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Microbiological Evaluation of Meadow Chernozem Carbonate Soil under Various Fertiliser Systems

Abstract. Microorganisms play an important role in shaping soil fertility, maintaining its performance as a bio-inert matter, and are an indicator of qualitative changes in the soil. As a result of anthropogenic impact on the soil, the number and species composition of the microbiota is decreasing. The purpose of the study was to assess the number of different physiological groups of microorganisms on meadow chernozem carbonate soil under different pea fertilisation systems. The number of different groups of soil microorganisms was estimated according to the method

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of D.H. Zvyagintsev by sowing soil suspension on solid culture media. It was found that the most favourable indicators of micro biocenosis and the highest yield of peas of the Tsarevych variety were formed by the residual effect of organic fertilisers and $N_{45}P_{45}K_{45}$. The variant without fertilisers induces the development of organic matter mineralisation processes in meadow chernozem carbonate soil by the content of pedotrophic and humate-decomposing organisms and formed the lowest pea yield. The introduction of only mineral fertilizers for pea cultivation formed a high rate of humus accumulation at the level of $K_{ac.}$ =1,6-1,9

Keywords: microbiological activity of the soil, meadow chernozem carbonate soil, microorganisms, peas, fertiliser systems, organic matter

RELEVANCE

The biological activity of the soil is determined by soil microorganisms, which are an important component of the biological cycle of substances. The study of the soil microbiome makes it possible to understand and identify patterns of organic matter transformation processes, fertility, ecological and phytosanitary conditions (Tonkha *et al.*, 2017). Important factors influencing these processes and plant productivity are the use of various fertiliser systems, treatments, land reclamation agents, etc. (Tonkha *et al.*, 2019).

ANALYSIS OF RECENT RESEARCH AND PUBLICATIONS

The microbiological state is an indicator of changes in soils and an indicator of their ability to regenerate and rehabilitate themselves. Research by S.Yu. Bulyhin and O.L. Tonkha (2018) revealed that the basis for improving highly efficient farming systems and management of microbial processes in chernozems under different anthropogenic uses is a comprehensive assessment of the microbial microbiota, studying the biodiversity and spatial and functional structure of these organisms.

Long-term soil fertilisation significantly increased soil microbial biomass and dehydrogenase activity. Organic fertilisers had a greater effect on biomass and activity compared to mineral fertilisers (Nannipieri *et al.*, 2003). Soil microbiological activity is closely related to soil organic matter (Demianiuk, 2018). Wheat straw left on the soil increased nitrogen content and resulted in an upward trend in organic carbon content (Patyka *et al.*, 2014).

In the studies by Campbell *et al.* (1991), the use of green manure and by-products increased the number of azotobacter, reduced the specific content of melanin-synthesising micromycetes by 12.4, and reduced soil phytotoxicity by 16.8% compared to the variant without organic matter. They also point out that optimisation of plant mineral nutrition leads to a slowdown in the mineralisation of humus, total organic matter of the soil and nitrogen compounds.

The positive impact of organic fertilization systems has been documented in works by (Chu *et al.*, 2007; Malynovska *et al.*, 2014; Tonkha *et al.*, 2017; Demydenko *et al.*, 2017). At the same time, when by-products are removed from the field, the balance of organic matter becomes deficient and amounts to -0.19 t/ha, and the deficit of nutrients increases by 125% (Pylypenko *et al.*, 2016). Among the indicators that assess the biological activity of soil, the most complete should be considered the total number of microorganisms in the soil environment, although it can only assess the potential activity, which will be high under favourable environmental conditions and low under unfavourable conditions (Yeshchenko *et al.*, 2011).

Aside from tillage, crop rotation, predecessors, tillage, fertiliser application, etc. have a significant impact on the biological activity of the soil. As the intensity of biochemical processes increases, crop productivity rises, organic matter accumulates in the soil, and its physical and chemical properties and fertility improve (Tonha *et al.*, 2017; Malynovska *et al.*, 2014).

