



Research Article

Molecular and elemental compositions of humic acids of arable soils in north forest-steppe zone of Russia

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Abstract

The degradation of arable lands is becoming widespread and leads to significant loss of soil organic matter (SOM), expressed both in quantitative losses and in qualitative changes in the molecular and elemental composition of SOM. In this regard, a study of soils in the forest-steppe zone of Russia was conducted on the territory of the proposed Carbon Polygon in the Tambov Region and its humus state was characterised. Four contrasting types of land use were selected: long-term arable land, fallow land (60 years of self-restoration), a shelterbelt site (as a zonal reference) and an eroded site with a buried arable horizon. Carbon and nitrogen stocks were investigated, the element and molecular composition of humic acids (HAs) being analysed. This set of sites allows us to

analyse different stages of SOM transformation under varying scenarios of land use, ranging from active degradation to post-agrogenic recovery. To analyse the molecular structure, the method of ^{13}C NMR spectroscopy was used. As a result of the study, it was found that the type of land use determines both the quantitative indicators of SOM and nitrogen stocks and the molecular structure of soil HAs. It was established that, in soils of fallow lands, a significant amount of SOM accumulates (up to $50.81 \pm 0.96 \text{ kg/m}^2$), which possesses a relatively high level of resistance to biodegradation, determined by a high concentration of aromatic structural fragments. It was noted that arable and fallow soils may have high carbon sequestration potential compared to zonal soil and eroded site. Long-term agricultural practices lead to the formation of relatively stable molecular complexes of HAs, which show signs of oxidative degradation of SOM due to an increased proportion of carboxyl structural fragments.

Keywords

^{13}C NMR spectroscopy, carbon polygon, Chernozems, soil organic matter, stabilization

Introduction

Humic substances and SOM plays a crucial role in providing the fertility of arable lands and is one of the key factors involved in regulating the global climate (Abdelrahman et al. 2023). Arable lands, alongside forests and wetlands, possess significant potential for accumulating additional carbon in soils and plant biomass, thereby reducing atmospheric CO_2 levels (Griscom et al. 2017). Similar carbon sequestration practices are also common in central European countries, where some of the land was used for coal mining (Badraghi et al. 2025). The molecular composition of HAs, as a primary component of SOM, is of substantial importance for soils intensively used in agriculture, as it determines the degree of SOM resistance to biodegradation (Mustafa et al. 2023). Understanding the processes of SOM stabilisation is critical for soils involved in agriculture, as they are prone to SOM loss due to unsustainable land use (Silveira et al. 2022). In Russia, 130 million hectares of agricultural land, including 85 million hectares of arable land, are at risk of erosion and disappearance, while in Western Europe, about 41 million hectares are under threat of erosion (Ivanov and Stolbovoy 2019). As a result of climate change and unsustainable land use, processes such as desertification, acidification, salinisation, contamination by priority toxicants are intensifying and a decline in SOM content is observed (Ivanov et al. 2021).

Global challenges related to climate change, land degradation and increasing the sequestration potential of natural and anthropogenically transformed lands have led to the development of natural-climate initiatives in Russia. Based on the 21st UN Framework Convention in Paris ("The Paris Agreement"), Russia agreed a long-term national "low-carbon" development strategy, which includes reducing the content of key climate-active gases in the atmosphere (IPCC 2021). As part of implementing this

agreement, the "4 per 1000" initiative was approved, aiming to increase SOM content in soils of arable lands (Ivanov et al. 2021).

Chernozems are the most fertile soils in the world used in agriculture worldwide and play a crucial role in international and national programmes aimed at their conservation and sustainable use (Organization 2021). They are predominantly distributed in Russia and Ukraine, with smaller areas present in south-western Poland, Hungary, Croatia, Czechia, Austria, Slovakia and Romania (You et al. 2023). The Chernozem belt of Russia is located in the forest-steppe and steppe territories, in the north and it is bounded by the taiga zone. The southern border in the west is limited by the mountain province of the Caucasus (43° N), while in the east, Chernozems extend as far as the Trans-Baikal Territory (Dobrovolsky and Urusevskaya 1984). The first major work on the study of Chernozems in Russia is the book "Russian Chernozem", written by the great Russian soil scientist - V.V. Dokuchaev. In this work, information about Chernozem-type soils in various regions of the country was summarised and systematised and the main differences between them were identified (Aparin 2008). According to the USSR classification of soils, up to 19 different types of Chernozems and seven types of meadow-chernozemic soils are distinguished; in the international classification IUSS WRB, these soil types correspond to the Chernozems group, as well as to the Phaeozems and Vertisols groups (USSR 1977, WRB 2022). Such great diversity is determined by climatic features, differences in hydrothermal regimes and the morphological features of carbonates within the soil profile (Smirnova et al. 2025). In terms of zonality, Chernozems are widespread in the forest-steppe and steppe zones, on carbonate-rich loams, as well as on marls, clays, flysch sediments, deluvia and, for meadow-chernozemic soils, fluvial sediments (Rojas et al. 2025). The Tambov Region is located in the zone of northern and typical forest-steppe. Several hypotheses exist regarding the formation of this territory (Chendev et al. 2022), namely: 1 – the advance of forest on to the steppe, which resulted in the transformation of meadow-chernozemic soils (Luvic Chernozems) into soils of forest genesis (Luvic Phaeozems) (Tanfil'ev 1896); 2 – soils of forest soils (Luvic Phaeozems) were an independent formation and their predecessors were forest-meadow soils that gradually transformed into soils of forest genesis (Akhtyrtsev 1979); 3 – the movement of forest areas within the steppe zone (Denisov 1935); and 4 – anthropogenic influence, which led to the expansion of the steppe area within the forest-steppe territory (Alexandrovsky and Alexandrovskaya 2005). The formation of such an extensive belt of highly productive ecosystems determines the carbon balance of the Eurasian territory. This is due both to the presence of vast areas of agricultural land, which act as hyperaccumulators of organic matter and to the presence of forest areas, which play a stabilising role for adjacent agroecosystems (re-distribution of water, protection from erosion, microclimate formation) (Chendev et al. 2026). Nevertheless, humus-accumulating horizons of Chernozems are used as a substrate in the production of quarry dumps, which leads to significant losses of organic matter (Abakumov and Frouz 2009).

Soils involved in agriculture are subject to intensive anthropogenic pressure (Chendev et al. 2017). Despite their potential to accumulate significant amounts of organic matter and,

thus, contribute to climate change mitigation, they can also be active emitters of climate-active gases (Lugato et al. 2018). SOM is dynamic in time and space and its components possess distinct characteristics and turnover rates within the ecosystem, which allows for the analysis of various stages of SOM transformation under land-use influence (Bezuglova et al. 2020). A large number of methods exist to determine the qualitative composition of organic matter, including fractionation, proton magnetic resonance, electron paramagnetic resonance and gel chromatography (Kopecký et al. 2021). One effective method for analysing the molecular composition of SOM is studying the structure of HAs using ^{13}C NMR spectroscopy (Cao et al. 2011). This method, combined with elemental analysis, helps determine the maturity of organic matter and its resistance to biodegradation, which is especially crucial for agricultural soils. In this context, the hypothesis of this work is that the organic matter of soils used in agriculture possesses a relatively high level of resistance to biodegradation, explained by its high maturity. Therefore, the aim of the work was to study the molecular composition of HAs from soils forming on the territory of a proposed Carbon Polygon in the Chernozem zone of Russia. The following tasks were set to achieve this aim: 1) to identify the physicochemical characteristics and carbon and nitrogen stocks of the studied soils; 2) to investigate the elemental and molecular composition of HAs extracted from the soils; and 3) to assess the degree of stabilisation of SOM in soils with long-term agricultural use.

