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

**UNIVERSITATEA DE ŞTIINȚE AGRICOLE
ŞI MEDICINĂ VETERINARĂ
"ION IONESCU DE LA BRAD" IAŞI**

LUCRĂRI ŞTIINȚIFICE

Vol. 57

NR. 1

SERIA HORTICULTURĂ

EDITURA "ION IONESCU DE LA BRAD"

IAŞI 2014

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

"Ion Ionescu de la Brad" Publishing House Iaşi
ISSN–L=1454-7376
(Print)-ISSN 1454-7376
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(CD-ROM) = ISSN 2069 – 847X

CONTENT

CUPRINS

CHAOS CONTROL OF CHAOTIC CHEMICAL SYSTEMS

CONTROLUL SISTEMELOR CHIMICE HAOTICE

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Abstract. *Chemical systems can exhibit chaotic behaviour and this fact is very important for chemical processes and for biological structures. From this point of view the control of these phenomena have a great practical impact despite the fact that it is very dificult; this is the reason the theoretical models are useful in these situations. The control using these models can give the informations about the selfcontrol inside the biological structures where the behaviour of the dynamic systems is realized by a feedback mechanism. The main aim of this paper is to study the synchronization of two chemical chaotic systems proposed by Samardzija and which is based on 9 rections and 3 intermediary species, using the adaptive feedback method of control. The transient time until synchronization depends on initial conditions of two systems, the strength and the number of the controllers.*
Key words: *chaotic chemical system, chaos control*

Rezumat. *Sistemele chimice pot avea comportare haotică şi acest fat este foarte important pentru procesele chimice şi structurile biologice. Din acest punct de vedere controlul acestor fenomene are un mare impact in ciuda faptului că este foarte dificilş acesta este motivul pentru care modelele teoretice sunt utile în aceste situaţii. Contolul sistemelor pe baza acestor modele poate da informaţii despre autocontrolul din structurile biologice unde comportarea sistemelor dinamice se realizează printr-un mecanism de feedback. Scopul principal al acestei lucrări este de a studia sincronizarea a două sisteme chimice propuse de Samardzija care se bazează pe 9 reacţii şi 3 specii intermediare, folosind o metodă de control de tip feedback. Timpul după care se obţine sincronizarea depinde de condiţiile initiale ale celor doua sisteme si de intensitatea controler-ului.*
Cuvinte cheie: *sistem chimic haotic, controlul haosului*

INTRODUCTION

Chemical reaction systems have become one of the favorite domains to study nonlinear systems, both experimentally and theoretically. These systems can exhibit chaotic behaviour and this fact is very important for chemical processes and for biological structures. From this point of view the deliberate control of these phenomena have a great practical impact despite the fact that it is very dificult; this is the reason the theoretical models are useful in these situations. In

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addition, the control using these models can give the informations about the selfcontrol inside the biological structures where the behaviour of the dynamic systems is realized by a feedback mechanism. Over the last decade, there has been considerable progress in generalizing the concept of synchronization to include the case of coupled chaotic oscillators especially from technical reasons. When the complete synchronization is achieved, the states of both systems become practically identical, while their dynamics in time remains chaotic. Many examples of synchronization have been documented in the literature, but currently theoretical understanding of the phenomena lags behind experimental studies (Grosu, 1997; Grosu et al., 2008; Lerescu et al., 2004; Lerescu et al., 2006; Oancea et al., 2009; Oancea et al., 2011). The main aim of this paper is to study the synchronization of two chemical chaotic systems based on the adaptive feedback method of control. One of these chemical models was proposed by Samardzija and it is based on 9 rections and 3 intermediary species.

THEORY

The model proposed by Samardzija represents some chemical reactions and its mechanism consists in the following elementary steps (Wang and Chen, 2010):

1. $A_1 + X + 2Z \xleftrightarrow{k_1} X + 3Z$
2. $X + Y \xleftrightarrow{k_2} 2Y$
3. $A_2 + X + Z \xleftrightarrow{k_3} Z + P_1$
4. $A_3 + X + Z \xleftrightarrow{k_4} X + P_2$
5. $A_4 + 2Y \xleftrightarrow{k_5} 3Y$
6. $2Z \xleftrightarrow{k_6} P_3$
7. $A_5 + X \xleftrightarrow{k_7} 2X$
8. $Y \xleftrightarrow{k_8} P_4$
9. $A_6 + Z \xleftrightarrow{k_9} 2Z$

The time evolution of the intermediary species X, Y , and Z is given by a nonlinear system of equations:

$$\begin{aligned}
\frac{dx_1}{dt} &= -k_2 x_1 x_2 - k_3 x_1 x_3 + k_7 x_1 \\
\frac{dx_2}{dt} &= k_2 x_1 x_2 + k_5 x_2^{\,2} - k_8 x_2 \\
\frac{dx_3}{dt} &= k_1 x_1 x_3^{\,2} - k_4 x_1 x_3 - 2k_6 x_3^{\,2} + k_9 x_3
\end{aligned} \tag{1}$$

This system has a chaotic behaviour, for the following constants:

$k_1 = 1$ $k_2 = 2$ $k_3 = 1.5$ $k_4 = 20$ $k_5 = 0.8$

k_6=12.85 k_7 = 45 k_9=514.2

1. Chaotic dynamics of chemical system

Coosing k_8 as a control parameter, we can know dynamics of this system.

For k_8 =50 the strange attractor for this system is given in the figure 1.

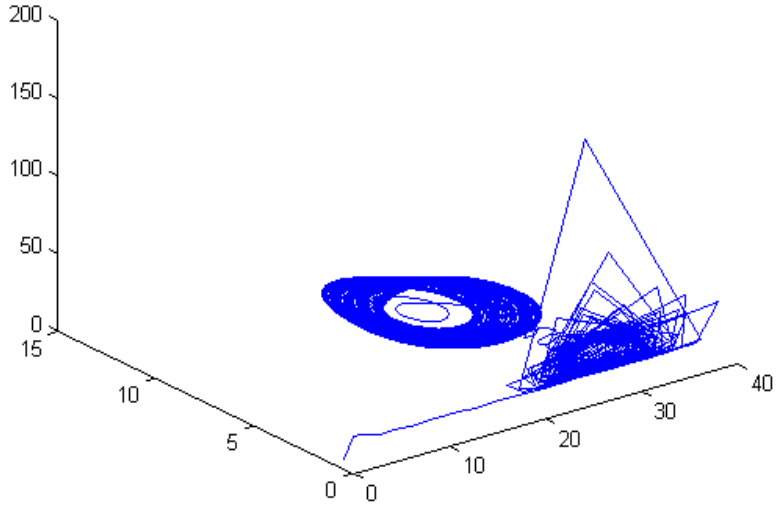
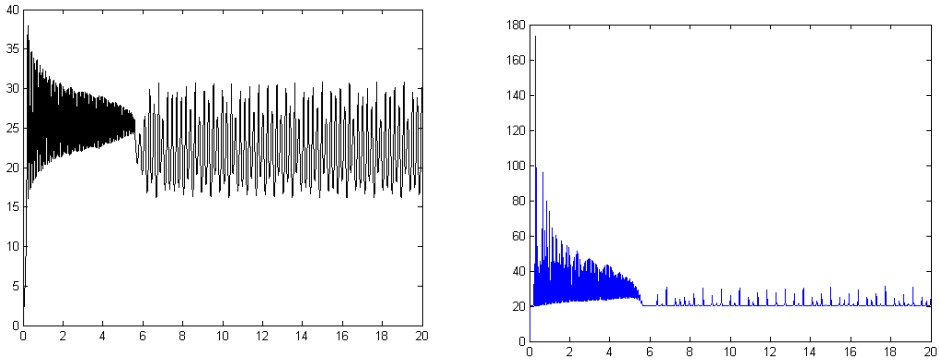


Fig. 1 – Phase portrait of (x_3, x_1, x_2,) for system (1) with initial conditions 1 1 1

The dynamics of the this chaotic chemical system is given in figure 2.



a

Fig. 2 – a-x_1(t); b- x_3(t) for k_8 =50;

The chaotic behavior is sustained by Lyapunov exponents from figure 3.

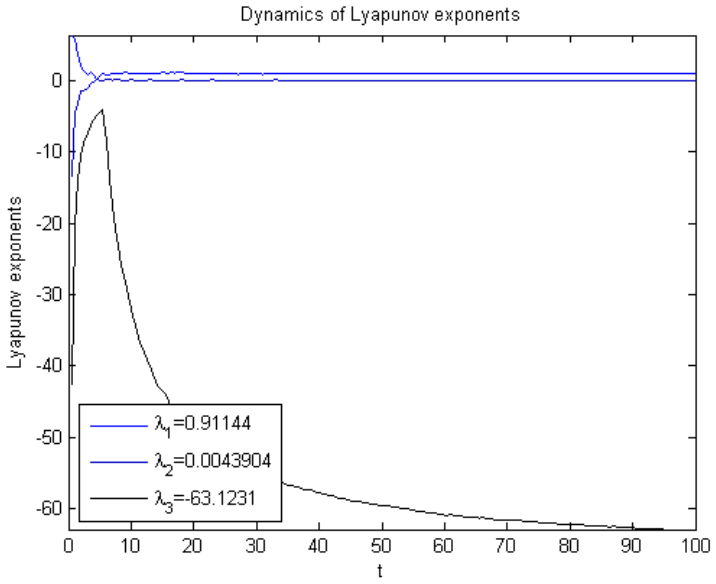


Fig. 3 – The Lyapunov exponents

This system is very sensitive to initial condition. Then we choose k_8 =43

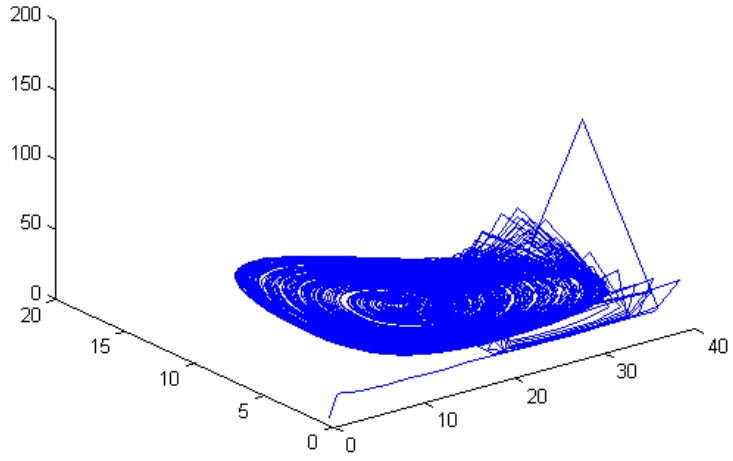
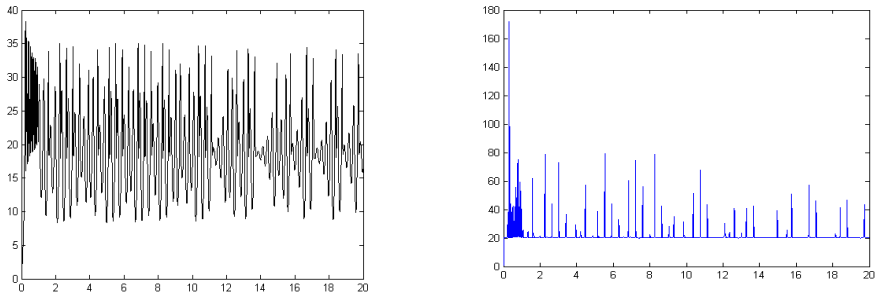


Fig. 4 – 3D attractor (x_3, x_1, x_2) for k_8 =43

Figure 5 shows the changes of the variable x and z with time for k_8 =43.



a b

Fig. 5 – a-x_1(t); b- x_3(t) for k_8 =43;

2. Synchronization of two chaotic systems

To synchronize two identical chemical syststems we followed the method proposed by Guo et al. [8], Hu and Xu [9], based on Lyapunov-Lasalle theory. Let be a chaotic non-autonomous system:

$$\dot{x} = f(x,t) \quad \text{where } x = (x_1, x_2 \ldots)^T \in R^n$$

is the state vector of the system and $f = (f_1, f_2 ..)^T \in R^n$ is the non-linear vector field of the system, which is considered as a driving system.

For any $x = (x_1, x_2 \ldots)^T \in R^n$ and $y = (y_1, y_2 \ldots)^T \in R^n$

there exists a positive constant l such that:

$$\left| f(x,t) - f(y,t) \right| \le l \max \left| x_i - x_j \right| \quad \text{i, j=1,2...n}$$

The slave system will be: $\dot{y} = f(y,t) + z(z_1, z_2, \ldots)$ where $z(z_1, z_2, \ldots)$ is the controller. If the error vector is e = y - x, the objective of synchronization is to make

$$\lim \left\| e(t) \right\| \to 0$$

$$t \to +\infty$$

The controller is of the form: $z_i = \varepsilon_i (y_i - x_i)$ and $\dot{\varepsilon}_i = -\gamma_i e_i^2$, i=1,2...n

and γ_i , i = 1,2,. . . , n are arbitrary positive constants.

RESULTS AND DISCUSSION

According this method of synchronization, the slave system for this chemical system will be:

$$\frac{dy_1}{dt} = -2y_1y_2 - 1.5y_1y_3 + 45y_1 + z_1(y_1 - x_1)$$

$$\frac{dy_2}{dt} = 2y_1y_2 + 0.8{y_2}^2 - k_8y_2 + z_2(y_2 - x_2)$$

$$\frac{dy_3}{dt} = y_1{y_3}^2 - 20y_1y_3 - 25.7{y_3}^2 + 514.2y_3 + z_3(y_3 - x_3) \qquad (2)$$

and for the control strength:

$$\dot{z}_1 = -(y_1 - x_1)^2$$
$$\dot{z}_2 = -(y_2 - x_2)^2$$
$$\dot{z}_3 = -(y_3 - x_3)^2 \qquad (3)$$

Figures 6-9 demonstrate the syncronization of the two chemical systems.

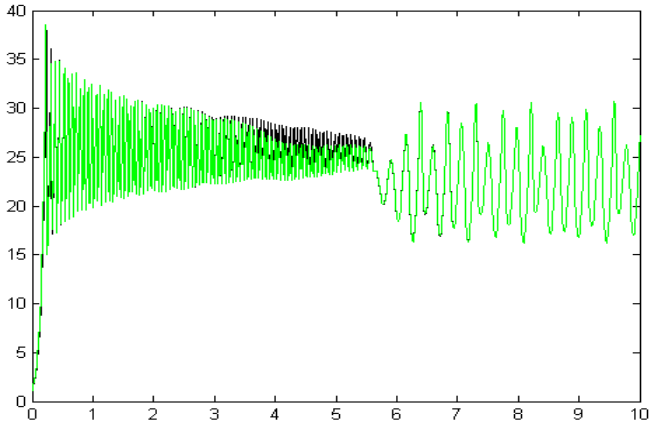


Fig. 6 – x1(t)- black y1(t)- green[$x_1(0)$=1, $x_2(0)$=1, $x_3(0)$=1; $y_1(0)$ =1.1; $y_2(0)$ = 1.1 $y_3(0)$ = 1.1; $z_1(0)$ =1; $z_2(0)$ = 1 ; $z_3(0)$ =1]

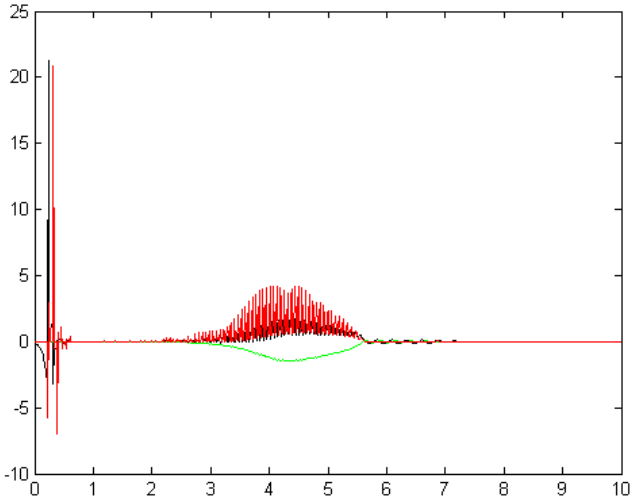
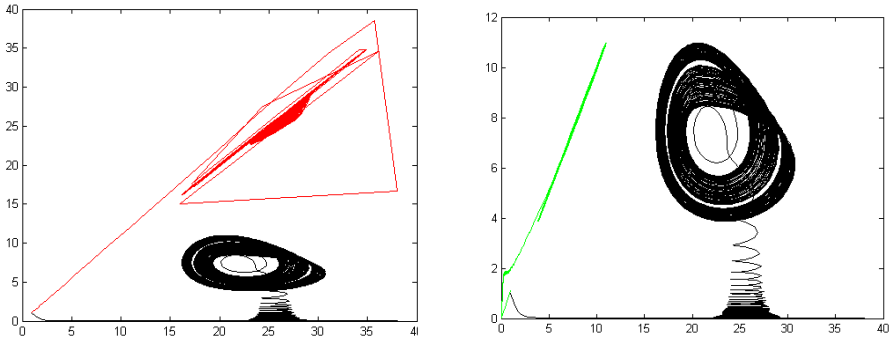


Fig. 7 – Synchronization errors between master and slave systems [$x_1(0)$=1, $x_2(0)$=1, $x_3(0)$=1; $y_1(0)$ =1.1; $y_2(0)$ = 1.1 $y_3(0)$ = 1.1; $z_1(0)$ =1; $z_2(0)$ = 1 ; $z_3(0)$ =1]



a b

Fig. 8 – Phase portrait of a) :(x_1, x_2-black) and (x_1,y_1-red); b): (x_1, x_2-black) and (x_2,y_2-green) for two systems [$x_1(0)$=1, $x_2(0)$=1, $x_3(0)$=1; $y_1(0)$ =1.1; $y_2(0)$ = 1.1 $y_3(0)$ = 1.1; $z_1(0)$ =1; $z_2(0)$ = 1 ; $z_3(0)$ =1]

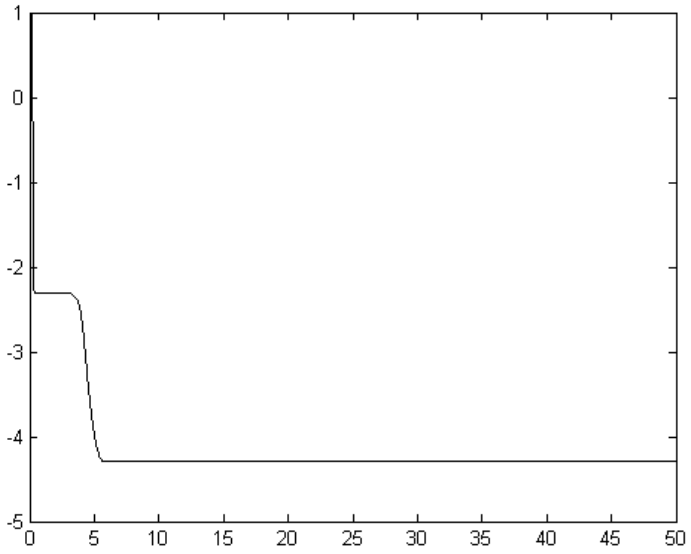


Fig. 8 – The control strength z_1[$x_1(0)$=1, $x_2(0)$=1, $x_3(0)$=1; $y_1(0)$ =1.1; $y_2(0)$ = 1.1 $y_3(0)$ = 1.1; $z_1(0)$ =1; $z_2(0)$ = 1 ; $z_3(0)$ =1]

Debin Huang (2005), by testing the chaotic systems including the Lorenz system, Rossler system, Chua's circuit, and the Sprott's collection of the simplest chaotic flows found that we can use a single controller to achieve identical synchronization of a three-dimensional system (for Lorenz system this is possible only we add the controller in the second equation).

For these systems we achieved the synchronization if one controller is applied only in the first or in the second equation (fig. 9).

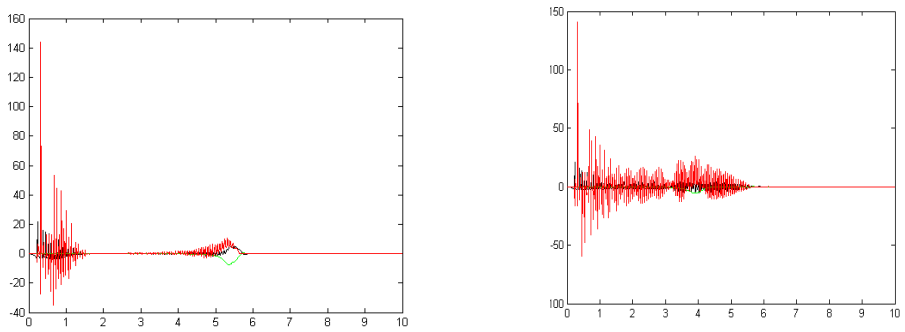


Fig. 9 – Synchronization errors between master and slave for chemical systems with one controller [$x_1(0)=1$, $x_2(0)=1$, $x_3(0)=1$; $y_1(0)=1.1$; $y_2(0)=1.1$ $y_3(0)=1.1$; a- $z_1(0)=1$; b- [$x_1(0)=1$, $x_2(0)=1$, $x_3(0)=1$; $y_1(0)=1.5$; $y_2(0)=1.5$ $y_3(0)=1.5$; $z_2(0)=1$]

CONCLUSIONS

In this work we analized the dynamics of the Samardzija system wich is based on 9 rections and 3 intermediary species and we realized the synchronization of two systems using an adaptive feedback method. The transient time until synchronization depends on initial conditions of two systems, the strength of the controllers and their number. Then we can control this chemical system in accordance with recent debates of Wang and Chen (2010) about full global synchronization and partial synchronization in a system of two or three coupled chemical chaotic oscillators.

