# SOIL BIOTA QUALITY IMPROVEMENT BY INTRODUCING INTO THE CROP ROTATION OF LEGUME-CEREAL GRASS MIXTURES

## ÎMBUNĂTĂȚIREA CALITĂȚII BIOTEI SOLULUI PRIN INTRODUCEREA ÎN ROTAȚIE A AMESTECURILOR DE IERBURI LEGUMINOASE ŞI GRAMINEE

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***Abstract.*** *The effective restoration of biota in degraded arable chernozems occurs as a result of the multiannual grasses application. The use of alfalfa and ryegrass mixture during 3-5 years helps to restore the natural soil biota activity at the expense of exudates contribution in soil and roots of perennial crop residues. This procedure has favored the revival of population of invertebrates and microorganisms. The number of invertebrates on average was 2.5 times higher compared with the control, total biomass - by 1.6 times. This method is especially effective to restore the Lumbricidae family. Their number increased by 3.0 and biomass by 2.0 times. Invertebrates' diversity has improved. The microbial biomass content and dehydrogenase activity in the 0-25 cm layer were increased in average by 1.4 times. Perennial grass mixtures can be used in rows of perennial fruit crops for the biota's restoration in degraded soils.*
**Key words:** *soil biota, perennial grasses, restoration*

***Rezumat.*** *Restabilirea efectivă a numărului şi biomasei biotei cernoziomurilor arabile degradate are loc în rezultatul înţelenirii cu ierburi multianuale. Utilizarea amestecului de lucerna+raigras timp de 3-5 ani contribuie la restabilirea naturală al activităţii biotei solului din contul aportului în sol al exudatului rădăcinilor şi resturilor vegetale al plantelor multianuale. Acest procedeu a favorizat renaşterea populaţiei de nevertebrate şi microorganisme. Numărul nevertebratelor în medie a fost de 2,5 ori mai mare comparativ cu martorul, biomasa totală – de 1,6 ori. Această metodă este deosebit de efectivă pentru restabilirea fam. Lumbricidae. Numărul lor se măreşte de 3,0 ori, iar biomasa – de 2,0 ori. Diversitatea nevertebratelor s-a îmbunătăţit semnificativ. Conţinutul biomasei microbiene şi activitatea dehidrogenazei în stratul 0-25 cm s-a majorat în medie de 1,4 ori. Amestecurile de graminee perene pot fi folosite în rândurile culturilor pomicole multianuale pentru restaurarea biotei în solurile degradate.*
**Cuvinte cheie:** *biota solului, graminee perene, restabilire*

## INTRODUCTION

Soil biota plays an important role in the generation of soil ecosystem services and land productivity (Lavelle et al., 2006; Barrios, 2007). Soil invertebrates and microorganisms provide intermediate services in the agriculture, supply

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nutrients to plant (Fragoso et al., 1997), maintain the soil structure, improve the water infiltration, and participate in the soil organic matter decomposition (Swift et al., 2004; Barrios, 2007). Indices of the soil biota are the global indicators of soil quality and sustainability of ecosystems. The preservation of soil ecosystem services depends upon the preservation of soil biota. This may be achieved by using of environmentally friendly agricultural technologies based on the utilization of perennial grasses.

***The purpose of this research*** was to determine the influence of the perennial legume-cereal grass mixtures cultivation on the biota's recovery process in chernozems degraded as the result of the long-term agricultural utilization.

## MATERIAL AND METHOD

***Experimental sites.*** The soil biota has been tested in two experimental sites located in different zones of the Republic of Moldova.

The first site was located in the center of the country, in the Ivancha village, Orhei region. The long-term arable soil with crop rotation without fertilizers (control), and grass mixtures (ryegrass+lucerne and ryegrass+sainfoin) cultivation were tested. Plots under mixtures of perennial grasses (ryegrass+lucerne) were founded in the autumn of 2007. The mixture of ryegrass+sainfoin was seeded in the autumn of 2010.

The soil is the leached chernozem with humus content of 3.43 % and pH = 6.6 in the 0-25 cm layer.

The second site was located in the southern zone, in the Tartaul de Salchie village, Cahul region. These were plots with long-term arable (control) and mixture of ryegrass and lucerne and mixture of ryegrass and sainfoin. Plots under mixtures of perennial grasses (ryegrass+lucerne and ryegrass+sainfoin) were founded in the spring of 2010.

