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DEGRADATION, REHABILITATION, AND CONSERVATION OF SOILS

Soil Biophilic Elements (C, N, P) and Microbial Activity in Forest Parks of Moscow and Suburban Forests

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Abstract—In six forest parks of Moscow and four suburban forests (5 plots each, $n = 50$), soil physical, chemical, and microbial properties of the upper 10-cm layer were assessed in combination with vegetation properties. The contents of carbon (C), nitrogen (N), and phosphorus (P) in soil and microbial biomass were determined. It was revealed that soil bulk density; pH value; and contents of N-NO_3^-,Ca, and heavy metals (Pb, Cu, Ni, Zn) increase in forest parks of Moscow in comparison with those in suburban forests. In the soils of forest parks, a decrease in the microbial biomass C (C_{mic}) content, basal respiration (BR), and microbial C and N availability (C_{mic}/C, N_{mic}/N, BR/C) took place. The changes in soil microbial properties were mainly driven by the decrease in abundance of leaf litter and the available soil C content (13–35% of the explained variance). The microbial response of soil microorganisms to input of low molecular weight organic substrates (carbohydrates, carboxylic and phenolic acids, amino acids, amino sugars) in forest parks and suburban forests did not differ significantly. In the soils of forest parks, no changes in microbial mineralization and immobilization of P (P_{mic}, P_{mic}/P) were found. The impact of urbanization on the forest ecosystems led mainly to a decrease in the intensity of soil C and N cycles. Apparently, these changes were caused by the recreational activity and management practices applied to green spaces in the city, which led, in particular, to a decrease in the amount of forest litter in urban parks compared to suburban forests.

Keywords: urban soils, green infrastructure, microbial biomass, mineralization of organic matter, soil pollution

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INTRODUCTION

Forest parks, squares, and gardens of cities are a key component of urban green infrastructure and play a huge role in mitigating negative urbanization effects [55, 63]. The green infrastructure of the city contributes to the reduction of chemical pollution [32] and CO_2 concentration in the atmosphere [72], sustains the microclimate [55] and biodiversity [56], creates optimal water regime, and improves the aesthetic component [42].

In forest parks, in contrast to other green areas, a special land use is organized, which allows preserving the vegetation and soil covers. Hence, the soils of urban forest parks are analogous to natural soils to a certain extent, since they have a similar profile [6]. However, their chemical and biological properties can differ significantly from the reference soils of suburban forests because of the impact of various factors of the urban environment. It has been shown that the soils of urban forest parks are characterized by a higher pH value and an increased content of organic and inorganic pollutants compared to suburban forests [21, 37, 57, 71]. The soils of the recreational areas of the city are often compacted, and a decrease in the density of the grass cover, the thickness of the plant litter, and the biodiversity of mesofauna have been noted [3, 10, 59]. The content of nitrogen (N) and phosphorus (P) in the soils of urban forest parks can increase due to the additional input of these elements with emissions from vehicles and plants, walking pets, and NP fertilizers [9, 30, 36, 65]. A meta-analysis of nitrogen compounds precipitated from the atmosphere revealed that in the cities of East Asia, Europe, and North America, their content in the soils is higher than that in the soils of corresponding suburban areas [36, 54]. In cities, emissions of nitrogen dioxide (NO_2) from industrial enterprises can be a significant source of soil nitrogen. For example, in Moscow industrial emissions supply 134 kg $N-NO_2$ ha/yr [14]. Thus, in urban forest parks, a decrease in the input of organic carbon (C) to the soil with plant litter takes place together with an increase in the input of nitrogen and phosphorus, which can introduce a certain imbalance in the cycles of these biophilic elements.