REFERENCES

1. **Grosu I., 1997** -*Robust Synchronization*, Phys. Rev. 56, pp. 3709-3712
2. **Grosu I., Padmanaban E., Roy P. K., Dana S. K., 2008** - *Designing Coupling for Synchronization and Amplification of Chaos*, Phys Rev Lett 100, 234102, pp. 1-4
3. **Lerescu A.I., Constandache N., Oancea S., Grosu I., 2004** - *Collection of master-slave synchronized chaotic systems*, Chaos Soliton Fract., 22(3), pp. 599-604
4. **Lerescu A.I., Oancea S., Grosu I., 2006 -** *Collection of Mutually Synchronized Chaotic Systems,* Physics Letters A, 352, pp. 222-228.
5. **Oancea S., Grosu F., Lazar A., Grosu I., 2009 -** *Master–slave synchronization of Lorenz systems using a single controller*, Chaos, Solitons and Fractals, 41, pp. 2575-2580
6. **Oancea S., Grosu I., Oancea A.V., 2011 -** *Biological control based on the synchronization of Lotka-Volterra systems with four competitive species*, Rom. J. Biophys, 21(1), pp. 17-26
7. **Wang J.W., Chen A.M., 2010 -** *Partial synchronization in coupled chemical chaotic oscillators*, Journal of Computational and Applied Mathematics, 233, pp. 1897-1904
8. **Guo W., Chen S., Zhou H., 2009 -** *A simple adaptive-feedback controller for chaos Synchronization*, Chaos, Solitons and Fractals, 39, pp. 316–321
9. **Hu M., Yang Y., Xu Z., Guo L., 2008 -** *Hybrid projective synchronization in a chaotic complex nonlinear system*, Mathematics and Computer in Simulation, 79, pp. 449-457
10. **Huang D., 2005 -** *Simple adaptive-feedback controller for identical chaos synchronization*, Phys. Rev. E, 71, 037203

SYNTHESIS OF SULFOCHLORIDE DERIVATIVES OF THE ARYL OXYALKYL CARBOXYLIC ACIDS AS INTERMEDIATES IN OBTAINING COMPOUNDS WITH BIOLOGICAL POTENTIAL

SINTEZA SULFOCLORURILOR ACIZILOR ARIL-OXIALCHIL CARBOXILICI CA INTERMEDIARI ÎN OBȚINEREA UNOR COMPUȘI CU POTENȚIAL BIOLOGIC

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Abstract. *Aryl - oxyalkyl carboxylic acids and their derivatives are compounds with high biologic potential, having various pharmacological properties or auxin - type growth regulator action. The pharmacological tests determined that the presence of substituted or unsubstituted sulphonamidic groups in the phenoxyacetic derivatives confers them a toxicity which in most cases is negligible; they also have high bioavailability and can be used as effective growth stimulants for various plant species at low concentrations. Since the R_1, R_2 - substituted phenoxyacetic acid esters and their sulfochlorides are intermediates used in the synthesis, this paper presents the general scheme of the process for the chlorosulfonation of the considered aryl - oxyalkyl carboxylic esters, the mechanism of the reaction, the obtaining method and the sulfochlorides yields for the methyl esters of fenil-1,2-dioxiacetic, fenil-1,3-dioxiacetic şi fenil-1,4-dioxiacetic acids in the reaction with the chlorosulfonic acid.*
Key words: *phenoxyacetic, methyl esters, sulphochloride, growth regulator*

Rezumat. *Acizii aril-oxialchil carboxilici şi derivaţii lor fac parte dintre compuşii chimici cu un potenţial biologic ridicat, având diverse proprietăţi farmacologice sau regulatoare de creştere de tip auxinic. Prin teste farmacologice efectuate asupra compuşilor sintetizaţi s-a determinat că prezenţa grupărilor sulfonamidice, substituite sau nesubstituite, în moleculele derivaţilor fenoxiacetici conferă produselor finale o toxicitate de cele mai multe ori neglijabilă; de asemenea, au o biodisponibilitate ridicată şi pot fi folosite ca produse stimulatoare de creştere eficiente pentru diverse specii de plante, în concentraţii mici. Deoarece atât esterii acizilor fenoxiacetici R_1,R_2-substituiţi cât şi sulfoclorurile acestora sunt intermediari folosiţi în sinteză, în această lucrare au fost realizate studii referitoare la schema generală a procesului de clorosulfonare pentru esterii aril-oxialchil carboxilici luaţi în considerare, mecanismul de reacţie, metoda de obţinere şi randamentele în sulfocloruri pentru esterilor metilici ai acizilor fenil-1,2-dioxiacetic, fenil-1,3-dioxiacetic şi fenil-1,4-dioxiacetic în reacţia cu acidul clorsulfonic.*
Cuvinte cheie: *fenoxiacetic, esteri metilici, sulfoclorură, regulator creştere*

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The aryl - oxyalkyl carboxylic acids and their derivatives are chemical compounds with high biologic potential, having various pharmacological properties. Numerous compounds from this class enter into the composition of drugs with different use:

- the radical of the 2,3-dichloro phenoxyacetic acid is contained in the structures of two diuretic drugs: *Tricrinafen* and *Edecrin*;

- the radical of the 2,6-dichloro phenoxyacetic acid enters in the structure of *Lofexidine*, with central antihypertensive action;

- various anti-inflammatory drugs have phenoxyacetic or α-phenoxypropionic structure: *Ibufenac* (derives from phenoxyacetic acid), *Paduden* (derives from α-phenoxypropionic acid), *Aclofenac*, *Fenclofenac*, *Fenprofen*, *Percluson*;

- A number of products with antibacterial activity have in their structure phenoxyacetic radicals (*Penicillin V*), α-phenoxypropionyl (*Feneticiline*) and α-phenoxybutyryl (*Propiciline*);

- *Meclofenoxat* with a psycho energizing action is a derivative of p-chloro phenoxyacetic acid, as well as Iproclozide, used as an analeptic for the thymus (Oniscu, 1988);

- a number of derivatives of phenoxyisobutyric acid have lipid-lowering properties: *Beclobrat*, *Clofibrat*, *Fenofibrat*, *Teofibrat*;

Some phenoxyalkyl carboxylic derivatives have remarkable applications in agriculture, like the acids: 2,4- dichloro phenoxyacetic (2,4-D acid), 2,4,5-trichloro phenoxyacetic (2,4,5-T acid) with selective herbicide action and α-(2,4,5-trichloro phenoxy)-propionic acid (2,4,5-TP acid) used as selective herbicide in corn and cotton cultures.

Used in small doses, these products have growth regulating action. In case of exceeding the effective dose, phenoxyacetic acids turn into defoliants. For example, the 1 : 1 mixture between 2,4,5-T acid and 2,4-D acid butyl esters was used by U.S.A. as defoliant in the Vietnam War, under the name of "orange agent" (Neamtu and Irimie, 1991).

The α-(2-methyl, 4-chloro phenoxy)-propionic acid (2M-4CP acid) destroys weeds resistant to other herbicides, being used in cereal crops protection.

The α-(2,4-dichloro phenoxy)-propionic acid (2,4-DP acid) stimulates fruit growth and prevents their fall before harvest (Comanita et all., 1986).

A series of γ-phenoxybutyric acids are highly selective herbicides: γ-(2,4-dichloro phenoxy)-butyric acid (2,4-DB acid) or γ-(2-methyl, 4-chloro phenoxy)-butyric acid (MCPB acid) etc.

C. Oniscu and coworkers synthesized a series of esters, amides and hydrazides of the phenoxyalkyl carboxylic acids, as well as derivatives of the phenoxyalkyl carboxylic acids with sulphonamide group (Oniscu, 1968; Botez and Oniscu, 1972), which represent a new class of growth stimulators. From this class of compounds, two substances with auxinic action were tested in sugar beet, carrots, grapevine, and roses cultures, with remarkable results, allowing the

product 2-sulphonamide, 4-chloro phenoxy-acetic acid's approval as sugar beet crop growth stimulator under the name of ASFAC.

Pharmacological tests performed on other novel compounds synthesized by the same group determined that their toxicity is almost zero; also they have a high bioavailability and remarkable neurostimulation, antidepressants and anticonvulsants properties (Nigovic et all., 1996).

A particularly valuable compound obtained from research in the class of phenoxyalkyl carboxylic sulphonamides derivatives is dimethylaminoethyl hydrochloride ester of the 2-chloro-4-sulphonyl dimethylamido phenoxyacetic acid (Romener), an efficient metabolic regulator of nerve cell, energizer, antidepressant and antipsychotic.

MATERIAL AND METHOD

The general scheme for obtaining the derivatives of aryl- oxyalkyl carboxylic sulphonamides comprises the following steps:

- R-phenoxyacetic acids obtained from the corresponding phenols by condensation with monochloroacetic acid in an alkaline solution (NaOH);
- obtaining the methyl, ethyl etc. esters of these acids;
- esters' chlorosulfonation;
- chlorosulfonated esters' condensation with ammonia, substituted amines or other compounds with amino groups (Dumitrascu, 1998)

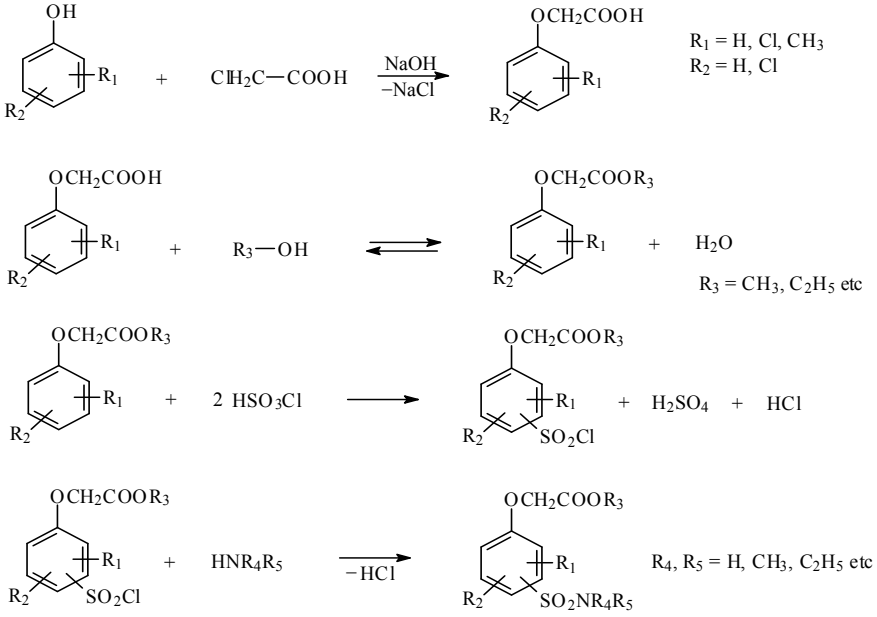

Fig. 1 - The reaction scheme for obtaining sulphonyl amido-phenoxyacetic derivatives

The same steps of the general scheme for obtaining the derivatives of aryl oxyalkyl carboxylic acids sulphonamides are followed also to synthesize compounds

containing in their structure two oxyacetic groups grafted in various positions of the aromatic nucleus.

Since the R1, R2-substituted phenoxyacetic acid esters and their sulfochlorides are intermediates used in the synthesis of all the compounds prepared by the sequence of reactions described above, studies were conducted on obtaining them.

Chlorosulfonation reaction mechanism, according to the literature (7), is shown in the diagram below (fig. 2) indicating that in the first step takes place the sulphonation with SO_3 and in the next step the sulphonic acid is converted to sulphochloride.

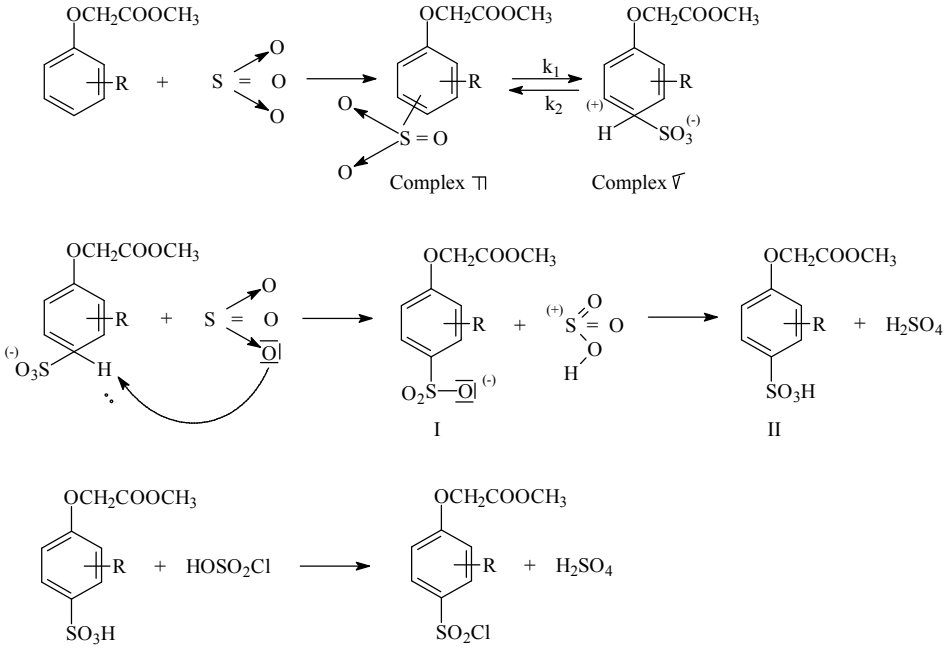

Fig. 2 - Chlorosulfonation reaction mechanism

Obtaining the sulfochlorides of the phenoxyacetic and phenyl-dioxyacetic acids methyl esters was performed according to the scheme shown in figure 1.

The general procedure for preparation is as follows:

- over 0.7 moles of chlorosulfonic acid cooled to 0 - 50°C are added in small portions, with continuous stirring, 0.1 moles methyl ester, so that the temperature does not exceed 5^0C;
- after the addition of the ester, the reaction mixture is maintained at the same temperature 30 - 40 min and then the temperature is raised to values specific to the type of the ester, maintaining the temperature for 90 - 100 minutes, when the formation of the sulphochloride takes place;
- finally, the mixture is cooled to 5 - 6^0C and poured into a mixture of water and ice, under vigorous stirring, to destroy the unreacted chlorosulfonic acid and to precipitate the sulphochloride;
- the obtained sulfochloride is filtered, water washed until pH = 6.5, is recrystallized from a mixture of water - acetone (2: 1 volume ratio) or benzene, then is dried at temperatures $\leq 40^0$C.

RESULTS AND DISCUSSIONS

According to the literature data on the chlorosulphonation of the aryl-oxyacetic acids (Oniscu, 1968), the chlorosulphonation of the phenyl dioxyacetic esters with chlorosulphonic acid is a process of pseudo equilibrium which can be kinetically described by the equation:

$$\frac{C_{SCl}}{C_{E_S}} = \frac{k_1}{k_2} \frac{C_{E_0}(M-1) - C_{SCl}}{C_{C_0} + C_{SCl}} \qquad (5)$$

in which: C_{SCl}=momentary concentration in sulphochloride (moles/l); C_{Es} = momentary concentration in sulphonic ester (moles/l); C_{Eo} = initial concentration of the ester (moles/l); C_{Co}=initial concentration of the sulfuric acid in the chlorosulphonic acid; M=the ratio between the concentration of the chlorosulphonic acid and the ester's concentration; k_1=rate constant for the formation of sulphochloride (l/moll·h); k_2=rate constant for the transformation of sulphochloride into sulphonic acid, determined by the sulfuric acid (l/mol · h).

Also, the literature (Oniscu, 1968) states that the molar ratio between the aryl-oxyalkyl carboxylic esters and the chlorosulfonic acid in the process of chlorosulphonation is 1:7. Based on these data, we initially set the optimal value in the chlorosulphonation process of the phenyl dioxyacetic esters of 1: 6. Under these circumstances, we observed the influence of reaction temperature on the chlorosulphonation of the phenyl dioxyacetic esters.

The chlorosulphonation of the ***phenyl-1,2-dioxyacetic acid's methyl ester*** was done by treating 0.6 mole of chlorosulphonic acid to 0.1 moles ester at a temperature of 0 - 5^0C. After merging the reactants, we raised the temperature at different values and then maintained it for one hour. Finally, the reaction mixture was diluted with ice + water mixture when the sulphochloride ester is separated.

We worked with temperature between 15 – 35^0C, and the obtained results are presented in table 1.

Table1.

Temperature (^{0}C)	15	20	25	30	35
η (%)	55	75	86	94	88

The chlorosulphonation of the ***phenyl-1,3-dioxyacetic acid's methyl ester*** followed the same steps described above, with the completion of the reaction at temperatures in the range of 10 – 25^0C. The obtained results are presented in table 2.

Table 2.

Temperature (^{0}C)	10	15	20	25
η (%)	60	85	92	89

The chlorosulphonation of the ***phenyl-1,4-dioxyacetic acid's methyl ester*** was also carried out under the conditions shown above and the completion of the reaction was carried out at temperatures between 35 – 60^0C, for one hour. The obtained results are presented in table 3.

Table 3.

Temperature (^{0}C)	35	40	45	50	60
η (%)	32	60	72	80	75

Representing graphically using the coordinates η - t^0C, the data presented in the tables above shows that for each case there is an optimum temperature range (fig. 3).

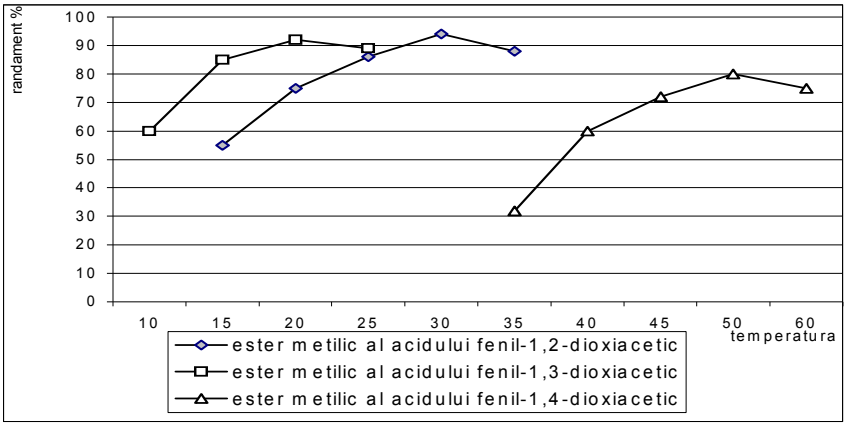

Fig. 3 - The variation of efficiency with temperature in the chlorosulphonation process

The minimum reaction temperature, of approximately 20^0C, is noted for the phenyl-1,3-dioxyacetic ester, fact explained by the concurring orientation of the two oxyacetic acid groups existing in the phenyl ring.

CONCLUSIONS

1. The molar ratio between the reactants in the chlorosulphonation process was established at 1:6 (dioxyacetic ester:chlorosulphonic acid);

2. The optimum reaction temperatures for the three obtained esters were: 30^0C for the phenyl-1,2-dioxyacetic acid's methyl ester when we obtained the best efficiency value – 94%, 20^0C for the phenyl-1,3-dioxyacetic acid's methyl ester when we obtained the highest efficiency value of 92% and 50^0C for the phenyl-1,4-dioxyacetic acid's methyl ester when we obtained 80% efficiency.

REFERENCES

1. Botez Gh., Oniscu C.,**1972** - *Brevet RO nr. 56123*

2. Comăniță Eugenia, Șoldea Camelia, Dumitrescu Elena, **1986** - *Chimia și tehnologia pesticidelor*. Editura Tehnică, București.

3. Dumitrașcu Adina,**1998** – *Teză de doctorat*, Univ. Tehnică "Gh. Asachi" Iași

4. Neamțu G., Irimie F., **1991** - *Fitoregulatori de creștere – Aspecte biochimice și fiziologice,* Editura Ceres, București.

5. Nigovic B., Kojik-Prodic Biserka, Antolic S., Tomic Sanja, Puntarec V., and Cohen J.D., **1996** - *Structural Studies on Monohalogenated Derivatives of the Phytohormone Indole-3-acetic Acid(Auxin)*, Acta Cryst., B52, 332-343.

6. Oniscu C., **1968** - *Cinetica reacției de clorosulfonare a esterilor acizilor clor-fenoxiacetici,* Bul. Instit. Polit. Iași, 8 (XIX), 231

7. Oniscu C., **1988** – *Chimia și tehnologia medicamentelor*, Ed. Tehnică, București

PYRROLOPYRIDAZINE DERIVATIVES SUBSTITUED WITH FLUOR: SYNTHESIS AND FLUORESCENT PROPRIETIES

DERIVAŢI PIROLOPIRIDAZINICI SUBSTITUIŢI CU FLUOR: SINTEZĂ ŞI STUDIUL PROPRIETĂŢILOR FLUORESCENTE

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Abstract. *1,2-diazines derivatives are invaluable materials in the fields of medicine (such as anti-HIV, antiviral and anticancer, antibacterial and antifungus medicines), opto-electronics (compounds with liquid crystal properties and highly fluorescent derivatives: sensors and biosensors, electroluminescent materials, lasers) and agriculture (herbicidal activity and the grow up factor for plants). 1,3-Dipolar cycloaddition is one the most important methods of constructing the pyrrolopyridazine, in classical conditions and using microwave irradiation. For pyrrolopyridazine derivatives was studied the absorption and emission spectra, in ethanol, chloroform and cyclohexane solutions at room temperature.*
Key words: *pyrolopiridazine derivatives, fluorescence, 3+2 dipolar cycloadditions.*

Rezumat. *Derivaţii 1,2-diazinici sunt compuşi cu proprietăţi deosebite în medicină (anti-HIV, medicamente antivirale şi împotriva cancerului, proprietăţi antibacteriene şi antifungice), cu proprietăţi opto-electronice (compuşi cu proprietăţi de cristale lichide şi produse derivate foarte fluorescente: senzori şi biosenzori materiale electroluminiscente, lasere) şi în agricultură (compuşi cu activitate erbicidă şi stimulatori în creşterea şi dezvoltarea plantelor). Reacţiile de cicloadiţie 1,3-dipolare sunt cea mai accesibilă metodă în sinteza derivaţilor pirolopiridazinici, în condiţii clasice şi sub acţiunea microundelor. Pentru derivaţii sintetizaţi au fost înregistrate spectrele de absobţie şi emisie, în etanol, cloroform şi ciclohexan la temperatura camerei.*
Cuvinte cheie*: derivaţi pirolopiridazinici, fluorescenţă, cicloadiţii 3+2 dipolare.*

INTRODUCTION

1,2-diazines are reviewed in literature for their applications: compounds with different biological activities (anticancer, antituberculosis, antimicrobial, antihypertensive etc.), opto-electronics properties (fluorescent derivatives used as sensors and biosensors, electroluminescent materials, lasers and other semiconductor devices) and compounds with liquid crystal properties (Mangalagiu, 2011). Herbicidal activity and grow up factor for plants are also reviewed (Mitsumori et al., 2005; Valeur, 2002).

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In a preliminary communication (Zbancioc et al., 2006; Butnariu et al., 2009; Tucaliuc et al., 2013) is presented the synthesis and spectral analysis of pyrrolopyridazine derivatives. The reaction pathway involves, in the most frequent cases, a Huisgen [3+2] dipolar cycloaddition of ylides to dipolarophiles (activated alkenes and alkynes).

However, this strategy has some disadvantages: lack of control over stereo- and regioselectivity, long reaction times, high energy consumption, and sometimes, low yields.

During the last few decades microwave irradiation (MW) has become an increasingly valuable tool in organic chemistry, since it offers a versatile and facile pathway in a variety of syntheses.

Furthermore, interphase transfer catalysis reactions under MW conditions have the great advantage of using small amounts of, or even no organic solvents ('solvent free'), such reactions are more environmentally friendly and generate less side products (Van der Eycken et al., 2006; Loupy, 2002).

The aim of this work was to study the relationship between optical properties and structure (the effect of substituents and conjugation).

MATERIAL AND METHOD

The strategies adopted for construction of fluorescent derivatives, are depicted in figure 1 and 2. The preparation of all derivatives (**9a, 9b', 9b'', 9c, 10a, 10b, 10c)** involves two steps: initially N-alkylation of the pyridazine (**1**), fig. 1, followed by a 3 +2 dipolar cycloaddition of diazinium ylides (**8a-8b**) (generated *in situ* from the corresponding salts) to the corresponding dipolarophiles (activated alkenes and alkynes nonsymmetrical substitued: ethyl 4,4,4-trifluorocrotonate and ethyl 4,4,4-trifluorobutinoate), fig. 2.

