatography using a monolithic type column. *Journal of Chromatography A*, 973: 221–227
2. **Downey M.O., Hanlin R.L., 2010** - Comparison of Ethanol and Acetone Mixtures for Extraction of Condensed Tannin from Grape Skin. *South African Journal of Enology and Viticulture*, 31: 154-159.
3. **Escribano - Bailon Maria Teresa, Celestino Santos-Buelga, 2003** – *In Methods in Polyphenol Analysis*. The Royal Society of Chemistry Cambridge.
4. **Jackson F.S., McNabb W.C., Barry T.N., Food Y.L., Peters, J.S., 1996** - The condensed tannin content of a range of subtropical and temperate forages and the reactivity of condensed tannin with ribulose-1,5-bis-phosphate carboxylase (Rubisco) protein. *Journal of the Science of Food and Agriculture* 72,: 483-492.
5. **Lapornik B., Prosek M., Wondra A.G., 2005** - Comparison of Extracts Prepared from Plant By-Products Using Different Solvents and Extraction Time. *Journal of Food Engineering*, 71: 214-222.
6. **Luque de Castro M.D., Garcia-Ayuso L.E., 1998** - Soxhlet extraction of solid materials: An outdated technique with a promising innovative future. *Analytica Chimica Acta*. 369: 1-10
7. **Palma M., Piñeiro Z., Barroso C., 2001** - Stability of phenolic compounds during extraction with superheated solvents. *Journal of Chromatography A* 921(2): 169-174.
8. **Palma M., Taylet L.T., 1999** - Extraction of polyphenolic compounds from grape seeds with near critical carbon dioxide. *Journal of Chromatography A* 849(1): 117-124.
9. **Schieber A, Keller P, Carle R., 2001** - Determination of phenolic acids and flavonoids of apple and pear by high-performance liquid chromatography. *J Chromatogr* 910(2): 265–273.
10. **Yilmaz Y., Toledo R.T., 2005** - Oxygen Radical Absorbance Capacities of Grape/Wine Industry Byproducts and Effect of Solvent Type on Extraction of Grape Seed Polyphenols. *Journal of Food Composition and Analysis*, 19: 41-48.

## COMPARATIVE STUDY OF SOME WHITE ROMANIAN AND CYPRIOT WINES

### STUDII COMPARATIVE ALE UNOR VINURI ALBE CIPRIOTE SI ROMÂNEȘTI

**COLIBABA Lucia Cintia**<sup>1</sup>, **COTEA V.V.**<sup>1</sup>, **KOKKINOFTA Rebecca**<sup>2</sup>,  
**LUCHIAN Camelia Elena**<sup>1</sup>, **CODREANU Maria**<sup>1</sup>  
e-mail: cintia\_colibaba@yahoo.co.uk

**Abstract:** *The Republic of Cyprus, a country well known to the Romanian consumers for its special sweet wines Commandaria, produces also still wines that are much appreciated. The specific climatic conditions of Cyprus, subtropical climate - Mediterranean and semi-arid type with very mild winters (on the coast) and warm to hot summers, have a colossal influence on the obtained wines. Snow is possible only in the Troodos Mountains in the central part of the island. Rain occurs mainly in winter, with summer being generally dry. The aim of this study is to analyze from a physical-chemical point of view some typical Cypriot wines and, in parallel, some Romanian wines. The wines are either commercial or authentic samples (produced in the laboratory by applying classical technologies).*

**Keywords:** Cypriot wines, Romanian wines, structural analysis

**Rezumat:** *Republica Cipru, o țară cunoscută consumatorilor de vinuri datorită vinului special Commandaria, produce de asemenea și vinuri liniștite extrem de apreciate. Condițiile climatice specifice țării și anume climatul subtropical, mediteranean și semi-arid, cu ierni foarte blande și veri foarte călduroase, joacă un rol foarte important în dezvoltarea structurii vinurilor. Precipitațiile apar rar, mai ales iarna, zăpada se găsește doar în munții Troodos. Scopul acestui studiu este analiza comparativă a unor vinuri albe din Cipru și din România, pentru a determina structura acestora. Probele analizate sunt atât comerciale dar și experimentale (obținute prin metode cunoscute, în laborator).*

**Cuvinte cheie:** vinuri cipriote, vinuri românești, analize structurale

## INTRODUCTION

Climate is the most important factor in the success of all farming systems, with a major influence in determining the crop's quality for a given region as well as a measure of quality control of plant products and management of economic sustainability (Cotea and Sauciuc, 1985; Jones, 2008).

Cyprus has a subtropical climate – Mediterranean and semi-arid type (in the north-eastern part of the island) with very mild winters (on the coast) and

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> State General Laboratory, Nicosia, Cyprus

warm to hot summers. Snow is possible only in the Troodos Mountains in the central part of island. Rain occurs mainly in winter, with summer being generally dry. Cyprus has one of the warmest climates in the Mediterranean part of the European Union – situated in C III b EU area. The average annual temperature on the coast is around 24 °C during the day and 14 °C at night. Generally, summers last about eight months, beginning in April with average temperatures of 21-23 °C during the day and 11-13 °C at night, and ending in November with average temperatures of 22-23 °C during the day and 12-14 °C at night, although in the remaining four months temperatures sometimes exceed 20 °C. Sunshine hours on the coast are around 3400 per year, from an average of 5-6 hours of sunshine per day in December to an average of 12-13 hours in July.

Up until 2004 all Cyprus wines were basically considered as table wines. Accession to the European Union, however, has necessitated new legislation that would classify the wines produced on the island. Four viticultural regions have been identified which are designated to produce wines of controlled appellation of origin:

**1. Akamas Laona** (north-west coast of Cyprus) - both red and white wines. In the case of red wines at least 85% of the blend should be derived from one of the two local varieties of Maratheftiko and Ofthalmo and for white wines 85% of the blend should come from the local variety Xynisteri.

**2. Vouni Panayias – Ambelitis** (western part of the island at an altitude of over 800 metres ) both white and red wines may be produced. White wines must use Xynisteri as the basic constituent (at least 85%). Red wines may be produced in two ways: the basic constituent must be either one of the two indigenous varieties of Maratheftiko or Ofthalmo to a level of at least 85%, or the local Mavro variety (at least 60%) supplemented by over 30% of one of specified foreign varieties (Cabernet sauvignon, Cabernet franc, Syrah, Merlot etc.).

**3. Pitsilia** (32 villages all situated on the slopes of Madari, Papoutsia and Maheras mountains) - production of white wines, Xynisteri must constitute at least 85% of the blend and for red wines either Maratheftiko or Ofthalmo must constitute at least 85% of the blend, or the blend may be made up of 60% Mavro and 30% of one of the specified foreign varieties.

**4. The wine villages of Limassol** (southern slopes of the Troodos mountains - 20 villages aka wine villages). Prominent in the wine villages region are two subregions - Afames and Laona. The production of both white and red wine in the wine villages region is based on the same grape varieties as in the Pitsilia region.

Terroir is a highly important concept in viticulture because it relates the sensory attributes of wine to the environmental conditions in which the grapes are grown (Van Leeuwen *et al.*, 2004).



## MATERIAL AND METHOD

The Cypriot wine varieties taken into study were represented by :

1. Xynisteri -the prevailing white grape variety of the Cypriot vineyard. It produces excellent light-coloured white wines, with low alcohol levels and acidity, especially when it is cultivated in such regions as Laona of Akamas, Ambelitis, Vouni Panayia, Pitsillia. Its grapes do not have an intense aroma. Xynisteri wines are not suitable for extended ageing. This is the only other variety apart from Mavro that is used for making Commandaria.
2. Spourtiko - The name Spourtiko means "bursting", a reference to its berries whose fragile skin splits easily. It is a variety with a short vegetative cycle with medium-sized, loosely-packed cluster with big berries of golden-yellow colour. Spourtiko is also the variety that can give a solution to the problematic pollination of Maratheftiko. Aromatically is a neutral variety, so the floral aromas of its wine seems to derive exclusively from the alcoholic fermentation.
3. Promara – White, very rare grape variety, with big and compact cluster and big berries. Resists very well to the drought and the calcareous soils that characterize the Cypriot vineyard. It has great resistance to the cryptogamic diseases and to the enemies of the vine because of its thick skin. Disposes aromatic precursors that can be expressed in the wine, by using the appropriate winemaking techniques giving wines characterized by aromas of watermelon and exotic aromas. Its wines present well-balanced acidity and the capacity to mature in oak barrels.

The Romanian wine varieties taken into study were represented by Grasă de Cotnari, Sauvignon blanc, Pinot gri, Riesling italian, Aligoté, Tămăioasă românească, Fetească regală, Traminer roz, Francusă, Neuburger, Muscat Ottonel, all harvested from the Ampelographic Collection of USAMV Iasi.

The analytical methods used to determine these parameters were in accordance with the European standards and those imposed by the OIV (OIV, 2013).

## RESULTS AND DISCUSSIONS

The results are presented in table 1 (Cypriot wine samples) and table 2 (Romanian wine samples). The Cypriot areas where the wines have been produced are also mentioned.

The white grape varieties of Cyprus produce wines with an alcoholic concentration that ranges from 5.3%vol. to 15.01% vol. The climatic conditions that influence a high accumulation of sugars stand at the base of this, as well as legislative and technological processes allow wines with a lower concentration of alcohol. The Romanian samples range between 10.08% vol. and 14.1%vol.

The acidity of the analysed samples is between 3.83 g/L tartaric acid (Cypriot Spourtiko variety – sample no. 11) and 9.14 g/L for Aligoté (sample no. 5) in Romania. The intense metabolism of acids in grapes is also a results of extreme climatic conditions, especially heat. The pH is equilibrated in all wines, as well as the volatile acidity, showing no signs of future microbiological problems.

The remanent sugars show that the majority of analysed samples are dry, a small percentage surpassing the Romanian legislative benchmark of 4 g/L.



Table 1

## Physical-chemical analysis of Cypriot wine samples

Cyprus wine samples	Free SO <sub>2</sub> (mg/L)	Total SO <sub>2</sub> (mg/L)	Vol. acidity (g/L)	Total acidity (g/L)	Density	Alcohol (vol.%)	Remanent sugars (g/L)	Total dry extract	Non-reductive extract	pH
Promara - Lemesos 2012	13.01	100.22	0.19	5.31	0.9962	10.23	2.62	25.3	22.68	3.6
Chardonnay – Pafos 2012	8.45	83.1	0.31	6.29	0.997	9.96	4.68	27.1	22.42	3.5
Morokanella – Lemesos 2012	5.71	47.71	0.17	4.72	0.9954	9.83	4.48	22.4	17.92	3.6
Spourtiko – Lemesos 2012	10.5	52.05	0.18	4.13	0.9958	8.06	2.47	17.7	15.23	3.68
Xynisteri – Pafos 2012	7.53	93.6	0.3	4.03	0.994	9.73	1.82	18.5	16.68	3.8
Sauvignon blanc – Lemesos 2013	9.82	107.3	0.44	6.09	0.9921	13.92	6.68	26.3	19.62	3.25
Promara – Lemesos 2013	11.19	95.88	0.18	5.01	0.9918	10.37	1.6	15	13.4	3.4
Promara – Lemesos 2013	7.99	82.18	0.19	5.26	0.9913	12.23	5.63	19	13.37	3.06
Morokanella – Lemesos 2013	15.52	139.7	0.29	4.86	0.9933	8.07	0.91	11.3	10.39	3.34
Chardonnay- Pafos 2013	5.94	117.5	0.22	5.21	0.9938	12.01	1.28	25	23.72	3.6
Spourtiko – Lemesos 2013	6.39	124.4	0.19	3.83	0.9973	5.3	0.9	12.6	11.7	3.4
Xynistery – Lemesos 2013	7.08	148.1	0.19	3.88	0.995	7.5	0.84	13.9	13.06	3.5
Xynisteri – Coumandaria 2013	7.31	39.49	0.63	4.81	0.9878	15.01	2.04	18.3	16.26	3.8
Xynisteri – Coumandaria 2013	8.9	79.22	0.37	4.91	0.9905	12.25	1.37	17.2	15.83	2.9

Table 2

## Physical-chemical analysis of Romanian wine samples

Romanian wine samples – Ampelographic Collection USAMV Iasi	Free SO <sub>2</sub> (mg/L)	Total SO <sub>2</sub> (mg/L)	Vol. acidity (g/L)	Total acidity (g/L)	Density	Alcohol (vol.%)	Remanent sugars (g/L)	Total dry extract	Non-reductive extract	pH
Grasa de Cotnari 2014	62.09	147.25	0.25	8.55	0.9905	11.6	1.7	19.8	18.1	3.02
Sauvignon blanc 2014	160.94	259.79	0.29	8.94	0.9895	11.24	1.1	16.2	15.1	3.08
Pinot gri 2014	65.52	138.57	0.33	6.68	0.9879	14.49	4.81	21.6	16.79	3.24
Riesling italian 2014	103.41	159.35	0.29	7.07	0.9886	11.83	0.77	15.7	14.93	2.91
Aligoté 2014	105.24	145.73	0.33	9.14	0.9909	10.08	0.72	16.2	15.48	3.03
Tamaioasa romaneasca 2014	75.11	138.57	0.31	6.93	0.9943	11.63	15.47	29.4	13.93	3.12
Feteasca regala 2014	40.63	103.64	0.43	6.92	0.9911	13.94	6.63	23.7	17.07	3.21
Traminer roz 2014	33.79	82.87	0.25	6.73	0.9871	14.1	1.67	18.5	16.83	3.23
Francusa 2014	86.74	132.4	0.41	8.54	0.9886	11.87	0.63	15.7	15.07	2.98
Neuburger 2014	61.63	138.11	0.45	7.71	0.9922	12.44	10.63	26.8	16.17	2.9
Muscat Ottonel 2014	43.6	109.34	0.33	6.43	0.9895	12.2	1.34	19	17.66	3.21

The total dry extract and non-reductive extract have values specific for white wines, exception being the wine samples where a short maceration or *batonnage* was applied (Chardonnay).

The free sulphur dioxide varies a lot between the Romanian and the Cypriot samples, in the latter being in very small quantities (5.71 mg/L to 13.01 mg/L) while in the Romanian samples these are much higher.

The low quantities of free sulphur dioxide can also be explained by the *milléssime* of the wines (some are from 2012, some from 2013).

## CONCLUSIONS

White Cypriot wines vs. Romanian wines show the huge influence the climatic conditions have on the wine-making sector. Therefore, the highest alcoholic concentration 15,1 %vol. (Cypriot sample) compared to 14,4 %vol. (Romanian sample).

The lowest total acidity is found in a Cypriot sample -3,83 g/L compared to 6,43 g/L (Romanian sample).

The highest non-reductive extract for white wines is 23,7 g/L (Cypriot sample) compared to 18,1 g/L (Romanian sample).

**Acknowledgements:** *The wine samples were provided by the project PN-II-RO-CY-2013-1, grant no. 764/2014.*

## REFERENCES

1. Cotea V.D., Sauciuc J., 1985 - *Tratat de oenologie*, vol. 1, Ed. Ceres, București.
2. Jones V. Gregory et al., 2005 - *Climatic change and global wine quality* vol.73, no. 3, pp. 319-343, Kluwer Academic Publishers
3. Van Leeuwen.C., Friant P., Chone X., Tregoat O., Koundouras S., Dubordieu D., 2004 - *Influence of climate, soil, and cultivar on terroir*, Am. J. Enol. Vitic. 55(3):. 207-217.
4. \*\*\* OIV, 2013 - *Recueil des methodes internationales d'analyse des vins et des mouts*, Office International de la Vigne et du Vin, Editura O.I.V., Edition Officielle, Paris.

## STUDIES ON THE BEHAVIOR OF THE CHERRIES IN THE STORAGE WITH MODIFIED ATMOSPHERE OF CARBON DIOXIDE

### STUDII PRIVIND COMPORTAREA FRUCTELOR DE VIȘIN LA PĂSTRAREA ÎN ATMOSFERĂ MODIFICATĂ ÎN DIOXID DE CARBON

**VERINGĂ Daniela<sup>1</sup>**

**e-mail:** veringa.daniela@yahoo.com

**Abstract.** *A number of previous studies have established that the transportation and storage of fruit at low temperatures is insufficient in many cases and has to ensure optimal part of the gaseous medium characterized by certain concentrations of oxygen, carbon dioxide and nitrogen. The researchs hows the results obtained from the fruit of the cherry maintained in a modified atmosphere of carbon dioxide at various concentrations and for different periods of maintenance. We intended to correlate the administration of carbon dioxide with the exposure time to his treatment so thatby alternating carbon dioxide – rich atmosphere to reduce losses. The experiments revealed a series of issues related to weight loss which had a decreasing proportion related to the size of the CO<sub>2</sub> concentration. Regarding the organoleptic analysis were found differences between the experimental values used.*

**Key words:** carbondioxide, cherry, storage

**Rezumat.** *O serie de cercetări anterioare au stabilit că transportul și păstrarea fructelor la temperaturi scăzute este în multe cazuri insuficientă și trebuie să se asigure o componentă optimă a mediului gazos caracterizat de anumite concentrații ale oxigenului, dioxidului de carbon și azotului. Cercetările efectuate prezintă rezultatele obținute la fructele de vișin menținute în atmosferă modificată cu dioxid de carbon în diferite concentrații și diferite perioade de menținere. S-a urmărit corelarea administrării dioxidului de carbon cu timpul de suprapunere la acest tratament astfel încât prin alternarea atmosferei bogate în dioxid de carbon cu atmosfera normală, să se reducă pierderile. În urma efectuării experimentelor au rezultat o serie de aspecte referitoare la pierderile în greutate care au avut o evoluție descendentă proporțional cu mărimea concentrației de CO<sub>2</sub>. În ceea ce privește analiza organoleptică s-au constatat diferențe între valorile experimentale utilizate.*

**Cuvinte cheie:** dioxid de cabon, fructe de vișin, păstrare

## INTRODUCTION

Transporting perishable fruit in modified atmospheres have been studied in recent decades by several researchers. They found that the transport and storage of fruit at low temperatures is insufficient in many cases and has to ensure optimal

---

<sup>1</sup>Research and Development Institute for Industrialization and Marketing of Horticultural Product, București, Romania

component of the gaseous medium characterized by certain concentrations of oxygen, carbon dioxide and nitrogen, the use of gas activities having an inhibitory effect on metabolic and pathogens (Peleg, 1985; Ryall and Pentzer, 1979; FAO., 1988).

The effect of carbon dioxide in high concentrations on the preservation of fruit and vegetables is known for a long time and is differentiated not only in terms of species but also on the various actions positive or negative exerted on fruit (Thayer and Rajkowska, 1999; Thompson, 1998 Pratella *et al.*, 1991).

The essential aspect of positive action highlighted by research in recent years relate mainly to the possibility of preventing parasitic deterioration and slowing the natural process aging.

The negative aspect is the pathophysiological type (phytotoxicity and induction of metabolic alterations).

In research done was attempted correlation of CO<sub>2</sub> management administration with time of submission to this treatment to eliminate shortcomings discovered.

## MATERIAL AND METHOD

The research was conducted in laboratory conditions using the variety cherries 'Nana', regarding simulating conditions on the transport of cherries in modified atmosphere with carbon dioxide through intermittent treatment with this gas.

Were performed following conditioning room maintained:

V<sub>01</sub>- Control variant - Desiccator with normal atmosphere in keeping with the duration of 3 days; V<sub>02</sub>- Control variant - Normal atmosphere keeping in desiccator lasting 6 days; V<sub>1</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 10% for 1 day and 3 days hold time; V<sub>2</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 20% for 1 day and 3 days hold time; V<sub>3</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 30% for 1 day and 3 days hold time; V<sub>4</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 10% for 2 days and hold time 3 days; V<sub>5</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 20% for 2 days and hold time 3 days; V<sub>6</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 10% for 3 days and hold time 3 days; V<sub>7</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 10% for 1 day and 6 days hold time; V<sub>8</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 20% for 1 day and 6 days hold time; V<sub>9</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 30% for 1 day and 6 days hold time; V<sub>10</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 10% for 2 days and 6 days hold time; V<sub>11</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 20% for 2 days and 6 days hold time; V<sub>12</sub> - Modified atmosphere with CO<sub>2</sub> at a concentration of 30% for 2 days and 6 days hold time.

There were observations and determinations in dynamic, on: appearance of fruit; firmness; organoleptic analysis; weight loss; fruit plant health.

## RESULTS AND DISCUSSIONS

The research was conducted in laboratory conditions, cherries were sorted before starting experiments. They were carried out 3 replications for each version being stored in climate room at a temperature of 2 C. After one day (Table 1) or

two days (Table 2), carbon dioxide was removed by ventilation, with the aid of a compressor.

The first control on plant health of the fruit was performed after 3 days for variants V01, V1, V2, V3, V4, V5, V6, organoleptic analysis repeating it after 2 days (it is believed that within two days the fruit will be sold). During control was determined also the weight loss progress. The second inspection was carried out after 6 days for variants V02, V7, V8, V9, V10, V11, V12 followed by the same procedures.

Table 1

**Losses realized in experiments on the behavior of cherries after treatment with CO<sub>2</sub> during one day**

Option	Symbol	Total loss		Write-downs by					
				Weight loss		Impairment losses soaking		Mildwed losses	
		3 days	6 days	3 days	6 days	3 days	6 days	3 days	6 days
Control	V <sub>0</sub> <sub>1</sub>	16,2	-	3,4	-	12,8	-	-	-
Modified atmosphere with CO <sub>2</sub> -10%	V <sub>1</sub>	12,34	-	1,04	-	11,3	-	-	-
	V <sub>7</sub>	-	22,06	-	1,91	-	20,15	-	-
Modified atmosphere with CO <sub>2</sub> -20%	V <sub>2</sub>	11,25	-	0,99	-	10,26	-	-	-
	V <sub>8</sub>	-	17,55	-	1,7	-	15,85	-	-
Modified atmosphere with CO <sub>2</sub> -30%	V <sub>3</sub>	10,18	-	0,85	-	9,33	-	-	-
	V <sub>9</sub>	-	16,28	-	1,68	-	14,6	-	-

Table 2

**Losses realized in experiments on the behavior of cherries after treatment with CO<sub>2</sub> during two days**

Option	Symbol	Total loss		Write-downs by					
				Weight loss		Impairment losses soaking		Mildwed losses	
		3 days	6 days	3 days	6 days	3 days	6 days	3 days	6 days
Control	V <sub>02</sub>	-	26,9	-	4,6	-	22,1	-	0,2
Modified atmosphere with CO <sub>2</sub> -10%	V <sub>4</sub>	13,4	-	0,98	-	12,6	-	-	-
	V <sub>10</sub>	-	18,57	-	1,86	-	16,71	-	-
Modified atmosphere with CO <sub>2</sub> -20%	V <sub>5</sub>	12,11	-	0,9	-	11,21	-	-	-
	V <sub>11</sub>	-	17,45	-	1,68	-	15,77	-	-
Modified atmosphere with CO <sub>2</sub> -30%	V <sub>6</sub>	11,41	-	0,81	-	10,6	-	-	-
	V <sub>12</sub>	-	17,17	-	1,65	-	15,52	-	-

Analyzing the data in Tables 1 and 2 we find:

- a downward trend of losses by weight, in proportion to increase in carbon dioxide concentration and with duration of treatment;
- for variants with CO<sub>2</sub> treatment during 2 days, weight loss percentage is slightly lower than the duration of treatment option for 1 day. The same observation applies to softening and, also, total losses, exception being 30% solution where the results were weaker;
- mold appeared only in control version (6 days of storage), so CO<sub>2</sub> atmospheres totally inhibit mold attack.

For evaluating the organoleptic, there were held tests after each Control version. Organoleptic analysis was repeated after 2 days (during the fruit is sold) under the same conditions. The appreciation was recorded in charts, highlighting: appearance, firmness and taste of the fruits, according tasting sheets (Table 3).

Table 3

**Appreciation of cherries organoleptic qualities after three days keeping in modified atmospheres**

Organoleptic qualities	V <sub>01</sub>	V <sub>1</sub>	V <sub>2</sub>	V <sub>3</sub>	V <sub>4</sub>	V <sub>5</sub>	V <sub>6</sub>
Apperance	12,85	12,00	13,14	13,71	13,57	15	12,42
Firming	27	28	29	30	28	29	29
Taste	38,57	34,71	36,85	33,14	37,14	35,57	27,14
Total	78,42	74,35	78,99	76,85	78,71	79,57	68,56
Ranking	4	6	2	5	3	1	7

In terms of appearance of V<sub>5</sub> variant (20% CO<sub>2</sub> and two days treatment) has the maximum score, other variants fits between nice and very beautiful.

Regarding firmness, variant V<sub>3</sub> (30% CO<sub>2</sub> and 1 day treatment) is close to very good (maximum score 35) and the other, between good and very good (range 28-35 points).

In terms of taste control, Control version, has the highest score and falls near good qualifier. The other variants are between acceptable and good (30-40 points).

Doing all scores, ranked, is the first version V<sub>5</sub> (20% CO<sub>2</sub> and 2 days treatment).

Table 4

**Appreciation of cherries organoleptic qualities after 3 days in modified atmosphere and 2 days in normal atmosphere**

Organoleptic qualities	V <sub>01</sub>	V <sub>1</sub>	V <sub>2</sub>	V <sub>3</sub>	V <sub>4</sub>	V <sub>5</sub>	V <sub>6</sub>
Apperance	12,6	10,6	12,6	11,4	11,4	13,2	13,8
Firming	21	26,6	27	27,2	28	28,2	30,8
Taste	38,5	36,14	37,87	30,71	35,57	37,14	30,0
Total	72,1	73,34	77,47	69,31	74,97	78,54	74,6
Ranking	6	5	2	7	3	1	4



Version  $V_5$  (20%  $\text{CO}_2$  with 2 days treatment) has the most beautiful appearance, even after the second test.

$V_{01}$  and  $V_6$  variants are between nice (12 points) and very nice (15 points). Regarding firmness, the version with the highest score is  $V_6$  (30%  $\text{CO}_2$ , 2 days treatment).

For taste, Control version kept the highest score and other variables approaching adjective "good".  $V_3$  and  $V_6$  variants had the qualifier "acceptable".

The first place is version  $V_5$  (20%  $\text{CO}_2$  and 2 days treatment), than closely was  $V_2$  variant (20%  $\text{CO}_2$  and 1 day treatment) and  $V_4$  variant (10%  $\text{CO}_2$  and 2 days treatment).

After 6 days of maintenance in modified atmospheres and two days in normal atmosphere have made the same tests which are presented in Table 5 and Table 6.

Table 5

**Appreciation of cherries organoleptic qualities after 6 days in modified atmosphere**

Organoleptic qualities	$V_{02}$	$V_7$	$V_8$	$V_9$	$V_{10}$	$V_{11}$	$V_{12}$
Appearance	14	14,14	14,57	14,57	15	15	14,57
Firming	24	26,71	30	30	31	32	26,71
Taste	45,71	41,42	40,0	37,14	38,57	38,57	37,14
Total	83,71	82,27	84,57	81,71	84,57	85,57	78,41
Ranking	4	5	2	6	2	1	7

Appearance has maximum at 2 versions:  $V_{10}$  and  $V_{11}$ , the others approaching „Very Nice”.

Version  $V_{11}$  (20%  $\text{CO}_2$  and 2 days treatment) has the best score at the farm. In terms of taste, version Control has the highest score.

The variants  $V_7$  and  $V_8$  had between good and very good (40-50 points).

After ranking,  $V_{11}$  (20%  $\text{CO}_2$  and 2 days treatment) ranks first, followed by variants  $V_8$  and  $V_{10}$  with the same score.

Table 6

**Appreciations of cherries organoleptic qualities after 6 days in modified atmospheres and two days in normal atmosphere**

Organoleptic qualities	$V_{02}$	$V_7$	$V_8$	$V_9$	$V_{10}$	$V_{11}$	$V_{12}$
Appearance	13	13	13,5	13,5	13,5	14	13
Firming	26,83	29,16	30,0	29,16	29,16	30,0	29,16
Taste	43,33	41,33	40,0	33,66	40,66	40,33	36,33
Total	83,16	83,49	83,5	76,32	83,32	84,33	78,49
Ranking	5	3	2	7	4	1	6

After the second test, version  $V_{11}$  (20%  $\text{CO}_2$  and 2 days treatment) looks the most beautiful.

In terms of firmness,  $V_8$  and  $V_{11}$  variants are between good and very good. Taste has good value in Control version.

Consulting ranking, we found that version V<sub>11</sub> (20% CO<sub>2</sub> and 2 days treatment) is on the first place, followed by the V<sub>8</sub> variant (20% CO<sub>2</sub> and 1 day treatment).

After being held for five days in exicator, under high relative humidity and temperature in the laboratory (cca. 20°C) shows that fruits go through a physiological degradation process, which externalizes by highlighting surface stains fade. Referring to plant health, finds that after 5 days at 20 ° C, with 30% CO<sub>2</sub> and 2 days treatment, has the lowest percentage of fruit affected by pathogens.

Also, between the variants with 20 and 30% CO<sub>2</sub> and 1 day treatment, the differences are very small. After 9 days of maintenance in these conditions, the fruit surface was covered with a rich *Rhizopus mycelium stolonifer*.

## CONCLUSIONS

Laboratory tests undertaken followed correlation of carbon dioxide during the administration of allegiance to the treatment of cherries, so that by altering carbon dioxide-rich atmosphere with normal atmosphere to eliminate or reduce the deficits detected in previous tests.

The conclusions after carrying out experiments with cherry are:

- losses decreased with increasing CO<sub>2</sub> concentration and duration of maintenance treatment;
- weight loss in variants with CO<sub>2</sub> are lower by 60% - 70% compared to losses in control variant;
- organoleptic properties of cherries are rated with the highest scores at variant with 20% CO<sub>2</sub> and 2 days treatment;
- the percentage of fruit affected by pathogens decreases with increasing CO<sub>2</sub> concentration and duration of maintenance treatment with 50-70% compared to Control variant;
- Optimal treatment for cherry version for transport is considered to be 20% CO<sub>2</sub> atmosphere maintained for 2 days and a temperature of approx. 2° C.

## REFERENCES

1. Cociu V., Oprea Șt., 1989 - *Metode de cercetare în ameliorarea plantelor pomicele*. Ed. Dacia, Cluj, p. 29, 124-129.
2. Davarynejad G.H., Szabo Z., Nyeki J., Szabo T., 2008 - *Phenological Stages, Pollen Production Level, Pollen Viability and in vitro Germination Capability of Some Sour Cherry Cultivars*. Asian Journal of Plant Sciences, 7: 672-676.
3. Tofan Clemansa, 2001 – *Igiena și securitatea produselor alimentare*. Editura AGIR, Bucuresti, p. 56.
4. \*\*\*, 2009 – *Agricultura UE*. Comisia Europeană.

## RESEARCH ON EVOLUTION OF THE MAIN PESTS OF PLATANE IN ROMANIA

### CERCETĂRI PRIVIND EVOLUȚIA PRINCIPALILOR DĂUNĂTORI AI PLATANULUI ÎN ROMÂNIA

**BĂLĂNESCU (NEACȘU) Irina Ioana<sup>1</sup>, ROȘCA I.<sup>1</sup>**

**e-mail:** ioanrosca\_usamv@yahoo.com

**Abstract.** In recent years, in Bucharest and in other urban centres in our country began to be placed *Platanus* spp. trees. In the area of Bucharest, in the year 2014 and 2015 was an extension of two dangerous *Platanus*' pests [*Thesycamore* lace bug (*Corythucha ciliata* Say, 1832 - Hemiptera: Tingidae) and leaf-mining moth (*Phyllonorycter platani* Staudinger, 1870 - Lepidoptera: Gracilariidae)]. Research has followed the evolution of these pests in the nursery from Bolintin Deal and identify other potential pests that have the potential to attack the *Platanus* spp.. In order to carry out effective and well timed control methods, bio monitoring studies of the pests are necessary. The biology, ecology and the vulnerable life periods of the pests were studied. It was noted intensity and frequency of infested leaves, calculating the degree of attack, these indicators are variable, depending on the pest life cycle or the period studied. For *Corythucha ciliata* recorded population was between the 2 and 8 exemplars/leaf attacked, while for *Phyllonorycter platani* attack was between 441 and 549 leaves attacked, regarding the number of mine / leaf between 1 and 10 and ranges of attacked leaf surface is between 1.4 and 5.5/attacked leaf. Are presented other pests identified in the nursery.

**Key words:** evolution of *Platanus* pests in nursery.

**Rezumat.** În ultimii ani, în București și în alte centre urbane din țara noastră au început să fie plantați copaci de *Platanus* spp. În zona din București, în anul 2014 și în 2015 a fost o extensie a doi dăunători periculoși ai platanului "[Ploșnița dantelată a platanului (*Corythucha ciliata* Say, 1832 - Hemiptera: Tingidae) și molia minieră a frunzelor (*Phyllonorycter platani* Staudinger, 1870 - Lepidoptera: Gracilariidae)]. Cercetarea a urmărit evoluția acestor dăunători în pepiniera de la Bolintin Deal și identificarea altor dăunători potențiali care au capacitatea de a ataca *Platanus* spp. În scopul de a pune la punct metode eficiente și oportune de control, sunt necesare studii de monitorizare a biologiei dăunătorilor. Biologia, ecologia și perioadele de viață vulnerabile ale dăunătorilor au fost studiate. S-a evaluat intensitatea și frecvența frunzelor infestate, s-a calculat gradul de atac, acești indicatori sunt variabili, depinzând de ciclul de viață a dăunătorilor sau perioada studiată. Pentru *Corythucha ciliata* s-a înregistrat populația care a fost între 2 și 8 exemplare / frunză atacată, în timp ce pentru *Phyllonorycter platani* atacul a fost între 441 și 549 de frunze atacate, în ceea ce privește numărul de mine pe frunze acesta a fost

---

<sup>1</sup> University of Agronomic Sciences and Veterinary Medicine of Bucharest, Romania

între 1 și 10 și intervale de suprafață frunze atacate este între 1,4 și 5.5. Sunt prezentați și alți dăunători identificați în pepinieră.

**Cuvinte cheie:** evoluția dăunătorilor platanului în pepinieră

## INTRODUCTION

The Sycamore lace bug, *Corythucha ciliata* (Say, 1832), is a nearctic tingid that feeds on the underside of the leaves of *Platanus spp.* (called sycamore trees in North America and platanes in the Old World). In North America, the area of occurrence of the Sycamore lace bug extends throughout eastern USA and eastern Canada (Halbert and Meeker, 1998), and the species was introduced and is successfully established in Europe and Asia. *Corythucha ciliata* species has been reported in Europe in 1964, in Padova, Italy, and Romania of Kis, in 1990. The greatest danger of this pest is the association brought two fungi *Ceratocystis fimbriata* Ellis and Halst. form platani J. M. Walter and *Apiognomonina (Gnomonia) veneta* Sacc. and Speg. that through synergy with pests can cause decline and death of trees. The two pests have, in addition to their herbivorous character, a negative impact on the visitors of the parks or streets both from the point of view of aesthetics and hygiene and much more sycamore lace bug can be an agent of insect-caused dermatosis considered subjects who visit or live near wooded areas or trees which are infested.

*Phyllonorycter platani* was originally described from northern Italy by Staudinger in 1870 (Šefrov, 2001). Deschka (1984, 1995) believes that its autochthonous (origin) area includes the Balkans and western Asia in accord with the distributional area of the primary host plant (*Platanus orientalis* L.). This distributional area reaches, however, far to the east crossing Middle East up to Central Asia. Deschka (1995) believes that the sudden spread of *Phyllonorycter platani* was started round 1970. But this invasion began obviously decades sooner. It was followed by several distributional waves with the rise of numerous locally limited populations representing the secondary distributional centres. *Phyllonorycter platani* Stdgr., has been recorded in Romania for the first time at Bucharest in 1970 (Drăghia, 1970; Rakosy *et al.*, 2003) or in 1996 by Sandru after Ureche Camelia (Ureche, 2006) Larvae of pest, develop mines large and very distinctive, with several mines often on one leaf in leaves.

## MATERIAL AND METHOD

Identification of the pests was done in nursery "Bolintin Deal" at the north of Giurgiu district, 18 km away from Bucharest, near Bucharest- Pitesti highway, in a small company in the field of ornamental plants nursery, its geographical coordinates are 44° 27' 33" North, 25° 49' 16" East.

The climate is continental, with very hot summers, and not very important quantities of precipitation which fall as showers, and cold winters with strong blizzards and frequent worm periods. The average rainfall is 650-700 mm / in most rainy month from the year is June (maximum of 24 hours was 103.2 mm at 21.VII.1978).

There are years when rainfall was doubled, and years when rainfall decreases, appearing drought and rainfall is 250-300 mm, annual average temperature for this

area was 10.2°C average temperature of January was -2.3oC, and the average temperature in July was 23.7°C, (thermal amplitude was 21.4oC). In 2007 the business was started by importing plants from Italy, Holland and France.

Experience has taught them that imported plants that have started their life cycle in a certain climate, have lower rooting rates and the plants develop slower. Thus, in 2009 the owners decided to produce their own plants in Romania. The first author started the business by cultivating her first 2000 sq. m. with plants in the field. Now company is growing and at present the production covers over 2 ha plants, (1 ha of container fields) and it's financial data shows that in 2014 sales were 93,500 Ron and in 2015 sales (till 01 October) were 55,000 Ron.

Structure and volume of number of tree sold is increasing from 2014 to 2015 (Tab. 1).

Table 1

**Structure and volume of number of tree sold is increasing from 2014 to 2015**

<b>Names of species</b>	<b>No.trees/2014</b>	<b>No. trees/2015</b>
<i>Platanus acerifolia</i>	3000	7500
<i>Rosa spp.</i>	3000	1000
<i>Prunus nigra</i>	1000	300
<i>Betula alba</i>	200	100
<i>Magnolia spp.</i>	500	50
<i>Thuja orientalis</i>	600	600
<i>Thuja occidentalis "Smaragd"</i>	300	300
<i>Tilia tomentosa</i>	1000	150
<i>Acer saccharinum</i>	500	1000

**Fig. 1 - Images of Platanus trees batch from nursery**

The extremely vast assortment of plants, about 50 species are grown, ranges from deciduous tree to conifers, climbers, roses, topiary, coming in various sizes from small and medium up to unique specimens (*Platanus acerifolia*, *Thuja occidentalis* "Smaragd", *Picea pungens* "Glauca globossa", *Acer platanoides*, roses, *Prunus cerasifera* "Nigra", *Juniperus spp.* etc.). Financial data shows that nursery Bolintin is viably because there are, in 2014 sales of 93,500 Ron and in 2015 sales (till 01 October) 55,000 Ron.