The cycles of biophilic elements in the soil are closely related to the activity of soil microorganisms, which are involved in the decomposition of organic matter (OM) thereby contributing to plant nutrition [7, 70]. The respiration activity of the microbial community determines the rate of OM mineralization, providing regulating and supporting ecosystem functions associated with the turnover of biophilic elements [1, 12, 66, 69]. The microbial activity (basal respiration) is most often estimated by the rate of CO_2 production under optimal hydrothermal conditions without an additional energy source (organic substrate) application. The ability of microorganisms to decompose certain organic compounds can be analyzed by their respiration response to the input of specific readily available C-containing substrates, which characterizes the microbial catabolic activity [29].

Soil microbial activity largely depends on the content of biophilic elements, which can change in urban environment due to their additional input [33, 35, 41]. There are controversial results in the scientific literature about the effect of extra input of N and P on the soil microbial activity and, accordingly, the rate of OM mineralization and carbon stabilization, which is largely determined by the climatic features and the ecosystem type [33, 35]. In this aspect, the functioning of urban soils in the forest parks of the temperate zone has been poorly studied. Based on the evolutionary-economic theory of microbiome functioning [18], we suppose that the additional input of readily available N and P to the soil of urban forest parks should lead to a noticeable decrease in microbial respiration activity aimed at obtaining these elements from more complex OM pools. Such changes should also lead to a reduction in the share of microbial biomass in the total contents of C, N, and P in the soil of forest parks compared to the reference soils. At the same time, the additional input of C-containing readily available substrates (carbohydrates, carboxylic acids, etc.) into the soil of forest parks should compensate for the imbalance of its biophilic elements, that is, contribute to an increase in the C content in relation to N and P. Therefore, we expect that the respiration response of microbial community in the soils of urban forest parks to the input of an additional source of energy in the form of readily available C-substrates should be at the level of that in suburban reference forests. Thus, the first hypothesis of our study will be examined by assessing the basal respiration and the ratio of microbial biomass CNP to the total pool of these elements in the soil, and the second one—by evaluating the respiration response to the input of various organic substrates into the soil.

OBJECTS AND METHODS

Sampling points. Moscow has an area of ~2500 km^2 (since 2012), including the areas of "New Moscow"; the city population is 12 million people. Moscow is the largest city in Europe with a developed green infrastructure. Green plantings occupy almost 49% of its area (of which 175 km^2 are specially protected areas) and provide ecological, recreational, sanitary, and city-forming functions [68]. Moscow is located in the central part of the East European Plain (56° N, 37° E) and is characterized by a temperate continental climate with the mean annual air temperature of 5.0°C and the mean annual precipitation of 696 mm. Moscow oblast is dominated by taiga and mixed forests on zonal soddy-podzolic soils (Albic Retisols) [5].

On the territory of "Old Moscow" (~1000 km^2) within the Moscow Automobile Ring Road (MKAD), the following forest parks were chosen for our study: Aleshkinskii forest, Bitsevskii forest park, Forest Experimental Dacha (Petrovsko–Razumovskii Reserve), Troparevskii Landscape Reserve, Lianozovskii park, Southwestern forest park, the areas of which are 240, 2208, 257, 16, 219, and 102 ha, respectively (Table S1). The suburban areas (Klyaz`ma forest, a forest area near the city of Lytkino, a forest area near the village of Radiotsentr, and Shishkin Forest) were selected mainly in the south and north of the city at a distance of 8–40 km from the nearest urban study point (Fig. 1). The choice of forest parks is due to the presence of the texture-differentiated soddy-podzolic soil with a reference profile, the dominant zonal type for automorphic positions on loams under mixed forests. Preliminarily, the study sites with loamy soil-forming rocks were selected according to the map [8]. Then, in the field, reconnaissance of the selected study objects was carried out, 40–50-cm soil test pits were dug (to the upper part of the BT textural horizon of the soddy-podzolic soil) according to the classification and diagnostics of soils in Russia [16].