When the dipolarophile was ethyl 4,4,4-trifluorocrotonate (*trans*-isomer, nonsymmetrically diactivated olefine) the reactions involved additional stereo and regiochemical problems, in one therm chorochemistry (Epiotis, 1978). While for ylides **8a** and **8c** the reaction occur chorospecifically, for ylide **8b** (R = Cl) they occur choroselectively**,** after flash chromatography and crystallization from an appropriate solvent, we recovered an inseparable mixture of two regisomers (**9b'**and **9b''**, 1:1)**.**

The reaction with ethyl 4,4,4-trifluorobutinoate leads to the aromatised pyrrolopyridazine **10a-c**. Aromatisation of the initially hyrogenated diazine **iii** occurs spontaneously and could be explained by oxidative dehydrogenation.

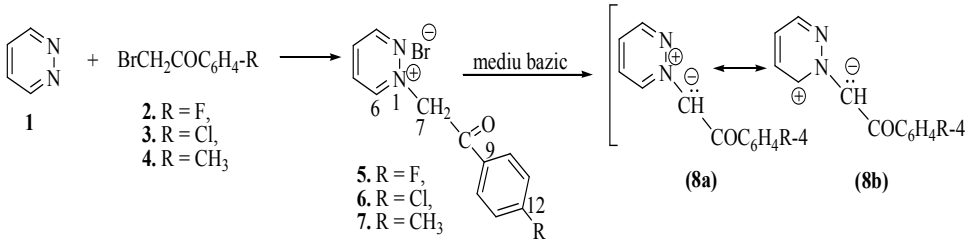

Fig. 1 - N-alkylation of the pyridazine.

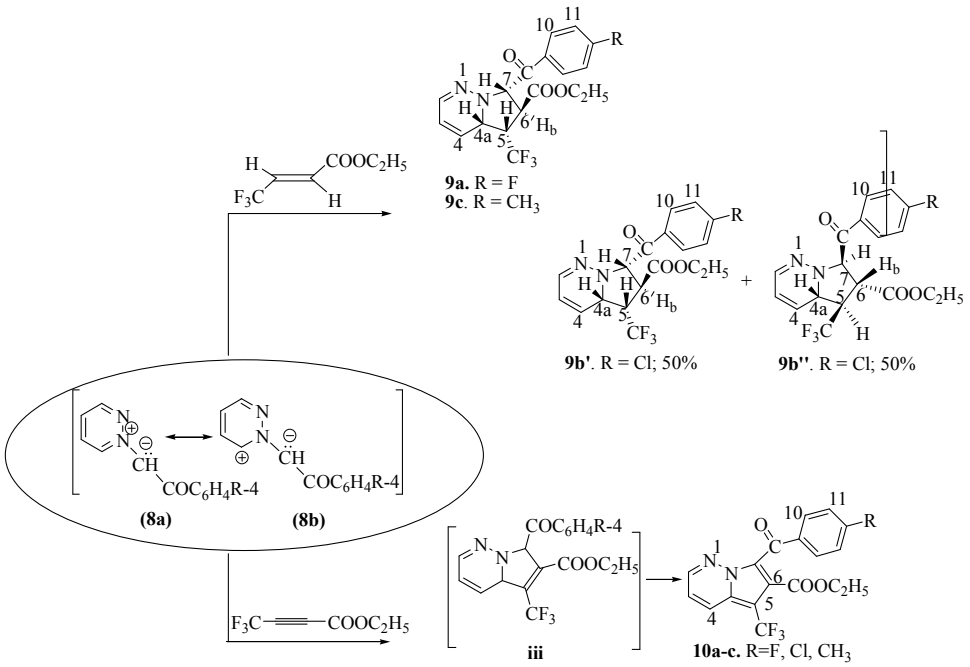

Fig. 2 - 3 +2 dipolar cycloaddition of diazinium ylides.

MW assisted reactions were carried out using a monomod reactor (STAR-2, CHEM corporation, USA). Table 1 lists the optimized conditions, under MW and classical heating. Using MW irradiation, in liquid phase, the best results were obtained applying a constant irradiation power (25% of the full power of the magnetron, 50 W) and varying the temperature ("power control").

Attention was then focused on interphase transfer catalysis reactions. In this study, the solid phase was a mixture of potassium fluoride and N-(p-R-phenacyl)-pyridazinium bromides; the liquid phase consisted of dipolarophiles dissolved in trioctyl-methyl-ammonium chloride–Aliquat 336 (a tensioactive compound that acts as transfer catalyst). The resultant biphasic system is subjected to the action of microwaves using the monomode reactor at 50 W. The best results have been obtained by applying a constant temperature and varying the irradiation power („temperature control").

We presume that the MW heating approach is more effective in [3+2] dipolar cycloaddition reactions due to two factors: the mode of action under MW irradiation and the structure of the ylide intermediate.

It is well known that the magnetic field component of MW radiation is responsible for the dielectric heating effect. The greater the dipole moment of the molecule, the larger the effect of the MW energy will be. The ylides having a 1,2-dipolar structure are excellent dipoles and, therefore, the efficiency of MW heating increases considerably when compared with classical heating.

The results listed in table 1, show the efficiency of the MW irradiation in comparison with the classical heating: the yields were increased in some cases, and the amount of solvent required was reduced.

Table 1

Cycloaddition reactions of pyridazinium ylides with activated alkenes and alkynes under microwave heating and classical conditions

Compd.	Classical		Microwaves			
	Reaction time/min	Yield %	Liquid phase		Interphasic transfer catalysis (KF-Aliquat)	
			Reaction time/min	Yield %	Reaction time/min	Yield %
9a	180	14	5	10	15	-
9b'+9b''	180	16	5	11	15	-
9c	180	9	5	7	15	-
10a	180	38	5	59	15	56
10b	180	41	5	59	15	52
10c	180	46	5	68	15	58

All reagents and solvents employed were of the best grade available and were used without further purification.

The structure of the compounds was proved by spectral analysis: the 1H NMR and 13C NMR spectra and two-dimensional experiments 2D-COSY, 2D-HETCOR(HMQC), long range 2D-HETCOR (HMBC) were recorded on a Bruker Avance 400 DRX spectrometer at 400/100 MHz. Chemical shifts are given in parts per million (δ-scale), coupling constants (J) in hertz and downfield shift from internal tetramethylsilane (δ 0.00 ppm). The IR spectra were recorded on an FT-IR Shimadzu Prestige 8400s spectrophotometerin KBr. Melting points were determined using an electrothermal apparatus and are uncorrected. Flash chromatography was performed with Aldrich 230e400 mesh silica gel. TLC was carried out on Merck silica gel 60-F-254 plates.

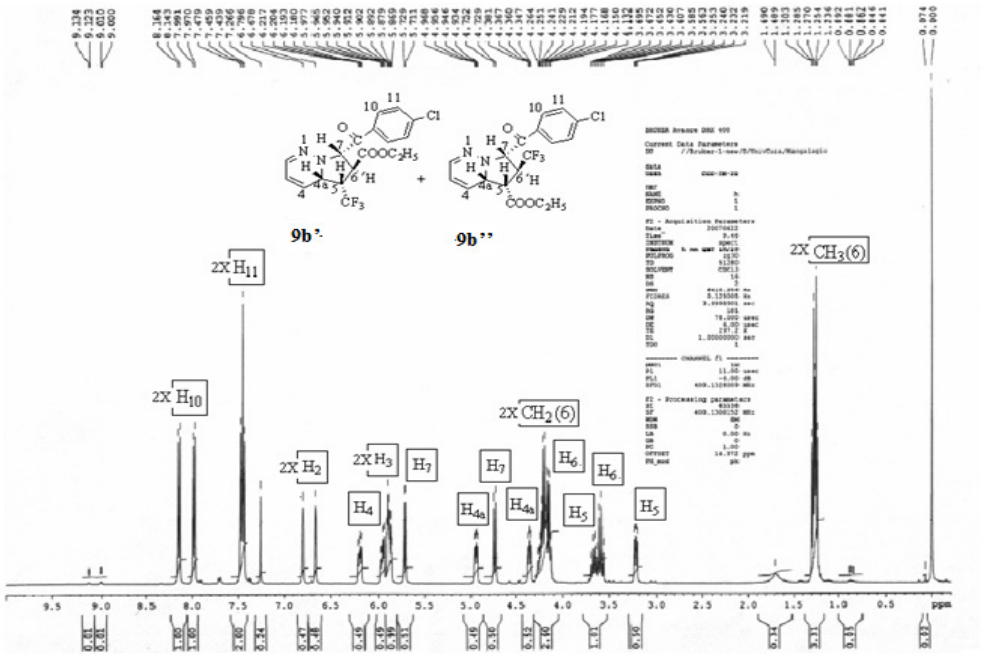

Fig. 3 - 1H-NMR spectrum for compounds **9b'+9b''**.

In the next stage of our work, we studied the absorption and emission spectra of the obtained compounds. The spectra of all the compounds were recorded in ethanol, chloroform and cyclohexane solutions at room temperature.

The fluorescence spectra were recorded with a Turner Bio Systems fluorimeter using FluoOpticalKitID PN: 9300-043 SN: F2000000BB5A4C2D SIG: UV with λ_{ex} = 365 nm and λ_{em} = 410–460 nm.

Relative quantum yields were determined by using anthracene in ethanol (= 0,27 at 25° C) (Parker, 1986). Although, compounds are relatively similar in molecular structure, exhibit clear differences in their experimental absorption and emission spectra, as summarised in table 2.

Table 2

λmax (nm) of absorption spectra and relative quantum yields (%) of piridazine derivatives

Comp.	Fluorescence (λ_{max}, nm) (quantum yield %)			Absorption (λ_{max}, nm)		
	Etanol	Cloroform	Ciclohexan	Etanol	Cloroform	Ciclohexan
9a	420	416	Insolubile	315	320	Insolubile
9b'+9b''	418	414	Insolubile	314	322	Insolubile
9c	430	424	Insolubile	318	320	Insolubile
10a	450	447	Insolubile	330	327	Insolubile
10b	452	449	Insolubile	332	325	Insolubile
10c	456	453	Insolubile	332	331	Insolubile

RESULTS AND DISCUSSIONS

The results listed in table 1 show the efficiency of the MW irradiation in comparison with the classical heating: the yields were increased in some cases, and the amount of solvent required was reduced.

As shown in table 2, the componds are blue emitters (λ_{max} of fluorescence around 420-456 nm, λ_{max} of absorption around 320-331 nm) and have low quantum yield.

The effect of conjugation and the presence of double bonds in azaheterocycles compounds determine fluorescence and quantum yields of the analyzed compounds.

If pyrroloderivatides were fully aromatised, then the quantum yield was extremely high (Zbancioc et al., 2010).

CONCLUSIONS

1. We report a fast, efficient and straightforward method for preparation of fluorescent derivatives containing the piridazine ring, , both in liquid phase and interphasic transfer catalysis.

2. The microwaves induceds a remarkable acceleration of the [3+2] dipolar cycloaddition reaction of pyridazinium ylides to activated alkene and alkyne and allowed a general and facile method for the preparation of pyrrolopyridazine derivatives.

3. Stereo-, regio- and chorochemistry of the cycloadditions were studied.

4. The compounds obtained and tested posses fluorescent proprieties (λ_{max} of fluorescence is around 420-456 nm, λ_{max} of absorption is around 320-331 nm).

5. A certain influence of the substituents concerning absorption and fluorescent properties were observed: the subtituent from the position 5 being important for fluorescence.

RFERENCES

1. **Butnariu R., Mangalagiu I., 2009 -** *New pyridazine derivatives: Synthesis, chemistry and biological activity*. Bioorg. Med. Chem., 174, pp. 2823-2829.
2. **Epiotis N.D., 1978 -** *Theory of Organic Reactions*. Springer: Berlin, p. 34.
3. **Loupy A., 2002 -** *Microwaves in Organic Synthesis*, Wiley, Weinheim, Germany.
4. **Mangalagiu I. I., 2011 –** *Recent Achievements in the Chemistry of 1,2-Diazines,* Curr. Org. Chem., 15, pp. 730-752.
5. **Mitsumori T., Craig I. M., Martini, I. B. Schwartz B. J., Wudl F., 2005 -** Macromolecules, 38, pp. 4698–4704.
6. **Parker C. A., 1986 -** *Photoluminescence of Solutions*, Elsevier, Amsterdam.
7. **Tucaliuc R., Cotea V. V., Niculaua M., Tuchilus C., Mantu D., Mangalagiu I. I., 2013 -** *New pyridazine-fluorine derivatives: Synthesis, chemistry and biological activity. Part II*. European Journal of Medicinal Chemistry, 67, pp. 367-372.
8. **Valeur B., 2002 -** *Molecular Fluorescence*, WileyVCH, Weinheim.
9. **Van der Eycken E., Kappe O., 2006 -** *Microwave-Assisted Synthesis of Heterocycles*, Springer-Verlag, Berlin, Heidelberg, Germany.
10. **Zbancioc G., Mangalagiu I., 2006-** *Microwave-Assisted Synthesis of Highly Fluorescent Pyrrolopyridazine Derivatives,* Synlett, 5, pp. 804-806.
11. **Zbancioc G., Mangalagiu I., 2010 –** *Pyrrolodiazine derivatives as blue organic luminophores: synthesis and properties,* Tetrahedron, 66, pp. 278-282.

TECHNIQUES FOR ESTABLISH OPTIMAL VALUES OF DIMENSIONING VARIABLES USED IN CORRECT PRINTING OF THE TECHNICAL DRAWING

TEHNICI DE STABILIRE A VALORILOR OPTIME ALE VARIABILELOR DE COTARE PENTRU IMPRIMAREA CORECTĂ A PLANȘELOR DE DESEN TEHNIC

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Abstract. *The achievement, with the computer graphics help, by using AutoCAD software, of the technical drawings is apparently a well-known issue by many of users. But the multitude of stages that lead, finally to the printed form of a technical drawing, starting with the loading and establishing the properties of the layers and ending with the printing process, makes this action to be not an easy task. From practical experience, the authors know the fact that between all the stages required to obtain the printed form of a drawing the phases that may pose the most problems are in number of three. The first is the one that refers to the choosing a standard format for printing. The second takes into account the case when the users can not represent or represent incorrectly, on the same format, multiple representations using different scales. Finally, the third concerns the situation in that the users can't indicate the representation scale. Taking to account these inconvenient, the authors have developed a working way that comes to support of users and facilitate the understanding process of the stages listed above.*
Key words: *technical drawing, dimensioning, representation scale, format, CAD*

Rezumat. *Realizarea asistată de calculator, utilizând programul AutoCAD, ale planşelor de desen tehnic este un domeniu aparent pe deplin cunoscut de mulți utilizatori. Dar multitudinea de etape ce conduc, în cele din urmă la forma tipărită a unui desen tehnic, începând cu încărcarea şi stabilirea proprietățile straturilor şi terminând cu procesul de imprimare, face ca această acțiune să nu fie o sarcină ușoară. Din experiența practică, autorii cunosc faptul că dintre toate etapele necesare pentru a obține forma tipărită a unui desen, fazele care pot prezenta cele mai multe probleme sunt în număr a trei. Prima este cea care se referă la alegerea unui format standard pentru tipărire. Cea de a doua ia în considerare cazul în care utilizatorii nu pot reprezenta sau reprezintă în mod incorect, pe acelaşi format, mai multe reprezentări folosind scări diferite. În cele din urmă, a treia se referă la situația în care utilizatorii nu pot indica scara de reprezentare. Luând în considerare aceste inconveniente, autorii au dezvoltat un mod de lucru care vine în sprijinul utilizatorilor şi facilitează procesul de înțelegere a etapelor enumerate mai sus.*
Cuvinte cheie: *desen tehnic, cotare, scară de reprezentare, format, CAD*

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INTRODUCTION

Realisation assisted by computer, by using the computer program AutoCAD, of the technical drawings is an area where "everyone knows" because any graphical representation is essentially a conglomeration of line segments, rectangles, convex polygons, circles, arcs, ellipses and so on, and if the one who achieves the drawing knows both AutoCAD and the rules of the civil or industrial technical drawing, he may easily obtain the desired drawing.

It is known the fact that the achievement, by the classical method, using the technical drawing instruments (ruler, compass, pencil), of the technical drawings assume:

- choosing of a standard format of representation;
- establishment of one or more representation scales;
- reducing all distances to the appropriate scales;
- fitting to page of graphical representations;
- representation of the elements;
- achievement dimensioning.

Realisation assisted by computer, by using the computer program AutoCAD of the technical drawings, is totally different face to classical way and, if one of stages is not known or fully understand, the risk that the drawing be incorrectly is high.

From experience, the authors established that, during the time of technical drawing achievement, when AutoCAD is used, from the multitude of stages, the great majority of users:

- does not set correctly, depending on the used unit measure for the drawing, the unit measure for the standardised formats;
- does not fit correctly the graphical representation into printing format;
- cannot represent or represents incorrectly, on the same drawing, more graphical representations realised to different scales;
- cannot specify the adopted scale or scales.

Taking to account these inconvenient, the authors developed a strategy of work that comes to support the AutoCAD users and facilitate the understanding of stages listed above.

MATERIAL AND METHOD

The working mode proposed by the authors is designed as a flowchart and for the correct performance of all steps listed above AutoCAD users simply must carefully follow the logical scheme and to execute the steps outlined.

In order to verify the effectiveness of the logical scheme the authors have proposed to test it on several types of representations, both assembly drawings and details drawings, because in this way it is possible to cover a large range of representation scales (Table 1) and standard formats (Table 2). (Slonovschi A., Prună L., 2010, 2013).

Table 1

The representation scales commonly used and areas for their use

Areas of use of the representation scales	Commonly used representation scales
Construction details	1:1, 1:2, 1:5, 1:10, 1:20
Working drawings	1:20, 1:50, 1:100
Site plan	1:200, 1:500
Site layout plan	1:1000, 1:2000

Table 2

The standard layouts and theirs size according to adopted unit of measure, millimetre, centimetre, and meter

The standardized formats	Sizes		
	millimetre (mm)	centimetre (cm)	meter (m)
A_4	210 x 297	21 x 29.7	0.21 x 0.297
A_3	420 x 297	42 x 29.7	0.42 x 0.297
A_2	594 x 420	59.4 x 42	0.594 x 0.42
A_1	841 x 594	84.1 x 59.4	0.841 x 0.594
A_0	1189 x 841	118.9 x 84.1	1.189 x 0.841

Testing was done on a group of 27 students.

Each student received the next two AutoCAD files (*.dwg):

- metal pole S_1; representation and basic details and capital.
- plan formwork - reinforcement plan GS_2.

The general characteristics of the two drawings are: all representations are made on 1:1 scale, there isn't any dimension placed on drawing and isn't established any standard format.

Regarding on the first drawing:

- was realized in millimetres;
- the representation of the pole does not change and does not scales;
- basic and capital details does not change but must be done to a bigger scale face to the existing one.

Regarding to the second drawing:

- was realized in centimetres;
- the representation of the beam does not change and does not scales;
- the sections through the beam does not change but must be represented to a bigger scale than the existing one.

The logical schema shown in figure 1 was made in such way that, in both cases, the students to resolve the next requirements:

- to establish, according to the units of measure used to the realization of the drawing, the units of measure for the standardized formats;
- to optimally fit, to a standardized format, the assembly, the metal pole S_1 and the plan formwork - reinforcement plan GS_2;
- to specify the adopted format;
- to specify the standardized representation scale that was adopted for the assembly;

to establish for details (basic details and capital and the sections through the beam) an optimum standardized scale;

- scaling the joining details with a value of scale chosen in such way that to the print, the details to be represented to the scale established to the previous step;
- to verify, in both situations, if the assembly and the details drawing are optimal framed to the chosen standardized format;
- in case that, the represented elements exceeded the chosen format, the students must adopt a new format or they must modify the representation scale of the details until all are fit within chosen format.

Because the logical schema is complex, the authors took the decision to note the boxes with numbers and letters and to explain separately the content of these.

Figure 1 represents the first part of logical schema through that the users are directed to individual branches in that all reports are done according to the adopted units of measure, millimeter, centimeter or meter.

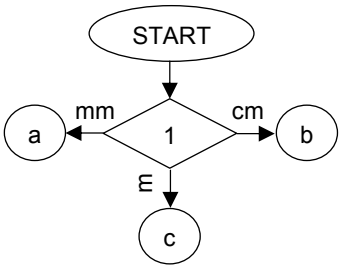

Fig. 1 - The first part of the logical schema

The components of this part of logical schema are:

- 1 – units of measure in that the drawing was made;
  - ✓ a – the branch in that all settings and values are according to millimetre;
  - ✓ b – the branch in that all settings and values are according to centimetre;
  - ✓ c – the branch in that all settings and values are according to meter.

If the branch "a" is continued the logical schema looks like in figure 2.

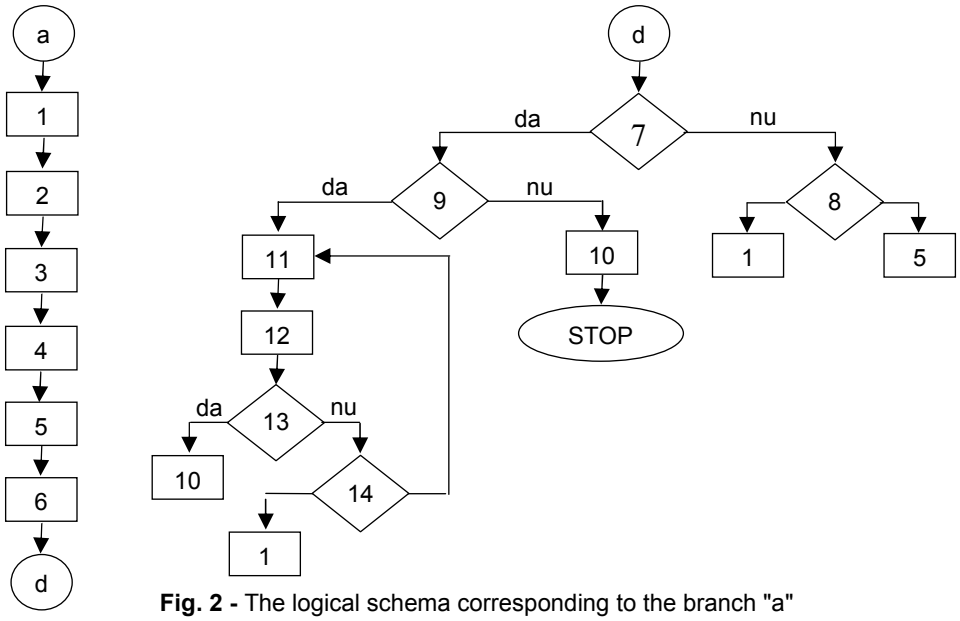

Fig. 2 - The logical schema corresponding to the branch "a"

The components from branch "a" of the logical schema are:

- 1 – one of formats A_4, A_3, A_2, A_1, A_0 are adopted;
- 2 – whatever format the title block has the sizes 185 x 40 (mm);
- 3 – the format and the title block are represented, using the command **RECTANGLE** with the sizes chosen previously;
- 4 – The representation scales can be reported to the values 1, 2, 5 or multiples with 10, above unit or below par of these numbers, thus:
  - ✓1:1;
  - ✓2:1, 5:1, 10:1;
  - ✓1:2, 1:5, 1:10;
  - ✓1:20. 1:50, 1:100, etc.
- 5 – will adopt a scale;
- 6 – the scale of the format and title block is modified using SCALE command, with a value inversely proportional of representation scale value, presented in step 3, thus:
  - ✓ if the adopted scale is 1:1 the scaling factor has value 1;
  - ✓ if the adopted scale is 1:n the scaling factor has value n;
  - ✓ if the adopted scale is n:1 the scaling factor has value 1/n.
- 7 – shall be determined whether the format adopted in step 1 and scaled in step 6 best fits in the representation received;
- 8 – it changes the adopted format in step 1 or the chosen scale in step 5;
- 9 – the drawing contains representations that could be realized at different scales?;
- 10 – it continues with the dimensions settings realization and with the dimensioning of the drawing;
- 11 – shall be determined a scale for realization of the details , according with table 3:

Table 3

The schema of determining of the representation scales

The representation scale of the assembly, adopted in step 5	The possible representation scale of the detail	Ratio of scales	The possible representation scale of the detail
1:100	1:50	100 / 50 = 2	2
	1:20	100 / 20 = 5	5
	1:10	100 / 10 = 10	10
1:50	1:20	50 / 20 = 2.5	2.5
	1:10	50 / 10 = 5	5
	1:5	50 / 5 = 10	10

- 12 – it scales detail/details with a value of the scale factor according to the ratio between the representation scale of the assembly and desired representation scale of detail/details;
- 13 – shall be determined if, both plane of assembly and detail/details scaled are optimally fit in the adopted format at step 1;
- 14 – it changes the representation format adopted at step 1 or the chosen representation scale of the detail at stage 11.

