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ASSESSMENT OF VARIATIONS CAUSED BY BIOLOGICAL ACTIVITIES IN THE GREATER CAUCASUS FOREST SOILS USED FOR AGRICULTURE

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SUMMARY

For the first time, the determination of biological activity parameters found in the upper horizon (0–20 cm) of the mountain gray forest soil succeeded at the agrocenosis and natural biogeocenosis of the Sheki-Zagatala Economic Region, Azerbaijan. The use of integrated indicators of the ecological and biological state of soils (IIEBSS) summarized the variations in biological activities (humus content and reserves, microbial biomass carbon content and reserves, activity of hydrolase, and oxidoreductase enzymes) of the arable soils. A 45% IIEBSS decrease was evident in the cultivated soils due to soil degradation processes leading to a disruption of their ecological functions and a decline in their fertility. The pioneering study made a comparative assessment of the variations caused by the biological activity in the upper horizon of mountain gray forest soils' biogeocenosis in the southern macroslope of the Greater Caucasus. Studies on soil biological indicators are few, and this research is vital to develop and boost ecotourism.

Keywords: Soil biological activities, soil degradation, soil fertility, mountain gray forest soils, Greater Caucasus, Sheki-Zagatala Economic Region, soil use for agriculture

Key findings: The progressive study predominantly depended on the effective use of forest soils with decisive importance. The study revealed soil stability was low in cultivated cenoses under long-term anthropogenic

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INTRODUCTION

The soil cover of the mountain zone has always attracted the attention of researchers due to the originality of its properties, the conditions of its formation, and the general patterns of its geographical distribution. The greatest anthropogenic impact beset the flat and foothill zones, and large arable areas have lost their natural appearance due to intensive agricultural production (Allison and Vitousek, 2005). Soil involvement in active agriculture has a negative influence on its physical, chemical, and biological properties, which, in turn, causes the development of degradation processes, leading to the disruption of ecological functions in soils with reduced natural fertility (Acosta-Martínez and Tabatabai, 2000; Adamczyk *et al.*, 2014).

Agriculture happens in conditions of complex relief and diverse natural landscapes with dissimilar bioclimatic conditions (Asgarova and Hasanova, 2022). In accordance with natural and economic characteristics, the territory of the economic region underwent division into three agroecological zones, i.e., plain, foothill, and mountain, which considerably differed in climate, vegetation, and soil cover (Bunyatova *et al.*, 2025). Despite the ruggedness of the relief, a fairly large area of land contributed to agricultural use, amounting to ≈60% of the entire territory of the Republic, including 43% occupied by the arable land.

Agrogenic soils are mostly useful for highly productive grain crops, as well as for oilseeds, vegetables, and fruit crops. This work is part of a series of scientific studies aimed at studying the biological activity of soils in the plain and foothill areas of the Sheki-Zagatala Economic Region (41°15'34"N 47°11'17"E), subject to long-term agricultural impact.

Past studies cover in sufficient detail the issues related to the morphological, physical, and agrochemical properties of the republic's mountain soils. However, studies of the soil's biological characteristics, particularly in mountain gray forest soils common in foothill areas, have not progressed to date. The terraced landscapes of the southern slopes of the Greater Caucasus are territories playing a

vital role in the development of tourism in the country. (Hasanova, 2015; Ewing *et al.*, 2020; Hasanova and Abasova, 2024a).

Biological parameters' determination allows researchers to establish the general level of biological activity of arable and natural mountain soils and draw certain conclusions about the soil resistance to anthropogenic loads (Bao, 2000). By assessing biological activity, it seems very promising to determine such informative parameters, i.e., humus content, physiological activity of microbial biomass, and the activities of the two classes of enzymes: hydrolases (invertase, phosphatase, and urease) and oxidoreductases (dehydrogenase and catalase) (Hasanova *et al.*, 2021; Hasanova and Mammadova, 2023). Soils, when used rationally in agriculture for cultivation, acquire new features. One should consider the soil resources of the planet, as well as our region, are inadequate; therefore, one of the most critical problems is the efficient use of the land reserves (Figure 1). Preservation of soil samples in a soil museum from natural and cultivated communities is crucial for the research of young soil scientists and ecologists of future generations (Mammadova *et al.*, 2020).

