

## THE STUDY OF VEGETABLE EXTRACT ACTION OF *LOLIUM PERENNE* L. ON SOME PERENNIAL GRAMINAES CHEMICAL COMPOSITION

## STUDIUL ACȚIUNII EXTRACTULUI VEGETAL DE *LOLIUM PERENNE* L. ASUPRA COMPOZIȚIEI CHIMICE A UNOR GRAMINEE PERENE

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**Abstract:** The purpose of the researches is represented of determination of allopathic effect realized of *Lolium perenne* L. on perennial gramineas *Festuca rubra* L., *Dactylis glomerata* L. and *Poa pratensis* L. In the vegetal preparation of *Lolium perenne* L. have been found compounds with allopathic character which corresponds to some other researches realized on global level. The studied biologic material is represented of four perennial gramineas species: *Dactylis glomerata* L., *Lolium perenne* L., *Poa pratensis* L. and *Festuca rubra* L., studied in laboratory conditions and in vegetation pots. The experience have been established in three repetitions. Excepting the witness that have been watered only with running water, the other vegetation pots have been splashed with deafly alcoholic pirolizidinici alkaloids white extract, respectively ergotic obtained from the aerial vegetal part of *Lolium perenne* L. specie. The extract have been applied in three different shots, shot of 10 ml/pot, 40 ml/pot and 80 ml/pot. The novelty degree of the paper and personal contribution results from the fact that on national level, perennial gramineas that have been studied have not been completely characterized from allopathic point of view. Between the factors that threaten the biodiversity and to which must get the profile research are allelochemical compounds. Their structure and their way of action are different and can built start points for realizing new herbicides. The paper importance results from the fact that through these experiments, have been proved the alkaloids influence (on changing the quality parameter of plants that have been studied) did reflected through an a small decrease on crude protein contain.

**Rezumat:** Scopul cercetărilor îl reprezintă determinarea efectului alelopativ exercitat de *Lolium perenne* L. asupra gramineelor perene *Festuca rubra* L., *Dactylis glomerata* L. și *Poa pratensis* L. În preparatul vegetal de *Lolium perenne* L. s-au regăsit compuși cu caracter alelopativ ceea ce corespunde cu alte cercetări realizate pe plan mondial. Materialul biologic studiat este reprezentat de patru specii de graminee perene: *Dactylis glomerata* L., *Lolium perenne* L., *Poa pratensis* L. și *Festuca rubra* L., studiate în condiții de laborator și în vase de vegetație. Experiența a fost instalată în trei repetiții. Cu excepția matorului care a fost udat doar cu apă de la robinet, plantele din celelalte vase de vegetație au fost stropite cu extract slab alcoolic de alcaloizi pirolizidinici, respectiv ergotici obținut din partea vegetală aeriană a speciei *Lolium perenne* L. Extractul a fost aplicat în trei doze diferite, doze de 10 ml/ghiveci, 40 ml/ghiveci și 80 ml/ghiveci. Gradul de noutate al lucrării și contribuția personală rezultă din faptul că pe plan național, gramineele perene luate în studiu nu s-au caracterizat complet din punct de vedere alelopativ. Printre factorii care amenință biodiversitatea și spre care trebuie să se îndrepte cercetarea de profil, se află și compușii alelochimici. Structura și modul lor de acțiune sunt diverse și pot constitui puncte de plecare pentru fabricarea de noi erbicide. Importanța lucrării rezidă din faptul că, prin aceste experimente, s-a demonstrat că influența alcaloizilor asupra modificării indicelui de calitate a plantelor luate în studiu s-a reflectat printr-o ușoară scădere a conținutului de proteină brută.

**Key words:** perennial gramineas, allelopathy, lolinici acizi, crude protein  
**Cuvinte cheie:** graminee perenne, alelopatie, acizi lolinici, proteină brută.

## INTRODUCTION

The allelopathy attracts many researchers from various disciplinary branches and from all around the world, which proves that this field of science must be studied through an interdisciplinary approach.

Both the similar chemical composition and the coexistence of different plant species can be strongly affected by interactions between them (INDERJIT AND CALLAWAY, 2003).

The existence of inhibition areas around perennial gramineae (*Festuca rubra L.*, *Lolium perenne L.*, *Poa pratensis L.* and *Dactylis glomerata L.*) and the decreasing of the diversity of the plants' species that are composing the meadows, and also the changes that are suffered by the composition of the soil from these habitats are suggesting the involvement of some chemical compounds.

