Scientific Papers. Series A. Agronomy, Vol. LXIV, No. 1, 2021
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

INFLUENCE OF YARA MINERAL FERTILIZER PRODUCTS ON GROCHEMICAL INDICATORS AND MICROBIOLOGICAL ACTIVITY IN SOILS AT COMMON WINTER WHEAT

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Abstract

Experiment was carried out on a test field in the village of Gurkovo, on an area of 9 ha, in three variants. Fertilization was performed with fertilizer products: Yara Mila Triple (16% N, 16% P_2O_5, 16% K_2O), Yara Vera Amidas (40% N, 14% SO_3) and Yara Bela Sulfan (24% N, 15% SO_3, 11 % CaO). A variety Avenue of common winter wheat was used. Macronutrients (digestible forms of N, P, K) in the soil during the tillering and maturity phenological phases of wheat shows quantitative dynamics, but between the second phase and the harvesting phase the values of macronutrients in all fertilization variants decrease, which is related to their good absorption. The used fertilizer products increase the biogenicity of soils, as established and regrouping of the separate groups of microorganisms in the composition of total microflora - reduces the amount of non-spore-forming bacteria at the expense of development of bacilli, actinomycetes and micromycetes. The increase of mineral nitrogen compounds in soil during the maturity phase to some extent inhibits the development of microorganisms, but not their mineralization activity.

***Key words**: mineral fertilization, microorganisms, macronutrients.*

INTRODUCTION

The usage of easily assimilable and quickly operating mineral fertilizers, in effective combinations and fertilizer norms, appropriate weather and duration of fertilization improve the soil fertility, the quality and yield of the agricultural production. The soil fertilization increases the reserves of nutrient substances in the upper soil layer, as well as the quantity of the organic carbon (Karcauskiene and Repsiene, 2009), which leads to changes in the quantity and activity of the soil microflora (Giacometti et al., 2013). The changes in the microbial communities may be used for prognostication the effects of the ecosystemic disturbances by organic and conventional management practices (Bending et al., 2000; Bruggen-Van and Semenov, 2000; Poudel et al., 2002), since the microbial community possesses potential for faster growth and exchange, because of which is a more reactive component of the soil ecosystem for external stress, than the plants and animals (Panikov, 1999). The soil microorganisms are sensitive to changes in the surrounding environment (Schinner and Sonnletner, 1996) and are indicators that the microbial population changes after the fertilization (Hyman et al., 1990). The bringing in of mineral fertilizers as a whole increases the total quantity of the microorganisms in the soil (Milanov and Yorova, 1980; Donov et al., 1992; Prescott et al., 1992; Hart and Stark, 1997; Forge and Simard, 2001; Meena et al., 2014), as main share in the content of the general microflora after fertilization occupy the non-spore forming bacteria and bacilli, and less presented are the actynomycetes and micromycetes (Bogdanov et al., 2015; Naskova et al., 2015; Plamenov et al., 2016; Malcheva et al., 2018a; Malcheva et al., 2019a). The accumulation of microbial biomass in the initial soil-forming process in recultivating objects, especially after fertilization, is due mainly to the active development of the non-spore forming bacteria (Stefanova and Petrov, 2019). The non-spore forming aerobic and anaerobic bacteria play basic role for decomposition of green wastes in the separate phases of their composting (Malcheva et al., 2018b; Kostadinova and Dyakov, 2019), as well as after application of compost and biochar in the soil (Malcheva et al., 2019b; Malcheva et al., 2020). Other authors determine that the increase of the total microflora after fertilization is within short-term plan and is frequently followed by decrease of the microbial biomass and activity (Ohtonen, 1992; Smolander et al., 1994; Périé and Munson, 2000).

Yara Mila Triple is a balanced NPK 16/16/16 formula, appropriate for all crops, which are not sensitive to chlorine. The Yara Mila Triple granules are elaborated in such way, so there are not losses of nutrient substances flying off with storage, transportation and fertilizer bringing in. Yara Vera Amidas is based on covering of hot granulated carbamide with molten sulphur. It contains 40% total nitrogen, out of which 35% is in amidic form, which has to transform into an assimilable form for the plants - from ammonium to nitric, and 14% sulphur. The granulated form of Yara Vera Amidas allows even distribution on the soil surface and thus the uneven assimilation by the crops is avoided. Yara Bela Sulfan is a complex nitrogen fertilizer for simultaneous application of three important nutrient elements - nitrogen, sulphur and calcium. The ammonium nitrate is combined with calcium sulphate and thus is reached to the content of 24% nitrogen (out of which 12% nitrate and 12% ammonium), 15% sulphur and 11% calcium. The granules are with size, which allows thick and even covering of the area. The nutrient substances in Yara Bela Sulfan are completely soluble and provide the optimum dynamics of the absorption.