Harmful insect species were noted according to usual methods. Because we done our observation in nursery, number of leaves of *Platanus* trees was considered as average 45. It was taken into consideration 4 batch (Fig. 1) of *Platanus* trees (761 trees). Samples were collected in the field, and more detailed processing was carried out in laboratory conditions. Species of insect pests were determined in the laboratory. Microscopic techniques were used to determine of some species.

Finally, the found species were classified systematically, but in this paper will be presented only four species *Corythucha ciliata* Say, 1832 (Sycamorelace bug); *Phyllonorycter platani* (Sycamore leaf-miner pest); *Metcalfa pruinosa* (citrus flatid planthopper) and *Lymantria dispar* (Gypsy Moth) are moths in the order *Lepidoptera*, family *Erebidae*.

In order to avoid damages noted in 2014, in 2015 it was applied an expensive scheme of treatments, 5 applications (Tab. 2).

Table 2

Scheme of treatments applied in nursery during year 2015

No. of treatment	Data of treatment	Comercial product (active ingredients)	Dose/10 l water
1	6.05	Confidor 200 SL (imidacloprid 200 gr/litru) Dursban 50 W (chlorpyrifos 50%)	5 ml 10 ml
2	25.05	Mimic 2 LV (tebufenozide 24%) Topsin 70 WDG (tiofanat metil 41-60%) Dithane M 454 (mancozeb 80%) Atonik SL (nitroguaiacolat de sodiu 1 gr/kg; orto-nitrofenolat de sodiu 2 gr/kg; paranitrofenolat de sodiu 3 gr/kg) Lannate 20 SL (metomil: 200 g/l) Nissorun 10 WP (hexitiazox 10%)	5 ml 10 g 20 g  10 ml 20 ml 5 g
3	20.06	Confidor 200 SL (imidacloprid 200 gr/litru) Topsin 70 WDG (tiofanat metil 41-60%) Dithane M 454 (mancozeb 80%) Atonik (nitroguaiacolat de sodiu 1 gr/kg; orto-nitrofenolat de sodiu; 2 gr/kg; paranitrofenolat de sodiu 3 gr/kg) Lannate 20 SL (metomil 200 g/l) Nissorun 10 WP (hexitiazox 10%)	5 ml 10 g 20 g  10 ml 20 ml 5g
4	29.07	CONFIDOR 200 SL 200 SL (imidacloprid 200 gr/litru)	5 ml
5	29.10	Confidor Energy (deltametrin 21-40%) Decis Mega 50 EW (deltametrin 50 g/l) Novadim Progress (dimetoat 400 g/l) Kumulus DF (sulphur 80%)	5 ml 10 ml 20 ml 30 g



## RESULTS AND DISCUSSIONS

The *Platanus* species have few specialized phytophagous pests. During our observations, in 2014, it were registered on *Platanus acerifolia* the following pests: *Corythucha ciliata* Say, 1832 (Sycamorelace bug), order *Hemiptera*, family *Tingidae*; *Phyllonorycter platani* (Sycamore leaf-miner pest) Staudinger, 1870, order *Lepidoptera* family *Gracilariidae*; *Acalyptris platani* Muller- Rutz, 1934, order *Lepidoptera*, family *Nepticulidae*; possible *Epirrita autumnata* Borkhausen, 1794 (autumnal moth) order *Lepidoptera*, family *Geometridae*; probably *Acleris forsskaleana* Linnaeus 1758 (Maple Leaf-tier Moth) *Lepidoptera*, *Tortricidae*; *Fagocyba cruenta* Herrich-Schaffer, 1838, order *Homoptera*, family *Cicadellidae* and *Drepanosiphum platanoidis* (= *platanoides*) (Common Sycamore Aphid) order *Homoptera*, family *Drepanosiphidae*. During our observations, in 2014 and 2015, it wereregistered on *Platanus acerifolia* the following main pests: *Corythucha ciliata* Say, 1832 (Sycamorelace bug), order *Hemiptera*, family *Tingidae*; *Phyllonorycter platani* (Sycamore leaf-miner pest) Staudinger, 1870, order *Lepidoptera* family *Gracilariidae*, in 2015 was observed attack of *Metcalfa pruinosa* Say, 1830 (citrus flatid planthopper) order *Hemiptera*, suborder *Auchenorrhyncha*, superfamily *Fulgoroidea*, family *Flatidae* and *Lymantria dispar* L. 1758, (Gypsy Moth), order *Lepidoptera*, family *Erebidae*.

*Corythucha ciliata* Say, 1832 (Fig. 2), it was observed from August, but it's attack was no heavy. In 2014 from 761 trees, in 18 August, only 5 (1.31%) were attacked with 6 leaves with pest colonies, in 29 August, 9 (1.18%) with 19 leaves with pest colonies, in 18 September, 48 (6.31%) with 122 leaves with pest and in 16 October, 47 (6.18%) with 144 leaves with pest colonies. Maximum number of adults and nymphs/leaf was 45. In 2015 due to treatments applied in all 5 variants from table 2 only few exemplars, without importance, were observed. The sycamore lace bug is the only lace bug listed as feeding on *P. occidentalis* according to the world hostlist for lace bugs (Drake and Ruhoff, 1965).

*Phyllonorycter platani* Staudinger, 1870 (Fig. 3) it was identified from large mines, often having several larvae in one leaf, generally underside between veins (Fig. 4). The upper side becomes mottled. On upper side can be over veins (British leaf miners). The mine begins as an epidermal corridor, sometimes several cm in length. This widens into a shallow, greyish green, irregularly lobed blotch. The fully developed mine is an orange brown tentiform mine with a number of length folds. Almost all mines are lower-surface. The upperside of the mine then is a mottled oval, because the larva here and there has eaten holes in the roof of the mine, i.e., the palissade parenchyma. Pupation inside a white cocoon. It was observed, in 2014, on 30 August as initial attack with small larvae in leaves mines, which become large and very distinctive, with several mines often appearing on one leaf in October (Fig. 5).

The moths fly in late April to May and in August. Wingspan (distance from one wingtip to the other wingtip) is 8-10 mm. The pupae overwinter in mines in fallen leaves and there are 2-3 adult flights/year first is starting in late April to May and the second in August. In 2014 Pest attack was no heavy. In 18 August and 16 October

from 761 trees 222 (29.17%) were attacked by pest, from these 41.38% had only 1 leaf attacked with onemine, 6.9% with two mines, 17.24% with threemines and 34.48% with five mines.



**Fig. 2** - Sycamore lace bug (*Corythucha ciliata*), adults, larvae and frass

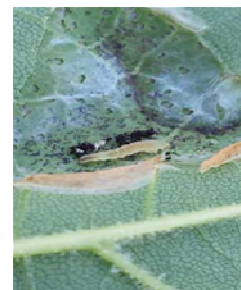
In 2015 main points of biology of *Phyllonorycter platani* are: May 5, adults flight; June 18, appearing of larvae; July 3, existence of larva+pupa+adult; August 13, existence of larva+adut; September 1 and October 19, larve's presence. The moths's larvae (Fig. 5) is colourless and transparanthave a head capsule and chewing mouthpartswith opposable mandibles, six thoracic legs andabdominal legs. In leaf mine, larvae made twotypes of frass: small, light brown granulescattered throughout the mine, and larger blackish brown grains in an elongate clump. The light spots are windows the larva has eatenin the palissade parenchyma, the roof of themine.



**Fig. 3** - Sycamore leaf-miner



**Fig. 4** - Sycamore leaf-miner pest attack



**Fig. 5** - Sycamore pest, adultas mines on underside leaf surface leaf-miner larvae

*Metcalfa pruinosa* (Fig. 6) is a polyphagous planthopper, recently introduced from North America, which can cause serious damage on grapevine. *Metcalfa pruinosa* feeds on a wide variety of trees and shrubs. Distribution: North America, Cuba, Central and South America; adventive in Europe. European hosts for *Metcalfa*



*pruinosa* include through many others *Acer campestre* L., *Acer platanoides* L., *Clematis vitalba* L. and others.

Regarding of economic importance, of considerable concern in orchards and vineyards in its introduced range, particularly in southern Europe. *Metcalfa pruinosa*, overwinters as eggs inserted in woody tissue or under bark. First nymphs are found on the leaves and stems in May, while adults are present from July to October. Adults are 5–8 mm long, with large moth-like wings, bluish in colour, covered with waxy powder. Dense populations of nymphs cause stunting of the shoots, while those of adults produce large quantities of honeydew on which sooty mould develops. *Metcalfa pruinosa* attack in 2015 was almost insignificant, adults were observed for the first time on June 07 and from 100 analysed trees, when 160 adults were observed.



Fig. 6 -Metcalfa pruinosa adult



Fig. 7 – Attack of Lymantria dispar larvae

*Lymantria dispar* L. 1758 is native to temperate forests in western Europe. This moth is an important defoliator on broad-leaf and conifer trees. Male moths are dark buff and fly readily during the day. Females are white with black, wavy markings, they have robust abdomens and do not fly, and their wingspan can reach 5 cm. The larval stage (caterpillar) is hairy, and a mature larva is 50-65 mm long with a yellow and black head. The pupal stage is dark reddish-brown and is held in place to some object by small strands of silk. Egg masses deposited by females during July overwinter on trees, eggs hatch from late April through early May. Small first instar larvae do not feed right after they hatch and can be dispersed by wind. Young larvae feed on foliage (fig. 7) and remain on host plants night and day. Pupation takes place during late June and early July. Adults start emerging in late June with peak emergence in mid-July. This pest produces one generation a year in Romania.

In nursery it was registered a weak attack, few larvae nests and of course a light defoliation (defined as 0 to 30% loss of foliage) and has little effect on the health of trees, defoliation being barely detectable.

From all five variants presented in Table 2, variant 5 consisting of Confidor Energy (deltametrin 21-40%) 5 ml; Decis Mega 50 EW (deltametrin 50 g/l) 10 ml; Novadim Progress (dimetoat 400 g/l) 20 ml; Kumulus DF (sulphur 80%) 30 g. in 10 l water has the greatest efficiency 98% after 3-4 days from treatments.

## CONCLUSIONS

During our observations, regarding *Corythucha ciliata* in 2014 from 761 trees, in 18 August, only 1.31% were attacked with 6 leaves with pest colonies, in 29 August, 1.18% with 19 leaves with pest colonies, in 18 September, 6.31% with 122 leaves with pest and in 16 October, 6.18% with 144 leaves with pest colonies. Maximum number of adults and nymphs/leaf was 45. In 2015 due to treatments applied only few exemplars, without importance, were observed.

In 2014 *Phyllonorycter platani* attack was no heavy. In 18 August and 16 October from 761 trees 29.17% were attacked by pest, from these 41.38% had only 1 leaf attacked with one mine, 6.9% with two mines, 17.24% with three mines and 34.48% with five mines, in 2015 main points of biology of *Phyllonorycter platani* are: May 5, adults flight; June 18, appearing of larvae; July 3, existence of larva+pupa+adut; August 13, existence of larva+adut; September 1 and October 19, larve's presence.

*Metcalfa pruinosa* attack in 2015 was almost insignificant (adults were observed for the first time on June 07 and from 100 analysed trees, when 160 adults were observed, in nursery it was registered a *Lymantria dispar* weak attack, few larvae nests and of course a light defoliation.

There are variants of chemical treatments for controlling of main pests.

**Acknowledgments:** Thanks, in this way, to the company Nursery Bolintin, for its support in realising experiments and permission to use the data obtained.

## REFERENCES

1. Deschka G., 1984 - *Phyllonorycter (Lithocolletis) platani* (Stauinger 1870). Jber Steyrer Ent. Runde, pp. 21-31.
2. Deschka G., 1995 - *Schmetterlinge als Einwanderer. Einwanderer - Neue Tierarten erobern Östeneich*. Snpfia 37 zugleich Kataloge des ÖÖ. Landesmuseum N. E., 84, pp. 77-28.
3. Drake C.J., Ruhoff F.A., 1965 - *Lacebugs of the World: A Catalog* (Hemiptera: Tingidae). Smithsonian Institution, Washington, D.C., United States National Museum Bulletin 213, 634 p.
4. Drăghia I., 1970 - *Nouvelles contributions a la connaissance des insectes mineurs de Bucharest et de ses environs*. Trav. Mus. Hist. nat. „Grigore Antipa“. Bucarest. 10: 235-240.
5. Halbert S.E., Meekef J.R., 1998 - *The Sycamore Lace Bug, Corythucha ciliata* (Say) (Hemiptera: Tingidae). Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Entomology Circular No. 387, 2 p.
6. Rakosy et al., 2003 - *Catalogul Lepidopterelor Romaniei/Verzeichnis der Schmetterlinge Rumaniens*. Soc. Lepid. Rom. Cluj-Napoca. Tipar Romsver, Cluj- Napoca, 447 p.
7. Šefrov H., 2001 - *Phyllonorycter platani* (Staudinger) - A review of its dispersal history in Europe (Lepidoptera, Gracillariidae). Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis Sborník Mendelovy Zemědělské a Lesnické Univerzity v Brně, IL (5): 71-75.
8. Ureche C., 2006 - *Invasive leaf miner insects in Romania*. IUFRO Working Proceedings of the Workshop 2006, Gmunden/Austria.

## OBSERVATION ON THE STRUCTURE AND ECOLOGICAL PARAMETERS OF THE POPULATION OF INVERTEBRATES IN PLUM ORCHARDS

### OBSERVAȚII PRIVIND STRUCTURA ȘI PARAMETRII ECOLOGICI AI POPULAȚIEI DE INVERTEBRATE ÎN PLANTAȚIILE POMICOLE DE PRUN

**BUTNARIU Gianina<sup>1</sup>, TĂLMACIU M.<sup>1</sup>, TĂLMACIU Nela<sup>1</sup>, HEREA Monica<sup>1</sup>**

**e-mail:** gianina.butnariu@yahoo.com

**Abstract:** During the research in Vasile Adamachi Iasi stationary were studied two plum plantation, one represented by the Rivers early variety and other Stanley varieties. To prepare the structure and ecological parameters representative entomofauna of invertebrates from the plantation, were set six traps soil type Barber and during the research were collected species belonging to the orders: Coleoptera (*Dermestes lanarius*, *Polydrosus amoenus*, *Tomoxia biguttata*, *Anisodactylus binotatus*, *Harpalus distinguendus*, *Armadillidium vulgare*, *Galeruca pomonae*, *Pseudophonus rufipes*), Hymenoptera (bees, wasps, ants), Arachnida, Heteroptera (*Pyrhocoris* sp.), Lepidoptera, Gastropoda (snails) and Isoptera. Larger differences, appear significant from year to year which shows that the greatest influence on entomofauna have in general environmental conditions from one year to another in this case were very different.

**Key words:** Coleoptera, plum plantation, useful entomofauna

**Rezumat:** În perioada de cercetare la staționarul Vasile Adamachi Iași au fost luate în studiu două plantații de prun, una reprezentată de soiul Rivers timpuriu, iar cealaltă de soiul Stanley. Pentru întocmirea structurii și a parametrilor ecologici reprezentativi entomofaunei de nevertebrate din plantație, au fost fixate șase capcane de sol de tip Barber, iar pe parcursul cercetărilor au fost colectate specii aparținând ordinilor: Coleoptera (*Dermestes lanarius*, *Polydrosus amoenus*, *Tomoxia biguttata*, *Anisodactylus binotatus*, *Harpalus distinguendus*, *Armadillidium vulgare*, *Galeruca pomonae*, *Pseudophonus rufipes*), Hymenoptera (albine, viespi, furnici), Arachnida, Heteroptera (*Pyrhocoris* sp.), Lepidoptera, Gastropoda (melci) și Isoptera. Diferențe mai mari, semnificative apar de la un an la altul ceea ce denotă că influența cea mai mare asupra entomofaunei o au în general condițiile de mediu care de la un an la altul în cazul de față au fost foarte diferite.

**Cuvinte cheie:** Coleoptera, plantații de prun, entomofauna utilă.

## INTRODUCTION

Plum tree characterized as the tree of life or hope, is spread from the plains to the hills and sometimes to the Carpathian foothills. Over the years, its products have established plum and livelihood of farmers while contributing to the country's reputation. High adaptability to different climates and soil, made plum tree grow and produce spontaneously or cultivated area of distribution and variety is virtually

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

limitless variety. In this context, Romania has become the country with the largest fruit production in the Balkans and in Europe. Current level protection requirements is not only save the crop, but the fight must be carried out to increase the percentage of fruit quality, perfectly healthy, with no accumulation of toxic residues in fruits and their preparations (Minoiu and Lefter, 1987, Lăcătușu and Pisică, 1980 )

### MATERIAL AND METHOD

For the collection of the biological material were used soil traps type Barber. This consisted of placing in the soil of 6 recipiente has been placed a solution of formalin (40%) diluted with water to a concentration of 5% (Minoiu and Lefter, 1987).

The location of traps was made on two rows at a distance of 12 meters between the rows and 6 meters between traps by 3 traps per row.

The sampling procedure was done in each of the three years of observation (2013, 2014 and 2015) in the period from May to August, at intervals of about 10-20 days. At each harvest the collected insects were placed in gauze cloth, each sample separately and replaced or supplemented then the liquid in the trap. The material was then tagged, of the label specifying: data collection, the number of traps , the stationary and variety. In laboratory the material was cleaned of plant debris and then washed under running water, it is selected the order or species.

### RESULTS AND DISCUSSIONS

The centralizing results useful and harmful entomofauna found in plum groves of stationary Vasile Adamachi, jud. Iasi at the two varieties studied Rivers timpuriu and Stanley, during the three years using soil traps type Barber.

The summary of obtained results in two varieties stationary of Vasile Adamachi Iasi in 2013. After to applying the method in the two varieties were collected a total of 30 species with a total of 393 samples (Table 1). Of the 30 species collected, a total of 10 species were common in both varieties. They were *Dermestes laniarius*, *Heteroptera* (bedbugs) *Polydrosus sericeus* Schall, *Coccinella 7punctata*, *Homoptera* (cycads) *Polydrosus amoenus* Schall, *Tomoxia biguttata*, *aenea Amara*, *Harpalus calceatus* Duft., *Anisodactylus binotatus* F .. The species with the highest number of samples collected were: *Dermestes laniarius*, with 70 samples, *Heteroptera* (bugs) with 66 samples, *Coccinella septempunctata* L. and *Polydrosus sericeus* Schall.cu 41 samples, *Cyanis cinerea*, 34 samples, *Tomoxia biguttata* with *Homoptera* (cycads) with 15 samples and *calceatus Harpalus* Duft with 14 samples, *Harpalus distinguendus* with 12 samples, 11 samples obtain *Polydrosus amoenus* Schall and *Amara aenea* with a total number are 10 samples. The other species have been collected a number between 2 and 7 samples. In 2014, the two were collected plum plantation of 45 species with a total of 1,094 samples (Table 2). Of the 51 species collected species (Chatened du Gaetan, 1990, Panin, 1951, Safavi, 1968), numbers 22 species were common in both plantations. They were: *Araneide*, *Diptera* (adults), *Hymenoptera* (wasps), *Harpalus distinguendus*, *Hymenoptera* (bees), *Pseudophonus griseus*, *Coccinella conglobata*, *Anisodactylus binotatus*, *Calathus fuscipes*, *Carabus coriaceus*, *Adalia bipunctata*, the species with the largest number of specimens collected were: *Orthoptera* (120), *Hymenoptera* (bees (106)), *Pseudophonus rufipes* (86), *Harpalus distinguendus* (81),

*Hymenoptera* (ants (53)) *Lepidoptera* (51), *Hymenoptera* (wasps (48)), *Pseudophonusgriseus* (46) *Araneide* (42), *Homoptera* (cycads) (36), *Diptera* (adults) (31) *Galeruca tanacetii* (30), *Amara aenea* (29), *Coccinella conglobata* (28), *Anisodactylus binotatus* (25) *Opatrum sabulosum* (26), *Ontophagus ovatus* (20). Another species collected were recorded from 2 to 20 samples.

Table 1

The structure of collected species in 2013

No.	Name of species	V. Adamachi stationary		Total samples
		Rivers	Stanley	
1.	<i>Dermestes lanarius</i>	42	28	70
2.	<i>Heteroptera (ploşnita)</i>	38	28	66
3.	<i>Cyaniris cyanea F.</i>	34	-	34
4.	<i>Polydrosus sericeus Schall</i>	26	15	41
5.	<i>Coccinella septempunctata</i>	14	27	41
6.	<i>Homoptera(cicade)</i>	12	2	14
7.	<i>Polydrosus amoenus Schall</i>	9	2	11
8.	<i>Tomoxia biguttata</i>	8	7	15
9.	<i>Chilopoda longitarsis</i>	7	-	7
10.	<i>Amara aenea</i>	7	3	10
11.	<i>Harpalus distinguendus</i>	12	-	12
12.	<i>Cymindis vaporariorum L.</i>	6	-	6
13.	<i>Hymenoptera (viespi)</i>	5	-	5
14.	<i>Ophonus azureus</i>	5	-	5
15.	<i>Necrophorus vespillo L.</i>	3	-	3
16.	<i>Harpalus calceatus</i>	3	11	14
17.	<i>Armadillidium vulgare</i>	3	-	3
18.	<i>Anisodactylus binotatus F.</i>	2	3	5
19.	<i>Balaninus glandium</i>	2	-	2
20.	<i>Podonta nigrita F.</i>	2	-	2
21.	<i>Galeruca pomonae</i>	2	-	2
22.	<i>Halyzia 22 punctata</i>	-	4	4
23.	<i>Stomodes gyriscollis</i>	-	4	4
24.	<i>Anisodactylus signatus</i>	-	3	3
25.	<i>Amara eurynota</i>	-	3	3
26.	<i>Calathus melanocephalus</i>	-	3	3
27.	<i>Coccinella hieroglyphica</i>	-	2	2
28.	<i>Ontophagus ovatus</i>	-	2	2
29.	<i>Lixus iridis Ol</i>	-	2	2
30.	<i>Eremotes ater L</i>	-	2	2
Total 49 species		242	151	393

In 2015, the two stationary were collected a total of 51 species with a total of 1936 samples (Table 3). Of the species collected species, numbers 22 species were common to both varieties (Chatened du Gaetan, 1990, Panin 1951, Rogoianu and Perju, 1979). They were *Dermestes lanarius*, *Coleoptera* (larvae), *Hymenoptera* (wasps), *Heteroptera* (*Pyrrhocoridae*) *Polydrosus sericeus* Schall., *Calathus fuscipes* Goeze., *Gastropoda* (snails), *Adalia bipunctata*, *Coccinella septempunctata*, *Anisodactylus binotatus*, *Orthoptera* (grasshoppers), *Homoptera* (cycads), *Hymenoptera* (bees), *Harpalus distinguendus* Duft.

Table 2

## The structure of collected species in 2014

No.	Name of species	V. Adamachi stationary		Number of samples
		Rivers	Stanley	
1	<i>Pterostichus niger</i> Schall	3	-	3
2	<i>Araneida</i>	33	9	42
3	<i>Diptera (adulti)</i>	23	8	31
4	<i>Opatrum sabulosum</i>	26	-	26
5	<i>Hymenoptera (viespi)</i>	36	12	48
6	<i>Harpalus distinguendus</i>	13	68	81
7	<i>Gastropoda (melci)</i>	13	2	12
8	<i>Tomoxia biguttata</i>	12	-	12
9	<i>Hister purpurascens</i>	10	-	10
10	<i>Hymenoptera (albina)</i>	92	14	106
11	<i>Galeruca tanacetii</i>	25	5	30
12	<i>Harpalus calceatus</i>	12	7	19
13	<i>Lepidoptera (larve)</i>	37	14	51
14	<i>Dermestes lanarius</i>	18	-	18
15	<i>Hymenoptera (furnici)</i>	33	20	53
16	<i>Armadiidium vulgare</i>	3	-	3
17	<i>Orthoptera (lăcuste)</i>	87	33	120
18	<i>Diptera (larve)</i>	17	-	17
19	<i>Ceutorhynchus crucifer</i>	3	-	3
20	<i>Blaps lutifera</i>	6	-	6
21	<i>Orthoptera (Gryllus)</i>	3	3	6
22	<i>Onthophagus taurus</i>	17	-	17
23	<i>Polydrosus sericeus</i> Schall	16	-	16
24	<i>Homoptera (cicade)</i>	31	5	36
25	<i>Carabus scabriusculus</i>	5	-	5
26	<i>Leipidoptera (adulti)</i>	9	3	12
27	<i>Amara aenea</i>	11	18	29
28	<i>Pseudophonus griseus</i>	8	38	46
29	<i>Coccinella conglobata</i>	21	7	28
30	<i>Anisodactylus binotatus</i>	7	18	25
31	<i>Calathus fuscipes</i> Gaeze	4	4	8
32	<i>Polydrosus amoeus</i>	12	-	12
33	<i>Carabus coriaceus</i> L	6	2	8
34	<i>Onthophagus ovatus</i>	20	-	20
35	<i>Homoptera (afide)</i>	8	-	8
36	<i>Collembola</i>	2	-	2
37	<i>Adalia bipunctata</i>	3	4	7
38	<i>Heteroptera (ploșnițe)</i>	10	-	10
39	<i>Pseudophonus rufipes</i>	-	86	86
40	<i>Carabus scabriusculus</i>	-	5	5
41	<i>Coccinella 7 punctata</i>	-	5	5
42	<i>Chilocorus longitarsis</i>	-	2	2
43	<i>Coleoptera (larve)</i>	-	2	2
44	<i>Podagrica fuscicornis</i>	-	3	3
45	<i>Stafilinide</i>	-	2	2
Total 51 species		695	399	1094



Table 3

## The structure of collected species in 2015

No.	Name of species	V. Adamachi Stationary		Number of samples
		Rivers	Stanley	
1.	<i>Dermestes lardarius</i>	41	21	62
2.	<i>Lepidoptera (larve)</i>	89	38	127
3.	<i>Lepidoptera (adulti)</i>	12	9	17
4.	<i>Coleoptera (larve)</i>	6	8	14
5.	<i>Hymenoptera (viespi)</i>	122	165	278
6.	<i>Cymindis vaporariorum</i>	14	-	14
7.	<i>Nothyophylus palustris</i>	11	-	11
8.	<i>Diptera (larve)</i>	51	-	51
9.	<i>Araneida</i>	144	151	259
10.	<i>Heteroptera (ploșnițe)</i>	5	-	5
11.	<i>Heteroptera (Pyrrhocoridae)</i>	19	27	46
12.	<i>Polydrosus sericeus</i> Schall.	3	85	88
13.	<i>Copris lunaris</i>	8	-	8
14.	<i>Calathus fuscipes</i> Goeze.	11	25	36
15.	<i>Anatis ocellata</i>	12	-	12
16.	<i>Chrysopa</i>	8	-	8
17.	<i>Gasteropode (melci)</i>	50	14	54
18.	<i>Adalia bipunctata</i>	20	3	23
19.	<i>Coccinella 7 punctata</i>	12	5	17
20.	<i>Anysocactylus binotatus</i> F.	30	10	40
21.	<i>Homoptera (afide)</i>	12	5	17
22.	<i>Galeruca pomanae</i>	31	15	46
23.	<i>Harpalus calceatus</i> Duft.	46	-	46
24.	<i>Collembola</i>	3	-	3
25.	<i>Chilopora longitarsus</i>	12	-	12
26.	<i>Orthoptera (Gryllus)</i>	29	26	55
27.	<i>Ceutorhynchus crucifer</i>	3	-	3
28.	<i>Tomoxia biguttata</i>	9	14	23
29.	<i>Ontophagus ovatus</i>	15	-	15
30.	<i>Diptera (adulti)</i>	9	11	20
31.	<i>Hymenoptera (furnici)</i>	48	38	86
32.	<i>Orthoptera (lăcuste)</i>	80	34	114
33.	<i>Galeruca tanaceti</i>	11	-	11
34.	<i>Homoptera (cicade)</i>	6	19	25
35.	<i>Hymenoptera (albina)</i>	33	23	56
36.	<i>Opatrum sabulosum</i> L.	6	-	6
37.	<i>Hister purpurascens</i>	15	-	15
38.	<i>Armadillidium vulgare</i>	33	-	33
39.	<i>Miriapode</i>	3	-	3
40.	<i>Blaps letifera</i>	11	-	11
41.	<i>Pseudophonus rufipes</i>	36	-	36
42.	<i>Carabus coriacus</i> L.	2	-	2
43.	<i>Amara aenea</i>	9	-	9
44.	<i>Coccinella 14 punctata</i>	9	-	9
45.	<i>Harpalus distinguendus</i> Duft.	3	3	6
46.	<i>Pterostichus niger</i> Schall.	-	5	5
47.	<i>Ceuthorrhynchus crucifer</i>	-	6	6
48.	<i>Pedinus femoralis</i>	-	2	2
49.	<i>Coccinella conglobata</i>	-	21	21



50.	<i>Chilocorus similis</i>	-	4	4
51.	<i>Stomodes gyriscollis</i>	-	3	3
<b>Total: 51 species</b>		<b>1146</b>	<b>790</b>	<b>1936</b>

The species with the largest number of collected samples were: *Hymenoptera* (wasps) (278), *Lepidoptera* (larvae) (127) *Orthoptera* (grasshoppers) (114) *Polydrosus sericeus* (88), *Hymenoptera* (ants) (86) *Dermestes lanarius* (62), *Hymenoptera* (bees) (56), *Orthoptera* (*Gryllus*) (55) *Gastropoda* (snails) (54), *Diptera* (larvae) (51) *Harpalus calceatus* Duft. and *Galeruca pomanae* (46) *Anysodactylus binotatus* (40), *Calathus fuscipes* and *Pseudophonus rufipes* (36), *Armadilidium vulgare* (33). Other species have been collected from 2 to 25 samples.

In the research period (2013-2015) were collected 3964 specimens belonging to nine systematic order. The highest number of specimens collected was recorded in 2015 with 2301 copies and lowest number samples was recorded in 2013 (415). In 2014 were collected 1266 samples. A rate of 5,81%, followed by the group of *Diptera* with a total 128 samples representing 3,12% and a total of 48 samples at a rate of 1,19% was registered isopod insect group.

In total, in the two orchards from Vasile Adamachi Iasi stationary and two varieties during the research were collected 4091 samples.

### CONCLUSIONS

Research was conducted in plum orchards in the period 2013-2015 in two plantations, namely: the *Rivers timpuriu* variety and the *Stanley* variety.

It was studied invertebrate fauna belonging to different classes and phyla, most collected species belonging to the class *Hexapoda* (insects) using soil traps type Barber;

In the period 2013-2015 at the two varieties situation on the number of collected samples separately for each order is as follows: *Coleoptera* with 38,37%, *Hymenoptera* with 22,21%, *Orthoptera* with 12,58%, *Homoptera* 4,37, *Arahnida* 8,34%, *Heteroptera* 3,3%, *Lepidoptera* 5,81%, *Diptera* 3,12% and isopods with 1,19%.

**Acknowledgements:** This paper was published under the frame of European Social Found, Human Resources Development Operational Programe 2007-2013, project no. POSDRU/159/1.5/S/132765.

### REFERENCES

1. Chatened du Gaetan, 1990 - *Guide des Coleopteres d'Europe*. Delacrois et Niestlé, Paris.
2. Minoiu N., Lefter Gh., 1987 - *Bolile și dăunătorii speciilor sâmburoase*, Editura Ceres, București;
3. Lăcătușu Matilda. Pistică C., 1980 - *Biologia dăunătorilor animalii*, Editura Didactică și Pedagogică, București;
4. Panin S., 1951 - *Determinatorul coleopterelor dăunătoare și folositoare din R.S.R.*, Ed. de Stat. , București;
5. Rogojanu V., Perju T., 1979 - *Determinator pentru recunoasterea daunatorilor plantelor cultivate*. Editura Ceres, Bucuresti.
6. Safavi M., 1968 - *Etude biologique et ecologique des Hymenopteres des œufs des punaises des cereales*. Entomophaga.

## THE PROSPECT OF MOLECULAR APPROACHES IN FUNDAMENTAL RESEARCH AND MANAGEMENT OF INSECTS

### ROLUL VOLATILELOR EMISE DE PLANTE ÎN „PREGĂTIRE” (PRIMING) LA ATACUL ERBIVORELOR

**TALMACIU M.<sup>1</sup>, ENEA C.I.<sup>2</sup>, BRUDEA V.<sup>3</sup>,  
TALMACIU Nela<sup>1</sup>, HEREA Monica<sup>1</sup>**  
e-mail: mtalmaciu@yahoo.fr

**Abstract.** *Jasmonates are the main signals in plant defence against herbivores, also being new weapons in plant defences. Oral secretions of insects are molecular patterns that trigger tritrophic relationships plant-pest-entomophagous, by releasing volatile. Priming plant by influence of volatile or internal signals produced by jasmonates, provides an increased resistance to pest attack. Only molecular approach in fundamental researches and in the management of the pests, assures knowledge of life phenomenology between plant and insect.*

**Key words:** *biotic stress, parasitoids, priming.*

**Rezumat.** *Jasmonații sunt principalele semnale în apărarea plantelor împotriva erbivorelor, de asemenea, fiind considerate arme noi în apărare de către plante. Secrețiile orale ale insectelor sunt modele moleculare care determină relațiile tritrofice plantă-dăunător-entomofagi, prin eliberarea volatilelor. Amorsarea de plante prin influența semnalelor volatile sau a celor interne produse de jasmonați, oferă o rezistență crescută la atacul dăunătorilor. Abordarea moleculară nu numai în cercetările fundamentale ci și în gestionarea dăunătorilor, asigură cunoașterea fenomenologiei vieții între plante și insecte.*

**Cuvinte cheie.** *Stress biotic, insect parasite, “priming”*

## INTRODUCTION

Plants have developed various strategies for defense against phytophagous and pathogens. Although some of these strategies are constituents, present all the time, others are induced phytophagous attack (Frost *et al.*, 2008). Defenses induced must be adaptive they are costly to implement and there is a variability spatial and temporal phytophagous and pathogens (the plants do not always experience the attack), defending differentiated against various enemies (the defense against one increases susceptibility to other).

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> Agricol Research and Development Station of Suceava, Romania

<sup>3</sup> Agricol Research and Development Station of Suceava, Romania

The offset this vulnerability at some plants has a first specific training in relation to environmental signals that indicate the likelihood increasing attack. In defense plant "preparation" (priming) is a physiological process that makes the plant to respond more quickly or aggressively to biotic or abiotic stress future. This training acquired is called "ready state" (primed state). Training may be initiated as a response to environmental signals that indicate the likelihood increase biotic stress, but may persist ready state due to residual effect, following initial exposure to stress. A classic example, a pest induce hypersensitive response, but induction is more effective if the plant has experienced a previous attack. The trees in the ready state persists in several seasons, the phenomenon is called "delayed-induced resistance", which can accelerate the induction of resistance in the presence attacks.

Any signal indicating the presence phytophagus can be a start for induction training. Most signals are usable plant volatiles induced phytophagus (HIPVS) and a subset of volatile organic compounds (VOCs) emitted in response. Finally internal signals are transmitted through the vascular system, or externally by plant volatiles. In case of injury in plants, information is transmitted through the phloem and signaling molecules xilem transported from the place attacked, systems, regions unchallenged. Systems can effectively transmit information signals from the attack site (wound) in regions vulnerable to attack. Signaling internal systems is performed Jasmonic acid (JA) and its conjugated forms.

In the following we present experiments that clarifies the role of plant volatiles in the preparation, plant-herbivore enabled tritrofica relations and parasite-dependent gene sets JA.

Engelbert H et al. (2007) mentions that regulates volatiles at least three genes responsible for serving platform octadecanoidă Jasmonic acid. Maize plants friendly, wounds exposed to volatiles from *Spodoptera littoralis*, regulate the expression of genes defense is enabled for new volatile emissions. Corn rootworm prepared by feeding the growth rate (RGR) was lower parasitoid wasp *Cotesia marginiventris* and was attracted by the abundance of volatiles released by plants activated. The preparation can influence the dynamics of mediation combinations defense phytophagus by direct or indirect defenses, via third trophic interactions.

## MATERIAL AND METHOD

The first successes in handling the emission of volatile insect repellent

It has been shown that it can manipulate the emission of volatile signals by maize roots attacked by worm *Diabrotica virgifera* western roots, attracting predatory nematode *Heterorhabditis megidis* (Hiltpold and Turlings, 2008). Corn and ancestral lines in Europe are very attractive at this nematode through the issuance of (E) - $\beta$ -caryophyllene, but American varieties have lost this signal, the nematode larvae control agent. To restore nematode attraction, corn lines were converted to emission constituent (E) - $\beta$ -caryophyllene, significantly reducing the attack.

Since the composition of volatile terpenoids dominate HIPV, were the first targets for manipulation is obvious that increasing the attractiveness of crops for

natural enemies of pests calls for targets specific compounds. Recent publications have shown that it is possible to manipulate genetically engineered production of attractants. Kappers *et al.* (2005) brought strawberries FaNES1 gene (a gene synthesis linalool / nerolidol) *Arabidopsis thaliana* to launch the (3S) - (E) -nerolidol attracting dust mite predators. In other studies, introduced a gene maize TPS10 (terpenes synthase) in A.th. for sesquiterpene volatile issue, launched the attack caterpillars (Schnee *et al.*, 2006). The transformed plants were more attractive to females parazitoizii, who used corn for finding host volatiles, but after the wasps were paired with host plant volatiles. A third notable achievement is the introduction synthase gene in *Arabidopsis* (E) -B-farnesene, issuing sesquiterpenic producing aphid control, having a role repellent and attract enemies (Pickett *et al.*, 2006).

The induction of protease inhibitors (PIs) by acid Jasmonic . This is a regulators of protein, plays an important role in defending plants with the target insect's digestive canal in digestion and absorption disturbance (Felton, 2005). In turn, herbivores induce post-translational modifications of the proteins, inhibiting their defensive functions by increasing their stability in the stomach. Microarray technology has revealed the genes encoding these proteins palette adjusted grazing.

