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Soil enzymatic responses to long-term fallowing in Southern Taiga Forests

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Abstract

The largest area of land in Russia is located in the fallow state, there is a change in plant communities, physico-chemical parameters of soils and changes in the enzymatic activity of soils. To analyze the condition of fallow and undisturbed soils, we studied different-aged changes in the main physico-chemical parameters of soils, analyzed the features of morphological structure of soils, and also studied the enzymatic activity of soils of such classes of enzymes as hydrolase and oxidoreductase. Sampling was carried out from the upper humus-accumulative horizons of 13 soil sections of the Leningrad and Novgorod regions of Russia. As a result of research, it was revealed that transition of lands to fallow state leads to transformation of soils towards zonal series of soils. Soil transformation is accompanied by a decrease in pH value, content of biogenic elements, with an increase in the content of carbon and biogenic elements in old-age plots. The study of enzyme activity in soils showed that the activity of the studied enzymes at different sites varies differently, depending on land use. Significantly higher activity of oxidoreductases class was noted for soils in which transformation of wood residues takes place and O horizon is formed. A comparative assessment of the biological activity of the studied soils was given using the indicator of total relative enzymatic activity (indicator representing the total biochemical activity of soil based on enzyme analysis). According to the comparative assessment of soil biological activity, it was found that the biological activity increases with increasing time of soils being in fallow state. Thus, to restore soil biochemical activity and agroecosystem stability, long (30-year) fallow periods with secondary forest formation should be maintained, which provides neutral pH, organic carbon accumulation, and maximum enzymatic activity superior to both recently abandoned and arable lands.

Keywords: Hydrolase, North-West of Russia, Oxidoreductase, Podzols, Retisols.

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Introduction

Worldwide, the area of abandoned land is 220 million hectares, of which Russia accounts for almost 20% (Lurie et al., 2010). This high percentage of abandoned land was caused by the economic depression that occurred in Russia in the early 1990s, due to political factors. Therefore, since the early 1990s, there has been a stable trend in the country's agriculture to remove previously sown arable land from active agricultural rotation (Nechaeva, 2023). Some of these lands were built up by cities and industrial facilities. But the largest part, about 91% of arable land in Russia, after being abandoned underwent a process of restoration (Lurie et al., 2010). Being a favorable environment for the restoration of natural landscapes, post-agricultural lands began to develop in the direction of formation of natural ecosystems, given the natural climatic zone and overgrow meadow, shrub and woody vegetation (Sorokina, 2016; Dmitriev and Lednev, 2016; Telesnina, 2017).

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Abandonment of agricultural land is an increasingly important problem in European countries (van der Sluis et al., 2014; Gabarrón-Galeote et al., 2015; Vacquie et al., 2015; Ustaoglu and Collier, 2018). Very acute, this issue is also in China, where large-scale agricultural land abandonment is taking place. According to the authors (Liu and Li, 2017), this is due to the fact that, as a result of China's ongoing rapid urbanization, an increasing number of rural workers are moving to urban areas, significantly reducing the rural agricultural workforce and contributing to farmland abandonment. Thus, the relevance of a comprehensive study of post-agricultural land processes is determined by the large scale of their distribution and high potential of their cultivation (Shchukin et al., 2018; Kalinina et al., 2018).

Since in parallel with the restoration of zonal vegetation, in the course of post-agrogenic evolution there is a change in their biological properties (Ovsepyan et al., 2017; Kurganova et al., 2021), the need to study the enzymatic activity of fallow soils is beyond doubt. According to various authors (Valkov et al., 1999; Zvyagintsev, 2005), enzymatic activity reflects the intensity of biological, physiological, biochemical processes occurring in the soil and is considered as a derivative of the combination of abiotic, biotic and anthropogenic factors of soil formation (Sun et al. 2022; Peng et al., 2024). Therefore, the biological activity of soil is crucial importance in the process of soil formation, development and degradation. The most frequently used indicator of potential biological activity is the soil enzymatic activity (Galstyan, 1974; Gorbtsova et al., 2017; Gedgafova et al., 2023).

Some of the most commonly considered enzymes are the classes of hydrolases (invertase, phosphatase, urease) and oxidoreductases (dehydrogenase, catalase). Dehydrogenases (DHA) is an indicator of oxidative metabolism in soil and microbial activity. The functioning of dehydrogenases is related to many biochemical processes in soil, including greenhouse gas emissions. Catalase (CAT) is an enzyme that catalyzes the decomposition of hydrogen peroxide into water and oxygen (Lemanowicz et al., 2020). Some of the most commonly studied hydrolytic enzymes are phosphatase (PHA), urease (Ure) and invertase (INV). PHA enzyme catalyze the hydrolysis of phosphoric acid esters and phosphoric acid anhydrides with the release of soluble phosphate from organic phosphorus and improvement of the phosphorus cycle. Thus, PHA activity can be used as an indicator of inorganic phosphorus availability to plants and microorganisms. Another hydrolytic enzyme Ure catalyze the hydrolysis of urea to CO_2 and NH_3 and helps to control soil quality as influenced by management practices and nitrogen content after urea application (Adetunji et al., 2017). Another hydrolytic enzyme INV is often used as an indicator of carbon cycling, which cleaves carbohydrate polymers, releases simpler sugars, thereby increasing soluble nutrient content of soil, mediates carbon transformations and produces major energy sources for soil microorganisms (Sardans et al., 2008). Soil enzymes affect essential soil processes such as plant decomposition, mineralization of soil organic matter, and release of nutrients into the soil through decomposition of organic residues and microbial activity (Dotaniya et al., 2019; Sobucki et al., 2021). Enzymes can be used to assess changes in the microbial community due to environmental changes or anthropogenic activities such as land use changes (Vikram et. al., 2024). It is known that due to the rapid response of the enzyme pool to many environmental changes, enzymatic activity is detected well before changes in other soil quality indicators (Zhang et al., 2015). The hypothesis of this work is that there will be a change in soil enzyme activity as a result of land use change and transition to fallow state. This study aimed to evaluate enzymatic activity across fallow soils of different ages in the Southern Taiga, and to apply a relative assessment approach.

Material and Methods

Study area

This study was carried out in the northwestern region of Russia, as follows Leningrad and Novgorod regions. The research area is located within the final moraine zone of the Valdai glaciation, characterized by widespread moraine deposits. The study areas are located on the plain and characterized by a low elevation difference of about 20 meters.