The integral indicator of the ecological and biological state of the soil (IIEBSP) formulation, applied to obtained data, allows scientists to determine the biological activity of cultivated and natural mountain gray forest soils. Likewise, it helps researchers assess the effect of agrotechnical measures on their biological properties (Babaev *et al.*, 2015; McDaniel *et al.*, 2013). The progressive study sought a comparative assessment of the biological activity indicators in the upper horizon (0–20 cm) of mountain gray forest soils of agrocenoses and natural biogeocenoses of the southern macroslope of the Greater Caucasus, Azerbaijan (Hasanova and Abasova 2024a; Hasanova and Asgarova, 2024b; Hasanova *et al.*, 2025).

MATERIALS AND METHODS

The object of the study was mountain gray forest soils formed in the foothills of the



Figure 1. Forest soil monoliths from forest meadow soils (41°15'34"N 47°11'17"E) in Soil Museum at Institute Soil Science and Agrochemistry of Azerbaijan.

southeastern part of the Greater Caucasus, Azerbaijan. The study area covers part of the broad-leaved forest belt and the area occupied by mountain gray forest soils (899 km²). Soil-forming rocks were the tertiary sedimentary deposits (limestones, sandstones, clays, and marls) (Mammadova *et al.*, 2024). The study area revealed characteristics of a moderately humid climate with warm summers and cold winters (41°15'34"N 47°11'17"E; 41°14'14"N 47°09'29"E, etc.).

Mountain gray forest soils formed under beech-hornbeam-oak forests (Kazeev and Kolesnikov, 2012). The natural vegetation cover of treeless slopes and foothill plains had representatives of forb-grass communities (Moreau *et al.*, 2019). Soil is the richest reservoir of biodiversity on the planet. Microbiological examination continued on 35 soil samples taken from the arable horizon from 32 sites (Howard *et al.*, 2017).

On the leveled areas of the relief, the described soils with favorable agrophysical and agrochemical properties served as arable lands for perennial plantings, pastures, and hayfields. In decades past, the arable lands have been the main source of small commodity producers on short-term lease terms in Azerbaijan (Nasirova *et al.*, 2022). In the current economic conditions, tenants do not comply with the required level of agricultural technology, do not use a rational crop rotation

system, and do not follow the standards for applying organic and mineral fertilizers, which eventually harshly affect the properties of the cultivated soils (Sparks *et al.*, 2020).

Collection and analysis of the soil samples to determine physical and biological properties proceeded as per recommended methods in ecology and soil science (Rowell, 1999). The soil samples collected came from the 0–20 cm horizon using envelopes at the natural biogeocenoses and agrocenoses in the first 10 days of July 2023 (Figure 2). Mixed samples totaling seven reached collection from the maize fields. Cartographic materials utilization helped determine the location of sampling points. The soils' classification diagnostics succeeded according to genetic classification. Laboratory and analytical studies performed included determining the humus content (%) following Tyurin's method in Nikitin's modification, by the potentiometric method, field moisture, soil density, and by the gravimetric method (Ismayil *et al.*, 2025).

Humus reserves in the 0–20 cm soil layer received calculation using the soil density indicators. A method for determining the rate of substrate-induced respiration (SIR) aided in assessing the physiological activity of microbial biomass (Shayakhmetova *et al.*, 2023). The SIR rate estimation employed the intensity of microorganism respiration after enriching the soil with a glucose solution (0.2 ml/g of dry

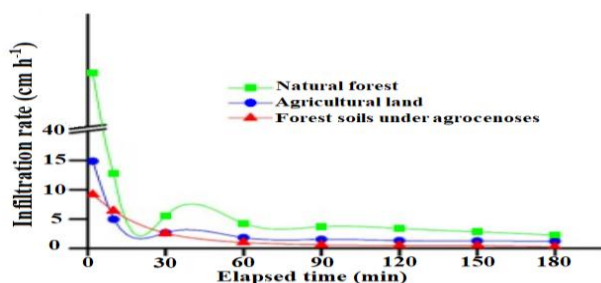


Figure 2. Variation in soil infiltration rate of natural forest, agricultural, and forest soils under agroecosystems.

