Changes in gray forest soil organic matter pools under anthropogenic load in agrocenoses

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Abstract. The relevance of research is determined by a complex of factors that shape the change in the humus system of gray forest soil dynamics (content, total reserves, values variability of labile and stable humus pools, humification processes direction) under the influence of mineral fertilizers and chemical reclamation in the long-term ecotope conditions. The purpose of the research was to assess the impact of systematic anthropogenic loading on changes in soil organic matter pools. Research methods included long-term stationary studies, laboratory determination of organic matter content and humus group composition, and statistical data analysis. It has been proven that liming practically did not contribute to the increase of the total humus content, however, due to the coagulating effect, certain changes in labile and more stable forms of humus substances occurred in its qualitative composition. It has been established that the cultivation of crops cultivation in crop rotation with a purely mineral fertilizer system did not lead to a significant organic matter accumulation in the soil, despite the increase in crop yield levels under appropriate conditions and the accumulation of a larger vegetative mass of root and post-harvest residues entering the soil, but its losses were significantly reduced. The use of organic components (green manure, by-products) in combination with moderate rates of mineral fertilizers on a liming background (humus content 1.63%), provided a clear tendency to expand humus reproduction by 0.16%, compared to the initial level, and by 15% and 12% more than in the version with no fertilizers and just mineral system. Under such a fertilizing system, there is a tendency to increase more stable humic acids. The increase in humification of organic matter was achieved through the application of fertilizers in limed backgrounds. In this case, the humus formation type exhibited characteristics of humate-fulvate, with a value of 0.9, which is characteristic of soils of forest origin in the northern Forest-Steppe region with the presence of soil-forming factors specific to that territory. The article materials are of practical value for agricultural producers in the development of an innovative strategy for preserving the

environment safety, and soil fertility under the influence of elements of energy-saving technologies for crops.

Key words: humus, ecotope, agrocenosis, soil fertility, humic acids, fertilizers.

INTRODUCTION

Soil and plant cover are one of the main components of the environment, which perform vital biosphere functions in nature and form a single system. The loss of soil fertility, and its degradation deprives plants of the ecological foundations of their existence. Therefore, restoring the fertility of degraded soils is establishing the natural ecological balance of territories disturbed by humans because of irrational economic activity (Kucher et al., 2022; Kyfyak et al., 2022). Changes in forms of land management and ownership, which became the main content of transformations in the agricultural sector of Ukraine in recent years, unfortunately, negatively affected the soil fertility, which lost a significant part of organic matter (Spriazhka et al., 2022). The most important indicator that determines the soil fertility level is the humus content (Boyko et al., 2019; Tkachenko et al., 2020). To preserve arable soil fertility, it is necessary to regularly apply organic fertilizers, which allow optimizing the humus state, which is closely correlated with its physical and chemical properties (Patyka et al., 2017; Kucher, 2018). For fresh organic matter humification, the ratio of carbon to nitrogen in it is important. The optimal mode of plant residue transformation is the ratio (C:N) 26-30:1 (Kalinichev & Iskrenova-Tchoukova, 2011; Dmytrenko et al., 2021).

In Ukraine, the most common soils are soils of forest origin (Havryliuk et al., 2022). According to various data, the area of gray forest soils in Ukraine reaches 4.7–5.5 million hectares (about 9% of the country's territory). The variety of conditions for these soils' formation, discussions about their genesis, and properties are the reasons for the different approaches to the classification of gray forest soils' emergence. Their share in the soil cover of the country is more than 33% and reaches 25% of all agricultural land (Polupan et al., 2005). These soils are most common in the right-bank part of the Forest-Steppe zone. They represent a valuable agricultural fund, the rational use of which is not possible without a detailed study of their humus state, and physical and chemical properties in general (Tsyuk et al., 2018, 2021, 2022; Kaminskyi et al., 2022).

In soil science, the problem of such soil genesis is still debatable and unresolved (Gadzalo et al., 2017; Kiryushin, 2019). Today, the discussion of a new forest soil classification continues, some scientists separate light gray and gray forest soils from dark gray podzolized soils, while combining the latter into one group with podzolized chernozems (Ivanyuk, 2017; Voitovyk et al., 2023).