The logical schema corresponding to the branch "b" (drawing realized in centimetres) is different from the logical schema corresponding to the branch "a" only by the first two steps, because both the format sizes and the title block sizes must be

given in centimetres.

The logical schema corresponding to the branch "c" (drawing realized in meters) differs from the logical schema corresponding to the branch "a" only by the first two steps, because both the format sizes and title block sizes must be given in meter.

RESULTS AND DISCUSSIONS

As a result of the application by the students in the two groups of the logical schema, were consisted the following:

- a rate of 85.19% can follow the logical schema and:
  - ✓ can establish correctly the standardized printing format and the unit of measure used for specifying the sizes of the format;
  - ✓ can represent correctly, on the same drawing, multiple representations realized to different scales;
  - ✓ can indicate the representation scale or scales adopted in order to achieve the representations of the assembly and detail.
- a rate of 14.81% can follow the logical schema and:
  - ✓ can correctly establish the printing standardized format of the drawing and the unit of measure used for specify the sizes of the format;
  - ✓ can indicate the adopted scale in order to achieve the assembly representations;

The students from the second group (14,81%) cannot understand why is necessary to represent, on the same drawing, multiple realisations at different scales. From the discussions with them, the authors, have deduced that the students don't know the technical drawing rules regarding to the representation, on the same drawing, of multiple drawings realized at different scales and the advantages given by this type of representations.

Following a brief presentation of the rules and advantages and this small percentage of students was able to correctly represent, on the same drawing, multiple representations at different scales.

CONCLUSIONS

1. The logical scheme proposed by the authors is an indispensable tool for those who want to realize, correctly and rapidly, a series of stages that precede the printing process of any drawing;

2. The breakdown on the types of units of measure (millimetre, centimetre, and meter) presents the advantage that the logical schema can include a diverse array of types of representation and may be successfully used by any student or specialist that wishes to correctly print a technical drawing.

REFERENCES

1. Slonovschi A., Prună, Antonescu I., 2010 – *Desen tehnic pentru construcții.* Ed. PIM, Iași, pp. 22, 27-32, 35-36.

2. Slonovschi., A., Prună L., 2013 - *The International Conference on Engineering Graphic and Design.* Timișoara 13 - 15 June 2013, pp. 27-30.

USING MOODLE TO COLLECT AND ANALIZE THE STUDENT FEEDBACK FORMS FOR TEACHER EVALUATION

UTILIZAREA MOODLE PENTRU COLECTAREA ŞI ANALIZAREA FIŞELOR DE EVALUARE A CADRELOR DIDACTICE

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Abstract. *At the end of each academic semester, the students from USAMV Iasi are asked to fill in a standard assessment sheet to state their opinion on the quality of teaching performed by every teacher they had classes with. The paper proposes the use of the Management Information System capabilities of the Moodle e-learning environment to colect and analize these feedback sheets. This would lead to saving materials, time, and subsequent processing effort.*
Keywords: *Moodle, e-learning, feedback*

Rezumat. *La fiecare sfârşit de semestru, studenții USAMV Iași sunt rugați să completeze câte o fișă de evaluare pentru fiecare cadru didactic cu care au avut ore în semestrul respectiv. În prezent, acest lucru se face folosind formulare de hârtie. Lucrarea propune utilizarea capabilităților de management al informațiilor ale mediului de e learning Moodle pentru colectarea și centralizarea in format electronic a acestor fișe de evaluare. Acest lucru ar duce la economie de materiale, de timp și de efort ulterior pentru procesarea și analiza informațiilor primite.*
Cuvinte cheie: *Moodle, e-learning, feedback*

INTRODUCTION

At the end of each academic semester, the students from USAMV Iasi are asked to fill in a standard assessment sheet to state their opinion on the quality of teaching performed by every teacher they had classes with. Presently, this is done using paper forms that are subsequently processed.

Using the Management Information System capabilities that Moodle has can be used to collect and process such forms in electronic form.

PROPOSED APPROACH

In the beginning of the 2012-2013 academic year, a Moodle e-learning platform was installed and put into use at The University of Agricultural Sciences and Veterinary Medicine of Iasi. Its end users are the university students who benefit from the courses developed by their teachers. More than 4200 users are now registered: the students in the bachelor cycle, and the respective teaching staff.

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Moodle has good Management Information System capabilities (Hollowell, 2011) that allows using it to colect and analize these feedback sheets in electronic form. This would lead to saving materials, time, and subsequent processing effort.

The general idea is that in Moodle 2.1 the teacher can build, among other activities that he includes within a course, an activity that asks students to state their oppinions on different matters, for example on the level of the course material, or on the quality of the teaching activity. Such an activity is (of course) called *feedback*. It consists of a series of questions that are included within a form. There are several (Rice, 2011) types of questions that can be included: multiple choice (rated or not rated), numeric answers, short text answers etc.

A feedback can be signed or anonymous as the teacher decides when building the activity.

This is how feedbacks work. Students are asked to execute the feedback activity in a certain period of time. Each student fills in the electronic form and return it to the teacher. When the completion time is over, the teacher will look over the received answers. He also has the possibility to view statistiscs that Moodle automatically performs on the set of answers. It is also possible for the teacher to export these statistics in Excel format for further processing.

Following is **the proposed approach** for solving the teaching evaluation task. We will use personas to describe the process. These personas are:

- *Admin* – the Moodle platform administrator
- *Head of Department* – the head of the teaching department in the university where the evaluation takes place
- *Teacher* – a member of the the respective department
- *Students* – the students that had classes with Teacher

The staring point is the building of a template course, say **Evaluation 0**. Admin creates and saves it for further utilisation in several copies. This course contains a single activity: a feedback. This is built accordig to the calssical paper form used to evaluate teaching activity within the university. Figure 1 shows a section of the feedback form (in Romanian). All the question showed in this section are of the type *multiple choice – single answer*. There are also (Rice, 2011; https://docs.moodle.org/21/en/Main_page) other types of questions included in the form: numeric answers for sating levels of satisfaction, and text answers for making personal suggestions.

Using the **Evaluation 0** course, *Admin* will create a course regarding *Teacher*, say **Evaluation *Teacher***. The *Students* would be later enrolled in this course. But also *Admin* sets this course to be managed by *Head of Department*, not by *Teacher*. In other words, through the **Evaluation *Teacher*** course *Head of Department* will ask *Students* for their opinion about *Teacher*.

Admin will create (Büchner, 2011) a separate course for each *Teacher* in the department, and all of these courses will be managed by *Head of Department*.

The students enrolment to the evaluation courses should be made by , but is likely that he will ask Admin for some help in this matter. To accomplish the

enrolments, *Admin* will need to receive from *Head of Department* the groups of *Students* corresponding to each *Teacher*. had classes with must possess this information.

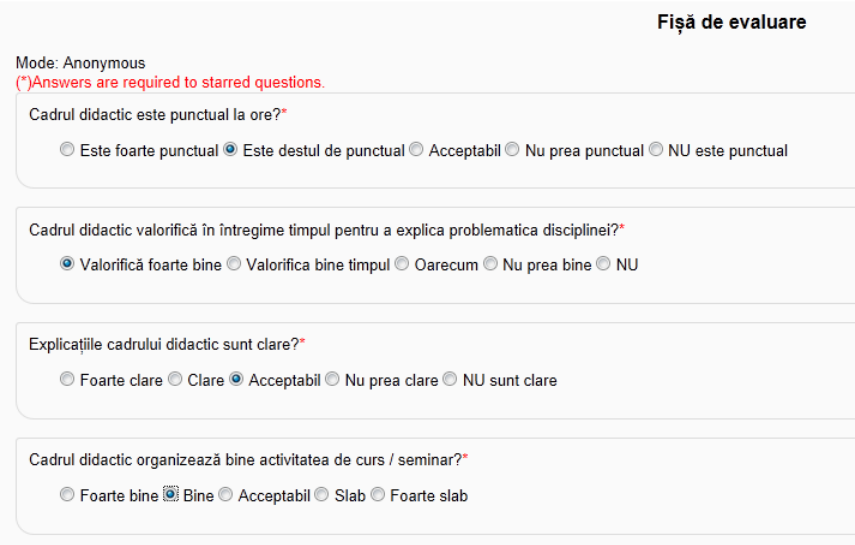

Fig. 1 - A section of the feedback form (in Romanian)

Putting into operation the evaluation process means to ask students to fill in the evaluation forms. Generally speaking, a student that logs in to the e-learning site will only see the list of courses that he is enroled in. Among these, he will see one **Evaluation *Teacher*** course for each *Teacher* he had classes with. These courses will become visible within a certain period of time which is set by *Head of Department* who manages them. In this period of time all the students should access and perform the feedback activities.

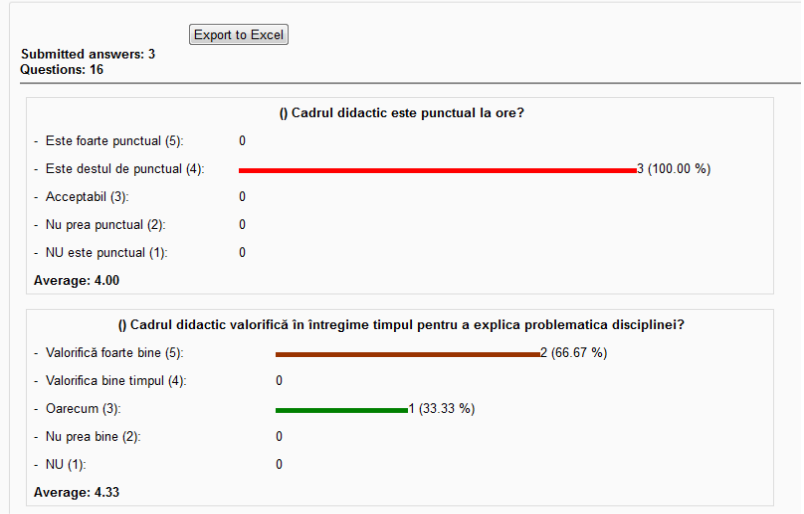

Fig. 2 - Statistics performed on the collected answers

After the closing of the evaluation interval, *Head of Department* will process the feedback for each member of the department. To this effect, he will enter (as course manager) every **Evaluation *Teacher*** course and perform the available statistics. He can also export in Excel those statistics. Figure 2 shows an example of such statistics performed with fictive test data.

CONCLUSIONS

A procedure was proposed for collecting the assessment forms that state the students opinion on the quality of teaching. This procedure involves using the Management Information System capabilities of the Moodle e-leasrning environment.

The first real world application of the described procedure is planned for the end of the first semester of the university year 2014-2015 in the Department of Sciences of The University of Agricultural Sciences of Iasi.

REFERENCES

1. **Büchner A., 2011** - *Moodle 2 Administration*. Packt Publishing Ltd. ISBN 978-1-84951-604-4
2. **Hollowell J., 2011** - *Moodle as a Curriculum and Information Management System.* Packt Publishing Ltd. ISBN 978-1-849513-22-7
3. **Rice W., 2011** - *Moodle 2.0 E-Learning Course Development*. Packt Publishing Ltd. ISBN 978-1-849515-26-9
4. ******* - *MoodleDocumentation* - https://docs.moodle.org/21/en/Main_page

GENETIC DIVERSITY BY LOCAL VARIETY OF PEANUT BASED ON ISOFLAVONES, TOTAL FAT, AND UNSATURATED FATTY ACID CONTENT CHARACTERS

DIVERSITATEA GENETICĂ A UNOR VARIETĂȚI LOCALE DE ARAHIDE, PE BAZA CONȚINUTULUI CARACTERISTIC DE IZOFLAVONE, GRĂSIMI ŞI ACIZI GRAŞI NESATURAȚI

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Abstract. *Genetic diversity of peanut germ plasm is important for plant breeder in making decision regarding selection and method of plant breeding. The data of isoflavones, fats, and unsaturated fatty acid characters were used to estimated genetic diversity. This study was expected to provide information of peanut breeding in the future. The result of this study showed that the genetic diversity of 22 peanut accesions was narrow.*
Keywords*: genetic diversity, accession, peanut, unsaturated fatty acid, isoflavone*

Rezumat*. Diversitatea genetică a germoplasmei arahidelor este importantă pentru amelioratori în ceea ce priveşte metodele de selecţie şi cultivare. Conţinutul de izoflavone, grăsimi şi acizi graşi nesaturaţi a fost folosit pentru a estima diversitatea genetică. Studiul a avut ca scop obţinerea de informaţii referitoare la cultivarea şi ameliorarea arahidelor. Rezultatele studiului arată ca distribuţia diversităţii genetice a celor 22 de cultivare este îngustă.*
Cuvinte cheie*: diversitate genetică, cultivar, arahide, acizi nesaturati graşi, izoflavone.*

INTRODUCTION

Peanut crop is the second most important food crops after soybean. The composition of peanut seeds of economically important are 12-33% carbohydrate, 20-30% protein, and lipid / fat 40-50% (Salisbury and Ross, 1991) as well as mineral deposits such as Calcium, Chloride, Ferro, Magnesium, Phosphorus, Potassium and Sulfur (Sudjadi, 2001). Potential content contained in this placing peanuts as a highly nutritious food crops.

High oil content in peanuts is economically desirable characteristics. Peanut plants known to contain a variety of fatty acid compounds. The presence of fatty acids serve as a source of high energy, so that the peanuts are classified as food crops. The main components include the bean seed protein and fat (Baker, 2002). Fatty acids containing high energy (produce more ATP).

In America and Europe, is known for its peanut oil content containing unsaturated fatty acids (omega 3 and omega 6). In Indonesia, a study of omega 3,

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omega 6 and omega 9 in peanuts has not been done. Research is still oriented cultivation techniques and obtaining high yields (Rashad and Anhar, 2007; Wanget et al., 2002). Because of the need for research on the development of food crops, especially groundnuts very necessary.

In the peanut oil contained compounds which are natural antioxidants tocopherol and effective in inhibiting the oxidation process peanut oil. The tocopherol compounds contained in the unsaturated fatty acids of peanut oil (Ketaren, 1986). The presence of double bonds in the structure of unsaturated fatty acid compounds influence the importance of the resulting compounds. The closer the location of the double bond in the carbon chain structure, such as omega 3 and omega 6, the easier it reacts and the greater nutritional value than other unsaturated fatty acids. Proportionally, saturated fatty acids abundant in animal protein, whereas unsaturated fatty acids found in many plant proteins, including the peanut oil.

Flavonoids and isoflavonoida is one class of secondary metabolites found in many plants, particularly from groups *Leguminaceae*. The content of flavonoids compounds in the plant itself is very low, around 0.25%. These compounds are generally in a state of bound / conjugation with sugar compounds (Snyder and Kwon, 1987). Isoflavone compounds are widely distributed in the plant parts, both in the roots, stems, leaves, and fruit, so that these compounds also unwittingly consumed in the daily diet. In fact, because of its ubiquitous distribution in the plant it is said that when a virtually normal diet without containing flavonoids. It shows that flavonoids are not harmful to the body and even otherwise may provide health benefits.

Studies have shown that isoflavones have antioxidant properties equivalent to the well-known antioxidant vitamin E. The antioxidant power of isoflavones can reduce the long-term risk of cancer by preventing DNA damage. Genistein is an isoflavone among the most powerful antioxidants in soybean, followed by daidzein.

MATERIAL AND METHOD

The research method used was a randomized block design experiment with the treatment of 22 accessions of peanuts in 2 replications, the first planting season and planting season III. The analysis was performed on total fat content, content of Omega-3 Fatty Acids, Fatty Acid Content of Omega-6, Omega Fatty Acid Content-9. Isoflavone content analysis is only done during the growing season I.

The analysis carried out in the Laboratory of Food Crop Post-Harvest in Cimanggu Indonesian Ministry of Agriculture, Bogor in April 2012 and in July 2013.

The material analyzed was a sample of 22 peanut accessions (70 accessions selected from peanuts available based representation of regions in Indonesia peanut spread) with 2 replications, so the number of samples analyzed was 44 samples at each crop growing season (planting season the first and third growing season).

Analysis of laboratory testing performed in the laboratory of the Center for Post-Harvest Food Crops Ministry of Agriculture of Indonesia in Bogor. Tools and materials used laboratory analysis followed the standard operating procedure the international level.

Laboratory and Data Analysis

Laboratory analyzes the content of total fat, polyunsaturated fatty acids (Omega 3, 6, and 9), and isoflavones using samples of peanut seed as much as 100 grams of each accession were observed. Analysis using GC method and Sochlet. Chemicals and reagents used were sulfuric acid 1.25%, 3.25% sodium hydroxide, hexane, sulfuric acid, boric acid 4%, indicator conway, 0.1N hydrochloric acid, sodium hydroxide in methanol, boron triflorida 20 %, saturated sodium chloride and selenium mixture.

The tools used in this analysis is soklet, furnaces, ovens, tube destruction, a set of distillation equipment, electric bath, rotary evaporator, desiccator, filter paper, and other glassware and gas chromatography can separate the components by means of a carrier gas and is recorded as a function of time by the detector (McNair and Bonelli, 1997). Chromatography also provide a short analysis of the sensitivity ppm (Khopkar, 1990) Hitachi brand-263.50 with FID detector. System carrier gas in gas chromatography filter typically contains a molecule of water and other impurities that would be seen in the results recorder (Skoog, 1994).

The observations made by an analysis of the total fat content, the content of unsaturated fatty acids (omega 3, omega 6, omega 9) and isoflavones. Observations were made on samples of peanut seeds that have been dried to a water content of 10-15%.

Data were analyzed statistically by analysis of variance and PCA.

RESULTS AND DISCUSSION

The data results of laboratory analysis of total fat, omega-3, omega-6, omega-9, and isoflavones analyzed variance with randomized block design method. Quadratic mean error value of each character Anova Barlet tested for homogeneity between growing seasons, are in Table 1.

Table 1

Barlet Test Results of 22 Peanut Accession Character Quality

Variance	KTe MT 1	KTe MT 3	Combine	Barlet Test	Criteria
Total Fat	21.30038	35.7779	28.53914	4.555362	inhomogeneous
Omega-3	0.912674	3.977573	2.445124	34.17637	inhomogeneous
Omega-6	19.58171	30.59719	25.08945	3.383465	homogeneous
Omega-9	29.45109	45.58558	37.51834	3.242786	homogeneous

Barlet test results in two cropping seasons on the character quality of the results showed that only two characters are homogeneous. The second character is the homogeneous character of omega-6 and omega-9. Thus the character can be analyzed both combined in the second growing season. The characters are not homogeneous character of omega-3 and total fat content. Both of these characters were analyzed independently of each growing season. In this research, an analysis of the isoflavone content. In the statistical analysis only independent analysis conducted in the first growing season.

Results of laboratory analysis of total fat, unsaturated fatty acids and isoflavones in the two cropping seasons are presented in Table 2.

The results of analysis of diverse quality of the 22 accessions of groundnut is shown in Table 3 Almost all the character quality of the results showed a narrow genetic diversity unless the character content of Omega-3. Character content of Omega-3 has a broad genetic diversity and extensive phenotypic diversity.

Character quality of the results is controlled by many factors, both genetic and environmental interactions, or both. Phenotypic diversity of all the character qualities calculated results is to have a wide variety of categories. This suggests environmental factors strongly influence the quality of the appearance of the character 22 peanut accessions studied.

Table 2

Average content of total fat, unsaturated fatty acids, and isoflavones

Accession	Total Fat	Omega 3	Omega 6	Omega 9	Isoflavons
Atambua	38,3975	2,163	35,03225	35,58225	166,31
Bm 3	36,635	1,5095	28,861	22,0685	288,05
Bm 4	33,73	1,6075	28,283	29,18125	514,01
Gajah	38,04	2,87475	32,6865	33,17375	256,76
Gorontalo A	38,87	2,3735	33,9455	38,21225	420,29
Gorontalo B	38,53	2,206	32,1565	32,734	143,31
Gorontalo C	42,5025	1,923	28,7045	35,812	379,14
Jerapah	41,3325	1,829	33,357	37,18725	247,25
Kanonang Merah	39,03	2,016	27,53075	31,21475	441,53
Kanonang Putih	43,5	2,82425	37,694	38,08025	347,29
Kefa Timor	39,6175	2,0635	33,50725	30,0805	199,89
Kinali Merah	38,465	2,44825	31,13075	32,8115	395,5
Kinali Putih	38,45	1,899	27,06675	36,29775	208,86
Larantuka	35,68	3,320667	40,46933	35,55933	438,62
Madura 1	36,4975	4,064	37,67925	37,15025	152
Siborongborong	39,065	1,3365	30,668	33,9705	152,02
Sima	34,09	2,0845	35,67	36,755	277,81
Soe Timor	35,6325	1,554	30,23525	29,5555	122,98
Sumba Timor	41,2075	4,16175	36,366	32,96675	210,39
Tondegesan Merah	40,265	3,10025	37,243	35,406	272,22
Tondegesan Putih	43,65	1,69	32,4545	35,28175	270,92
Tuban	40,7025	2,35525	31,472	33,17725	342,69

A superior crop varieties should be supported by the potential quality of the results. The quality of peanut yield determined by including the fat content of unsaturated fatty acids and isoflavones content. The results of a study of 22 peanut accessions in two growing seasons, in Table 3, showed a narrow genetic diversity and extensive phenotypic diversity in the character of the total fat content, isoflavones, omega-6, omega-9 and omega-3 (season two). While the character content of Omega-3 in the growing season of the genetic and phenotypic diversity extensive. The results of this study are supported by research over several years of testing at several locations, species strain Valencia peanuts produce oleic acid slightly higher or almost equal to the cultivar New Mexico Valencia C (Burow and Ayers, 2012). Research on the growth of three types of groundnut showed oil content and fatty acids were not significantly different (Raheja et al., 1987), as well as research on four peanut varieties originating from different regions in Pakistan (Akhtar et al., 2005) on the fatty acid composition showed a narrow genetic diversity. Variation of the ratio of high and low grade oleic peanut type in Spanish indicates that factors other than genetics may be involved in determining the ratio of oleic / linoleic (ratio O / L) with appropriate (Lopez et al., 2001). It is claimed that the character quality of peanut plants is strongly influenced by the environment.