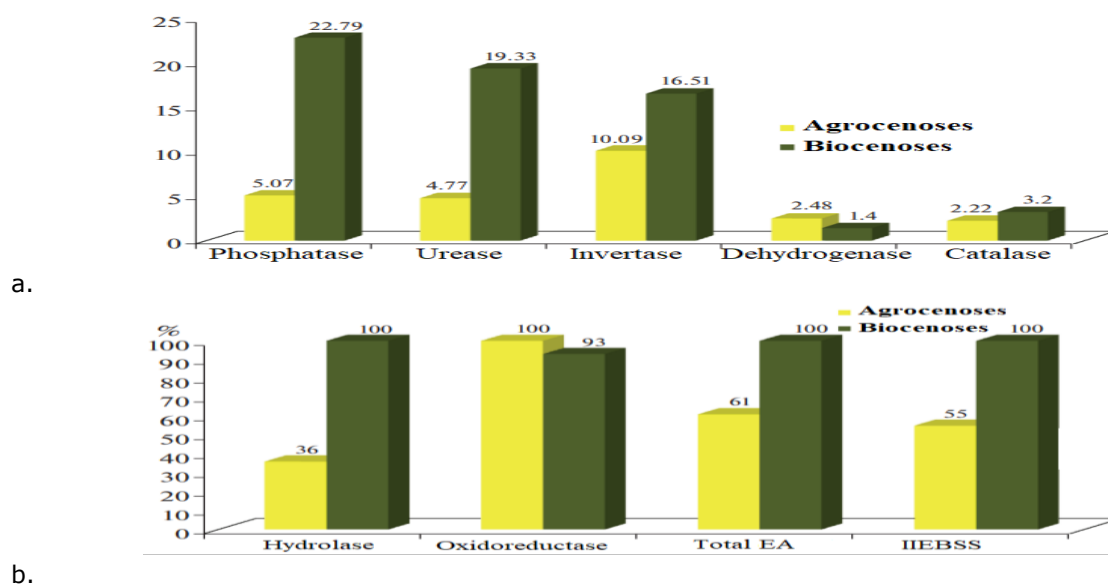


Figure 3 a. Changes in average values of enzyme activity in the upper horizon (0–20 cm) of mountain gray- forest soils as a result of agricultural use. b. Total relative activity of hydrolases, oxidoreductases, enzymes of two classes, and the value of IIEBSS of mountain gray-forest soils (in 0–20 cm horizon).

Note: a. Phosphatase activity (Pa) is expressed as mg P₂O₅/100 g soil/h, urease – mg /10 g soil/day, invertase – mg glucose/g soil/day, dehydrogenase – mg TPP/10 g soil/day, catalase – mg O₂/g soil/min. b. Total EA – Total enzymatic activity; IIEBSS – Integral indicator of the ecological and biological state of soils.

soil; titer 0.05 g of glucose). The detection of the carbon content of the soil microbial biomass continued by the following formula.

$$C_{\text{micr}} (\mu\text{kg C/g soil}) = \text{SIR} (\mu\text{CO}_2/\text{gsoil/h}) \times 40.04 + 0.37$$

The carbon reserves of microbial biomass in the 0–20 cm soil layer sustained verification using bulk density data. The proportion of microbial biomass carbon ($C_{\text{micr}}:C_{\text{org}}$ [%]), as calculated, was the ratio of

the microbial biomass carbon content to the total organic carbon content in the soil. The activities of the different enzymes, i.e., dehydrogenase, invertase, phosphatase, and urease, attained distinction by the colorimetric method, and catalase by the gasometric method, according to the methods of Khaziev (Figure 3). The obtained biological indicators bore assessment according to the Gaponyuk-Malakhov scale. Calculating the total relative

enzymatic activity engaged the approach according to the Zvyagintsev method (Shukla and Varma, 2011; Kazeev and Kolesnikov, 2012).

For a comparative assessment of the biological activity of natural and arable mountain gray forest soils, using the IIEBSS calculation method allowed for integrating the relative values of all the studied indicators. The obtained data analysis proceeded with the Statistica 10.0 program. The reliability of the differences in the studied soil characteristics of agro- and biogeocenoses assessment used the Student's t-test at a significance level of $p \leq 0.05$. Data visualization ran using the MS Excel 2013. The different data tables and graphs show the mean values and standard error (SE) (Sinsabaugh *et al.*, 2008; Sinsabaugh, 2010).

RESULTS AND DISCUSSION

Soil enrichment with organic matter provided a noticeable enhancement in microbial activities and expressed an increase in the rate of basal respiration and the microbial biomass content (Ramazanova, 2017). The most prominent variations in these indicators during the restorative succession resulted in the upper soil layer (0–10 cm). Their maximum values were evident in the soil of a 30-year leyland corresponding to a small-leaved forest, where easily decomposable litter provides a large reserve of accessible substrate for the microbial community (Shukurov *et al.*, 2025). An important factor of soil fertility with a certain influence on the soil processes, including biological ones, was the reaction of soil solution (The State of the World's Forests, 2020, 2019).