Criteria for selecting research objects: (1) homogeneity of the composition of soil-forming rocks (mantle and moraine loams), (2) flat territory without signs and potential danger of erosion processes (slope ≤5°), (3) similar species composition of the forest stand, (4) zonal soil type with natural arrangement of genetic

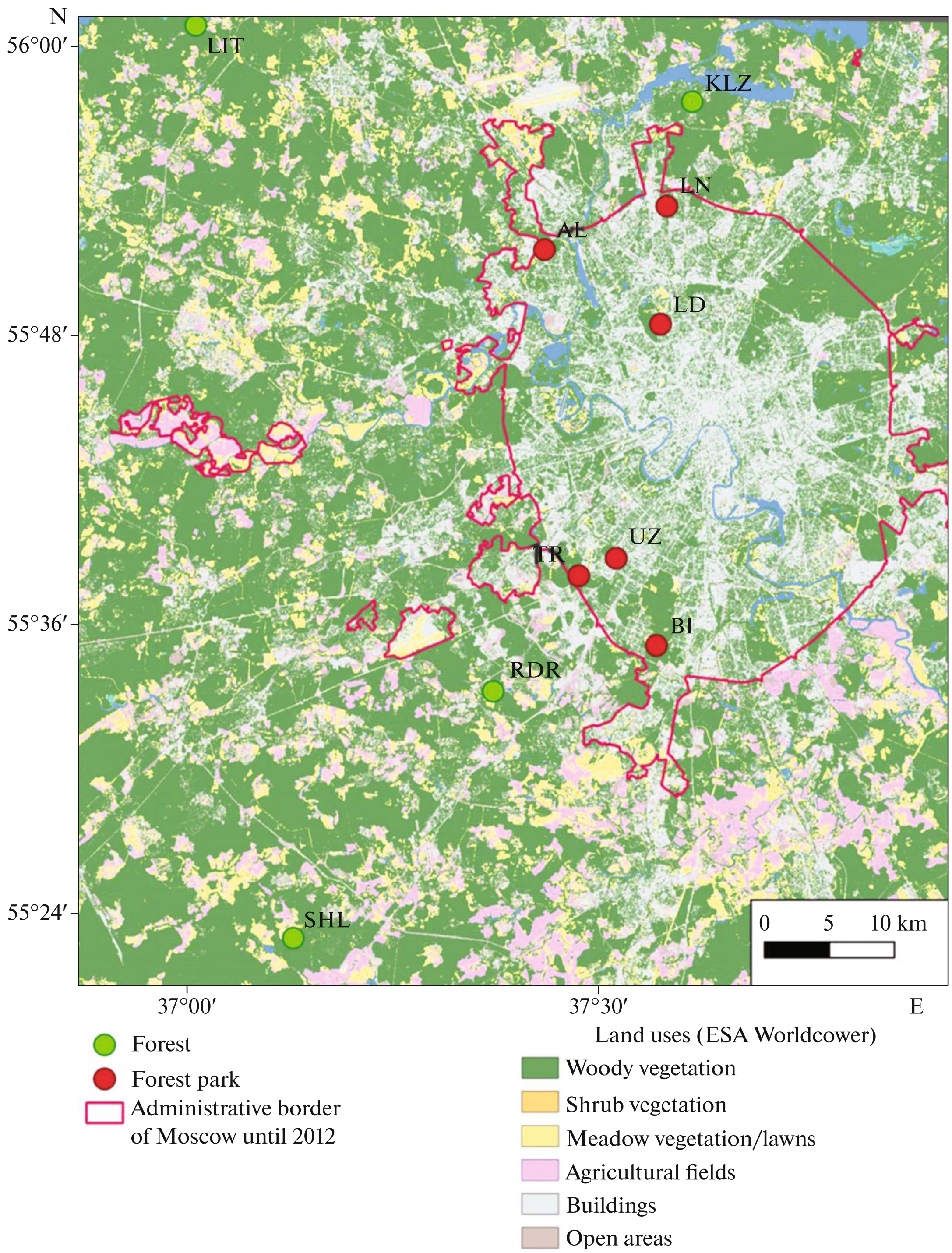

Fig. 1. Schematic map of the location of study objects: urban forest parks (LN is Lianozovskii park; AL is Aleshkinskii forest; LD is Forest Experimental Dacha; UZ is South–Western forest park; TR is Troparevskii landscape reserve; BI is Bitsevskii forest park) and suburban forests (LIT is forest area near Lytkino; KLZ is Klyaz`ma forest; RDR is forest area near the village of Radiotsentr; SHL is Shishkin Forest).

horizons, and (5) the age of the forest stand of at least 60 years.