The region located within the southern taiga bioclimatic zone, where Podzols and Retisols dominate well-drained watersheds, while Histosols and Gleysols prevail in waterlogged areas. Climatic conditions feature an average annual precipitation of 587 mm, with evapotranspiration reaching around 430 mm. The mean annual temperature is 4.3 °C.

The Ban'kovo village area (Leningrad region) exhibits an extended agricultural history, with fallow soils ranging in age from 40 to 120 years. All investigated soils developed on uniform parent material - water-glacial sandy loams with puddled structure, underlain by red-brown moraine loams at a depth of 70-80 cm. The natural vegetation of the study area is a pine-birch-blackberry forest.

Soils of 20-year-old fallow, agricultural and undisturbed lands were studied in the area of Belogorka village (Leningrad region). The relief of this territory is characterized by relatively leveled surface. Soil-forming rocks are represented by local red-colored moraines with significant admixture of Devonian rocks. The study area belongs to the southern taiga subzone. Pine forests with admixture of spruce, birch and aspen prevail.

In the area of Borovichi city (Novgorod region), re-involved 30-year-old soils of fallow lands, agricultural soils and undisturbed soils were investigated. The study plots are located at a distance of no more than 700 m from each other and represent a single agro-landscape of the former agro-holding. The study areas are presented in Figure 1.

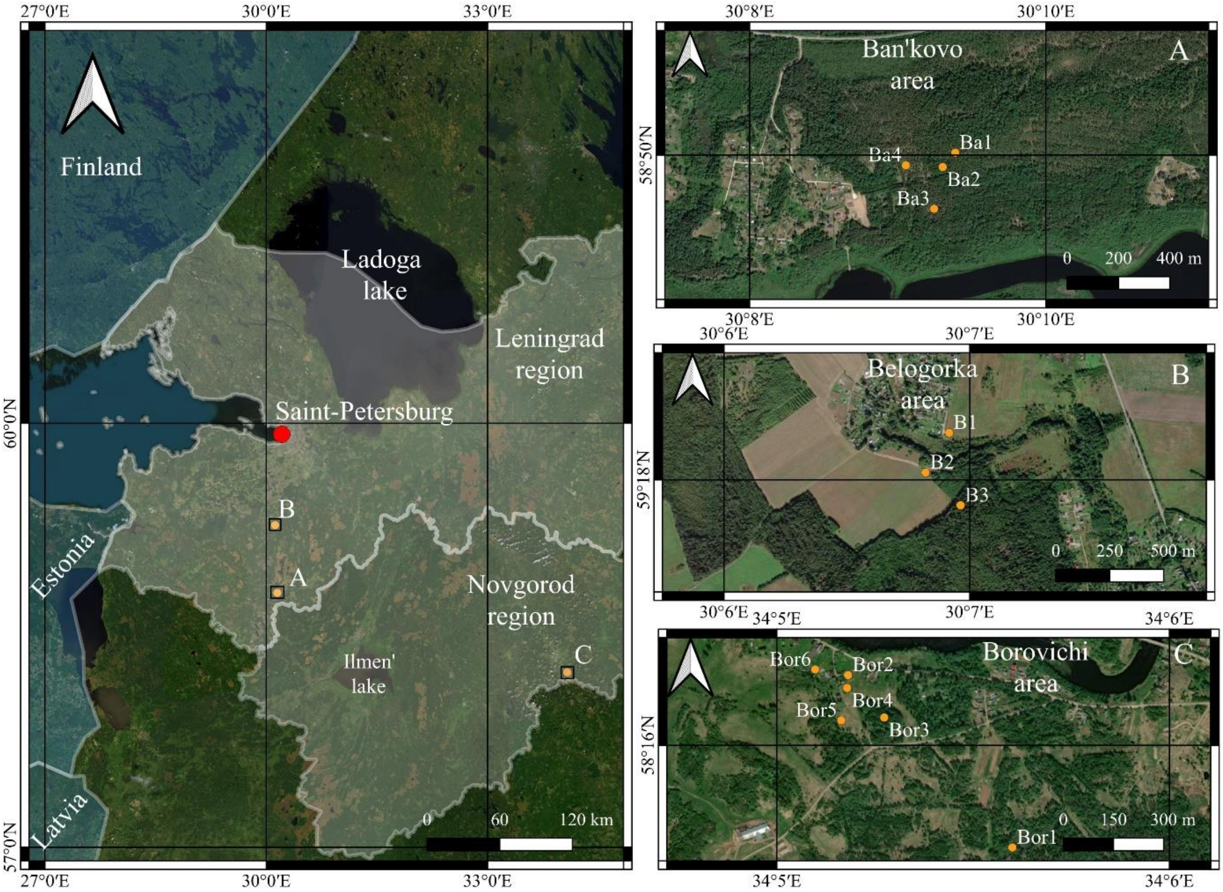


Figure 1. Location of the study area in the north-west of Russia

The selection of data is determined by the scope of soils of fallow lands formed on sandy parent materials (Ban'kovo), as well as silt-clay materials (Belogorka). Such diversity allows us to consider the features of transformation of the two leading soil types of the taiga-forest zone. The site in the Novgorod region (Borovichi) is characterized by soil formation as a result of transition to fallow state 30-years-ago, as well as by different type of agricultural use.

Differences in soil formation conditions and type of agricultural use determine the diversity of studied soils. The use of Podzols and Retisols in agriculture has a significant impact on the formation and biological indicators of soils, which are subject to ongoing changes and therefore require close attention and comprehensive study. Description of the studied soils are presented in Table 1.

Methods

Soil samples were collected from the key work sites to analyze physico-chemical and biological parameters of soils. Sampling was carried out from all studied soil horizons in order to trace the dynamics of enzymatic activity in soils of fallow lands and reclaimed fallow soils. The depth of sampling ranged from 0 to 70 cm and depended on the presence in the soils of the processes associated with the transition to fallow state. The comparison of the main physicochemical parameters and enzymatic activity was carried out for the upper humus-accumulative horizons (0-30 cm). The relatively large scale of the study allowed to identify different stages of soil formation in fallow and re-involved soils. Soils were classified according to the international classification IUSS WRB (2022).