Table 3

Diversity of Quality Character of 22 Peanut Accessions

Growing season	Two growing season		One growing season			
			MT 1		MT 2	
Character	σ_g	σ_f	σ_g	σ_f	σ_g	σ_f
Omega-6	-1,79	43,83				
SD	5,27	4,52				
	Narrow	Wide				
Omega-9	-1,91	37,71				
SD	5,24	4,68				
	Narrow	Wide				
Isoflavon			6262,99	18362,06		
SD			4421,22	3630,77		
			Narrow	Wide		
Total Fat			4,62	25,93	-8,47	27.31
SD			6,33	4,50	7,96	2,78
			Narrow	Wide	Narrow	Wide
Omega-3			0,89	1,80	-0,86	3,12
SD			0,44	0,40	0,89	0,33
			Wide	Wide	Narrow	Wide

Analysis of isoflavone content narrow genetic diversity while extensive phenotypic diversity. A similar study by Kirakosyan et al. (2007) on the concentration of isoflavone content in seeds and seedlings on 20 peanut genotypes originating from different geographical sources show a wide diversity. In their

study they also found isoflavone content in groundnut seeds 0.8-fold higher concentration than in the isoflavone content of peanut seeds.

The results of PCA to 5 characters on the quality of the two main components are mapped on a graph as in Figure 1 Biplot Biplot graph shows the distribution of the data character that describes the four main groups of characters that affect the population of 22 peanut accessions were observed. In four distribution groups are groups consisting of two characters, and there are also only consists of a single character. Some characters who are members of the group showed that these characters are-jointly affect changes in a population.

Table 4

Eigenvalue, percentage of variation and percent kumulatif 5 character quality results

PC	Total Eigenvalue	% of Variance	Cumulative %
1	1,897	37,935	37,935
2	1,33	26,6	64,535

Table 4 shows the cumulative percent of the highest value is to PC2. The highest percentage of variance indicated that PC1 character states with high values on PC1 is the most influential character in the diversity of the population of 22 peanut accessions.

Table 5

Component Value Matrix 5 characters at 22 peanut accessions

Characteristics	PC	
	1	2
Omega-6	**0,917**	0,159
Omega-3	**0,888**	
Total Fat	-0,127	**0,923**
Omega-9	0,495	**0,649**
Isoflavon	-0,156	

Table 5 shows the most influential character is the character of Omega-6 and Omega-3 on PC 1 column variation caused by the character of Omega-3 and Omega-6 are jointly affect the diversity of the population of 22 peanut accessions. Biplot diagram (Figure 1) clarify the role of the character of Omega-3 and Omega-6 which together are in quadrant I.

Temperature is the main controller which increase the solubility of oxygen in the water along with the drop in temperature that provides O2 as the recipient of a hydrogen atom essential for the process of unsaturation in the ER so that yield more unsaturated fatty acids. The formation of fatty acids is much faster on the state of the light than in the dark. Plants or parts of plants that received the higher light intensity will result in a higher fatty acid. Ability accession accept a variety of light and temperature cause the expression of diverse fatty acid content.

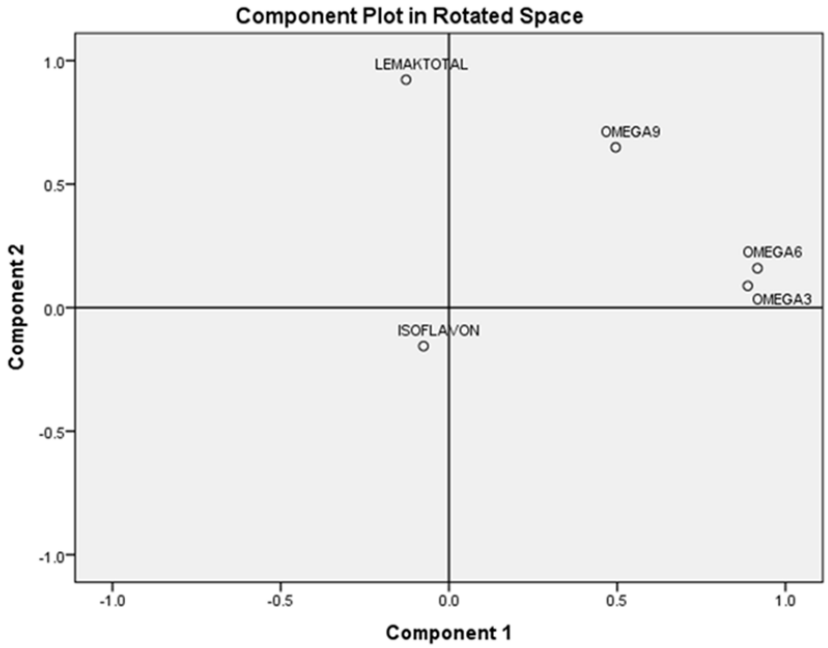

Fig. 1 - Biplot Graphic Pattern Spread the character quality of the 22 accessions of groundnut

The results of PCA for 22 accessions of peanuts on two main components Biplot mapped on a graph as shown in Table 5 and Table 6 and Figure 2 Biplot graph illustrates the distribution of accessions that clustered and distributed almost linear. The results showed that there were 13 accessions most defining quality of the diversity in the population 22 peanut accessions in the study.

Table 6

Eigenvalue, percentage of variation and the percentage of 22 accessions of cumulative groundnut

PC	Total Eigenvalue	% of Variance	Cumulative %
1	11,467	52,124	52,124
2	10,525	47,839	99,963

The data in Table 6 shows that the PC 2 has a higher cumulative percentage values of PC 1, but the higher percetage of variance shows by PC1, 52,124% indicating that accessions having the highest score on PC1 are most influential on the population diversity of 22 accessions of groundnut based character quality results.

Table 7 shows all accessions had values ≥ 0.5 on PC1 and PC2 which states that the accession-accession together create diversity in the population. The

diversity of each accession are displayed on the PC showed extensive phenotypic diversity.

Table 7

Component Value Matrix 22 peanut accessions based on the character quality of the results

Accession	PC		Accession	PC	
	1	2		1	2
Bm 4	0,784	0,621	Tondegesan merah	0,731	0,682
Kanonang merah	0,774	0,633	Tondegesan putih	0,727	0,687
Larantuka	0,77	0,638	Jerapah	0,717	0,697
Kinali merah	0,767	0,642	Kinali putih	0,708	**0,706**
Gorontalo A	0,766	0,643	Soe timor	0,617	**0,786**
Gorontalo C	0,759	0,65	Gorontalo B	0,639	**0,769**
Tuban	0,755	0,656	Siborongborong	0,647	**0,762**
Bm 3	0,753	0,657	Madura	0,648	**0,76**
Kanonang putih	0,747	0,664	Atambua	0,661	**0,75**
Sima	0,738	0,674	Kefa	0,7	**0,714**
Gajah	0,734	0,68	Sumba timor	0,705	**0,709**

The results of the PCA analysis of the population of 22 accessions of groundnut in the form of scores on PC1 and PC2 biplot mapped on a graph as shown in Figure 2.

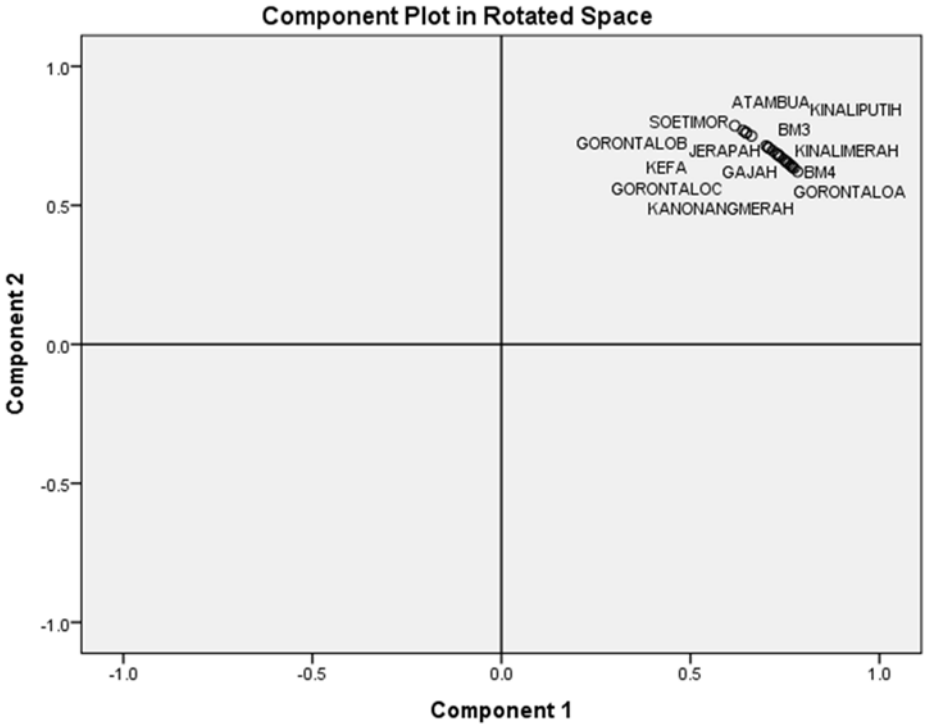

Fig. 2 - Biplot Pattern Chart 22 Spread peanut accessions based on the character quality

Distribution of 22 peanut accessions based on PC1 and PC2 show all accessions form a group on quandran I which showed a narrow genetic diversity of all accessions were observed.

CONCLUSIONS

Phenotypic diversity of all characters analyzed the quality of the results is large, while genetic diversity is narrow, because the metabolism of fats and unsaturated fatty acids and isoflavones are highly influenced by environmental factors so that the selection is based on the diversity of character qualities ineffective results.

Acknowledgements: *Thanks to the Center for Food Crops Republic of Indonesia, in Cimanggu Bogor-Indonesia, which has helped in the process of analysis of the total fat content, the content of unsaturated fatty acids (omega-3. omega-6 and omega-9) and the isoflavone content .*

REFERENCES

1. **Akhtar H., Hamid S., Khan J.I., 2005** - *Comparative Study of Fatty Acid Composition and Characterization of Fixed Oil of Four Peanut Varieties Available in Pakistan*. Natural Product Sciences 11(4), pp. 225-228.
2. **Baker G. L., 2002** - *Flavor Formation and Sensory Perception of Selected Peanut Genotypes (Arachis hypogaea L.) as Affected by Storage Water Activity, Roasting, and Planting Date.* Florida: The University of Florida.
3. **Burow M. A., 2012** - *Development of High-Yielding, High-Oleic acid Valencia Peanuts. American Peanut* Volume 44 Proceedings.14. Raleigh, North Carolina: The American Peanut Research and Education Society, Inc.
4. **Ketaren S., 1986** - *Pengantar Teknologi Minyak dan Lemak Pangan*. Jakarta: UI-Press.
5. **Kirakosyan A., Kaufman P.B., Duke J. A., Seymour E.M., Warber S., Bolling S.F., 2007** - *Production of Isoflavones in Seeds and Seedlings of Different Peanut Genotypes*. Crop Science. 47 (2), pp. 717-719.
6. **Lopez Y, Smith O.D., Senseman S.A., Rooney W.L., 2001** - *Genetic Factors Influencing High Oleic Acid Content inSpanish Market-Type Peanut Cultivars*. Crop Science. 41, pp. 51-56.
7. **McNair H. M., Bonelli E.J., 1988** - *Dasar Kromatografi Gas.* Malang: Bandung: ITB Bandung.
8. **Raheja R.R., Batta S.K., Ahuja K.L., Labana K.S., Singh M., 1987** - *Comparison of Oil Content and Fatty acid composition of Peanut genotype differing in growth habit.* Plant Foods for Human Nutrition 37, 103-108.
9. **Rasyad A., Anhar A., 2007** - *Interaksi Genetik x Lingkungan untuk Berbagai Komponen Hasil dan Stabilitas Hasil pada Beberapa Varietas Padi Lokal Sumatera Barat.* Zuriat, vol.18, no. 2, p. 100.
10. **Salisbury F.B., Ross C.W., 1991** - *Plant Physiology.* Belmont, California: Wadsworth Publishing Company.
11. **Skoog D.A., West D.M., Holler F.J., 1994** - *International Edition Analytical Chemistry an Intrduction.* 6th Edition. Sounders College Publishing, Appendix 4.
12. **Snyder H.E., Kwon T.W. 1987** - *Soybean Utilization.* AVI Book, New York, pp. 104-111.

13. **Sudjadi M., Supriati Y., 2001 -** *Perbaikan Teknologi Produksi Kacang Tanah di Indonesia.* Buletin Agrobio. 4(2), pp. 62-68.
14. **Wanget S.A., Setiamihardja R., Hermiati N., Yuwariah Y., 2002** - *Pola Pewarisan Karakter Warna Kulit Biji dan Jumlah Biji per polong Tanaman Kacang Tanah.* Bandung, Indonesia: Program Pascasarjana Universitas Padjadjaran. Tesis.

THE NEED TO CONTINUE VEGETABLES BREEDING IN ROMANIA IN THE YEARS 2015-2025

NECESITATEA CONTINUARII AMELIORĂRII LEGUMELOR ÎN ROMÂNIA ÎN PERIOADA 2015-2025

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Abstract*: Improved varieties have had a main role in the increases in yield and quality of vegetable crops. In this respect, the vegetable varieties are quickly replaced by new ones. During the last years, an immense quantity of new knowledge on the genetic diversity of vegetables and the utilization of genetic resources, breeding methods and techniques, and utilization of modern biotechnologies in vegetables crop breeding has accumulated.*
Unfortunatelly, în our country, we have lost a great part of genes stock; many breeding laboratories have not enough modern devices and international cooperation is not too strong. But we beleive that the breeding vegetable in our country is very necessary and Ministry of Agriculture must to grant more funds for this activity. We discuss what is possible to do în the next years in order to solve the main problems of this field: researchers training; devices for laboratory and field; breeding objectives în main cultivated vegetable species.
Keywords: *breeding vegetable, new varieties, objectives, breeding team, pathogens resistance.*

Rezumat: *Soiurile ameliorate au avut un rol principal în creşterea producţiei şi calităţii legumelor. Structura soiurilor şi hibrizilor de schimbă foarte repede, cultivarurile vechi fiind înlocuite de cele noi. În ultima perioadă s-au acumulat multe cunoştinţe în domeniul diversităţii genetice a legumelor, a utilizării resurselor genetice, a metodelor şi tehnicilor de ameliorare, inclusiv prin utilizarea biotehnologiilor moderne. Din păcate, în ţara noastră am pierdut o parte din resursele genetice la legume, laboratoarele rămase nu au dotarea necesară şi cooperarea internaţională în acest domeniu a slăbit. Dar credem că ameliorarea legumelor în România este foarte necesară şi are nevoie de fonduri pentru relansare. În articol abordăm ce este necesar de întreprins în anii viitori pentru rezolvarea problemelor din acest domeniu: pregătirea cercetătorilor, procurarea de echipamente pentru câmp şi laborator, principalele obiective care se cer rezolvate.*
Cuvinte cheie: *ameliorarea legumelor, cultivare noi, obiective de ameliorare, echipe de cercetare, rezistenţa la patogeni*

1. THE NEED TO ACHIEVE A NEW BREEDING PROGRAMMES OF VEGETABLE SPECIES

The production and consumption of vegetables have risen sharply in recent decades. Worldwide, in the last 15 years, the production has increased by 15%, a growth rate higher than in other groups of plants. The increasing of the production and

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areas in the last decade is a continuation of the trend occurred after the Second World War. In the period 1961-2000 the areas planted with vegetables and melons have increased by 153% and the total production by 253% (Voican et al., 2006).

The vegetables are an important part of a healthy and varied nutrition, enriching the body with more substances which may prevent diseases, such as vitamins (A, B1, B2, C, PP), antioxidants and minerals (Dumitrescu et al., 1998). As a result, we expect that in the near future, the need of vegetable products will increase. The improvement activity has had and will continue to have a crucial role in increasing production, in diversifying the product range, in the quality of vegetable products. From this point of view, the market of vegetable seed is very dynamic, there is a strong competition between the producing companies. Varieties and vegetable hybrids generally have a short life, being quickly replaced by new cultivars. Therefore, genetics and breeding methods, but - especially novelties, are of great interest to the breeders of the vegetable species, researchers and students.

Especially after 1980, an enormous amount of information on the genetic diversity of vegetable genetic resources has been accumulated, of the breeding methods and techniques, of the development and use of modern biotechnology. The activity of obtaining new cultivars is currently the attribute of large specialized companies, in which the powerful teams of specialists with top equipment activate who keep the secrecy of the improvement techniques for maintaining competitive advantage. Unfortunately, the Romanian researchers' access to all current knowledge of genetics, biotechnology and breeding techniques vegetables has declined drastically in the last 20 years and just in the period when the most important findings are obtained.

Vegetable breeding activity in Romania decreased in intensity due to the unprecedented decrease of the research funds and, hence, the number of researchers; both the laboratory equipment and mechanical means in the field have become obsolete or have been decommissioned and the area has become unattractive for the young graduates of faculties.

However, the improvement vegetables activity should not be discontinued in pending better times. The researchers who still activate get good results, despite the difficulties. They need to join young people to acquire basic technique and try to obtain varieties and new hybrids with superior characteristics, adapted to the local culture conditions and responding to the Romanian consumer preferences.

2. MAIN RESULTS OBTAINED BY ROMANIAN BREEDERS OF VEGETABLE DURING 1995-2014 PERIOD

The main argument in the possibility of reviving the work of improvement, is that the breeders of the vegetable species from Romania have achieved in the last 25 years numerous successes in the difficult conditions of transition to market economy, followed by a prolonged economic crisis. Very valuable varieties for consumption tomatoes of summer-autumn and for the tomatoes for processing have continued to appear. After a long period in which early tomato hybrids for field and greenhouses missed, at SCDL Buzau, the Siriana F1 hybrid was created, which extends in production due to high yielding, to the earliness and outstanding organoleptic qualities (Scurtu and Lăcătusu, 2013).Breeders from SCDL Buzau are proud of not only the Siriana hybrid but also of the cherry tomato creations also a novelty in the domestic assortment. Nationally, we notice new varieties of chili pepper, bell pepper, long pepper and eggplant approved in the last 15 years that make up a variety conveyor that can cover long periods of consumption with fresh fruit from the early to the late varieties. In this group of vegetables the first F1 hybrids appeared, productive, uniform

as vigor of plants, with the fruit shape and size that is very attractive (Andra and Felicia F1 hybrids, with eggplants). At pumpkin vegetables appeared the first F1 hybrid zucchini and "cornichon" cucumbers, and at peas and beans the native assortment now includes varieties with different growing seasons for different destinations (fresh consumption or industrialization) and for different methods of culture. Another remarkable result of the last years is the getting of the first Romanian hybrid carrot that joins several other new varieties obtained from the breeders from ICDLF Vidra and SCDL Buzau.

A review of the most significant results of the improvement of vegetables is shown in Table 1, which shows the percentage of Romanian varieties and hybrids in production. It can be observed that scientific research in Romania has provided growers valuable varieties and hybrids, which in many cases have more weight in production than the imported cultivars. At the same time, it should not be overlooked that with the assortment destined to the crops for greenhouses, a field that will be expanded more and more in the future, the achievements are very modest. With the early cabbage, cucumbers, early tomatoes, lettuce and other crops for greenhouses, the domestic varieties weight is between 5 and 10%.

Table 1

The domestic vegetable varieties weight (%, 2012)

Species	Romanian Cultivars Weight, (%)	Species	Romanian Cultivars Weight, (%)
Tomatoes (for open field)	90	Onion	50
Pepper (tomato-pepper, long pepper, cayenne pepper)	90	Garden Peas	30
Eggplants	95	Garden Beans	15
Early cabbage	0	Other vegetables (for open field)	92
Summer and autumn cabbage	85	A total of field vegetables	60
Cucumbers and squashes (for open field)	20	Early tomatoes (protected 2 cycles)	5
Root vegetable (carrot, parsnip, parsley, celery, beetroot, radishes)	50	Other protected crops (cucumbers, peppers, eggplant, lettuce, zucchini, beans)	5

It is only in the period 1990-2012, with all the difficulties related to the decreasing numbers of researchers, the precarious material basis and the insufficient funding, the research in the field has managed to create and introduce into production a large number of varieties and hybrids, as shown in Table 2.

The improvement activity in the surveyed period got 227 new cultivars to 38 vegetable species, but the number of F1 hybrids is still modest (only 18 hybrids to 209 varieties).

There are numerous examples of varieties and vegetable hybrids created in Romania and approved both by growers and consumers. The best known are (the cultivars marked with an asterisk are recommended for the culture in plastic tunnels): *Siriana F1*, Pontica 102, Viorica, Darsirius,Buzău 47, Kristinica, Carisma*, Coralina*,* for tomatoes*; Cornel 209, Asteroid 204* for round pepper; *Bârsan, Galben Superior, Ceres, Arum, Buzău 10*, Vidra 9** for bell pepper; *Siret* for long pepper; *Andra F1*, Luiza, Contesa, Daniela, Belona**

Drăgaica , Buzău H1* and Eleonora* for eggplants*; Triumf F1 and Bucovina F1* for carrot*; Bistrița* for celery; *De Buzău* for onion*; Buzoiana, Mocira, Poiana* for cabbage; *Auria Bacăului*, Menuet, Ioana** for garden beans*; Vidra 187, Armonia, Işalnița 60, Diana* for garden peas; *Sirius F1*, Ierprem*; Cornişa* F1 for cucumbers.

The main features and characteristics of the new cultivars are most often higher than those made previously.

There are many pathogens attacking some vegetable species and only at few cultivars we can find the authors' mention on resistance or tolerance to attack. It is noted that most claims of tolerance or resistance to diseases are found in peppers and eggplants, while in other species, no such claims, so growers do not know whether or not such cultivars possess resistance. The lack of claims relating to certain pathogens expresses most often, the authors' uncertainty regarding the degree of tolerance or resistance due to the inability of testing on controlled infections.