The soil is the ordinary chernozem with humus content of 3.16 % and pH = 7.1 in the 0-25 cm layer (Cerbari et al., 2012).

Soil samples were collected from the 0-25 cm layer of the experimental plots during 2010-2012 years.

***Status of invertebrates.*** The state of invertebrates was identified from test cuts by manually sampling the soil layers to the depth of soil fauna occurrence (Gilyarov and Striganova, 1987). The identification of invertebrate's diversity at the level of families was carried out with the implementation of Gilyarov and Striganova's method (1987).

***Microbiological properties.*** The microbial biomass carbon was measured by the rehydratation method based on the difference between carbon extracted with 0.5 M $K_2SO_4$ from fresh soil samples and from soil dried at 65-70$^oC$ for 24h, with Kc coefficient of 0.25 (Blagodatsky et al., 1987). $K_2SO_4$ - extractable carbon concentrations in the dried and fresh soil samples were measured simultaneously by dichromate oxidation; $K_2SO_4$-extractable carbon was determined at 590 nm spectrophotometrically.

***Enzymatic activity.*** The (potential) dehydrogenase activity was determined by the colorimetric technique on the basis of triphenylformazan (TPF) presence from TTC (2, 3, 5-triphenyltetrazolium chloride) added to air-dry basis of soil (Haziev, 2005).

The biological indices were evaluated statistically using the variation analysis. Statistical parameters of the state of soil invertebrates were calculated taking into account the depth of soil fauna occurrence, parameters of the microbial biomass and dehydrogenase activity – for the layer of 0-25 cm.

## RESULTS AND DISCUSSIONS

The application of perennial legume-cereal grass mixtures led to the restoration of the total number of invertebrates and the *Lumbricidae* family (Table 1; Figure 1, 2). The post-restoration recovery of the invertebrate community is slow. The biomass of populations under grass mixture (1-2 year-old) was smaller than those under traditional arable management, but the number of invertebrates sometimes was significant. This demonstrates the reproduction of young invertebrates' populations and the occurrence of the initial stage of successional changes.

*Table 1*

**Statistical parameters of invertebrates in the leached chernozem in conditions of the land management with perennial grasses**

| Index | Unit measure | $\bar{x}$ | $S^2$ | V, % | $S_{\bar{x}}$ | Confidence intervals, $p \le 0.05$ | n |
|---|---|---|---|---|---|---|---|
| Control (arable) | | | | | | | |
| Number of invertebrates | ex $m^{-2}$ | 55.0 | 966.18 | 56.5 | 8.98 | 35.2-74.8 | 12 |
| Number of *Lumbricidae* family | | 38.0 | 781.09 | 73.6 | 8.07 | 20.2-55.8 | 12 |
| Biomass of invertebrates | g $m^{-2}$ | 13.3 | 235.43 | 115.4 | 4.43 | 3.5-23.1 | 12 |
| Biomass of *Lumbricidae* family | | 8.3 | 197.34 | 169.3 | 4.06 | 0-17.2 | 12 |
| Ryegrass+lucerne (3-5 year-old) | | | | | | | |
| Number of invertebrates | ex $m^{-2}$ | 136.4 | 2227.38 | 34.6 | 14.92 | 102.7-170.1 | 10 |
| Number of *Lumbricidae* famil | | 112.4 | 1722.49 | 36.9 | 13.12 | 82.7-142.1 | 10 |
| Biomass of invertebrates | g $m^{-2}$ | 21.0 | 142.08 | 56.8 | 3.77 | 12.5-29.5 | 10 |
| Biomass of *Lumbricidae* family | | 16.8 | 126.33 | 66.9 | 3.55 | 8.8-24.8 | 10 |
| Ryegrass+sainfoin (1-2 year-old) | | | | | | | |
| Number of invertebrates | ex $m^{-2}$ | 80.0 | 3814.40 | 77.2 | 25.21 | 15.2-144.8 | 6 |
| Number of *Lumbricidae* family | | 64.0 | 3104.00 | 87.1 | 22.75 | 5.5-122.5 | 6 |
| Biomass of invertebrates, g $m^{-2}$ | g $m^{-2}$ | 4.2 | 7.41 | 64.8 | 1.11 | 1.4-7.1 | 6 |
| Biomass of *Lumbricidae* family | | 3.4 | 7.41 | 80.1 | 1.11 | 0.6-6.3 | 6 |