Table 1. Description of the studied soil profiles

Soil ID	Horizon*	Depth, cm	Description	Location	Coordinates	Soil name**
Ba1	E_1	4-10	Mineral horizon with features of loss of silicate clay and iron, light color	Benchmark forest, undisturbed soil	N58.832090 E30.153881	Stagnic Podzol (arenic)
	E_2	10-20	Mineral horizon with features of loss of silicate clay and iron			
Ba2	O_e	0-3	Organic material consisting brganic material consisting by moderate decomposed organic residues	120 years old fallow lands	N58.831588 E30.153031	Plaggic Podzol (arenic)
	A_p	3-28	Mineral horizon with the accumulation of humified organic matter			
Ba3	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	80 years old fallow lands	N58.830129 E30.152454	Plaggic Podzol (arenic)
	B_s	30-70	Mineral horizon with illuvial accumulation of sesquioxides			
Ba4	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	40 years old fallow lands	N58.831647 E30.150548	Plaggic Podzol (arenic)
	B_s	30-70	Mineral horizon with illuvial accumulation of sesquioxides			
B1	A_p	2-34	Mineral horizon with the accumulation of humified organic matter	20 years old fallow lands	59.305955 E30.118322	Plaggic Retisol (loamic)
	E	34-65	Mineral horizon with features of loss of silicate clay and iron			
B2	A_p	0-42	Mineral horizon with the accumulation of humified organic matter	Agriculture lands	N59.304304 E30.116394	Plaggic Retisol (loamic)
	B_t	42-70	Mineral horizon with illuvial accumulation of silicate clay			
B3	A/E	5-25	Transitional mineral horizon with features of accumulation of humified organic matter and loss of silicate clay	Benchmark forest, undisturbed soil	N59.302943 E30.119252	Retisol (loamic)
	B_t	25-50	Mineral horizon with illuvial accumulation of silicate clay			
Bor1	A_h	0-30	Mineral horizon with the accumulation of humified organic matter	Benchmark forest, undisturbed soil	N58.265268 E34.091997	Podzol (arenic)
Bor2	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	30 years old fallow lands used as garden	N58.269883 E34.083628	Plaggic Podzol (arenic)
Bor3	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	30 years old fallow lands, secondary forest	N58.268745 E34.085468	Plaggic Podzol (arenic)
Bor4	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	30 years old fallow lands used as pasture	N58.269535 E34.083580	Plaggic Podzol (arenic)
Bor5	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	30 years old fallow lands used as hayfields	N58.268671 E34.083279	Plaggic Podzol (arenic)
Bor6	A_p	0-30	Mineral horizon with the accumulation of humified organic matter	130 years old agriculture lands	N58.270034 E34.081949	Plaggic Podzol (arenic)

* Jahn et al. (2006) ; ** IUSS WRB. (2022)

The content of carbon and nitrogen was determined on CHN analyzer, pH was determined by potentiometric method. Particle-size distribution of soils was determined according to Bowman and Hutko (2002). The FAO recommended method (FAO, 2023) was used to estimate microbial activity, basal respiration (BR). The enzymatic activities (PHA, INV, URE and DHA) was determined by colorimetric method, CAT - gasometrically according to Galstyan's methods modified by Kazeev (Kazeev et al. 2012). The analog methods are presented in the work of Guan et al. (1986). During the analysis of urease, it was noted that its activity was below the measured level, therefore, it was not included in further calculations of total relative enzymatic activity. This is due to the low activity of urease in soils that have been subjected to anthropogenic influence relatively recently. The physicochemical parameters and enzymatic activity were analyzed in three repetitions.

To calculate the total relative enzymatic activity (TREA) (Kazeev et al., 2003) of soils, the maximum value of each of the indicators in the sample is taken as 100% and the value of the same indicator in the others samples is represented as a percentage relative to it (1):

$$B1 = \left(\frac{Bx}{Bmax}\right) * 100\% \tag{1}$$

where, B1 - relative score of the indicator;
Bx - actual value of the indicator;
Bmax - maximum value of the indicator.

The relative values of several indicators are then summarized. In this study: activity of the studied soil enzymes. Then the average evaluation score of the studied indicators characterizing an individual sample is calculated (2):

$$Bmean = (B1 + B2 + B3 \ldots + Bn)/n \tag{2}$$

where, Bmean - average evaluation score of indicators;
n - number of indicators.

TREA (3) is calculated similarly to formula (1):

$$TREA = \left(\frac{Bmean}{Bmean\ max}\right) * 100\% \tag{3}$$

where, Bmean - average evaluation score of all indicators;
Bmean max - maximum evaluation score of all indicators.

Statistical processing of the data was performed in the Prism 10.2.2.397 software package (GraphPad software). The statistical test of two-way ANOVA ($p<0.0001$), Spearmen rank correlation ($p<0.1$), Pearson's correlation ($p<0.1$) as well as linear and nonlinear regression model (polynomial regression (2nd degree)) were used in this work. Linear and non-linear regression model was used for Ba2-Ba4 samples, due to the presence of different ages fallow lands, among which it is possible to conduct this analysis.

Results and Discussion

The features of soil formation on long-term fallowing lands

The vicinity of Ban'kovo village characterized by distribution of sandy parent materials by water-glacial origin and formation of podzol. In the studied 120-year-old podzol (Ba2) from Ban'kovo village the formation of transitional podzol horizon in the form of whitish layer, which is formed under the arable mineral humus-accumulative horizon, is noted. At earlier stages (80- and 40-year-old soils of fallow lands) this process is not pronounced. Formation of secondary podzol horizon is caused by change of plant communities from meadow to forest, formation of aggressive humus acids, destruction of mineral grains, which is accompanied by removal of iron and aluminum oxides and their accumulation in the middle horizon Bs.

For soils formed in the vicinity of Belogorka village (B1-B3), where the soil-forming rocks are represented by red-colored Devonian clays, more active rates of transformation are observed, in soils of fallow lands 20-years-old, formation of transition horizon with secondary eluviation is noted. The formation of this horizon is caused by excessive moistening in the boreal belt.

The studied soils in the Borovichi area are characterized by a long history of agricultural use, the area of ploughed land in this territory exceeded 80% during the Soviet period, and even those spruce forests that appear to be primary have been subjected to a very significant anthropogenic impact. The soils of the Bor2-Bor4 have loose initial E and Bs horizons, hence they were involved in arable horizons during Soviet times and show no signs of degradation during the following thirty years. Only the soil under the hayfield (Bor5) has some signs of degradation as follow secondary podzolization in the old arable horizon. Soils of agricultural lands can be subjected to significant changes as a result of transition to fallow state, this is caused by the change of plant communities, waterlogging, loss of organic matter and transformation of soils towards zonal series (Kalinina et al., 2019). The studied soils in the area of Ban'kovo village are less subject to transformation, this is due to the features of soil formation on red-colored sandy moraine sediments, on which the process of secondary podzolization can take more than 300 years (Litvinovich, 2009).

The studied soils are characterized by different degree of fallow state and type of agricultural use, which allows to study temporal and spatial dynamics of enzymatic activity.