Efficiency improvement of the nitrogen fertilization is a prime issue because of the low efficiency of nitrogen. The sharing in portions of the fertilization with nitrogen has for purpose to provide the wheat quantitatively with this macroelement according to its necessities in each phase of development, to synchronize the availability of soluble nitrogen in the soil and the plant necessities (Sticksel et al., 2000; Golba et al., 2013), and also to increase the nitrogen usage efficiency. This production experiment is used on great number of farms notwithstanding the technique and purpose of production. During the recent years the purpose is to be changed the physical properties of conventional water-soluble phosphorous fertilizers, to be decreased the fixation of phosphate anions in the soil and to be increased the efficiency of the fertilizer phosphorus with assimilation by the plants.

The purpose of the study is to be determined the impact of Yara mineral fertilizer products on agrochemical indicators and microbiological activity in soils with common winter wheat.

MATERIALS AND METHODS

The experiment is carried out at an experimental field in the territory of village of Gurkovo, on area of 90 decare, in three variants. The fertilization is carried out with the fertilizer products: Yara Mila Triple (16% N, 16% P_2O_5, 16% K_2O), Yara Vera Amidas (40% N, 14% SO_3) and Yara Bela Sulfan (24% N, 15% SO_3, 11% CaO). The used variety is common winter wheat Avenue (selection of company Limagrain). The sowing is carried out on 11 October 2018 with sowing norm 600 germinating seeds/m^2.

Within the frameworks of the experimental area is carried out pro-sowing bringing in of Yara Mila Triple (16% N, 16% P_2O_5, 16% K_2O) - 25 kg/decare (4 kg/decare N active substance, a.s.). The distribution of the experimental variants, each of which with area of 30 decare is as follows:

Variant 1 (Field 1):

First fertilization 09 March 2019: Yara Vera Amidas (40% N) - 20 kg/decare (8 kg/decare N a.s.);

Second fertilization 18 April 2019: Yara Bela Sulfan (24% N) - 25 kg/decare (6 kg/decare N a.s.).

Total active substance nitrogen for the whole period of vegetation: 18 kg/decare N.

Variant 2 (Field 2):

First fertilization 09 March 2019: Yara Vera Amidas - 15 kg/decare (6 kg/decare N a.s.);

Second fertilization 18 April 2019: Yara Bela Sulfan - 25 kg/decare (6 kg/decare N a.s.).

Total active substance nitrogen for the whole period of vegetation: 16 kg/decare N.

Variant 3 (Field 3):

First fertilization 09 March 2019: Yara Vera Amidas - 14 kg/decare (5.6 kg/decare N a.s.);

Second fertilization 18 April 2019: Yara Bela Sulfan - 20 kg/decare (4.8 kg/decare N a.s.).

Total active substance nitrogen for the whole period of vegetation: 14.4 kg/decare N.

Before setting of the experiment, during the vegetation and after harvesting of the crop are taken soil samples for each of the variants for determining the content of macronutrient elements in the soil - content of ammonium and nitrate nitrogen, mobile phosphates and assimilable potassium.
The content of ammonium nitrogen (NH_4-N) is determined photometrically with indophenol blue as a result of extraction with calcium dichloride ($CaCl_2$) solution. The nitrate nitrogen (NO_3-N) is determined photometrically with Nitrospectral as a result of extraction with calcium dichloride ($CaCl_2$) solution.
The content of phosphorus and potassium is determined by a double lactate method of Egner-Rheem. The method is based on extraction of the mobile compounds of phosphorus and potassium by a solution of calcium lactate $(CH_3CH.OH.COO)_2Ca$, which is buffered with hydrochloric acid to pH 3.5-3.7, with proportion soil-solvent 1:50 and time of interaction 90 min.
The values of the soil reaction are measured potentiometrically by a pH-meter in compliance with the requirements of the methods for measuring soil pH as per the international standard ISO 10390.
The microbiological activity of the soil includes determining of non-spore forming bacteria, actinomycetes, micromycetes, bacillary microflora, bacteria, which are assimilating mineral nitrogen. They are determined by the method of dilution and culture on solid nutrient media (МПА and САА), cultivation in thermostats and next reporting of colony forming units (CFU), recalculated per 1g absolutely dry soil. The statistical analysis includes calculation of average value out of three repetitions, standard diversion and variation coefficient (C.V.).