In 2020, five flat plots (10 × 10 m) were selected in different parts (north, south, west, east, and the center) of forest parks and suburban forests. Vegetation was described at each plot, namely forest stand species composition, crown and undergrowth density, grass layer and litter projective cover, dominant grass layer species. Mixed topsoil samples were taken from the upper (0–10 cm) layer using an envelope (five points) sampling pattern. Additionally, topsoil samples were taken to determine bulk density. A total of 50 study points were selected: 30 were in forest parks (6 × 5 plots) and 20 (4 × 5 plots)—in suburban areas. Geographical coordinates and descriptions of the vegetation at the study sites are given in Table S1.

Fresh soil samples were delivered to the laboratory and sieved through a 2-mm mesh size to exclude roots and solid inclusions. The samples were divided into two parts: (1) fresh wet samples for microbiological analysis (storage at +4°C) and (2) dried to an air-dry state for physical and chemical analyses.

Methods. *Vegetation cover.* The crown density of trees and shrubs, the projective cover of the grass layer and leaf litter were visually assessed as a percentage of the total area of the selected plot. The dominant tree species and the composition of the grass layer are given in Table S1.

Soil chemical properties. The contents of total carbon and nitrogen were determined by IR spectroscopy after dry combustion in an oxygen flow (1100°C; analyzer CHNS-932 LECO Corg, USA); then, the C/N ratio was calculated. The contents of total P, K, Mn, Ca, and heavy metals (Pb, Cu, Ni, Zn) were measured using a portable X-ray fluorescence analyzer (Olympus Vanta C, USA). The contents of ammonium ($N\text{-}NH_4^+$) and nitrate ($N\text{-}NO_3^-$) nitrogen in the soil were determined according to GOST (State Standard) 26489-85 and GOST 26951-86, respectively. The contents of available forms of phosphorus (P_{av}) and potassium (K_{av}) were analyzed by the Olsen method (LibraS6 spectrophotometer, Biochrom Ltd., Great Britain and FPA-2-01 flame photometer, Russia). Dissolved organic carbon and dissolved total nitrogen were considered available (C_{av}, N_{av}) for microorganisms and were determined in a 0.05 M K_2SO_4 extract from soil samples (5 g soil and 20 mL solution) using a Shimadzu TOC-VCPN analyzer (Shimadzu Corp., Japan) [60]. The pH values were measured in the soil water (soil : water = 1 : 2.5) suspension by the potentiometric method (Expert-pH pH-meter, Russia).

Soil physical properties. Under field conditions, soil sample from the upper 10-cm layer was placed into a metal cylinder (153.86 cm^3) with preserving its natural structure. The obtained sample was dried (105°C, 8 h), and its weight was determined (GOST 28268-89), and then the soil bulk density was calculated. The particle size distribution of soil samples was determined in soil water suspensions after ultrasonic dispersion by laser diffraction using a Microtrac S3500 Bluewave diffractometer (USA) [15].