Physico-chemical characteristic of studied soils

The studied undisturbed soils (Ba1, B3, Bor1) are characterized by acidic reaction, with relatively high content of carbon represented in the form of coarse forms of humus. Relatively high pH value was characterized by soils (Bor2-Bor6) in the area of Borovichi, which is associated with the young age of transition to fallow state, as well as their use in the form of gardens, pastures, and for growing crops. Soils of fallow lands were characterized by a higher pH level, indicating that agricultural influence has a long-term effect on the soil and does not contribute to a sharp change in the acid-base balance of soils (Table 2). However, according to Litvinovich (2009) fallow soils formed on water-glacial and lake-glacial sediments can change acidity relatively rapidly, which is due to active leaching processes and migration of Fe and Al oxides. In the first three decades, soils of fallow lands can retain their acid-base properties even if fertilizer application is stopped (Dymov et al., 2018).

Table 2. Physico-chemical parameters of studied soils.

Soil ID	Horizon	pH	C, %	BR, $mgCO_2$-C/g h^{-1}	N-NH_4^+	N-NO_3^-	P	K	Sand	Silt	Clay
					mg/kg				%		
Ba1	E_1	4.98	4.98	0.25	3.88	0.36	29	25	92	4	4
	E_2	5.91	5.91	0.19	2.83	0.36	188	16	86	11	3
Ba2	O_e	5.58	4.70	-	9.43	2.12	205	28	88	7	5
	A_p	5.64	1.39	1.66	2.46	0.45	140	23	90	6	4
Ba3	A_p	5.68	0.62	1.63	1.36	0.36	222	505	93	4	3
	B_s	5.71	0.35	1.03	1.31	0.06	187	264	93	3	4
Ba4	A_p	5.89	1.44	1.44	1.10	0.36	276	397	89	7	4
	B_s	5.85	0.38	0.63	0.68	0.18	106	522	90	7	3
B1	A_p	4.87	1.44	0.09	4.45	1.84	210	103	70	24	6
	E	5.25	0.62	0.06	0.68	1.60	43	107	67	25	8
B2	A_p	5.69	1.48	0.45	5.97	6.32	255	470	72	22	6
	B_t	5.66	0.53	1.05	1.15	1.54	125	264	69	15	16
B3	A/E	4.42	2.28	1.28	3.09	0.33	19	692	78	17	5
	B_t	4.79	0.82	0.34	1.83	0.45	241	560	75	19	6
Bor1	A_h	5.73	0.83	0.82	4.08	0.27	25	10	88	5	7
Bor2	A_p	6.11	1.40	0.05	6.49	0.49	59	13	75	18	7
Bor3	A_p	6.67	2.55	2.03	11.78	1.06	70	69	77	16	7
Bor4	A_p	6.37	2.42	1.60	11.52	2.31	161	195	86	11	3
Bor5	A_p	6.23	2.21	1.45	7.91	0.70	93	93	85	10	5
Bor6	A_p	6.87	3.77	1.27	5.92	3.61	499	505	83	11	6

Based on the data on carbon content in soils of fallow lands, we can note that the highest carbon content corresponds to Plaggic Podzol formed in the vicinity of Borovichi city. This is due to the different type of land use, development of humus-accumulative process, as well as fertilizer application. Soils of fallow lands, which are in the process of long-term self-restoration, are characterized by the formation of litter horizon (Ba2 O_e), in which weakly decomposed plant residues are accumulated. This horizon is formed above the old arable horizon. Younger variants of fallow land soils are characterized by the presence of old ploughing horizon from the surface, in which humus accumulation and transformation takes place, the carbon content in them varies in a wide range from 0.62% in Ba3 A_p horizon with the age of 80-years-old to the highest carbon content of 1.48% in B1 A_p horizon with the age of 20-years-old. Soils that had not recently come out of agricultural influence were characterized by the highest carbon content 40-years-old (Ba4 1.44% carbon content) as well as 20 years old (B1 1.48%). Regression analysis was performed to identify the relationship between the content of carbon and biogenic elements in old-arable soil horizons and the age of the deposit (Figure 2).

The obtained model shows that for this site the minimum of carbon content (Figure 2A) is observed between 80-81 years, after this time there is an increase in carbon content in the old-arable horizon. This may be due to active accumulation of organic matter as a result of transformation of plant residues that accumulate in the overlying O horizon.

Based on the analysis of basal respiration in soils, it was noted that the highest level is observed in old ploughed humus-accumulative horizons (Ba3, Ba2), where active transformation of organic matter takes place. Younger fallow soils were characterized by lower values of basal respiration.

From distribution of N-NH_4^+ it was noted that the highest content of ammonium form of nitrogen is characteristic of secondary forest plots (Bor3), as well as pasture (Bor4), fallow soils not used in agriculture were characterized by significantly lower content of N-NH_4^+, which may be associated with the cancellation of mineral fertilizer application. According to the obtained regression model (Figure 2E) for the Ban'kovo site, the N-NH_4^+ content was found to increase significantly with the age of the fallow state.

The highest content of N-NO_3^- was observed in the soil used in agriculture (B2), which is due to the application of mineral forms of fertilizers to the soil. The same is followed for phosphorus in the soil, the highest content was observed in the soil of agricultural land (Bor6). At the same time the lowest content of this element was found in horizons of undisturbed soils (Ba1 E_1, B3 A/E). According to the linear regression model (Figure 2D) for the Ban'kovo site, it was found that N-NO_3 content increases with age.

The highest content of potassium was noted in undisturbed soil B3 A/E, which is due to its relatively high content in soil-forming rocks. The distribution of potassium (Figure 2C) as a function of time for the Ban'kovo site is described by a nonlinear polynomial regression of the second degree, which shows that the maximum of potassium content falls on 65-70 years, after this time there is a decrease in the content. Phosphorus is characterized by a decrease in content with increasing age of the deposit, which is described by a linear regression equation (Figure 2B). According to the data of particle-size distribution, most of the studied soils belong to sands and sandy loam, except for soils formed in the area of Belogorka village (B1-B3), here the content of sand decreases and the content of dusty and clay particles increases, which is due to the formation of soils on deposits of Devonian red-colored clays.