Materials and Methods

The study area

The study area is located within Tambov Region, in the central part of the Oka-Don Plain. The relief is flat with slopes, at an altitude of 130-160 m above sea level. The territory belongs to the floodplain and above-floodplain terraces of the Tsna River. Quaternary deposits are represented by alluvial sands and glacial loams (Karandeeva 1957). The relief features weakly expressed gullies and ravines (see Fig. 1). The climate is temperate continental. The average annual temperature is $+6^{\circ}\text{C}$. A stable snow cover forms in late November or early December, with a snow depth of 20-40 cm. The average annual precipitation is 550 mm. The moisture coefficient ranges from 0.7 to 0.8. According to the Köppen-Geiger climate classification, the northern part of the region falls under the hot-summer humid continental climate (Dfa), while the southern part has a warm-summer humid continental climate (Dfb) (European Food Safety Authority (EFSA) et al. 2026). The study area is located within the forest-steppe zone of the East European Plain. However, due to intensive agricultural activity, the natural landscapes have undergone significant changes. Originally, the area was covered by oak-linden forests, but as a result of ploughing and the establishment of shelterbelts, arable land now dominates most of the territory. The main tree species chosen for creating the shelterbelts are silver birch (*Betula pendula*) and aspen (*Populus tremula*).

The most common soils in this territory are Haplic Chernozem and Luvisol Chernozem, which form on watersheds and sparsely occupied by meadow-steppe vegetation. Luvisol Greyzem Phaeozem, where fragments of broadleaved forests are preserved in the

vegetation cover and, in some areas of shelterbelts, amongst anthropogenically transformed soils, Luvic Chernozem are distinguished. Parent materials are represented by cover loess-like loams and clays overlying moraine deposits. The CaCO_3 concentration ranged from 0.01 to 0.4%. A description of the studied soils is provided in Suppl. material 1. During the fieldwork, it was determined that the following soils are developing on the territory of the proposed Carbon Polygon: Luvic Chernozem (Loamic) (P1-P2), Luvic Greyzemic Phaeozem (Loamic) (L1), Luvic Greyzemic Phaeozem (Loamic) on buried Chernozem (Loamic) (E1) and Luvic Greyzemic Phaeozem (Humic, Loamic) (L2) (see Suppl. material 2).

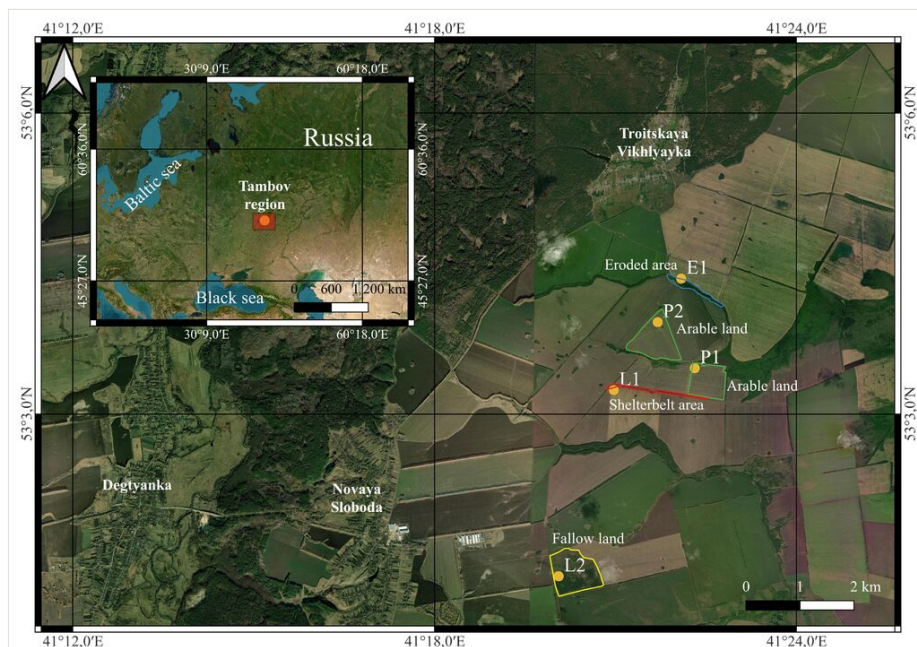


Figure 1.

The study area of Tambov Region, Russia. P1-P2 – Luvic Chernozem (loamic); L1 – Luvic Greyzemic Phaeozem (loamic); L2 – Luvic Greyzemic Phaeozem (humic, loamic); E1 – Luvic Greyzemic Phaeozem (loamic) on buried Chernozem (loamic).

Sampling strategy

The forest-steppe zone of Russia is characterised by a high degree of agricultural development, which has led to significant transformation of natural soils. In this regard, a study site was specifically selected for this research where several genetically contrasting soil profiles, differing in the type and duration of anthropogenic impact, co-exist within the same landscape. The key criterion for selecting the site was the ability to trace different pathways of SOM transformation under comparable lithological and geomorphological conditions.

The selection of arable, fallow, natural as well as eroded lands is based on the landscape diversity within the territory of the proposed Carbon Polygon (Carbon supersite). A carbon polygon is a specially equipped natural area where scientists measure how ecosystems (forests, wetlands, soils, seas) absorb and release greenhouse gases primarily CO₂ and CH₄. The territory of the proposed Carbon Polygon in the Tambov Region is intended to fill gaps in the Russian Carbon Polygon network, which currently has almost no coverage of the Chernozem zone in southern Russia. The sampling strategy was designed to capture the diversity of land-use types typical for this territory, including arable land, fallow land, shelterbelts and eroded areas. The eroded area within the territory of the proposed carbon polygon began to form about 70 years ago, when a firebreak channel was excavated on arable land, as a result of which part of the arable soils was buried under overburden material. On the arable lands, the main crops cultivated are winter and spring wheat; after harvesting, plant residues are ploughed back into the soil to a depth of up to 30 cm. Thus, agricultural fields are ploughed annually. The selected sites represent key scenarios for carbon stock assessment and SOM transformation monitoring within the future polygon. Soil samples from five soil profiles were collected from all diagnosed horizons (14 samples) to assess the concentration and stocks of carbon and nitrogen. For the arable soils (P1–P2), two soil profiles and four soil samples were collected (Ap–Bt); for the soil shelterbelt area (L1), three soil samples were collected (Ah–E–Bt); for the fallow soil, three soil samples were collected (Ah–A/E–Bt); and for the eroded area soil (E1), four soil samples were collected (Ah–Bt–[Ap]–[Bt]). For analysing the molecular composition of HAs, samples were used from the upper humus-accumulative horizons, transitional horizons, as well as a buried humus-accumulative horizon (7 samples). Soil sampling was carried out in mid-April 2025, which allowed for sample collection from arable land areas (P1-P2). Samples for chemical analyses were stored at +4°C to preserve their main soil parameters. Soils were ground and passed through a 2 mm sieve to obtain the fine earth fraction.

Laboratory analysis

The pH_{H2O} value was determined by potentiometric method in water suspension (in ratio 1:2.5) (FAO 2021). The particle size distribution of the samples was determined by the sedimentation method (Bowman and Hutka 2002). Soil organic carbon (SOC) was determined by the Turin method (FAO 2021b). Total nitrogen (TN) concentration was determined by the Kjeldahl method (FAO 2021c). Carbonate concentration in the soil was determined by the acidometric method. C, H and N in HAs concentration were determined on a CHN analyser (EA3028-HT EuroVector, Pravia PV, Italy). Carbon and nitrogen stocks were determined by the formula:

$$\text{Carbon stock} = p \cdot C \cdot H \cdot 0.1,$$

where, carbon stocks (kg/m²); p-horizon density (g/cm³); C- carbon concentration (%); 0.1 - conversion factor; H – depth, cm.

$$\text{Nitrogen stock} = p \cdot N \cdot H \cdot 0.1,$$

where, nitrogen stocks (kg/m^2); p - horizon density (g/cm^3); N - nitrogen concentration (%); 0.1 - conversion factor; H – depth, cm.

HAs were extracted from the studied soils by the method of the International Society for the Study of Humic Substances using sodium hydroxide (NaOH) solution in modification of Vasilevich et al. (2019). All studied HAs were pre-treated to remove carbonates during decalcification. Elemental composition of HAs is the percentage concentration of elements C, H, N and O calculated on an ash-free sample. The Van Krevelen diagram (Rodríguez-Murillo et al. 2017) was used for graphical analysis of elemental composition (in H/C-O/C coordinates). Oxygen concentration was calculated from:

$$O = 100 - (C + H + N),$$

where C, H and N concentrations were obtained by CHN analyser.