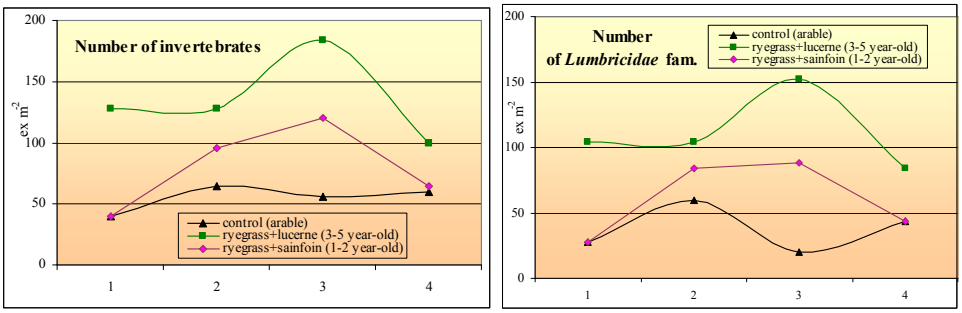


**Fig. 1** - Dynamics of invertebrates and *Lumbricidae* family in the leached chernozem by the perennial grasses' application:
1 – 12.05.2010; 2 – 25.05.2011; 3 – 13.09.2011; 4 –14.06.2012

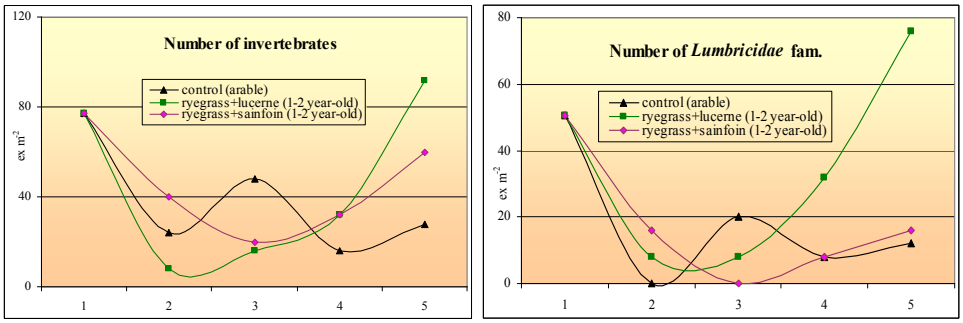


**Fig. 2 -** dynamics of invertebrates and *Lumbricidae* family in the ordinary chernozem by the perennial grasses' application:
1 – 23.04.2010; 2 – 24.06.2010; 3 – 17-18.05.2011; 4 – 20.09.2011; 5 – 24.04.2012

Statistically significant growth of zoological indicators has been registered after the third year of investigations on plots with grass mixtures of ryegrass and lucerne. The number of invertebrates on average was 2.5 times higher compared with the control plot, the total biomass – 1.6 times respectively. This method is especially effective for restoring the *Lumbricidae* family. Earthworm populations in the grass cultivation by ryegrass and lucerne during 3-5 years were significantly larger than those in the arable management, both in terms of earthworm abundance and biomass. Their number in the leached chernozem increased by 3.0 and biomass – by 2.0 times. The population growth over 5 years constitutes 74.4 ex $m^{-2}$ (744.000 ex $ha^{-1}$) or 14.9 ex $m^{-2}$ (149.000 ex $ha^{-1}$) annually.

The diversity of invertebrates was different depending on the agricultural management. The grass cultivated soil is characterized by a greater diversity of invertebrates. In addition to the *Lumbricidae* family, species of the *Formicidae, Gloremidae, Scarabaeidae, Elateridae, Geophilidae, Araneae, Coccinelidae* and *Carabidae* families were found. *Lumbricus terrestris* and *Allolobophora terrestris* species are the most typical representatives of the *Lumbricidae* family in the chernozem under perennial grasses. The abundant presence of the *Formicidae*

family representatives is observed. In general, the soil under grass mixture with ryegrass and lucerne contains 5-6 families of invertebrates, while the soil under arable only 2-4 families of edaphic fauna.

The use of phytorestoration procedures with legume-cereal grass mixtures creates advantageous conditions for the existence and functioning of autochthonous microorganisms and activates the internal reserves of microbiological systems of degraded soils. Microbial biomass in chernozems increases significantly from 216.2-275.8 to 262.7-388.3 μg C $g^{-1}$ soil according to the average data (Table 2). The maximum values have been registered in the leached chernozem with the application of the mixture of ryegrass and lucerne grass mixtures during 3-5 years.