Forecasting the humus state as an integral indicator of soil fertility has recently received in-depth research (Beregniak et al., 2023). Under anthropogenic influence, the humus is transformed, primarily its labile part, which is removed by water and pyrophosphate extractors (Snakin & Pfisyazhnaya, 1997).

Mobile organic substances serve as a biochemically active component of the soil's organic matter, significantly influencing processes of structural formation and energy accumulation. These compounds are characterized by an elevated content of nitrogen functional groups and a lower carbon content, including carboxyl and phenolic groups. The quantity of mobile humic substances reflects the soil's bioproductive potential and

can indicate the direction of soil processes. The dynamics of labile organic matter content in the soil is closely linked to the input of fresh organic material. The decomposition of these substances contributes to the formation of mobile compounds, which act as a reservoir for the mineralization and accumulation of unstable humic substances (Kachmar et al., 2019a).

The application of fertilizers and the input of labile carbon have important consequences for soil organic matter mineralization due to changes in its fractional composition, which in turn affects soil fertility and carbon sequestration (Wu L et al., 2023). The application of organic fertilizers (60 tons per ha of manure) on gray forest soil increases the relative content of deeply humified humic substances within the humus composition, consequently enhancing soil fertility (Litvinova et al., 2019a; Litvinova et al., 2023b).

Soluble organic matter (SOR) of humus is a heterogeneous phase that includes simple short-chain organic compounds that represent a group of nonspecific substances (fatty acids, organic acids, amino acids, sugars) (Dolenko et al., 2017; Wells & Stretz, 2019). It is believed that the soluble organic matter of humus contains components that are extremely mobile, capable of chemical transformation, controlling several chemical, physical, and biological processes that occur in the soil environment (Litvinova et al., 2020; Symochko, 2020; Litvinova et al., 2023a).

Although mobile organic matter usually makes up less than 1% of its total amount, it plays a very important role in the biogeochemical cycle of carbon, nitrogen, and phosphorus, as well as in the transport of mineral substances and its influence on the general ecological state of the environment. In addition, it is a stabilizing factor of soil colloids and aggregates. The formation and mobility of soluble organic matter in agricultural soils depend on many environmental factors (climate, hydrological conditions, microbiological activity) and anthropogenic factors (tillage, mineral nutrition, organic fertilizing, and liming) (Nuzzo & Sánchez, 2013; Litvinova et al., 2019b; Kachmar et al., 2019b; Ivanova et al., 2023). Therefore, the study of the above factors influence on the humus content and its fractional composition is extremely relevant (Tonkha et al., 2019).

The research objective is to determine how the composition of organic humus components changes when different fertilizer systems (mineral and organo-mineral) are used in the context of regular lime application on gray forest coarse-loamy soil.

MATERIALS AND METHODS

The research was carried out during 2018–2020 based on the Department of agro-soil science and soil microbiology of the National Scientific Center of the 'Institute of Agriculture of the National Academy of Agrarian Sciences', founded in 1992. The experiment studied the effect of different doses and forms of calcium-containing meliorants, organic and mineral fertilizers, and their combinations on the properties of gray forest coarse light loamy soil for growing winter wheat. With the onset of 2006 (the beginning of the III rotation of crop rotation), the reconstruction of the experiment was carried out to adapt it to modern conditions, and repeated liming was also carried out. An experiment placed in three fields of a 7-field crop rotation. The scheme of the experiment includes ten variants in a quadruple repetition, the sowing area is 60 m^2 (10×6), and the accounting area is 24 m^2 (6×4).

The experiment studied the influence of meliorants, organic (green manure and plant by-products), and mineral fertilizers on the properties of gray forest soil and crop rotation productivity (Option 1 – With no fertilizers (control); $2 - CaCO_3$ (1.0 Hr); $3 - N_{60}P_{30}K_{60}$; $4 - N_{60}P_{30}K_{60} + CaCO_3$ (1.0 Hr); $5 - N_{120}P_{60}K_{120} + CaCO_3$ (1.0 Hr); 6 - green manure (mustard 15 t ha⁻¹) + CaCO_3 (1.0 Hr); 7 - green manure + by-products + NPK + CaCO_3 (1,0 Hr)). Accordingly, a single dose of the active substance is for winter wheat of the Kraevyd variety $- N_{60}P_{30}K_{60}$.