The study of soil biological activity is important in researching the processes of organic matter transformation and evaluating pea fertilisation systems. Each element of mineral nutrition has a specific value (Ushkarenko, 2008). Studies have shown that the yield of peas (*Pisum sativum* L.) does not depend on the levels of application of nitrogen, phosphorus, and potash fertilisers, but only on the on the amount of water in the soil. The minimum values of nutrients in soils – 73 mg/kg of mineral nitrogen, 10 phosphorus (by Olsen) and 60 mg/kg of exchangeable potassium – did not affect pea yields. The authors concluded that the current high fertiliser application rates for peas need to be reconsidered, as they may be unnecessary and unprofitable (Kakar *et al.*, 2002; Amjad *et al.*., 2004).

Peas have the ability to symbiosis with nodule bacteria and maintain a positive nitrogen balance in agriculture. Currently, the question of the impact of different fertiliser rates on the number of microorganisms involved in the transformation of nitrogen and carbon remains unresolved.

The purpose of this study was to assess the number of different physiological groups of microorganisms on meadow chernozem carbonate soil under different fertilization systems.

MATERIALS AND METHODS

The research was conducted within the stationary experiment of the Agronomic Research Station of the National University of Life Sciences of Ukraine in a six-crop rotation with the following crops: peas – winter wheat – corn – soybeans – spring barley – corn for grain. The soil of the experimental site is meadow chernozem carbonate low-humus coarse-pulverulent-medium loamy on loess-like loam. The total humus content in the soil of the experimental plots was 4.09-4.50%. The experiment studied fertilisation variants: 1) no fertilisers (control); 2) residual effect of manure +$N_{15}P_{15}K_{15}$; 3) residual effect of manure +$N_{30}P_{30}K_{30}$; 4) residual effect of manure +$N_{45}P_{45}K_{45}$; 5) $N_{30}P_{30}K_{30}$. The pea variety is Tsarevych. The soil cultivation system is surface tillage. Soil sampling was performed in the 0-20 and 20-40 cm soil layers in May.

The selection and preparation of soil samples for the study of aerobic microbiota in the laboratory was carried out in accordance with DSTU ISO 10381-6-2001. The number of different groups of soil microorganisms was estimated according to the method of D.H. Zvyagintsev by sowing soil suspension on solid culture media. The total number of microorganisms that decompose organic compounds containing nitrogen was studied on meat-peptone agar (MPA). Microorganisms that assimilate mineral forms of nitrogen were studied on a starch-ammonia medium (SAM). The number of microorganisms that synthesize melanins on Chapek's medium at pH = 5.0, decompose humates on sodium humate medium, pedotrophs on soil agar. Statistical data processing was performed using the Statistica software package.

RESULTS AND DISCUSSION

Different intensity of soil use has led to changes in the number of microorganisms engaged in the nitrogen cycle. In the meadow chernozem carbonate soil, the number of ammonifiers depended on the fertiliser system and soil layer (Table 1).

Table 1. Number of ammonifying and amylolytic microorganisms in meadow chernozem carbonate soil under different fertilisation methods, million. CFU/g of soil

Fertiliser variant	Soil layer, cm	Ammonifying	Amylolytic
No fertilisers (control)	0-20	5.75 ± 1.26	0.36 ± 0.06
	20-40	26.11 ± 2.48	0.50 ± 0.02
$N_{30}P_{30}K_{30}$	0-20	12.57 ± 0.10	0.41 ± 0.02
	20-40	3.01 ± 0.10	0.51 ± 0.02
Organic residual effect+ $N_{30}P_{30}K_{30}$	0-20	7.00 ± 0.79	0.43 ± 0.00
	20-40	20.52 ± 1.00	1.03 ± 1.41
Organic residual effect+ $N_{45}P_{45}K_{45}$	0-20	8.45 ± 0.45	0.45 ± 0.12
	20-40	29.58 ± 2.01	0.67 ± 0.01
Organic residual effect+ $N_{15}P_{15}K_{15}$	0-20	27.66 ± 1.03	0.91 ± 0.02
	20-40	30.65 ± 4.60	0.98 ± 0.09

A larger number of microorganisms that decompose organic and synthesize mineral forms of nitrogen was obtained in a layer of 20-40 cm, with the exception of variant $N_{30}P_{30}K_{30}$. The difference compared to the 0-20 cm layer was 10-80%. The highest number of ammonifying microorganisms in the layer of 20-40 cm was observed with the application of $N_{15}P_{15}K_{15}$ in combination with the residual effect of organic matter, and the lowest with the application of only mineral fertiliser system $N_{30}P_{30}K_{30}$. This leads us to the conclusion that mineral fertilisers alone have a negative impact on soil organic matter.

