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Study on the influence of Lumbrical and Lumbrex bio-fertilizers over an artificial grassland of red fescue (*Festuca rubra* L.)

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Abstract

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In the period 2014-2016, a field experiment was conducted to establish the influence of the Lumbrical (150 and 200 ml/m^2) and Lumbrex (150 and 200 ml/da) on the yield, height and chemical composition of red fescue grassland. The preparations included in the experiment have a positive effect on the productivity of red fescue. During the study period, the soil treatment of the plants increased the yield of grass crops to a higher extent compared to the foliar. The amount of fresh and dry mass in the variant with Lumbrical 150 ml/m^2 exceeded significantly the non-treated control by 34.5 and 37.0% ($P < 0.01$). In the grasslands with Lumbrical 200 ml/m^2, the values were 24.9% (fresh mass) and 30.2% (dry mass) higher than the control ($P < 0.05$). The treatment with Lumbrical and Lumbrex increased the height of the grassland of *Festuca rubra* L. The influence of Lumbrical bio-fertilizer at a dose of 200 ml/m^2 increased the average stem length by 14.9% in comparison with the control. Crude protein content in grasslands with soil and leaf bio-fertilization exceeded the control variant by 11.9% (Lumbrex 150 ml/da) to 20.8% (Lumbrical 150 ml/m^2). Mineral, phosphorus and crude fat content in the variants with Lumbrical 200 ml/m^2 is higher with 14.7%, 16.7% and 17.3% respectively, in comparison with the non-treated control. The introduction of the granulated fraction (200 ml/m^2) reduced the fiber concentration of the grassland by 7.5%.

Keywords: Lumbricus rubellus; *Festuca rubra* L.; organic farming; bio-fertilization

Introduction

Perennial meadow grasses mainly form the turf of the grass cover. Their forage qualities are closely related to morphology, adaptive abilities and resistance to the cultivation environment. Most of them are absolutely autotrophic plants and react positively to the soil application of biological products containing easily soluble minerals, micro and macro elements. This positive reaction stimulates the growth and development of the root system in plants and facilitates the direct absorption of nutrients by plant individuals (Drevon and Hartwig, 1997; Atiyeh et al., 2000; Tang et al., 2001). The introduction of organic and environmentally friendly products is a priority of organic farming and is related to improving the long-term biological activity of the soil and its natural fertility. Yancheva and Manolov (2003) define organic production as the most popular modern form of sustainable agriculture. The priority of a number of scientific studies both abroad and in Bulgaria is to stimulate the production parameters (Popov, 2014) and to improve the biometric indicators (Popov and Yordanov, 2012) of crops to increase the yield and quality of the forage biomass (Todorov, 2005; Masheva and Mihov, 2008). Red fescue is a major structural component in the composition of meadows and pastures in the area of the Central Balkan Mountain. In the grassland, the species is often dominated by high share participation (88-98%), which favors the formation of relatively stable and long-lasting mixed grassland (Mitev and Goranova, 2008; Mitev et al., 2010).

The productivity and quality of red fescue treated with bio-fertilizers depend on their species, dose and application method, nutrient content of the soil as well as the climatic conditions of the area in which the crop is cultivated (Jančovič et al., 2004; Rotar et al., 2010; Samuil et al., 2011).

Lumbricus rubellus compost enriches and favors the course of biochemical and physical processes in the soil, creating better conditions for plant growth (Makulec, 2002; Arancon et al., 2004). The application of this type of biofertilization in forage production technology implies the improvement of the quality indicators and the nutritional value of the main forage species in the mountain areas.

Hence the topicality of the present study comes, whose main objective is to study the effect of Lumbrex and Lumbrical bio-fertilizers on the productivity, height of grassland and quality of red fescue forage in the Central Balkan Mountain.

Materials and Methods

The survey was conducted during the period 2014-2016 in the experimental field of the Department of 'Mountain Grass Associations and Maintenance of their Biological Diversity' at Research Institute of Mountain Stockbreeding and Agriculture (RIMSA), Troyan. Lumbrex and Lumbrical biofertilizers were tested for foliar and soil application on artificial grassland of red fescue (*Festuca rubra* L.), Ryder cultivar.