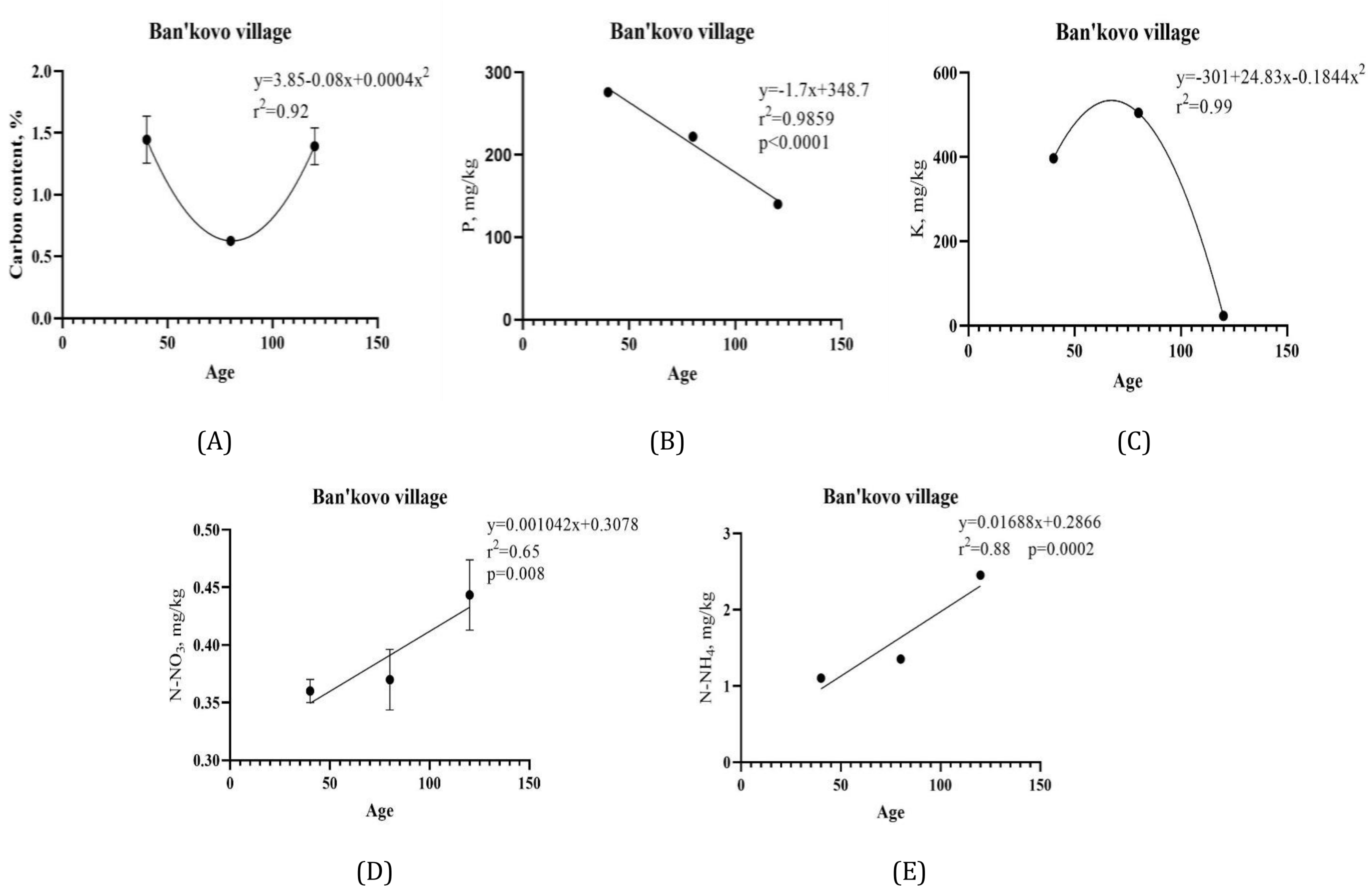


Figure 2. Changes in carbon and biogenic elements content depending on the period of their abandonment. A – carbon content (%), B – P, mg/kg, C – K, mg/kg, D – N-NO_3, mg/kg, E – N-NH_4, mg/kg.

Spearman rank correlation analysis was applied to identify the correlation between the investigated physico-chemical parameters (Figure 3).

According to the obtained data, we can note that there is no high correlation among the studied parameters except for the content of sand and dust. This is due to the different age of soils of fallow lands and features of soil transformation in the conditions of transition to fallow state.

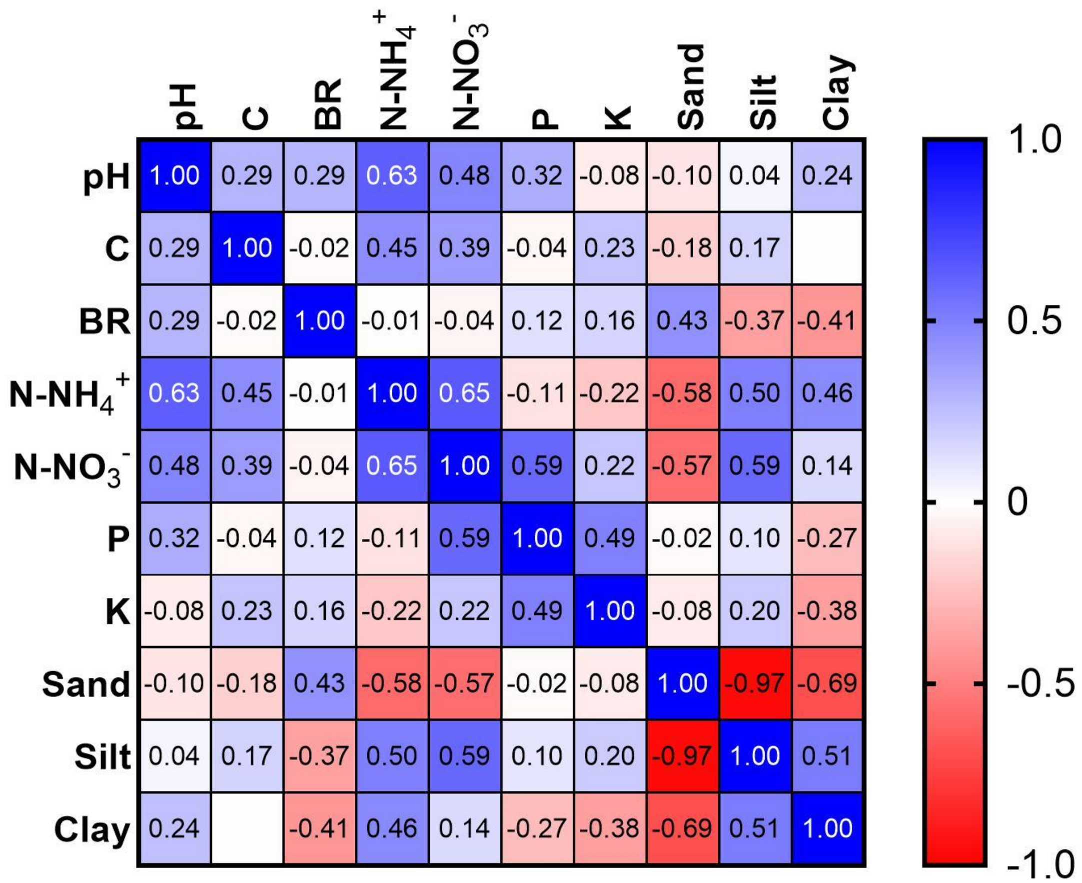


Figure 3. Spearman rank correlation graph for physico-chemical parameters of soils.