The H/C ratio was calculated by the following formula:

$$H/C = (H, \%/1.01) / (C, \%/12.01).$$

The O/C ratio was calculated by the following formula:

$$O/C = (O, \%/16) / (C, \%/12.01).$$

The degree of oxidation was calculated from:

$$w = 2 * ((O/16) - (H/1.01)) / (C/12.01),$$

where C and H concentrations were obtained by the CHN analyzer.

Solid-state spectra of HAs were determined by CP/MAS ^{13}C -NMR spectroscopy on a Bruker Avance 500 NMR spectrometer (Massachusetts, United States) in a 4 mm ZrO_2 rotor. After obtaining the HAs spectra, the analysis of the results was performed using the MestReNova software package (Mestrelab Research, Santiago de Compostela, Spain). In the obtained spectra, eight groups of structural fragments were identified: 0-46 ppm – Alkyl C; 46-60 ppm – O, N-Alkyl; 60-105 ppm – CH_2OH , CHOH ; 105-144 ppm – Aromatic C; 144-164 ppm – O, N-Aromatic; 164-183 ppm – COO-R ; 183-190 ppm – Aromatic C=O ; 190-204 ppm – C=O . Based on these, the hydrophobicity index ($\text{AL h,r} + \text{AR h,r}$) and the humification index (C,H-AL/O,N-AL) were calculated (Lodygin et al. 2014).

Statistical analysis

Statistical analysis of the data (see Suppl. material 3, Suppl. material 4) was performed using ANOVA, followed by post-hoc pairwise comparisons to evaluate differences amongst soil profiles and horizons. The results are shown as the mean $\pm$ SD and statistically significant differences ($p < 0.05$) are noted by different letters in the corresponding figures. Statistical data analysis and visualisation were performed in GraphPad Prism 9 (GraphPad software LLC).

Results and Discussion

Features of the structure and transformation of soils under long-term agricultural use

The studied soils exhibit significant differences in both morphological structure and chemical composition (see Table 1). Long-term agricultural use leads to substantial transformation, noted in reduced acidity, increased SOC and decreased nitrogen concentration, as well as levels of biogenic elements compared to zonal soil analogues (Smirnova et al. 2025). The CaCO₃ concentration in the soils had a relatively low value, which varied from 0.04% to 0.40%. The analysis of intraprofile pH distribution revealed that arable soils P1-P2 are characterised by a slightly acidic pH reaction, which remains almost unchanged within the profile. This is due to active anthropogenic impact and the periodic application of physiologically acidic fertilisers. The shelterbelt soil (L1) was characterised by a sharp decrease in pH in the E horizon, which is explained by an active leaching process, while, in the underlying horizon, the pH value increases due to the presence of carbonates in the lower horizons and parents materials. The same trend is characteristic of both fallow soils (L2) and the eroded site (E1). However, the eroded site is characterised by a decrease in pH in the buried humus-accumulative horizon [Ap], reflecting the history of agricultural management. According to ANOVA, the pH of eroded soil (E1) differed significantly (p-value < 0.05) from the other soil profiles. This supports our thesis that land-use history exerts a strong influence on subsequent soil development.

Table 1.														
The physicochemical properties (mean ± SD) of studied soils. Each soil sample was analysed in triplicate, except for particle size distribution, concentration of carbonates (CaCO ₃) and bulk density (p), which were analysed once. ArL – arable land; ShA – shelterbelt area; FIL – fallow land; ErA – eroded area.														
Soil ID Landuse type	Hor.	pH	Particle size distribution, %			SOC	TN	C/ N	P ₂ O ₅	K ₂ O	N-NH ₄ ⁺	N-NO ₃ ⁻	p	CaCO ₃
			Sand	Dust	Clay									
P1 ArL	Ap	5.48 ± 0.16	5	42	53	4.26 ± 0.15	0.29 ± 0.01	17	38.1 ± 1.3	171.9 ± 8.1	5.56 ± 0.04	0.09 ± 0.01	1.406	0.16
	Bt	5.32 ± 0.31	17	55	38	0.47 ± 0.02	0.05 ± 0.01	11	10.1 ± 1.0	105.2 ± 2.5	1.94 ± 0.12	0.15 ± 0.01	1.548	0.10
P2 ArL	Ap	5.53 ± 0.44	42	23	35	3.45 ± 0.10	0.25 ± 0.01	16	53.1 ± 3.1	96.8 ± 3.9	7.05 ± 0.56	0.09 ± 0.01	1.172	0.18
	Bt	5.53 ± 0.12	45	15	40	1.88 ± 0.01	0.15 ± 0.01	15	49.2 ± 3.3	93.1 ± 2.2	5.37 ± 0.37	0.19 ± 0.02	1.179	0.14

Soil ID Landuse type	Hor.	pH	Particle size distribution, %			SOC	TN	C/ N	P ₂ O ₅	K ₂ O	N-NH ₄ +	N-NO 3 ⁻	p	CaCO ₃
			Sand	Dust	Clay	%	mg/kg		g/cm 3	%				
L1 ShA	Ah	5.17 ± 0.07	37	10	53	3.37 ± 0.13	0.47 ± 0.01	8	101.2 ± 5.3	374.7 ± 14.7	14.1 ± 0.1	0.97 ± 0.09	0.712	0.10
	E	4.49 ± 0.12	62	10	28	0.44 ± 0.03	0.08 ± 0.01	6	82.1 ± 1.1	189.7 ± 6.0	5.11 ± 0.81	5.55 ± 0.03	1.559	0.01
	Bt	5.19 ± 0.25	37	30	33	0.26 ± 0.04	0.04 ± 0.01	7	15.4 ± 1.4	137.4 ± 9.0	2.78 ± 0.39	0.12 ± 0.01	1.589	0.08
L2 FIL	Ah	4.72 ± 0.29	42	20	38	5.26 ± 0.10	0.34 ± 0.01	18	49.3 ± 2.4	118.7 ± 4.2	7.82 ± 0.18	0.03 ± 0.01	0.964	0.14
	A/E	4.94 ± 0.11	50	25	25	5.08 ± 0.18	0.31 ± 0.01	19	21.4 ± 2.4	86.3 ± 4.1	4.53 ± 0.87	0.03 ± 0.01	1.260	0.12
	Bt	5.32 ± 0.32	65	25	10	0.90 ± 0.02	0.06 ± 0.01	21	39.2 ± 2.1	80.8 ± 6.9	4.07 ± 0.49	0.03 ± 0.01	1.261	0.10
E1 ErA	Ah	6.26 ± 0.04	35	25	40	2.39 ± 0.10	0.20 ± 0.01	14	159.1 ± 7.1	276.1 ± 20.8	13.17 ± 0.72	0.03 ± 0.01	1.184	0.21
	Bt	6.76 ± 0.25	52	23	25	2.38 ± 0.06	0.10 ± 0.01	27	63.9 ± 3.0	10.0 ± 1.0	4.98 ± 0.67	0.06 ± 0.01	1.277	0.40
	[Ap]	5.89 ± 0.50	45	22	33	3.32 ± 0.06	0.26 ± 0.01	15	65.0 ± 3.2	84.7 ± 7.1	4.4 ± 0.3	0.01 ± 0.01	1.299	0.17
	[Bt]	6.26 ± 0.40	35	27	38	0.36 ± 0.01	0.07 ± 0.01	6	101.8 ± 4.3	80.3 ± 8.7	2.97 ± 0.13	0.03 ± 0.01	1.520	0.16

As a result of prolonged ploughing and unsustainable land use, soils P1-P2 undergo active degradation, expressed in a decline of carbon concentration (3.45–4.26% for P1-P2) compared to a soil in the process of self-restoration (5.25% for L2) (see Suppl. material 5). The arable soils are characterised by the presence of a plough pan at a depth of about 30 cm, as well as over-compaction. Intensive ploughing has affected the zonal process of clay particle eluviation. For example, Luvis Chernozems (Loamic) (P1) exhibit a relatively high concentration of clay particles in the upper ploughing horizon (52.5%), while in the underlying horizon, the clay concentration drops to 37.5%. The obtained data are confirmed by the results of Chendev et al. (2020), who indicate that the conversion of arable soils into shelterbelts leads to a decrease in bulk density and clay content. This may be due to the formation of this soil in a microdepression, into which clay particles can be transported as a result of slope erosion. Depending on climatic features and land-use type, there are numerous mechanisms of degradation; however, they are