The protease inhibitors (PIs) or proteins caused by arthropod attack (AIPS) can be adjusted to multiple hormonal signaling, including Jasmonic acid, salicylic acid and / or ethylene (Zhu *et al.*, 2008). Numerous research confirms jasmonates role in the induction of protease inhibitors to attack by herbivores or volatiles emitted by neighboring plants.

Farmer and Ryan (1990) specifies the communication between plants using Meja volatile induce synthesis of protease inhibitors in the leaves. These defensive responses, local or systemic were awarded before that date, ethylene volatile. Methyl jasmonatul applied to the surface of the tomato leaves induces the synthesis of protease inhibitors defense and neighboring plants.

Induction of protease inhibitors (PIs) in wounds, which harm the stomach digestive proteases insects, is the best example of defensive proteins whose synthesis is closely linked to acid signaling Jasmonic (Steppuhn and Baldwin, 2007). Other defensive proteins polyphenol oxidases are dependent Jasmonic acid, threonine deaminases, arginases and stored vegetative proteins. Many other proteins with activity against insects stability and proteases activity in the digestive system. All these components of metabolic should be seen not as separate entities, which operates in a synergistic defense system.

## RESULTS AND DISCUSSIONS

The negative performance of PIs are caused by overproduction phytophagus stomach digestive proteases insects, essential amino acids depleting and reduce growth. Reducing combinations increase may be due to toxic effects of anti-nutritive or antifeedant. For example, entering the wounds polifenoloxidaze act as PIs. Plant enzymes can exert anti-nutritional effects on insects by disrupting homeostasis phytophase amino acids in the digestive tract, but can be extended to other classes of plant-derived nutrients, including lipids, carbohydrases and vitamins.

The study of plant-insect interactions, especially in systems multitrofica offers the possibility to identify semiochemicals, can induce plant defense

responses. In this respect identified cis-jasmonats with great effects and persisting regulation of gene expression associated with plant defenses (Pickett, 2008). Grains have a wide range of induction levels of resistance caused by cis-jasmonate. Some varieties have proven resistant to aphids, thereby identifying a compound jasmonate induced by cis-6-methyl-5-hepten-2-one, the repellent effect at the same time, the attraction of parasitoids. In wheat, the increase of production of the hydroxamic acid (benzoxazinoidă) contribute to its resistance, and this is exploited by the breeding program.

Resistance is enabled induced chemical plants, pathogens and plants phytophagus and prepare for a better defense, joining in good agricultural practices. Tested resistance induction usefulness in combating biotic agents and increase plant productivity by treatment with BIONR, Oxyxom<sup>TM</sup>, or statements Reynoutria MessengerR sachaliensis- MilsanaR and BTH. Also in the induction of resistance to attack cis-jasmonatul bring phytophagus agricultural plant for resistance levels.

By presenting the summary of the signaling mechanisms and inducible defenses in plants at the molecular level is found jasmonats, as coordinators of these mechanisms play an important role in this direction and deepening this research from a fundamental perspective will solve practical issues of management. Undoubtedly that this research approach can not be achieved only in broad team of researchers at the Molecular genetic, biochemical, metabolic, etc. proteonomic will highlight the molecular mechanisms accuracy and ability to change in the interests of preserving the integrity of the plant. Knowledge of molecular phenomena can not be achieved without the use of proper laboratory techniques, improved in recent decades, supported by bioinformatics.

The observations at the macro level, biometric evaluations, weight can not satisfy us. Approach "omice" "genomice, proteonomice, lipidonomice, metabolomice, transcriptomice" are necessary for understanding biological phenomena. Using the genomes sequenced plant, *Arabidopsis thaliana* as tomatoes, tobacco etc, the mutant gene silencing or overexpressing may clarify the signaling chain and biotic stresses preparedness measures and inducing defense.

The researches present and future to establish signaling pathways of the biotic environment of receptor proteins stimulated the factors connecting the nodes of interference of various plant hormones, to decision makers in triggering transcripts (activation sets genes dependent on plant hormones, plant defense default) in the production of secondary metabolites involved in defense.

In preparing students during the license, but especially by masters must increase the proportion of subjects with levels of molecular genetic approaches, biochemical, environmental, etc., for a real knowledge of the phenomena of life and no surface.

## CONCLUSIONS

1. Jasmonates (JAS) are the main signals for adjusting resistance to phytophagous insects, representing new weapons and rapid responses against insect attack produced.

2. Plants can recognize provocateurs derived from feeding herbivores, which is associated molecular patterns phytophagous (HAMPs) compounds flags that can trigger defense. Chemical instigators of oral secretions of insects form plays an important role in the qualitative and quantitative responses of plants.

3. Biotic receptors signals by connecting factors mediating or regulating plant hormones biosynthesis genes and their expansion biosynthesis of secondary metabolites mediate transcription. These events include a reconfiguration transcriptomic with changes in gene transcription levels of defensive and growth; release of volatile organic compounds (VOCs) that functions as an indirect defense and disturbing accumulation of secondary metabolites herbivores or pests nicotine, trypsin and protease inhibitors (TPIs).

4. The secretions of herbivores triggers a plant-herbivore relationships tritrofica-entomophagus through the release of volatiles from the leaves injured, attracting entomofagii. Also, plants can recognize buildup triggering relationship tritrofică plant ponte-deposit ponte- entomophagus oofagi. Practical applications of management can be achieved in under tritrofica relations, introducing genes that increase the number of manufacturing certain volatile entomophagous etc.

5. The induction of resistance in the attacks include direct defense by the synthesis of toxic chemicals or tasteless and indirect defenses by producing volatile or honeydew nectar. Some chemical changes in the leaves injured acts as signs of injury in areas of the plant are not challenged or neighboring plants. Recognising these signals initiate preparation, which included changes at the molecular level, leading to the so-called state prepared leaves unchallenged. Leaves in ready state are able to respond more quickly and vigorously attack herbivores. Induction of resistance is more costly in energy terms, than plants pregătită.a state.

6. Jasmonates plays an important role in the production of protease inhibitors, resulting in overproduction of digestive proteases in the insect stomach, depleting essential amino acids and reduce growth. Reducing combinations increase may be due to toxic effects of anti-nutritive or antifeedant.

## REFERENCES

1. **Browse J., 2005** - *Jasmonate: an oxylipin signal with many roles in plants*. Vitam. Horm., 72: 431-456.
3. **Demole E., Lederer E., Mercier D., 1962** – *Isolement et détermination de la structure du jasmonate méthyle, constituant odorant caractéristique de l'essence de jasmin*. Helv. Chim. Acta, 45: 675-685.
4. **Engelberth J., Seidl Adams, Schütz J.C., Tumlinson J.H., 2007** - *Insect elicitors and exposure to green leafy volatiles differentially upregulate major octadecanoides and*

- transcript of 12-oxophytodienoic acid reductases in Zea mays*. Mol Plant Microbe Interact, 20: 707-716.
6. **Frost C.J., Mescher M.C., Carlson J.E., Consuelo M. De Moraes, 2008** - *Plant defense priming against herbivores: getting ready for a different battle*. Plant Physiology, vol. 146: 818-824.
7. **Hilker M., Stein Claudia, Schröder R., Varama M., Mumm R., 2005** - *Insect egg deposition induces defense responses in Pinus sylvestris: characterisation of the elicitor*. The Journal of experimental Biology, 208, p.
9. **Howe G.A., Jander G., 2008** - *Plant immunity to insect herbivores*. Annu Rev Plant Biol, 146: 801-803.
10. **Little D., Caroline Gouthier-Darimont, Brussow, Raymond Ph., 2007** - *Oviposition-induced changes in Arabidopsis genome expression*. Joint International Workshop on: PR –proteins and induced resistance against pathogens and insect, The Netherlands.
12. **Maffei M.E., Mithöfer A., Boland W., 2007**- *Before gene expression early events in plant-insect interaction*. Trends Plant Sci, 12: 310-316.
13. **Mithöfer A., Boland W., 2008** - *Recognition of herbivory-associated molecular pattern*. Plant Physiology, vol. 146: 825-831.
14. **Pickett J.A., 2008**. *Constitutive and induced plant-plant signaling: hypotheses and practical developments*. State College, Pennsylvania, Symposium, International Society of Chemical Ecology, USA, Summary.
15. **Schmelz E.A., Engelberth J., Alborn H.T., Tumlinson III J.H., Teal P.E.A., 2009** - *Phytohormone-based activity mapping of insect herbivore-produced elicitors*. PNAS, vol. 106: 653-657.
16. **Steppuhn A., Baldwin I.T., 2007** - *Resistance management in a native plant: nicotine prevents herbivores from compensating for plant protease inhibitors*. Ecol. Lett, 10: 499-511.
17. **Vick B.A., Zimmerman D.C., 1984** - *Biosynthesis of jasmonic acid by several plants species*. Plant Physiol., 75: 458-461.
18. **Wu J., Hettenhausen C., Schuman M.C., Baldwin I.T., 2008** - *A comparison of two Nicotiana attenuata accessions reveals large differences in signaling induced by oral secretions of the specialist herbivore Manduca sexta*. Plant Physiol, 146: 927-939.
19. **Zheng S.J., Dicke M., 2008** - *Ecological genomics of plant-insect interactions: from gene to community*. Plant Physiology, vol. 146: 812-817.



## ANTIOXIDANTS BUCKTHORN OIL, ADJUVANT IN MILD COGNITIVE DYSFUNCTION THERAPY

### ANTIOXIDANȚII ULEIULUI DE CĂTINA, ADJUVANȚI ÎN TERAPIA DISFUNȚIEI COGNITIVE MEDII ȘI UȘOARE

**BODESCU Maria-Mădălina<sup>1</sup>, BODESCU Oana Maria<sup>2</sup>,  
TĂLMACIU M.<sup>2</sup>, HEREA Monica<sup>2</sup>**  
e-mail: bodescumm@yahoo.com

**Abstract:** Buckthorn oil contains many of the elements essential for health: vitamin C, vitamin E, B vitamins, vitamin A, calcium, magnesium and iron. All these precious ingredients are added and essential fatty acids omega-3, omega-6, omega-7 and omega-9. Buckthorn is a natural source of antioxidants and therefore its effectiveness to protect against oxidative stress has been shown by some researchers. The antioxidant properties of seabuckthorn were determined in vitro by Geetha, Sai Ram Singh Ilavazhagan, and Sawhney (2012). The research included 31 patients with mild cognitive impairment and medium recruit and private clinic of patients with psychiatric profile during 1 june -30 august 2014 following two lots First lot included 20 patients receiving specific medication and capsules antidemential seabuckthorn oil (900 mg) and lot II (control group) included 11 patients being treated with medication specific antidemential. I noticed a significant improvement in oxidative status by modifying SOD ( $p = 0.001$ ) and cognitive status by modifying the MMSE ( $p \leq 0.001$ ) reduced. Antioxidants are an important adjunct in the treatment of mild cognitive impairment and media.

**Key words:** seabuckthorn antioxidants, oxidative stress, cognitive dysfunction, Buckthorn oil, antioxidants, vitamin C, vitamin E,

**Rezumat:** Uleiul de cătină conține multe dintre elementele esențiale pentru sănătate: vitamina C, vitamina E, vitamine din complexul B, vitamina A, calciu, magneziu și fier. La toate aceste ingrediente prețioase sunt adăugați și acizi grași esențiali omega-3, omega-6, omega-7 și omega 9. Cătina este o sursă naturală de antioxidanți și, prin urmare, eficacitatea sa a proteja împotriva stresului oxidativ a fost demonstrat de către unii cercetători. Proprietățile antioxidante ale catinii au fost determinate controlat de către Geetha, Sai Ram Singh Ilavazhagan, și Sawhney. Cercetarea a inclus 31 de pacienți cu insuficiența renală ușoară cognitivă și recrută într-o clinică privată în intervalul 01 iun - 30 august 2014 în două loturi, primul lot a inclus 20 de pacienți care au primit medicație specifică și capsule de ulei de cătină

---

<sup>1</sup> "Grigore T. Popa" University of Medicine and Pharmacy of Iasi, Romania

<sup>2</sup> University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

*antidemență (900 mg) și lotul II (grupul de control) a inclus 11 de pacienți tratați cu medicamente specifice antidemență. Am observat o îmbunătățire semnificativă a stării oxidative prin modificarea SOD ( $p = 0,001$ ) și statutul de cognitive prin modificarea MMSE ( $p \leq 0.001$ ) ce s-a redus. Antioxidanții sunt un adjuvant important în tratamentul de insuficiența renală ușoară cognitivă și anti demență.*

**Cuvinte cheie:** *catina, stres oxidativ, disfuncții cognitive, antioxidanți, vitamina C, vitamina E.*

## INTRODUCTION

Sea buckthorn is a berry bush introduced into the culture of Romania. In the last 40 years has been shown that this fruit, harvesting difficult and highly perishable fresh, contains a number of valuable substances for human metabolism and an important role in regulating biological characteristics and chemical composition (Cakir, 2004).

Certain researches for this fruit plant have been made lately to present a special interest, pursuing the development of hybrid technologies for separation, concentration and purification of sea buckthorn extract, in order to obtain concentrated active ingredients with applications in food, pharmaceutical and cosmetic (Sa, 2007).

It has special activity in chronic hepatitis, urogenital disorders, neurological and psychiatric disorders. In the specific literature highlights the exceptional work and the role anemic stagnation and regression of various eye diseases (hemeralopia, presbyopia, cheratomalacie, myopia, astigmatism, hyperopia, glaucoma, cataracts) due to its high in beta-carotene (Folstein M.F *et al.*, 1975 Centenaro *et al.*, 1977).

For external use, use in dermatological disorders (psoriasis), atopic disorders and inflammatory ENT component in the topical treatment of eczema, burns, thermal and chemical frostbite, allergodermia, slow wound healing. It is the only known natural protective activity against solar radiation or other nature (Ecotech, 2010). Concentrate active principles are used with spectacular results in geriatrics, treat depression, Parkinson's disease, tumors, adenomas and leukemia (Ulian, 2009).

## MATERIAL AND METHOD

The research included 31 patients with mild cognitive impairment patients recruited from the profile private psychiatric clinic during 1<sup>st</sup> June -30<sup>th</sup> August 2014 following two groups. Group I included 20 patients receiving medication specifically antidemential and oil capsules Seabuckthorn (900 mg) and group II (control group) included 11 patients being treated with medication specific antidemential.

Evaluation of clinical and laboratory status and oxidative done for each patient before inclusion in the study and after 6 months.

The evaluations were carried out by medical history and laboratory evaluation which included: determining blood pressure, heart rate, dosage MMSE and SOD. Subject Inclusion criteria were:

- Disease known retrieval easy and moderate cognitive impairment (MMSE 25-20);

- the patients consent to participate in the study.

Exclusion criteria were represented

- Retrieval disease absence;

- refuses from patients to participate in the study.

MMSE test is the most common tool used to assess cognitive function. Folstein MMSE was developed in 1975 to assess the mental state of psychiatric patients and to differentiate the origin of their organic or functional pathology.

## RESULTS AND DISCUSSIONS

The results of the comparative analysis of cognitive status and oxidative status between the two groups at the beginning of the study have revealed that there are no differences statistically significant  $p > 0.05$  (Table 1).

Evaluation of cognitive and oxidative status in patients in group I

Table1

**Comparative analysis of cognitive status and oxidative status between the two groups**

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
MMSE	Equal variances assumed	,14	,70	-,072	41	,943	-,020	,27	-,58	,54
	Equal variances not assumed			,069	18,57	,945	-,020	,29	-,62	,58
SOD	Equal variances assumed	,09	,76	-,449	41	,655	-2,34	5,20	-12,86	8,17
	Equal variances not assumed			-,447	19,86	,660	-2,34	5,23	-13,26	8,58

Mean MMSE initial enrollment in the study was  $22.55 \pm 0.79$ , the value being 13.9% higher than the average GI registered at 6 months,  $25.6 \pm 0.12$  (Table

2), the difference between the two areas being highly significant (95% CI 0.11 to 0.70;  $p = 0.009$ ) (Table 3).

Table 2

Mean of MMSE and SOD in patients in group I

		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	MMSE before	22,5,5	31	,79	,14
	MMSE after	25,6	31	,69	,12
Pair 2	SOD before	359,74	31	15,27	2,74
	SOD after	404,66	31	13,59	2,44

a. LOT = LOT I

Table 3

Comparative analysis of cognitive status and oxidative status patients in group I before and after 6 months

		Paired Differences							
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference		t	df	Sig. (2-tailed)
					Lower	Upper			
Pair 1	MMSE before - MMSE after	,40	,81	,14	,11	,70	2,80	30	,009
Pair 2	SOD before - SOD after	12,77	21,81	3,91	4,77	20,77	3,26	30	,003

The difference between the two environments is highly significant (95% CI 4.77 to 20.70;  $p = 0.003$ ) (Table 4)

Table 4

Comparative analysis of cognitive status and oxidative status patients in group II before and after 6 months

		Paired Differences							
		Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference		t	df	Sig. (2-tailed)
					Lower	Upper			
Pair 1	MMSE before - MMSE after	-,09	1,15	,33	-,82	,640	-,276	11	,78

Pair 2	SOD before - SOD after	5,25	20,02	5,78	-7,47	17,97	,908	11	,38
--------	------------------------	------	-------	------	-------	-------	------	----	-----

a. LOT = LOT II

Evaluation of cognitive status and oxidative patients in group II

Mean MMSE initial enrollment in the study was  $22.47 \pm 0.87$ , the value being 5.01% higher than the average MMSE recorded 6 months,  $23, 5 \pm 0.96$ , the difference between two areas not significant (95% CI 0.82 to 0.64;  $p = 0.078$ )

Mean SOD initial enrollment in the study was  $362.08 \pm 15.44$  of value being 5.09% higher than the average recorded 6 months sod of  $380.18 \pm 16.28$ , the difference between two areas not significant (95% CI -7.44 to 20,717,970;  $p = 0.038$ ).

The results of the comparative analysis of cognitive status and oxidative status between the two groups at 6 months from the beginning of the study have revealed that there are differences statistically significant  $p < 0.05$  both when the mean MMSE scores, and the environment SOD (Table5).

Table5

**Comparative analysis of cognitive status and oxidative status between the two groups after 6 month**

Levene's Test for Equality of Variances			t-test for Equality of Means							
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
MMSE	Equal variances assumed	,14	,00	-,072	41	,003	-,020	,27	-,58	,54
	Equal variances not assumed			-,069	18,57	,005	-,020	,29	-,62	,58
SOD	Equal variances assumed	,09	,01	-,449	41	,005	2,34	5,20	-12,86	8,17
	Equal variances not assumed			-,447	19,86	,005	2,34	5,23	-13,26	8,58

From studies made by various authors, return in the legs buckthorn juice is about 75%. It is worth noting that the seeds and peel results after extracting the juice can be dried to 10-12% moisture. These after-grinding can be subjected to oil extraction. The maximum amount of oil found in fruit peel and seeds.

It is to note that only the amount of vitamin C in sea buckthorn is 8 times higher than in peaches, 20 times higher than in the big tree, 80 times higher than in tomatoes, and 200 times higher than the amount of vitamin C contained in grapes. Therefore buckthorn is called the treasure of vitamin C.

## CONCLUSIONS

I noticed a significant improvement in oxidative status by modifying SOD ( $p = 0.001$ ) and cognitive status by modifying the MMSE ( $p \leq 0.001$ ) reduced the studio group than the control group. Antioxds have an important role in the treatment of mild cognitive impairment.

Seabuckthorn is a natural reservoir of antioxidants and therefore its effectiveness to protect against oxidative stress has been shown by some researchers. The antioxidant properties of seabuckthorn and immunity are definite.

## REFERENCES

1. **Cakir Ahmet, 2004**, -Essential oil and fatty acid composition of the fruits of *Hippophae rhamnoides* L. and *Myrtus communis* L. from Turkey. Bioch.System and Ecology 32: 809–816.
2. **Centenaro G., Capietti G., Pizzocaro F., Marchesini A., 1977** -. The fruit of Sea buckthorn *Hippophae rhamnoides* as a source of vitamin C. Atti Soc Ital Sci Nat Mus. Civ. Stor. Nat. Milano, 118: 371-378.
3. **Folstein M.F., Folstein S.E., McHugh P.R., 1975** - Mini-Mental State: a practical method for grading the cognitive state of patients for the clinician. J. Psychiatr Res. 12: 189–198
4. **Sa H., 2007** - Catina farmacia completă din grădină, Woodhead Publishing, no 11.
5. **Teng E.L., Chui H.C., 1987** - The Modified Mini-Mental State (3MS) Examination. J. Clin. Psychiatry.;48: 314-318.
6. **Ulian C., 2009** - Catina, cel mai puternic antioxidant, <http://www.aplr-doctorat.blogspot.com>
- 7.\*\*\* **ECOTECH (2010). Compoziția chimică a catinei**. International Conference: Cooperation to Reduce Emissions. Analysis. Questions. Best Technologies and Practices, Almaty, Kazakhstan, 03 november 2010.

## REPRESENTATIONS OF THE GARDEN OF EDEN IN CHRISTIAN SACRED ARCHITECTURE

### REPREZENTĂRI ALE GRĂDINII RAIULUI ÎN ARHITECTURA SPAȚIULUI SACRU CREȘTIN

**CEHAN Agata Mihaela<sup>1</sup>, GHEORGHIȚĂ Constanța Carmina<sup>1</sup>**  
e-mail: agata.cehan@yahoo.com

**Abstract.** According to the Bible, the path of humanity began in a garden, an enclosed precinct, a hideaway, a shelter, simultaneously a place of genesis and death. In the architecture of the Christian churches, Heaven, or Paradise, is often represented as a garden with lush and spontaneous vegetation within which dominates the Tree of Knowledge. The Garden of Eden is thus a place of the reconstruction of the man, the centre of the soul. This study aims to analyse the visual representations of the Garden of Eden in the decorative arts of the Christian worship space.

**Key words:** symbol, Eden, garden, Paradise, cross-tree, sacred-space, sacred architecture

**Rezumat.** Potrivit Bibliei, drumul omenirii a început într-o grădină, spațiu împrejmuit, refugiu, adăpost, loc al genezei și al morții în același timp. În arhitectura spațiilor sacre creștine, Raiul sau Paradisul este reprezentat de cele mai multe ori ca o grădină cu vegetație luxuriantă și spontană în mijlocul căreia tronează Pomul Cunoașterii. Grădina Raiului este locul de restaurare a ființei, centrul sufletului. Acest studiu își propune analiza reprezentărilor vizuale ale Grădinii Raiului în plastica decorativă a spațiului de cult creștin.

**Cuvinte cheie:** simbol, Eden, paradis, copacul-cruce, spațiu sacru, arhitectură sacră creștină

## INTRODUCTION

According to the Bible, the path of humankind began in a garden, an enclosed precinct, a hideaway, a shelter, a place of genesis and death. In the architecture of the Christian churches, Heaven, or Paradise, is often represented as a garden with lush and spontaneous vegetation within which dominates the Tree of Knowledge. "Now the Lord God had planted a garden in the east, in Eden; and there he put the man he had formed. The Lord God made all kinds of trees grow out of the ground—trees that were pleasing to the eye and good for food. In the middle of the garden were the tree of life and the tree of the knowledge of good and evil. A river watering the garden flowed from Eden; from there it was separated into four headwaters." (Gen. 2: 8-10)

In the book of *Genesis* Eden is described as being a beautiful garden, enclosed with circular walls, with well-doer waters and fruit-bearing trees. From the center located the Paradise's well spring the four rivers of the Earth: Pison, Gihon,

---

<sup>1</sup>„Gh. Asachi” Technical University of Iași, Romania



Hiddekel and Euphrates. In this garden, the Lord planted the tree of knowledge of good and evil. After committing the original sin, this sacred space will be banned for man and will be protected by a cherubim with a flaming sword.

The Christian sacred space has an architectural component at the same time with a symbolic one. The symbol implies representation, being most often a sign or an image associated with a few words or abbreviations of the item that it represents. Symbols have always helped in finding or identifying certain values, beliefs, ideas or creed. (Cehan and Gheorghiu, 2014)

## MATERIAL AND METHOD

Eden or Garden of Eden are frequently represented in the iconography of Christian worship spaces. In general, we call icon "any visual representation of an object crafted using various forms of art (drawing, painting, sculpture, etc.) [...] Thus within the broad concept of the Icon, understood as a visual representation of something, are included also statues, sculptures, engravings, reliefs, etc." (Braniște, 1993)

This paper identifies and interprets different visual representations of the Garden of Eden, in the decorative art of Christian worship space. As research methods were used: theoretical and photographic documentation, observation and case study.

## RESULTS AND DISCUSSIONS

The Holy Fathers identified in their teaching about divine reasons the whole world as a symbol "a translucent circumstance of divine transcendence" (Streza, 2010). Each symbol is a microcosm, an entire world, his perception being subjective, everyone sees and understands what can and wants to see and understand. "...the whole intelligible world presents itself mysteriously imagined in the symbolic faces of the sensible world, for those who have eyes to see; and the entire sensible world, if studied with a mind fond of knowledge, in its very reasons being included the intelligible world." (Maxim Mărturisitorul) The open space, without limitation, generates insecurity, fear, chaos. "...a well-defined space [...] is made up of three parts: *THE WORLD AXIS, THE ORDERED SPACE and THE BORDER. Beyond BORDER there is CHAOS*" (Biciușcă, 2008). Once its limits are perceived, it begins to be perceived and understood. This is why the Paradise is imagined as an enclosed garden in which the primary essential elements are ordered around the *Tree of Knowledge*.

The association between *World Tree* and *the Tree of Life*, as a divine manifestation, is found in the Christian tradition, Tree of Life having as sap the heavenly dew, while its fruit transmit a touch of immortality. In the Christian iconography appears the symbol of the cross with leaves, or the Tree-Cross, which stands on top of a mountain in the center of the world, resuming the image of the Cosmic Tree or of the world.

The cross is the most common sacred symbol, representing one of the fundamental reference points of cultural and religious history of the world, believed to be a geometric variant of the *World Tree. World axis, ladder or tree*, all these meanings conveys a symbolism of ascension in the *center of the world*, a bond

between the layers of universal existence. The cross symbolizes the *Center* and the directions or lines of power departing from this center. *Sign of signs*, the cross is a model of cosmos totality, but also of man or anthropomorphic deity. Its most elevated meanings are related with the universal history of Christianity, the cross being assimilated with the *Cosmic Tree*, in its quality as *Center of the World* symbol. (Evseev, 2001)

Transitional space between the faithful world and the church, the precincts of the ecclesiastical spaces seem to be a reinterpretation of what the Holy Scripture describes as "garden eastward in Eden" (Genesis 2:8). The Church, centrally located, seems to subordinate the whole precinct's composition, just as "the tree of life also in the midst of the garden, and the tree of knowledge of good and evil." (Genesis 2:9). Extrapolating, we can say that the metaphor of the "tree of knowledge" is represented on several levels. The central vertical axis of a church, defined by the nave and the dome on the central tower, support of Jesus Pantocrator, represents the image of an *Axis Mundi*, the link between the realm of death (horizontal plane, the floor of the church), the terrestrial world (space defined by the church walls) and the realm of promises (everything that is above). In the same way we may consider the church itself as an object, as a representation at the scale of the whole ensemble of this 'tree of knowledge', and further, even at a larger scale, the whole ecclesiastic ensemble as an *Axis Mundi* for the landscape. (Gheorghiță and Grigorovschi, 2013)



**Fig. 1** -The Garden of Eden, German author unknown

([https://en.wikipedia.org/wiki/Hortus\\_conclusus#/media/File:Meister\\_des\\_Frankfurter\\_Paradiesg%C3%A4rtleins\\_001.jpg](https://en.wikipedia.org/wiki/Hortus_conclusus#/media/File:Meister_des_Frankfurter_Paradiesg%C3%A4rtleins_001.jpg))

In medieval iconography, in the Eden Garden, enclosed within walls, *Hortus Conclusus* (Fig. 1), Virgin Mary is represented with the child and the angels. Hence derive the closed gardens from the enclosure of the monasteries. The tree in the upper left corner of the painting is a cherry tree which refers to the blood of Jesus shed on cross; the may-lilies from the close-up and the lilies from the middle right and

the white flowers symbolize the innocence of the Virgin Mary, the iris in the background, a flower of the Virgin, appears in scenes of the Annunciation; roses (top right) symbolizes Mary "the rose without thorns", not touched by original sin; strawberries (table below) represent the food of happiness; peony (below, middle) is the symbol of spiritual wealth and honor.

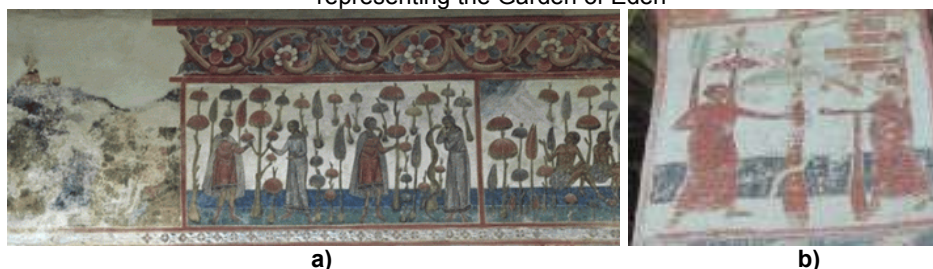
According with the painting orthodox Erminia, the representations of Eden are present in the icons illustrating the *Creation of Man*, *The Creation of Adam*, *Creation of Eve*, *Adam and Eve Trampling Commandment*, *Expulsion from Paradise* and *Lamentations of Adam and Eve* (Dionisie din Furna, 2000). (Fig. 2, 3, 4) A different representation of the Garden of Eden is found on the east wall of the narthex, in the lower register left of the icon of *The Last Judgment*. (Fig. 4) Heaven is depicted as a garden surrounded by walls of "crystal, gold and gems all around", where it can be found „*Distinguish trees, beautifully decorated and flying birds of many kinds*”, in the center of which the Mother of God is sitting in a „gold stall” surrounded by two angels dressed like deacons. (Dionisie din Furna, 2000)

In the Garden of Eden, the first humans, Adam and Eve, are allowed to taste all fruits and plants, but the fruit of the *Tree of Life* and the *Tree of the Knowledge of Good and Evil*. According to some interpretations, the forbidden fruit is represented as an apple. From a symbolic perspective, the apple has as a starting point its own inside with the five alveoli arranged in the form of five-pointed star, which is why the initiated ones have considered it the fruit of knowledge and freedom (Chevalier and Gheerbrant, 1994). According to its nearly spherical shape, it would generally signify the mundane desires or the giving in to these desires. The Prohibition made by God would put the man on guard against the predominance of those desires that drag him toward material life opposite to the spiritual life, which is in fact the meaning of evolution. The apple would be the symbol of this knowledge and laying it in front of a need: to choose (Chevalier and Gheerbrant, 1994)

The apple, tree and fruit, occupies one of the most important places in the plant code of symbols of different nations. It's a symbol of life, of youth and love, but also of sin, of carnal temptations, the ambivalence of the symbol being due to the fact that, before becoming a feminine symbol, the apple was one of the essentials emblems of the Great God of Earth. Because the main zoomorphic aspect of the God of Earth is the snake, the biblical myth associates the apple with the snake that will tempt Eve. This is how the apple became one of the symbols of the sin. The biblical text does not mention exactly which was the fruit of "knowledge and sin", some older painters drawing in the middle of Heaven a fig tree, or a peach, or a quince, as a fruit of temptation. Another theory holds that the constant association of apple with the forbidden fruit has been made because of the fact that in Latin, *malus* has a double meaning: *apple*, but also *bad*. The third negative symbolic interpretation of the apple as tree and fruit is based on the fact that early Christians were suspicious and granting negative connotations to the pagan objects and symbols, including apple which holds one of the most privileged places, because of its red color, mixed with the green and the yellow, being a symbol of life and youth (Evseev, 2001).



**Fig. 2** - Icons on the north wall of the monastery Sucevița representing the Garden of Eden



**Fig. 3** - Representations of the Garden of Eden  
a) Voroneț Monastery

([https://fotopoeziemuzica.files.wordpress.com/2008/04/imag2010\\_jpgvoronet-fatada-nordica-adam-si-eva-in-rai-scene-din-viata-maicii-domnului.gif?w=468](https://fotopoeziemuzica.files.wordpress.com/2008/04/imag2010_jpgvoronet-fatada-nordica-adam-si-eva-in-rai-scene-din-viata-maicii-domnului.gif?w=468))

b) Moldovița Monastery

([https://commons.wikimedia.org/wiki/Category:Moldovita\\_murals#/media/File:Moldovita\\_murals\\_2010\\_09.jpg](https://commons.wikimedia.org/wiki/Category:Moldovita_murals#/media/File:Moldovita_murals_2010_09.jpg))



**Fig. 4** - Last Judgement, Voronet, overview and representation of the Garden of Eden  
(<https://rasvancristian.files.wordpress.com/2008/10/voronet-1-imagine-mare.jpg>)

The fig tree is present in the Garden of Eden. According to the Old Testament, after the Fall, the fig leaf becomes the first piece of clothing of Adam, symbol of the acknowledged male sexuality: *"Then the eyes of both of them were opened, and they realized they were naked; so they sewed fig leaves together and made coverings for themselves."* (Gen.3: 7) The fig tree is a symbol of marriage, fertility, sexual attraction, feminine principle, spiritual enlightenment and truth (Evseev, 2001).

The peach tree, also known as "Persian apple" is a symbol of spring and of gentleness. In China and Japan, it represents an ancient symbol of the immortality, being associated with the Tree of Life (Evseev, 2001).

## CONCLUSIONS

The world created and represented in the Christian iconography painting is entirely oriented toward eternity, toward the beyond. By simplicity, schematic and linear shaping of the portraits, the characters give an impression of remoteness from the real world. Unlike the western religious art, more realistic, oriented in the precise reproduction of the man and nature, in the Orthodox iconography both the nature, the man and the animal world are represented not in their „natural” as in the "spiritual" one. So the icon is not an exact copy of reality, it is neither portrait nor photo. The icon is the image, the prototype and the symbol of the divine creation, the real world being represented as a transfigured cosmos.

In its various representations of Christian iconography, Garden of Eden appears as a space whose purpose is recreation and spiritual ascent. It represents a sacred space, a protected place, enclosed by symbolic walls or fences, and it is guarded at times by higher beings, as the angel at the gates of the Earthly paradise. Garden of Eden symbolizes the condition of beatitude in which the human ancestors lived before the Fall. It is a state of perfection, to which the man is desperately trying to return, as it shown by the numerous journeys in the search of the Lost Paradise.

## REFERENCES

1. **Biciușcă Florin, 2008** - *Geometria simbolică*, Colecția Spații Imaginate, Ed. Paideia, București, pp. 9-10
2. **Braniște E., 1993** – *Liturgica generală*, ed. Institutului Biblic și de Misiune al Bisericii ortodoxe Române, București, 415 p.
3. **Cehan A. M., Gheorghiță C.C., 2014** – *Simbolul viței de vie în arhitectura spațiului sacru*, Lucrări Științifice USAMV Iași , seria Horticultură, vol. 57, nr. 1, Iași, Ed. Ion Ionescu de la Brad, pp. 265-270
4. **Chevalier J., Gheerbrant A., 1994** - *Dicționar de simboluri*, vol. II, București, Editura Artemis 284 p.
5. **Dionisie din Furna, 2000** – *Erminia picturii bizantine*, Ed. Sophia, București, pp. 69-70; 218;
6. **Evseev I., 2001** - *Dicționar de simboluri și arhetipuri culturale*, Ed. Amarcord, Timișoara, p. 47-48; 112-113; 176; 149;
7. **Gheorghiță Constanța Carmina, Grigorovschi Mircea, 2013** - *The Importance of Mineral-Vegetal Ratio in Structuring the Landscaped Space Of Some Iași Churches* în European Journal of Science and Theology: 9(4).
8. **Sfântul Maxim Mărturisitorul** - *Mystagogia* 21, pp 912, 697 A
9. **Streza L., 2010** - *Simbolul liturgic în arhitectura bisericii, locaș de închinare*, Revista Teologica, nr.2, pp.113-125.



## WAYS OF IMPLEMENTING IN THE LANDSCAPING EDUCATION SOME REHABILITATION STUDIES FOR OUTDOOR SPACES OF UNIVERSITIES

### MODURI DE APLICARE ÎN ÎNVĂȚĂMÂNTUL PEISAGISTIC A UNOR STUDII DE REABILITARE A SPAȚIILOR EXTERIOARE AFERENTE UNIVERSITĂȚILOR

**DASCĂLU Doina Mira<sup>1</sup>,**  
e-mail: doinamira@yahoo.com

**Abstract.** Nowadays, there are not enough theoretical studies regarding the conception of a specific landscape design for universities outdoor spaces. In this context, we have initiated a thematic research on dysfunctions, needs and ways of landscaping recovery/rehabilitation and development for universities outside spaces. In order to correlate research with specialized landscaping education, we tried to apply the results of these studies in the education of the students from Landscape Specialization within the Horticulture Faculty of USAMV Iasi. The paper analyse selected results of landscaping solutions for virtual spaces and the ways these were applied to real university outdoor spaces of Iași town. We aimed to create multifunctional concepts in order that these solutions can be applicable - sequentially or in whole composition design - to other similar cases

**Key words:** landscaping, education, revitalization, university spaces

**Rezumat.** În prezent, există un deficit de studii teoretice privind conceperea unui design peisagistic specific pentru spațiile exterioare ale universităților. În acest context, am inițiat o cercetare tematică privind disfuncțiile, nevoile și modalitățile de recuperare/reabilitare și dezvoltare pentru spațiile exterioare aferente universităților. În scopul de a corela cercetarea cu învățământul de specialitate peisagistică, am încercat să aplicăm rezultatele acestor studii în educația studenților de la Facultatea de Horticultură - Specializarea Peisagistică - USAMV Iași. Lucrarea analizează câteva modalități în care soluții virtuale de amenajare au fost aplicate în cazul real al spațiilor unei universități ieșene. Conceptele propunerilor selectate sunt multifuncționale, pentru ca aceste soluții să fie aplicabile - secvențial sau complex - în alte cazuri similare.