RESULTS AND DISCUSSIONS

The results of the carried out agrochemical analyses of soil samples with wheat are presented in Table 1.
The obtained data of the carried out agrochemical analyses before setting the experiment with common winter wheat prove poor reserve of the soil with macroelements and slightly acid reaction of the soil. The values are close in the three fields, which is important for the scientific reliability of the obtained results.

Table 1. Content of macroelements in the soil in different phases of development with wheat

№	Crop	Variant	pH	Mineral N		Assimilable P and K	
				NO_3-N	NH_4-N	P_2O_5	K_2O
Pre-sowing							
1	Wheat	Field 1	5.07	10.2	7.05	8.65	28
2		Field 2	4.9	10.3	6.98	9.03	28.5
3		Field 3	5.01	11.2	7.11	8.72	28.1
Tillering phase							
1	Wheat	Field 1	4.94	15.6	7.1	10.7	30.9
2		Field 2	5.2	10.9	6.48	14.9	34.4
3		Field 3	5.04	15.4	16.7	8.92	31.6
Maturity phase							
1	Wheat	Field 1	5.24	10.3	29.2	8.7	29
2		Field 2	5.01	14.9	31.3	12.5	28.5
3		Field 3	5.07	32.2	45.1	6.18	28.3
Harvesting phase							
1	Wheat	Field 1	5.21	8.9	25.4	5.2	26.5
2		Field 2	5.07	11.2	25.6	10.1	25.4
3		Field 3	5.1	28.3	37.3	4.3	26.2

It is determined by the soil analysis that the content of ammonium nitrogen, in all studied phases with the wheat is with relatively close values (4.94-5.24 mg/kg). In phase tillerering, field 2 distinguishes with a slightly higher value than the other two fertilizer variants. Field 1 has superiority in the reserve with ammonium nitrogen during the next two phases. The highest values of nitrate nitrogen in phase tillerering is determined in field 1, and in phase ear formation and after harvesting at field 3, where field 1 is with the lowest results. The comparatively close values of the nitrate nitrogen in phase tillering in the three fields (10.9-15.6 mg/kg) must be reported and its categorical increase in the next two phases in field 3 (around three times more in comparison with the other fertilizer variants). The obtained results in all phases of development and after harvesting are categorical regarding the values of assimilable phosphorus and potassium. The quantity of the studied macroelements is with the highest values at field 2. An exception is the results of the agrochemical analyses after harvesting, where the highest reserve of assimilable phosphorus is field 1.

The not big difference in the values of the studied soil macroelements pro-sowing and after harvesting, as well as their dynamics among the separate wheat phases of development, show the exceptionally good assimilability of the brought in fertilizer products.

On the grounds of the obtained results we may affirm that the quantity of the macroelements in the soil during the first two phases have their quantitative dynamics, but between the second phase and phase harvesting the values of all macroelements in all fertilization variants decrease, which is connected with their good assimilation.

The results of the microbiological analysis are presented in Table 2. The study is in seasonal dynamics, during the three phenological phases of the wheat development - tillering, ear formation, harvesting. An analysis of a control sample is carried out in the autumn before setting of the experiment.

Table 2. Quantity and qualitative composition of the soil microflora (x 10^3 cfu/abs. dry soil)

Crop	Phase	Variant	Total microflora	Non-spore-forming bacteria	Bacilli	Actinomycetes	Micromycetes	Bacteria absorb mineral nitrogen
Wheat	Control	Field 1	2916	2142	396	234	144	3024
		Field 2	2520	1836	378	198	108	2700
		Field 3	2286	1584	360	216	126	2718
	Tillering	Field 1	6300	3192	588	2352	168	2604
		Field 2	5756.4	2902.8	656	2000.8	196.8	2328.8
		Field 3	7155	2130	930	1695	2400	1800
	Maturity	Field 1	3547.8	2284.2	712.8	437.4	113.4	1539
		Field 2	2608	1696	368	352	192	1936
		Field 3	2316.6	1620	372.8	210.6	113.4	1296
	Harvesting	Field 1	3998.4	2570.4	621.6	672	134.4	1747.2
		Field 2	3132.4	1968	442.8	524.8	196.8	2050
		Field 3	2851.2	1814.4	486	307.8	243	1490.4