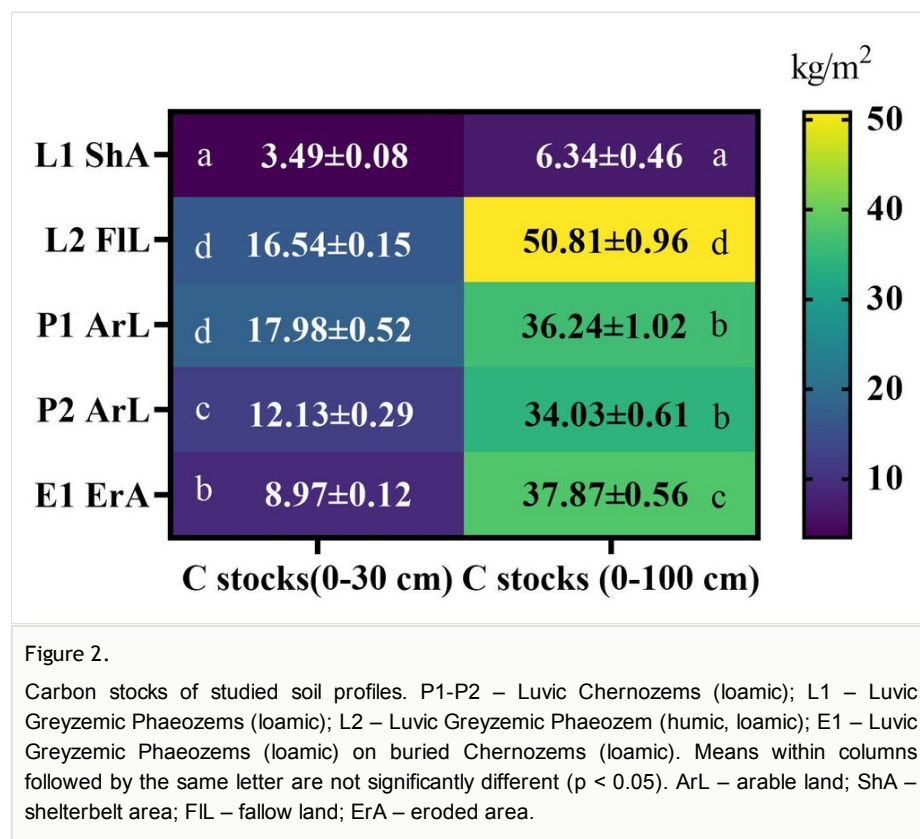
almost universally united by the loss of SOM, the restoration of which is a fundamental and separate challenge (Chendev et al. 2020). In Australia, for example, approximately 51% of carbon in the upper 10 cm soil layer has been lost over the past 40 years (Luo et al. 2010).

The soil forming in the shelterbelt area, Luvic Greyzemic Phaeozems (Loamic) (L1), represents the zonal variant of soil formation. Under conditions of high anthropogenic pressure, weakly transformed soils in the Tambov Region remain only in shelterbelt areas and on territories of nature reserves (Chendev et al. 2020). The soil pit preserves all genetic horizons (Ah-E-Bt), in contrast to agricultural soils, where the E horizon is absent. This soil is characterised by a relatively low carbon concentration ($3.367 \pm 0.133\%$), by the annual incorporation of carbon-rich crop residues into the ploughed layer of agricultural soils, which enhances organic matter accumulation. However, it shows the highest nitrogen concentration ($0.467 \pm 0.006\%$) amongst all studied soils, along with high levels of available potassium (374.68 ± 14.75 mg/kg K_2O), ammonium nitrogen (14.1 ± 0.1 mg/kg $N-NH_4^+$) and nitrate nitrogen (5.55 ± 0.03 mg/kg $N-NO_3^-$). However, it shows the highest nitrogen content ($0.467 \pm 0.006\%$) amongst all studied soils, along with high levels of available potassium (374.68 ± 14.75 mg/kg K_2O), ammonium nitrogen (14.1 ± 0.1 mg/kg $N-NH_4^+$) and nitrate nitrogen (5.55 ± 0.03 mg/kg $N-NO_3^-$) (see Suppl. material 6). The high content of exchangeable nitrogen forms indicates their active accumulation in the soil due to microbial activity. The transition of soils to a fallow state leads to their self-restoration towards zonal variants. Specifically, an eluviation process is initiated, which is presented in the re-distribution of the sand fraction along the profile and the formation of a transitional A/E horizon. An important feature of these soils is a decrease in density compared to agriculturally used soils, an increase in porosity and an increase in the biodiversity of soil fauna, which is reflected in the increased content of SOM.

Soil erosion is the third most important factor in arable land degradation and affects about 29.9% of all agricultural land, making this problem widespread worldwide (Panagos and Katsoyiannis 2019). In the vicinity of Tambov, the erosion site (E1) was formed artificially, initially as a fire channel, but, as a result of slope erosion, soil degradation occurred. A characteristic feature of this site is the presence of a buried old-arable horizon, on the surface of which Luvic Greyzemic Phaeozems (loamic) develop. It was noted that the buried Chernozems (loamic) were not affected by significant changes at the morphological level, the A/E transition horizon did not begin to develop here compared to its fallow analogues and the minimum carbon content amongst the studied soils was noted here ($2.390 \pm 0.100\%$ for Ah and $3.320 \pm 0.061\%$ for [Ap]). According to calculations by Maltsev and Yermolaev (2020), the territory of the Tambov Region, where agricultural lands predominate (60.3% of the region's area), can lose, on average, up to 0.45 kg/m² per year of soil due to erosion. This can lead to serious losses of SOM, which will affect the low-carbon development of the region (Burova et al. 2026).

The key indicators for assessing the sequestration potential of a territory are carbon and nitrogen stocks in the soil. The calculation of carbon stocks (see Fig. 2) was carried out

both up to the "active" zone (0-30 cm), which is more dependent on the characteristics of land use and for the "passive" zone (0-100 cm).



As a result of calculations of carbon stocks in the soil, it was revealed that the smallest stock ($3.49 \pm 0.08 \text{ kg/m}^2$ for 30 cm and $6.34 \pm 0.46 \text{ kg/m}^2$ for 100 cm) are characteristic of zonal Luvic Greyzemic Phaeozems (loamic) formed on the territory of the shelterbelt. It is explained by the contrasting genesis and land-use history of the studied sites. Unlike soils of arable lands, which receive annual inputs of crop residues and fallow soils, which accumulate organic matter under natural vegetation, the shelterbelt soil lacks regular organic inputs. The largest carbon stocks are typical for the soil of fallow lands ($16.54 \pm 0.15 \text{ kg/m}^2$ for 30 cm and $50.81 \pm 0.96 \text{ kg/m}^2$ for 100 cm), which was significantly different ($p < 0.05$) from the studied soil profiles. Over approximately 60 years of self-restoration, a significant accumulation of carbon has occurred, indicating that the abandonment of ploughing and the successional change of vegetation is a powerful mechanism for carbon sequestration. Similar results were obtained by Chendev et al. (2020), who note that the formation of soils in shelterbelts on former arable land leads to an increase in carbon stocks compared to soils used in agriculture. According to Kalinina et al. (2019), initial stage of post-agrogenic transformation is characterised by intensive accumulation of SOM. However, after the ecosystem reaches a certain age or the limit of its carbon storage capacity, accompanied by a transformation of the molecular composition of