The main target processes for the allelopathic substances are: cell division, permeability and stability of the membranes, production and balance of vegetal hormones, protein production, photosynthesis and respiration (RIZVI ET AL. 1992; FERGUSON ET AL. 2003). These effects that are slowing and even stopping some fundamental processes for plants, entail competitiveness and, at the same time, superiority to the allelopathic plant even if it has limited access to nutritional resources.

Chemical studies conducted on perennial gramineae and not only, have showed that the substances that are causing the apparition of allelopathy are secondary metabolites of vegetal origin that are a part of the classes of alkaloids, isoprenoids, flavonoids, phenols, terpenoids and glucosinolates.

These substances are virtually present in all plant tissues, but in practice they are concentrated in leaves, roots and vegetal remains. (BOUTON, 2005)

It is important to identify the classes of allelopathic compounds and also the concentrations respectively the mechanisms by which they reach the environment, so that we can form a general view of their function as precise as possible.

Recent researches (CASTELLS ET AL. 2004; INDERJIT AND WEINER, 2001) suggest that allelopathic substances can indirectly intermediate the chemical interactions between plants due to change of soil composition.

The pyrrolizidine alkaloids (lolines) are toxic products that are found in plants like: *Lolium perenne L.*, *Festuca rubra L.*, *Dactylis glomerata L.* and *Poa pratensis L.* The main representatives of this class of chemical compounds, N-formylloline respectively N-acetylloline have allelopathic properties such as germination inhibition at mono and dicotyledonous seed (DAVIES ET AL. 1993; LATCH&TAPPER, 1988). When these alkaloids are found in sufficiently high concentrations, they induce negative effects on the development of plants that are grown in symbiosis with the perennial plants (PÉREZ ET AL. 1991, TSANUO ET AL. 2003, FERGUSON ET AL, 2003).

## MATERIAL AND METHODS

The studied biological material is represented by four species of perennial gramineae: *Dactylis glomerata L.*, *Lolium perenne L.*, *Poa pratensis L.* and *Festuca rubra L.*, studied under laboratory conditions and in vegetation pots.

The experience has been installed in three repetitions. Excepting the control who was watered only with tap water, the plants from the other vegetation vases were sprayed with low alcoholic extract of pyrrolizidine alkaloids, respectively ergotics obtained from the aerial part of the *Lolium perenne L.* The extract was applied in three different doses, doses of 10 ml/pot, 40 ml/ pot and 80 ml/pot.

The determination of the ergovaline respectively lolinic alkaloids content in the vegetal extract was determined by chromatographic analysis.

## RESULTS AND DISSCUSION

Analyzing the experimental data presented in Tables 1,2 and 3 it can be observed that spraying plants with *Lolium perenne L.* extract leads to changes in their chemical composition, changes that vary with the applied dose.

The concentration values of NFL (N-formylloline), NAL (N-acetylloiline) and EGV (ergovaline) increase by the increasing of the dose of extract.

At *Festuca rubra L.*, the NFL increased regardless of the applied dose. Thus, the dose of 10 ml increases it by 1 µg/g, the dose of 40 ml increases it by 1.02 µg/g and 80 ml by 1.03 µg/g. The same happens with NAL, N-acetylloiline which increases by the increasing of the applied dose, thus the application of the first dose of extract leads to an increase of 1 µg/g, at a dose of 40 ml/pot the growth is 1.02, and at 80 ml/pot of 1.03 µg/g. Same trend of increased concentrations of NFL and NAL is also observed at the other studied perennial graminas that were sprayed with the three different doses of extract.

In what concerns the other alkaloid, the ergovaline, it has a tendency to accumulate only at *Dactylis glomerata L.* at a dose of 80ml/pot. If EGV (ergovaline) is absent in *Dactylis glomerata L.* plants used as controls in the other variants it is observed an increase of 0.070 µg/g. This EGV accumulation can be explained by taking into account the higher polarity of the ergovaline molecule as compared with that of N-acetylloiline.

The application of some doses of *Lolium perenne L.* extract caused a slight decrease in crude protein content; the decrease of crude protein is most evident in plants that were sprayed with a dose of 80 ml/pot. For example at *Festuca rubra L.* by applying a dose of 80 ml/pot the crude protein decreases by 1.04 times. A decrease in crude protein is also recorded at *Dactylis glomerata L.* when sprayed with a higher dose in an amount of 80 ml/pot. Thus, the crude protein content at the treated *Dactylis glomerata L.*, is reduced 1.1 times as compared to the *Dactylis glomerata L.* control. Treated *Poa pratensis L.* shows a decrease in crude protein by 1.02 times as compared to the control. A decrease in crude protein is also observed at the other doses of 10 and 40 ml/pot. There is only one exception, in the case of *Poa pratensis L.*, that when it was sprayed with a 10 ml dose, registered a slight increase in crude protein.