**Cuvinte cheie:** peisagistică, educație, revitalizare, incinte universitare

## INTRODUCTION

The economic crisis affects higher education institutions in Romania by the lack of funds for rehabilitation and modernization, therefore outdoor spaces belonging to these institutions are mostly degraded in terms of landscape.

Abroad, many exterior spaces of universities are multifunctional designed to provide students propitious conditions for study and relaxation, but also to provide a large variety of activities in order to save funds. For example, one of

---

<sup>1</sup>University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

the most interesting multifunctional landscaping example is the concept of Turenscape Group for Shenyang Jianzhu University Campus in China. The Turenscape designers faced with important financial difficulties and the limitations of an agricultural site conditions. Finally, the solution correlated the relaxation and education of the students with a productive landscape, where the farming processes became part of educative and relaxation process, for all interested (Krauel, 2006).

Despite these landscape design projects for exterior academical spaces, there are few theoretical studies regarding the analysis and conception of specific landscape design for universities outdoor spaces. In this context, we have initiated a research on thematic disfunctions, needs and ways of landscaping recovery and development for outside spaces of universities.

In order to correlate research with specialized landscaping education, we tried to apply the results of these studies in the education of the students from Landscape Specialization within the Horticulture Faculty of USAMV Iasi. The paper analyse selected results of landscaping solutions for virtual spaces and also for existing spaces of universities outdoor spaces. We aimed to create multifunctional concepts in order that these solutions can be applicable - sequentially or in whole composition design - to other similar cases.

## **MATERIAL AND METHOD**

The study started from identifying issues and needs, raised by the loss of many outdoor spaces of Romanian universities, due mainly to their degradation and disuse. In the same time, the necessary background support was created by documentary studies about some specific landscape design for universities outdoor spaces, supplemented with comparative analysis. We applied some of these studies in the landscape architecture education, in order to develop students design creativity. Students were directly concerned, since these type of arrangement were for their benefit, were intended to create a high degree of quality for their academic life.

Starting from these premises, we have conceived several design themes whose purpose was to develop students creative personality. These themes are developed in virtual spaces for the students of second and third academic years, as useful exercises to improve their landscaping skills. For the fourth year and also for the license projects we created themes for existing situations. One of the existing cases selected for this paper was the rehabilitation of three exterior courtyards of "Al. I. Cuza" University of Iasi. The setting of needs and existing dysfunction, both for virtual and existing spaces, led finally to the fulfilment of few solving concepts. In order to prove the range of planning types diversity in landscaping design, this study presents selected concepts for development and revitalization.

## **RESULTS AND DISCUSSIONS**

The themes developed in virtual spaces was created for the students of second and third academic years after preliminary studies. In order to experience how the research ideas can be applied to improve landscape design education in a specific area, we have chosen as basic theme of landscape design "Landscape



design of academic interior yard”, having square shape of 18 m x 18 m. For the beginning of the design process it was necessary a synthetic presentation of the research ideas for the students (Fig. 1).



**Fig. 1** - Presentation of the research ideas for the students - photo by Dascălu

Then we started a general survey, conducted among students, staff and the auxiliary staff, setting what would be their main needs for leisure and teaching activities in outdoor spaces of the university. Together with students, we analyzed the dynamic of needs and leisure, personal items that may influence the design concept. The most important has been found the cultural and aesthetic needs.

Based on collected options, we established together with students, which would be the functionalities possible to be converted into a spatial concept. The selected functions were the relaxation and leisure, educational and informative functions, social-cultural functions, aesthetic functions, sanogene functions. Further, the multifunctionality of all the elements has been found as the best solution possible to be applied for the limited area of the virtual court.

The next step was the correlation of functions with conceptual aesthetic forms (Petrovici and Nica, 2012). The games of geometric shapes strongly distinguished the solutions. Each student has chosen some games in agreement with his personality, applying it in the virtual space of the yard. To illustrate the results, we present below some of the students selected solutions.

Using successive design surveillance and individual improvements of each solution, we have developed the students courage to use aesthetic forms in order to express landscape functions (Jurov, 2006). Were used games of circles and squared forms combinations (Fig. 2), circles and organic/natural forms (Fig. 3), polygonal and angles games, angles and curves combinations and other combinations.

Further, we have expanded the research to the fourth year and also for the license projects, increasing the difficulty of exercises, to solve problems in existing situations and sites. The goal of developing and refining students creative

personality was achieved by explaining and applying personalized guidance and design corrections to each artistic temperament of students, for all landscaping concepts and arrangements.



**Fig. 2 -** Student project with circles and squared forms-photo by Dascălu



**Fig. 3 -** Student project with circles and organic forms-photo by Dascălu

In the case of the exterior courtyards of “building B”, belonging to “Al. I. Cuza” University of Iasi (Fig. 4), the investigation of the existing situation revealed dysfunction and planning needs, helping us to establish efficient landscaping solutions. After the restoration of “building A” this university had no enough funds for rehabilitation of “building B” nor for indoor spaces, nor for exterior spaces. In the back area of the “building B” there are three exterior courtyards in an advanced stage of decay, unequipped and almost deserted (Fig. 5). The selected proposals provide two versions of courtyard planning, with various functional areas, designed as multifunctional spaces. We guided the design conceptions, each of the two projects being differentiated by original shapes and features, giving personality to the compositions (Fig. 6).



**Fig. 4** - Existent situation - [www.uaic.ro](http://www.uaic.ro) - processed photo by Dascălu



**Fig. 5** - Existent situation-courtyard in advanced stage of decay-photo by Dascălu



**Fig. 6** - Examples from yard projects: left version 1; right version 2 - photo by Dascălu

The spaces were designed both for students and teachers, also for university employees. These spaces are specifically conceived to provide various possibilities for outdoor activities such as: active and passive recreation, leisure, relaxation, self-study and outdoor classes, cultural/educational workshops and other extracurricular activities. In this context, the types of landscape furniture have been studied to provide opportunities for all users, of having different activities. Resting areas and places were designed for individual, collective and

semicolective use, depending on the types of outdoor activities. Some of the effects of morphological landscape elements - such as ground, water and vegetation - were conceived to create either study atmosphere, or to stimulate communication and socialization. To separate rest areas were created differences of land level, water basins, decorative walls with or without water games, colonnades, portals of various materials. These elements have contributed also to directing interior perspectives, helping to a better perception of spatial proportions of the yards. The land was systematized by proposing vertical difference of max. one meter, solved by terracing, steps and other elements judiciously chosen. We avoid using too large variety of furniture, materials, shapes and colors, which could corrupt the aesthetical harmony, or may create confusion effects by over-saturation of perception (Dascălu, 2011). We tried in these design concepts to control the perception of space distances, the distance between the viewer and all points of interest, also to proportionate the rhythm of routes intervals. All these elements have a great importance because they influence the composition, giving balance and harmony for overall final results (Jurov, 2006). In this context were studied carefully the various types of networking between all elements of landscaping that can provide personality-all subtle morphological items, like light and color, play an important role in achieving benefic psychological effects.

## CONCLUSIONS

1. The aims of an efficient correlation of scientific research with specialized education, was verified through the creativity of students to solve the virtual exercises and the practical applications for the existing cases. All solutions presented in this paper are based on our own original design research, verified during the design hours through practical guidance of the students.

2. The students projects for the courtyards belonging to "Al. I. Cuza" University of Iasi proved that a sustainable design can be applied in other similar cases, namely limited and degraded universities external spaces, only by creating multifunctional solutions, for a wide range of outdoor activities.

*Acknowledgments.* We thank you for student collaboration: Sucila Radu (Fig. 2), Moraru Mihaela (Fig. 3), Huza Iulian and Stanciu Mitrut (Fig. 6).

## REFERENCES

1. Dascălu Doina Mira, 2011 - *Landscape effects of urban furniture textures*. Bul. USAMV Cluj Napoca, Hort. 68(1): 324-331.
2. Jurov C., 2006 - *Arhitectura ambianțelor. Elemente de teorie cognitivă și psihologie ambientală în arhitectură*. Editura Capitel, București, p. 85-93.
3. Krauel J., 2006-*The art of landscape*. Ed. Links, Barcelona, p. 28-33.
4. Petrovici Liliana, Nica R.M., 2012- *Function and meaning in contemporary landscape design*. Lucr. St. USAMV Iasi, seria Hort., 55(2): 587-593.

## SUSTAINABLE COMMUNITY REHABILITATION WITH FORESTRY SYSTEMS

### REABILITAREA SUSTENABILĂ A COMUNITĂȚILOR PRIN SISTEME DE CULTURI FORESTIERE ÎN ROMÂNIA

NICA R.M.<sup>1</sup>, DUMITRAȘCU Aurora Irina<sup>1</sup>,  
VÎNĂU Nely<sup>1</sup>, CORDUBAN C.G.<sup>1</sup>

e-mail: nica.razvan@tuiasi.ro

**Abstract.** Development of green areas at the periphery of towns was a concern for city planners since the middle of the XIX<sup>th</sup> century. Sustainable design of urban systems as ecological entities calls for increasing the percentage of planted areas as well as community oriented projects. The control of urban sprawl is basically unattainable, the trend of housing developments at the periphery being noticeable in later years in all major towns in Romania. The need to juxtapose green areas in-between housing developments is just as pressing as the one for public functions. A solution would be to intervene with a pattern of small forestry plantation patches of fast growing trees, in addition to extending the natural forests. Urban forestry provides means for sustainable tree harvesting, additionally stabilizing the concentration of carbon dioxide in the atmosphere at microclimatic level. Forestry plantation could be beneficial for the community in terms of socioeconomics, providing a sustainable source of income. The authors propose means of implementing such forestry systems with regard to socio-economic aspects of community rehabilitation.

**Key words:** sustainable, community rehabilitation, forestry plantations

**Rezumat.** Dezvoltarea de zone verzi, la periferia orașelor a fost o preocupare pentru planificatorii orașelor de la mijlocul secolului XIX. Proiectarea durabilă a sistemelor urbane ca entități ecologice solicită creșterea ponderii suprafețelor plantate, precum și proiecte orientate spre comunitate. Controlul expansiunii urbane este, în principiu, de neatins, tendința de evoluție a construcțiilor de locuințe la periferie fiind vizibilă în ultimii ani în toate orașele importante din România. Nevoia de a juxtapune spații verzi între dezvoltările de locuințe este la fel de presantă ca cea pentru funcții publice. O soluție ar fi intervenția cu un model de zone mici de plantații forestiere cu specii de copaci copaci cu creștere rapidă. Plantațiile forestiere oferă mijloace sustenabile pentru recoltarea copacilor, stabilizând concentrația de dioxid de carbon în atmosferă la nivel microclimatic. Culturile forestiere sunt benefice și pentru comunitate din punct de vedere al socio-economic, oferind o sursă durabilă de venit.

**Cuvinte cheie:** sustenabilitate, reabilitarea comunității, culturi forestiere

## INTRODUCTION

Horizontal sprawl of cities through progressive dwellings developments in the suburbs must be joined by an effort to extend the green areas, with special

---

<sup>1</sup>"Gheorghe Asachi", Technical University of Iasi, Romania



care to the forests that surround towns, forming the green belt, thus balancing the air quality and reducing pollution.

In addition to the care for extending natural forests, forestry plantations with fast growing tree species can successfully meet this challenge. Forest plantations combine the economic benefits (sustainable source of biomass, by increasing the share of energy from renewable energy sources) with the ecological ones (storing solar energy in wood mass and fixation of carbon dioxide from the atmosphere), representing alternatives to forestry and the unavoidable alteration, even through sustainable management policies of their ecosystem balance. The environmental benefits of forestry plantations include, aside from the priority objectives previously set, the phyto-remediation of polluted land with household waste and heavy metals, recovery of agricultural land damaged by excessive fertilization, reduction of pollution of groundwater, thus projecting a natural filter (Rosu et al, 2009).

Developing and expanding forests are obligations and regional priorities in order to achieve ecological balance locally, nationally and globally, being carried out through the National Afforestation Program, (Petrescu-Mag, 2011). In this regard, short rotation forestry plantations on land outside the forest is a concern at European, regional and national levels (Cosofret and Danila, 2014).

In an extended review of urban forestry, Roy et al, 2012, evaluates the potential of urban trees of ameliorating the environmental degradation resulting from rapid urbanisation. The review extrapolates from previous research the services of urban forests, that include:

- life support, energy conservation and opportunities for aesthetic experiences, citing ecological economics literature;
  - mitigating carbon pollution, improving urban air quality, attenuating storm-water flooding, conserving energy and reducing noise, among others, citing urban forest and environmental literature;
- but also the disservices, that include:
- financial, health and maintenance burdens upon urban residents, citing environmental and social studies;

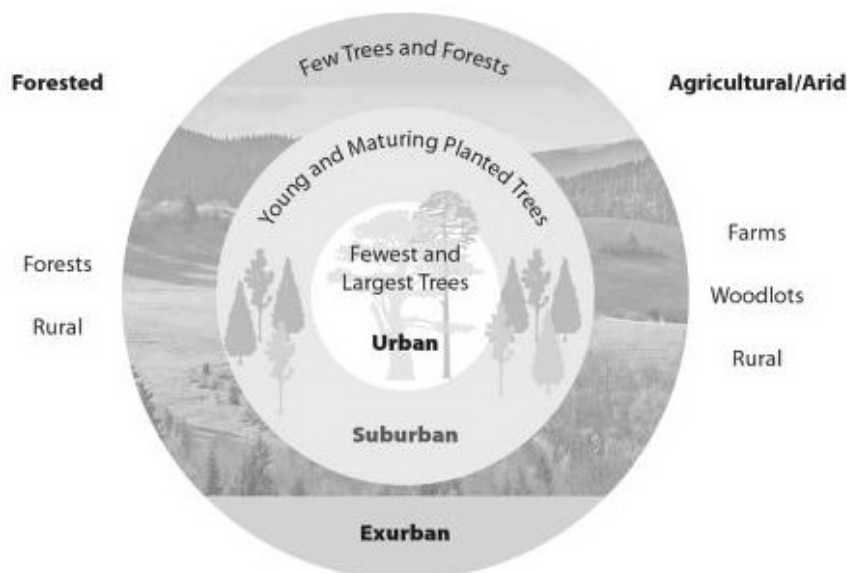
Tree plantations, though encouraged through governmental policies, in an effort to sequester carbon, have raised recently several questions regarding social and ecological effects. Orderly rows of trees have increased sevenfold over the last two decades, assuring new means of production in societies with deprived labour forces. In a sustainability concerned world, their ecological benefits have been praised, although some critics have voiced concerns over the negative impact of exotic species introduction, high water consumption and even negative social and economic impacts (Rudel, 2009).

## **MATERIAL AND METHOD**

The space occupied by forests in relation to city centre can be classified in four major areas:

- urban;
- sub-urban;
- ex-urban;
- rural;

with the development of green belts and the possibility of extending them through plantations in the sub-urban and ex-urban areas, (fig. 1) (Konijnendijk *et al*, 2006)



**Fig. 1** - Land use and the urban forest (Miller *et al.*, 2015)

The density (number of trees on a patch of land) varies according to the zone patterns after:

- urban area is characterized by the lowest density of trees, which are found in parks and residential areas, with the greatest degree of arboreal maturity;
- suburban area has a higher density, with the greatest potential for increasing forested areas, particularly in residual perimeters, characterized by the new built neighbourhoods young plantations and conserved forest patches particularly in non-agricultural lands;
- ex-urban zone is located at the junction of suburban area with rural one, is defined in the specific literature as: peri-urban forest, anthropogenic-natural interface or urban-rural interface and is characterized by a patchwork of farmland, woodland, with industrial or commercial areas. In the forested areas, tree numbers will be decreasing, while in agricultural areas, they will be growing in number, (Helms, 1998).

In Romania, the need to juxtapose green areas in-between housing developments is just as pressing as the one for public functions. A solution would be to intervene with a pattern of small forestry plantation patches of fast growing trees, in



addition to extending the natural forests. Urban forestry provides means for sustainable tree harvesting, additionally stabilizing the concentration of carbon dioxide in the atmosphere at microclimatic level. Forestry plantation could be beneficial for the community in terms of socioeconomics, providing a sustainable source of income.

The authors propose means of implementing such forestry systems with regard to socio-economic aspects of community rehabilitation.

## RESULTS AND DISCUSSIONS

### City forested belt, historical and current trends

The green city belt represents the forested suburban and ex-urban area, with the role of providing an environment for recreation of the urban population and improving air quality by absorbing carbon dioxide from the atmosphere and, as a secondary objective, providing wood fibre by a sustainable management (Miller *et al*, 2015).

In Europe, the first concerns for the wooded areas in the proximity of cities date back to Renaissance and Baroque, with the demolition of the city walls, and migration of the aristocracy to villas and palaces in the suburbs.

The concept of forested city belt in the current meaning arises on the industrial revolution. In response to the first chaotic development and uncontrolled occupation of land by the early nineteenth century, cities in the UK make great efforts to expand green areas on the periphery, by introducing forested belts and satellite settlements.

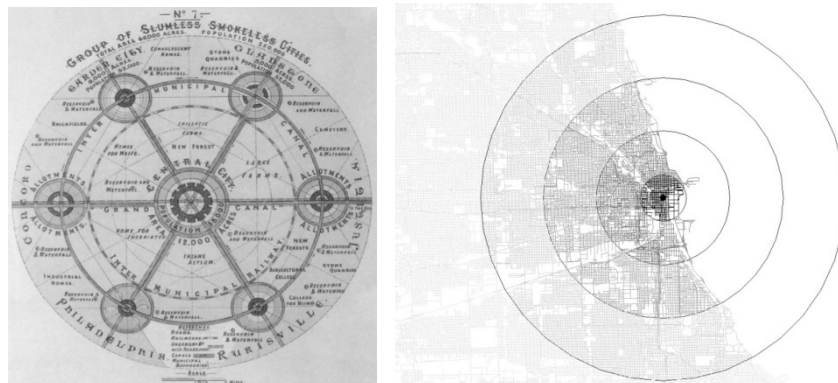
The "Romantic Landscape" pursued by this process becomes an area of leisure enjoyed by all classes. With the expansion of industrialization across the continent, similar patterns are repeated in many European cities (Lawrence, 1993).

Early twentieth century finds the city in search of solutions to social and urban issues. The development of large industrial cities, a great number of housing projects, often unsanitary and population migration from rural green to urban gray, all lead to a heterogeneous landscape. Moreover, the quality of urban life generates controversy on how to structure the city and how people can interact with the natural inhabitant. Thus, two patterns of green city planning emerge:

- the progressive model, developing the concept of functional segregation, separating residential areas from industrial areas, integrating the city with no apparent limit landscape;
- the cultural model, supporting the idea that the key to metropolitan chaotic expansion resides in the construction of new satellite towns, with concentric green rings surrounding the city (Choay, 2002).

The cultural model promotes the idea of city green belt, continuing on the path set by British urban planners in the XIX<sup>th</sup> century. Among the prominent figures of this movement could be mentioned Ebenezer Howard, that brought the concept of garden city, R.Unwin and B.Parker, that through their projects both in the United Kingdom and the United States have implemented the concept of the

city's green belt. A follower of the Chicago School, W.E. Burges in the interwar period is advancing a new theory in urban sociology and ecology, publishing in 1925 the model theory of concentric circles. Following an analysis carried along a decade of Chicago's development, proposed an ideal spatial organization for a city that has the start in the urban core, bordered by a green area perimeter (fig. 2)



**Fig. 2 - Concentric models, Ebenezer Howard and W.E. Burges**  
(commons.wikimedia.org, <http://www.gutenberg.org>)

Currently, many cities, both in the developed and in the developing world take efforts to expand the forested belt: Beijing has expanded green areas from 3.2% in 1947 to 26.9% in 1990; in Shenyang, China, growth in the last decade of green areas was of 8%, consisting mostly of young tree species. Policies to increase the green belt are followed up in major European cities as well, Dublin and Vienna monitoring the levels of fine particles per cubic meter, compared with the number of trees in the extended urban area, (Miller *et al.*, 2015).

### **Green belt, design considerations**

Forested landscapes in urban region tend to create their own aesthetic, but landscapers can manipulate the effects through arrangement of vegetation to further consider simplicity, balance, focalization, unity, proportion and rhythm. A good design must integrate elements such as:

- connectivity between various areas;
- integration of open space;
- uses of paths for circulation;
- uses of vegetation as visual cues, such as focal points of interest, (Bell *et al.*, 2005).

People's choices may vary when considering the size of vegetation, tree density, canopy considerations. Various studies showed that people tend to prefer moderately dense forests over paths of few trees or over-dense ones, that their perception is largely influenced by their distance from the trees and varies with age and education. More mature individuals enjoy more dense stocking and a

natural look of the forest. Graduates of science schools are more inclined to prefer a neat and organized landscape, while art graduates enjoy natural-like environments (Miller et al., 2015).

Considering the above, it is safe to assume that from an aesthetic point of view, orderly rows of trees can be associated with the existing tree stock in an effort to extend the city's forest belt. After all, similar landscape solutions have been practiced for the classical orderly alleys in European parks for centuries.

## CONCLUSIONS

The progressive urban model claimed a vertical city that is lost in the landscape, and the culturalist urban model supports the idea of a precise city limitation through a green belt.

Nowadays, in the current context of excessive urbanization, it seems increasingly difficult to achieve a physical separation or a necessary green transition for the city functional zones and the surroundings. Realization of green belts around cities is not a new concern, but one that has been applied successfully, either based on the urban theories described above, but mostly based on the experiences of city dwellers for centuries. Forest plantations may accompany urban developments by creating new green spaces of parks with ornamental trees in the suburbs, bringing social benefits as well.

## REFERENCES

1. **Lawrence H. W., 1993** - *The Neoclassical Origins of Modern Urban Forests*, *Forests and Conservation History* 37: 26-36;
2. **Helms J. (Ed.), 1998** - *The Dictionary of Forestry*, Washington D.C., Society of American Foresters
3. **Choay F., 2002** - *Urbanismul - Utopii și realități*, Ed. Paideea, 34 p;
4. **Bell S., Bloom D., Rautamaki M., Castel-Branco C., Simpson A., 2005** - *Design of Urban Forests*, *Urban Forests and Trees*, Springer, Berlin, pp. 149-186;
5. **Konijnendijk C.C., Ricard R.M., Kenney A., Randrup T.B., 2006** - *Defining urban forestry – A comparative perspective of North America and Europe*. *Urban Forestry & Urban Greening* 4(3- 4): 93-103;
6. **Rosu C., Mihai Filat M., Chira D., 2009** - *Cultura plopilor, a salciilor si a altor specii forestiere in zona inundabila a Dunarii*, Editura Tehnica Silvica, pp. 5-12;
7. **Rudel T. K., 2009** - *Tree Farms: Driving Forces and Regional Patterns in the Global Expansion of Forest Plantations*, *Land Use Policy*, 26, Elsevier, pp. 545-550;
8. **Petrescu-Mag, 2011** - *Protectia mediului în contextul dezvoltării durabile. Legislație și instituții*, Editura Bioflux, Cluj-Napoca, România; pp. 214-220;
9. **Roy S., Pickering C., Byrne J.A., 2012** - *A systematic quantitative review of urban tree benefits, costs, and assesment methods across cities in different climatic areas*, *Urban Forestry & Urban greening*, Researchgate, 56 p;
10. **Cosofret C., Danila I.C., 2014** - *Sustenabilitatea culturilor cu specii cu ciclu scurt de productie pe terenuri din afara fondului forestier, proiectul Stroma*, *Bucovina Forestiera* 14(2): 252-254 pp;
11. **Miller R.W., Hauer R.J., Werner L.P., 2015** - *Urban Forestry, Planning and Managing Green Spaces, Third Edition*, Wavelang Press, Long Grove Illinois, pp. 5-22, 64-82.

## MATTERS REGARDING THE IDENTITY OF SPACE BETWEEN DESIGN AND PERCEPTION

### APRECIERI PRIVIND IDENTITATEA SPAȚIULUI ÎNTRE CONCEPȚIE ȘI PERCEPȚIE

**ȘTEFAN Diana<sup>1</sup>**

**e-mail:** stefan\_g\_diana@yahoo.com

**Abstract.** *In this paper the author studies the scientific literature in order to clarify the terms of design and perception in relation to the identity of space. The scientific literature highlights four theoretical directions on the relationship between designs – perceptions – the identity of space. For example, the identity of space is a function of urban – architectural reality, of sentimental - emotional reality and a variation of determinants. In this regard the variation of determinants is a function connected with the mass of receptors (residents, tourists, non-tourists, professionals etc.) and with the mass of transmitters (buildings, streets, urban furniture, green spaces, billboards etc.). The authors appreciate that people give form to the environment as the environment forms people, stressing the interdependence of the ways and their living environment. Adding the analysis it's determined that the individual has a certain model after which operates in urban environments (mental maps), there is a variation of the determinants that conditions his perception, certain elements that make him close to the environment (territoriality), meanings are assigned to the space that surrounds him and is judged by peers according to the environment in which he lives. The sum does not follow a set of rules, laws and general principles of perception that leads to good urban design of urban – architectural reality, both contributing to the clarity of the identity of a space.*

**Keywords:** design, perception, identity.

**Rezumat.** *În lucrare autorul studiază literatura de specialitate cu scopul de a clarifica termenii de concepție și percepție în relație cu identitatea spațiului. Literatura de specialitate pune în evidență patru direcții teoretice privind relația concepție – percepție – identitatea spațiului. Spre exemplu, identitatea spațiului este o funcție a realității urbanistice – arhitecturale, a realității emoționale – afective și a schemei determinanților. În acest sens schema determinanților este o funcție legată de masa receptorilor (rezidenți, vizitatori, non-vizitatori, profesioniști etc.) și a masei emițătorilor (clădiri, străzi, mobilier urban, spații verzi, panouri publicitare etc.). Autorul apreciază faptul că oamenii dau forma mediului înconjurător și mediul înconjurător formează oamenii, subliniind interdependența dintre modul și mediul de viață al acestora. Însușind analizele efectuate s-a stabilit că individul are un anumit model după care își desfășoară activitățile în spațiul urban (hărți mentale), există o schemă a determinanților ce îi condiționează percepția, anumite elemente îl fac apropiat de mediul înconjurător (teritorialitatea), atribuie semnificații spațiului ce îl înconjoară și este judecat de semenii în funcție de mediul în care trăiește. Din sumă nu rezultă o serie de reguli, legi și principii generate de*

---

<sup>1</sup> ”Ion Mincu” University of Architecture and Urbanism, Bucharest, Romania

*percepție care duc la o bună concepție a realității urbanistic – arhitecturale, ambele contribuind la claritatea identității unui spațiu.*

**Cuvinte cheie:** concepție, percepție, identitate.

## INTRODUCTION

Research on the relationship between designs – perception – the identity of space, with man as unit, were made mainly during the 1960-1970. Authors who have distinguished themselves in this field are Kevin Lynch with “*The Image of the City*”, Donald Appleyard with “*Ambient et Comportament*” (1977), Christian Norberg-Schulz with “*Habiter. Versune architecture figurative*” (1984), Edward Hall (1966) and others.

**Design** is the process by which a team of specialists comes with solutions and improvements to the current problems of urban – architectural reality. The team of specialists consists in a number of people trained in science and varied disciplines such as urbanism, architecture, sociology, economics, mathematics, geography, ecology etc., which work together to resolve given problems. This interaction results in a sum of factors, principles and laws underlying the future design.

The speech regarding **perception** is sporadic, oscillating between subjectivism and objectivism, between personal poles and urban space poles. Perception has a fundamental reason in “need for localization” (Tolman, 1951) and is directly linked to individual experience, with familiarity, with mood, with performed activities and with the configuration of the environment. Studies have shown that perception is operational, responsive and inferential.

Analysing the literature in the field it’s considered that until now were obtained data on perception, individual behaviour in urban space, but has not yet reached a sum of rules, laws and principles arising from this data that would lead to a better design of space that it will cause it’s strong identity.

## MATERIAL AND METHOD

To highlight the relationship between designs – perception – the identity of space, the author used as research methods the bibliographic study, phenomenology, semiotics and the method of combining several theories.

Man, the unit of measurement of the relationship mentioned above, perceive the urban – architectural reality not only visually (images) but also by auditory, olfactory and tactile means, aspect referred by phenomenology.

The individual assigns meanings to urban - architectural reality generated by signs, symbols and connotations. This award constitutes the sentimental – emotional reality. This aspect is studied by semiotics.

## RESULTS AND DISCUSSIONS

Studies on the relationship between design – perception – the identity of space has as a starting point the determining plan which includes determinants assign to the mass of receptors (individuals) and determinants assign to the mass of transmitters (urban – architectural reality) (Fig. 1).

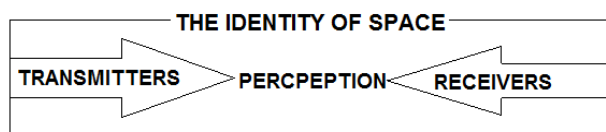


Fig. 1 –The relationship between determining plan – perception – the identity of space.

Depending on the receiver and the transmitter the determining plan include the following:

- The mass of receptors:
  - sex, age, religion;
  - residents, tourists, non-tourists, professionals, disabled;
  - the geographical area of origin;
  - the availability of the individual to spend time indoors or outdoors;
  - cultural differences (differences in perspective of space between an individual from a northern city and one from a southern city);
  - the experience of the individual in different places;
  - education, the level of income, the level of stress etc. .
- The mass of transmitters:
  - the shape of the object, accessibility, style, height, size;
  - the area is built, the period in which it was built;
  - material, colour, light etc.

The bibliographic study highlights four important models in the relationship between designs – perception – the identity of space.

There is **mental maps** pattern set by Kevin Lynch in 1960 in his book “*The Image of the City*”. Following the studies it concluded that users understand the surroundings in a consistent and predictable form, mental maps of five elements: routes (roads, streets, sidewalks and other paths), edges (perceived boundaries, walls, buildings etc.), districts (relatively large sections of the city distinguished by a certain identity or character), nodes (crossings, focal points etc.), landmarks (easily identifiable objects that serve as reference points).

This theory gathers the thinking of the individual and his way of orientation in space in a simplified scheme that does not include factors that cause the individual to choose certain routes, edges, districts, nodes, landmarks in a given area and how it interprets these chosen elements. A person who uses the city, carrying various tasks, selects certain aspects of their environment in conducting those tasks. Set in an x point of the town, the individual who must operate in a y point of the city, will choose a route that is not necessarily conditioned by its dimension but by personal experience.

The idea of identifying any person with a particular place and its reporting to it was stated by Christian Norberg-Schulz in “*Habiter. Vespune architecture figurative*” (1984).

Identification refers to the quality of things and orientation to their spatial correlation. Architectural works are objects that embody the core value of human identification, reproducing the world as it is. Identifying always refers to everyday actions, therefore depends on psychological orientation function.

The elements of architectural language which one identifies with may be concentrated in three areas: morphology, topology and typology. Morphology is how the form was built. Spatial order translated into organization is stated by topology. Typology expresses manifestations of habitat types.

The individual can identify with one, more or all aspects of architectural language. If man is reflected in many aspects of urban – architectural reality, the identity of space becomes more clear and forceful.

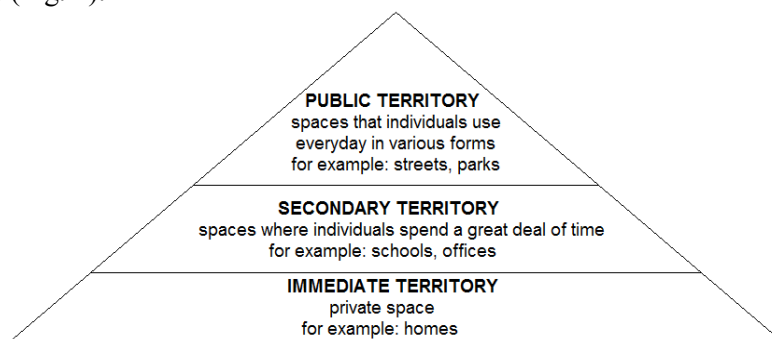
Identification as well as mental maps aren't presented by their authors as concepts that encompass the entire relationship between design – perception – the identity of space, it still remains neglected the factors that influence them.

**Proxemics theory** defined by Edward Hall in 1966 is designating how the use of space depends on its quality of territory and its cultural components.

Man assigns meanings to physical space, as such human territory is eminently social. The territory itself doesn't exist, it has no reality except by one who uses it and makes it an object of knowledge. The territory is a physical space cutting in areas defined subjectively by the quality of relations established by it.

*“Territoriality is the basis of stable social organization”* (Edney, 1975).

There are three types of territories: the primary territory occupied constantly by the same person or group of people (for example: homes), the secondary territory which is not clear who belongs to, a relatively large number of people have access (for example: a certain place in the classroom) and public territory over which individuals do not believe they have rights when they don't occupy it (for example: a park bench). The distinction between these categories contribute to understanding the sentimental – emotional reality of individuals associated to the territory and how an individual will react when their territory is invaded (Fig. 2).



**Fig. 2** – The importance of bringing closer the territory to the individual.



Individuals derive part of their identity from these territories. Protecting territory by individuals, in fact of their self, analogies with the animal world. The individuals are more successful in different situations where their territory is: in football we have to do with a “surprise” when the foreign team wins. Changes over centres, routs, landmarks etc. can be perceived as an invasion of space which may lead to a negative reaction, reason underlying territoriality.

People use markers recognizable by everyone, such as objects or signs that indicate possession of the territory by a particular individual.

Proxemics exposes the attachment to territory and the way individual attribute it (markers that are recognizable to everyone). It remains to be seen whether the concept of territory changes according to media, economic, political influences and if it differs from one culture to another.

New factors influencing the relationship between design – perception – the identity of space are formulated by Jean Baudrillard in “*Module on Simulacra and Simulation*” explaining the distinction between reality and semblance loss.

In postmodern culture our society has become so confident in models and maps that all contact with the real world that preceded the map is lost. The new factors of postmodern culture that influence the relationship mentioned above are: media culture, exchange – value relationship, multinational capitalism, urbanization, language and ideology.

**Media culture** represented by television, movies, magazines, billboards and the internet is concerned not only about sharing information, but also with the interpretation of individual privacy for himself, causing him to address to peers through the lens of media. Man no longer purchase goods according to their needs, but because of the desire increasingly polished by promos, which take the individual a step away from the reality of his own body and the world around him.

Entry into capitalism meant that the individual has stopped to think of goods purchased through the **utilization – value ratio**, property started to lose its material reality. Instead it began to be seen after value and in what it can be changed, its exchange value, assets are regarded by the individual through the **exchange – value ratio**. Multinational capitalism results in loss of basic notion of the goods consumed. Much of the consumers of Starbucks brand, recognized worldwide as a distributor of coffee, wouldn’t be able to recognize a coffee tree and to know the process of obtaining the finished product.

**Urbanization** leads to an alienation of man from the natural environment.

**The language**, in subtle ways, keeps the individual away from reality. Postmodernism understands the ideology as a support to human perception of reality. There is nothing outside ideology, at least nothing that can be expressed by an articulated language. Because people are so reliable on language to structure their own perception, any representation of reality is always already ideological sham, always already built by simulacra.

This hypothesis is sounding the alarm on the true urban – architectural reality and the susceptible sentimental – emotional reality. As a conclusion to

those mentioned above, the relationship between designs – perception – the identity of space is strongly influenced by the society characteristics, which are always evolving.

## CONCLUSIONS

1. Based on determinants plan the study highlights four models in the relationship between designs – perception – the identity of space. The models listed are: mental maps, identifying an individual with a particular place and its responding to it, proxemics and the new factors of postmodern world.

2. Adding the analysis it's determined that the individual has a certain model after which operates in urban environments (mental maps), there is a variation of the determinants that conditions his perception, certain elements that make him close to the environment (territoriality), meanings are assigned to the space that surrounds him and is judged by peers according to the environment in which he lives. The sum does not follow a set of rules, laws and general principles of perception that leads to good urban conception of urban – architectural reality, both contributing to the clarity of the identity of a space.

## REFERENCES

1. **Afrăsinei Alexandra, 2011** - *Despre lectura orașului*, "Ion Mincu" University of Architecture and Urbanism, Bucharest, PhD Thesis.
2. **Appleyard Donald** - *Notes on Urban Perception and Knowledge*. Extras de pe internet pe 15 Septembrie, 2015, de la adresa: [http://edra.org/sites/default/files/publications/EDRA02-Appleyard-97-101\\_0.pdf](http://edra.org/sites/default/files/publications/EDRA02-Appleyard-97-101_0.pdf).
3. **Baudrillard Jean, 1981** - *Module on Simulacra and Simulation*.
4. **Boncu Ștefan** - *Teritorialitatea și spațiul personal. Comportamentul uman în condiții de aglomerație*, Bucharest University, Social Psychology, course 33.
5. **Ghenciulescu Ștefan, 2004** - *Identitate și continuitate în plan urban și teritorial. Două studii de caz: București și Elveția*, "Ion Mincu" University of Architecture and Urbanism, Bucharest, PhD Thesis.
6. **Golledge, Reginald G.; Zannaras Georgia** - *The Perception of Urban Structure: An Experimental Approach*. Extras de pe internet pe 15 Septembrie, 2015, de la adresa: [http://www.edra.org/sites/default/files/publications/EDRA02-Golledge-111-117\\_0.pdf](http://www.edra.org/sites/default/files/publications/EDRA02-Golledge-111-117_0.pdf).
7. **Lynch Kevin, 1960** - *The Image of the City*.
8. **Norberg-Schulz, Christian, 1984** - *Habiter. Versune architecture figurative*, Gruppo Editoriale Electa, Milano.
11. **Popliceanu Erika-Eva, 2012** - *Relația biunivocă dintre comportamentele socio-umane și spațiul public urban*, "Ion Mincu" University of Architecture and Urbanism, Bucharest, PhD Thesis.