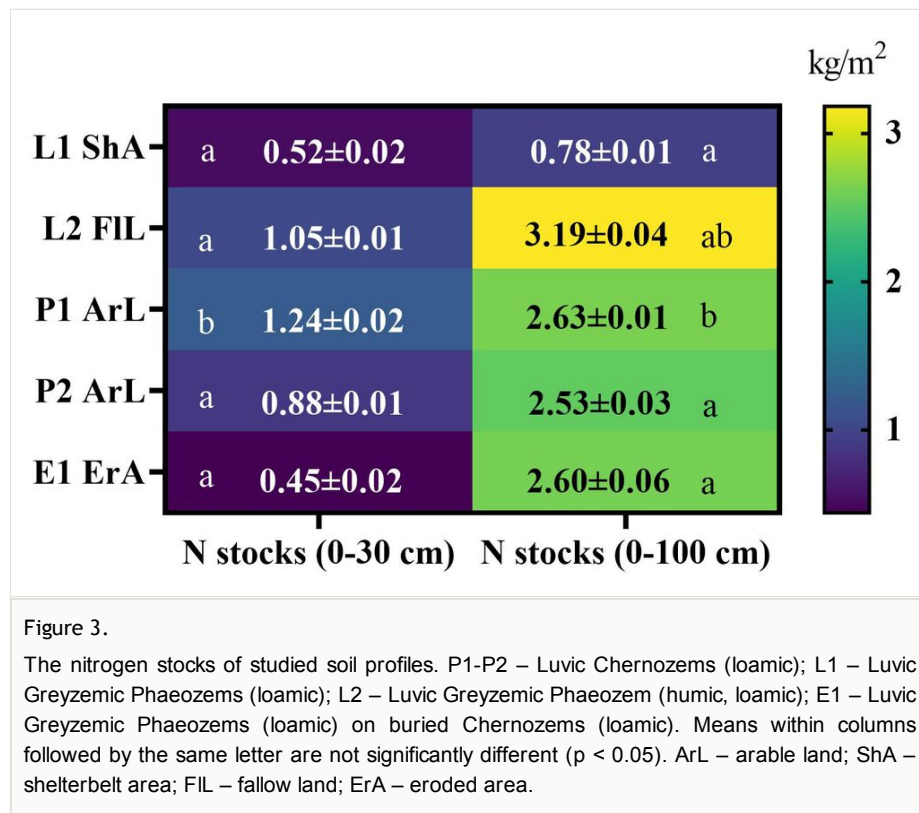
organic matter, SOM stocks may decline to background levels. This pattern is of critical importance for low-carbon development policies. This non-linear trend must be taken into account when developing low-carbon economy strategies. Accordingly, reclamation mechanisms should be aimed not only at stimulating carbon accumulation, but also at preserving already sequestered SOM. Carbon stocks in agricultural soils were relatively close to each other on a scale of 1 metre (36.24 ± 1.02 and 34.03 ± 0.61 kg/m²) and it had no significant differences. These stocks are lower than the values obtained for the soil of fallow lands (for a layer of 1 metre). It is important to note that the soils of agricultural lands may have a high sequestration potential in relation to natural Luvic Greyzemic Phaeozems (loamic). The eroded soil (E1) is characterised by relatively low C stocks in the 0-30 cm layer (8.97 ± 0.12 kg/m²), which is attributed to the slow formation of the modern Ah horizon. However, the 0-100 cm layer shows a relatively high C stocks value (37.86 ± 0.56 kg/m²), explained by the presence of a buried old-arable horizon.

According to various studies, agricultural land soils are considered a reliable carbon sequester (Poeplau et al. 2020, Fey and Wiermann 2025). At the same time, this potential can be increased by using various agricultural practices, namely irrigation methods, crop rotation changes and fertilisation (Amelung et al. 2020, Skadell et al. 2023). Land cultivation leads to carbon loss and not only the quantity, but also the quality of crops changes (Guo and Gifford 2002). This thesis is confirmed by our data; the conversion of soils to a fallow state leads to a significant ($p < 0.05$) increase in the content and stocks of SOM. However, it was noted that prolonged exposure of soils to a fallow state can lead to degradation of soils, because, as a result of vegetation change and the development of zonal features of soil formation, the transformation of soils towards zonal analogues will develop (Polyakov et al. 2025).

As a result of the calculation of nitrogen stocks (see Fig. 3), it was found that it generally repeats the dynamics of carbon stocks. Luvic Greyzemic Phaeozems (loamic) formed on the territory of the forest belt are characterised by a relatively low value of N stocks in the 1 m layer, which indicates the depletion of nitrogen in the soils. In front of this, the highest N stocks are typical for the soils of fallow lands (3.19 ± 0.04 kg/m²). The highest nitrogen stocks in the "active" layer of 0-30 cm are typical for the soil of agricultural lands (P1), which significantly differs ($p < 0.05$) from the studied soils. This is due to the use of organic forms of fertilisers and reclamation techniques. According to Koković et al. (2021), it was found that the use of fertilizers in combination with plant residues leads to long-term sequestration of carbon and nitrogen in soils, due to the activation of microbial biomass.

An important indicator of the quality of SOM is the C/N ratio. Based on this indicator, it was found that the C/N ratio of 14-15 is typical for arable horizons of soils P1–P2 and is characterised as an optimal wide range, which is due to the formation of biodegradation-resistant SOM. On the contrary, the Luvic Greyzemic Phaeozems (loamic) (L1) is characterised by a very narrow ratio of C/N – 7, which indicates the formation of a mobile organic material that is rapidly involved in the carbon cycle. According to Kovaleva et al. (2023), soils formed under shelterbelts in the Tambov Region are characterised by narrow C/N ratios (4–9), which is due to the development of eluvial–illuvial processes, as

well as a decrease in carbon and nitrogen concentration compared to arable soils. The soil of fallow lands has the widest and optimal C/N (15-16) ratio, active humification processes occurring here. The erosion site (E1) is characterised by a relatively wide ratio for horizons Ah and [Ap], 12 and 13, respectively.



Elemental composition of HAs of soils under the long-term agricultural use

The elemental composition of HAs is the most important indicator, which provides information on intramolecular oxidation/reduction processes, as well as on the structure of HAs molecules. Based on the analysis of the elemental composition of soil HAs, it was revealed that the studied preparations are characterised by a heterogeneous composition, which indicates the dynamics of intramolecular processes and varying degrees of transformation of organic matter (see Table 2).

Anthropogenic effects, namely agricultural practices, have a strong impact on the structure of HAs. HAs of soils used in agriculture (P1-P2) are characterised by a reduced carbon concentration (51.62-54.36%) and a predominance of oxygen (39.84-43.00%), indicating active intramolecular oxidation and the initial stages of degradation of soils. An increase in oxygen concentration is a characteristic stage of HAs maturation and can serve as an indicator of agrogenic transformation (Lodygin and Abakumov 2022). On the

contrary, the highest carbon concentration was observed in HAS samples of soils from fallow land (L2 Ah) and the eroded site (E1 Ah, [Ap]); however, the E1 Ah sample is characterised by a predominance of hydrogen in the molecular composition of HAS, which indicates a low maturity of SOM and its availability to microorganisms. HAS samples from soils of L2 fallow lands are characterised by a relatively low hydrogen concentration (3.48-3.61%), which indicates a high carbonisation of the structure of HAS molecules and is a characteristic stage of the transformation of SOM and the formation of biodegradation-resistant molecular complexes. For HAS of soil forming under the forest (L1), in addition to a relatively high hydrogen concentration, a relatively low value of the oxidation index ($w = 0.071 \pm 0.027$ for L1 Ah) was noted, which indicates a low maturity of organic molecules and zonal features of humification in Greyzemic Phaeozems.

Table 2.

The elemental composition (mean $\pm$ SD) of HAS isolated from studied soils. Each humic acid sample was analysed in duplicate. P1-P2 – Luvic Chernozems (loamic); L1 – Luvic Greyzemic Phaeozems (loamic); L2 – Luvic Greyzemic Phaeozem (humic, loamic); E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic). w - the degree of oxidation. ArL – arable land; ShA – shelterbelt area; FIL – fallow land; ErA – eroded area.