The same trend is also evident in the case of the NDF (neutral detergent fiber) and ADF (acid detergent fiber) variations, except for *Festuca rubra L.* which registered an increase of the acid detergent fiber (ADF) percentage when sprayed with *Lolium perenne L.* extracts.

Table 1.

The chemical composition of the control plants

Specia	Cenuşa %	CP %	NDF %	ADF %	NFL ug/g	NAL ug/g	EGV ug/g
<i>Festuca rubra L.</i>	11.37	17,98	70.78	28.45	1550	771	0.129
<i>Lolium perenne L.</i>	11.10	17.78	69.35	29.75	541	127	0.071
<i>Dactylis glomerata L.</i>	10.80	18.78	74.89	31.45	0,090	0,050	ned
<i>Poa pratensis L.</i>	10.12	16.45	73.09	31.56	0,045	0,015	ned

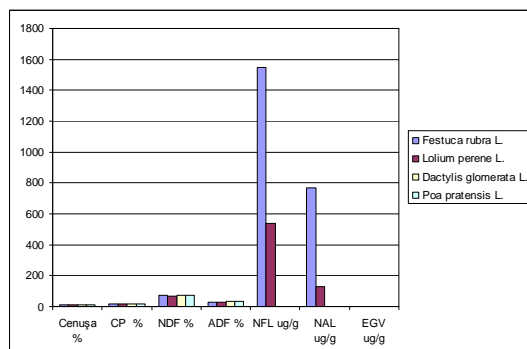


Figure 1. Graphic representation of the chemical composition of the control plants

Table 2.  
The chemical composition of the plants after spraying them with *Lolium perenne L.* extract in a dose of 10ml/pot

Specia	Cenusa %	CP %	NDF %	ADF %	NFL ug/g	NAL ug/g	EGV ug/g
<i>Festuca rubra L.</i>	11.25	17.72	70.11	37.82	1563	775	0.129
<i>Dactylis glomerata L.</i>	10.74	18.27	74.05	29.12	0,090	0,050	ned
<i>Poa pratensis L.</i>	9.77	16.71	71.88	31.49	0,045	0,015	ned

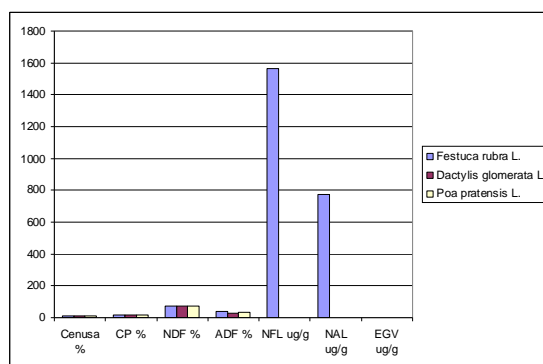


Figure 2. Graphic representation of the chemical composition of the plants after spraying them with *Lolium perenne L.* extract in a dose of 10ml/pot

Table 3.  
The chemical composition of the plants after spraying them with *Lolium perenne L.* extract in a dose of 40ml/pot

Specia	Cenusa %	CP %	NDF %	ADF %	NFL ug/g	NAL ug/g	EGV ug/g
<i>Festuca rubra L.</i>	11.85	17.48	69.21	37.52	1580	780	0,128
<i>Dactylis glomerata L.</i>	10.54	17.88	73.24	28.87	0,123	0,081	ned
<i>Poa pratensis L.</i>	10,11	16.42	71.16	30.78	0,075	0,021	ned

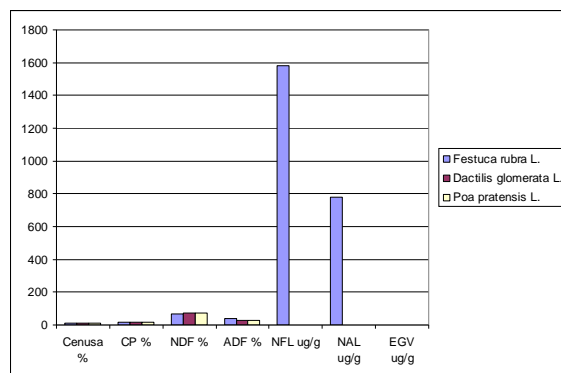


Figure 3. Graphic representation of the chemical composition of the plants after spraying them with *Lolium perenne L.* extract in a dose of 40ml/pot

Table 4.