Enzymatic activity of soil

To characterize the enzymatic activity of soils of fallow land as well as soils with different types of use, the catalytic activities of enzymes of hydrolase (INV, PHA, Ure) and oxidoreductase (DHA, CAT) classes were studied (Figure 4, 5). According to the data obtained, the enzyme activities of soils have high variability Cv ranges from 68.13% for DHA to 220.9% for Ure. The high variability of these enzymes has been noted by other researchers (Gorobtsova et al., 2017), who attribute this with the high variability of soil formation conditions, anthropogenic impact, as well as the time of soils being in fallow state. According to the statistical analysis, significant differences ($p<0.0001$) in enzyme activity among the upper humus-accumulative horizons of the studied soils were determined.

Among the enzymes of hydrolase class, the highest activity was observed for PHA, for Bor6 (159.48 mgP_2O_5/100 g/1 h) in agricultural soils. The high activity of PHA in agricultural soils was due to the application of mineral forms of phosphorus. In fallow soils, PHA activity decreased with the age of transition to fallow state, with very strong activity in undisturbed soils (Ba1 E_1, B3 A/E). The high PHA activity in undisturbed soils may be due to active transformations of organo-mineral components in podzol horizons (E). According to the Ure activity, it was found that no Ure activity was observed in most of the studied soils. The highest activity was observed in secondary forest (Bor3), as well as in Ba2 O_e, thus Ure activity is associated with the formation of forest litter on fallow land soils. Similar results were obtained in a study on enzyme activity on secondary forests formed in place of fallow land conducted in China's Danxia Province, which showed that reclaimed land contained more hydrolytic enzymes than arable soils (Wang et al., 2023). According to the data obtained during the analysis of INV activity it was noted that the highest activity level was observed in soils of different types of use Bor4 (pasture), Bor3 (secondary forest), Bor2 (garden), as well as soil used in agriculture (Bor6). Among soils of fallow lands, it can be noted that INV activity was higher in old-aged fallow soils (Ba2 O_e) and decreased with decreasing age of transition to fallow state.

DHA activity was found to be very weak in most of the studied soil samples, the highest activity was observed in Ba2 O horizon (5.83 mgTFF/10 g/24 h). Our data are confirmed by Gorobtsova (2017), who found that in undisturbed horizons DHA activity is higher than in arable horizons. DHA activity depends significantly on the type of land use, as well as on the quality of organic matter, the content of labile forms of humus leads to an increase in DHA in soils (García-Ruiz et al., 2008). CAT activity averaged 1.35 mgO_2/1 g/ 1 min, and was weak in most of the studied samples, the average activity level was recorded in sample Bor 3 (secondary forest). The activity level decreased from undisturbed soils to fallow soils. The activity of oxidoreductases was higher in soils with restoration of woody vegetation species and in soils with formation of litter horizon O, where transformation of plant residues takes place.