Soil ID* Land-use type	Horizon	C, %	N, %	H, %	O, %	C/N	H/C	O/C	w
L1 ShA	Ah	55.60 $\pm$ 0.33	2.88 $\pm$ 0.08	4.36 $\pm$ 0.08	37.19 $\pm$ 0.33	22.53 $\pm$ 0.77	0.933 $\pm$ 0.012	0.502 $\pm$ 0.007	0.071 $\pm$ 0.027
L2 FIL	Ah	58.28 $\pm$ 0.54	2.02 $\pm$ 0.13	3.61 $\pm$ 0.03	36.12 $\pm$ 0.63	33.77 $\pm$ 1.81	0.737 $\pm$ 0.013	0.465 $\pm$ 0.012	0.194 $\pm$ 0.012
	A/E	55.20 $\pm$ 0.08	2.01 $\pm$ 0.04	3.48 $\pm$ 0.11	39.34 $\pm$ 0.01	32.03 $\pm$ 0.66	0.750 $\pm$ 0.025	0.535 $\pm$ 0.001	0.320 $\pm$ 0.023
P1 ArL	Ap	54.36 $\pm$ 0.63	2.15 $\pm$ 0.10	3.65 $\pm$ 0.10	39.87 $\pm$ 0.64	29.49 $\pm$ 1.67	0.799 $\pm$ 0.013	0.551 $\pm$ 0.015	0.302 $\pm$ 0.044
P2 ArL	Ap	51.62 $\pm$ 0.25	2.01 $\pm$ 0.04	3.38 $\pm$ 0.06	43.03 $\pm$ 0.23	30.02 $\pm$ 0.41	0.779 $\pm$ 0.018	0.626 $\pm$ 0.006	0.473 $\pm$ 0.005
E1 ErA	Ah	58.48 $\pm$ 0.06	3.07 $\pm$ 0.01	4.38 $\pm$ 0.06	34.10 $\pm$ 0.01	22.23 $\pm$ 0.08	0.891 $\pm$ 0.013	0.438 $\pm$ 0.001	-0.015 $\pm$ 0.012
	[Ap]	57.97 $\pm$ 0.96	2.17 $\pm$ 0.04	3.80 $\pm$ 0.32	36.09 $\pm$ 1.24	31.11 $\pm$ 1.05	0.778 $\pm$ 0.053	0.468 $\pm$ 0.024	0.157 $\pm$ 0.100

An important parameter characterising intramolecular processes and the degree of maturity of HAS is the H/C-O/C ratio (see Fig. 4). The directional lines on the diagram indicate the theoretical pathways of hydrogenation/dehydrogenation and reduction/oxidation processes. These shifts reflect the processes of structural condensation and maturation of HAS during the transformation of SOM.

According to the diagram obtained, it was noted that HAS of soils affected by erosion (E1 Ah) and forest soils (L1 Ah) are characterised as the least mature, which is due to the predominance of aliphatic structural fragments. HAS of soils of agricultural lands (P1-P2)

are in the process of maturation, condensation processes are actively taking place here; however, relatively high O/C values indicate their relatively high oxidation state. At the same time, this trend is typical for most HAs of soils of agricultural lands, due to the peculiarities of fertilisation and land use (Lodygin and Abakumov 2022). According to the oxidation index (w), these samples are characterised by the presence of carboxylic aromatic fragments ($w = 0.302\text{--}0.468$), which indicates a low degree of degradation of SOM. HAs of soils of fallow lands (L2) are characterised by a relatively low ratio of H/C and O/C, indicating their mature structure. As a result of the transition to the fallow state, condensation of the most stable molecular complexes in the environment occurred. The same trend is typical for the buried old-arable horizon E1 [Ah]; here, the selection of the most biodegradation-resistant molecular complexes could occur. The L2 A/E transition horizon is characterised by a slight shift to the oxidation region, which is caused by active humus-illuvial processes.

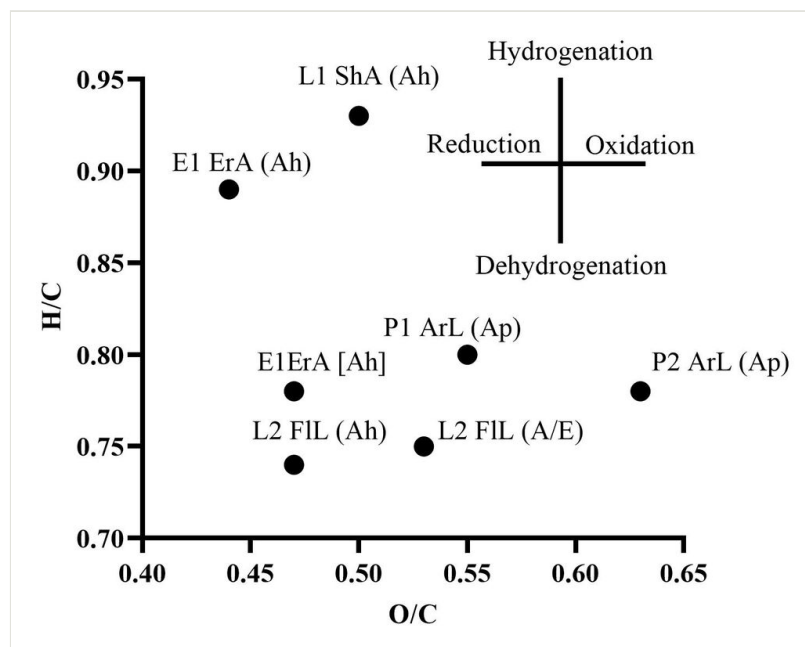


Figure 4.

Distribution fields of elemental composition indices in H/C-O/C coordinates of HAs. P1-P2 – Luvis Chernozems (loamic); L1 – Luvis Greyzemic Phaeozems (loamic); L2 – Luvis Greyzemic Phaeozem (humic, loamic); E1 – Luvis Greyzemic Phaeozems (loamic) on buried Chernozems (loamic). ArL – arable land; ShA – shelterbelt area; FIL – fallow land; ErA – eroded area.

Our results are confirmed by the data of Banach-Szott et al. (2021); a decrease in the H/C ratio reflects the degree of condensation of organic matter, while the lowest values were typical for Chernozems and the highest for HAs of zonal forest soils, which is reflected in our study. HAs of agricultural soils are characterised by a high degree of maturity compared to HAs of natural ecosystems, due to the conditions of origin of HAs, the type of

land use and the quality of humification precursors (Sánchez-Monedero et al. 2002; Dergacheva et al. 2012). For the O/C index and the degree of oxidation (w), it was noted that the highest ratio is typical for HAs of Chernozems in arable lands, which indicates the oxidised nature of HAs molecules (Banach-Szott et al. 2021).

Molecular composition of HAs isolated from soil under long-term agricultural use

The molecular composition of HAs is an important parameter necessary for analysing the structure and assessing the stability of SOM in the ecosystem. The ^{13}C NMR spectra are presented in Fig. 5.

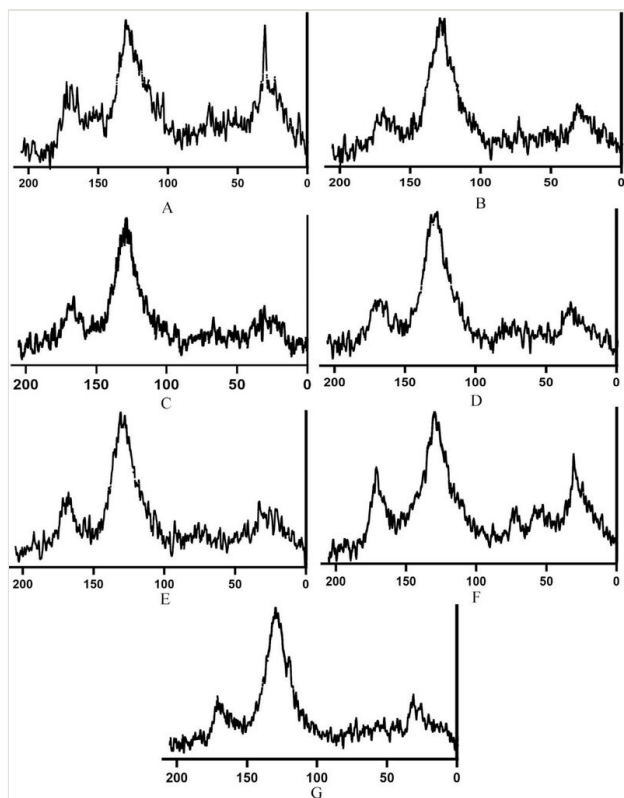


Figure 5.