The experimental results showed natural mountain gray forest soils in the upper humus-accumulative horizon had a slightly acidic reaction (Talibi and Hasanova, 2022). Agricultural use caused some alkalization in the soil solution; however, the differences in the pH_{H₂O} value for natural and cultivated soils (11%) were nonsignificant ($t = 1.74$, $P = 0.12$). Bulk density is one of the physical indicators influencing the activity of biological soil processes. The average density of arable

horizons corresponded to the level of medium-compacted soil and significantly exceeded (34%) the average density of the upper horizon of natural mountain gray forest soils ($t = 8.99$, $P = 0.00$).

This study used the principal component method (Zhao *et al.*, 2024) to integrate and visualize the information obtained in the research. These include the variations in a wide range of properties at four different depths of the old-arable horizon in five objects forming the chronological series of fallow soils. Considering the entire set of studied properties, old-age mixed and young small-leaved forests, as well as arable soil and 15-year-old leyland, showed obvious distinction (Figure 4).

The youngest 10-year-old fallow land occupies an intermediate position between the arable land and 15-year-old fallow land. Factor 1 that was significantly correlated with the content of C_{org} ($r = -0.91$) and N_{total} ($r = -0.90$), phosphatase activity ($r = -0.93$), and pH ($r = 0.76$) describes 55.0% of the total information, and the ordinate axis clearly separates forest soils from arable soils and younger fallow lands, where meadow vegetation predominates. Factor 2 had the most considerable association with the content of C_{mic} ($r = -0.79$), which was responsible for only 19.6% of the revealed patterns. Thus, in the southern zone, during the restorative fallow succession, within the entire old-arable horizon, a consistent increase in the content of organic matter existed.

As a consequence, an increase in their microbial and enzymatic activity was prominent in the upper soil layer (0–10 cm) of the old-growth forest. It represented the subclimax stage of the post-agrogenic forest restoration, which was in a state of dynamic equilibrium to achieve, and post-agrogenic soils may need many years (Macnunlu *et al.*, 2025). During post-agrogenic succession, a noteworthy increase in the stratification of the C_{org} and N_{total} content in the 0–30 cm layer emerged, especially pronounced at the stage of formation of a closed stand and indicating the restoration of the carbon-sequestering function of soils at later stages of post-agrogenic development.

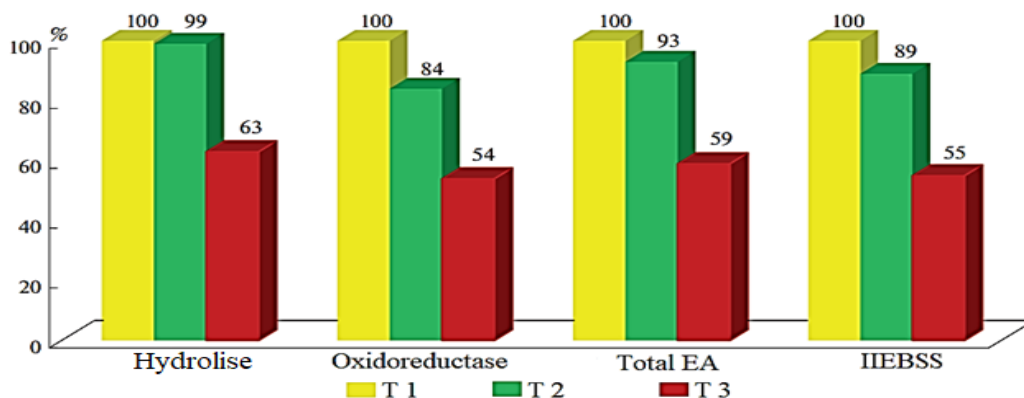


Figure 4. Total relative activity of hydrolases, oxidoreductases, enzymes of two classes, and IIEBSS of the upper horizons (0–20 cm) of mountain meadow-steppe soils at different stages of pasture digression.

Note: Total EA – Total enzymatic activity; IIEBSS – Integral indicator of the ecological and biological state of soils.

The highest values of the stratification index (0–5:5–10, 0–5:10–20, and 0–5:20–30) appeared in the soil of an old-growth forest, which was fundamentally different in the structure of its organo profile.