Lumbrical biological fertilizer is an environmentally friendly product resulting from the processing of organic waste by the red earthworm (*Lumbricus rubellis*) and applied modern biotechnology. The product is with high protein content (Markov, 2015) containing useful microorganisms, macro and micro elements in high concentrations. Organic bio-fertilizer Lunbrex contains humic and fulvic acids, macro and micro elements in certain concentrations. It is applied the generally accepted technology for cultivation of meadow grasses for forage.

Experimental variants are: 1. Control /nontreated/; 2. Lumbrical – 150 ml/m^2 (1 ml = 0.58 g); 3. Lumbrical – 200 ml/m^2 (1 ml = 0.58 g); 4. Lumbrex – 150 ml/da; 5. Lumbrex – 200 ml/da. The necessary soil cultivations were applied prior to sowing to create artificial grasslands. The sowing of the independent crop was carried out in the second half of March, manually scattered using the blocking method in 4 replicates, with a plot size of 5 m^2 and a sowing rate of 2.5 kg/da at 100% purity and germination rate. After sowing, the areas were rolled. The fertilizing process was carried out in the tasseling stage.

The following indicators were observed:

• Yield of fresh and dry mass (kg/da) – determined by mowing, weighing of grass in the different replications for each harvest plot followed by drying the samples to a constant weight at 105°C and recalculated for 1 da.

• Height (cm) – measured for different mowing at the harvest period of the grassland, plants were measured from the soil surface to the top of the highest stems along both diagonals of each parcel at 4 points, as the average values were calculated on the basis of that data.

• Basic chemical composition (g kg^{-1} DM) of grassland – dry and ground grass biomass was analyzed according to *Weende* analysis: crude protein (CP), crude fiber (CF), ether extract – crude fats (EE), mineral content (Ash), nitrogen-free extractable (NFE), calcium (Ca) and phosphorus (P).

For the statistical processing of yield data was used software product: Analysis Toolpak for Microsoft Excel 2010.

Results and Discussion

Productivity and height of red fescue grassland treated by bio-fertilizers

Essential for the accumulation of biomass is the type and rate of growth of forage grasses. Red fescue initially develops slowly, with its maximum development reaching the third, fourth year. It is too vulnerable to drought during the period of active vegetation. There is low intensity of growth and survival (Vateva and Stoeva, 2010).

The introduction of the studied biotor does not reliably affect the yield of fresh and dry mass in the first vegetation, which can be related to the above-mentioned biological features of the crop – Table 1. In the year of sowing, we slightly notice the values of the indicator in the variants with Lumbrical 150 and 200 ml/m^2 and Lumbrex 200 ml/da, but with an unreliable difference from the control.

In the second and third vegetation the soil intake of the granulated fraction of Lumbrical 150 ml/m^2 resulted in a significant ($P < 0.05$) effect for increasing the productivity of the grass. The yield in the control variant was exceeded by 36.7 and 42.0% (fresh mass) respectively and 32.4 and 54.6% (dry weight) respectively. On average, for the three-year experiment period, the fresh and dry mass of the variation exceeds the untreated control by 34.5 and 37.0% ($P < 0.01$), respectively.

In grasslands with a higher experimental dose (Lumbrical 200 ml/m^2), the yields of fresh and dry weight in the second and third vegetation also exceed the control, but with less and unproven differences. The average annual yields of this variant marked positive differences in fresh and dry mass yields compared to the 24.9 and 30.2% ($P < 0.05$) respectively.

Foliar treatment with Lumbrex at doses of 150 and 200 ml/da did not affect the yield of the red fescue.