^{13}C NMR spectra of HAs isolated from studied soils of Tambov Region. Ox – Chemical shifts, ppm. Oy – Intensity. P1-P2 – Luvic Chernozems (loamic) arable land; L1 – Luvic Greyzemic Phaeozems (loamic) shelterbelt area; L2 – Luvic Greyzemic Phaeozem (humic, loamic) fallow land; E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic) eroded area.

Based on ^{13}C NMR spectroscopy, the content of various structural fragments of HAs was calculated (see Table 3).

Table 3.

Concentration of structural fragments in the studied soil samples. P1-P2 – Luvic Chernozems (loamic); L1 – Luvic Greyzemic Phaeozems (loamic); L2 – Luvic Greyzemic Phaeozem (humic, loamic); E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic). 0-46 ppm – Alkyl C; 46-60 ppm – O,N-Alkyl; 60-105 ppm – CH₂OH, CHOH; 105-144 ppm – Arom C; 144-164 ppm – O,N-Arom; 164-183 ppm – COO-R; 183-190 ppm – Arom=O; 190-204 ppm – C=O. AR - sum of aromatic fragments; AL - sum of aliphatic fragments; AL h,r + AR h,r, % - degree of hydrophobicity; C,H-AL/O,N-AL - degree of humification of SOM. ArL – arable land; ShA – shelterbelt area; FIL – fallow land; ErA – eroded area.

Soil ID Landuse type	Hor.	Chemical shifts (ppm), % of ¹³ C										AR/ AL	AL h,r +AR h,r	C,H - AL/ O,N -AL
		0-46	46-60	60-105	105-144	144-164	164-183	183-190	190-204	AR	AL			
L1 ShA	Ah	26	5	6	44	8	9	1	1	53	47	1.13	70	2.36
L2 FIL	Ah	16	2	5	60	7	7	2	1	69	31	2.23	76	2.28
	A/E	12	2	6	59	9	10	1	1	69	31	2.23	71	1.50
P1 ArL	Ap	12	2	7	58	9	10	1	1	68	32	2.13	70	1.33
P2 ArL	Ap	15	1	6	59	7	10	1	1	67	33	2.03	74	2.14
E1 ErA	Ah	23	6	9	41	8	11	1	1	50	50	1.00	64	1.53
	[Ap]	16	3	5	59	8	7	1	1	68	32	2.13	75	2.00

According to the data obtained, it was revealed that the main functional groups in the HAs are the hydrophobic group Alkyl C (0-46 ppm), Arom C (105-144 ppm) and the hydrophilic group COO-R (164-183 ppm). Most of the studied HAs are characterised by the predominance of aromatic structural fragments, with the exception of HAs of the modern humus horizon E1 Ah, where the ratio of aliphatic and aromatic structural fragments is the same. It was found that the highest concentration of aromatic structural fragments corresponds to the HAs of the soils of fallow lands (L2 Ah-A/E), the AR/AL ratio being 2.23, which is confirmed by the analysis of the elemental composition and the H/C ratio. The predominance of aromatic structural fragments indicates a relatively high degree of resistance of SOM to biodegradation and its maturity. According to Shein et al. (2011), the quality of the SOM of the fallow Chernozems, as well as the physico-chemical properties is higher, which confirms our study. The relatively high concentration of aromatic structural fragments is primarily due to the advanced degree of humification and the selective preservation of recalcitrant lignin-derived and condensed aromatic compounds originating from the precursors of humification. Farkhodov et al. (2025) stated that prolonged ploughing leads to a decrease in the content of groups of alcohols, esters and carbohydrates in the HAs, which leads to a decrease in the aliphatic periphery. Our previous work (Suleymanov et al. 2022) showed that HAs of SOM that is exposed to water erosion may be characterised by a relatively high concentration of aromatic structural fragments. This is due to removing the oxygen-containing functional groups

which leads to its long-term conservation of SOM. The relatively low value of aromatic fragments is due to the oxidative degradation of SOM and an increase in the proportion of carboxylic ($-\text{COO-R}$) HAs fragments. This can be caused by the active agricultural influence (after harvesting, plant residues are ploughed back into the soil), as well as the input of organic forms of fertilisers, which leads to an increase in the concentration of aliphatic structural fragments. The lowest AR/AL ratio (1.00) was found in the HAs of the modern humus horizon of eroded soil (E1 Ah), which indicates a weak degree of transformation of HAs and the predominance of humification precursors rather than products of a "secondary" nature in the HAs structure. This is reflected in the relatively low concentration of aromatic structural fragments (41%, the lowest amongst the studied HAs), as well as the high concentration of hydrophilic groups of structural fragments ($-\text{CH}_2\text{OH}$, CHOH , $-\text{COO-R}$). A special place is occupied by the HAs of the forest soils (L1 Ah), the AR/AL ratio is relatively low (1.13) and it is the characteristic stage of SOM transformation in the forest zone under this type of climate. The low proportion of aromatic structural fragments may be due to the initial quality of humification precursors and the duration of their transformation. According to Banach-Szott et al. (Banach-Szott et al. 2019), the quality of forest litter largely influences the quality of SOM. Thus, in the study by Banach-Szott et al. (Banach-Szott et al. 2019), it was noted that, in soils under zonal tree species (*Quercus robur* L. and *Picea abies* L. Karsten), the composition of HAs was characterised by a higher proportion of aliphatic structural fragments than under thuja litter (*Thuja plicata* D. Don ex Lamb.), which is planted for decorative purposes. The same trend was noted for HAs of Chernozems, which also seems to indicate a zonal variant of soil formation (Kholodov et al. 2011). The HAs sample of forest soils is characterised by a maximum concentration of the hydrophobic Alkyl C group, as well as a relatively low concentration of hydrophilic structural fragments ($-\text{COO-R}$). Thus, we can note that the transition of soils to a fallow state is characterised by an increase in the maturity of SOM. For the L2 A/E transition horizon, there is a decrease in the concentration of hydrophobic aliphatic fragments (by 4%, relative to the overlying L2 Ah), as well as an increase in hydrophilic structural fragments (by 3%, relative to the overlying L2 Ah); this may indicate the initial process transformation of SOM towards zonal variants and decline of resistance of SOM to biodegradation. Agriculture leaves a strong imprint on the structural organisation of HAs of soils, which is expressed both in an increase in the proportion of aromatic structural fragments due to active humification processes and an increase in the mobility and solubility of HAs due to the presence of hydrophilic oxygen-containing groups ($-\text{COO-R}$). Various types of land cultivation, transition to a fallow state and erosion have a serious impact on the molecular structure of soils; this is due to ploughing, land degradation and fertilisation (Vishnyakova and Ubugunov 2023).

To analyse the humification processes and the rate of hydrophobicity, a diagram of integral indicators of HA was prepared (see Fig. 6).

According to the graph obtained, it was revealed that the largest proportion of carbon entering the hydrophobic part of HAs molecules is characteristic of SOM formed in fallow lands, which indicates its high turnover time in the environment. Similarly, to this HAs sample, a high level of hydrophobicity is typical for HAs of arable soil (P2 Ap), as well as

buried old-arable horizon (E1 [Ah]). At the same time, HAs of the forest soil is characterised as less hydrophobic, with a relatively high level of hydrophilic compounds and aliphatic structural fragments, which leads to a rapid turnover of SOM in the environment.

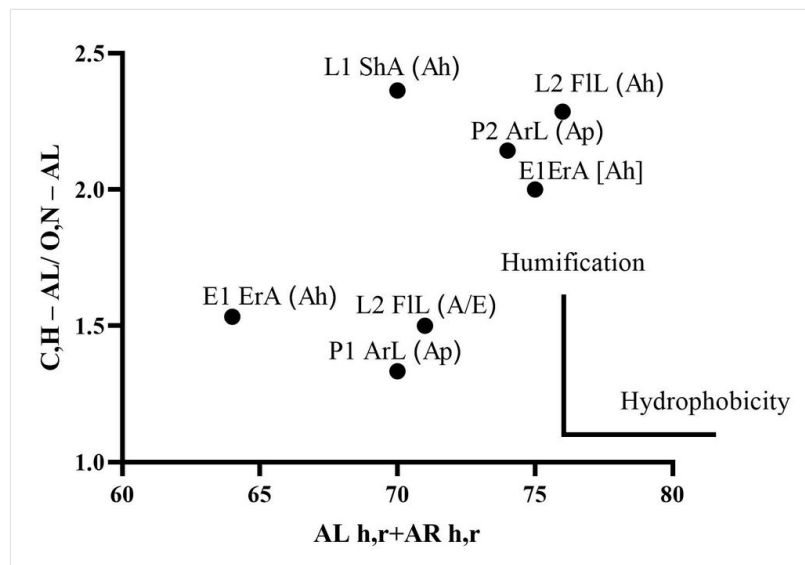


Figure 6.