The linear trends assessment with the change in the C_{org} content showed that over 30 years of post-agrogenic succession, a significant increase resulted in the entire old-arable layer (except for the 20–30 cm layer), with the highest growth rate of 0.66 g C/kg soil per year in the upper soil layer (0–5 cm) (Hasanova *et al.*, 2021c). A substantial rise in the N_{total} content was evident in the 0–5 and 20–30 cm soil layers with linear regression coefficients of 0.049 and 0.029 g N/kg soil per year, respectively. During the restorative succession, the increase in the C_{org} and N_{total} content in deeper soil layers was less prominent. For the soils of chronoserries, a negative correlation occurred between the content of C_{org} , N_{total} , and the C/N ratio with the pHKCl value in the 0–5 and 5–10 cm layers ($r = -0.76 \dots -0.92$, $P < 0.06$).

Soil enzymes, both hydrolytic and oxidoreductase, play a vital role in the transformation of organic matter (Williams *et al.*, 2000). Phosphatase activity enhancement happened many times during post-agrogenic restoration, especially in the upper soil layer of the arable horizon, and appeared primarily associated with an increase in the organic

matter content, with further confirmation by a remarkable positive correlation between these indicators (Hasanova, 2021a). The oxidoreductase activity during post-agrogenic succession does not have an obvious defined trend; however, its peaks correspond to the highly productive meadow (peroxidase). Moreover, the young small-leaved forest may have a connection with an elevated supply of easily decomposable plant litter with different biochemical compositions, such as grasses and small-leaved plants. Extracellular enzymes mediate the degradation, transformation, and mineralization of soil organic matter (Sinsabaugh, 2010). Over 30 years of post-agrogenic succession, the most significant increase in almost all the considered indicators was notable in the upper soil layer. In deeper soil layers, the variations in the analyzed soil properties during restorative succession become less pronounced. Thus, the assumptions made at the beginning of the research achieved full confirmation (Figure 5). The frequency of trees, shrubs, forbs, grass, and lianas in the Sheki, Gabala, Gakh, Zagatala, and Balakan regions' forests appears in Figure 6 (41°38'01" N 46°38'36" E; 41°43'06" N 46°24'59" E; 41°06'12" N 47°00'15" E; 40°58'53" N 47°50'45" E; 41°25'00" N 46°55'00" E). The biomass of trees in the Zagatala Region is greater than its other neighboring regions. The abundance of

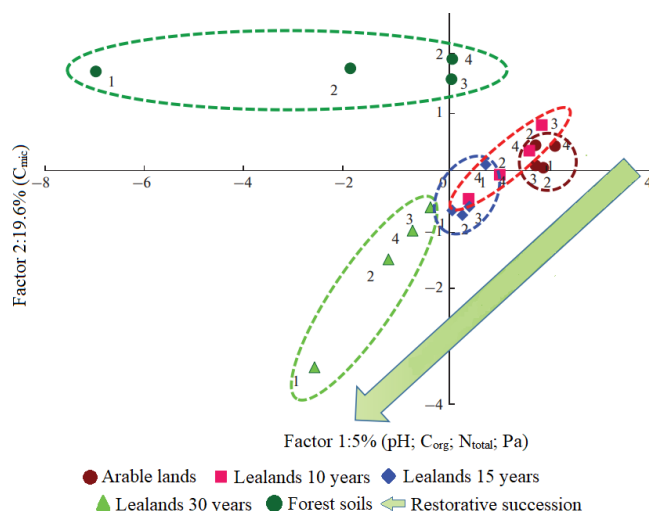


Figure 5. Distribution of study objects using the principal component method using the values of various soil characteristics at different depths: 1 = 0–5 cm, 2 = 5–10 cm, 3 = 10–20 cm, 4 = 20–30 cm.

Properties: pH – potential acidity of soils; C_{mic} – microbial biomass content; C_{org} – organic carbon content; N_{total} – total nitrogen content; Pa – Phosphatase activity ($\text{mg P}_2\text{O}_5/[\text{100 g soil h}])$.