Table 2

Vegetable Varieties and Hybrids Registered in the Catalogue during 1990-2012

Species	Number	Observations	Species	Number	Observations
Witloof cicory	2	-	Carrot	5	Hybrids – 2
Pepper	37	Chili pepper –13 Mild pepper – 12 Long pepper –11 Cayenne pepper1	Fennelflower	1	-
Okra	2	-	Parsley leaves	1	-
Basil	1	-	Parsley Root	1	-
Cuccumbers	20	Hybrids – 7	Watermelon	2	-
Onion	17	Hybrids – 2 Red Onion – 3 Chives – 1	Melon	3	-
Chicory	2	-	Parsnip	1	-
Savory (thime)	3	Wild thyme – 1	Eggplants	11	Hybrids – 2
Cauliflower	2	Autumn	Sweetcorn	1	-
Zucchini	6	Hybrids 2	Rhubarb	1	-
Tomatoes	28	Hybrids – 3 Cherry Type – 2 For processing –8 Fresh consumption-15	Redishes	6	Early – 1 Summer – 4 Summer-autumn – 1
Cabbage	12	Hybrids – 2 Autumn – 9 Summer – 1	Lattuce	8	-
Dwarf Beans	17		Sage	1	-

Species	Number	Observations	Species	Number	Observations
Climbing Beans	5		Beetroot	2	-
Fennel	1		Spinach	3	-
Kohlrabi	2		Origanum	1	-
Lovage	1		Dill	2	
Orache	3		Celeriac	2	-
Peas	10		Garlic	4	-

3.THE MAIN OBJECTIVES OF IMPROVING VEGETABLES IN THE NEXT PERIOD

The success of a long-term breeding programme is related to the adoption and implementation of a coherent strategy. Random and uncertain funding with values that may decrease from one year to another, cannot lead to competitive results with what is already obtained in the traditional countries and companies. As in any strategy we must start from: a) what it is (where we are); b) to establish our goals; c) how we shall achieve the proposed objectives.

a) One must first start from the existing situation, showing the strengths, weaknesses, opportunities and threats in the system that allow us to determine exactly where we are. We can easily identify, among the *strengths*, the existence of the organized research units and of a number of specialists with knowledge and passion for improvement. Other strengths relate to:

- the experience gained during almost 50 years of functioning of the vegetable research network;
- the accumulation within the institute's portfolio and the research stations of some patents on some valuable varieties (exclusivity);
- the growers' knowledge of some varieties developed in the country and the existence of a constant demand for these due to the credibility of the research stations, gained over time;
- the existence of the stations' sites that contain important information that growers need.

Among *weaknesses* (much more numerous than strengths) one may identify the lack of funding which means the small number of staff and the impossibility of creating multidisciplinary teams, low wages, the lack of equipment and means of production in the field, difficulties in accumulating strictly specialized knowledge, the small number of international partners, the sharp decrease of the germ-plasma sources, etc.

The main *opportunities* are the relatively large areas cultivated with vegetables (around 150,000 hectares), the relatively high cost of vegetable seed from abroad, the consumers' attachment to the Romanian vegetables, characterized by high quality nutrition and taste - and the emergence of some European research funds for 2014-2020;

- the ability to regain the land taken illegally in court, the land required for the isolation in space and the producing of the basic seed.

The program has many *threats* related to the market conquest by large international companies and the difficulty of competing with them without a good marketing programme in place.

- the insufficient concern or lack of concern of the state authorities to support this activity sector;
- the land and the stations' buildings are located in the space within or in close proximity to cities, have a very high value, and therefore of particular interest for the "hunters in the real estate", with permanent risk of losing their ownership;

- the danger of losing indigenous genetic heritage, extremely valuable, collected and improved over 50 years of activity.

b) The second factor to be considered is related to the mission of breeding (where we want to go), i.e. assuming clear objectives: what species will be improved; to what directions of culture and whose purpose will be to the new creations addressed (open field crops, greenhouse or solar, for fresh consumption or industrialization), what place their own creations in the cultivated area will occupy, etc. It is very important for what kind of creations we turn our efforts, respectively to what features and qualities will print to the new creations so that they can be requested by vegetable producers and consumers.

c) The third factor to be noted in the strategy is how we achieve the proposed objectives (providing financial resources, researchers training, sources of germ-plasma, the field and the laboratory equipment, marketing programmes, multiplication, the approval and certification of creations).

The main issue on which we will focus more is about setting goals for improvement, which eventually will make the size and expenditure of each species breeding programme.

Often, there are, in every species, many current and future goals that a breeding programme must solve. The breeder should identify important issues affecting the production and quality of each species in the area that new cultivars will be cultivated. The goals should be prioritized and worked on many projects the budget allows us, the personnel and material basis. Almost all the breeding programmes should aim at three objectives: the resistance to pathogens and insects, the increase of production and the improving of quality (Prohens and Nuez, 2008). Each of these has, in each species, many research directions.

The large number of species does not allow us to detail the myriad of issues concerning the improvement objectives. Significant progress on long-term in breeding programmes cannot be achieved without the development of “in vitro” culture techniques, which allow overcoming barriers of incompatibility to hybridization between cultivated species and wild relatives from which specific resistance genes should be introduced. Other targets for improvement should relate to improving resistance to soil salinity which is or may become a problem of land cultivated with vegetables, and increasing resistance to low or too high temperatures. Throughout the world is already working to introduce resistance to insects’ attack or other pests (mites, nematodes) and it is possible that such creations to get in our country. Therefore, the future improvement teams should be able to address these new issues.

REFERENCES

1. **Dumitrescu M., Scurtu I., Stoian L., Glăman Gh., Costache M., Lăcătuş V., 1998** - *Producerea legumelor* Ed. Artprint,.
2. **Gallais A., Bannerot H., 1992** - *Amelioration des especes vegetales cultivee* -INRA France.
3. **Prohens J., Nuez F**., **2008** - *Handbook of Plant Breeding-Vegetables-* Springer.
4. **Scurtu I., Lăcătuş V., 2013** - *Romanian Vegetable Growing– Present and Prospective for 2020-2025* Management strategies, year VI, special issue.
5. **Voican V., Scurtu I., Costache M., Lăcătuş V., Stoian,L., Roman T., Dumitrescu M. 2006** - *Cultura legumelor în câmp,* Ed. Phoenix, Bucuresti.

DYNAMICS OF THE FLAVONOIDS CONTENT IN SOME TOMATO CULTIVARS FROM NORD - EAST ROMANIA

EFECTUL STRESULUI SALIN ASUPRA DINAMICII CONȚINUTULUI DE FLAVONOIZI A UNOR POPULAȚII LOCALE DE TOMATE DIN NORD-ESTUL ROMÂNIEI

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Abstract. *The aims of the present work were to determine effects of salt stress on the flavonoids content in some tomato cultivars, to find the differeces of salt response in these cultivars. The impact of excess soil salinity on crop productivity is sometimes disastrous which results, in the identification and design of new genotypes of plants tolerant to osmotic stress conditions. Flavonoids, the most common group of polyphenolic compounds that are found ubiquitously in plants. The biological material was represented by local tomatoes populations collected from areas with saline soils from Moldavia region and compared with commercial type salt-tolerant tomato. The bifactorial experience was conducted in a pots experiment in randomized blocks with four repetitions. Ten tomato genotypes studied were subjected to salt stress for a period of 30 days is constantly wetted with saline solution to a concentration of 100 mM and 200 mM. Determination of flavonoids content of leaves was done by the spectrophotometric method. The results show that seven genotypes maintain a high level of flavonoids in the exposure of the two different solutions concentration compared to untreated.*
Key words: *Lycopersicon esculentum, salinity stress, flavonoids.*

Rezumat. *În cadrul acestui studiu a fost analalizat efectul stresului salin asupra conținutului de flavonoizi din frunzele unor genotipuri de tomate. Impactul salinității solului asupra productivității este de cele mai multe ori devastator, ceea ce duce la identificarea unor noi genotipuri de tomate rezistente la salinitate. Flavonoizii reprezintă grupul cel mai comun de compuşi polifenolici care se găsesc în plante. Materialul biologic a fost reprezentat de populații locale de tomate colectate din diferite areale cu soluri saline din regiunea Moldovei şi un soi martor rezistent la salinitate. Experiența de tip bifactorial cu 4 repetiții a fost înființată în vase de vegetație. Pentru o perioadă de 30 de zile, 10 genotipuri de tomate şi un soi rezistent la salinitate au fost supuse stresului salin. Plantele au fost udate cu soluție salină de 100 mM şi 200 mM. Metoda utilizată pentru determinarea conținutului de flavonoizi din frunze s-a realizat prin metoda spectrofotometrică. Rezultatele arată că la 7 genotipuri se menține un nivel ridicat de flavonoizi în cazul expunerii la cele două concentrații diferite de soluții saline față de varianta netratată.*
Cuvinte cheie: *Lycopersicon esculentum, stres salin, flavonoizi.*

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INTRODUCTION

Flavonoids, the most common group of polyphenolic compounds that are found ubiquitously in plants. These are widely distributed in plant fulfilling many functions. Flavonoids and other plant phenolics are especially common in leaves, flowering tissues and woody parts such as stems and bark. They are important in plant for normal growth development and defense against infection and injury. Flavonoids are the most important pigments for flower coloration producing yellow or red/blue pigmentation in petals (Khatiwora et. al., 2010).

Flavonoids represent one of the largest and most studied classes of phenylpropanoid-derived plant specialized metabolites, with an estimated 10.000 different members. Structurally, they consist of two main groups, the 2-phenylchromans (the flavonoids, including flavanones, flavones, flavonols, flavan-3-ols, and anthocyanidins) and the 3-phenylchromans (the iso- flavonoids, including isoflavones, isoflavans, and pterocarpans). Flavonoids act as attractants to pollinators and symbionts, as sunscreens to protect against UV irradiation, as allelochemicals, and as antimicrobial and antiherbivory factors. Their importance in plant biology goes beyond their specific functions within the plant (Dixon and Pasinetti, 2010).

In plants, polyphenol synthesis and accumulation is generally stimulated in response to biotic/abiotic stresses, such as salinity. Indeed, polyphenolic compounds participate in the defence against reactive oxygen species (ROS), which are inevitably produced when aerobic or photosynthetic metabolism is impaired by environmental stresses. Halophytes are naturally salt-tolerant plants that may be potentially useful for economical (oilseed, forage, production of metabolites) applications (Ksouri et. al., 2007).

To prevent the potential cytotoxic effects of ROS, the stimulation of antioxidant systems can assist in plant protection from oxidative stress. Plants have developed antioxidant enzymes such as superoxide dismutase, ascorbate peroxidase, glutathione reductase, catalase, peroxidase and non-enzymatic scavengers like glutathione, ascorbic acid, carotenoids and flavonoids which regularly maintain ROS balances within the cell. Flavonoids are reported as antioxidant agents by scavenging ROS, which are functioned by virtue of the number and arrangement of their hydroxyl groups attaches to ring structures. Their ability to act as antioxidants depends on the reduction potentials of their radicals and accessibility of the radicals (Chutipaijit et. al., 2009).

Some physiological responses to salt stress have been used in determining salt tolerance of plants. Plant hormone levels, antioxidant enzyme activities, pigment contents, osmotic potential reduction, gas exchange characteristics, total soluble protein contents and proline, amounts were determined in different tolerant and sensitive plant varieties at wide range of salt concentrations (Doganlar et. al., 2010). The reported results suggest that an increase of the total phenolic content in rice genotypes as a result of salt stress protects plants from oxidative damage. It was swon that the total flavonoid content increased in salt-stressed

seedlings of salt tolerant rice varieties by 6.34-7.31% and 1.72-3.48% in alt sensitive plants, which indicates that probably flavonoides similarly to proline compounds serve a protective role under stress conditions (Parvaiz, 2013).

MATERIAL AND METHOD

The research was conducted under greenhouse condition and the analysis in the Laboratory of Plant Physiology, Faculty of Agriculture, from USAMV Iaşi.

The biological material was represented by local tomatoes populations collected from areas with saline soils from Moldavia region and compared with commercial type salt-tolerant tomato (*Ursula* $_{F1}$) from Israel.

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

For determination of photosynthetic pigments of leaves we used a UV spectrophotometer type – 1800. The content of chlorophyll pigments was assessed by light absorption ability of the acetone extract of pigments (1%) in the visible spectrum (320-325 nm).

Fig. 1 - The biological material for research (local tomatoes populations)

RESULTS AND DISCUSSIONS

After 15 days the application of saline treatments, the experimental results showed for the plants watered with water only, values of flavonoid

pigment content appreciated on the ability to absorb light in the 320 nm wavelengthn, between 0.66 and 3.23 a.u. In seven of the genotypes studied values between 1.93 and 3.23 a.u. are superior to the variety *Ursula* $_{F1}$, resistant to salinity. The exposure to 100 mM *NaCl* caused an increase of absorbance in 6 genotypes (*Ursula* $_{F1}$, *Copalău* $_{2}$, *Copalău* $_{3}$, *Copalău* $_{4}$, *Copalău* $_{5}$, *Moşna* $_{3}$), subjected to salt stress compared to the control which is between 2.20 and 3.26 a.u. and compared to salt-tolerant type *Ursula* $_{F1}$ (1.70 a.u.) 100 mM a single cultivar (*Moşna* $_{3}$) showed higher values, namely 3.26 a.u. The others 9 genotypes had lower values between 1.70 and 3.13 a.u. The exposure to 200 mM *NaCl* caused an increase of absorbance values at 7 genotypes (*Ursula* $_{F1}$, *Dorohoi* $_{8}$, *Copalău* $_{2}$, *Copalău* $_{3}$, *Copalău* $_{4}$, *Copalău* $_{5}$, *Moşna* $_{3}$) subject to salt stress compared to the control which is between 2.10 and 3.23 u.a, and compared to salt-tolerant type Ursula F1 (2.63 a.u.) 200 mM, 4 genotypes (*Şcheia, Dorohoi* $_{4}$, *Dorohoi* $_{6}$, *Dorohoi* $_{8}$) showed higher values between 2.86 and 3.23 a.u. It is noted these genotype: *Ursula* $_{F1}$, *Copalău* $_{2}$, *Copalău* $_{3}$, *Copalău* $_{4}$, *Copalău* $_{5}$, *Moşna* $_{3}$ maintain a high level of flavonoids in the exposure of two different saline concentrations compared untreated variant (fig. 2).

After 30 days the application of saline treatments, the experimental results showed for the plants watered with water only, values of flavonoid pigment content appreciated on the ability to absorb light in the 320 nm wavelengthn, between 0.00 and 3.30 a.u. In 3 of the genotypes studied values between 2.90 and 3.30 a.u. are superior to the variety *Ursula* $_{F1}$ (2.86 u.a).

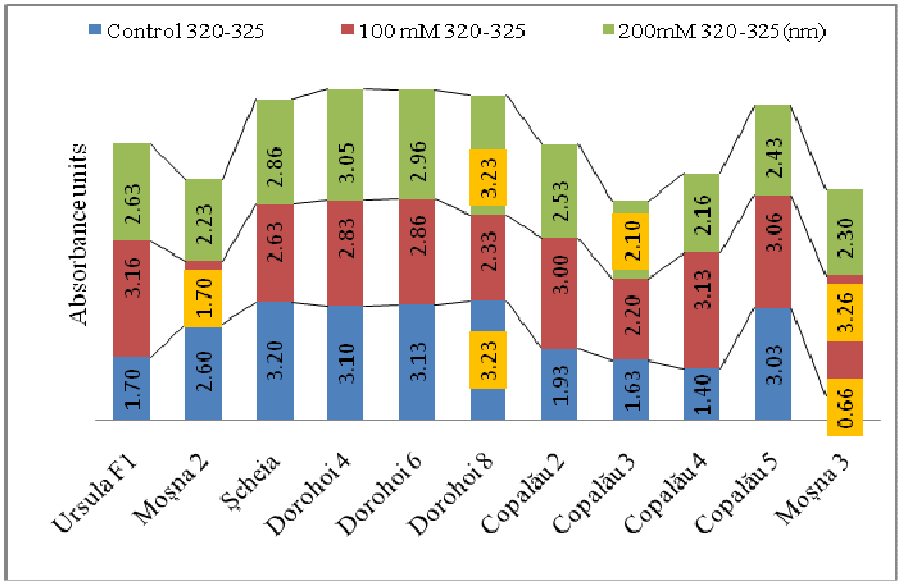

Fig. 2 - The content of flavonoid, after 15 days of saline treatment

The exposure to 100 mM *NaCl* caused an increase of absorbance in all genotypes subjected to salt stress compared to control and to *Ursula* $_{F1}$ values is the same for all genotypes, namely 4.00 a.u.

The exposure to 200 mM *NaCl* caused an increase of absorbance in 9 genotypes compared to control, with values between 2.73 and 3.56 a.u. A single genotype (*Copalău* $_5$) with a value less than the correspondent untreated variant. Compared with *Ursula* $_{F1}$ (3.30 a.u.) 200 mM, 7 genotypes (*Şcheia, Dorohoi* $_4$, *Dorohoi* $_6$, *Dorohoi* $_8$, *Copalău* $_3$, *Copalău* $_4$, *Moşna* $_3$) showed the hight values between 3.33 and 3.56 a.u. (fig. 3).

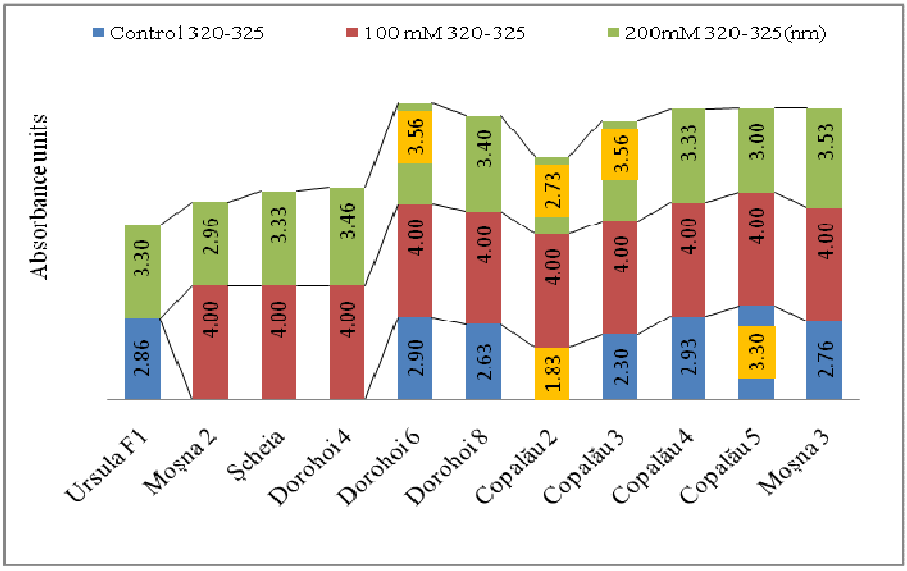

Fig. 3 - The content of flavonoid, after 30 days of saline treatment

CONCLUSIONS

After 15 days of saline treatment it is noted that's genotype: *Ursula* $_{F1}$, *Copalău* $_2$, *Copalău* $_3$, *Copalău* $_4$, *Copalău* $_5$, *Moşna* $_3$ maintain a high level of flavonoids in the exposure of two different saline concentrations compared untreated variant.

After 30 days of saline treatment the exposure to 100 mM *NaCl* caused an increase of absorbance in all genotypes subjected to salt stress compared to control and the exposure to 200 mM *NaCl* caused an increase of absorbance in 9 genotypes compared to control.

Compared with commercial type salt-tolerant tomato (*Ursula* $_{F1}$) after 15 days of saline treatment , 5 genotypes (*Moşna* $_3$, *Şcheia, Dorohoi* $_4$, *Dorohoi* $_6$, *Dorohoi* $_8$) showed a higher level of the flavonoid contents and after 30 days it is noteds that the exposure to100 mM *NaCl* showed a higher level for the all ten

genotypes. The exposure to 200 mM, showed a higher values for 7 genotypes (*Şcheia, Dorohoi $_4$, Dorohoi $_6$, Dorohoi $_8$, Copalău $_3$, Copalău $_4$, Moşna$_{3)}$* .

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

REFERENCES

1. **Chutipaijit. S, Cha-Um S.**, **Sompornpailin K., 2009 -** *Differential accumulations of proline and flavonoids in Indica rice varieties against salinity***.** Pakistan Journal of Botany, 41(5), pp. 2497-2506.
2. **Dixon Richard A., Pasinetti Giulio M., 2007** - *Flavonoids and Isoflavonoids*. Plant Biology to Agriculture and Neuroscience, 154(2), pp. 453-457.
3. **Doganlar Z. B., Demir K., Basak H, Gul I., 2010** - *Effects of salt stress on pigment and total soluble protein contents of three different tomato cultivars*. African Journal of Agricultural Research 5(15), pp. 2056-2065.
4. **Khatiwora E., Vaishali B. Adsul, Manik M. Kulkarni, Kashalkar R.V., Deshpande N.R., 2010** - *Spectroscopic determination of total phenol and flavonoid contents of Ipomoea carnea* International Journal of ChemTech Research, 3 (2), pp 1698-1701.
5. **Ksouri R., Megdiche W., Debez A., Falleh H., Grignon C., Abdelly C., 2007** - *Salinity effects on polyphenol content and antioxidant activities in leaves of the halophyte Cakile maritime.* Plant Physiology and Biochemistry 45, pp. 244-249.
6. **Parvaiz A., Azooz M.M, Prasad M.N.V., 2013** - *Ecophysiology and responses of plants under salt stress.* Springer 15, p. 298-299.

CHLOROPHYLL CONTENT INDEX OF SOME NE-ROMANIA *PHASEOLUS VULGARIS* L LOCAL CULTIVARS, UNDER SALT STRESS

INDICELE CONȚINUTULUI DE CLOROFILĂ A UNOR POPULAȚII LOCALE DE FASOLE DIN NE-ROMÂNIEI, EXPUSE STRESULUI SALIN

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Abstract. *Soil salinity is a significant limiting factor affecting crop productivity in many areas of the world. The development of salt-resistant crops has made very limited progress despite tremendous efforts. From this reason, the capitalization of salt tolerance variability in local landraces should be a first step in plant breeding for the improvement of salt tolerance of cultivated species. Phaseolus vulgaris L is a species particularly sensitive to salt stress A decrease in chlorophyll content under salt stress is reported as a commonly phenomenon in various studies. For this reason, the aim of this work was to determinate the effect of NaCl excess on chlorophyll content index at 19 common bean local landraces, at different growth stages, as an indicator of salt stress tolerance. After NaCl treatments, the most salt sensitive local landraces were Tudora 1, Balş 1, Copalău 2 and 3, Iezer 2, Moşna 6, and the most tolerant Coropceni 1 and 2, Iezer 4 and 5, Moşna 3 and 5, Săveni 1 and 7 and Truşeşti 5.*
Key words: *salt stress, Phaseolus vulgaris, chlorophyll content index*

Rezumat. *Salinitatea solului reprezintă un factor limitativ important pentru productivitatea culturilor agricole, în multe regiuni ale globului. Cu toate eforturile depuse, crearea de plante agricole rezistente la salinitate a înregistrat mici progrese. Din acest motiv, capitalizarea variabilității genetice, referitor la rezistența la salinitate a populațiilor locale, poate constitui un prim pas în lucrările de ameliorare, în vederea îmbunătățirii toleranței la salinitate a speciilor cultivate. Phaseolus vulgaris L este o specie deosebit de sensibilă la stres salin. Reducerea conținutului de clorofilă, sub efectul stresului salin, este descrisă ca un fenomen comun, în numeroase studii. Din acest considerent, scopul lucrării de față a fost determinarea efectului excesului de NaCl asupra dinamicii indicelui conținutului de clorofilă în cazul a 19 populații locale de fasole, ca un indicator al toleranței la stres salin. După tratamentul cu NaCl, cele mai sensibile cultivare s-au dovedit a fi: Tudora 1, Balş1, Copalău2 şi 3, Iezer 2, Moşna 6, iar cele mai tolerante: Coropceni 1 şi 2, Iezer 4 şi 5, Moşna 3 şi 5, Săveni 1 şi 7 şi Truşeşti 5.*
Cuvinte cheie: *stres salin, Phaseolus vulgaris, indicele conținutului de clorofilă*

INTRODUCTION

Soil salinity is a significant factor affecting crop productivity in many areas of the world. The decreases of plant growth and yield, depend on the plant

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species, salinity levels, and ionic composition of the salts (Husain et al., 2010, Slabu et al., 2009).