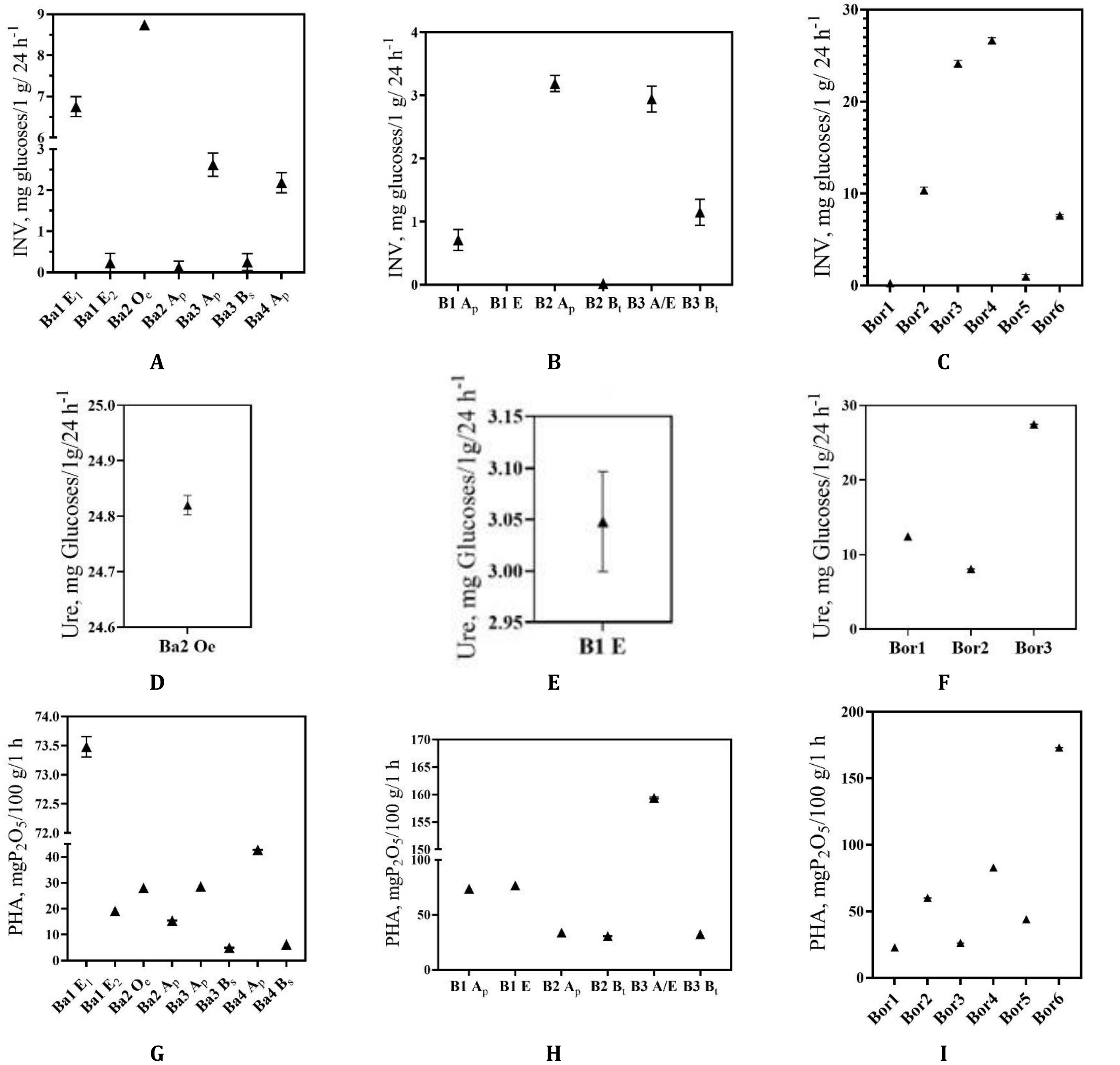


Figure 4. Soil enzymatic activity indices (INV, Ure, PHA) of undisturbed and fallow lands, as well as lands with different types of use ($p<0.0001$). A, D, G – Ban'kovo, B, E, H – Belogorka, C, F, I - Borovichi

Pearson's correlation was performed to analyze the correlation relationship between physicochemical parameters of soils and enzymatic activity. It was found that for $N\text{-}NH_4^+$ content has a high level of correlation with INV ($r=0.84$, $p<0.0001$) and with CAT ($r=0.82$, $p<0.0001$). It is observed that clay content has a relatively high level of correlation with DHA ($r=0.66$, $p=0.01$) and URE ($r= 0.61$, $p=0.02$). Figure 6 shows the dependence of enzymatic activities on physicochemical parameters of soils.

The comparison of the studied soils shows that almost all enzymes at all studied sites lost their activity down the soil profile, which is in accordance with the results of other researchers (Gorobtsova et al., 2015).

To identify the total biological activity of the studied soils, the total relative enzymatic activity was calculated according to Zvyagintsev (1978) and Kazeev (2003) and used in the works of Gorobtsova (2015). Two enzymes from the hydrolase class (phosphatase and invertase) and oxidase class (dehydrogenase and catalase) were used in the calculation of total relative enzymatic activity in this work. The activity of Urease was not included in the calculation because at most points, it showed no activity. The Figure 7 presents diagrams showing the total relative enzymatic activity of the upper horizons of soils in the studied plots. The obtained data show that in the area of Bankovo village the total enzymatic activity of soils increases when the period of fallow state from 40 to 120 year increases. The increase in enzyme activity in the process of postagrogenic evolution is also mentioned in the work (Kazeev et al., 2020). It shows that enzyme activity starts to increase in the first year of fallow state and continues to recover throughout the time after the cessation of agrogenic impact.

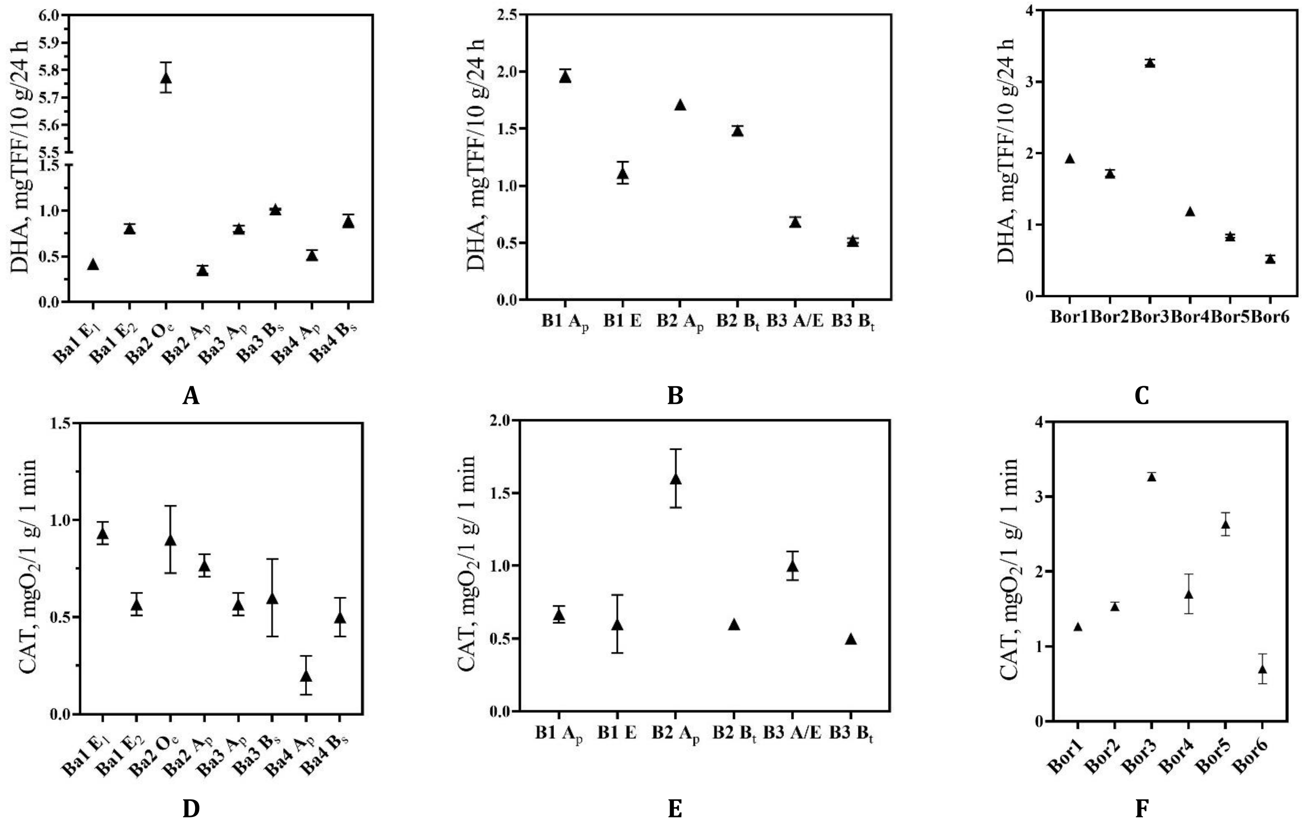


Figure 5. Soil enzymatic activity indices (DHA, CAT) of undisturbed and fallow lands, as well as lands with different types of use (p<0.0001). A, D – Ban'kovo, B, E – Belogorka, C, F – Borovichi.