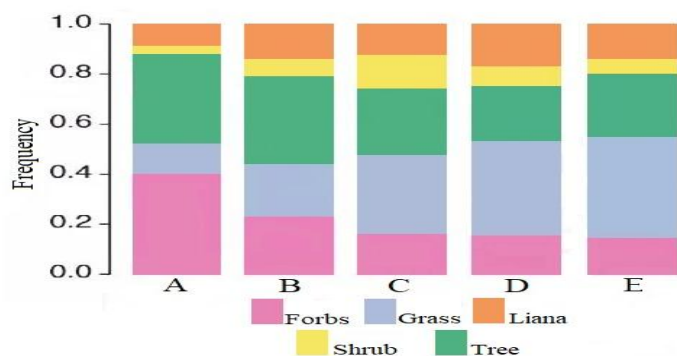


Figure 6. Frequency of forbs, lianas, trees, shrub, and grasses in main regions of Greater Caucasus. Note: A - Zagatala Region ($41^{\circ}38'01''$ N $46^{\circ}38'36''$ E); B - Balakan Region ($41^{\circ}43'06''$ N $46^{\circ}24'59''$ E); C - Sheki Region ($41^{\circ}06'12''$ N $47^{\circ}00'15''$ E); Gabala Region ($40^{\circ}58'53''$ N $47^{\circ}50'45''$ E); Gakh Region ($41^{\circ}25'00''$ N $46^{\circ}55'00''$ E). Source: by Authors in May, 2024.

phytomass determines the high biodiversity in these areas. (Figure 6).

Phosphorus is one of the main macronutrients that is part of such essential macromolecules as DNA, RNA, ATP, phospholipids, and some coenzymes. Phosphorus affects many biochemical processes in plants (Ramazanova, 2016). When it is lacking in plants, the synthesis of proteins and carbohydrates is inhibited, growth

is delayed, and a noticeable decrease in yield occurs. Sufficient phosphorus content ensures acceleration in plant growth and development, the formation of reproductive organs, and the maturation of plants, increasing crop yields and quality. One of the reasons for the high amount of enzymes in mountain forest soils correlates with the high content of elements in the biomass. (Figure 7).

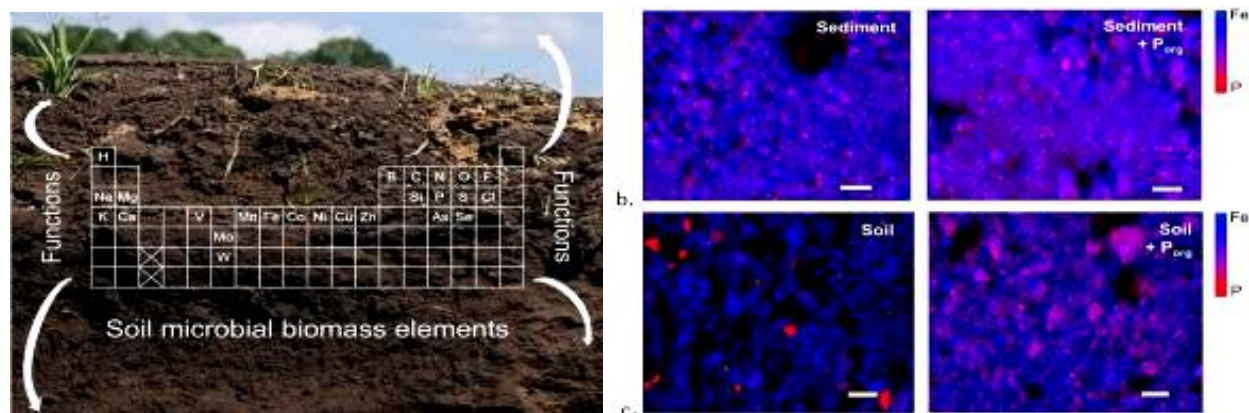


Figure 7. Soil microbial biomass elements and intensity mapping of P_{org} and Fe_{org} in forest natural soils.

Note: (a) Biomass of forest soil elements' functions; (b) and (c) μ-XRF counting intensity mapping for Fe_{org} (blue, maximum intensity = 400 for c or 800 for d) and P_{org} (red, maximum intensity = 20,000) in sediment or soil before (left) and after (right) 10 days of reaction with ATP (ATP-P, 300 μM or 9.3 mg L⁻¹).

CONCLUSIONS

The microbial metabolic coefficient gradually increases during the 30-year restorative succession of former arable soils, showing an association with an escalation in the reserves of incoming litter accessible for microorganisms' rapid decomposition, as well as with its high biochemical diversity. The ratio of basal respiration to the C_{org} that characterizes the available organic matter for decomposition by microorganisms in the upper soil layer of the former arable horizon during the first 30 years of restoration succession provided 2–4 times enhancement. Likewise, it revealed a notable correlation with the C_{mic} content.

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