In terms of the number of amylolytic microorganisms, the difference between some variants of the systems did not exceed 5%. Evaluating the degree of enrichment by the number of ammonifiers according to the method of D.G. Zvyagintsev, it is necessary to note that all fertiliser variants were characterized as poor.

According to H.O. Iutynska, 2006, pedotrophic microorganisms are involved in the decomposition of peripheral chains of humus molecules, and deep destruction is carried out by humate-decomposing microorganisms (Table 2).

Table 2. The number of pedotrophic, humate-decomposing microorganisms and micromycetes in meadow chernozem carbonate soil under different fertilisation variants, million CFU*/g soil

Fertiliser variant	Soil layer, cm	Pedotrophic	Humate-decomposing	Micromycetes, thousand CFU/g of soil.
No fertilisers (control)	0-20	11.22 ± 0.83	4.45 ± 0.61	6.19 ± 0.01
	20-40	17.73 ± 2.93	18.33 ± 0.89	10.32 ± 1.49
$N_{30}P_{30}K_{30}$	0-20	10.35 ± 1.15	3.99 ± 0.10	22.96 ± 0.99
	20-40	4.77 ± 1.13	2.04 ± 0.59	13.23 ± 0.00
Organic residual effect+ $N_{30}P_{30}K_{30}$	0-20	8.56±0.07	1.63 ± 0.05	9.05 ± 0.49
	20-40	14.65 ± 0.51	6.59 ± 0.10	18.93 ± 2.49

Table 2, Continued

Fertiliser variant	Soil layer, cm	Pedotrophic	Humate-decomposing	Micromycetes, thousand CFU/g of soil.
Organic residual effect+ $N_{45}P_{45}K_{45}$	0-20	6.80±0.21	5.53±0.15	35.85±0.50
	20-40	23.79±0.01	14.43±2.16	47.46±5.54
Organic residual effect+ $N_{15}P_{15}K_{15}$	0-20	18.74±1.15	8.99±1.28	69.79±1.54
	20-40	16.93±0.55	13.20±0.87	38.15±2.55

Note: * – thousand. CFU/g of soil

The highest number of the above microorganisms was observed under the residual effect of organic matter with full mineral fertilization in the norms of $N_{45}P_{45}K_{45}$ and $N_{15}P_{15}K_{15}$, and also in the control. Thus, when organic fertilisers are applied, organic matter is restored, and without fertilisers, humus decomposes. The application of $N_{30}P_{30}K_{30}$ mineral fertiliser alone resulted in the lowest number of pedotrophic microorganisms in the 20-40 cm layer. The difference between the variant with the highest values was 4.6 times. The highest number of micromycetes was observed in the organic-mineral fertiliser system with $N_{15}P_{15}K_{15}$ in the layer 0-20 cm and amounted to 69.79±1.54 thousand. CFU/g, which is 10 times less compared to the control.

An in-depth analysis of the structure of the microbial community under different fertiliser variants made it possible to determine the direction of microbiological processes in the soil (Table 3).

Table 3. Indicators of the direction of microbiological processes in meadow chernozem carbonate soil under different fertilisation variants, million CFU*/g of soil

Fertiliser variant	Soil layer, cm	Pedotrophy coefficient	Organic matter accumulation coefficient
No fertilisers (control)	0-20	2.0	0.40
	20-40	0.7	0.32
$N_{30}P_{30}K_{30}$	0-20	0.8	1.60
	20-40	1.6	1.94
Organic residual effect+ $N_{30}P_{30}K_{30}$	0-20	1.2	0.89
	20-40	0.7	0.89
Organic residual effect+ $N_{45}P_{45}K_{45}$	0-20	0.8	2.91
	20-40	0.8	1.12
Organic residual effect+ $N_{15}P_{15}K_{15}$	0-20	0.7	2.52
	20-40	0.6	1.19

Note: * – thousand. CFU/g of soil

The pedotrophicity coefficient reflects the functionality of the soil microcenosis structure and points to the degree of organic matter assimilation. The highest rates were observed in the upper layer of the control, in the lower layer of the $N_{30}P_{30}K_{30}$ variant and in the upper layer of the variant with the residual effect of organic matter $+N_{30}P_{30}K_{30}$ (K_{ped} = 2.0, 1.6 and 1.2, respectively), all other rates are in the same range (limits from 0.6 to 0.8). Analysis of the data shows that in variants with higher values, the intensity of decomposition of soil organic matter, in particular humus compounds, increases, which promotes the development of autochthonous microbiota and increases mineralisation processes from the general fund.