Organic fertilizers, namely by-products of the precursor (lupine straw - 2-4 t ha⁻¹) and green manure (green mass of oil radish 15–18 t ha⁻¹) were plowed during the main tillage. Nitrogen fertilizers were applied in ammonium nitrate form (DSTU 7370:2013), and early spring feeding was carried out. Phosphorous fertilizers in granulated superphosphate form and potash - in potassium chloride form. Chemical amelioration was carried out, lime was applied according to the value of hydrolytic acidity in a full dose (1.0 SI) (defect 4.5–6.0 t ha⁻¹ with a CaCO₃ content of about 50%), and it was applied under plowing.

For a more objective assessment of the specified factors' influence on the soil humus state, the obtained post-variant results were compared not only with the absolute control but also tied to the initial state. For this purpose, a full-profile soil section was dug on a fallow site (fallow age 29 years), which is located next to the experimental field, in which soil samples were taken along horizons along the entire profile.

Botanical studies for such a long time in this area indicate that a successional change of plant communities took place. Immediately after removal from agricultural cultivation, weeds begin to grow intensively in these areas. In the past, arable land receives a significant amount of plant material due to a more developed root system and a larger volume of phytomass than cultivated plants. Thanks to such dynamics of changes in the plant coenosis and the long modern processes of the organoprofile formation, fallow soils in terms of their properties are as close as possible to the category of 'virgin soil'.

Agrochemical selection of soil samples was carried out according to DSTU ISO 10381-1:2004 and DSTU 4287:2004; determination of organic matter (humus) content according to DSTU 4289:2004; humus group composition according to the Tyurin method in the modification of Kononova and Belchikova (DSTU 7855:2015); available (labile) organic matter (DSTU 4732:2007). Analytical works were carried out in the laboratory of ecological land safety, product quality, and environment of the 'Institute of Soil Protection of Ukraine'. Mathematical processing of analytical studies results of the soil and yield data was carried out by the dispersion and correlation-regression method, using Microsoft Excel and Statistica 6.0 programs.

RESULTS AND DISCUSSION

Humus content and its total reserves are integrated indicators of soil formation. When assessing the humus condition of soils, it is especially important to study the parameters of profiled carbon accumulation considering the depth and strength of humus horizons. The relevant regularities are outlined in the works of several authors, where the results of research on the humus content of forest-origin soils along the vertical profile and a sharp decrease in humus with depth compared to chernozems are presented (Dmytruk & Demyd, 2019). Gray forest soils are characterized by a clear profile

differentiation according to the eluvial-illuvial type and a peculiar humus accumulation in them. According to our data, from the generalized humus reserves (124 t ha^{-1}), which the studied soil has on a long-term fallow, 51.6% is contained in the HE horizon (0–29 cm) and 78.3% in the HE+lh horizons with a total thickness of 0–56 cm. Therefore, most humus reserves are concentrated mainly in the upper layer (Table 1).

Research option	Fallow for 1992–2019, %	Humus content, %	± to control, %	The difference is the humus content on a fallow area		
				%, ±	In %,	
					\pm to the initial	
With no fertilizer (control)	1.47 ± 0.02	1.40 ^b	_	-0.07	-4.8	
CaCO ₃ (1.0 Hr)		1.42 ^b	+0.02	-0.05	-3.5	
$N_{60}P_{30}K_{60}$		1.45 ^b	+0.05	-0.02	-1.6	
N ₆₀ P ₃₀ K ₆₀ + CaCO ₃ (1.0 Hr)		1.50 ^b	+0.10	+0.03	+0.2	
N ₁₂₀ P ₆₀ K ₁₂₀ + CaCO ₃ (1.0 Hr)		1.50 ^b	+0.10	+0.03	+2.0	
green manure $+$ CaCO ₃ (1.0 Hr)		1.62 ^{ab}	+0.22	+0.15	+10.2	
green manure + by-products +		1.63 a	+0.23	+0.16	+10.9	
$N_{60}P_{30}K_{60} + CaCO_3 (1.0 Hr)$						
average		1.52				
Sx		0.04				
V%		8.57				
S		0.13				

Table 1. The organic matter (humus) content in gray forest soil (layer 0–20 cm) depending on different levels of loading by fertilizers and meliorants

Means with the different letter are highly significantly different according to the Fisher's test ($P \le 0.05$).