Fertilization increases the biogenicity of soils in comparison with the studied controls (unfertilized samples) in different degree during the separate phases of the wheat development. The total quantity of microorganisms is the highest with the fertilization during phase tillering - around 2 times with variants 1 and 2, around 3 times with variant 3. In the same order decreases the total microflora during phase ear formation, but the quantity of the microorganisms remains higher in comparison with the controls. A slight increase of the total quantity of microorganisms is observed during phase harvesting in comparison with phase ear formation. Consequently regarding the seasonal dynamics the development of the microorganisms is higher during the spring in comparison with the summer. It is drawn attention as a tendency that with the fertilization, applied in variant 1, the total quantity of microorganisms is preserved higher during the three phases, with variant 2 intermediate in comparison with the other two variants, and with variant 3 - the highest during phase tillering and the lowest with the other phases. Generally this tendency correlates with the quantity of the nitrogen and sulphate compounds brought in with the fertilizers - the highest quantity with variant 1, follows variant 2 and the lowest with variant 3. The chemical analysis shows that the content of assimilable nitrogen compounds is the highest with variant 3, follow variant 2 and variant 1. Consequently the assimilation of the nitrogen compounds by the wheat plants follows the ascending order: variant 1 > variant 2 > variant 3, which presents and the development of the microorganisms as a whole. Thanks to the ammonification the soil is enriched additionally with ammonium and ammonium salts available for the plants. Respectively the yields of the fields are relatively in the same ascending order. The development of the microorganisms also decreases by depletion of the reserve of assimilable phosphorus and potassium - it is higher with phase tillering, and lower with the other two phases.

Highest percent in the content of the total microflora with the fertilized variants occupy the non-spore forming bacteria - 50-65%, with exception of variant 3, phase tillerering - 30%. The share of the non-spore forming bacteria is 69-74% with the control samples, and next in decreasing order are the bacilli, actynomycetes and micromycetes. While with the fertilized variants is observed regrouping of the micro-organisms - the actynomycetes occupy higher percent share than the bacilli during phases tillerering and harvesting. The micromycetes (the moulded fungi) also increase their quantity with fertilization, but their percent share remains the lowest in the content of the total microflora, with exception of variant 3, phase tillerering, where their quantity is the highest of all studied groups of microorganisms. The highest moisture of the soil is determined with this variant, which is a factor contributing for the better development of the moulded fungi. This regrouping of the microorganisms with phases tillerering and harvesting has impact on the decomposition of the organic substances in the soils. On the one hand the higher quantity of the non-spore forming bacteria and baccilli (ammonifiers) increases the initial stages of the organic matter destruction. Bacilli are also spore-forming species, which increases their resistance to changes (sometimes unfavourable) conditions of the environment. On the other hand the increase of the actinomycetes and micromycetes quantity shows that they also include actively in the decomposition of the organic matter, as with the assimilation of plainer substrata (carbohydrates, proteins, lipids, starch), as well as with the accumulation of more complex and difficult for assimilation substrata - mainly cellulose, hemicellulose, polysaccharides, when the speed of decomposition is also slowed down (final states of deconstruction). Some strains non-spore forming bacteria are less resistible to the direct impact of the nutrient substances, brought in with the fertilizers under the form of nitrogen and sulphur compounds, even more so they do not form spores surviving extreme conditions, “they stress” and die or they decrease their activity - not always higher quantity means higher activity. The reproduction and activity of the microorganisms are influenced by variety of factors - moisture, temperature, pH, reserve of nutrient substances, soil pollution, vegetation and etc. It is determined that the increase of the mineral nitrogen compounds in the soil during phase ear formation suppresses the development of the microorganisms, but not their activity - with the lowest total quantity of microorganisms and bacteria, assimilating mineral nitrogen with variant 2 and 3 during phase ear formation are determined the highest values of the mineralization coefficient in comparison with the same variants with the other phases. The quantity of bacteria, assimilating mineral nitrogen, however, is higher with the controls, than with the fertilized samples, which means that in some degree the speed of decomposition is impeded after fertilization - the values of the mineralization coefficient with the samples are higher (Figure 1).