The coefficient of organic matter accumulation characterises the intensity of the accumulation of organic compounds in the soil. The highest accumulation rates were observed in the upper layers of the "residual effect of organic matter$+N_{45}P_{45}K_{45}$ and „residual effect of organic matter $+N_{15}P_{15}K_{15}$" ($K_{ac.}$ = 2.91 and 2.52, respectively), slightly lower rates were observed in variant $N_{30}P_{30}K_{30}$ in both layers and in the lower layers of the variants "residual effect of organic matter$+N_{45}P_{45}K_{45}$" and "residual effect of organic matter$+N_{15}P_{15}K_{15}$"($K_{ac.}$ = 1.60, 1.94, 1.12, and 1.19, respectively). The lowest rates were observed in the tilth layer unfertilised ($K_{ac.}$ = 0.40 and 0.32). Thus, when growing peas on meadow chernozem carbonate soil, the best results in humus accumulation are observed with a combination of organic and mineral fertilizers.

The highest yield of peas of the Tsarevych variety was obtained on the variant "residual effect of organic matter $+N_{45}P_{45}K_{45}$" and amounted to 2.94 t/ha, which is 35% more than the control. Organo-mineral fertiliser variants with $N_{15}P_{15}K_{15}$ and $N_{30}P_{30}K_{30}$ formed a higher yield than the variant without fertilisers and the indicators were 2.45 and 2.63 t/ha, respectively.

CONCLUSIONS AND PROSPECTS

The study demonstrated changes in the microbiological parameters of meadow chernozem carbonate soil under different fertilisation systems in the Right-Bank Forest-Steppe. Specific features affecting the processes of humus formation under different fertilisation systems used on pea crops were determined.

It was found that the low agrochemical profile of the variant without fertilisers adversely affects the humification processes in the soil and causes the rapid development of microorganisms engaged in the destruction of the peripheral and nuclear parts of humus substances. The study revealed a high number of pedotrophic and humate-decomposing microorganisms and the lowest coefficient of organic matter accumulation ($K_{ac.}$ = 0.40 and 0.32). Applying only mineral fertilisers during pea cultivation formed a high rate of humus accumulation at the level $K_{ac.}$ = 1.6-1.9. The most favourable conditions for the accumulation of organic matter in the meadow chernozem soil and the highest pea yields of the Tsarevych variety are obtained with the residual effect of organic fertilisers and $N_{45}P_{45}K_{45}$.

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Мікробіологічна оцінка лучно-чорноземного карбонатного ґрунту за різних систем удобрення

Анотація. Мікроорганізми відіграють важливу роль у формуванні родючості ґрунту, підтримці його продуктивності як біоінертної речовини та є індикатором якісних змін у ґрунті. Внаслідок антропогенного впливу на ґрунт чисельність та видовий склад мікробіоти зменшується. Метою дослідження була оцінка чисельності різних фізіологічних груп мікроорганізмів на лучно-чорноземному карбонатному ґрунті за різних систем удобрення гороху. Чисельність різних груп ґрунтових мікроорганізмів визначали за методом Д. Г. Звягінцева шляхом висіву ґрунтової суспензії на тверді живильні середовища. Встановлено, що найсприятливіші показники мікробіоценозу та найвищу врожайність гороху сорту Царевич формували варіанти із залишковою дією органічних добрив та $N_{45}P_{45}K_{45}$. Варіант без добрив індукує розвиток процесів мінералізації органічної речовини в лучно-чорноземному карбонатному ґрунті за рахунок вмісту педотрофних і гуматрозкладаючих організмів і формує найнижчу врожайність гороху. Внесення лише мінеральних добрив під горох формувало високі темпи гумусонакопичення на рівні $K_{ac.}$ =1,6-1,9

Ключові слова: мікробіологічна активність ґрунту, лучно-чорноземний карбонатний ґрунт, мікроорганізми, горох, системи удобрення, органічна речовина