Diagram of integral indices of molecular composition of studied HAs. P1-P2 – Luvic Chernozems (loamic); L1 – Luvic Greyzemic Phaeozems (loamic); L2 – Luvic Greyzemic Phaeozem (humic, loamic); E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic). ArL – arable land; ShA – shelterbelt area; FIL – fallow land; ErA – eroded area.

As a result of the analysis, we confirm the main hypothesis of the study, agriculture leads to an increase in the stable carbon pool in the soil compared to the zonal variants, but it is characterised by an oxidised state. The most stable forms of humus are formed as a result of the transition to a fallow state, which leads to the conservation of SOM. HAS samples of the forest soil and modern eroded soil are characterised as the least mature, this is due to both the relatively young state (E1 Ah) and the zonal features of humus formation (L1 Ah). Our data show that the transition from arable land to fallow state leads to an increase in both the stocks and the resistance of SOM to biodegradation. According to the obtained data, a clear distinction was revealed between modern and buried HAs in the eroded profile (E1), which provides direct vision into the timescales of humification. The data show that, even as a result of erosion, buried SOM can stay in stable form, while newly-formed SOM requires decades to achieve a similar level of stability.

Limitation of study

This study is not intended to extrapolate quantitative data to the entire forest-steppe zone, but to identify molecular mechanisms of HAs transformation under different types of land

use. In this study, soil bulk density data obtained in a single replicate were used to calculate SOC and nitrogen stocks, which may lead to underestimation or overestimation of carbon and nitrogen stocks due to the high heterogeneity of anthropogenically transformed soils. According to Fowler et al. (Fowler et al. 2023), the use of a single replicate for bulk density measurement at a depth of up to 30 cm can introduce up to 17% variation in soil carbon and nitrogen stock values. Triplicate measurements for the studied physicochemical parameters and HAs were performed under laboratory conditions. The analysis of a limited number of well-characterized model sites is a widely accepted approach in genetic soil science for studying qualitative changes in organic matter (Kholodov et al. 2011, Chimitdorzhieva et al. 2026). Nevertheless, this study was conducted on a limited number of soil profiles ($n = 5$), each representing a specific land-use type within a single proposed carbon polygon. While this sample size allows for in-depth molecular characterisation of contrasting soil types, it does not permit broad statistical generalisation across the entire forest-steppe zone. The results should, therefore, be considered as preliminary evidence requiring validation on larger and spatially distributed datasets in future studies.

Conclusions

Based on the conducted research, it was revealed that the soils of agricultural lands may have a high potential for carbon sequestration. This is due to the accumulation of a significant ($p < 0.05$) amount of SOM (up to $36.24 \pm 1.02 \text{ kg/m}^2$ for P1 0-100 cm) compared with background forest soils ($6.34 \pm 0.46 \text{ kg/m}^2$ for L1 0-100 cm). HAs of agricultural soils (P1-P2) are characterised by a relatively high concentration of aromatic structural fragments (the AR/AL ratio reaches 2.13). However, as a result of tillage practices and applying organic fertilisers, there is a relatively high concentration of hydrophilic carboxylic structural fragments (up to 10% – COO-R) which indicates the slightly degraded nature of the SOM. It was found that the maximum C-stocks correspond to the soils of fallow lands, accumulating up to $50.81 \pm 0.96 \text{ kg/m}^2$ for L2 0-100 cm. The molecular composition of HAs of soils of fallow lands (L2) showed a maximum concentration of aromatic structural fragments (AR/AL – 2.23) and a relatively low concentration of hydrophilic groups of structural fragments, indicating the stabilisation of SOM. It has been revealed that erosion has a significant impact on both buried and modern SOM. Soils subject to erosion were characterised by relatively high C-stocks (up to $37.86 \pm 0.56 \text{ kg/m}^2$ for Er1 0-100 cm), such a high value being due to the presence of a buried old-arable horizon. The HAs of the buried organic material (E1 [Ah]) were characterised by a relatively high level of resistance to biodegradation (AR/AL – 2.13), while modern SOM (E1 Ah) was characterised by a weak degree of maturity and condensation (AR/AL – 1.00, H/C – 0.89). The zonal variant of Luvic Greyzemic Phaeozems (loamic) (L1) was characterised by the lowest stocks of carbon and nitrogen, 6.34 and 0.78 kg/m^2 , respectively, as well as a relatively low degree of maturity of SOM (AR/AL – 1.13, H/C – 0.93), which is due to the zonal type of soil formation and is characteristic of the forest-steppe zone of Tambov Region.

Future work will focus on studying the heterogeneity of soil formation across the proposed carbon polygon, including spatial and temporal variability in soil properties and on assessing its carbon sequestration potential in order to evaluate the site's suitability for carbon monitoring and long-term carbon storage.

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Author contributions

Vyacheslav Polyakov – Conceptualisation, methodology, software, validation, investigation, data curation, writing—original draft preparation, visualisation.

Maxim Kulikovskiy – Conceptualisation, investigation, writing—review and editing, supervision, project administration.

Mikhail Bukovsky – Validation, investigation, writing—review and editing, data collection.

Egor Artukhov – Investigation, literature search, data collection.

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Svetlana Chebotaryova – Formal analysis, literature search, data collection, data analysis.

Anastasiia Muraveva - Formal analysis, literature search, data analysis.

Evgeny Abakumov – Conceptualisation, methodology, validation, resources, writing—review and editing, supervision, project administration, funding acquisition.

Conflicts of interest

The authors have declared that no competing interests exist.

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Supplementary materials

Suppl. material 1: The morphological structure of studied soils

Authors: Vyacheslav Polyakov

Data type: Morphological

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Suppl. material 2: Soils formed on proposed carbon polygon of Tambov Region

Authors: Vyacheslav Polyakov

Data type: Images

Brief description: The soils formed on proposed carbon polygon of Tambov Region. P1-P2 – Luvic Chernozems (loamic) arable land; L1 – Luvic Greyzemic Phaeozems (loamic) shelterbelt area; L2 – Luvic Greyzemic Phaeozem (humic, loamic) fallow land; E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic) eroded area.

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Suppl. material 3: Chemical soil properties

Authors: Vyacheslav Polyakov

Data type: Raw data

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Suppl. material 4: Raw data for NMR

Authors: Vyacheslav Polyakov

Data type: Raw data

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Suppl. material 5: Carbon content of studied topsoils and buried soil horizon

Authors: Vyacheslav Polyakov

Data type: Data

Brief description: The carbon content of studied topsoils and buried soil horizon. Means within columns followed by the same letter are not significantly different ($p < 0.05$). P1-P2 – Luvic Chernozems (loamic) arable land; L1 – Luvic Greyzemic Phaeozems (loamic) shelterbelt area; L2 – Luvic Greyzemic Phaeozem (humic, loamic) fallow land; E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic) eroded area.

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Suppl. material 6: Figure S3

Authors: Vyacheslav Polyakov

Data type: Data

Brief description: The content of biogenic elements of studied topsoils and buried soil horizon. Means within columns followed by the same letter are not significantly different ($p < 0.05$). P1-P2 – Luvic Chernozems (loamic) arable land; L1 – Luvic Greyzemic Phaeozems (loamic) shelterbelt area; L2 – Luvic Greyzemic Phaeozem (humic, loamic) fallow land; E1 – Luvic Greyzemic Phaeozems (loamic) on buried Chernozems (loamic) eroded area.

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