The chemical composition of the plants after spraying them with *Lolium perenne L.* extract in a dose of 80ml/pot

Specia	Cenusa %	CP %	NDF %	ADF %	NFL ug/g	NAL ug/g	EGV ug/g
<i>Festuca rubra L.</i>	11.75	17.24	68.78	36.95	1600	790	0,128
<i>Dacyilis glomerata L.</i>	10.27	17.03	72.35	27.45	0,187	0,121	0,070
<i>Poa pratensis L.</i>	10.07	16.05	69.21	28.25	0,128	0,035	ned

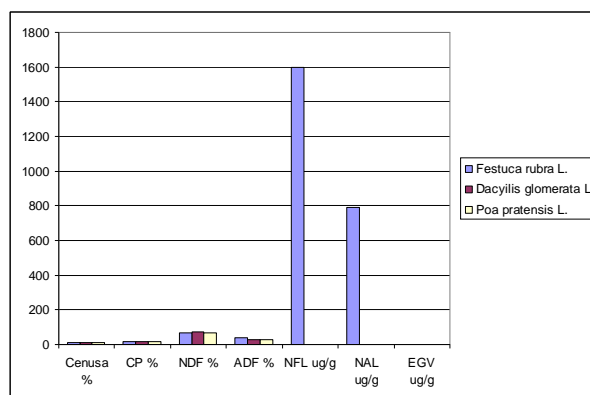


Figure 4. Graphic representation of the chemical composition of the plants after spraying them with *Lolium perenne L.* extract in a dose of 80ml/pot

### CONCLUSIONS

1. In the *Lolium perenne L.* extract allelopathic compounds were found, corresponding to other research conducted worldwide
2. From the many alkaloids which are known in the literature, the following have been identified:

NFL- N-formylloline, NAL- N-acetylloine, EGV-ergovaline.

3. The *Lolium perenne* L. extract is characterized by an increased content of p-cumaric acid and p-phenolic acid derivatives.

4. The influence of the alkaloids on the modification of the quality index in the first's phenophases of development was reflected by a slight decrease in crude protein content.

#### BIBLIOGRAPHY

1. BOUTON, F., 2005 - *Mise en évidence du potentiel allélopathique de la graminée Festuca paniculata dans les prairies subalpines*. Masters Report: Université Joseph Fourier.
2. EINHELLIG, F.A., 1995 - *Allelopathy: current status and future goals*. In "Allelopathy: Organisms, Processes, and Applications" (Inderjit, K.M.M. Dakshini and F.A. Einhellig, Eds.); ACS Symposium Series 582, American Chemical Society, Washington, DC, 1-25.
3. FERGUSON, J.J., and RATHINASABATHI. H., 2003. - *Experimental design for the study of allelopathy*. Plant and Soil 256 : 1-11.
4. INDERJIT C., and WEINER J., 2001 - *Plant allelochemical interference or soil chemical ecology? Perspectives in Plant Ecology, Evolution and Systematics*, 4: 3-12.
5. MORRIE CRAIG, A. DAN BILICH, JEANNETTE T. HOVERMALE, RONALD E. WELTY, 1994 - *Improved extraction and HPLC methods for ergovaline from plant material and rumen fluid*. J Vet Diagn Invest 6:348-352.
6. MOUBARAK, A. S., E. L. PIPER, C. P. WEST, AND Z. B. JOHNSON, 1993 - *Interaction of purified ergovaline from endophyte-infected tall fescue with synaptosomal ATPase enzyme system*. J. Agric. Food Chem. 41:407.
7. PERELLINO, N. C., J. MALYSZKO, M. BALLABIO, B. GIOIA, AND A. MINGHETTI, 1992 - *Direct biosynthesis of unnatural ergot alkaloids*. J. Nat. Prod. (Lloydia) 55:424.
8. PERELLINO, N. C., J. MALYSZKO, M. BALLABIO, B. GIOIA, AND A. MINGHETTI, 1993 - *Identification of ergobine, a new natural peptide ergot alkaloid*. J. Nat. Prod. (Lloydia) 56:489.
9. THOMPSON. E.N., STUEDMANN, J.A., HILL N.S., 2001 - *Anti-quality factors associated with alkaloids in eastern temperate pasture*. Journal of Range Management, 54:474-489.
10. YATES, S. G., PETROSKI, R. J., AND POWELL, R. G., 1990 - *Analysis of loline alkaloids in endophyte-infected tall fescue by capillarygas chromatography*. J. Agric. Food Chem. 38:182-185.
11. CASTELLS M., PENUELAS A., AND VALENTINE G.J., 2004 - *Are phenolic compounds released from the Mediterranean shrub Cistus albidus responsible for changes in N cycling in siliceous and calcareous soils?* New Phytologist, 162: 187-195.