## ANALYSIS OF THE GREEN AREAS WITH UNLIMITED ACCESS AND THEIR IMPACT ON THE POPULATION FROM IAȘI CITY

### ANALIZA SPAȚIILOR VERZI CU ACCES NELIMITAT ȘI IMPACTUL ACESTORA ASUPRA POPULAȚIEI DIN MUNICIPIUL IAȘI

**SANDU Tatiana<sup>1</sup>, TROFIN Alina-Elena<sup>1</sup>,  
PANTAZI Viorica<sup>2</sup>, UNGUREANU Elena<sup>1</sup>**  
e-mail: tatiana\_sandu69@yahoo.com

**Abstract.** The current area of Iasi is approx. 3.770 ha of which the green system occupies approx. 826.80 ha (both green spaces within city limits and outside the city) and only 362.60 ha represent unlimited access green spaces [approx. 44% of the total area of green space in Iasi and only 9.61% of the area attributed to the green system (Inside and outside town)]. From the analysis made for the most representative unlimited access green spaces in Iasi, results the existence of a small number of green system elements and an uneven distribution with very different shares from a neighborhood to another, from one area to another. This situation is the result of a sequential urban planning approach, held in various periods of time without solutions correlation and without to promoting the idea of linking the town and its outskirts. The comparative study regarding accessibility, service range and radius of influence of the main types of unlimited access green spaces in Iasi, completed with the analysis performed using a conducted opinion survey (survey covers a total of ten items, each with 3 ÷ 15 possible answers), led to the deduction of deviations from the indicators used in assessing urban green spaces.

**Keywords:** unlimited access green spaces, Iași, population, assessment indicators.

**Rezumat.** Suprafața actuală a municipiului Iași este de cca. 3.770 ha din care sistemul verde ocupă cca. 826,80 ha (atât spații verzi intravilane cât și extravilane) și numai 362,60 ha reprezintă spațiile verzi cu acces nelimitat [cca. 44% din suprafața totală de spațiu verde din municipiul Iași și doar 9,61 % din suprafața destinată sistemului verde (intravilan și extravilan)]. Din analiza făcută a celor mai reprezentative spații verzi cu acces nelimitat din municipiul Iași în parte, rezultă existența unui număr restrâns de elemente de sistem verde și o repartitie neuniformă, cu ponderi foarte diferite de la un cartier la altul, de la o zonă la alta. Această situație este rezultatul unei abordări urbanistice secvențiale, desfășurată în diverse etape de timp, fără corelări de soluții și fără a se promova ideea de legătură între intravilan și extravilan. Studiul comparativ privind accesibilitatea, raza de deservire și raza de influență ca distanță a principalelor tipuri de spații verzi cu acces nelimitat din municipiul Iași, completat cu analiza realizată cu ajutorul sondajului de

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

<sup>2</sup> Iasi City Hall-S.C. Public Services S.A. Iasi, Romania

*opinie efectuat (sondajul cuprinde un număr de zece itemi, fiecare cu 3 ÷ 15 variante de răspuns), au condus la deducerea unor abateri de la indicatorii utilizați în evaluarea spațiilor verzi urbane.*

**Cuvinte cheie:** spații verzi cu acces nelimitat, Iași, populație, indicatori de evaluare.

## INTRODUCTION

The current area of Iasi is approx. 3.770 ha of which the green system occupy about 826.80 ha (both green spaces within city limits and outside the city) and 362.60 ha represent the unlimited access green spaces (approx. 44% of the total area of green space in Iasi (Sandu *et. al.*, 2003).

According to the World Health Organization, the share of green space per inhabitant must be 50 square meters of green space

## MATERIAL AND METHOD

Territory analysis of the green space with unlimited access from Iasi was carried out between May 2014 - May 2015 through the work of visual monitoring of the current status of these types of green areas in the municipality of Iasi, more complex observations being carried out in collaboration with Green Spaces department specialists of the Iasi City Hall, as part of a broader collaboration.

The used evaluation indicators and the analysis of urban green spaces at the local level, as indicated by the European Union, refers to:

**a) Indicators concerning the urban green space availability:** total surface green / blue, green areas and water share in the city, the area of green space / inhabitant etc. In some cases more detailed indicators are used to provide information about form, function types of green space (*Practical Evaluation Tools for Urban Sustainability – Green Blue* citat de Chiriac D., et al., 2008).

**b) Indicators concerning the accessibility of green spaces,** or their proximity, measured by the percentage of the population that is located less than 15 minutes' walk from the urban green areas or in other cases, at 150 m from home (*Proiectului „COST Action C11 – Green Structure and Urban Planning” integrat în al V-lea Program-cadru al Uniunii Europene cu privire la evoluția spațiilor verzi*, citat de Chiriac, 2008).

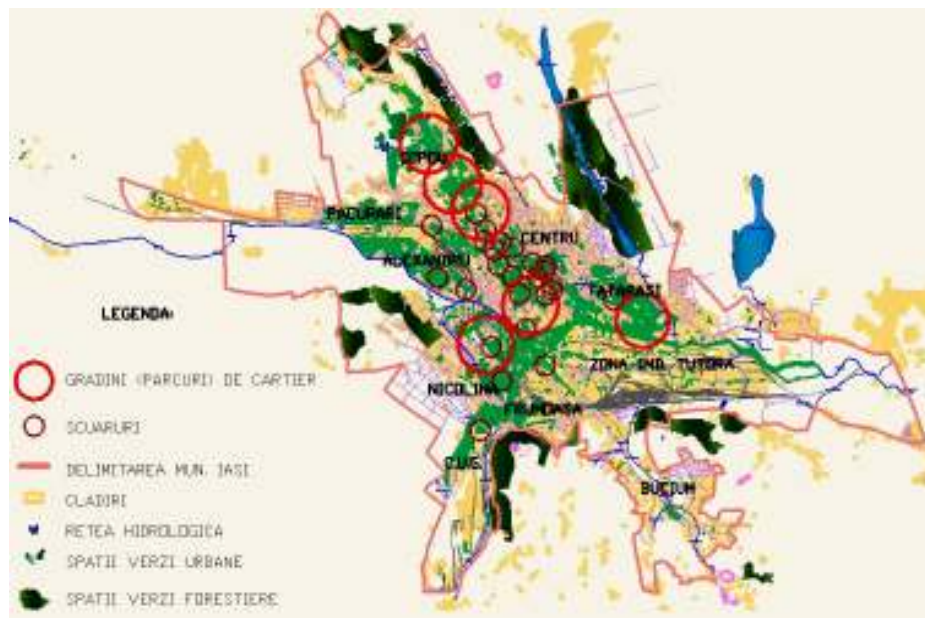
There was developed a comparative study on accessibility, range of service and range of influence as the distance of the main types of green spaces with unlimited access from Iași City and supplemented by the analysis performed using the poll conducted from March to May 2015 (the survey covers a number of ten items, each with 3 ÷ 15 answers) on a total of 523 respondents, which led to the deduction of deviations from the indicators used in assessing urban green spaces.

## RESULTS AND DISCUSSIONS

The most representative unlimited access green spaces from Iași City fulfilling the surface indicator are shown in Figure 1. The main factor that determines the recreational value of the analyzed territory is its accessibility. In this respect it is considered the extent endowment of the territory with communication routes, their modernization, the frequency of transport links and the distance to them, ș.a. (Sandu, 2009).

It is considered that for Iasi, the potential areas for short-term recreation in green spaces within city limits have an accessibility contained in a timeframe between 6 ÷ 25 minutes, by public transportation, car and foot traffic.

Urban green space efficiency is assessed by the range of influence, as distance and range of influence, as a service. Usually these limits do not have a geometric shape, but have a varied path, which is determined in relation to the opportunities for access to the considered green space and are influenced by the existence of natural or artificial obstacles.



**Fig.1** - Unlimited access green spaces distribution from Iași City (original)

The comparative analysis of the representative unlimited access green spaces in the municipality of Iasi (Table 1), tries to highlight the issues of accessibility for the urban green system elements and the degree of meeting the needs of the population of Iasi.

There is an obvious imbalance in terms of the geographical spread of these facilities, mostly located in areas Copou and center. Squares fall in rule as a share, but their distribution is not so satisfactory. The planted road strips are deficient by approx. 70 ha, which is quite serious because their implementation does not require such difficulties as creating new parks and gardens is. Data analysis shows that Iasi has a major deficit (approx. 216 ha) of areas occupied by parks and a deficit of approx. 90 ha in terms of neighborhood gardens.

It was noted that only for the areas inventoried in this study, the total number of visitors is estimated to be approx. 34.340 visitors, without the contribution of those from recreational areas outside the city.

Table 1

**The degree of meeting the needs of population materialized in access paths and entries to the main unlimited access green spaces** (according to norm no 112/1976)

Unlimited access green space	No. entries	From which:		No. access paths	Satisfies population
		Main entries	Secondary entries		
Gardens (parks) inside Iași					
Copou	4	1	3	4	yes
Expoziției	4	2	2	5	yes
Universității „Al. I. Cuza”	4	2	2	2	yes
Palas	9	3	6	2	yes
Tătărași	8	4	4	4	yes
Squares inside Iași					
Casa de Cultură a Studenților	4	1	3	2	yes
Piața Independenței	5	1	4	3	yes
Tg. Cucu-Biserica Bărboi	4	1	2	5	yes
Palatul Culturii	5	1	4	2	yes
Anastasie Panu-Elena Doamna	5	2	3	3	yes
Teatrul Național	5	1	4	3	yes

From the map being observed in Figure 2 we can notice that there are surfaces out of range of service, resulting in the fact that planted areas are undersized and part of the population does not benefit from them (eg neighborhoods Alexandru cel Bun, Bucium, Frumoasa, Moara de Vânt entirely as well as the neighborhoods Nicolina, C.U.G., Obreja, Păcurari-partially).

Estimations have led to the conclusion that the percentage of population of Iasi, within the zone of influence as distance, attending at a time the unlimited access green spaces and recreational areas related to city is approx. 45,300 visitors (about 13% of the total population), 5-7% less than the current standards for a city of Iasi category (Sandu *et al*, 2009).

The survey (questionnaire) conducted in Iasi population comprises a set of questions on people's perceptions about current and past status of urban green spaces and the activities that it undertakes in the space of these facilities. To better relevance of the answers that closely match the situation met in the field, subjects were interviewed in all six areas of Iasi.

Most of the answers given by the respondents notified malfunctions recorded in the green areas from Iași City which are actually an accumulation of failures, errors and shortcomings, from each urbanistic unit (neighborhood) that is part of the municipality. If at the district level these disturbances may be somewhat dimmed by use of an adjacent green space unit facilities, overloading its function, at the municipal level, the phenomenon does not allow this kind of "transfer", the effects being found in living standards and quality of urban life.



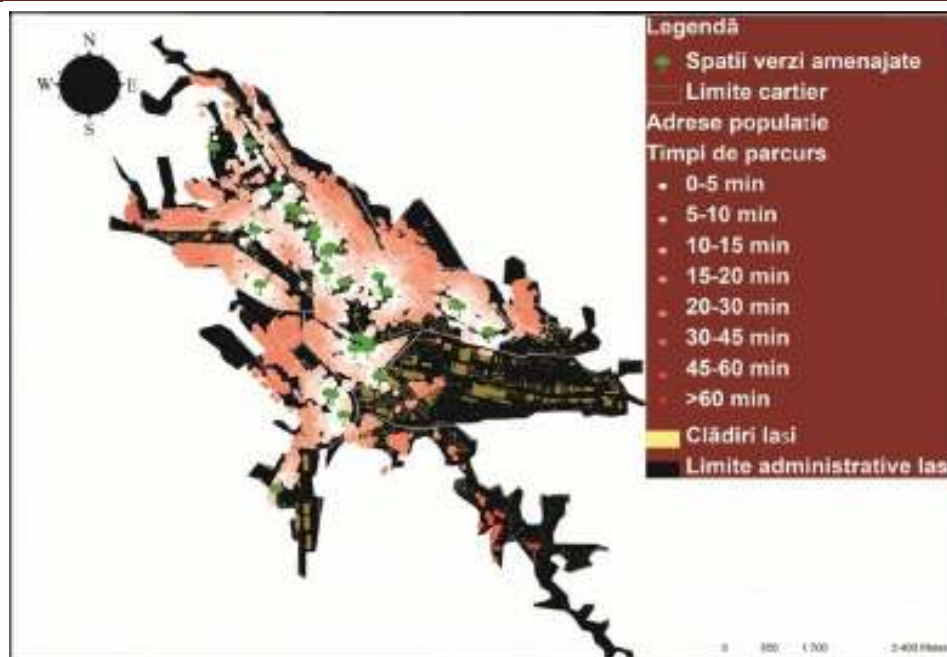


Fig. 2 - Distances / time to the nearest landscaped green space in Iași (original)

## CONCLUSIONS

The comparative study of accessibility, service range and range of influence as distance for the main types of unlimited access green spaces from Iași City and not the least the analysis conducted through the carried out surveys have allowed deducting the following conclusions:

- Deviations from the standards of unlimited access green spaces per capita:
  - in the city of Iași no landscaped green space meets the conditions of a city park. Both city parks and neighborhood parks are lacking.
  - the gardens under the administration of Iași Department of Public Services account for 4% of the 15% regulated settings and are concentrated in study zone I (Copou-Center) and isolated in zones IV and V (respectively Tătărași and Nicolina neighborhoods).
  - the existing squares represent approx. 45% of the total needed in the city, the deficit being about. 58 ha. Even if the squares are better represented, they are still missing in about half of the neighborhoods Bucium, Frumoasa, Galata, Moara de Vânt, Obreja, Țicău-Sărărie, Păcurari.
  - street lining are a category of green spaces relatively well represented in city of Iași, though there are areas (Moara de Vânt, Obreja) where they are missing or are underrepresented.



- according to norm 112/1973, the requirement for the Iasi city street plantations is approx. 70 ha. The deficit of around 32 ha is found particularly in neighborhoods Păcurari, Alexandru cel Bun, Galata, Socola, Nicolina Nicolina-CUG and in the gaps of existing alignments.

- the most representative alignments and strips are in Center and Copou area and yet they cover only approx. 55% of the necessary area.

- Deviations from accessibility standards:

- in the absence of town and neighborhood parks, whose functions are carried out by the gardens in the neighborhood, it appears that the most adequate is the case of Copou district, where there is a "chain" of gardens and parks (Copou, Expozitie) that meet the criteria of accessibility.

- to these is added the patch "Ghica Voda" and the gardens of Copou institutions which together constitute one of the greenest areas of a city in Romania.

- The functioning status of unlimited access green spaces:

- some of the green areas with unlimited access from Iași City do not have clear landscape and architectural compositions or do not have facilities to determine a specific profile.

- the wearing of the green spaces contribute to a non- corresponding urban aspect by: poor execution, lack of concern for the care and maintenance of green spaces, s.o.

- Non ensuring required environmental and aesthetic quality by the existence of poor neighborhoods, degradation of vegetation fund (scattered salt in winter, untreated disease and pest attacks stationing pollutants etc.) and environmental inappropriate use of plants from Iași City.

## REFERENCES

1. Chiriac D., Humă Cristina, Stanciu Mariana, 2008 - *Spațiile verzi - o problemă a urbanizării actuale*, Revista Calitatea vieții, XX, nr. 3–4, București
3. Constantinescu A.C. și col. 1993- *Starea factorilor de mediu în județul Iași – privire critică*. Revista „Omul și mediul înconjurător”, Iași.
4. Sandu Tatiana, Hovîrneanu Cristi-Irina, Bernardis R., 2003 - *Studiu privind situația spațiilor verzi în municipiul Iași și posibilitățile de îmbunătățire a stării acestora*, Lucrări Științifice Vol. 46, Seria Horticultură, Iași.
5. Sandu Tatiana, Trofin A.E., 2009 - *Accesibilitatea și raza de deservire a celor mai semnificative spații verzi cu acces nelimitat din cadrul municipiului Iași*. Lucrări Științifice. Vol. 52, Seria Horticultură U.S.A.M.V. Iași.

## STUDIES REGARDING THE BEHAVIOR OF SOME SPECIES OF *SEDUM* UNDER THE STRESS CONDITIONS INDUCED BY ROOF CULTURE

### STUDII PRIVIND COMPORTAREA UNOR SPECII DE *SEDUM* LA CONDIȚIILE DE STRES INDUSE DE CULTURA PE ACOPERIȘ

NEGREA Roxana<sup>1</sup>

e-mail: roxana.acfrance@gmail.com

**Abstract.** The principles of green roof landscaping, essential in developing cultivating projects, can be successfully applied knowing the full characteristics of the species that are used. Taking this into consideration, the main objective of this research is to enrich the assortment of ornamental used in the landscaping of the green roofs with new species, resilient to the stress caused by the existing cultures present on roofs. The biological material used was composed of 108 mature specimens of *Sedum spurium* 'Fuldaglut' and *Sedum reflexum* 'Angelina' cultivated in 3 types of substrate in containers, placed on the roof. Analyses carried out in the experiment have shown the ability to adapt to the stress conditions of the two species of *Sedum* by determining peroxidase and catalase activity. As a result of the study performed we were able to determine which of the two species is more adaptable to the roof culture, and also the adequate substrate for elaborating the cultivating schemes.

**Key words:** *Sedum*, roof culture, substrate, stress.

**Rezumat.** Principiile amenajării acoperișurilor verzi, esențiale în elaborarea schemelor de plantare, pot fi aplicate cu succes doar cu condiția cunoașterii în amănunt a caracteristicilor speciilor care intră în compoziția lor. Luând în calcul aceste considerente, obiectivul central care face subiectul acestei lucrări este acela de a îmbogăți sortimentul de plante ornamentale utilizate în amenajarea acoperișurilor înverzite cu specii rezistente la stresul datorat condițiilor de cultură existente la nivelul acoperișurilor. Materialul biologic utilizat a fost format din câte 108 exemplare mature de *Sedum spurium* 'Fuldaglut' și *Sedum reflexum* 'Angelina' cultivate în containere amplasate pe acoperiș, în trei tipuri de substrat. Analizele efectuate în cadrul experimentului au evidențiat capacitatea de adaptare la condițiile de stres a celor două specii de *Sedum* prin determinarea activității peroxidazei și a catalazei. În urma studiului efectuat s-a putut determina care din cele două specii este mai pretabilă culturii pe acoperiș, cât și substratul cel mai adecvat pentru elaborarea schemelor de plantare.

**Cuvinte cheie:** *Sedum*, cultură pe acoperiș, substrat, stres.

## INTRODUCTION

Growing flower species on rooftops has encouraging advantages in terms of environment, education system and community life, boosting the populations solidarity in order to achieve a framework for proper long-term management of

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iași, Romania

premises. As a result, traditionally, gardens and green areas in Romania, have long been planted with numerous species of rustic plants, some of them from the spontaneous flora, but with high decorative value (Zaharia *et al.*, 2013), which determined them to be later on "adopted" and frequently cultivated on roofs, or in certain areas (corresponding to the specific habitat of the plants), or wide spread, in the case of those with greater adaptability or which are commonly found in the spontaneous flora.

These qualities of flower species, especially of those belonging to the *Sedum* genus, decorative due to the beauty of the flowers and leaves, contribute to increasing the ornamental quantity of towns, as they can be used both for outfitting ground level spaces and in compositions successful carried out for roof gardens. This combination of business with pleasure is encouraged and highly developed in several US cities, Canada and Australia. A major importance in the sustainable maintenance of this algorithm is to maintain these plantations in the best possible condition at roof level by identifying and reducing the stress they face as much as possible (Haggas, 2006).

One of the reasons why not every plant may be used for growth on rooftops is hydric stress (Emilsson, 2008), which has a pronounced influence on the metabolism of phosphorus compounds in leaf tissue, inducing reduction of their content, especially of etheric carbohydrates during vegetative phenophases (Iordănescu, 1988). Despite this reason, there are many examples of plants cherished and cultivated in Romanian gardens, who because of ability to adapt can be used successfully in the creation of green roof: *Sempervivum tectorum* L., *Arabis caucasica* L. *Vinca major* L. (Negrea *et al.*, 2014). These species are among those with high ecological plasticity, which can be grown in the most various climatic conditions (Haggas, 2006). This feature is due to the large adaptability to environmental conditions in which they formed - from open places with strong insolation and severe humidity (Stefan *et al.*, 2013), to shady forests with moist soils.

In order to have an answer as accurately as possible about the emergence of the phenomenon of stress, manifested due to the culture medium from the roof (Nagase and Dunnett, 2011), we have to correlate the biochemical processes with the structural aspects, the lack of a fair enzyme activity is both the result of an imbalance and an adverse biochemical reactions (Weisany *et al.*, 2012). The whole complex of biochemical factors, substances responsible for upsets due to both environment and culture due to various substrates, is an important means of assessing the incompatibility between species from the experiment and different types of substrate.

Reported to the particular ecopedological conditions on which flowering species are grown, not always the most favorable, the culture site is the main way to fight the limiting factors of the soil (Levitt, 1980).

## MATERIAL AND METHOD

The biological material used was composed of *Sedum spurium* 'Fuldaglut' and *Sedum reflexum* 'Angelina', both purchased in pots of 12 cm from a specialized nursery. *S. spurium* 'Fuldaglut' is a vigorous perennial species, 10-15 cm tall with semi persistent

leaves, with whole or slightly serrated edges and pink or fuchsia star-shaped flowers (Zaharia, 2010). *S. reflexum* 'Angelina' is also perennial, 5 to 10 cm in height, with succulent and 1-2 cm long leaves, light-green mixed with yellow flowers, hermaphrodite, with 5 sepals and 5 petals, simple, succulent, grouped in inflorescences. The literature shows that fruits are polifollicles (Smydo, 2006), but none of the studied specimens, did not produce fruits the first year of planting.

The 216 mature uniform vegetal material (108 specimens for each species) has been transplanted in the spring of 2014 on the roof of a building belonging to the University of Agricultural Sciences and Veterinary Medicine of Iasi (fig.1.), by mounting them in 18 containers of 80/480 cm. In order to also make a comparative analysis between their development, the experimental scheme for each species includes three variants (type of culture substrates), with four repetitions each and 9 plants per repetition:  $V_1$  (control) - forest soil,  $V_2$  - mixture of Novobalt peat (43%), coconut fiber (30%), composted bark (23%), alginate (4%),  $V_3$  - blonde peat (40%), brown peat (30%), sand (10%) and forest soil (20%).

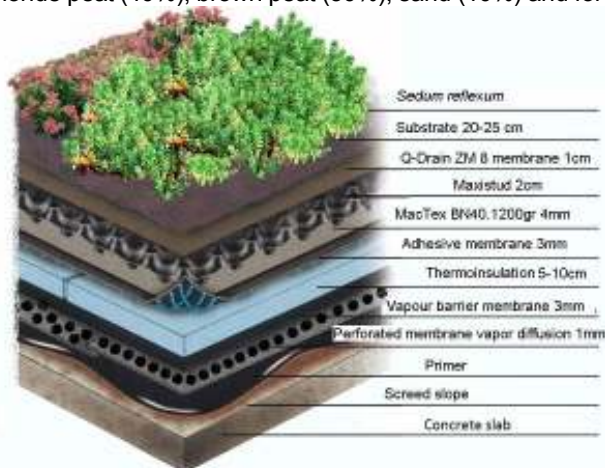


Fig. 1 - Section through roof-mounted alveoli

The overall objective of the research pursued in this paper was to conduct biochemical research on the plant - growth substrate association, to highlight the expression of adaptability phenomenon to various substrate. The research is focused on catalase and peroxidase activity, as indicators of adaptability to stress induced by specific roof conditions (drought, sunstroke etc.).

Peroxidase activity (POD) was then determined according to the method described by Brad et al. (Iordanescu and Dumitru, 1988) in which the reaction medium contains 0.1 mL ascorbic acid, 1 mL of distilled water, 2 mL and 1 mL prepared benzidine reactive protein. The method is based on measuring the color intensity of the product of oxidation with hydrogen peroxide under the action of peroxidase. To determine the activity of catalase (CAT) iodometric titration method was used. The hydrogen peroxide left decomposed after a certain time of incubation, oxidizes the potassium iodide.

## RESULTS AND DISCUSSIONS

The opportunity of the study is achieved by increasing green areas in urban landscape in terms of continuous demographic growth. Landscape architecture, being directly related to ensuring the ecological balance of the environment, has as main

objective the preservation and development of landscapes and their associated values for the benefit of current and future generations. The scientific research addressed in this paper focused on some plants with ornamental characteristics, recording the way they adapt and develop in the agro-meteorological Iasi area, with the purpose of including them on green rooftop culture (fig. 2).

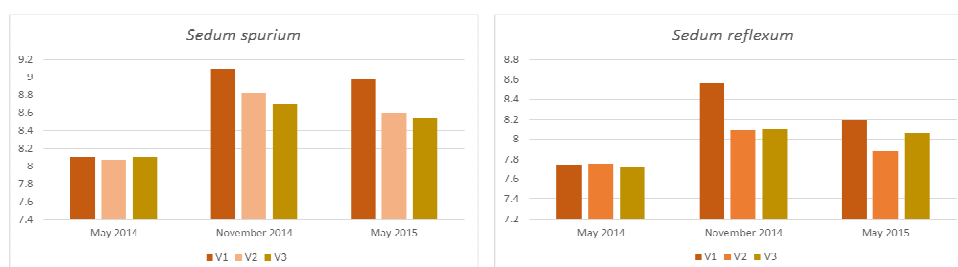


**Fig. 2** - *Sedum spurium* 'Fuldaglut' (a) and *Sedum reflexum* 'Angelina' (b) at the moment of planting

The determination of the enzymatic activity is a practical way to assess the level of stress, being used as a method of evaluating the ability of the plant to respond to this stress factor induced by the type of substrate. By studying the manner of their adaptability some modifications have been observed in enzyme activity.

Peroxidase activity average values determined two weeks after establishing the experimental crop was 8.09 U/mg protein for *Sedum spurium* and 7.75 U/mg protein for *Sedum reflexum* (fig. 3).

Following the measurements performed for the species *Sedum spurium*, the highest values of POD, were obtained from the control variant  $V_1$  situated on the substrate made from the forest soil, both in November and in May (9.10 respectively 8.98 U/mg protein), showing the stress faced by them. Although the differences are small compared to the control (by 3.4-5.3%), the lowest value of POD was recorded in plants located on the substrates  $V_2$  and  $V_3$ , which is a clear indication of the fact that this types of substrate reduces stress (fig. 3).



**Fig. 3** - Peroxidase activity (POD) during a year of vegetation (U/mg protein)

In the case of the *Sedum reflexum* species, by analyzing the data concerning POD activity it could be observed that the plants located in the substrate consisting of forest soil from variant  $V_1$  manifested the highest stress in November reaching the value of 8.57 U/mg protein, while the substrates of the embodiments  $V_2$  and  $V_3$  peroxidase activity decreased by 5.7-5.9% compared to the control (fig. 3). In May, peroxidase activity decreases for all variants, but maintained higher values for  $V_1$ .

Two weeks after planting, catalase activity (CAT) determined for the two species recorded close relative values, ranging from 4.38 U/mg protein for *Sedum spurium* and 4.82 U/mg protein for *Sedum reflexum*, in the three variants of substrate (fig. 4).

Tests carried in November 2014 and May 2015 indicate different values for catalase activity, depending on the variant and species (fig. 4). Both the *Sedum spurium* and the *Sedum reflexum*, the lowest catalase activity was recorded in the variants  $V_2$  and  $V_3$ . In May 2015 the values of CAT were similar for the three variants (between 4.69-4.77 U/mg protein for *Sedum spurium* and 5.11-5.25 U/mg protein for *Sedum reflexum*). In all measurements, variant  $V_1$  recorded the highest values of catalase activity (fig. 4).

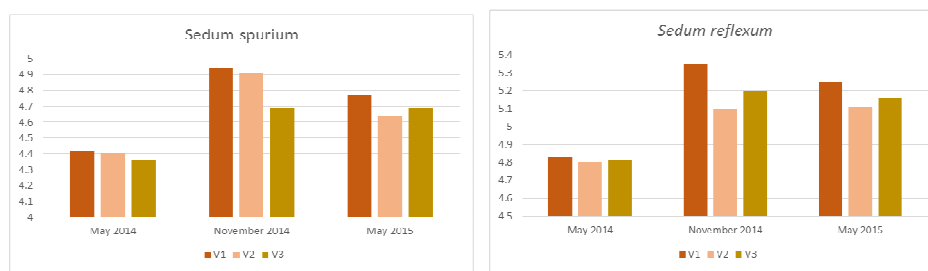


Fig. 4 - Catalase activity (CAT) during a year of vegetation (U/mg protein)

The analyzes and the tests carried out on the two investigated species showed a more intense enzyme activity in the case of plants located on the substrate consisting of forest soil ( $V_1$ ).

Knowing that the intensification of catalase activity is conditioned by increasing the quantity of hydrogen peroxide accumulated in tissues, a specific phenomenon under stress conditions, the results show the continued existence of adaptation processes in plants, given the fact that this enzyme is involved in the mechanism of adaptation of the plants to drought.

## CONCLUSIONS

In terms of rooftop culture the plants are subject to additional factors stress, since values of peroxidase and catalase activity registered immediately after planting material on the rooftop, in the first decade of May 2014, are lower than those recorded after 6 respectively 12 months after planting (November 2014 and May 2015).



The relatively small differences registered between the variants indicate little influence of substrate type on adaptation to growing conditions on the roof of *Sedum spurium* 'Fuldaglut' and *Sedum reflexum* 'Angelina'. However, lower levels of peroxidase and catalase activity of the variants V<sub>2</sub> and V<sub>3</sub>, we recommend the substrates consisting of Novobalt peat (43%), coconut fiber (30%), composted bark (23%), alginate (4%) and those composed of blonde peat (40%), brown peat (30%), sand (10%) and forest soil (20%).

**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*

## REFERENCES

1. **Emilsson T., 2008** - *Vegetation development on extensive vegetated green roofs: Influence of substrate composition, establishment method and species mix*, Ecological Engineering, 33 (3) : 265–277.
2. **Haggas C., 2006** - *Green Roof Plants: A Resource and Planting Guide*, The Booklist, ISSN 00067385, 10/2006, Volume 103, Issue 3, p. 14.
3. **Iordănescu D., Dumitru I.F., 1988** – *Biochimie practică*, Editura Didactică și Pedagogică, București.
4. **Levitt J., 1980** - *Responses of plant to environmental stresses* // New York, London: Acad. Press. Vol. 2. pp. 607-621;
5. **Nagase Ayako, Dunnett N., 2011** - *The relationship between percentage of organicmatter in substrate and plant growth in extensive green roofs*, Landscape and Urban Planning, ISSN 0169-2046, 2011, Volume 103, Issue 2, pp. 230 - 236.
6. **Negrea Roxana, Draghia Lucia, Ciobotari G., 2014** - *The influence of some culture systems on the ornamental value of Sedum spurium 'Fuldaglut' and Sempervivum tectorum species*. Lucrări Științifice, Seria Horticultură, Ed "Ion Ionescu de la Brad" Iasi, Vol. 57, Nr. 1, pp. 217.
7. **Stefan M., Munteanu N., Stoleru V., Mihasan M., 2013** - *Effects of inoculation with plant growth promoting rhizobacteria on photosynthesis, antioxidant status and yield of runner bean*. Romanian Biotechnological Letters Vol. 18, No.2, București, pp. 8132-8144.
8. **Smydo J., 2006** - *6,000 Sedum plants green up roof of refurbished building*, Pittsburgh Post –Gazette, ISSN 1068-624X, 10/01/2006, pag. B.1.
9. **Zaharia A., 2010** – *Cercetări privind stabilirea valorilor optime ale factorilor de mediu șitehnologici în procesul de înmulțire și cultură în containere a speciilor din genurile Sedum și Sempervivum folosite în amenajarea spațiilor*, Teză de doctorat U.S.A.M.V Cluj- Napoca;
10. **Zaharia Alina, Draghia Lucia, Cârstea Oana-Mariana, Chelariu Elena-Liliana 2013** – *Studies regarding the behavior of same wild ornamental species introduced in culture in pedoclimatic conditions from the Iași area*, Lucrări științifice USAMV Iasi, Vol. 56, Nr. 2, Seria Horticultură, pp. 251-256;
11. **Weisany W., Sohrabi Y., Heidari G., Siosemardeh Adel, Ghassemi Golezani K., 2012** - *Changes in antioxidant enzymes activity and plant performance by salinity stress and zinc application in soybean (Glycine max L.)*. Plant Omics Jurnal, Iran ISSN:1836-3644, pp. 60-65.



## INFLUENCE OF HYDRAULIC CONNECTION OF LAKE – SEA SYSTEM ON FLORA AND FAUNA OF THE COASTAL LAKES, AREA "SŁOWINSKI NATIONAL PARK", POLAND

### INFLUENȚA CONEXIUNII HIDRAULICE LAC –MARE ASUPRA FAUNEI ȘI FLOREI DIN LACURILE DE COASTĂ, ZONA „PARCULUI NAȚIONAL SŁOWIŃSKI”, POLONIA

BĂLAN Anca<sup>1</sup>, LUCA M.<sup>1</sup>, AVRAM M.<sup>1</sup>

e-mail: anca\_balan2003@yahoo.com

**Abstract.** *This paper presents studies and research on spatial hydrotechnics Słowiński National Park area in order to protect the environment. The main goal of the research in this paper is to realize the importance of undertaking regeneration work in small river valleys, in the section of the Baltic Sea coast and to increase current knowledge about the importance of ecohidologic operating conditions of these ecosystems. The two lakes coastal studied have different levels of communication with the sea and the various morphometric parameters. Creating environmental conditions in the two areas located to the existing fauna and flora by limiting lake-sea connection.*

**Key words:** *community site, conection hydrologic, rehabilitation, regularization works*

**Rezumat.** *Lucrarea prezintă studiile și cercetările privind amenajarea hidrotehnică a zonei Parcului Național Słowiński, Polonia în scopul protecției mediului prin renaturalizarea brațelor de râu. Scopul principal al cercetărilor, în prezenta lucrare este de a realiza importanța efectuării lucrărilor de regenerare, în văile râurilor mici, în secțiunea de coasta a Mării Baltice precum și de a crește stadiul actual al cunoștințelor despre importanța condițiilor ecohidrologice de funcționare a acestor ecosisteme. În cadrul acestor două lacuri se aplică diverse tehnologii biologice pentru realizarea și dezvoltarea unui habitat specific sitului natural.*

**Cuvinte cheie:** *situri naturale, conexiune hidrologică, reabilitare, lucrări de regularizare*

## INTRODUCTION

The coastline of the South Baltic Sea is rich in numerous and diverse bodies of water hydrology. These include lakes, coastal lakes genetic belonging. Baltic is located in the temperate climate zone.

Each of these lakes varies considerably from one another on biological and morphometric parameters. The differences are associated with the straight line distance to the sea Lake basin and the intensity of wave power caused the sea surface, morphometric parameters, the coefficient of exhibition, water quality and

---

<sup>1</sup> "Gheorghe Asachi" Technical University of Iași, România

water level in the lake basin. (Cieśliński, 2004; Trojanowski, 1990; Trojanowski *et al.*, 1991)



**Fig. 1** - Coastal estuaries of the Polish Baltic Sea

Polish Baltic Sea coast has a number of smaller or larger lakes such as Gardno, Łebsko, DOLGA Wielkie (lakes located in the Słowiński National Park) and Jamno, Kopan, Wicko, Sarbsko, Resko, Koprowo (outside the park). In spite of similar areas of location, they vary in type and geomorphological genesis, both in terms of hydrological conditions and the hydrochemical (Paturej, 2006).

## MATERIAL AND METHOD

Old river arms are analyzed lakes formed by meandering river or sea that have been closed. They can be supplied and drained in certain circumstances the hydrological regime. Structure and functioning of wetland ecosystems, including lakes is achieved in close interconnection directly or indirectly with a water source or an emissary. This interconnection is closely linked to fluctuations of water levels in source or emissary during floods or flow pulsations. (Junk *et al.*, 1989, Tochen *et al.*, 2000).

Hydrological processes involve changes in the chemical composition of water from lakes and its migration to other areas due to interchange water - sediment.

### Lake Gardno

Gardno Lake is located in the middle part of the Polish coast and is part of Słowiński National Park. Considering this is the second lake area coastal lagoons compared to Poland.

Lake Gardno is connected to the Baltic Sea from Łupawa channel with a length of 1500 m. It is separated by the sea water by a sand barrier it has a width of about 800-2000 (Trojanowski, 2003b).

Łupawa Channel and several smaller rivers: Bagiennica, Grabownica, Brodniczka and Brodna lakes supplying with water the lake Gardno. Total river inflow to the lake was estimated at 9.07 m s<sup>-1</sup>. In conclusion, Łupawa River provides about 90% of the total inflow of surface water from the basin, represented by about 286 • 10<sup>6</sup> m<sup>3</sup> annually. (2468.1 ha Olsztyn IRS 1966).

Gardno lakes are shallow, with average depth about 1.3 m (maximum depth 2.6 m). The lake covers an area of approximately 25 km<sup>2</sup> large. Emergency macroflora relates to 4% of the area of the lake.

The lake has two interconnections type Gardno lake - river. The first interconnection is Łupawa River in the east, south protected by a small pier.

This resulted in a moderate decline in river levels with damage and loss of habitat for many species of flora and fauna of value. The habitat was affected including floodplain (floodplain of the river).



**Fig. 2** - Location of Gardno lake: a - the location of sampling sites.

SOURCE: [www.national-geographic.pl](http://www.national-geographic.pl)



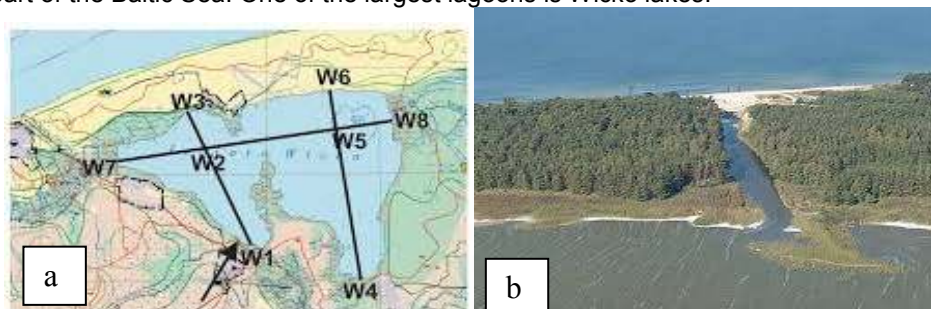
**Fig. 3** - View of channel Łupawa - spill in the Baltic Sea

#### Wicko lake

Locating the lake is in Słupsk Pomeranian Region near town Jarosławiec. Wicko lake is a lake who has two connections coastal hydrology.

The first connection is with the Baltic Sea by a canal and the second in the west is connected via a canal to Kopan lake regeneration. Wicko lakes are triangular with an area of 1059 hectares.