*Table 2*

**Microbial biomass (**μg C $g^{-1}$ soil**) in the long-arable chernozems in conditions of the land management with perennial grasses**

| Variant | $\bar{x}$ | $S^2$ | V, % | $S_{\bar{x}}$ | Confidence intervals, P ≤ 0.05 | n |
|---|---|---|---|---|---|---|
| Leached chernozem | | | | | | |
| Control (arable) | 275.8 | 4793.92 | 25.1 | 19.99 | 213.8-319.8 | 12 |
| Ryegrass+Lucerne (3-5 year-old) | 388.3 | 12681.43 | 29.0 | 32.51 | 316.8-459.8 | 12 |
| Ryegrass+sainfoin (1-2 year-old) | 279.3 | 2601.88 | 18.3 | 17.00 | 237.0-318.6 | 9 |
| Ordinary chernozem | | | | | | |
| Control (arable) | 216.2 | 851.00 | 13.5 | 4.84 | 205.8-226.6 | 15 |
| Ryegrass+lucerne (1-2 year-old) | 262.7 | 3758.50 | 23.3 | 15.83 | 228.7-296.7 | 15 |
| Ryegrass+sainfoin (1-2 year-old) | 282.7 | 3801.01 | 21.8 | 15.92 | 248.5-316.9 | 15 |

*Table 3*

**DEHYDROGENASE ACTIVITY (**mg TPF $10g^{-1}$soil $24h^{-1}$**) IN THE LONG-ARABLE CHERNOZEMS IN CONDITIONS OF THE LAND MANAGEMENT WITH PERENNIAL GRASSES**

| Variant | $\bar{x}$ | $S^2$ | V, % | $S_{\bar{x}}$ | Confidence intervals, P ≤ 0.05 | n |
|---|---|---|---|---|---|---|
| Leached chernozem | | | | | | |
| Control (arable) | 1.13 | 0.07 | 23.1 | 0.29 | 0.49-1.77 | 12 |
| Ryegrass+lucerne (3-5 year-old) | 1.58 | 0.09 | 18.5 | 0.09 | 1.38-1.78 | 12 |
| Ryegrass+sainfoin (1-2 year-old) | 1.29 | 0.17 | 31.5 | 0.14 | 0.97-1.61 | 9 |
| Ordinary chernozem | | | | | | |
| Control (arable) | 1.49 | 0.20 | 29.7 | 0.26 | 0.93-2.05 | 15 |
| Ryegrass+Lucerne (1-2 year-old) | 2.03 | 0.27 | 25.6 | 0.13 | 1.75-2.31 | 15 |
| Ryegrass+sainfoin (1-2 year-old) | 1.94 | 0.22 | 24.4 | 0.12 | 1.68-2.20 | 15 |

The use of the mixture of perennial grasses had a stimulating effect regarding to the dehydrogenase in both soils (Table 3). Dehydrogenase activity

increased on average by 1.4 times in the conditions of the ryegrass and lucerne mixture application. The cultivation of ryegrass and sainfoin mixture led to the stimulation of the dehydrogenase activity in the leached chernozem by 14%, in the ordinary chernozem – by 30% respectively.

## CONCLUSIONS

1. The effective restoration of the biota in degraded arable chernozems occurs as a result of the cultivation of perennial grasses. Perennial grass mixtures can be used both in arable soils and in rows of perennial fruit crops to create oases and migration corridors for the soil biota.

2. The restoration of the lost components of the soil biota in the long-term arable chernozems is a long process. The reproduction of young populations of invertebrates in conditions of the use of perennial grasses during 1-2 years testifies the initial phase of the biota's succession.

3. The use of the mixture of ryegrass and lucerne during 3-5 years has resulted to the growth of the number of invertebrates and *Lumbricidae* family by 2.5 and 3.0, their biomass – by 1.6-2.0 times in comparison with the traditional arable management. Invertebrates' diversity has improved, the number of invertebrates' families increased from 2-4 to 5-6. The microbial biomass content and dehydrogenase activity in the 0-25 cm layer increase in average by 1.4 times. The microbial biomass accumulates in the soil in amounts of 132.5 kg $ha^{-1}$ annually. The annual population growth of earthworms reaches of 14.9 ex $m^{-2}$.

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