Phaseolus vulgaris L. is one of the most important food plants, cultivated in several cultivars (Burescu and Toma, 2005), for its seeds rich in protein (15%), fat (2%) and starch (80%). Is a species particularly sensitive to the presence of NaC1 in the root zone (Greenway and Munns, 1980). During the last decades, the development of salt-resistant crops has made very limited progress despite tremendous efforts (Schubert et al., 2009). The investigation of stress exposed plants and subsequent identification of certain stress-specific biomarkers revealed important information about the limits of stress tolerance. (Gostin, 2007). The genetic variation in cultivated beans germplasm for salinity tolerance is limited (Gama et al., 2007). Although, it is important to test several cultivar regarding this quality. From this reason, the capitalization of salt tolerance variability in local landraces should by a first step in plant breeding for the improvement of salt tolerance of cultivated species.

Under salt stress, the decrease in chlorophyll content is a commonly reported phenomenon in various studies (Taffouo et al., 2009, Nazarbeygi et al., 2011, Mehr , 2013). Salinity and salt stress duration significantly affected photosynthesis (Bayuelo-Jimenez, 2012). For pinto bean found Ghassemi-Golezani et al. (2012) that under salt stress, the chlorophyll content index (CCI) of leaves diminished with progressing plant development at reproductive stages and during reproductive stages CCI of all cultivars decreased with increasing plant senescence. For faba bean found Slabu et al. (2009) that the reduction of chlorophyll in leaves after NaCl exposure may be explained as a result of high Cl^- concentrations in the chloroplasts, which is amplified by a simultaneously high Na^+ concentration. On the other hand, it is known that under salt stress, the plants have a reduced growth and an intense green color (Mengel, 1991), respectively a higher chlorophyll concentration in the leaves. These aspects can be explained by the biphasic response of plants to salinity, proposed by Munns (2002): a rapid, osmotic phase that inhibits growth of young leaves, and a slower, ionic phase that accelerates senescence of mature leaves. In the second phase of salt stress, NaCl decreased total chlorophyll concentration in the leaves by destruction of chloroplast structure and the instability of pigment protein complexes (Koyro, 2002). Plant adaptations to salinity are of three distinct types: osmotic stress tolerance, Na^+ or Cl^- exclusion, and the tolerance of tissue to accumulated Na+ or Cl^- (Munns and Tester, 2008). Therefore maintaining high levels of chlorophyll content in leaves can be an indication of greater salinity tolerance of plants.

The aim of this work was to determinate the effect of salt stress on chlorophyll content index at 19 common bean local landraces, at different growth stages, as an indicator for salt stress tolerance.

MATERIAL AND METHOD

A pot experiment with factorial arrangements on the bases of randomized complete block with three replications was conducted in 2014 to investigate the

salinity tolerance at 19 common bean local landraces. The cultivars were collected from NE Romania Iasi and Botosani districts from areas with saline excess. Six seeds were sown 3 cm deep in each pot, filled with 10 kg garden soil. The pots were placed in the greenhouse. The temperature variation in the greenhouse was 17- 30ºC. After the emergence were kept only three plants per pot. After the occurrence of the second true leaves, the plants were exposed to salt stress experimental variants: V1 – control, watered with water only; V2 - 4 watering with 1 liter 100 mM NaCl solution for each watering; V3 - 4 watering with 1 liter 200 mM NaCl solution for each watering. Plants were grown to full maturity.

Chlorophyll content index was measured in each plot by using a portable chlorophyll content meter (CCM-200, Opti-Sciences Inc., NH, USA). Ten measurements were taken for each pot. 4 determinations were made: d1 - after a week of exposure of plants to salt stress; d2 - alt flowering stage; d3 - beginning of pot development; d4 - ripening of fruit and seed.

RESULTS AND DISCUSSIONS

At the first determination (d1) control plants showed variations of CCI values between 11.7 (Truşeşti 5) and 21.0 (Copalău1). The treatment with 100 mM NaCl (V1) for one week led to an increase of the CCI values, 14.5 (Truşeşti) and 24.7 (Coropceni 1). The same behavior has been observed after the treatment with 200 mM NaCl, CCI values ranging from 12.0 (Coropceni 2) and 23.0 in Copalău 1 (table 1).

Table 1

Chlorophyll content index at vegtative growth stages (d1) and by flowering (d2)

	d1			d2		
Cultivars	**V1**	**V2**	**V3**	**V1**	**V2**	**V3**
Balş 1	17,9	19,2	19,5	21,1	24,1	24,0
Copalău 1	21,0	23,3	23,0	27,9	21,2	22,4
Copalău 2	15,2	17,1	17,6	15,3	17,2	16,1
Copalău 3	13,8	17,1	17,6	15,7	17,7	18,7
Codreni 1	16,3	17,3	13,0	14,4	14,1	13,1
Coropceni 1	17,5	24,7	17,2	24,3	19,2	16,7
Coropceni 2	17,7	17,7	12,0	21,0	16,6	17,7
Iezer 2	14,7	14,7	15,1	19,6	22,5	23,0
Iezer 4	14,3	18,7	18,1	20,0	24,3	17,4
Iezer 5	12,2	16,7	16,2	19,8	28,8	25,9
Moşna 1	18,6	23,9	17,8	17,9	25,6	17,0
Moşna 3	12,3	16,3	17,8	14,9	16,2	17,6
Moşna 4	13,4	17,7	14,7	14,7	23,2	20,7
Moşna 5	21,0	21,7	20,4	21,5	22,8	23,8
Moşna 6	12,3	17,0	13,7	16,7	19,4	18,0
Săveni 1	14,8	18,6	17,9	16,0	19,9	25,2

Săveni 7	12,2	18,4	19,5	16,3	16,4	25,5
Truşeşti 5	11,7	14,5	13,8	18,2	16,8	21,2
Tudora 1	16,6	17,8	18,8	19,6	22,6	17,8
MIN	**11,7**	**14,5**	**12,0**	**14,4**	**14,1**	**13,1**
MAX	**21,0**	**24,7**	**23,0**	**27,9**	**28,8**	**25,9**

The studies done by Beinsan C. et al, 2009 showed for the common beans an increase in chlorophyll content at lower level of stress and a decrease in more stressed variant. Dadkhah and Moghtader (2008) found for the sugar beet that under salt stress leaves appeared healthy and leaf chlorophyll content increased with increasing salinity. The increasing in chlorophyll can be explained due to salinity effect of leaf area. Leaves of stressed plants became thicker than unstressed plants. Thicker leaves contain more cells in a certain leaf area (Dadkhah and Moghtader, 2008). Also in the case of common bean salinity concentration caused considerable reduction of leaf area with differences between bean local landraces on different variant of osmotic stress (Beinsan et al., 2009).

The increase of CCI at vegetative growth phenophase could by explain by a leaf reduction under salt stress without a destruction of chloroplast.

At the flowering stage (d1) a growth of CCI was recorded in 11 of the analysed cultivars, in the rest of the cultivars was observed a reduction of CCI, after the treatment with 100 mM NaCl or with 200 mM NaCl (table 1).

At the beginning of pot development (d3), CCI values remain elevated in most cultivars, ranging between 14.3 and 28.4 (table 2) except cultivars Balş 1 and Tudora 1, wherein the plants are completely chlorotic.

Table 2

Chlorophyll content index at beginning of pot development (d3) and by ripening of fruit and seed (d4)

	d3			d4		
cultivars	**V1**	**V2**	**V3**	**V1**	**V2**	**V3**
Balş 1	22,0	3,3	2,5	12,4	0,0	0,0
Copalău 1	26,9	22,8	21,9	17,4	6,7	0,0
Copalău 2	21,6	17,8	16,7	6,3	0,0	0,0
Copalău 3	16,5	20,2	19,8	7,2	0,0	0,0
Codreni 1	14,3	12,2	11,1	6,5	8,9	0,0
Coropceni 1	28,4	25,5	24,5	14,4	10,0	4,6
Coropceni 2	23,8	18,1	21,1	9,3	7,4	6,0
Iezer 2	24,8	26,5	21,5	10,9	0,0	0,0
Iezer 4	16,3	20,1	15,1	12,2	6,1	6,5
Iezer 5	21,5	19,8	20,5	13,3	10,5	9,5
Moşna 1	22,9	18,1	17,8	0,0	0,0	0,0
Moşna 3	14,7	16,6	17,1	9,8	12,1	8,1

Moşna 4	17,0	20,1	14,8	9,9	3,9	0,0
Moşna 5	24,1	24,6	18,3	9,3	4,0	3,9
Moşna 6	17,3	20,3	14,1	9,2	0,0	0,0
Săveni 1	17,2	26,3	19,6	12,3	11,0	8,3
Săveni 7	16,6	22,1	23,4	10,3	5,0	5,9
Truşeşti 5	18,6	18,5	23,7	11,6	4,7	7,8
Tudora 1	20,4	0,0	0,0	17,6	0,0	0,0
MIN	**14,3**	**0,0**	**2,5**	**6,3**	**0,0**	**3,9**
MAX	**28,4**	**26,5**	**24,5**	**17,6**	**12,1**	**9,5**

At ripening of fruit and seed (table 2), normally occurs a decrease of CCI values by control variants, which are between 6.3 (Copalău 2) and 17.6 (Tudora 1). The treatments with 100 mM NaCl caused the drying of cultivars: Balş 1, Copalău 2 and 3, Iezer 2, Moşna 6. The treatments with 200 mM caused plants death from cultivars: Copalău 1, Codreni 1, Moşna 4. The decrease of CCI values after saline treatment may be explained by the toxic effect of Na^+ and Cl^- ions that disorganize the cells, especially chloroplasts, resulting a reduction of chlorophyll content.

After this salt treatments, the most tolerant cultivars, by which the chlorosis occurred less, were Coropceni 1 and 2, Iezer 4 and 5, Moşna 3 and 5, Săveni 1 and 7, and Truşeşti 5.

CONCLUSIONS

1. At the analysed cultivars, the control variants, watered with water only, the chlorophyll content index shows high variability.

2. NaCl treatment causes an increase in CCI, excepting of three local populations at 200 mM NaCl: Codreni 1, Coropceni 1 and Coropceni 2.

3. By the sensitive variants, elevated CCI is maintained until flowering and by the salinity tolerant variants until the pods formation.

4. After NaCl treatments, the salt sensitive local landraces were Tudora 1, Balş 1, Copalău 2 şi 3, Iezer 2, Moşna 6, and the most tolerant Coropceni 1 and 2, Iezer 4 and 5, Moşna 3 and 5, Săveni 1 and 7 and Truşeşti 5.

***Acknowledgments**: 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. Bayuelo-Jimenez J. S., N. Jasso-Plata, and I. Ochoa, 2012 - *Growth and Physiological Responses of Phaseolus Species to Salinity Stress*. International Journal of Agronomy doi:10.1155/2012/527673, pp. 1-14.

2. Beinsan C., Camen D., Sumalan R., Babau M., 2009 - *Study concerning salt stress effect on leaf area dynamics and chlorophyll content in four bean local landraces*

from Banat area. 44th Croatian and 4th International Symposium on Agriculture, Opatija, Croatia, 16-20 febr., pp. 416-419.
3. **Burescu P., Toma I., 2005** – *Manual de lucrări practice de botanică*. Ed. Universităţii din Oradea, pp. 346-347.
4. **Dadkhah A., Moghtader S. H., 2008** - *Growth and gas exchange response of sugar beet (*Beta vulgaris L.*) cultivars grown under salt stress.*,in :*Photosynthesis-energy from the sun,* Eds: J F Allen; et al. Ed. *Springer Netherlands,* p. 1432 -1436.
5. **Gama P. B. S., Inanaga S., Tanaka K., Nakazawa R., 2007** - *Physiological response of common bean (*Phaseolus vulgaris L.*) seedlings to salinity stress*. African Journal of Biotechnology, vol. 6 (2), pp. 79–88.
6. **Greenway H., Munns R., 1980** - *Mechanisms of salt tolerance in nonhalophytes*. Annu. Rev. Plant Physiol. 31, pp. 149-190.
7. **Ghassemi-Golezani K., Neda Nikpour-Rashidabad, Saeid Zehtab-Salmasi, 2012** - *Physiological performance of pinto bean cultivars under salinity*. IJPAES, vol 2(2), pp. 223 -228.
8. **Gostin Irina, 2007** – *Biomarkeri structurali la plante*. Ed. Univ. „Al I Cuza" Iasi, pp.17.
9. **Hussain N., Sarwar G., Schmeisky H., Al-Rawahy S., Mushtaque A., 2010** - *Climate change and management of cool season grain legume crops*. Eds: Yadav SS, McNeil DL, Redden R, Pati. Ed. Springer, Heidelberg, pp.178.
10. **Koyro H. W., 2002** - *Ultrastructural effects of salinity in higher plants*. In: *Salinity: Environment – Plants – Molecules*. Eds: A. Läuchli and U. Lüttge, Ed. Kluwer Academic Publ. pp.139-158.
11. **Mehr Z. S., 2013** - *Salt-induced changes in germination and vegetative stages of* Anethum graveolens L. Journal of Stress Physiology & Biochemistry Vol. 9(2), pp. 190 -198.
12. **Mengel K., 1991** - *Ernährung und Stoffwechsel der Pflanze*. Ed. G Fischer, Jena, pp. 267.
13. **Munns R., 2002** - *Comparative physiology of salt and water stress*. Plant, Cell and Environment 25, pp. 239–250.
14. **Munns R., Tester M., 2008** - *Mechanisms of Salinity Tolerance* . Annual Review of Plant Biology. Vol. 59: pp. 651-681.
15. **Nazarbeygi E., Yazdi H.L., Naseri R., Soleimani R., 2011** - *The effects of different levels of salinity on proline and a-, b- chlorophylls in canola*. Amer-Eurasian J. Agric. Environ. Sci., Vol.10, pp. 70-74.
16. **Slabu C.,. Zörb Ch, Steffens D., Schubert S., 2009** - *Is salt stress of faba bean* (Vicia faba) *caused by* Na^+ *or* Cl^- *toxicity*? J. Plant Nutr. Soil Sci.172, pp. 644–650.
17. **Schubert S., Anja Neubert, Antje Schierholt, Ali Sümer and Christian Zörb, 2009** - *Development of salt-resistant maize hybrids: The combination of physiological strategies using conventional breeding methods*. Plant Science 177, pp.196–202.
18. **Taffouo V. D., J. Kemdem Kouamou, L. M. Tchiengue Ngalangue, B. A. Nandjou Ndjeudji, A. Akoa, 2009** - *Effects of salinity stress on growth, ions partitioning and yield of some cowpea* (Vigna unguiculata L. *Walp*.) *cultivars.* International Journal of Botany, 5, pp.135-143.

CHLOROPHYLL CONTENT INDEX AND LEAF AREA OF SOME TOMATO LOCAL CULTIVARS FROM N-E ROMANIA, UNDER SALT STRESS

INDICELE CONȚINUTULUI DE CLOROFILĂ ȘI SUPRAFAȚA FOLIARĂ A UNOR POPULAȚII LOCALE DE TOMATE DIN NORD-ESTUL ROMÂNIEI, EXPUSE STRESULUI SALIN

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Abstract: *Soil salinity is an important abiotic stress factor seriously affecting plant productivity and survival. Photosynthesis and growt of many plants are inhibited under NaCl salinity. The research was conducted under greenhouse condition. The biological material was represented by four local tomatoes populations collected from areas with saline soils from Moldavia region and compared with commercial type salt-tolerant tomato. Tomato genotypes in the study were subjected to salt stress for a period of 30 days is constantly wetted with saline solution to a concentration of 100 mM and 200 mM. The chlorophyll content was determinated whit chlorophyll meter and the leaf area with portable scanner AreaMeter AM 300 – 0002. Analyzed the results based on biphasic model of growth response under salt stress proposed by Munns (1993), and found that tomatoes reacted similarly. The transition from phase I to phase II, is done by salt stress intensity, but mostly by cultivated genotype.*
Key words: *Soil salinity, tomato, leaf area, chlorophyll content.*

Rezumat: *Dintre factorii de stres din mediu, salinitatea rămâne principalul factor care pune sub semnul întrebării viitorul agriculturii. Procesul de fotosinteză pentru multe specii de plante este inhibat de concentrația NaCl. Experiența a fost înființă în vase de vegetație în condiții de seră. Au fost luate în studiu 4 genotipuri de tomate colectate din solurile saline ale Moldovei și un soi comercial rezistent la salinitate. Acestea au fost expuse stresului salin pe o perioadă de 30 de zile, fiind udate constant cu soluții saline de concentrație 100 mM și 200 mM. Conținutul de clorofilă al frunzelor a fost determinat cu ajutorul clorofilometrului iar, pentru suprafața foliară s-a utilizat aparatul portabil AreaMeter AM 300 – 0002. Analizând rezultatele pe baza modelului bifazic de reacție al plantelor la salinitate propus de către Munns (1993), s-a constatat că și tomatele se înscriu în același model. Trecerea de la faza I la faza a II-a, făcându-se în funcție de intensitatea stresului salin și de genotipul cultivat.*
Cuvinte cheie: *salinitatea solului, tomate, suprafață foliară, conținut de clorofila*

INTRODUCTION

Soil salinity is an important abiotic stress factor seriously affecting plant productivity and survival. Growth and development of glycophytes are negatively

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affected but halophytes tolerate high salt concentrations (Doganlar et. al., 2010). Salinity resistance of crops is determined by the physical properties of the soil, physiological particularities of plant, growth and development phase (Jităreanu, 2007).

The tolerance to Na^+ of plants under salt conditions (*NaCl*), the prevention of the replacement of Mg^+ with Na^+ and the continuous increase of chlorophyll amount is accepted as an important indicator of salt tolerance. It is stated that plants with high chlorophyll content under salinity stress are more tolerant to salt (Yaşar and Esra, 2012). The decrease in chlorophyll content of the leaves of the plants treated with *NaCl*, may be caused by the increase in the concentration of Cl^- in the chloroplast, which may be amplified simultaneously by increasing the concentration of Na^+, as a result of synergistic effect. An increase in the concentration of Mg^+ in the nutrient solution prevents effectively lowering the concentration of chlorophyll (Slabu, 2005).

Photosynthesis is one of the mostly affected factors due to salt stress (Babu et. al., 2011). The decline in photosynthesis due to salinity stress could be due to lower stomata conductance, depression in carbon uptake and metabolism, inhibition of photochemical capacity or a combination of all these factors (Zhani et. al., 2012).

Leaf area represent an important physiologic index in characterization of intensity to some metabolic process (groweing, transpiration, photosynthesis, respiration, etc) (Şumălan and Dobrei, 2002). Ciobanu (Popescu) and Şumălan (2009), shwoed that at the plant exposed to saline environment generally has the leaf area reduced.

MATERIAL AND METHOD

The research was conducted under greenhouse condition from USAMV Iași.

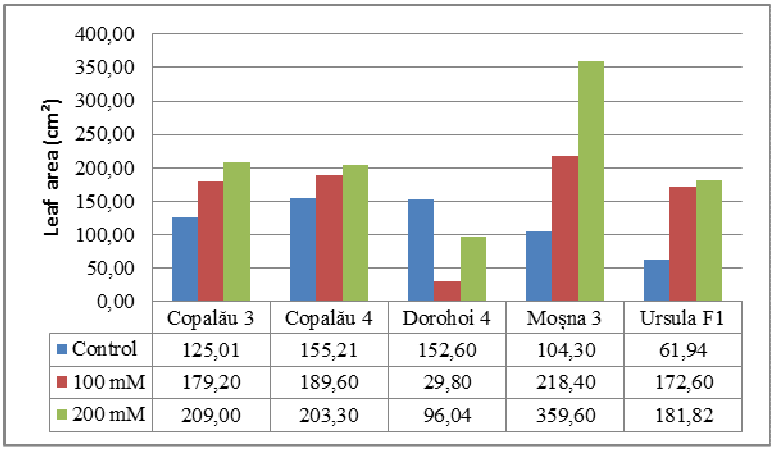
The biological material was represented by local tomatoes populations collected from areas with saline soils from Moldavia region and compared with commercial type salt-tolerant tomato (*Ursula* $_{F1}$) from Israel.

The bifactorial experience was conducted in a pots experiment in randomized blocks with four repetitions. Four tomato genotypes (*Copalău*$_3$, *Copalău* $_4$, *Dorohoi* $_4$, *Moşna* $_3$) studied were subjected to salt stress for a period of 30 days is constantly wetted with saline solution to a concentration of 100 mM and 200 mM.

The chlorophyll content was determinate whit chlorophyll meter and the leaf area with portable scanner Area Meter AM 300 – 0002.

RESULTS AND DISCUSSIONS

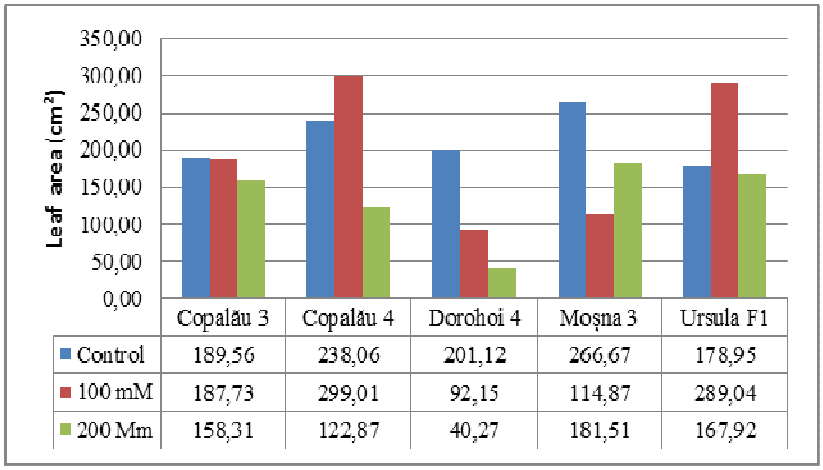
After 30 days, treatment with *NaCl* did not interfere with the foliar growth in the base of the stem. Found an increase in leaf area in the two concentrations of *NaCl* compared to control, in tomato genotypes under study, which shows that the leaves appeared before applying treatments with *NaCl*, grown in the absence of stress are not affected. An exception is genotype *Dorohoi*$_4$ exposed to a concentration of 100 mM (fig. 1).