A number of coastal lagoons and lakes are located along the entire southern part of the Baltic Sea. One of the largest lagoons is Wicko lakes.



**Fig. 4** - Location of Wicko lake: a - the location of sampling sites .b - general view of the Baltic Sea connection. SOURCE: [www.national-geographic.pl](http://www.national-geographic.pl)

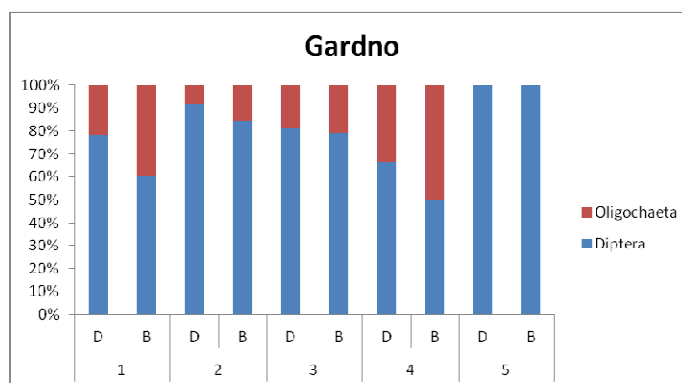
Ecological improvement of surface water must meet and maintain a normal relationship with the aquatic flora and fauna.

In the first phase were carried out research on the habitat condition affected areas without applying the regulation work to achieve interconnectivity.

## RESULTS AND DISCUSSIONS

Achieving regulation work for ensuring the interconnectivity with power sources and lakes with emissaries were allowed providing the conditions for growth of the habitat. The regularization of ecological achieved were integrated very well into the aquatic environment.

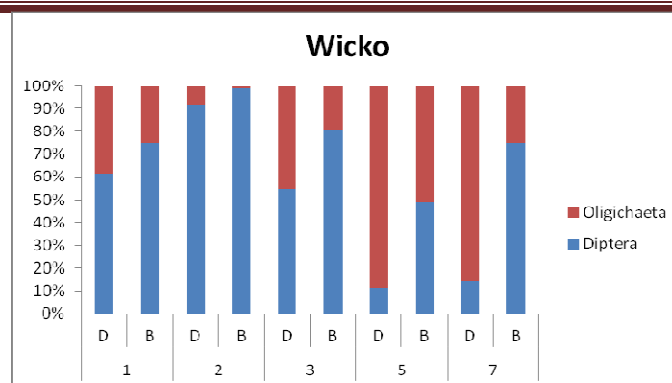
A first effect of achieving inter-connectivity was changing habitat conditions. This change is represented by changing parameters of water quality in lakes. A first result is the development of a number of existing species. The result of the analysis of samples of biological material taken from the field and analyzed in the laboratory for the Gardno lake are shown in Fig. 5.



**Fig. 5** - The structure of macrozoobenthos regarding the density and biomass in Gardno lakes

Following laboratory analysis was determined the following: sample 1, Plateaus Diptera density is 80% and 20% Oligochaeta; Biomass Diptera is 60% and Oligochaeta 40%. In sample 2, the density of the Diptera is 90% and 10% Oligochaeta; Biomass Diptera is 80% and 20% Oligochaeta. In sample 3, the density of the Diptera is 80% and 20% Oligochaeta; Diptera biomass is 78% and 22% Oligochaeta. In sample 4, the density of the Diptera is 70% and 30% Oligochaeta; Diptera biomass is 50% and 50% Oligochaeta. In the sample 5 and the biomass density of the Diptera is 100%.

The result of the analysis of samples of biological material taken from the field and analyzed in the laboratory for the Wicko lake are shown in Fig. 6



**Fig. 6** - The structure of macrozoobenthos regarding the density and biomass in Wicko lakes

In coastal lakes close to the Baltic Sea (Gardno, Wicko) was found by field and laboratory research densely Diptera and Oligochaeta (Fig. 5, 6, Table 1). They have adapted to the environment in lakes and have proved to be more tolerant with water that has a higher salinity.

Table 1

**Density of macrobenthos in the area studied indv/m<sup>2</sup>**

Taxon	Gardno	Wicko
<i>Diptera</i>	3,795.56	654.81
<i>Oligochaeta</i>	506.67	823.70
<i>Hirudinea</i>	-	-
<i>Crustacea</i>	-	-
<i>Gastropoda</i>	-	-
<i>Bivalvia</i>	-	-
<i>Trichoptera</i>	-	-
<i>Ephemeroptera</i>	-	-

Research has shown that Gammarus sp. It is affected by chemical quality of the water in both lakes. Degradation of habitat mainly influences the aerobic conditions. Mention that Gammarus sp. is the basic nutrition for migratory fish. It follows in this context the importance of achieving interconnectivity work and regularization of the riverbeds and lakes.

## CONCLUSIONS

1. Ensure optimal conditions for live in natural sites imposes the making a work regulation of lakes, rivers and canals. Between lakes must be provided a interconnectivity with power supply and emissary and adjusting hydraulic parameters.

2. The two lakes studied (Gardno, Wiczo) are supplied with brine who the life parameters degrading the life parameters of habitat and ecological environment.

## REFERENCES

1. **Amoros C., Bornette G., 2002** - *Connectivity and biocomplexity in waterbodies of riverine floodplains*. Freshwat. Biol., 47: 761-776.
2. **Bajkiewicz-Grabowska E., D. Borowiak, 2000** - *Anthropogenic and natural transformations of lakes*, vol. 2. (Eds), , KLUG-PTLim, Gdańsk, 67-70.
3. **Boulton A.J., Peterson C.G., Grimm N.B., Fisher S.G., 1992** - *Stability of an aquatic macroinvertebrate community in a multiyear hydrologic disturbance regime*. Ecology, 73: 2192-2207.
4. **Cieśliński R., 2003** - *Lake Łebsko in the light of current hydrochemical research*, *Limnological Review*, volume 3/2003, Institute of Geography of the Jan Kochanowski University, Kielce, 47–52.
5. **Corus C.P., 2001** - *Piling Handbook*, Internet Edition.
6. **Gibbins C.N, Dilks C.F, Malcolm R., Soulsby C., Juggins S., 2001** - *Invertebrate communities and hydrological variation in Cairngorm mountain streams*. Hydrobiologia, 462: 205-219.
7. **Jarosiewicz A., Ficek D., 2008** - *Seasonal nutrient dynamic in Łebsko lakes in mid-lake zone of the coastal Lake Łebsko*, Pol. Arch. Hydrobiol. 46: 257-275
8. **Junk W.J., Bayley P.B., Sparks R.E., 1989** - *The flood pulse concept in river-floodplain systems*. In Dodge, D. P. (eds). Canadian Special Publications of Fisheries and Aquatic Science, Proceedings of the International Large River Symposium, pp. 110-121.
9. **Luca M., Bălan Anca, Tamașanu F. , 2014** - Technical University "Gheorghe Asachi" of Iasi, Romania, *Restorat on and wathershed of lezer lake*, commune Sadova. (*Reabilitarea și amenajarea Lacului lezer, comuna Sadova. Proiect tehnic*) Tehnical Project, SC Polias-Instal Iași.

## ORGANOPHOSPHORUS PESTICIDE RESIDUES FROM SOIL AND CUCUMBER FRUITS

### REZIDUURILE DE PESTICIDE ORGANOFOFORICE DIN SOL ȘI FRUCTE LA O CULTURĂ DE CASTRAVEȚI

**BUHĂIANU Bianca<sup>1</sup>, MUNTEANU N.<sup>1</sup>, HURA Carmen<sup>2</sup>,  
GALEA (DELEANU) Florina-Maria<sup>1</sup>, STOLERU V.<sup>1\*</sup>**

**e-mail:** vstoleru@uaiasi.ro

**Abstract:** This paper presents a study on the content of organophosphorus pesticides (OPPs) in a cucumber crop, in two different cultivation systems, conventional (intensive) of Tg. Frumos area and "V. Adamachi" organic farm that belongs of UASVM Iasi. The aim of this study is to evaluate whether the content of OPPs correlate with cultivation system applied in poly-tunnels. OPPs residues were analyzed by atomic absorption spectrophotometry on Shimadzu GC-2100. As a result of analysis, in conventional farm were detected in soil and fruits residue of pesticides as: Omethoate, Phorate and Phosmet. Pesticide contents were within the maximum allowed by EC Regulation no. 396/2005.

**Key words:** Cucumis sativus, insecticides, contaminants, crop

**Rezumat:** Lucrarea de față prezintă un studiu cu privire la conținutul de pesticide organofosforice dintr-o cultură de castraveți, din două sisteme diferite de cultivare, convențional (intens chimizat) din zona Tg. Frumos și organic, din ferma "V. Adamachi", ce apartine USAMV Iasi. Scopul definit al acestui studiu este de a evalua măsura în care conținutul de pesticide organofosforice se corelează cu sistemul de cultivare aplicat în solarii. Determinarea reziduurilor de pesticide organofosforice s-a efectuat prin spectrofotometrie cu absorbție atomică, utilizând echipamentul Shimadzu GC-2100. În urma efectuării analizelor, au fost detectate în ferma convențională, atât în sol, cât și în legume următoarele reziduuri de pesticide: Omethoate, Phorate și Phosmet. Conținuturile de pesticide s-au încadrat în limita maximă admisă de Regulamentul CE nr. 396/2005.

**Cuvinte cheie:** Cucumis sativus, insecticide, contaminanți, cultură

## INTRODUCTION

Over the years and with the development of fast growing industry, and increase the number of population, the need to produce much food for its necessary. This has led to an intensive chemical treatment of crops, which with time has produced a massive land degradation (Munteanu *et al.*, 2010; Stoleru *et al.*, 2013). That makes to the accumulated of organic and inorganic contaminants in soils, which can react with each other thus producing new compounds, more

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

<sup>2</sup> National Institute for Public Health/Regional Center, Iasi, Romania



hazardous to human health, but also the flora and fauna which affected the environment (Stoleru, 2013; Dumitrescu *et al.*, 1998; Pădurariu, 2011).

Due to excessive soil degradation and the need increasing more food, the idea of organic farming is increasingly appreciated and "embraced" by producers and the number of organic farms increased from year to year (Stoleru, 2013; Munteanu *et al.*, 2010).

The using of pesticides in agriculture, most often irrational cause pollution of soil and products, on the extensive areas.

## MATERIAL AND METHOD

Research carried out in "V. Adamachi" farm from Iași (organic farm L1), and A.F. Maxim from Tg. Frumos (conventional farm L2), desiring thus, analyzes comparing in both systems of cultivation on the content of organophosphorus pesticides (Fig.1).



**Fig.1** The area of study UASVM Iași (L<sub>1</sub>) and AF Maxim Tg. Frumos (L<sub>2</sub>)

During to 2014 have been analysed the content for 23 OPPs (Mevinphos, Molinate, Heptenophos, Omethoate, Naled, Monocrothopos, Phorate, Fonofos, Metribuzin, Parathion-methyl, Metalaxyl, Malathion, Fenthion, Parathion-Ethyl, Isofenphos, Mecarbam, Penthoate, Myclobutanil, Fensulfothion, Phosmet, Phosalone, Demeton-S-Methyl-sulfoxide) on 20 soil samples and 10 samples of vegetables from the two farms (Tabel 1).

Sampling was carried in two phases: • Phase I - before planting (10-11 March);

- Phase II a – after the last harvest (9-10 August).

*Table 1*

Soil and fruit code samples				
Farm type	Location	No of samples	Soil code sample	Fruit code sample
<b>Organic</b>	UASVM Iași , Ph I	5	S <sub>1</sub> , S <sub>2</sub> , S <sub>3</sub> , S <sub>4</sub> , S <sub>5</sub>	C <sub>1</sub> , C <sub>2</sub> , C <sub>3</sub> , C <sub>4</sub> , C <sub>5</sub>
	UASVM Iași , Ph II	5	S <sub>11</sub> , S <sub>12</sub> , S <sub>13</sub> , S <sub>14</sub> , S <sub>15</sub>	
<b>Convențional</b>	A.F. Maxim , Ph I	5	S <sub>6</sub> , S <sub>7</sub> , S <sub>8</sub> , S <sub>9</sub> , S <sub>10</sub>	C <sub>1</sub> , C <sub>2</sub> , C <sub>3</sub> , C <sub>4</sub> , C <sub>5</sub>
	A.F. Maxim , Ph II	5	S <sub>16</sub> , S <sub>17</sub> , S <sub>18</sub> , S <sub>19</sub> , S <sub>20</sub>	

In the ecological system were applied differentiated fertilization with organic products (Orgevit-1500 kg /ha) and microbiological (Micoseed plus – 120 kg/ha), and for conventional farm was applied soluble fertilizer with Nutrispore, amount of 900 kg/ha, applied 10 by 10 days, started with bed preparation.

Biological material was represented by hybrid Artist, from Bejo Seed.

Analyses were carried out on samples of soil and fruit as well.

The method used in the research laboratory at the Sanitary Veterinary Iasi and determination of organophosphorus pesticides was performed by means of gas chromatography (GC) - Shimadzu 2010 equipped with detectors NPD and ECD.

## RESULTS AND DISCUSSION

As a result of the yield determinations, the highest number of fruit/plant (fr/pl) was in version fertilized with Orgevit (26 fr/pl), and those that were fertilized with Micoseed had a lower number of fruit/plant, respectively 17 fr/pl (Table 2)

Fruit weight is other important factor for the cucumber production. From this point of view have been remarked plants fertilised with Nutrispore (129,26 g/fr.).

Table 2

**Dynamics of fruits/plant and weight of fruit of cucumbers**

Experimenteal version	Fruits/plant	Weight of fruit (g)	Weight fruits/plant (kg)
Artist F1 x Orgevit	26	113,78	2,933
Artist F1 x Micoseed	18	129,03	2,238
Artist F1 x Nutrispore	17	129,26	2,326
Artist F1 x Control	16	111,27	1,779

In the table 3 are present the total yield till to 10 August, before the end of crop. Cucumber yield varied from 39,529 t/ha up to 65,171 t/ha.

Table 3

**Total yield of cucumber (2014)**

No.	Experimental version	Yield (t/ha)	Relatively yield (%)	Differeneces to Control and significance
1.	Artist F1 x Orgevit	65.171	164,87	25,642 <sup>***</sup>
2.	Artist F1 x Micoseed	49.728	125,80	10,199 <sup>*</sup>
3.	Artist F1 x Nutrispore	51.683	130,75	12,154 <sup>**</sup>
4.	Artist F1 x Control	39.529	100,00	0,00 <sup>*</sup>

LSD 5%=8.231 t/ha; LSD 1%=12.004 t/ha; LSD 0.1%=16.5151 t/ha;

**The content of organophosphorus pesticides from soil**

The results of the analysis of the OPPs content from soil samples collected during phase I, are shown in Table 4.

From the analysis carried out on the soils samples from V. Adamachi farm was not detected any type of OPPs. V Adamachi explore organic farming since 2006 and does not use synthetic chemical pesticides.

Results for soil samples from Tg. Frumos farm highlight the contents of Omethoate between 0.01mg/kg (S<sub>6</sub>, S<sub>8</sub>, S<sub>9</sub>) and 0.02 in S<sub>10</sub>. Also, have been found traces of Phorate in quantities of 0.001 mg/kg for S<sub>7</sub> and S<sub>9</sub>, 0.002 mg/kg for S<sub>6</sub> and S<sub>10</sub> and 0.003 mg/kg for the sample S<sub>8</sub>, and Phosmet: 0.005 mg/kg in S<sub>6</sub> and S<sub>10</sub>; 0.006 mg/kg in S<sub>7</sub> and S<sub>9</sub> and 0.007 mg/kg in sample S<sub>8</sub>.

Table 4

**The content of organophosphorus pesticides in soil samples (phase I)**

Pesticide name	Soil code sample (mg/kg)						Maximum admitted limit (mg/kg)
	S <sub>1</sub> -S <sub>5</sub>	S <sub>6</sub>	S <sub>7</sub>	S <sub>8</sub>	S <sub>9</sub>	S <sub>10</sub>	
Omethoate	Nd	0,01	0,015	0,01	0,01	0,02	0,02
Phorate	Nd	0,002	0,001	0,003	0,001	0,002	0,1
Phosmet	Nd	0,005	0,006	0,007	0,006	0,005	0,05

nd\*= below the detection limit of the device

The OPPs residues found in soil samples because were used against the species of *Tuta absoluta* Mey. and *Tetranychus urticae*. Due to the large number of applications, OPPs have been accumulate in the soil and thus were found in soil samples analyzed, under MAL.

The results for soil samples collected from Phase II are shown in Table 5. OPPs were not detected in large numbers, or in quantities too important in phase-II, show that are metabolized and thus has an improvement of soil quality. The Omethoate found in much lower: in S<sub>16</sub> of 0.02 mg/kg, in S<sub>17</sub> was found 0.007 mg/kg, in S<sub>18</sub> and S<sub>19</sub> was 0.005 mg/kg, and in S<sub>20</sub> found 0.01 mg/kg.

Table 5

**The content of organophosphorus pesticides in soil samples (phase II)**

Pesticide name	Soil code sample (mg/kg)						Maximum admitted limit (mg/kg)
	S <sub>11</sub> -S <sub>15</sub>	S <sub>16</sub>	S <sub>17</sub>	S <sub>18</sub>	S <sub>19</sub>	S <sub>20</sub>	
Omethoate	nd	0,02	0,007	0,01	0,005	0,01	0,02
Phosmet	nd	0,01	0,004	0,002	0,007	0,005	0,05

The contents for Phosmet were much lower than first phase, so that the new contents are: S<sub>16</sub> and S<sub>19</sub> – 0.007 mg/kg, S<sub>17</sub> and S<sub>20</sub> – 0.005 mg/kg, and S<sub>18</sub> –

0,002 mg/kg. After soil analyzes taken in second step proved that their content according to MAL.

Regarding from sampling at the farm "V. Adamachi "this time too were not found residues of organophosphate pesticides.

### The content of organophosphorus pesticides in fruits

Same as soil samples collected at the farm "V. Adamachi ", in fruit samples were not detected residues of organophosphate pesticides (Table 6).

In contrast to the soil samples from the AF Maxim, in cucumber fruit were found organophosphorus pesticides in only two samples taken in the analysis.

In samples C<sub>6</sub> and C<sub>9</sub> have been found residues of Omethoate and Phosmet in quantity of 0.01 mg/kg. Regarding to the Phorate content, this pesticide did not found. Because were found only 0.01 mg/kg OPPs residues in fruit samples, shows that any where the fruit harvested from AF Maximum allowed MAL in the content of pesticides.

Table 6

The content of organophosphorus pesticides in fruit samples

Pesticide name	Fruit code sample (mg/kg)				Maximum admitted limit (mg/kg)
	C <sub>1</sub> .C <sub>5</sub>	C <sub>6</sub>	C <sub>9</sub>	C <sub>7</sub> , C <sub>8</sub> ,C <sub>10</sub>	
Omethoate	nd	0,01	0,01	nd	0,02
Phosmet	nd	0,01	0,01	nd	0,05

## CONCLUSIONS

The highest yield was achieved in the experimental version fertilized with Orgevit, respectively 65.171 t/ha.

In soil samples collected in two stages from Adamachi farm have not been detected residues of OPPs.

In the samples taken from the AF. Maxim , have been detected three OPPs of the 23 taken into consideration: Omethoate, Phorate and Phosmet. In one sample was reached MAL respectively in the S<sub>10</sub> sample (0.02 mg / kg).

OPPS residue in fruits highlighted as part of soil content passes in fruit, so were traces of Omethoate and Phosmet in quantities of 0.01 mg / kg. Products obtained in organic cucumbers not pose a threat to human health in terms of the levels of OPPs. For the AF Maxim must be requires closer monitoring for the MAL risk.

## REFERENCES

1. Ciofu R., Stan N., Popescu V., Chilon P., Apahidean S., Horgoș A., Berar B., Lauer F.K., Atanasiu R., 2004 -*Tratat de legumicultură*, Editura Ceres, București.

2. Dumitrescu M., Scurtu I., Stoian L., Glăman G., Costache M., Dițu D., Roman T., Lăcătuș V., Rădoi V., Vlad C., Zăgorean V., 1998 - *Producerea legumelor*, Editura Artprint, București.
3. Hura Carmen, 2006 - *Ghid de laborator – Metode de analiză pentru produse alimentare*, Editura tehnică, științifică și didactică Cermi, Iași.
4. Indrea D., Apahidean S.A., Măniuțiu D., Apahidean Maria, Sima Rodica, 2012 - *Cultura legumelor Ediția a-III-a*, Editura Ceres, București.
5. Munteanu N., Bireescu L., Bulgariu D., Hura Carmen, Stoian L., Stoleru V., 2010 - *Monografia producției legumicole ecologice din Nord-Estul României: Posibilități și riscuri*, Editura Arhip Art, Iași.
6. Munteanu N., Stoleru V., Stan T., Stoleru Carmen- Maria, Aldescu Teodora, 2007 - *The perspectives of organic agriculture in Romania*. Lucrări Științifice USAMV Iasi, seria Horticultură Vol. 50(1): 555-560.
7. Pădurariu Eugenia-Anca, 2011 - *Studiul principalilor factori de risc într-un sistem ecologic de producere a legumelor*. Teza de doctorat, U.S.A.M.V. Iași.
8. Stoleru V., 2013 - *Managementul sistemelor legumicole ecologice*, Editura "Ion Ionescu de la Brad", Iași.
9. Stoleru Vasile, Munteanu Neculai, Hura Carmen, 2015 - *Organophosphorus pesticide residues in soil and vegetable, through different growing systems*. Environmental Engineering and Management Journal, 14 (6).

## NONWOVENS USED IN HORTICULTURE – OPTIMIZATION OF NEEDLE PUNCHING PROCESS PARAMETERS

### NEȚESUTE FOLOSITE ÎN HORTICULTURĂ – OPTIMIZAREA PARAMETRILOR PROCESULUI DE INTERȚESERE

**LUPU Iuliana Gabriela<sup>1</sup>, GROSU M.C.<sup>1</sup>, HOGAȘ H.I.<sup>1</sup>**  
e-mail: iuliana68lupu@yahoo.com

**Abstract.** Textile fabrics have a long history of use in agriculture. The term “agrotexile” now is used to categorize the woven, nonwoven and knitted fabrics used for agricultural and horticultural applications. Among the major sectors and specific applications that are considered to have potentials for jute/synthetic nonwovens in agriculture/horticulture are frost protectors. The purpose of this paper is to investigate the influence of needling punching process parameters on functional properties of nonwovens used as frost protectors. The study was focused on the influence of needling density and needle gauge on jute/polypropylene nonwoven density and water vapour permeability by using a central, composite design for second-order. The results show that the process parameters have a significant influence on nonwoven characteristics. The higher of process parameters values, the higher is fabric density. A less porous nonwoven have a lower water vapour permeability which means that the agrotexile conserves water by reducing evaporation.

**Key words:** agrotexile, nonwoven, composite design

**Rezumat.** Textilele au o lungă istorie a utilizării lor în agricultură. Termenul de „agrotexil” este folosit pentru a defini textilele țesute, nețesute sau tricotate utilizate în aplicații agricole și horticole. Printre principalele sectoare și aplicații specifice ale agriculturii/horticulturii în care se pot utiliza nețesute din iută/fibre sintetice se numără materialele de protecție la îngheț. Scopul acestei lucrări este de a investiga influența parametrilor procesului de interțesere asupra proprietăților funcționale ale nețesutelor utilizate pentru protecție. Studiul s-a axat pe influența densității de interțesere și a fineții acelor asupra densității și a permeabilității la vapori de apă utilizând un model matematic central compus rotabil de ordinul doi. Rezultatele studiului au indicat faptul că parametrii procesului de interțesere au o influență semnificativă asupra caracteristicilor studiate. Odată cu creșterea valorilor optimizate crește densitatea nețesutului. Un nețesut mai puțin poros are o permeabilitate la vapori mai mică ceea ce înseamnă că agrotexilul conservă apa prin reducerea evaporării acesteia.

**Cuvinte cheie:** agrotexil, nețesut, model compus

## INTRODUCTION

Agrotexiles are nowadays extensively being used in horticulture, farming and other agricultural activities. Agrotexiles play a significant role to help control

---

<sup>1</sup> "Gheorghe Asachi" Technical University of Iași, România

environment for crop protection, reduce variations due to climate or weather change and therefore they can generate optimum condition for plant growth.

Spun bonding and needle punching techniques are widely used for the production of nonwoven agrotextiles. Needle punching is a process for converting webs of fibers into coherent fabric structures, generally by means of barbed needles, which produce mechanical bonds within the web (Purdy, 1980).

Among the major applications that are considered to have potentials for jute/synthetic nonwovens in agriculture are frost protectors. Nonwovens are generally used in order to enhance crop growth and production by increasing both air and soil temperatures, reducing the water evaporation and wind damage. A medium weight nonwoven will give from 2 to 6 degrees of frost protection (Olle *et.al.*, 2010).

In order to understand more about the influence of needling process parameters on nonwoven characteristics it is essential to use mathematical modeling which is an investigation method of technological processes based on experimental data collection and processing. The present study is investigating the effect of two parameters on the functional characteristics of nonwoven that can contribute to reducing the water evaporation and of course the costs with water, labour etc. For this purpose, a central composite design for second-order model has been employed.

## MATERIAL AND METHOD

A blend of jute (75%) and polypropylene (25%) fibers was used to prepare nonwoven samples. These raw materials were chosen due their environment-friendliness, widespread presence in the market and cheap price. The jute fibers used had an average length of 80 mm and fineness 27 dtex and the polypropylene fibers of 50 mm and 7.7 dtex.

Nonwoven samples were prepared on a pilot plant single needle-punching machine Automatex, equipped with universal card type CA 500, cross-lapper model MFA 600 and needle-punching loom type MPR 600. The basis weight of the webs of jute/polypropylene fibers formed by carding and lapping process was controlled as 220 g/m<sup>2</sup>. The webs were fed to the needling zone on a needle loom type Automatex having Groz-Beckert needles with two barbs on each edge and various finenesses (gauge) (for e.g. 15x18x3xC222 G3017). The needle-punched fabrics were produced by the penetrating action of barbed needles which reorientation and intermingles the fibers from a horizontal to a vertical direction (Rusell, 2007).

The experiments took place under pilot unit conditioned. Before performing the measurements, the samples were conditioned at 65%, relative humidity and 20°C temperature for 24 h.

Mass (W) was tested according to the standard ISO 9073-1:1989 Textiles – Test methods for nonwovens – Part 1, whereas thickness (T) was tested according to the standard for nonwoven textile ISO 9073-2:1995. Density of nonwoven textiles was calculated from mass per unit area and thickness values, using the following equation:

$$\rho_N = \frac{W}{T} \left[ \text{kg} / \text{m}^3 \right] \quad (1)$$

The water vapour permeability of the nonwoven samples was measured according to the standard ASTM E96. Six circular specimens, 6 cm in diameter, were cut from each fabric. A 145 ml aluminium cup was filled with distilled water, covered with the fabric specimen. The whole assembly was then weighed and reweighed after a certain



elapse of time (3, 24, 48 and 72 hours). The water vapour permeability (WVP) was calculated using the following formula:

$$WVP = \frac{G/t}{A} \quad (2)$$

where  $G$  = weight change (g),  $t$  = time during which  $G$  occurred (h) and  $A$  = test area ( $m^2$ ).

In order to study the influence of the needling-punching process parameters on fabric density and water vapour permeability a central composite surface factorial design of two variables was used (Lupu *et al.*, 2013, Taloi, 1987). The useful limits of the two variables used in the central composite design, i.e. needling density and needle gauge, were selected by conducting a number of preliminary experiments. The limits, actual and coded values of factors are given in Table 1.

Table 1

Actual and coded values of independent variables

Variable	Symbol	Code				
		-1.414	-1	0	1	+1.414
Needling density (punches/cm <sup>2</sup> )	$x_1$	60	73	105	137	150
Needle fineness (gauge)	$x_2$	30	32	36	40	42

## RESULTS AND DISCUSSIONS

The obtained matrix was a 13-points central composite design, which consisted of a full factorial design  $2^2$  (4) plus five center points and four star points. Thus, the 13 experimental samples allowed the estimation of the linear, quadratic and two-way interactive effects of various factors on nonwoven properties.

The developed matrix design, including the coded values of the factors and actual values of different samples and their properties are shown in Table 2.

To correlate the effect of independent variables and response, the following second-order standard polynomial was considered (Cojocaru *et al.*, 1968):

$$Y = b_0 + b_1x_1 + b_2x_2 + b_{11}x_1^2 + b_{22}x_2^2 + b_{12}x_1x_2 \quad (3)$$

where  $Y$  represents the response and  $b_0, b_1, \dots, b_{12}$  are the coefficients of the model.

The coefficients of main and interactive effects were determined using the standard method (Cojocaru *et al.*, 1968). The regression coefficients of the proposed model for different parameters can be calculated. To establish the relationship between the independent variables ( $x_1$ ) and ( $x_2$ ) and the dependent variable ( $Y$ ), a regression analysis was performed as describe above. The regression coefficients were used in the quadratic-proposed polynomial model (Table 3) to determine the predicted response values. These coefficients have either positive or negative value, and accordingly have an effect on the experimental results. The coefficients of  $R^2$  multiple correlation and the  $F$ -values together with the response surface equations of the factorial design for second-order models after testing the regression coefficients by employing the student test are shown in Table 4.

To check the significance of multiple correlation coefficients, we used the  $F$  test (Taloi, 1987). Accordingly,  $F$ -ratios were calculated for a 95% level of confidence and two degrees of freedom ( $f_1 = 2$  respectively,  $f_2 = 10$ ) and then compared with the

corresponding tabulated values. If the calculated values of  $F$ -ratios exceed the corresponding tabulated values, then the independent variables had a significant influence on the dependent variable. The tabulated value of the  $F$ -ratio at 95% level of confidence was found to be 4.1 (Cojocaru *et al.*, 1968).

Table 2

Constructional details of experimental fabrics

Sample No	Needling density ( $x_1$ )	Needle gauge ( $x_2$ )	Fabric density ( $\text{kg/m}^3$ )	Water vapour permeability ( $\text{g/m}^2\cdot\text{h}$ )
1	-1	-1	65.259	15,706
2	1	-1	60.741	17,331
3	-1	1	63.769	17,266
4	1	1	67.082	14,753
5	-1.414	0	63.043	16,855
6	1.414	0	69.494	17,115
7	0	-1.414	61.689	16,053
8	0	1.414	66.053	17,050
9	0	0	65.955	10,130
10	0	0	64,489	11,070
11	0	0	65,048	11,105
12	0	0	67,026	9,814
13	0	0	64,519	10,312

Table 3

Quadratic proposed polynomial models

Quadratic proposed polynomial models	
(1) Fabric density $Y_1$ =	$65.423+0.99x_1+1.378x_2+0.203x_1^2-0.995x_2^2+1.958x_1x_2$
(2) Water vapour permeability $Y_2$ =	$10.49-0.065x_1+0.0409x_2+3.12x_1^2+2.903x_2^2-1.035x_1x_2$

The values of the multiple correlation coefficients (Table 4) between the experimental data and the predicted values illustrate a very good and significant correlation.

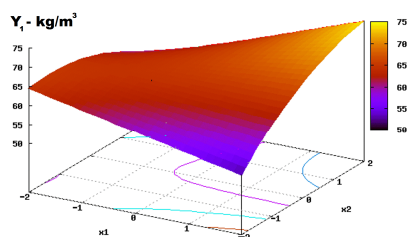
The fabric density equation  $Y_1$  (Table 4) reveals that the needling density ( $x_1$ ) and needle gauge ( $x_2$ ) have a significant influence on the characteristic described by the equation. Moreover, the coefficients of the first degree term have the same sign ( $b_1>0$ ,  $b_2>0$ ) indicating the effect of independent variables to be the same. The presence of second degree-term coefficient ( $x_2^2$ ) indicates a well-defined response surface. The effect of independent variables on fabric density is shown in Figure 1. In addition, a considerable influence has the interaction term  $x_1x_2$  whose positive coefficient shows an increasing tendency of the fabric density on the cumulative action of both parameters. As can be seen from Figure 1, the mathematical model  $Y_1$  describes a hyperboloid with saddle point having the following coded values:  $x_1 = -1.218$  and  $x_2 = -0.505$ . The actual values of the critical point obtained by optimization are  $x_1 = 66$  punches/cm<sup>2</sup> and  $x_2 = 32$  gauge for a fabric density  $Y_1 = 60.884$  kg/m<sup>3</sup>.

The higher the needling density for the same needle gauge, the higher is the fabric density due to the strong fiber peg formation as more number of fibers are arranged vertically (Sengupta and Sengupta, 2013). A higher fabric density means a higher compactness of the nonwoven. Regarding the increase of needle gauge, the action is softer than the action of needling density (as evidenced previously also Sengupta, 2005). The number of fibers at the surface pulled deep into the fabrics structure is smaller for the 40 gauge needle than for the 32 gauge needle (Lupu *et al.*, 2013) leading to increased fabric weight which means a higher fabric density, for the same needling density. Therefore, the critical point cannot be considered an optimum point for the nonwoven used as frost protector.

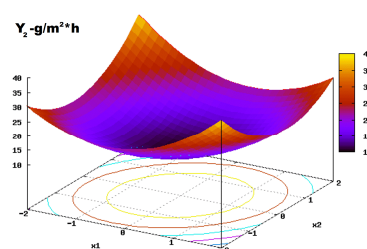
Table 4

**Response surface equation after testing of regression coefficients and evaluation of multiple correlation coefficient**

Response	Response surface equation	Coefficient of multiple correlation	F -ratios
$Y_1 =$	$65.423 + 0.99x_1 + 1.378x_2 - 0.995x_2^2 + 1.958x_1x_2$	0.8316	11.21
$Y_2 =$	$10.49 + 3.12x_1^2 + 2.903x_2^2 - 1.035x_1x_2$	0.988	204.59



**Fig. 1** - Effect of needling density ( $x_1$ ) and needle fineness ( $x_2$ ) on fabric density  $Y_1$  (kg/m<sup>3</sup>)



**Fig. 2** - Effect of needling density ( $x_1$ ) and needle fineness ( $x_2$ ) on water vapour permeability  $Y_2$  (g/m<sup>2</sup>.h)

Inspection of the equation of  $Y_2$  (Table 4) indicates an influence of needling density and needle gauge on water vapour permeability through a second-degree term. Also, the coefficients of the second-degree terms indicate a well-shaped response surface. A considerable influence has the interaction term. The negative sign of the interaction term coefficient indicates a decreasing tendency of the water vapour permeability to the cumulative action of both parameters. The effect of independent variables on water vapour permeability is presented graphically in Figure 2. The mathematical model of  $Y_2$  describes an elliptical parabolic dependency with a minimum point having  $x_1 = 0.00$  and  $x_2 = 0.00$  as coded values. The actual values of the critical point are  $x_1 = 105$  punches/cm<sup>2</sup> and  $x_2 = 36$  gauge for a water vapour permeability  $Y_2 = 10.490$  g/m<sup>2</sup>.h.

The increase in needle gauge over a certain value (36 gauge) is responsible for a smaller entanglement of the fibers resulting in a more bulky fabric structure. Thus, the water vapour permeability is increasing due to the higher number of voids through

which the water can be evaporated faster. Agrotextiles must conserve water by reducing evaporation. Hence, a minimum value for the water vapour permeability means a reduction of evaporation so the critical point can be considered an optimum. Another study has focused on the analysis of the same processing parameters (Lupu *et al.*, 2013) in order to establish the relationship between needling-punching process parameters (needling density and needle gauge) and thermal conductivity of the same obtained nonwovens. The water vapour permeability and thermal conductivity (main properties) can be changed by changing the two studied process parameters to ensure frost protection function of nonwovens.

## CONCLUSIONS

A series of cross-laid needlepunched nonwoven agrotextiles was produced based upon a central composite design for second-order model by varying the process parameters. We investigated the effect of two parameters on the physical properties of needlepunched nonwoven agrotextiles used as frost protectors. We found that the needling density and needle gauge have a significant effect on fabric density whereas the needle gauge has a major influence on water vapour permeability. The information available from contour diagrams regarding the interaction of parameters on fabric density and water vapour permeability is very useful for designing a needle-punched nonwoven fabric for agricultural applications.

Water vapour permeability is highly correlated with fabric density. A higher fabric density means a higher compactness of the nonwoven structure which can conserve water by reducing evaporation.

Using nonwovens in agricultural applications it is recommended based on several advantages like less expensive, easy to install and protects crops against low temperature and frost.

## REFERENCES

1. Cojocaru N., Clocotici V., Dobra D., 1968 – *Metode statistice aplicate în industria textilă*, Editura București, România
2. Lupu I.G., Cramariuc O., Hogas H.I., Hristian L., 2013 - *Parameters optimization for the production of needlepunched nonwoven agrotextiles*, The Journal of The Textile Institute, vol. 104, no.10, pp.1125-1131
3. Olle M., Bender I., 2010 - *The effect of non-woven fleece on the yield and production characteristics of vegetables*, Journal of agricultural science, no.1, pp. 24-29
4. Purdy A.T., 1980 - *Needle punching*, The Textile Institute Manchester, North Carolina State University, ISBN 10: 0900739320/0-900739-32-0
5. Rusell S.J., 2007 - *Handbook of nonwovens*, Woodhead Publishing Limited, Cambridge, England, ISBN-13: 978-1-85573-603-0 (book)
6. Sengupta S., 2005 - *Studies on tracking behaviour of jute needle-punched non-woven fabric*, IE (1) Journal-TX, vol. 86, pp.16-20
7. Sengupta S., Sengupta A., 2013 – *Electrical resistance of jute needle-punched nonwoven fabric – effect of punch density, needle penetration and area density*, Journal of The Textile Institute, Vol. 104, No.2, pp.132-139
8. Taloi D., 1987 – *Optimization of metallurgical processes*, Editura București, România

## STUDIES ON THE TOXIC EFFECT OF VARIOUS PHARMACEUTICAL PREPARATIONS OBTAINED FROM *HAMAMELIS VIRGINIANA*

### STUDII PRIVIND EFECTELE TOXICE ALE UNOR PREPARATE FARMACEUTICE OBȚINUTE DIN *HAMAMELIS VIRGINIANA*

**PRISĂCARU Cornelia<sup>1</sup>, PRISĂCARU Anca-Irina<sup>2</sup>**  
e-mail: corneliapris@yahoo.com

**Abstract.** *Hamamelis virginiana* L. (witch-hazel) is a shrub from Hamamelidaceae family cultivated in Europe for medical purposes. Due to its high tannins content (gallotannins, polycatechins, non-esterified gallic acid, procyanidins), flavonoides and saffrole-rich volatile oil, *Hamamelidis folium* and *Hamamelidis cortex* extracts are part of many pharmaceutical products for internal and external use. *Aqua Hamamelidis* is one of the favourite preparations in cosmetics and dentistry. The experiment presented in this study focuses on monitoring the hepatotoxic effects of pharmaceutical products and aqueous extractive solutions from *Hamamelidis folium* and *Hamamelidis cortex*. The trial was conducted on four groups of Wistar rats, to which various pharmaceutical preparations from vegetal products of *Hamamelis* were administered internally. At the end of the four weeks trial, it was noticed that the biochemical parameters important for the hepatic function and integrity were modified. The hepatotoxic effects are particularly important for the preparations obtained from *Hamamelidis cortex*.