The relevance of the gray forest soils study is determined by the fact that, along with their high fertility, they quickly lose it due to the irrational use of soils in agricultural production. Due to the low structure and unfavorable water-air properties, during the plowing of these soils, blocks are created that quickly settle and easily float. And yet such gray forest coarse silt-light loamy soils, which were formed on thick loamy loams, can be classified as 'especially valuable' soils of the Northern Forest Steppe.

When comparing the studied indicators with the initial level, completely different results were obtained, which indicate that the use of soils in agriculture without crop fertilizing leads to the inevitable predominance of humus decomposition processes over synthesis, which leads to a decrease in its content and a violation of the balance in agrocenoses. According to research results, this indicator was 4.8% relative to the initial level.

Studies conducted according to the proposed scheme established that liming had almost no effect on the total humus content. Thus, in the control variant and when applying lime at the full rate of hydrolytic acidity, the humus content in the soil was almost the same, 1.40-1.42%, respectively.

In their study, Tkachenko et al. (2020) confirm the research findings regarding the changes in the acidic properties of gray forest soil depending on different fertilizer systems in crop rotation and the long-term aftermath of chemical amelioration. They also emphasize the indispensable role of organic fertilizers in enhancing the buffering capacity of gray forest soils and neutralizing the acidifying impact of mineral fertilizers, both compared to the control variant and the initial level (Tkachenko et al., 2020).

Under the mineral system ($N_{60}P_{30}K_{60}$) only, despite a significant increase in crop yield and, accordingly, the accumulation of a larger vegetative mass of root and post-harvest residues entering the soil did not lead to its significant accumulation, although its losses were significantly reduced. A positive effect was not achieved with the separate application of meliorants and mineral fertilizers, and with their simultaneous application, a tendency to its increase was observed. A similar trend was observed during the application of double doses of $N_{120}P_{60}K_{120}$ on calcification background.

One of replenishing soil reserves with fresh organic matter in the conditions of a sharp shortage of organic fertilizers is the precursor by-products and green manure as a biological component of the natural environment. So, under such conditions, the total humus content during the years of research was at the level of 1.63%, which is 15% and 12% more, respectively, than in the version with no fertilizers and a purely mineral system. The relative increase in the total humus content by 0.16%, compared to the fallow value ($LSD_{05} = 0.12$), indicates that plowing the predecessor by-products and green manure once per crop rotation, in combination with the annual application of mineral fertilizers recommended doses, contributes to increasing the total humus content in gray forest soil, due to the tendency towards its slow accumulation. The variability degree for this characteristic was found at a weak level (V = 8.5%) and proves the indicator's stability. These results are corroborated by the research conducted by Yang et al. (2023), which established the highest relative content of humus-like components when organic amendments were applied.

The use of organic fertilizer components with moderate doses of mineral fertilizers in the ecotope was characterized by a clear tendency to recover, especially on calcified backgrounds. In this case, the extended humus recovery to the initial level was within 10% in percentage terms.