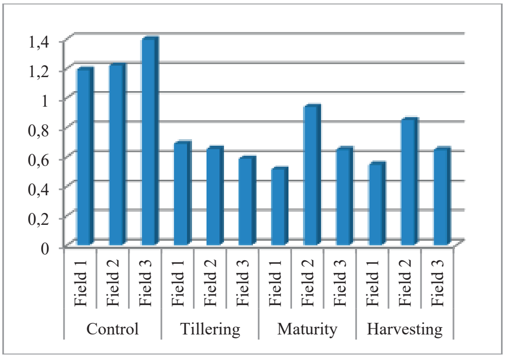

Figure 1. Mineralization coefficients

The statistical analysis showed that the extract is homogeneous - low values of standard deviation and coefficient of variation under 10% with all groups of microorganisms, for all variants and phases (Table 3).

The usage of Yara mineral fertilizers leads to increase of the content of macroelements in the soil in comparison with the control sample before setting of the experiment. The trend of increase continues until phase ear formation as in phase after harvesting is observed decrease of the reserve, which is an adequate process on basis the botanical characteristics of the wheat and the phases of vegetation. The best results are obtained with the variant fertilization of field 1 regarding ammonium nitrogen, field 3 - nitrate nitrogen, assimilable phosphorus and potassium - field 2.

On the grounds of the obtained results may be affirmed that the fertilizer variants suggested for testing have exceptionally favourable impact on the soil reaction and the content of macroelements in the soil. Their quantities after phase harvesting are sufficient to provide good initial development of a next crop on the same area in the plan of crop rotation.

Table 3. Statistical indicators: Standard deviation (STDEV), coefficient of variation (C.V., %)

Crop	Phase	Variant	Total microflora		Non-spore-forming bacteria		Bacilli		Actinomycetes		Micromycetes	
Statistical indicators			STDEV	C.V.	STDEV	C.V.	STDEV	C.V.	STDEV	C.V.	STDEV	C.V.
Wheat	Control	Field 1	4.546	3.820	0.816	3.711	0.471	3.722	0.000	0.000	5.888	3.505
		Field 2	3.559	3.489	0.000	0.000	0.816	7.423	0.471	7.443	4.320	2.880
		Field 3	6.532	7.423	1.700	8.359	0.000	0.000	0.471	6.428	5.099	3.377
	Tillering	Field 1	4.320	2.274	0.816	2.333	3.742	2.673	0.816	8.165	4.082	2.634
		Field 2	2.449	1.384	0.816	2.041	3.266	2.677	0.943	8.081	7.483	5.345
		Field 3	3.266	2.300	1.414	2.281	2.944	2.605	4.899	3.062	2.449	2.041
	Maturity	Field 1	3.742	2.654	1.247	3.530	1.247	3.024	0.471	4.041	4.967	5.228
		Field 2	3.266	3.081	0.816	3.550	0.816	3.711	0.943	8.081	6.164	5.095
		Field 3	3.266	3.266	1.414	6.149	0.943	7.071	0.000	0.000	1.633	2.041
	Harvesting	Field 1	2.160	1.412	0.471	1.263	1.633	4.082	0.471	5.657	7.071	4.562
		Field 2	1.414	1.179	0.816	2.722	0.471	1.458	0.816	6.804	4.320	3.086
		Field 3	1.633	1.458	1.247	4.204	0.816	4.297	1.247	8.134	2.828	3.074

The usage of the fertilizer products increases the biogenicity of the soils in comparison with the studied controls (unfertilized samples) in different degree during the separate phases of the wheat development. The total quantity of microorganisms is the highest with the fertilization during phase tillering - around 2 times with variants 1 and 2, around 3 times with variant 3. The biogenicity decreases during phase ear formation and harvesting, as the total microflora is the highest with variant 1, variant 2 follows, and variant 3 is with the lowest quantity. As a tendency is drawn attention that with the fertilization with variant 1 the total quantity microorganisms is preserved higher during the three phases, with variant 2 intermediate in comparison with the other two variants, and with variant 3 - the highest during phase tillering and the lowest with the other phases.