	Copalău 3	Copalău 4	Dorohoi 4	Moșna 3	Ursula F1
Control	125,01	155,21	152,60	104,30	61,94
100 mM	179,20	189,60	29,80	218,40	172,60
200 mM	209,00	203,30	96,04	359,60	181,82

Fig. 1 - Effect of salt stress on leaf growth in the basal third of the stem

The middle third of the stem, salinity affects leaf growth compared to control to 2 genotypes (*Dorohoi*$_4$, *Moşna*$_3$,) exposed to 100 mM *NaCl* concentration. In the exposure 200 mM all genotypes showed lower values of leaf area. Compared to *Ursula* $_{F1}$, salinity resistant varieties all untreated genotypes showed higher values. When exposed to excess salt *Copalău*$_4$ one genotype showed higher values (fig. 2).



	Copalău 3	Copalău 4	Dorohoi 4	Moșna 3	Ursula F1
Control	189,56	238,06	201,12	266,67	178,95
100 mM	187,73	299,01	92,15	114,87	289,04
200 Mm	158,31	122,87	40,27	181,51	167,92

Fig. 2 - Effect of salt stress on leaf growth in the middle third of the stem

The manifestation of the negative effects of excess salt, to the leaves of the middle, can be explained by the fact that the leaves have started to grow under salt stress effect, and by the fact that the ions are transported with the mineral water to areas where they accumulate increased sweating.

Increased leaf from the top stem was strongly affected by excess salt to the all cultivars studied (fig. 3).

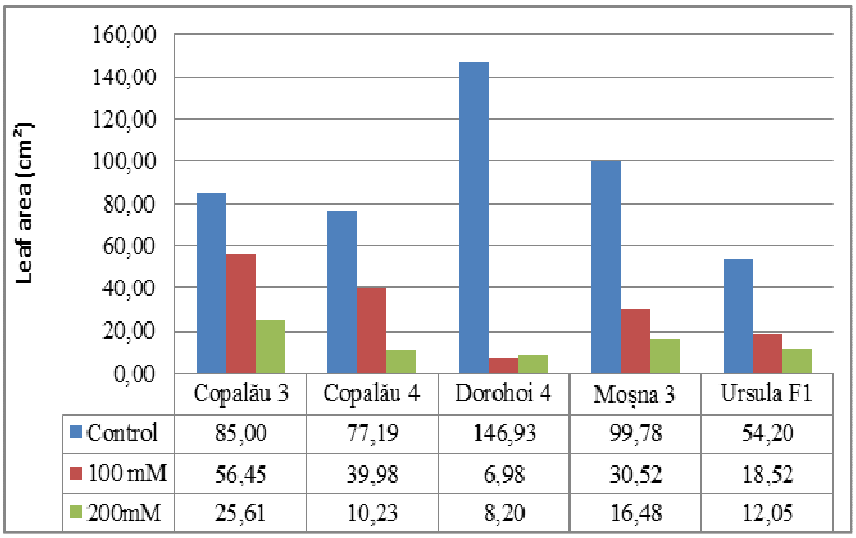


Fig. 3 - Effect of salt stress on leaf growth in the top of the stem

The chlorophyll content of leaves was determined as chlorophyll content index (CCI).

After 15 days salt stress has been found differences between the genotypes in the concentration of chlorophyll in the leaf level. In the case of tomatoes exposure to 100 mM concentration, in comparison with the plants watered only with water there is a lower value genotypes *Moşna*$_3$, *Copalău*$_3$ and an increase in chlorophyll index for the other genotypes.

When exposed to 200 mM the chlorophyll content is higher for all genotypes (fig. 4). This shows that plants to chlorophyll content index registers values higher compared with control variant, are in the phase osmotic stress.

After 30 days exposure to 100 mM, compared to control variant observed higher values of chlorophyll content of leaves for all genotypes, except genotype *Dorohoi*$_4$ which shows their maintenance during osmotic stress. In the exposure of 200 mM genotypes *Dorohoi*$_4$, *Moşna*$_3$ and *Ursula* $_{F1}$ have higher values compared to plants watered with water only, this means that they are still in the process of osmotic stress.

Genotypes *Copalău*$_4$ *Copalău*$_3$ has lower values compared with to control variant (fig. 5). In this case switch-on the second phase of stress, the ion toxicity, disturbances in the chloroplast. The transition from phase I (osmotic stress) to phase II (ions toxicity), is done by salt stress intensity, but mostly by cultivated genotype (Muuns, 1993).

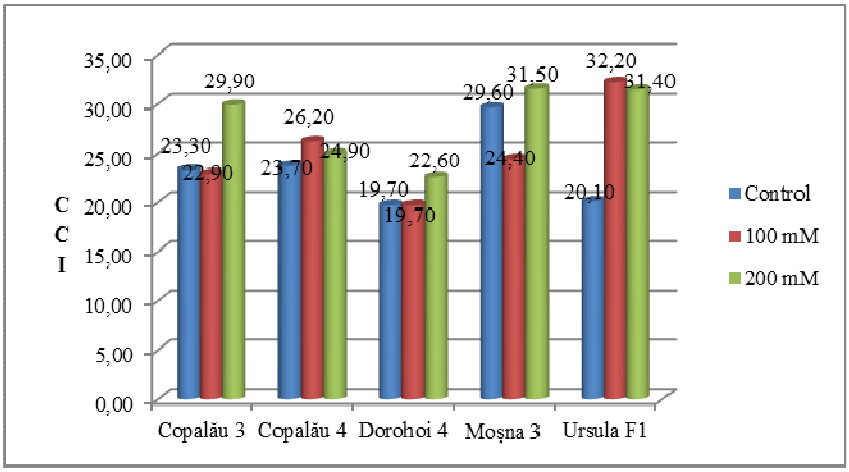


Fig. 4 - The index chlorophyll content after 15 days of exposure to salt stress

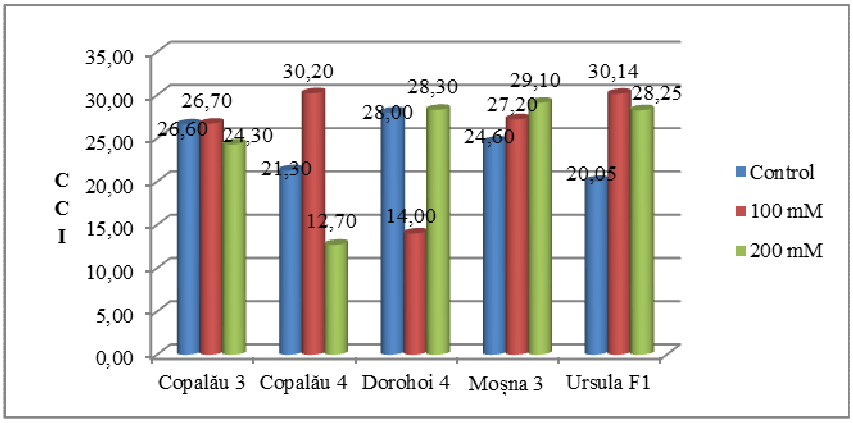


Fig. 5 - The index chlorophyll content after 30 days of exposure to salt stress

CONCLUSIONS

After 30 days, treatment with *NaCl* did not interfere with the foliar growth in the base of the stem but the middle third of the stem, salinity affects leaf growth compared to control to 2 genotypes. In the exposure 200 mM all genotypes showed lower values of leaf area.

Increased leaf from the top stem was strongly affected by excess salt to the all cultivars studied.

The plants to chlorophyll content index showing the higher values compared with control variant, are in the phase osmotic stress.

The transition from phase I (osmotic stress) to phase II (ions toxicity), is done by salt stress intensity, but mostly by cultivated genotypes.

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

REFERENCES

1. **Babu M.A., Singh D., Gothandam K.M., 2011** - *Effect of salt stress on expression of carotenoid pathway genes in tomato*. Journal of Stress Physiology & Biochemistry, 7(3), pp. 87-94.
2. **Doganlar Z.B., Demir K., Gul I., 2010** - *Effects of salt stress on pigment and total soluble protein contents of three different tomato cultivars*. African Journal of Agricultural Research, 5(15), pp. 2056-2065.
3. **Jităreanu C. D., 2007** - *Fiziologia plantelor*. Ed. Ion Ionescu de la Brad, Iasi.
4. **Munns R.,** 1993 - *Physiological processes limiting plant growth in saline soils: some dogmas and hypotheses.* Plant Cell and Environment 16, pp. 15-24.
5. **Popescu (Ciobanu) I., Şumălan R., 2009** - *The effects of the salinity stress on the growing rates and physiological characteristics to the Lycopersicum esculentum specie*. Buletin UASVM Horticulture, 66 (2), pp. 616-620.
6. **Slabu C.**, 2005 – *Physiologische Reaktion der Ackerbohne (Vicia Faba L.) auf Natrium und Chlorid*. Ed. Ion Ionescu de la Brad, Iasi.
7. **Şumălan R., Dobrei C., 2002** - *Fiziologie vegetală lucrări practice*. Ed. Marineasa Timișoara.
8. **Yaşar A., Esra S., 2012** -*The effect of salt stress on growth, chlorophyll content, proline and nutrient accumulation, and K/Na ratio in walnut*. Pakistan Journal of Botany, 44(5), pp. 1513-1520.
9. **Zhani K., Mariem B.F., Fardaous M., and Cherif H., 2012** - *Impact of salt stress (NaCl) on growth, chlorophyll content and fluorescence of Tunisian cultivars of chili pepper (Capsicum frutescens L.)*. Journal of Stress Physiology & Biochemistry, 8(4), pp. 236-252.

EXPERIMENTAL RESULTS ON RUNNER BEAN CULTIVATION (*PHASEOLUS COCCINEUS* L.) IN INTERCROPPING SYSTEM

REZULTATE EXPERIMENTALE PRIVIND CULTIVAREA FASOLEI MARI (*PHASEOLUS COCCINEUS* L.) ÎN SISTEM INTERCROPPING

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Abstract. *This paper presents the behavior of runner bean (Phaseolus coccineus L.) in three intercropping systems, with maize, sunflower and Jerusalem artichoke, compared to a pure culture system (available in three plant display variants - trellis with individual string on a single row, individual string trellis on two rows and synthetic net trellising). The results were evaluated based on the main morphological and phenological plant characteristics, including agro-productivity. The results revealed significant differences between the studied variants, demonstrating the superiority of pure culture growing on synthetic mesh trellis.*
Key words: *pure crop, associated crop, trellising system, runner bean yield*

Rezumat. *Lucrarea prezintă modul de comportare a fasolei mari (Phaseolus coccineus L.) în trei sisteme de intercropping cu porumb, cu floarea soarelui și cu topinambur, în comparație cu sistemul de cultură simplă (în trei variante de dispoziție a plantelor – palisat pe spalier cu sfoară individual într-un singur rând, palisat pe spalier pe sfoară individual pe două rânduri și palisat pe spalier cu plasă sintetică). Rezultatele au fost evaluate pe baza principalelor caracteristici morfologice și fenologice ale plantelor, inclusiv cele de agroproductivitate. Rezultatele au pus în evidență diferențe semnificative între variantele studiate, demonstrând superioritatea variantei de cultivare în cultură pură palisată pe spalier cu plasă sintetică.*
Cuvinte cheie: *cultură pură, cultură intercalată, sistem palisare, producție de fasole mare*

INTRODUCTION

Runner bean (*Phaseolus coccineus* L.) is an herbaceous, annual and out-crossing species, propagated, usually, by seeds, adapted to a relativ cool and humid climate. Such a climate it is not proper for the North – East region of Romania (Popa, 2010). To achieve cost-effective production, it is necessary to diversify the cultivation of this species, through optimization of design and period setting and the use of various support systems, such as runner beans' interleaving with other cultivated species (maize, sunflower, Jerusalem artichoke), whose stem is also supporting bean plants (Hamburdă et al., 2013).

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Intercropping is based on the management of plant interactions, in order to increase the productivity (Vandermeer, 1989; Wiley, 1979 a, b). The success of intercropping depends on thorough knowledge of the botanical, ecological and technological particularities of associated species, especially habitus, root morphology, requirements for water and nutrients. The purpose of this system is to put the growing crops in the same place throughout the growing season or at least for a period of time (Mousavi and Eskandari, 2011). The plants involved in intercropping may be seeded or planted at the same time or at different times (Ofori and Stern, 1987).

This paper presents the behavior of runner bean in three intercropping systems, with common maize (*Zea mays* L.), sunflower (*Helianthus annuus* L.), and Jerusalem artichoke (*Helianthus tuberosus* L.), compared to a pure culture system (available in three plant display variants - trellis with individual string on a single row, individual string trellis on two rows and synthetic mesh trellising). The need for this research stems from the fact that a standard technology for runner bean crop is not yet developed. Thus, it is an attempt to obtain cost-effective productions, by using different means of trellising and determining the most efficient trellising system.

MATERIAL AND METHOD

In accordance with the purpose and research objectives, we organized an experience, in 2013, in the Vegetable growing department's experimental field, at "V. Adamachi" farm, U.A.S.V.M. Iaşi.

The available biological material was the runner bean seeds (C_3 local population), seeds of maize (Flato hybrid), sunflower seeds (Tristan hybrid) and Jerusalem artichoke tubers (Topstar cultivar).

Organization of the experiment was in a randomized blocks device with three repetitions. The studied experimental factor was the trellising system with six graduations, in field conditions:

V_1 = on individual string trellis with double rows,
V_2 = on individual string trellis with a single row,
V_3 = on synthetic net trellis (17 cm mesh) in a single row,
V_4 = intercropping with common maize,
V_5 = intercropping with sunflower,
V_6 = intercropping with Jerusalem artichoke.

Each recurrence plot had a 6 m^2 surface area, which consists of two spaced rows of 1.0 m, ensuring a distance of 0.4 m between runner bean plants. For the pure culture, the system consisted of a reinforced concrete support trellis formed of pillars and a steel wire of about 2 cm on the top thereof.

The establishment of runner bean crop was conducted between 1.05 - 30.05. Sowing maize and sunflower, respectively planting artichokes were made about two weeks before sowing runner bean. Crop establishment was performed by direct seeding, three runner bean seeds/nest and two maize seeds or sunflower seeds/nest. At the time of emergence, there have been left, in each nest, two runner bean plants and one of maize and sunflower plants. Artichokes were planted by two tubers/nest, and at emergence time, only one stem/plant were allowed. Thus, in the case of trellising runner bean plants on maize/sunflower/Jerusalem artichoke, for each repetition, there were 30 nests of runner bean plants and 14 plants of

maize/sunflower/Jerusalem artichoke; in the runner bean mesh and string trellising systems, there were 30 nests of runner bean plants on each repetition.

The experimental culture was conducted according to technical rules arising from the specialized literature consulted (Munteanu et al., 1989, Stan N. et al., 2003, Ruști, 2007; Popa, 2010; Axinte et al., 2006). The culture was performed on well leveled ground; the soil is a cambic chernozem, well-stocked in nutrients, with an organic matter content of 3.2 - 3.4% and a pH of 6.5-6.8. Weather conditions during the growing season (months IV to IX) were characterized by an average temperature of 17.8^0C, an average relative humidity of 67% and a rainfall amount of 495.4 mm.

Works were carried out as recommended for common climbing beans (Munteanu et al., 1982; Ruști and Munteanu, 2008; Popa, 2010). The culture was not irrigated.

Basic research methods were the observation and the experiment, in which, for evaluating the performance of studied variants, biometric observations and measurements were conducted on the main morphological and phenological plant features, including agro-productivity. Production results were statistically interpreted according to the specialized literature (Jităreanu, 1994; Săulescu and Săulescu, 1967).

RESULTS AND DISCUSSIONS

Between the studied trellising systems, there was no significant differences in the morphological and phenological characterization of the runner bean plants.

Results in terms of morphological characterization: runner bean plant port is voluble; size (height) of the plant is over two meters; plant vigor is high; number of branches per plant is three to four; the foliage color is dark green; flower color is white; seed color is white; pods of widely varying size, with length of 8-17 cm and a width of 1.4 to 2 cm; it is important to point out that the pod's length correlates with the number of seeds/pod, longer pods having a higher number of seeds; seed size, measured by the length of the longest axis, varies between 15 and 22 mm; the number of seed per pod ranges from two to four.

Results in the phenological characterization: emergence is hypogeic, meaning it is achieved only by the epicotil growing above ground, the cotyledon leaves remaining in the soil; period from sowing to emergence was about seven to ten days, the period from emergence to appearance of the first real trifoliate leaves was around five to seven days, the period from emergence to the first flowers was about 32 - 35 days, the period from emergence to the first pods was about 67-70 days, the period from emergence to seed maturation was around 115-122 days and the period from emergence to the end of the vegetation period was around 130-140 days (tab. 1).

Table 1

Phenological characters (number of days)

Sowing - emergence	Emergence - first real trifoliate leaf	Emergence – first flowers	Emergence – first pods	Emergence – seed maturation	Emergence – end of vegetation
7-10	5-7	32-35	67-70	115-122	130-140

Results in terms of runner bean production - pure culture

Following the investigations, it appears that seed production ranged from 2733 kg/ha to 3325 kg/ha. Highest production was obtained in the variant with trellising runner beans on synthetic mesh (3325 kg/ha), which showed distinctly significant positive differences as compared to the mean (3080 kg/ha), while the lowest production variant was obtained on runner bean individual trellising on double rows (2733 kg/ha), a very significant negative differences from the control being highlighted. The second variant (individual string trellis with a single row), achieved yields within the average experimental variation limits (tab. 2).

Table 2

Results obtained in pure culture of runner bean (year 2013)

Variant			Yield of runner bean		Differences to mean (kg/ha)	Semnification
no.	specification		kg/ha	% of the mean		
1	pure culture	V_1	2733	88,7	-347	ooo
2		V_2	3182	103,3	+102	^{NS}
3		V_3	3325	107,9	+245	**
Mean			3080	100	-	-

^{w}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;
o,oo,ooo - negative significant at p ≤ 0.05, 0.01, 0.001, respectively
LSD 5% = 103,43 (kg/ha)
LSD 1% = 171,15 (kg/ha)
LSD0,1% = 320,34 (kg/ha)

Results in terms of runner bean production - intercropping

Following investigations, it appears that seed production ranged from 789 kg/ha and 3093 kg/ha (tab. 3).

Table 3

Results obtained in intercropping system (year 2013)

Variant			Runner bean yield		Differences to mean (kg/ha)	Semnification
no.	specification		kg/ha	% of the mean		
1	intercropping	V_4	1966	100,8	+16,6	^{NS}
2		V_5	3093	158,6	+1143,6	***
3		V_6	789	40,4	-1160,3	ooo
Mean			1949	100	-	-

^{w}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;
o,oo,ooo - negative significant at p≤0.05, 0.01, 0.001, respectively
LSD 5% = 271,37 kg/ha
LSD 1% = 449,03 kg/ha
LSD 0,1% = 840,47 kg/ha

Highest yield was obtained when interleaving runner bean with sunflower (3093 kg/ha), which recorded very significant positive differences as compared to the mean (1949 kg/ha), while the lowest yield was obtained in the intercalation of runner bean with Jerusalem artichoke (789 kg/ha), very significant negative differences from the control being highlighted. The third option (intercropping with maize) achieved yields within the average experimental variation limits.

Results in the production of runner bean - total experience

Following investigations, it appears that seed yield ranged from 789 kg/ha and 3325 kg/ha. Highest yield was obtained in the variant with trellising runner bean on synthetic mesh (3325 kg/ha), which recorded very significant positive differences as compared to the experimental mean (2515 kg/ha), while the lowest production was obtained when interleaving runner bean with Jerusalem artichoke (789 kg/ha), very significant negative differences from the control being highlighted (Tab. 4).

Table 4

Yield of runner bean per total experience (year 2013)

Variant			Runner bean production		Differences to mean (kg/ha)	Semnification
no.	Specification		kg/ha	% of the mean		
1	pure culture	V_1	2733	108,6	+218	x
2		V_2	3182	126,5	+667	xxx
3		V_3	3325	132,2	+810	xxx
4	intercropping	V_4	1966	78,1	-549	ooo
5		V_5	3093	122,9	+578	xxx
6		V_6	789	31,3	-1726	ooo
Experience mean			2515	100	-	-

[w]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;

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

LSD 5% = 163,26 kg/ha
LSD 1% = 232,08 kg/ha
LSD0,1% = 336,04 kg/ha

CONCLUSIONS

1. There were no significant differences in characterization of runner bean plants, regarding plant height, vigour of the plant, pod size, seed size, seed number in a pod, vegetation period.

2. The highest production results were obtained in the pure culture system (3080 kg/ha), in comparison with the intercropped culture (1949 kg/ha).

3 In terms of overall experience production, it ranged from 789 kg/ha (V_6 - intercropping with Jerusalem artichoke) and 3325 kg/ha (V_3 - trellising system on synthetic net).

***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. **Axinte M., Roman Gh.,V., Borcean I., Muntean L.S., 2006** - *Fitotehnie.* Editura "Ion Ionescu de la Brad", Iaşi.
2. **Hamburdă Silvia Brîndușa, Dascălu T., Munteanu N.**, **2013** – *Preliminary studies for new cultivation systems of runner bean (Phaseolus coccineus L.).* Lucrări științifice, seria Horticultură, USAMV Iași. 56 (2), pp. 167-172.
3. **Jităreanu G., 1994** – *Tehnică experimentală*. Editura "Ion Ionescu de la Brad", Iași, 167 pag.
4. **Munteanu N., Timofte Valentina, Timofte E., 1989** – *Aspecte tehnologice ale culturii fasolei de grădină urcătoare*. Cercetări Agronomice în Moldova, vol. 4, Iași.
5. **Mousavi S.R., Eskandari H., 2011** - *A general overview on intercropping and its advantages in sustainable agriculture*. J.Appl. Environ. Biol. Sci. 1 (11), pp. 482-486.
6. **Ofori F., Stern W.R., 1987** - *Cereal-legume intercropping system*. Advance in Agronomy. 41, pp. 41-90.
7. **Popa Lorena Diana, 2010** - *Cercetări privind agrobiologia speciei Phaseolus coccineus L. în vederea optimizării cultivării*. Teză de doctorat. USAMV Iaşi. 232 p.
8. **Ruști Gr., 2007**- *Cercetări privind îmbunătățirea tehnologiei de cultură a fasolei de grădină urcătoare (Phaseolus vulgaris L. var. comunnis L).* Teză de doctorat. U.Ș.A.M.V. Iași.
9. **Ruşti G., Munteanu N., 2008** – *Cultura fasolei de grădină urcătoare*. Editura „Ion Ionescu de la Brad", Iaşi.
10. **Stan N., Munteanu N., Stan T., 2003** – *Legumicultură*, vol III. Editura "Ion Ionescu de la Brad", Iaşi.
11. **Săulescu N.A., Săulescu N.N., 1967** - *Câmpul de experiență*- ediția a II a. Edit. Agro-Silvică de stat, Bucureşti.
12. **Vandermeer J.H., 1989** - The ecology of intercropping. Cambridge University Press, Cambridge, U.K.
13. **Willey R.W., 1979a** - *Intercropping–Its importance and research needs. Part 1. Competition and yield advantages.* Field Crop Abstr 32, pp. 1–10.
14. **Willey R.W., 1979b** - *Intercropping–Its importance and research needs. Part 2. Agronomy and research approaches.* Field Crop Abstr 32, pp. 73–85.