Table 1. Yield of fresh and dry mass of *Festuca rubra* L. treated with Lumbrical and Lumbrex by years and average for the period 2014-2016 (kg/da)

Variant	2014		2015		2016		2014-2016	
	kg/da	% compared to control	kg/da	% compared to control	kg/da	% compared to control	kg/da	% compared to control
Fresh mass								
Control	1350.0	100.00	2212.5	100.0	3425.0	100.0	2329.2	100.0
Lumbrical 150 ml/m^2	1512.5	112.04	**3025.0***	**136.7***	**4862.5***	**142.0***	**3133.3****	134.5**
Lumbrical 200 ml/m^2	1425.0	105.56	2825.0	127.7	4475.0	130.7	**2908.3***	**124.9***
Lumbrex 150 ml/da	1225.0	90.74	2037.5	92.1	3762.5	109.9	2341.7	100.5
Lumbrex 200 ml/da	1400.0	103.70	2150.0	97.2	3637.5	106.2	2395.8	102.9
LSD 0.05	316.07	23.41	619.3	28.0	1344.0	39.2	517.1	22.2
LSDD 0.01	443.66	32.86	869.3	39.3	1886.5	55.1	725.8	31.2
LSD 0.001	626.34	46.40	1227.2	55.5	2663.4	77.8	1024.7	44.1
Dry mass								
Control	294.3	100.0	742.4	100.0	730.9	100.0	589.2	100.0
Lumbrical 150 ml/m^2	308.6	104.8	**983.3***	**132.4***	**1129.8***	154.6*	807.2**	137.0**
Lumbrical 200 ml/m^2	**362.0**	123.0	889.9	119.9	**1050.0***	**143.7***	**767.3***	**130.2***
Lumbrex 150 ml/da	280.9	95.4	744.6	100.3	929.4	127.2	651.7	110.6
Lumbrex 200 ml/da	299.5	101.8	771.6	103.9	844.6	115.6	638.5	108.4
LSD 0.05	68.6	23.3	195.4	26.4	316.5	43.3	137.4	23.3
LSD 0.01	96.3	32.8	274.2	37.0	444.2	60.8	192.9	32.8
LSD 0.001	135.9	46.3	387.2	52.2	627.1	85.8	272.3	46.3

The composition of the tested bioreactors stimulates the growth and development of plants (Atiyeh et al., 2000; Makulec, 2002; Arancon et al., 2004). The results of the conducted triennial studies reported a positive influence of the preparations Lumbrical and Lumbrex on the biometric height indicator of plants (Table 2).

On average, for the 2014-2016 period, the height of the stems in variants with increased doses of soil (Lumbrical 200 ml/m^2) and foliar (Lumbrex 200 ml/da) feed have higher values and the premise that lumbar culture has a positive effect on the dynamics the growth and development of the grass.

The effect of the action of the granulated fraction at a dose of 200 ml/m^2 increased the average length of the stems of *Festuca rubra* L. by 14.9% relative to the control. The growth of plants in the variant is characterized by a low degree of variation – VC 11.8% (almost identical to the control – 11.6%) and non-significant deviation (SD – 5.2 cm) compared to standard (SD – 4.4 cm). According to the indicators: variation coefficient and degree of variation, grasses with applied leaf treatment of the liquid fraction Lumbrex are considerably higher and more equalized by the height of the grass. Red fescue demonstrates high homogeneity (up to 10-12%) in biodiversity variants. The liquid fraction of Lumbrex acts both as a source of nutrients and a growth stimulant for plants, which, according to Nickel (1982), strengthens the metabolism, activates the absorption of nutrients and redistributes them in the organism. This is also the reason for the minimal difference in the average values for the foliar treatment of grasslands at doses of 150 ml/da (43.4 cm) and 200 ml/da (43.5 cm) in comparison with the maximum (44.0 cm) in the variant with Lumbrical 200 ml/m^2.