The highest percent share in the content of the total microflora with the fertilized variants occupy the non-spore forming bacteria - 50-65%, with exception of variant 3, phase tillerering - 30%. Regrouping of the rest groups of microorganisms is determined in comparison with the controls - the percent participation of the actinomycetes is increased significantly, which take part in the final stages of the organic matter destruction. They are in higher quantity than the bacilli with phase tillering for all variants and phase harvesting for variants 1 and 2. The micromycetes are less presented in the content of the total microflora with exception with variant 3, phase tillering, where their quantity is the highest. The quantity of bacteria, assimilating mineral nitrogen is higher with the controls, than with the fertilized samples, which means that in certain degree the speed of decomposition is impeded after fertilization - the values of the mineralization coefficient with the controls are higher.

Similar results for distribution of the separate groups of microorganisms in the total microflora content, after mineral and organic fertilization and in the phases of composting, are determined also in other own and somebody else's researches (Naskova et al., 2015;

Plamenov et al., 2016; Malcheva et al., 2018a; 2018b; Malcheva et al., 2019a; 2019b; Stefanova and Petrov, 2019; Kostadinova and Dyakov, 2019; Malcheva et al., 2020).

The researches regarding the effects of the nitrogen fertilization on the microbial biomass remain contradictory. For example, Zhang et al. (2005) have observed significant increase of the microbial biomass up to two years with usage of nitrogen fertilizers at pastures in China, but Sarathchandra et al. (2001) announce for significant decrease of the microbial biomass at perennial pasture of New Zealand because of a two-year nitrogen fertilization. Meanwhile, Johnson et al. (2005) have determined that two year of application of N has not influenced the microbial biomass in soils in Scotland. These tendencies depend on the differences regarding the soil moisture, content of organic substances in the soil, total content of N, pH, duration of bringing in of N (Williams et al., 2007), but the specific basic factors are still not completely identified (Arnebrant, 1990; Zhang et al., 1998). The degree of the fertilization effect on the microbial biomass depends on pH according to researches of Geisseler and Scow (2014) - the fertilization decreases the microbial biomass with soils with pH under 5, but has a significantly positive effect with higher values of pH of the soil.

CONCLUSIONS

The usage of Yara mineral fertilizers leads to increase in the content of macroelements in the soil in comparison with the control sample before setting of the experiment. The trend of increase continues until phase ear formation as in phase after harvesting is observed decrease of the reserve, which is an adequate process on base the botanical characteristics of the wheat and the phases of vegetation. The best results are obtained with the variant fertilization of field 1 regarding ammonium nitrogen, field 3 - nitrate nitrogen, assimilable phosphorus and potassium - field 2. The quantities of the macroelements after phase harvesting are enough to provide good initial development of a next crop on the same area in the plan of crop rotation.

The usage of the fertilizer products increases the biogenicity of the soils in comparison with the studied controls (unfertilized samples) in different degree during the separate phases of the wheat development. The total quantity of microorganisms is the highest with the fertilization during phase tillering - around 2 times with variants 1 and 2, around 3 times with variant 3. The biogenicity decreases during phase ear formation and harvesting, as the total microflora is the highest with variant 1, follows variant 2, and variant 3 is with the lowest quantity. As a tendency is drawn attention that with the fertilization with variant 1 the total quantity of microorganisms is preserved higher during the three phases, with variant 2 intermediate in comparison with the other two variants, and with variant 3 - the highest during phase tillering and the lowest with the other two phases.

The highest percent share in the content of the total microflora with the fertilized variants occupy the non-spore forming bacteria - 50-65%, with exception of variant 3, phase tillering - 30%. Regrouping with the rest groups microorganisms is determined in comparison with the controls - the percent participation of the actinomycetes is increased significantly, which participate in the final stages of the organic matter destruction. They have higher quantity than the bacilli with phase tillering for all variants and phase harvesting for variant 1 and 2. The micromycetes are the less presented in the content of the total microflora, with exception of variant 3, phase tillering, where their quantity is the highest. The quantity of the bacteria, assimilating mineral nitrogen is higher with the controls, than with the fertilized samples, which means that in some degree the speed of decomposition is impeded after fertilization - the values of the mineralization coefficient with the controls are higher.

The soil microbiological activity depends on and is in correlation with complex of factors - moisture, temperature, pH, mechanical composition, nutrient reserve, choice of a fertilizer and variants of fertilization, type of vegetation, processing of the soil and etc. Since they are sensitive indicators, their study in dynamics with fertilization with different types of fertilizers in appropriate combinations and norms, with different vegetation is necessary with purpose evaluation of the fertilization impact on the microbiological and enzyme activity of the soil, and from there on the soil fertility.

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