**Key words:** *Hamamelidis folium*, *Hamamelidis cortex*, *Hamamelidis oil*, hepatotoxicity.

**Rezumat.** *Hamamelis virginiana* L. (nucul vrajitoarelor), arbust încadrat în familia Hamamelidaceae, este cultivat în scopuri medicinale în Europa. Datorită conținutului ridicat în taninuri (galotaninuri, policatechine, acid galic neesterificat, procianidoli), flavonozide și ulei volatil bogat în safrol, extractele din *Hamamelidis folium* și *Hamamelidis cortex* intră în compoziția a numeroase produse farmaceutice destinate administrării interne și externe. *Aqua Hamamelidis* constituie un preparat preferat în cosmetologie și stomatologie. Experimentul detaliat în acest articol științific se axează pe monitorizarea efectelor hepatotoxice ale produselor farmaceutice și soluțiilor extractive apoase din *Hamamelidis folium* și *Hamamelidis cortex*. Experimentul s-a efectuat pe patru loturi de șobolani Wistar, cărora li s-a administrat intern diferite preparate farmaceutice din produsele vegetale de *Hamamelis*. La finalul celor patru săptămâni s-a constatat modificarea unor parametri biochimici cu relevanță pentru integritatea și funcția hepatică. Efectele hepatotoxice sunt

---

<sup>1</sup> University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

<sup>2</sup> S.C. Fiterman Pharma, Iasi, Romania

deosebit de relevante pentru preparatele obținute din *Hamamelidis cortex*.

**Cuvinte cheie:** *Hamamelidis folium*, *Hamamelidis cortex*, ulei de *Hamamelis*, hepatotoxicitate.

## INTRODUCTION

*Hamamelis virginiana* is a shrub native from the eastern part of North America. The common name – witch hazel, comes from earlier times when native North Americans ascribed the plant with magic powers, probably due to its flowering characteristic. Witch hazel blooms later than any other native plant, generally in October through December but on occasion may start to bloom as early as September. The flowers have the ability to roll back up if the weather gets too cold and unfurl again when the weather is more favourable (Bone and Mills, 2012).

Over the years the plant has been highly appreciated as an astringent, venotonic, vasoprotective, anti-inflammatory and antibacterial remedy due to its high tannins content. The vegetal products, used in both traditional and modern medicine and cosmetics, are represented by the bark (*Hamamelidis cortex*) and leaves (*Hamamelidis leaves*) (Crellin and Philpott, 1990; Erdelmeier *et al.*, 1996; Hughes-Formella *et al.*, 1998). It is this high tannins concentration which makes the extract of witch hazel suitable mostly for external use. Nevertheless, there are food supplements on the European market administered internally for the improvement of peripheral circulation.

Tannins are generally regarded as safe due to their property of protein precipitation and their high molecular weight that impede their gastrointestinal or dermal absorption into the systemic circulation. Despite this, tannins have shown hepatotoxicity (Williamson and Manach, 2005).

The present study is part of a more extensive series of non-clinical experiments that stress the consumer's health and environmental protection and aims to establish the hepatotoxicity degree caused by ingestion of preparation containing *Hamamelidis folium*, *Hamamelidis cortex* and *Hamamelidis oleum*.

## MATERIAL AND METHOD

The experimental model included 4 groups of Wistar rats (average weight of 212.17 g), each group consisting of 5 animals. The experiment lasted 4 weeks (Table 1).

The first group of rats was the reference group which was kept in standard conditions. The animals of the second group were given 5% *Hamamelidis folium* infusion by gavage, in a dose of 2 g. The animals of the third group were administered 5% *Hamamelidis cortex* decoction in the same dose and by the same route. The fourth group was treated with Hamamelis oil (2 *guttas* diluted to 2 g with olive oil).

At the end of the experiment, blood samples were collected for the biochemical analysis which consisted in the assay of aspartate aminotransferase (AST), alanine aminotransferase (ALT), lactate dehydrogenase (LDH) and gamma-glutamyl transpeptidase (GGT).

Table 1

The experimental model				
Groups	5% HF <sup>1</sup> infusion	5% HC <sup>2</sup> decoction	Hamamelis oil	Biochemical parameters
Group 1	-	-	-	AST, ALT, LDH, GGT
Group 2	2 g	-	-	AST, ALT, LDH, GGT
Group 3	-	2g	-	AST, ALT, LDH, GGT
Group 4	-	-	2 guttes <sup>3</sup>	AST, ALT, LDH, GGT

<sup>1</sup>HF – *Hamamelidis folium*; <sup>2</sup>HC – *Hamamelidis cortex*; <sup>3</sup>diluted to 2 g with olive oil

## RESULTS AND DISCUSSION

The results obtained after the biochemical investigation of the parameters characteristic to the hepatic integrity are presented in Figs. 1-4. Transaminases are enzymes with a strictly cytosolic localization. This is why they are used as indicators of hepatic cytolysis. They are catalysts of the transamination reaction that allows the synthesis of aminoacids needed by the organism.

The evolution of ALT, as shown in Fig. 1, shows a slight insignificant increase of the activity from 18.123 UI (for the reference group) to 18.500 UI for group 2 (treated with 5% *Hamamelidis folium* infusion). Of higher significance is the increase of ALT to 20, 670 UI in group which was given the hamamelis oil. This augmentation may be explained by the migration of the enzyme from the cytosolic space to the extracellular liquid due to the hepatocyt membrane damage produced by the tannins from *Hamamelis virginiana*. This phenomenon is more important for the animals treated with decoction from *Hamamelidis cortex*.

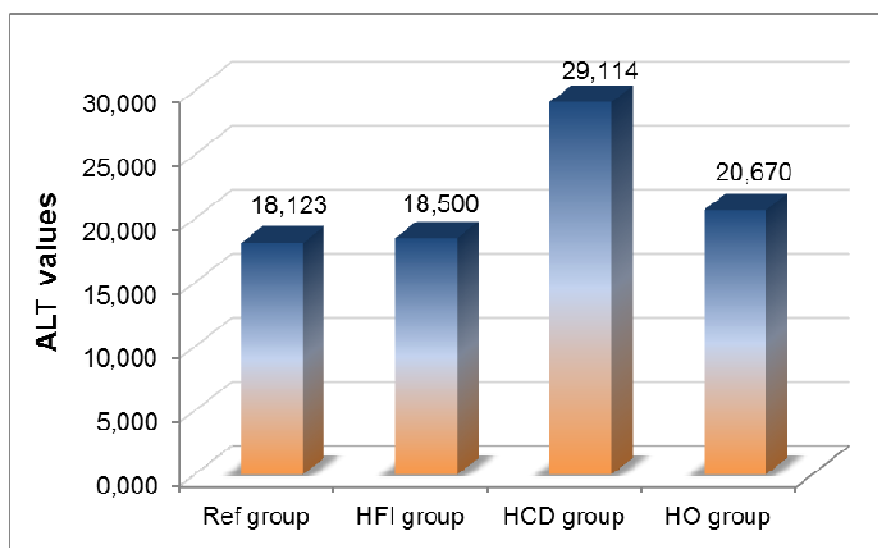


Fig. 1 - Evolution of ALT for the 4 experimental groups



The study of the second aminotransferase (AST) emphasize an increase of the serum activity for the animals that were given the infusion of *Hamamelidis folium* (from 48.999 UI, value characteristic to the reference group to 50.150 UI). Of great importance is the variation of AST in group 3 (treated with *Hamamelidis cortex* decoction), suggesting an increase of the hepatocyte permeability having as result the migration of AST in the serum (Fig. 2).

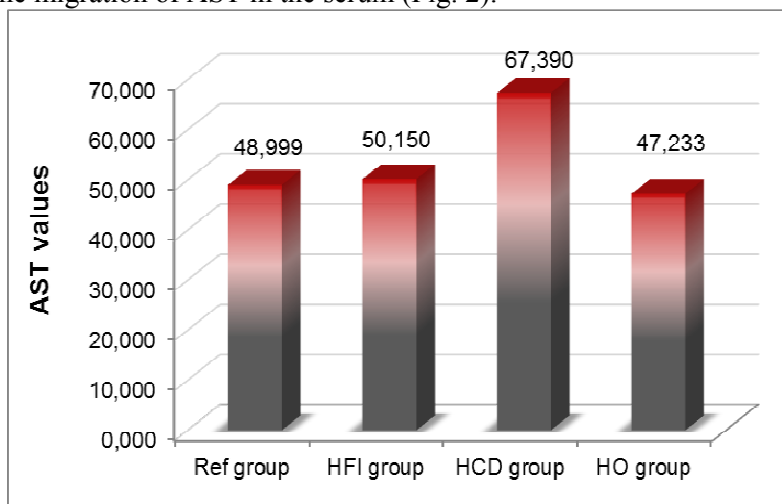


Fig. 2 - Evolution of ALT for the 4 experimental groups

The evolution of the third biochemical parameter (LDH) is in agreement with the evolution of the other parameters, revealing a significant increase of its activity in the serum of the third group which was given the decoction from the bark of witch hazel (2.291  $\mu\text{mol}/\text{ml}$  vs 1.557  $\mu\text{mol}/\text{ml}$  representing the value for the reference group) (Fig 3).

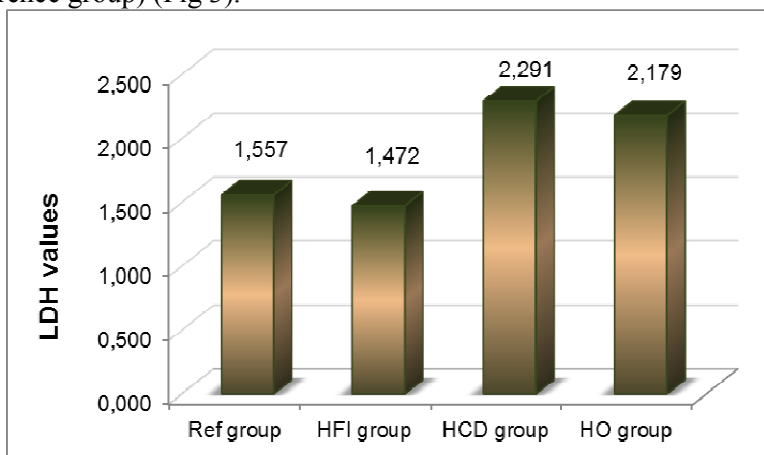


Fig. 3 - Evolution of LDH for the 4 experimental groups

The last of the studied parameters (GGT) is an enzyme that interferes the glicoproteic transport and is represented by five isoenzymes that have multiple diagnostic values (parameter of hepatic cytolysis, marker of ethylism and indicator of an oncogenic process). The evolution of this parameter (Fig. 4) sustains the previous results, the heighest value being reached by the third group (*Hamamelidis cortex* decoction).

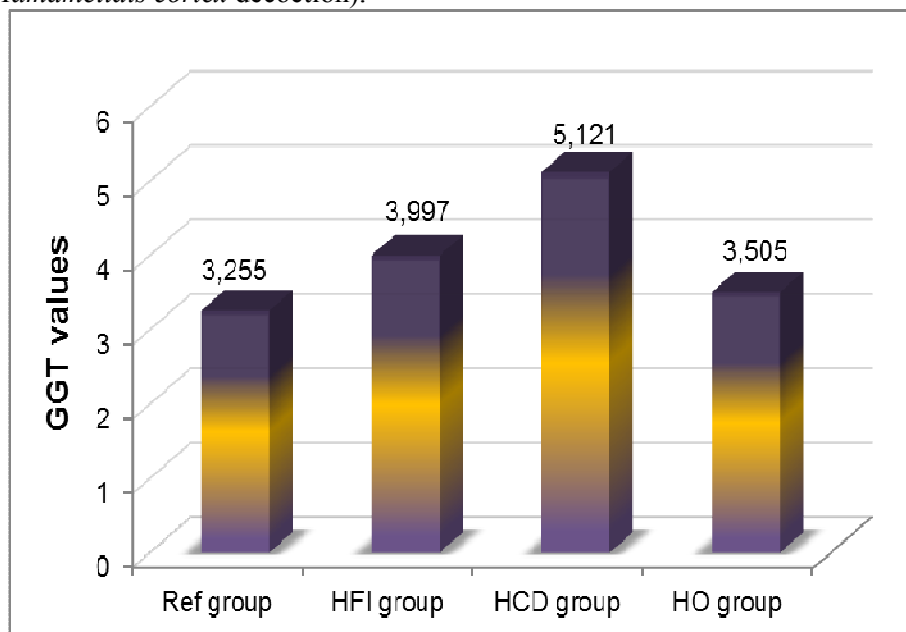


Fig. 4 - Evolution of GGT for the 4 experimental groups

## CONCLUSIONS

1. The 5% decoct of *Hamamelidis cortex* showed the highest hepatotoxic potential of all the studied preparations, inducing an increase of all the four biochemical parameters (aspartate aminotransferase, alanine aminotransferase, lactate dehydrogenase and gamma-glutamyl transpeptidase).
2. The oil extract of *Hamamelidis folium* is the second preparation when it comes to hepatotoxicity, producing a slight increase of aspartate aminotransferase and gamma-glutamyl transpeptidase activity.
3. The third studied preparation - 5% infusion of *Hamamelidis folium*, exerted its toxic potential only upon gamma-glutamyl transpeptidase.

## REFERENCES

1. Bone K., Mills S., 2012 - *Principles and Practice of Phytotherapy, Modern Herbal Medicine*, 2: *Principles and Practice of Phytotherapy*. Elsevier Health Sciences, 1051 pages.

2. **Crellin J.K., Philpott J., 1990** - *A Reference Guide to Medicinal Plants*. Duke University Press, 560 pages.
3. **Erdelmeier C.A., Cinatl J. Jr., Rabenau H., Doerr H.W., Biber A., Koch E., 1996** - *Antiviral and antiphlogistic activities of Hamamelis virginiana bark*. *Planta Med.* 62(3): 241-245.
4. **Hughes-Formella B.J., Bohnsack K., Rippke F., Benner G., Rudolph M., Tausch I., Gassmueller J., 1998** - *Anti-Inflammatory Effect of Hamamelis Lotion in a UVB Erythema Test*. *Dermatology* 196: 316–322.
5. **Williamson G., Manach C., 2005** - *Bioavailability and bioefficacy of polyphenols in humans. II. Review of 93 intervention studies*. *Am. J. Clin. Nutr.* 81(1): 243-255.

## THE PRESENCE OF HEAVY METALS IN RHUBARB ACCORDING BY TECHNOLOGY

### PREZENȚA METALELOR GRELE ÎN REVENT, ÎN FUNCȚIE DE TEHNOLOGIA APLICATĂ

**RĂILEANU Marcela<sup>1</sup>, COJOCARU Al.<sup>1</sup>, IPĂȚIOAIEI C.<sup>1</sup>,  
MUNTEANU N.<sup>1</sup>, STOLERU V.<sup>1</sup>**

**e-mail:** vstoleru@uaiasi.ro

**Abstract.** *Rhubarb( Rheum rhabarbarum L.) is a perennial vegetable plant, adapted to temperate climate in Romania but he is a little known and widespread. Such as rhubarb found favorable conditions for growth and development in our country, contributing to the diversification assortment, mentioning the advantage wich is not required establishment of culture each year. Through the application of differentiated technology, the content of heavy metal in rhubarb stalks differs of the cultivar and the harvesting period. Based on these considerations, the aim of the present study was to highlight the content of any heavy metals (Cd, Cr, Pb, Mn, Si) in one cultivar of rhubarb, variety Victoria. In all samples, the Cd content was below the detection limit, respectively below 10 ppm, and the other contaminants was respected in large the maximum admissible; is recommended in case of exceeding the maximum limit, vegetable product can't be consumed.*

**Key words:** *Rheum rhabarbarum, contaminants, fertilizers*

**Rezumat.** *Reventul sau rubarba este o plantă legumicolă perenă, adaptată climatului temperat al României fiind însă foarte puțin cunoscută și răspândită. Astfel că, reventul a găsit condiții bune de creștere și dezvoltare în țara noastră, contribuind la diversificarea sortimentului, prezentând avantajul că nu necesită înființarea culturii în fiecare an. În urma aplicării unor măsuri tehnologice diferențiate, conținutul de metale grele acumulat în pețiolii reventului, diferă în funcție de cultivar și de perioada de cultivare. Bazându-ne pe aceste considerații, scopul acestui studiu, a fost de a scoate în evidență prezența unor metale grele (Cd, Cr, Pb, Mn, Ni, Si) la un cultivar, respectiv soi Victoria, în funcție de tehnologia aplicată. În toate probele, conținutul de Cd a fost sub limita de detecție a aparatului, respectiv sub 10 ppm, iar ceilalți contaminanți au respectat în mare limitele maxim admise; deși este recomandat în cazul depășirii limitei maxime admise, produsele legumicole să nu fie consumate.*

**Cuvinte cheie:** *Rheum rhabarbarum, contaminanți, fertilizanți*

## INTRODUCTION

Rhubarb is a perennial vegetable plant, which can contribute to diversification of the assortment, thus the advantage of not requiring annual crop

---

<sup>1</sup>University of Agricultural Sciences and Veterinary Medicine of Iasi, Romania

establishment (Stan *et al.*, 2003).

Stalks are the edible part of the plant with a sweet-sour taste (2% carbohydrate) containing organic acids (ascorbic, tartaric, citric, malic, oxalic), minerals (0.5-0.6%) and 5% protein. Stalks of rhubarb are used in the preparation of compotes, jams, marmalades and other desserts. Rhubarb has a significant importance from ecological point of the view, as: being a perennial, does not require setting up each year; not requiring treatment plant; rhizomes exploring a large area of ground, so it can be cultivated and land with pesticide residues and heavy metals; and it can be grown on slopes, less fertile (Munteanu *et al.*, 2008; Stoleru *et al.*, 2014; Răileanu *et al.*, 2015).

The contaminants are the elements, which in large quantities or over limit can cause damage in terms of product quality and cause further illness and even environmental issues (Hura and Hura, 2007; Brădățan, 2007). Heavy metals, according to the degree of toxicity, acting differently on the physiological and biochemical processes. Downturn appearance and necrosis or chlorosis are some of the symptoms of heavy metal toxicity in plants. By eating contaminated food chemical, heavy metals such as neurodegenerative causes malfunctions (Hura, 2005; Brădățan, 2007).

Heavy metals, due to the potential toxic are danger for the environment and human health or animal. Heavy metals in the soil are usually naturally in low concentrations, which has a beneficial role (Zn, Mg, Se), but with increasing concentration, are toxic both to plants and animals or man. Currently, the doses are not established rhubarb and technological measures for the implementation of fertilization, leaf stalk consumable being the many contaminants can accumulate (Hura and Hura, 2007).

The aim of the study was to evaluate the influence of fertilization measures on the production and contaminants from edible.

## MATERIAL AND METHOD

An experiment with a rhubarb crop was carried out in an experimental plot at UASVM Iasi, during 2014-2015.

The experimental crop was established in 2013, by seedlings of 60 days, with a distance design of 100 x 75 cm (N= 47°11'34,09" E= 27°32'59,75"). Seedlings were been produce in concordance with specific literature, in a hot greenhouse.

The soil from the stationary is a mold cambic soil type, easy antropic, whit the following physicochemical properties, in the substrate of 0-60 cm: clay 32 %, ph=7,11; EC=252.3  $\mu$ S/cm<sup>2</sup>, CaCO<sub>3</sub>=1.03%, OM=28.23 mg/kg, C/N = 5,20, N=4.53 g/kg, P=106.66 ppm, iar CEC = 20.9 meq/100g.

To achieve its purpose, treatments were applied fertilizers in organic farming systems organic and conventional as follows: Orgevit® = 1100 kg/ha (applied in five stages, the first stage in early April and the following 10 in 10 days), Nutrifine®=700 kg/ha (applied in five stages, the first stage in early April and the following 10 in 10 days) and Micoseed®=60 kg/ha (applied in three stages, the first stage in early April and the following 10 in 10 days, that the treatment 4 and 5 was used Nutryaction®=5 l/ha)

Micoseed® It is a fertilizer based on microorganisms, particularly based *Glomus sp.* Nutrifine® a synthetic chemical complex type NPK 20-20-20. Orgevit® 100% organic poultry which is in the form of drops.

The total yield and dynamic of production, according with fertilization regimes, was carried out by weigh the stalks, for every harvest.

The biological material used for the analysis was represented stalk. Stalk is sweet, pink patches develop on a ribbed green, pink color is more intense at the bottom of the stem and faded green uniform top.

The principle of humidity determination is based to loss of the drying oven at a temperature of 102 ° C, until a constant mass. The loss will be calculated as a percentage relative to the initial mass of the sample.

Determination of heavy metals in vegetables is by dry mineralization. This consists in the destruction by carbonization and incineration (450-500 °C) oven sample; ash being thus passed by dissolving in dilute hydrochloric acid.

The results on the content of heavy metals in rhubarb stalks have been conducted at the UASVMB Timisoara.

## RESULTS AND DISCUSSIONS

### Results regarding to the total yield on rhubarb

The dates for total production are presented for both years in the Fig.1. In all experimental versions the yield is higher to Control.

The average yield of rhubarb, during 2014 period, ranged from 26.54 t / ha in the control version to 45.45 t/ha in the variant fertilized with Micoseed®. Positive significant differences to the control have been obtained in the versions fertilized with Orgevit® (31.08 t/ha) and the Nutrifine® (41.63 t/ha).

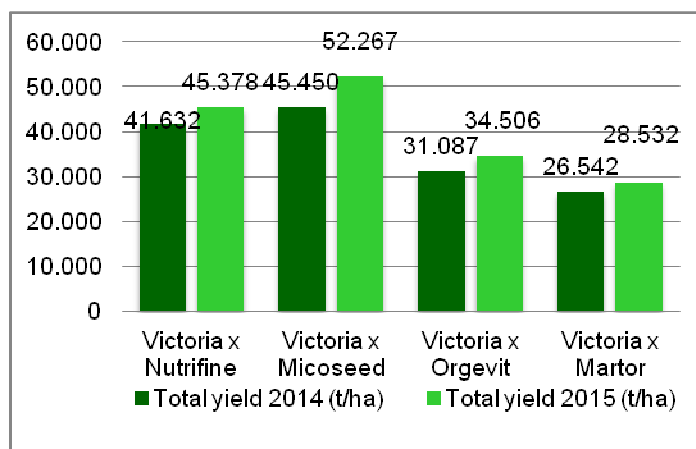


Fig.1 - Results for rhubarb yield, during 2014-2015

Average yield of rhubarb, during 2015 period, varied from 28.53 t / ha in control version to 52.26 t / ha in the same experimental version, fertilized with Micoseed®. Positive significant differences than to control were obtained by using Orgevit® (34.50 t/ha) and the Nutrifine® (45.37 t/ha) fertilizers

**Results on the dry matter content and humidity**

The dry matter content and water from vegetables, are the primary factors to the optimum development of metabolic processes. In the experimental study we can conclude, the maximum dry matter was obtained in the organic version (9.63%) and the lowest in the chemical treatment (7.85%). The humidity is inversely proportional with organic matter content.

Table 1

**Rezultate privind conținutul de dry matter and humidity**

Treatment applied	Biochemical analysis		Total %
	s.u. (%)	u. (%)	
Nutrifine	7,85	92,15	100
Micoseed	8,13	91,87	100
Orgevit	9,63	90,37	100
Un-fertilized (control)	9,14	90,86	100

According to the different references (Butnariu and Buta, 2014; Ciofu *et al.*, 2004; Stan *et al.*, 2003), the water content varies between 90.37% and 92.15% in rhubarb, which shows us that the results obtained are around values other authors.

**Results on the heavy metal contents in rhubarb**

The dates on heavy metal contents are shown in Table 2. The cadmium content was below 10 ppm, below the detection limit of equipment. The maximum limit allowable for Cd in vegetable leaf should be less than 50 mg/kg FW, which shows that the results not exceed the maximum admitted limit (MAL).

Chromium content in the experimental versions widely varied from 70 mg/kg FW in the fertilized version with Orgevit® up to 115 mg/kg FW in Control. The dates confirm that MAL is exceeded more two2 times in Control. In the fact not the fertilization system influences the accumulation of heavy metal in edible part of rhubarb. The content of Cr from stalks was around 80 mg / kg, in the both versions chemical respectively biological fertilized.

The higher quantity of chromium in the Control can be explained by the fact that this element comes from soil as residue. Provided with not fertilized crop, the root of the rhubarb has a physiological tendency to develop a deep root system and explore a large area of soil especially in the depth.

Thus, the rhubarb extracts nutrients along with different contaminants from soil, which have been at greater depths. Because the rhubarb is a perennial species, year by year the root system is larger.

By the application of fertilizers near the roots, plants explores a lower soil surface, the system is less developed and thus not of great depth takes some heavy metals.



Heavy metal contents in the rhubarb samples

Experimental version	Heavy metals contents					
	Cd (mg/kg)	Cr (mg/kg)	Pb (mg/kg)	Ni (mg/kg)	Si (mg/kg)	Mn (%)
Victoria x Micoseed	□ 10	80	13	53	29,03	0,26
Victoria x Orgevit	□ 10	70	8	49	29,39	0,30
Victoria x Nutrifine	□ 10	81	23	43	28,24	0,32
Victoria x Martor	□ 10	115	18	61	28,58	0,17
MAL	50	50	50	50	50	5

\*-MAL – maximum admitted limit

The lead content in all experimental versions allowed the maximum concentrations of 50 mg / kg. Nevertheless, the highest lead content was achieved in chemical fertilization, respectively 23 mg/kg FW.

Nickel content in the experimental stationary ranged from 43 mg/kg FW in the chemical fertilized version to 61 mg/kg FW to Control. From the results can be said that in the same case of chromium, nickel content is not influenced by fertilization regime applied more even in the unfertilized version. Thus, MAL was exceeded in control and microorganism version.

The content of silica slightly varies, depending on the experimental variant, the higher was determined in organic fertilizer 29.39 mg/kg FW and the lower in the chemical version, respectively 28.24 mg/kg FW.

In the case of manganese, the highest concentration was in chemical version of 0.32% and the lowest was registered in control sample 0.17%, which shows that Mn accumulation in the stalks of rhubarb is influenced by nutrition system. By using organic fertilizers and those of micro-organisms, to ensure concentration of Mn values between 0.26% and 0.30%, without exceeding the limit of 5%.

## CONCLUSIONS

Regards to total production, in 2015 was obtained significant positive difference (+ 14.98%) compared to 2014. The highest yield for version fertilized with Micoseed® (52.26 t/ha in 2015), and the lowest for control sample (28.53 t/ha in 2015).

Higher dry matter content in Orgevit® fertilized version (9.63%) signifies accumulation of nutrients in plants, including dry mass. The content of Cr and Ni higher in stalks of rhubarb from the control sample compared to fertilized variants indicates that the content is not influenced by fertilization regime.

In most experimental versions MAL for contaminants have been exceeded, except in any situations. In this case, further research must be separate for each nutrient. For products where limits have been exceeded, their consumption is not recommended as it can cause neurodegenerative diseases is recommended that harvesting be done earlier.

## REFERENCES

1. Brădățan Gheorghe, 2007 – *Siguranța alimentelor*. Editura "Ion Ionescu de la Brad", Iași.
2. Butnariu M., and Buta A., 2014 – *Chemical composition of Vegetables and their products*. Handbook of Food Chemistry. Springer, 1-49. DOI 10.1007/978-3-642-41609-5\_17-1.
3. Ciofu Ruxandra, Stan N., Popescu V., Chilom Pelaghia, Apahidean S., Arsenie H., Berar V., Lauer K.F. Atanasiu N., 2004 - *Tratat de legumicultură*. Ed. Ceres, București.
4. Hura Carmen, 2005 – *Contaminarea chimică a alimentelor în România, în 2004*, vol.4, Editura CERMI, Iași, ISBN; 973-667-142-9.
5. Hura Carmen, Hura B.A., 2007 – *Assessment of the heavy metals in the food from Romania*, International Congress of Toxicology (ICTXI), 15-19.07.2007, Montreal, Canada.
6. Munteanu N., Stoian L., Stoleru V., Fălticeanu Marcela, 2008 – *Baze tehnologice ale legumiculturii ecologice*. Editura "Ion Ionescu de la Brad" Iași, 182 pp., ISBN 978-973-147.
7. Răileanu Marcela, Stoleru V., Munteanu N., Butnariu M., 2015 – *Perspective privind conținutul de acizi organici în revent*. Simpozionul științific al Facultății de Horticultură, Iași, 2015. Lucrări științifice USAMV Iași, seria Horticultură, In press.
8. Stan N., Munteanu N., Stan T., 2003 – *Legumicultură*, vol. III, Editura " Ion Ionescu de la Brad" Iași.
9. Stoleru V., Munteanu N., Sellitto V.M., 2014 - *New approach of organic vegetable systems*. Aracne Publishing House, Italy.
10. \*\*\* Bioterra, 2001 – *Îndrumător pentru Agricultură Ecologică*. Asociația Bioterra, Cluj Napoca.

## ASSESSMENT OF BIOLOGICAL SOIL FATIGUE IN HORTICULTURAL MONOCULTURES

### EVALUAREA OBOSELII SOLULUI DIN PUNCT DE VEDERE BIOLOGIC ÎN MONOCULTURILE HORTICOLE

**Jose Ignacio MARÍN GUIRAO<sup>1</sup>, Francisco Martín USERO<sup>1</sup>,  
Victoria VELASCO ARROYO<sup>1</sup>, José Manuel RODRÍGUEZ ZAFRA<sup>2</sup>,  
Julio César TELLO MARQUINA<sup>1</sup>  
e-mail: jignaciomarin@gmail.com**

**Abstract.** Biological soil fatigue in horticultural monocultures awakes the highest interest and presents the highest discrepancy. Different studies associate it with soil pathogens as causing agents, while others determine it in the lack of these. This work deals with the assessment of fatigue existence, a phenomenon which can be appreciated in the field, but a numerical model that provides data about yield losses and lack of plant vigour has not been found. Trial planning consisted of adding fresh organic matter through biodisinfection techniques and providing knowledge about the phenomenon and its relation with the content of organic matter and soil microbiota. For this reason, soils of two greenhouses were compared, the differences between them were the type of crop (cucumber and tomato respectively) as well as the supplying or non supplying of organic amendments. The content of organic matter and the soil or telluric microbiota (fungi, bacteria and oomycetes) in the soils were studied, as well as its effect on cucumber and tomato seedlings under controlled conditions. The results showed that fatigue appeared in soil with low content of organic matter, which showed at the same time lower density and diversity of fungal population. The addition of fresh organic matter seems to reconstitute the productive capacity of the soils, and this mitigates the fatigue and monoculture effects.

**Key words:** Edaphic microbiota, fungi, bacteria, oomycetes, seedlings.

**Rezumat.** Oboseala solului din punct de vedere biologic în monoculturile horticole trezește cel mai mare interes și prezintă totodată și o mare diferență. Diferite studii asociază acest fenomen cu agenți patogeni din sol ca agenți care îl provoacă, în timp ce alții afirmă că fenomenul se produce și în lipsa acestora. Această lucrare se referă la evaluarea existenței oboselii solului, un fenomen care poate fi apreciat în domeniu, însă un model numeric, care să ofere date despre pierderile de randament și lipsa de vigoare a plantelor nu a fost găsit. Experimentul a constatat din adăugarea de materie organică în stare proaspătă prin tehnici biodezinfectie și furnizarea de cunoștințe despre fenomenul și relația acestuia cu conținutul de materie organică și microbiotei solului. Pentru aceasta, solurile de două sere au fost comparate, diferențele dintre ele au fost

---

<sup>1</sup> Research group AGR-200. University of Almería. Department of Agronomy. Almería, Spain.

<sup>2</sup> Granada La Palma Sociedad Cooperativa Andaluza. Carchuna, Spain

*de tipul de cultură (castraveți și tomate), precum și administrarea, sau nu, de amendamente organice. A fost studiat conținutul de materie organică din sol și microbiota solului (ciuperci, bacterii și oomicetelor), precum și efectul asupra răsadurilor de castravete și tomate în condiții controlate. Rezultatele au aratat ca oboseala solului a apărut în varianta cu conținut scăzut de materie organică, care a avut în același timp și densitate mai mică dar diversitate a populației fungice. Adăugarea de materie organică în stare proaspătă poate reconstitui capacitatea productivă a solurilor, iar acest lucru atenuează efectele oboselii solului în monocultură.*

**Cuvinte cheie:** Microbiota edafică, fungi, bacterii, oomicete, răsad

## INTRODUCTION

This work tries to assess the existence of fatigue in horticultural soils in monoculture, as well as quantifying its intensity, this phenomenon can be appreciated in the land, but a numerical model which allows us to obtain reliable data about yield losses and lack of plant vigour has not been found. The studies that consider the existence of the phenomenon date from the years 70-80, and base their empirical character on the observations made in the land, in many cases, by the farmers themselves, without giving a numerical quantification. For this reason, the first question that must be posed by any person interested in the study of soil fatigue is: how do authors define fatigue if there is not data that supports its existence? From here on, the phenomenon complexity of fatigue, considered as an agronomic concept and defined by different authors as "fertility disorder of soils due to multiple causes that can be accumulative, successive or simultaneous in the land" (Bouhot 1983c, Chen *et al.* 1991) that causes a partial reduction of productivity and it is attributed mainly to reiterated monoculture in the same plot (Cebolla & Maroto 2004, Scotto 1982). However, this ambiguous term has been used with different approaches throughout the years. So that, physical fatigue has been defined as the loss of soil fertility due to labours or amendments which cause losses in their structure, and with this, a soil impoverishment (Maroto 2000, Bouhot & Dumas 1982). Chemical fatigue is described as the modification of chemical parameters of soil due to different fertilizers (Casado 1925, Bodet 1982), as well as the negative effect of allelopathic substances in the root development (Maroto 2000). And, finally, biological fatigue which is due to microbiological imbalances in favour of some organisms and/or to the detriment of others (Casado 1925), which is the most studied and the most controversial. In this sense, the main difference is found in the "thin line" that separates the phenomenon soil fatigue, that affects the normal development of plants, from pathogenicity that implies the presence of pathogenic organisms that act as causal agents in the expression of disease. These two terms, soil fatigue and pathogenicity, seem to walk hand in hand in the first studies about fatigue. Different authors, after observing yield losses in land with symptoms attributed to soil fatigue, carried out studies which concluded that the presence of pathogens as responsible agents of

such losses (Bouhot 1975, Bouhot 1979, Bouhot 1982a, Bouhot 1982b, Meynard & Bouhot 1982, Hoestra 1982, Vigouroux 1982, Roudeillac 1982, Bouhot & Bonnel 1982), even in some of these cases, biological fatigue coincides with chemical fatigue (Bouhot 1982a, Bouhot 1982c, Gindrat *et al.* 1982). On the other hand, due to the intensification of the expression of fatigue phenomenon after the withdrawal of methyl bromide, disinfecting substance of horticultural soils which achieved "covering up" its adverse effects (Tello & Lacasa 2004, Lacasa *et al.* 1999, Guirao *et al.* 2004), new studies arose which showed the existence of the phenomenon in the absence of pathogens (Otto *et al.* 1994, Lacasa *et al.* 2002, Guerrero *et al.* 2004, Guerrero *et al.* 2014). In this way, these studies showed that monoculture can involve that some microorganisms increase their presence in soils, to the detriment of others, which can affect negatively production, and in the case of pepper, it can be reduced to 60% (Lacasa *et al.* 2002, Guerrero *et al.* 2004). So that, before the possible existence of fatigue, other studies showed how adding organic matter through biodisinfection in agricultural soils in monoculture, affected fungal soil microbiota which reached a new balance (Martínez *et al.* 2009). Furthermore, biodisinfection seems to improve physical-chemical characteristics of soil (Fernández *et al.* 2005). These results have not been corroborated until now with new studies that evaluate and quantify soil fatigue caused by monoculture. For this reason, this study allows us to explore a model to be characterised in the intensive crops under plastic in the Spanish southeast. The way the trials are set out consists of adding fresh organic matter through biodisinfection techniques to compost soil, and it tries to provide knowledge about fatigue phenomenon or soil exhaustion in monoculture and its relationship with the content of organic matter and the soil microbiota. To that end, soils of two greenhouses of the province of Granada were compared which carried out monoculture year after year, and that they differ one from the other by the type of crop (cucumber and tomato respectively), as well as the supply or non supplying of organic amendments to the soil. The content of organic matter and the soil or telluric microbiota (fungi, bacteria and oomycetes) in the soils was studied, as well as its effect on cucumber and tomato seedlings under controlled conditions. Furthermore, in the case of soil with low content of organic matter, the assessments were made after the first incorporation of organic amendments through biodisinfection, to compare the results obtained before and after the disinfecting amendment.

## MATERIAL AND METHOD

### Selected greenhouses. Location.

#### *Greenhouse 1. Soil with low content of organic matter.*

Greenhouse with "raspa y amagado" Almería type structure, located in the subtropical coast of Granada (Motril). This soil has been cultivated for 14 years with cucumber monoculture always with the same cultivation lines, and no organic matter has ever been supplied. In the first sampling carried out, the greenhouse showed high incidence of disease in plants caused by *Fusarium oxysporum* f.sp. *radicis cucumerinum*.