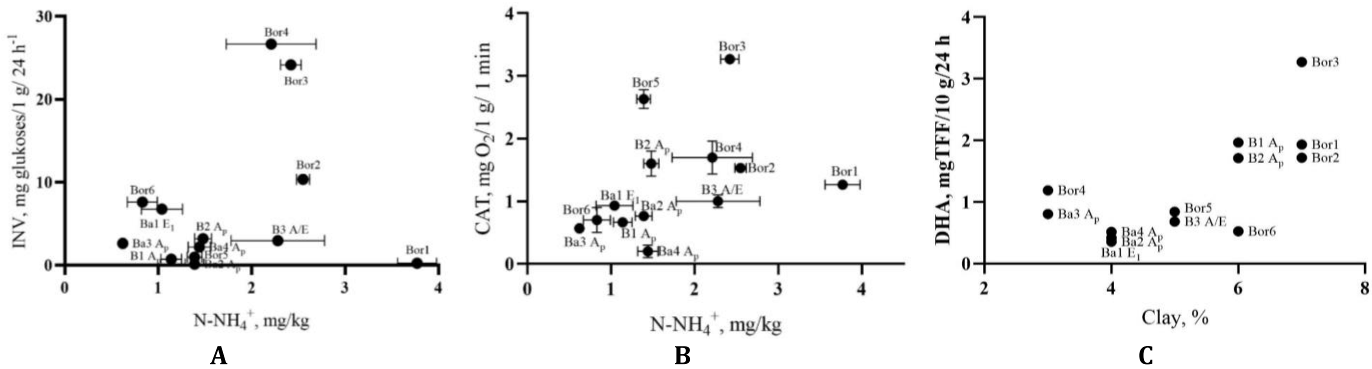
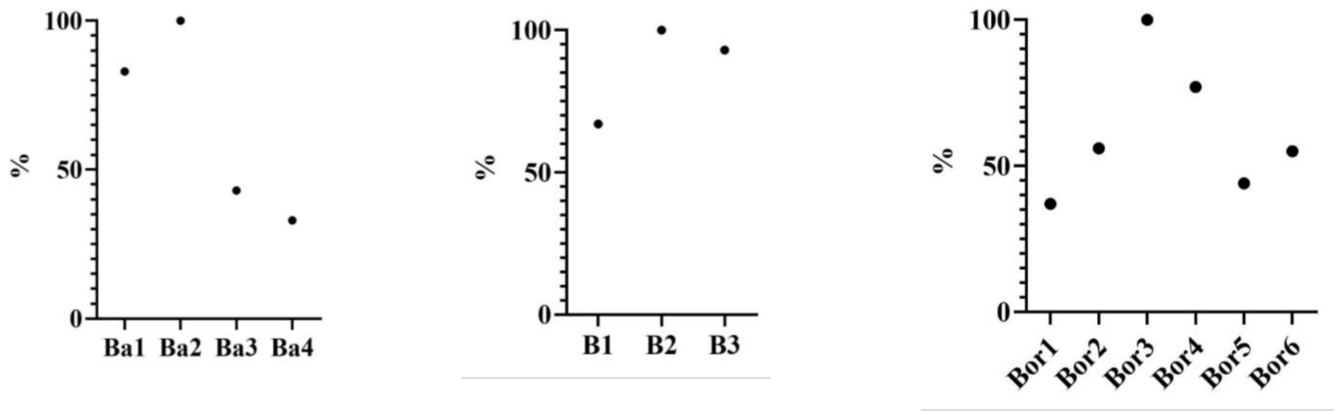


Figure 6. Relationship between physico-chemical parameters and enzymatic activity. A - Relationship between N-NH$_4^+$ and INV. B - Relationship between N-NH$_4^+$ and CAT. C - Relationship between Clay and DHA.

In comparison with the soils in the Bankovo area, in the soils formed in the vicinity of Belogorka village, the total enzymatic activity in the soils of agricultural land is higher than in the soils under 20-year fallow land. Such a high value of total enzymatic activity in arable soil was obtained due to high values of redox enzymes. Similar results were obtained in the study of fallow soils in Belgorod region (Ovsepyan et al., 2017), which revealed higher activity of redox enzymes in arable soils compared to fallow soils. Comparison of the total enzymatic activity of 30-year-old fallow soils under different land uses in the Borovichi area revealed that soils under secondary forest had the highest biological activity. The lowest values of total enzymatic activity in this area were found in the undisturbed soils under forest (Bor 1). This is probably due to the fact that the soil in this area is acidic, which is consistent with studies (Gedgafova et al., 2015; Gorobtsova et al., 2015) showing that enzyme activity is higher in soils with slightly alkaline reaction compared to acidic soils.



A **B** **C**

Figure 7. Total relative enzymatic activity (%) of the surface soil layer of the study sites.
A – Ban'kovo, B – Belogorka, C – Borovichi.

Comparison of all studied plots showed that the soil under the secondary forest formed on a 30-year fallow land has the highest total enzymatic activity. High biological activity at this site is due to the neutral pH, some of the highest values of organic carbon and nitrogen content, and basal respiration, which according to various authors (Kazeev et al., 2004; Gorobtsova et al., 2015) have a positive correlation with the total enzymatic activity of soils.

Conclusion

The transition of agricultural soils to fallow state induces complex changes in their biochemical and physicochemical properties. As soils remain uncultivated, several key transformations occur: pH gradually decreases due to natural podzol formation processes and shifts in vegetation cover, leading to the formation of E horizon and leaching of sesquioxides. The dynamics of biogenic elements follow a U-shaped pattern - initial depletion in early fallow stages gives way to gradual accumulation as the ecosystem stabilizes. Enzymatic activity undergoes significant modifications reflecting these ecological changes. Urease, dehydrogenase and catalase activities peak in mature soils of fallow lands, suggesting microbial communities retain memory of past agricultural use. Phosphatase shows maximal activity in arable soils due to residual effects of phosphorus fertilization. Invertase shows the most variable response, with highest activity observed across different land-use types, particularly in mature fallow systems.

The most biologically active soils are found in 30-year fallows state that have developed secondary forest cover. These systems combine neutral pH, high organic carbon content, and basal respiration, which is forms optimal conditions for enzymatic processes. Total enzymatic activity in such ecosystems often exceeds that of both recently abandoned fields and continuously cultivated lands. These patterns demonstrate that fallow succession drives non-linear changes in soil enzymatic profiles. Early fallow stages typically show low activity, while mature systems close or exceed the biological activity of native ecosystems.

Further research should focus on long-term monitoring of enzyme dynamics across different soil types and climatic zones to better predict fallow system recovery.

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