Not only the content and total reserves but also the group composition of humus reliably characterize its natural qualities in the zonal-genetic aspect of the environment. It is believed that the group composition of the humus is a stable feature that characterizes its genetic features. Recently, a lot of data has been obtained on the existence of certain dependencies between the qualitative humus composition and soil use nature (Skrylnyk et al., 2022). Therefore, there is a need to give a comprehensive humus characterization based on anthropogenic factors that influence the qualitative composition of the studied soil. The virgin soil composition is dominated by fulvic acids (FA), the share of which is 28.0% of the total content of humus acids with a low content of humic acids (HA) at 21.5%. The ratio of HA/FA is 0.76, such a ratio makes it possible to assess the humus type as humate-fulvate (Fig. 1). Several authors note that the longterm use of organic and mineral fertilizers intensifies the process of new humic acids formation and their mobile forms formation - the ratio HA/FA (the ratio of carbon humic to fulvic acids) is 0.80 in the control with no fertilizers and 1.2–2.0 in the fertilizer variants on the liming background (Litvinova et al., 2019b). Our studies have shown that the most favorable conditions for organic substances humification are created with the combined use of organic and mineral fertilizers, but the type of humus remains humusfulvate - the ratio of HA/FA is - 0.9, on the control version - 0.81 and the background of liming - 0.86. Long-term use of the studied fertilizing systems in generally accepted doses had little effect on the ratio between humic and fulvic acids. Only a trend in the change of these humic acid pools is observed. In the variant with no use of fertilizers, the fulvic acids content is at the level of virgin soil (28.4% of the total humic acids

content), and the content of more stable at 23.0%. The above-mentioned trend is associated with higher humus mineralization in arable lands and its destructive effect on the qualitative composition of humic substances. In their study, Borko et al. (2022)

indicated that such anthropogenic influence is associated with an increase in soil microbiological and enzymatic activity.

On the variants where fertilizers were not applied (soils of extensive agriculture), the regularity of humus formation enriched with the most soluble compounds, which have more pronounced acidic properties, was observed. In variants with green manure and by-products use, a tendency to increase more stable humic acids was observed. The increase in humus content was especially noticeable when they were applied on limed backgrounds. At the same time, the total humic acids content increased to some extent by 9% relative to the control variant. The ratio of HA/FA on these variants reached 0.9.

In general, these changes in the carbon group composition under the



Figure 1. Qualitative assessment of gray soil (layer 0–20 cm) humus condition depending on different levels of load by fertilizers and meliorants.

Note. Fertilizer option: $1 - \text{with no fertilizer (control)}; 2 - CaCO_3 (1.0 \text{ Hr}); <math>3 - N_{60}P_{30}K_{60}; 4 - N_{60}P_{30}K_{60} + CaCO_3 (1.0 \text{ Hr}); 5 - N_{120}P_{60}K_{120} + CaCO_3 (1.0 \text{ Hr}); 6 - \text{green manure} + CaCO_3 (1.0 \text{ Hr}); 7 - \text{green manure} + by-products + N_{60}P_{30}K_{60} + CaCO_3 (1.0 \text{ Hr}); 8 - \text{fallow area.}$

anthropogenic factors influence did not lead to a change in the humic-fulvate type of humus, which is characteristic of the forest-origin soils of the northern Forest Steppe with soil formation factors present in this area. Carbon extracted by water and hot water or extracted by a 0.2 N NaOH solution can be a reliable criterion for the content of active, easily soluble organic matter in the soil. The research conducted by Litvinova et al. (2019a) revealed that these organic matter fractions are primarily represented by compounds formed at the early stages of their transformation and sensitive pools of microbial biomass, which reflect any changes in the ecosystem.

Labile humic substances, which are biochemically the least stable, serve as an available source of nutrition for plants and, first, determine effective soil fertility. Their content in gray forest soil according to the application of stronger reagents is shown in the table (Table 2).

Even though, in all cases, different reagents were used for the extraction of the humic substances, an increase in their absolute values was established as more active reagents were used, and at the same time, the same general patterns were preserved. The organic substance that was extracted into the water extract, which to some extent imitates the soil solution, was observed in the highest concentration in the original soil, the version without the use of fertilizers (control), and in the mineral system. However, liming of unfertilized soil, as well as fertilized by green manure and plant by-products, contributed to the reduction of the above-mentioned humus fractions. However, in the

analysis conducted by Rosa & Debska (2018), there is noted a lack of changes in the content and proportion of mobile organic matter in the soil associated with the application of organic fertilizers. It was found that the main influence on the content and distribution of labile and water-soluble organic matter makes the method of land use and the related anthropogenic impact on the ecotope.