Basic chemical composition of grassland of red fescue, treated with bio-fertilizers Lumbrical and Lumbrex

The forage chemical composition gives a real idea of its nutritional value. The most prominent in this regard is the

Table 2. Height of grassland of *Festuca rubra* L., treated with fertilizers Lumbrical and Lumbrex, average for 2014-2016 (cm)

	Control	Lumbrical 150 ml/m^2	Lumbrical 200 ml/m^2	Lumbrex 150 ml/da	Lumbrex 200 ml/da
X	38.3	41.8	44.0	43.4	43.5
SD	4.4	3.0	5.2	4.0	2.9
VC	11.6	7.2	11.8	9.2	6.7
MIN	5.0	10.0	9.0	8.0	5.0
MAX	120.0	90.0	80.0	100.0	85.0

Table 3. Basic chemical composition of grassland of *Festuca rubra* L. treated with Lumbrical and Lumbrex, average for the period 2014-2016 (g kg^{-1} DM)

Variants	CP	CF	EE	Ash	NFE	Ca	P
Control	81.8	313.9	22.6	78.4	417.5	14.3	1.8
Lumbrical 150 ml/m^2	**98.8**	308.4	25.0	81.1	400.4	**16.3**	1.5
Lumbrical 200 ml/m^2	96.6	**292.0**	**26.5**	**89.9**	404.5	15.3	**2.1**
Lumbrex 150 ml/da	91.5	302.5	26.4	76.3	415.4	11.4	1.5
Lumbrex 200 ml/da	92.3	308.7	24.6	79.4	400.9	14.6	1.7
Average	92.2	304.2	25.2	81.4	409.4	14.3	1.7
SD	6.5	8.4	1.6	5.3	8.1	1.8	0.3

crude protein content in the dry matter composition. For the study period, the crude protein content of the soil and leaf application exceeded the control variant by 11.9% (Lumbrex 150 ml/da) to 20.8% (Lumbrical 150 ml/m^2) – Table 3. The values of the indicator in the variants with the imported granular fraction are higher than those with foliar application.

The degrading (active) function of humic and fulvoacids in the composition of the test preparations favors the release of nutrients in a form accessible to the plants. Fertilizing with the studied lumbrical product stimulates the development of red fescue and improves the quality of the harvested forage mass.

The results of the chemical analysis of the biomass indicate lower crude fibers (CF) content in the grassland with the variants with Lumbrical and Lumbrex compared to the non-treated control. For soil application of Lumbrical 200 ml/m^2, the fiber concentration in the dry matter decreased by 7.5% in comparison to the control and by 3.2% compared to the mean value of the indicator.

Nitrogen-free extractable substances (NFE), crude fats (EE), calcium and phosphorus are essential elements of the chemical composition of grass forage and are the factors determining its taste.

In the present study, the grassland with the higher dose (200 ml/m^2) of Lumbrical is also characterized by a higher crude fat content (26.5 g kg^{-1} DM), minerals (89.9 g kg^{-1} DM) and phosphorus (2.1 g kg^{-1} DM), and that of 150 ml/m^2 is richer than the macroelement calcium (16.3 g kg^{-1} DM) in comparison to the control.

There is an opposite trend towards the amount of nitrogen-free extractable substances in dry matter. Biological products, which were tested, did not have a positive effect on NFE synthesis. All variants of bio-fertilization have lower carbohydrate content than the control.

Conclusion

The products included in the experiment increased the productivity of forage biomass. The yield of fresh and dry mass in the variant with soil application of Lumbrical 150 ml/m^2 exceeded significantly the non-treated control by 34.5 and 37.0% ($P < 0.01$). In the grasslands with a higher experimental dose (Lumbrical 200 ml/m^2), the exceed of fresh and dry mass in comparison with the control was 24.9 and 30.2% ($P < 0.05$).

Red fescue has a greater degree of responsiveness to the soil fraction of the tested bio-fertilizers in terms of plant growth. The effect of Lumbrical at a dose of 200 ml/m^2 increased the average stem length by 14.9% in comparison to the control.

Crude protein content in grasslands with soil and foliar bio-fertilization exceeds the control by 11.9% (Lumbrex 150 ml/da) to 20.8% (Lumbrical 150 ml/m^2). The minerals, phosphorus and crude fat content in the 200 ml/m^2 in the variants with Lumbrical exceeded the non-treated control by 14.7%, 16.7% and 17.3%. The application of the granulated fraction (200 ml/m^2) reduced the fiber concentration of the grassland by 7.5%.

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