When the crop ended, a biodisinfection treatment was carried out through the incorporation of mustard+radish+manure+rest of the previous crop (including sick plants) and then later solarisation (biosolarisation) during approximately 3 months.

*Greenhouse 2. Soil with high content of organic matter.*

Greenhouse with "raspa y amagado" Almería type structure, located in the interior of Granada (Fornes). Tomato monoculture has been cultivated in this soil for 14 years. Tomato crop has always been planted in this soil with the exception of a cucumber crop during 3 seasons. Every year supplies of fresh organic matter are made to be composted in the soil. Specifically, mustard+radish+vetch which are added once the tomatoes have grown, in autumn-winter, and prior to this, they bury tomato plants with manure at the end of the harvest.

**Sampling.**

The samples were made following a determined spatial distribution, with the purpose of obtaining a sample as homogeneous as possible. Therefore, with the help of a spade, the samples of approximately 10 kg were taken at a depth of between 0 to 30 cm, at three different points within the same cultivation line, and then they were mixed and homogenized inside the same bag. The samples were taken in the centre of the cultivation line to avoid the possible "edge effect" of the corridor between cultivation lines.

The samples that were taken were the following:

Greenhouse 1: - Before the incorporation of organic matter (8/5/2014).

- After the incorporation of organic matter through biodisinfection (21/1/2015).

When samples were taken, organic matter had not been fully decomposed (the typical smell of geosmin produced by actinomycetes was appreciated).

Greenhouse 2: - A single sample was made (29/5/2014). The sample was taken when double stem plants had the third cluster opening.

**Determination of the percentage of organic matter in soils.**

Determination of organic matter was made through oxidation method with potassium dichromate and chloridric acid and a later valuation with Mhor's salt.

**Assesment of plant vigour in controlled environmental chamber.**

***Vegetable material***

Evaluations were made for the two horticultural species that are grown usually in the soils that are being studied: tomato (*Solanum lycopersicum* L. cv. Río Grande; Ramiro Arnedo S.A.), and cucumber (*Cucumis sativus* cv. Marketmore 76; Ramiro Arnedo S.A.). Therefore, seedlings of the two horticultural species were grown in each of the studied soils, with the purpose of determining the fatigue, if any, was specific for a particular species or if it was shown for both horticultural species.

***Description of trials***

Each horticultural species was sown separately at a rate of one tomato seed and one cucumber seed, in 175 ML volume pots (experimental unit) which contained the soil subject to study mixed with vermiculite at a rate 2:1 v/v, to avoid soil compaction which was observed in previous trials with non mixed soil.

Seeds were previously disinfected through immersion in a commercial dissolution of sodium hypochlorite at 20% during 15 minutes, and then they were rinsed and soaked during 48 hours in a wet chamber before sowing. Trials were carried out during 30 days in a controlled environmental chamber at 12.000 lux, photoperiod of 14 h light/day and maximum and minimum temperatures of 24,9±0,6 and 21,2±0,8 °C respectively. Plants were irrigated on demand, but they were not fertilised during the trials.

Five replications were made with each soil for each horticultural species, and the trials with the different soils were repeated twice over the trial period.

#### ***Evaluated parameters in seedlings***

- *Number of leaves.* Only the true leaves were considered, cotyledons as well as non fully developed leaves were rejected.

- *Length of the aerial part.* The total length of the aerial part of the seedling was considered as the distance between the part of the stem at the substrate level area and the apical bud of the seedling.

- *Aerial dry weight and root dry weight.* The aerial part was separated from the root part, and they were dried in a heater at 72°C during 48 hours. After this time, each part was weighed separately on a precision scale (Model PB 303-S, with 1mg sensibility).

- *Leaf area.* To calculate the leaf area, the free software ImageJ 1.48 (NIH Image, Maryland, USA) was used. Previously, the leaves and/or leaflets of each seedling were scanned (Scanner Epson Perfection 1240).

#### **Study of the fungal and bacterial microbiota and oomycetes.**

##### ***Sampling preparation***

Following the instructions of Tello *et al.* (1991) and Rodríguez-Molina (1996) the samples were subject to a drying, grinding and sieving process. After placing the samples in plastic trays, drying was done at environmental temperature, during a variable time (7-10 days) depending mainly on the humidity of the sample when arriving at the laboratory. A porcelain mortar was used for grinding, and for sieving a 200  $\mu$  mesh size sieve. The mortar and the sieve were washed and disinfected between the different samples flaming them with alcohol.

##### ***Analytical methods***

The analytical methods used to know the microbiological composition of soils were (Tello *et al.* 1991):

*Successive suspension-dilutions method for the evaluation of the total microbiota (fungi and bacteria).* In this case, soil is added to a culture medium in the form of a suspension of sterile water. The culture medium was acidified malt extract agar. Ten repetitions of each sample were made at the dilutions  $10^{-3}$  and  $10^{-4}$ . Incubation was carried out in the laboratory at room temperature during 4-7 days. After the indicated time, the fungi Colony-Forming Units (CFU) were counted and identified at the levels of the genera, as well as the CFU counting of the total bacteria present in each repetition. In this case, the  $10^{-4}$  dilution was chosen, the results are expressed as  $\times 10^4 \text{UFC} \cdot \text{g}^{-1}$  of dry soil.

*Komada's selective medium (1975) modified by Tello et al. (1991) to assess the Fusarium flora.* It's a selective culture medium for *Fusarium* genus which has three solutions. In this case, soil dilution is carried out directly in a dish with the melted culture medium. For each soil fraction or replication (4 in total), 4 dishes were prepared, which supposes a total of 16 dishes per sample. Incubation was carried out in the laboratory at room temperature during 4-7 days. After the indicated time, the CFU present in each Petri dish were counted and identified at the levels of the species.

*Vegetable trap method to evaluate the presence of oomycetes.* Immature petals of carnation were used. Five repetitions of each sample were made for each of the sampling carried out. Repetitions had 5 carnation petals. Incubation was made in the laboratory at room temperature during 4-5 days. After the indicated time, data was taken, determining the presence or absence of oomycetes in each of the replications, for this reason, presence was considered when oomycetes were found in at least one of the petals. The



results are expressed, for each of the samples carried out as a % of replications with presence of oomycetes.

#### Statistical Analysis

Student's t-test was carried out to compare the parameters evaluated in cucumber and tomato plants between soils. As they were parametric analysis, the assumptions of normality and homoscedasticity were checked previously. With the purpose of evaluating significant differences in the microbial variables analysed (Fungi CFU, Bacteria CFU) a non-parametric test was applied (Kruskal-Wallis), given that a data transformation was not found that fulfilled the assumptions of normality and homoscedasticity required by the parametric tests (due to the high variability of the values of the variables studied as effect of the heterogeneity that these variables show naturally in the soil). The statistical package used was Statgraphic Plus 5.1 (Manugistic Incorporate, Rockville, MD, USA) for Windows.

## RESULTS AND DISCUSSIONS

#### Percentage of organic matter on soils.

Soil of Greenhouse 1 showed a 0.54% content of organic matter before the incorporation of organic amendments through biodisinfection, and reached 1.56% after the application of the treatment. Soil of Greenhouse 2 showed a 4.46% content of organic matter.

#### Plant vigour in controlled environmental chamber

##### *Greenhouse 1 vs Greenhouse 2*

Table 1 showed that when plants were grown in tomato monoculture greenhouse soil, where during the last 14 years, organic amendments have been added (Greenhouse 2), in all the parameters evaluated, the values are significantly higher compared with the plants grown in cucumber monoculture soil without supplies of organic amendments (Greenhouse 1). These differences were shown in the case of cucumber plants as well as tomato plants.

Table 1

Values (average $\pm$ typical deviation) of the parameters considered for the assessment of cucumber and tomato plant vigour in controlled environmental chamber, depending on the soil of two greenhouses.

Greenhouse 1: soil with low content of organic matter and cucumber monoculture;

Greenhouse 2: soil with high content of organic matter and tomato monoculture.

	No. of leaves	Height (cm)	Root dry weight (g)	Aerial dry weight (g)	Leaf area (cm <sup>2</sup> )
<b>Pepino cv. Marketmore</b>					
Greenhouse 1	2,3 $\pm$ 0,5	4,01 $\pm$ 0,75 b	0,05 $\pm$ 0,01 b	0,15 $\pm$ 0,05 b	17,63 $\pm$ 9,51 b
Greenhouse 2	4,0 $\pm$ 0,0	6,08 $\pm$ 0,74 a	0,14 $\pm$ 0,04 a	0,34 $\pm$ 0,06 a	89,97 $\pm$ 14,16 a
<b>Tomate cv. Rio Grande</b>					
Greenhouse 1	2,4 $\pm$ 0,5 b	6,05 $\pm$ 1,44 b	0,05 $\pm$ 0,02 b	0,11 $\pm$ 0,04 b	19,54 $\pm$ 9,66 b
Greenhouse 2	4,4 $\pm$ 0,8 a	8,95 $\pm$ 0,95 a	0,10 $\pm$ 0,03 a	0,29 $\pm$ 0,05 a	92,52 $\pm$ 13,91 a

Different letters in the same parameter and horticultural species show significant differences ( $P \leq 0.05$ ) through t-Student test.

*Greenhouse 1 before and after the incorporation of organic amendments through biodisinfection*

Likewise, with reference to the results shown in Table 2, vigour of plants which were grown in cucumber monoculture soil without organic amendments supplies during 14 years (Greenhouse 1) improved after the first incorporation of organic amendments through biodisinfection (biosolarisation). In this way, in tomato as well as cucumber plants, the values of all the evaluated parameters are significantly higher after the addition of organic matter.

Table 2

**Values (average±typical deviation) of the parameters considered for the assessment of cucumber and tomato plant vigour in controlled environmental chamber, depending on the soil of Greenhouse 1 (Soil with low content of organic matter and cucumber monoculture) before and after the incorporation of organic matter through biodisinfection**

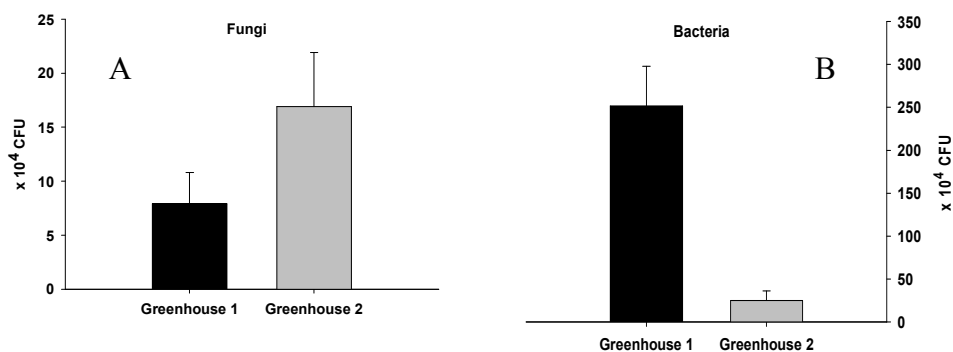
Greenhouse 1	No. of leaves	Height (cm)	Root dry weight (g)	Aerial dry weight (g)	Leaf area (cm <sup>2</sup> )
<b>Pepino cv. Marketmore</b>					
Before	2,3±0,5 b	4,01±0,75 b	0,05±0,01 b	0,15±0,05 b	17,63±9,51 b
After	3,1±0,3 a	5,31±0,64 a	0,08±0,02 a	0,26±0,04 a	48,29±22,33 a
<b>Tomate cv. Rio Grande</b>					
Before	2,4±0,5 b	6,05±1,44 b	0,05±0,02 b	0,11±0,04 b	19,54±9,66 b
After	3,2±1,1 a	7,82±0,54 a	0,07±0,01 a	0,19±0,03 a	51,64±27,52 a

Different letters in the same parameter and horticultural species show significant differences ( $P \leq 0.05$ ) through t-Student test.

**Fungal, bacterial microbiota and oomycetes.**

*Greenhouse 1 vs. Greenhouse 2*

Total fungal density grown in the greenhouse tomato monoculture soil in which during the last 14 years, organic amendments have been added (Greenhouse 2), showed significant differences ( $p$ -value<0,001 through Kruskal-Wallis test) compared with cucumber monoculture soil without supplies of organic amendments (Greenhouse 1), being the first case significantly higher (Figure 1A).



**Fig. 1 - Total fungi (A) and bacteria (B) density in the soils of Greenhouses 1 (soil with low content of organic matter and cucumber monoculture) and 2 (soil with high content of organic matter and tomato monoculture). Values (average±typical deviation) expressed in 10<sup>4</sup> CFU·g<sup>-1</sup> of dry soil. \*\*\* ( $p < 0,001$ ) through Kruskal-Wallis test**

Likewise, fungal diversity in soil of Greenhouse 2 was also higher, it showed 13 different fungal genera compared with the 5 genera present in the Greenhouse 1 (Table 3) soil. In this respect, in Greenhouse 2 soil (soil with a high content of organic matter) population density of the different genera expressed was well-balanced, and showed total values that, in general, did not differ very much between genera, except for a possible exception. In this case, the *Acremonium* spp. and *Fusarium* spp. genera showed the highest populations, and *Gilmaniella* spp. and *Drechlera* spp. are the fungal genera that showed the lowest populations. However, in Greenhouse 1 soil (soil with low content of organic matter) the fungal population density belonging to *Fusarium* spp. genus stood out, followed by those belonging to *Penicillium* spp. genus, while the presence of *Acremonium* spp. and *Aspergillus* spp. was minimum. Also it must be considered that all the genera present in Greenhouse 1 soil were also found in Greenhouse 2 soil.

Table 3

**Population density of fungi genera present in the soils of Greenhouse 1 (soil with low content of organic matter and cucumber monoculture) and of Greenhouse 2 (soil with high content of organic matter and tomato monoculture). Values (average±typical deviation) obtained from successive suspension-dilution technique and acidified malt extract agar, and expressed in  $10^4$  CFU·g<sup>-1</sup> of dry soil.**

	Greenhouse 1	Greenhouse 2
% Organic matter	0,54	4,46
Genera	x $10^4$ CFU·g <sup>-1</sup>	
<i>Acremonium</i> spp.	0,1±0,3	2,9±1,8
<i>Alternaria</i> spp.	--	0,4±0,7
<i>Amblyosporum</i> spp.	--	1,4±2,4
<i>Aspergillus</i> spp.	0,1±0,3	1,4±1,2
<i>Drechlera</i> spp.	--	0,2±0,6
<i>Fusarium</i> spp.	4,2±2,3	2,7±1,3
<i>Gilmaniella</i> spp.	--	0,1±0,3
<i>Mucor</i> spp.	--	0,3±0,5
<i>Paecylomices</i> spp.	--	1,7±1,6
<i>Penicillium</i> spp.	1,4±1,1	1,9±1,4
<i>Stachybotrys</i> spp.	--	0,3±0,7
<i>Stemphylium</i> spp.	--	1,9±1,7
Non Identified	0,7±0,9	1,7±1,2
Total Genera	5	13

Likewise, the soil of Greenhouse 2 also showed a higher diversity of fungi species belonging to *Fusarium* spp. genus, it showed 4 species compared with a single species present in Greenhouse 1 soil (Table 5). In this respect, it must be highlighted that the species present in greenhouse 1, *Fusarium oxysporum*, which showed a very high population density and in this case it was identified as *F.oxysporum* f.sp. *radicis-cucumerinum* (the causing agent of root and crown rot in cucumber plants) was not present in the soil of Greenhouse 2. In this last soil, the species present depending on the population density in decreasing order were:

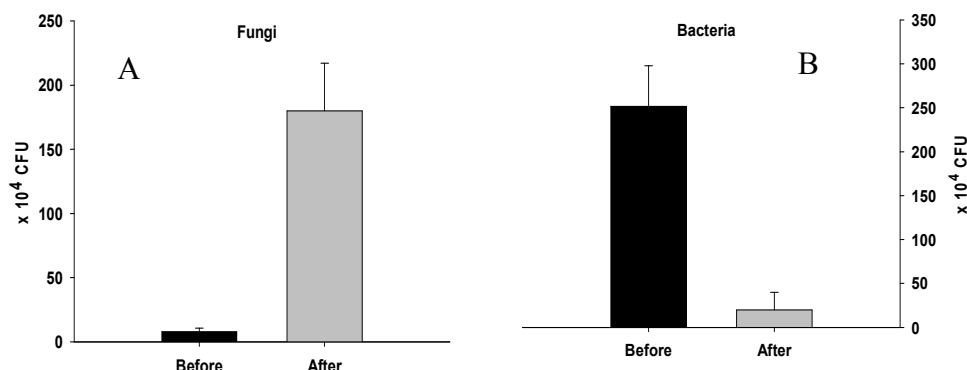
*F. solani*, *F. equiseti*, *F. dimerum* and *F. sambucinum* that did not show population values as high as *Fusarium oxysporum* in the soil of Greenhouse 1.

In the same way, the presence of oomycetes differs also depending on the soil considered (Table 7). In this case, in greenhouse 2 soil, which is rich in organic matter, there was presence of *Pythium* spp. (100% of presence) as well as *Phytophthora* spp. (40% of presence), while in the soil of Greenhouse 1, which is poor in organic matter, there was not presence of oomycetes.

However, on the contrary to the previous results shown, the total density of bacteria found in the soil of Greenhouse 1 (with low content of organic matter) was significantly higher ( $p$ -value<0,001 through Kruskal-Wallis test) than soil of Greenhouse 2 (with high content of organic matter) (Figure 1B).

*Greenhouse 1 before and after the incorporation of organic amendments through biodisinfection*

Total fungi density grown in greenhouse cucumber monoculture soil with low content of organic matter (Greenhouse 1) was significantly increased ( $p$ -value<0,001 through Kruskal-Wallis test) in the following season, after the first incorporation of organic amendments through biodisinfection (Figure 2A).



**Fig. 2** - Total fungi (A) and bacteria (B) density in the soils of Greenhouses 1 (soil with low content of organic matter and cucumber monoculture) Before and After the incorporation of organic amendments through biodisinfection. Values (average±typical deviation) expressed in  $10^4$  CFU·g<sup>-1</sup> of dry soil. \*\*\* ( $p$ <0,001) through Kruskal-Wallis test

In this case, fungal diversity was reduced from 5 genera present before the treatment to 3 genera detected after the treatment (Table 4). In this respect, it is noteworthy the shift of soil fungi genera which occurred after the treatment. In this sense, the genera that showed less importance before the treatment, *Acremonium* spp. and *Aspergillus* spp. stood out by their high population density after the treatment, and *Aspergillus* spp genus was the one stood out the most. On the other hand, after the treatment, there was no presence of *Fusarium* spp. genus fungi, which was the highest population density before biodisinfection was carried out.

Table 4

Population density of fungi genera present in the soils of Greenhouse 1 (soil with low content of organic matter and cucumber monoculture) Before and After the incorporation of organic amendments through biodisinfection. Values (average $\pm$ typical deviation) obtained from successive suspension-dilution technique and acidified malt extract agar, and expressed in  $10^4$  CFU·g<sup>-1</sup> of dry soil

Greenhouse 1	Before	After
% Organic matter	0,54	1,56
Genera	$\times 10^4$ CFU·g <sup>-1</sup>	
<i>Acremonium</i> spp.	0,1 $\pm$ 0,3	12,0 $\pm$ 10,3
<i>Aspergillus</i> spp.	0,1 $\pm$ 0,3	166,0 $\pm$ 37,8
<i>Fusarium</i> spp.	4,2 $\pm$ 2,3	--
<i>Penicillium</i> spp.	1,4 $\pm$ 1,1	--
Non Identified	0,7 $\pm$ 0,9	2,0 $\pm$ 4,2
Total Genera	5	3

Table 5

Population density of fungi species belonging to *Fusarium* spp. genera present in the soils of Greenhouse 1 (soil with low content of organic matter and cucumber monoculture) and of Greenhouse 2 (soil with high content of organic matter and tomato monoculture). Values (average $\pm$ typical deviation) obtained from Komada's selective medium (1975) modified by Tello et al. (1991) and expressed in CFU·g<sup>-1</sup> of dry soil

	Greenhouse 1	Greenhouse 2
% Organic matter	0,54	4,46
<i>Fusarium</i> sp.	CFU·g <sup>-1</sup>	
<i>F. oxysporum</i>	17552,8 $\pm$ 4215,0	--
<i>F. solani</i>	--	2622,0 $\pm$ 424,3
<i>F. equiseti</i>	--	1645,2 $\pm$ 170,3
<i>F. dimerum</i>	--	476,0 $\pm$ 108,4
<i>F. sambucinum</i>	--	78,3 $\pm$ 38,8
Total species	1	4

Table 6

Population density of fungi species belonging to *Fusarium* spp. genera present in the soils of Greenhouse 1 (soil with low content of organic matter and cucumber monoculture) Before and After the incorporation of organic amendments through biodisinfection. Values (average $\pm$ typical deviation) obtained from Komada's selective medium (1975) modified by Tello et al. (1991) and expressed in CFU·g<sup>-1</sup> of dry soil.

Greenhouse 1	Before	After
% Organic matter	0,54	1,56
<i>Fusarium</i> sp.	CFU·g <sup>-1</sup>	
<i>F. oxysporum</i>	17552,8 $\pm$ 4215,0	--
Total species	1	0

This last statement is corroborated by the results obtained in the assessments of the *Fusarium* flora through selective culture medium for *Fusarium* spp. genus (Table 6). Therefore, the high population of *Fusarium oxysporum*, which was present in the soil before the biodisinfection treatment, disappeared fully after the treatment.

The absence of oomycetes in greenhouse soil is clear after the treatment (Table 7).

Total density of bacteria borne by soil of Greenhouse 1 (with low content of organic matter) was significantly lower ( $p$ -value<0,001 through Kruskal-Wallis test) after biodisinfection treatment (Figure 2B).

Table 7

**Presence of oomycetes (*Pythium* spp. and *Phytophthora* spp.) in the soil of Greenhouse 1 (soil with low content of organic matter and cucumber monoculture) Before and After the incorporation of organic amendments through biodisinfection, and in soil of Greenhouse 2 (soil with high content of organic matter and tomato monoculture). Values expressed as a % presence.**

Oomycetes	Greenhouse 1 Before	Greenhouse 1 After	Greenhouse 2
<i>Pythium</i> spp. (% presence)	--	--	100
<i>Phytophthora</i> spp. (% presence)	--	--	40

The effect of soil fatigue on the decrease of plant development and/or production in horticultural crops is a topic that has not been dealt with very much in the current literature. For this reason, we have to turn to not very recent articles to find authors that, without referring to a specific pathogen action, confirm the existence of a fertility disorder of horticultural soils due to multiple reasons which can be accumulative, successive or simultaneous, and that it is translated into a complex syndrome that may cause yield losses, plant dwarfism, yellowing, underdevelopment and many others (Louvet, 1980; Bouhot 1983c, Tello *et al.* 2011). This phenomenon to which different authors linked to monoculture and/or repeated crops (Hoestra 1983; Massesse 1983; Sebillotte 1983; Messiaen *et al.* 1991; Cebolla & Maroto, 2004; Tello & Lacasa 2004), is not attributed to an exclusive cause due to its complexity, but it could have a close relationship with soil microbiota (Martínez *et al.* 2009; 2011), and therefore, with the levels of organic matter in cultivated soils, given that, the microbial community will depend on the available food. In this sense, the results obtained in the trials in a controlled environmental chamber with cucumber and tomato seedlings included in this study concluded higher vigour of plants grown in soils with supplies of organic amendments. In this case, in one of the studied soils (Greenhouse 1), the cucumber monoculture, which was repeated over time together, with the lack of supplies of organic matter, was translated into a deficient development of plants, possibly due to the adverse effects derived from the phenomenon of soil fatigue, which could be related with the nutritional state of the same (Bouhot 1983c, Chen *et al.* 1991). In this respect, since in this case the effects were not specific for cucumber plants (the lower development was also observed in tomato plants), in this soil, crop rotation, at least with tomatoes, would have not been enough to mitigate such effects. Likewise, in pepper monoculture developed in long-term cycles and very common in the fields of Cartagena, where organic amendments were not added to the soil, with the lack of pathogens, yield decreases were registered up to 60% of

the production. The reasons that caused these yield losses and were attributed to the phenomenon of soil fatigue were "covered up" for more than 15 years through the use of Methyl Bromide (Lacasa *et al.* 2002; Guerrero *et al.* 2004), an active matter that was withdrawn in Spain and the rest of developed countries in 2005, due to the commitments of the European Union with the Montreal Protocol.

On the other hand, the excellent development of the plants grown in the soil of the greenhouse where tomato monoculture was developed, but that added regularly organic amendments to the soil every year, make us think that the supplies of organic matter could be the reason of this "silencing" of the adverse effects derived from monoculture. Furthermore, this aspect is supported by the improvement observed in the plants grown in the first soil mentioned (very poor in organic matter) after the incorporation of different organic amendments through biosolarisation and by Guerrero *et al.*'s work (2014), in which the development of pepper plants grown in soil which showed specific fatigue on such crop, improved after applying biosolarisation treatments, reaching growths similar to those obtained in the treatments with Methyl Bromide. In this sense, it must also be taken into account that biosolarisation in greenhouse soils improves the physical and chemical characteristics of the same (Fernández *et al.* 2005; Núñez-Zofío *et al.* 2012).

Likewise, with reference to the microbiological content (fungi, bacteria and oomycetes) of the studied soils, soil with high content of organic matter, also had a higher amount and diversity of fungi than the soil in which the farmer did not add organic amendments. However, the bacterial population was significantly lower. In the same way, the incorporation of organic amendments through biosolarisation increased the total content of fungi in the soil with low content of organic matter, but a decrease was produced in the number of fungi genera as in the total content of bacteria. In this respect, the lower content of bacteria in the soils with higher amount of fungi, could be attributed to the inherent deficiencies of the analytical technique, since, probably the higher speed in the development and growth of some fungi genera compared with the bacterial community may limit the expression of this last one (Kirk *et al.* 2004). On the other hand, the decrease in the number of fungi genera after biosolarisation treatment may be attributed to the disinfecting power of the technique. In this sense, it is expected that only those genera with ability to resist such disinfecting power are present after the same. These same effects were found in Martínez *et al.*'s work (2009) in which general fungal microbiota, after different disinfection treatments (including biosolarisation), was reduced to *Aspergillus*, *Fusarium*, *Rhizopus* and *Penicillium* genera, being the most common and abundant *Aspergillus* and *Fusarium*. In this way, in our work, after the biosolarisation treatment, the presence of fungi belonging to *Aspergillus* spp is highlighted because it increased approximately 1000 times, compared with the population held before applying the treatment. Furthermore, it has to be considered that *Aspergillus* spp. are saprophytic fungi and decompose organic matter that participated actively in the nitrification



processes (Pateman *et al.* 1967), acting as the bacteria belonging to the *Nitrosomonas*, *Nitrosococcus* or *Nitrobacter* genera, among others, and played an important role in the leaching reduction of nitrates derived from the decomposition of organic matter. On the other hand, it is specially significant that the biodisinfection treatment carried out made completely inactive any *Fusarium oxysporum* propagule. In this way, during the campaign after the treatment, the cucumber plants grown in the greenhouse did not express any incidence of the disease caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum*, and showed higher vigour and yield (farmer's statement). Likewise, it must be highlighted that in the trials carried out in a controlled environmental chamber with the soil of that greenhouse before carrying out the biosolarisation treatment, disease was not expressed in the case of cucumber plants. Finally, the presence of oomycetes belonging to the genera *Pythium* spp. and *Phytophthora* spp. –recognised pathogens in horticultural crops, in the soil with high content of organic matter could be explained by a previous work (de Cara *et al.* 2009) carried out in the vicinity of the area in which the greenhouses, considered in this study, are located, and concluded that these microorganisms were introduced by irrigation water and passed from one greenhouse to another by contamination of the farm equipment. For this reason, farmers of this area use grafted plants, and therefore, these microorganisms appear in soil as common inhabitants.

## CONCLUSIONS

Considering the complexity inherent to "soil environment" and in order to understand better the changes occurred after the supply of organic matter through biodisinfection, soil must be considered as a "living thing" in which the shift of its dynamic balance (Tello *et al.* 2011) caused by monoculture and the lack of organic matter must be restored until reaching its regeneration ability, which will be closely linked to the final microbial balance and, therefore, to the availability of organic matter in the soil. For this reason, it would be advisable to supply fresh organic matter to compost in the soil repeatedly, every year at the end of the horticultural season in order to reach a new balance to avoid soil fatigue in plant development.

In summary, the results showed that fatigue appeared in the soil with low content of organic matter, which showed at the same time the lowest density and diversity of fungal population. Unlike the Guerrero *et al.*'s study (2014), in which the authors advised crop rotation to mitigate the effects of specific fatigue in pepper, in the tomato and cucumber monoculture soils of this study, the addition of fresh organic matter seems to reconstitute its productive capacity, and this mitigates the fatigue and monoculture effects. It is very significant that this aspect has not been dealt with before in the specialised literature consulted.

## REFERENCES

1. **Bodet JM., 1983** - *Fatigue des sols et céréalières et fourragères. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 37-43.
2. **Bouhot D., 1975** - *Recherches sur l'écologie des champignons parasites dans le sol.* Phytopathology, 7 (1). pp: 9-18.
3. **Bouhot D., 1979** - *Un test biologique à deux niveaux pour l'étude des fatigues de sol. Application à l'étude des nécroses des racines de céleri-rave.* Annales de Phytopathologie, 11 (1). pp: 95-109.
4. **Bouhot D., 1983a** - *État actuel des travaux sur la fatigue des sols en culture de betterave sucrière. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 29-36.
5. **Bouhot D., 1983b** - *Étude de la fatigue des sols dans les aspergeraies et le pépinières d'asperge. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 61-64.
6. **Bouhot D., 1983c** - *La fatigue des sols. Position du problème et principe du diagnostic. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 9-21.
7. **Bouhot D., Bonnel L., 1983** - *État actuel des travaux sur la fatigue des sols en culture de fraisier. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 71-75.
8. **Bouhot D., Dumas Y., 1983** - *Fatigue des sols en culture de tomates de plein champ. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 77-81.
9. **Casado C., 1925** - *El cansancio del suelo y la desinfección parcial.* Servicio de Publicaciones Agrícolas. Ministerio de Agricultura. Dirección General de Agricultura, Minas y Montes.
10. **Cebolla V., Maroto JV., 2004** - *La desinfección como medio de control de la fatiga del suelo.* Comunitat Valenciana Agraria 26, 21-26.
11. **Chen Y., Gamliel A., Stapleton JJ., Aviad T., 1991** - *Chemical, physical and microbial changes related to plant growth in disinfested soils. p.103-129. In: J. Katan and J.E. DeVay (eds.), Soil Solarization, CRC Press, Boca Raton, FL.*
12. **De Cara M., Peregrina I., Arellano J.A., Benavides J., Bueno M., Martínez A., Gómez J.M., Serrano Y., Guirado M.L., Rodríguez J.M., Santos M., Diánez F., Tello J.C., 2009** - *Patógenos asociados a la marchitez de tomate tipo "cherry" en la provincia de Granada. Estudio particular de las fuentes de inóculo de Phytophthora parasítica causante de la podredumbre del tallo.* Boletín de Sanidad Vegetal Vol.35, 2, 283-296.
13. **Fernández P., Guerrero M.M., Martínez M.A., Ros C., Lacasa A., Bello A., 2005** - *Effects of biofumigation plus solarization on soil fertility. Industrial crops and rural development. Proceedings of 2005 Annual Meeting of the Association for the Advancement of Industrial Croops,* 229-236.
14. **Gindrat D., Varady C., Neury G., 1983** - *Le déprérissement de l'asperge en Suisse romande. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 51-55.
15. **Guerrero M.M., Guirao P., Martínez-Lluch M.C., Tello J.C., Lacasa A., 2014** - *Soil fatigue and its specificity towards pepper plants in greenhouses.* Spanish Journal of Agricultural Research, 12(3), 644-652.
16. **Guerrero M.M., Lacasa A., Ros C., Martínez M.A., López J.A., Guirao P., Bello A., Torres J., Martínez M.C., González A., 2004** - *La reiteración de la biofumigación con solarización en la desinfección de suelos de invernaderos de pimiento. In:*

- Desinfección de suelos en invernaderos de pimiento.* Consejería de Agricultura, Agua y Medio Ambiente, Región de Murcia. Jornadas 16: 239-258.
17. Guirao P., Guerrero M.M., Ros C., Lacasa A., Beltrán C., Martínez M.C., Torres J., Oncina M., Contreras J., 2004 - *La reducción de dosis del bromuro de metilo en el cultivo de pimiento y el calendario de retirada.* In: *Desinfección de suelos en invernaderos de pimiento* (Lacasa A et al., eds). Consejería de Agricultura, Agua y Medio Ambiente, Región de Murcia. Jornadas 16: 61-78.
18. Hoestra H., 1983 - *La fatigue des sols en Hollande.* In: *La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 85-90.
19. Kirk J.L., Beaudette L.A., Hart M., Moutoglís P., Klironomos J.N., Lee H., Trevors J.T., 2004 - *Methods of studying soil microbial diversity.* Journal of Microbiological Methods, 58: 169-188.
20. Komada H., 1975 - *Development of a selective medium for quantitative isolation of Fusarium oxysporum from natural soil.* Plant Disease, 64: 450-454.
21. Lacasa A., Guerrero M.M., Ros C., Guirao P., Torres J., Bielza P., De Paco T., Contreras J., Molina R., Torné M., 2002 - *Desinfección del suelo de invernadero de pimiento con dicloropropeno+dicloropirina (Telopic EC). Dosis de aplicación y efectos del plástico de sellado.* Agrícola Vergel, 245: 256-266.
22. Lacasa A., Guirao P., Guerrero M.M., Ros C., Bello A., Bielza P., López J.A., 1999 - *Alternatives to methyl bromide for sweet pepper cultivation in plastic house in South-East Spain.* Proceedings 3rd International Workshop "Alternatives to Methyl Bromide for the Southern European Countries", December 7-10, Heraclion, Crete, Grecia: 41-44.
23. Louvet J., 1980 - *Maladies et fatigues de sol en cultures légumières.* Phytoma-Défense des Cultures 320: 19-21.
24. Maroto J.V., 2000 - *Elementos de horticultura general.* Ed. Mundi-prensa. 2ª edición. 424 pp.
25. Martínez M.A., Lacasa A., Tello J., 2009 - *Ecología de la microbiota fúngica de los suelos de los invernaderos y su interés agronómico.* Ministerio de Medio Ambiente y Medio Rural y Marino, Centro de publicaciones, Madrid, 374 pp.
26. Martínez M.A., Martínez M.C., Bielza P., Tello J., Lacasa A., 2011 - *Effect of biofumigation with manure amendments and repeated biosolarization on Fusarium density in pepper crops.* Journal of Industrial Microbiology & Biotechnology 38: 3-11.
27. Messiaen C.M., Blancard D., Rousell F., Lafon R., 1991 - *Les maladies des plantes maraîchères.* Ed: I.N.R.A. Paris.
28. Meynard J.M., Bouhot D., 1983 - *Confrontation de résultats du diagnostic au champ et du test biologique de fatigue des sols. Cas du blé.* In: *La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 45-49.
29. Núñez-Zofío M., Larregla del Palacio S., Garbisu C., 2012 - *Repeated Biodisinfection Controls the Incidence of Phytophthora Root and Crown Rot of Pepper While Improving Soil Quality.* Spanish Journal of Agricultural Research 10 (3): 794-805.
30. Otto G., Winkler H., Szabó K., 1994 - *Proof of actinomycetes in rootlets of species of Rosaceae from a SARD soil. A contribution to the specificity of replant diseases.* Acta Horticulturae 363: 43-48.
31. Pateman J.A., Rever B.M., Cove D.J., 1967 - *Genetic and Biochemical studies of nitrate reduction in Aspergillus nidulans.* Biochemical Journal 104 (1): 103-111.
32. Rodríguez-Molina M.C., 1996 - *Ensayo de caracterización de suelos agrícolas y forestales de Extremadura tomando como indicadores a Fusarium Link y Pythium Pringsheim: la representatividad del muestreo.* PhD thesis, EPSIA Madrid, Madrid, 209 pp.

33. Roudeillac P., 1983 - *Les dépérissements du fraisier et la fatigue du sol. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 65-70.
34. Scotto C., 1983- *Mesures susceptibles d'apprécier et de limiter les effets de la fatigue des sols dans le cas de reconstitution des vergers. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 99-111.
35. Sebillotte M., 1983 - *Potentialité du milieu et fatigue des sols. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris. 115-122.
36. Tello J.C.; Lacasa A., 2004 - *Las enfermedades de origen edáfico y su control en los pimentonales del Campo de Cartagena. Una interpretación retrospectiva del sexenio 1979-1985". Desinfección de suelos en invernaderos de pimientos.* Ed.: Comunidad Autónoma de la Región de Murcia, pp 11-26.
37. Tello J.C., Palmero D., de Cara M., Moreno A., Santos M. 2011 - *El suelo como "ente vivo" y su relación con las enfermedades de las plantas. In: Meco, R., Lacasta C., Moreno, M.M. (coords.) Agricultura Ecológica en Secano. Soluciones sostenibles en ambientes mediterráneos.* Ministerio de Medio Ambiente y Medio Rural y Marino, Junta de Comunidades de Castilla-La Mancha, SEAE, Ediciones
38. Tello J.C., Vares F., Lacasa A., 1991 - *Análisis de muestras, 39-48. In: Manual de laboratorio. Diagnóstico de hongos, bacterias y nematodos fitopatógenos.* MAPA, Madrid, 485 pp.
39. Vigouroux A., 1983 - *Fatigue de sol et accidents culturaux en arboriculture fruitière. In: La fatigue des sols. Diagnostic de la fertilité dans le systèmes culturaux.* Ed: INRA Paris, pp 91-97.

**Consilier editorial:**

**Vasile VÎNTU**

**Tehnoredactori:**

**Liliana ROTARU  
Liliana Elena CHELARIU**

**Corectori:**

**Lucia DRAGHIA  
Liliana ROTARU**

**Bun de tipar:**

**15.06.2015**

**Apărut:**

**2015**

**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:**

**Liliana ROTARU  
Liliana Elena CHELARIU**

**Readers:**

**Lucia DRAGHIA  
Liliana ROTARU**

**Imprimatur:**

**15.06.2015**

**Published:**

**2015**

**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**