Table 2. The organic matter (humus) mobile forms content in gray forest soil (layer 0–20 cm)
depending on different levels of loading by fertilizers and meliorants

	Aver. C	Water soluble		Soluble in			
Research option		%	% from %	% from %	% from		
			Aver. c	70	Aver. c		Aver. c
with no fertilizer (control);	0.81 ^{dc}	0.016 ^{ab}	1.97	0.025 ^{ab}	3.08	0.21 ^{ab}	25.9
CaCO ₃ (1.0 Hr);	0.82 ^{cd}	0.014 ^b	1.70	0.023 ^b	2.8	0.19 ^b	23.1
$N_{60}P_{30}K_{60}$	0.84 ^{bd}	0.017 ^a	2.02	0.027 ^a	3.21	0.22 ^a	26.1
N ₆₀ P ₃₀ K ₆₀ + CaCO ₃ (1.0 Hr);	0.87 ^b	0.015 _{ab}	1.72	0.024 ^{ab}	2.85	0.20 ^{ab}	22.9
N ₁₂₀ P ₆₀ K ₁₂₀ + CaCO ₃ (1.0 Hr);	0.87 ^b	0.016 ^a	1.83	0.027 ^a	3.10	0.22 ª	25.2
green manure +	0.93 ^a	0.010 °	1.07	0.017 °	1.82	0.14 °	15.0
CaCO ₃ (1.0 Hr);							
green manure + by-products +	0.94 ^a	0.012	1.27	0.020 bc	2.12	0.17 ^b	18.0
$N_{60}P_{30}K_{60} + CaCO_3 (1.0 Hr)$		bc					
Soil layer 0–20 cm from the	0.85 ^b	0.017 ^a	2.00	0.025 ^{ab}	2.94	0.21 ^{ab}	24.7
fallow area							
Sx	0.01	0.00		0.001		0.01	
V%	7.8	15.65		13.34		13.4	
S	0.03	0.00		0.00		0.03	

Means with the different letter are highly significantly different according to the Fisher's test ($P \le 0.05$).

Thus, liming practically did not contribute to the growth of the total humus content, however, due to the coagulating effect, certain changes in labile and more stable forms of humic substances occurred in its qualitative composition. The introduction of calcium into the soil led to a stronger fixation of humic substances since this element slows down the process of organic substances' transformation into mobile humic acids within the studied ecotope. At the same time, a certain reduction of labile humus fractions during liming is not a limiting factor of soil fertility since the highest levels of crop yields were obtained on limed versions.

CONCLUSIONS

It has been found that prolonged application of a complex of agronomic practices in the ecotope does not alter the humus type, which remains humate-fulvate with a value of 0.9, indicating the stability of soil formation processes.

In the control variant (without fertilizers), there is an observed pattern of organic matter formation enriched in highly soluble compounds with pronounced acidic properties.

The mineral fertilizer system ($N_{60}P_{30}K_{60}$) did not lead to an accumulation of soil organic matter, despite the greater vegetative mass of root and post-harvest residues entering the soil. However, losses of organic matter significantly decreased compared to the initial level, representing 0.2%.

With the systematic use of cover crops and agricultural by-products as a biological component of the agroecosystem, in combination with moderate mineral fertilization (N60P30K60) and periodic liming, there was a tendency to increase the overall organic matter content by 10% relative to the initial level and by 15% compared to the control without fertilizers.

An increase in the humification of organic matter was observed with the application of fertilizers in the presence of lime, leading to a humate-fulvate type of humus formation, with a value of 0.9, which is characteristic of forest-origin soils in the northern Forest-Steppe region with the prevailing soil-forming factors.

It was found that the fertilization system in combination with optimal liming rates (Cover crop + agricultural by-products + $N_{60}P_{30}K_{60}$ + CaCO₃ (1.0 Nr)) reduces the mobility of the humus system, increases the relative proportion of humic acids, and slows down the formation of water-soluble, hot-water-soluble, and labile organic matter compared to the control without fertilizers, within a range of 20%.

A promising direction for future research is the evaluation of soil nutrient status and carbon and nitrogen cycling indicators.

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