Soil microbial properties. The content of soil microbial biomass was determined by the methods of substrate-induced respiration (SIR) and fumigation-extraction (FE). The SIR method makes it possible to determine the carbon content of microbial biomass (S_{mic}-SIR) from the respiration response (CO_2 formation) of a soil sample enriched with glucose [2, 19]. The FE method is based on fumigation of a soil sample with chloroform (24 h) to destroy its microbial cells, followed by extraction with a 0.05 M K_2SO_4 solution [25, 45]. A soil sample without chloroform treatment (nonfumigated) is considered as a control. In the extractable solution from the fumigated and nonfumigated samples, the C and N contents were determined by a Shimadzu CN analyzer [58]. The C_{mic} content and microbial biomass nitrogen (N_{mic}) were calculated based on the difference between the contents of dissolved C and N in the fumigated and nonfumigated samples, divided by an empirical coefficient (0.45 and 0.54, respectively), taking into account their "incomplete transfer" from the soil to the solution [25, 49].

To determine the phosphorus content of microbial biomass (P_{mic}), soil (3 g) was placed into two 50-mL test tubes and 30 mL of distilled water was added to obtain soil suspension (soil : water = 1 : 10). An anion-exchange membrane and 0.3 mL of chloroform (fumigated sample) were added to one test tube with soil suspension, only 0.3 mL of water (nonfumigated sample) was added to the other tube, then they were tightly closed with lids, and placed on a laboratory shaker (200 rpm) for 24 h at room temperature. The surface of the membrane (551642S, VWR International, Darmstadt, Germany, the total area of each is about 8 cm^2) is saturated with a weak acid anion (bicarbonate), which contributes to the absorption of P from the suspension [26, 51, 73]. After this treatment, the membrane was removed from the suspension, washed with distilled water, transferred to 50 mL of a 0.25 M H_2SO_4 solution, and placed on the shaker (3 h) to isolate P into the solution. The P content in fumigated and nonfumigated solutions was determined by an inductively coupled plasma emission spectrometer (Avio 200, No. 68141-17 Singapore "Perkin Elmer Singapure Pte. Ltd.", 2019). The P_{mic} content was calculated from the difference in values for the fumigated and nonfumigated soils and corrected for the soil-specific extraction factor, which was 0.30 for the studied soils [22]. The use of such membranes makes it possible to avoid active sorption by the surface of clay minerals and organomineral complexes of the soil of phosphates released as a result of fumigation.

The proportion of C_{mic}, N_{mic}, and P_{mic} in the total content of these elements (C_{mic}/C, N_{mic}/N, P_{mic}/P, %) in the soil was determined. The ratio C_{mic}-SIR/C (%) was also calculated.

The CO_2 production by the soil (basal respiration, BR), which characterizes the potential rate of soil OM mineralization by microorganisms, was estimated at 22°C [46]. The specific respiration of microbial biomass (qCO_2 = BR/C_{mic}-SIR) and the specific rate of mineralization of total soil carbon (BR/C) were calculated.

The microbial response to addition of various organic substrates illustrating catabolic activity of the soil microbiome was assessed by the MicroResp™ technique [27, 62]. Soil samples (~0.5 g each) were placed into deep wells (945 μL) of a plate (96 wells) and added substrates (14 in total) of different groups: carbohydrates (D-glucose, D-fructose, D-galactose), carboxylic acids (L-ascorbic, citric, oxalic), amino acids (glycine, L-leucine, L-arginine, α-aminobutyric and L-aspartic acids), amino sugars (N-acetylglucosamine), and phenolic acids (lilac and vanillic). Each substrate was added to three wells of the plate (3 replicates). Then, the prepared indicator agar gel containingcresol red, potassium chloride and sodium bicarbonate was placed in the wells of another appropriate upper plate (well volume was 450 μL). The deep wells plate with soil was covered with a plate with indicator gel and incubated for 6 h at 25°C. During incubation, CO_2 released from the soil reacts with the indicator gel, changing its color (from pink to yellow). The color change of the gel was measured on a microplate reader (FilterMax F5, absorbance, λ = 595 nm). Absorption units were converted to the respiration response of the microbiome in μg C/(g soil h) [62].

The functional diversity of soil microorganisms was additionally assessed by the Shannon`s index: $H_{CLPP} = -\sum pi \times \ln pi$, where *pi* is the ratio of the response of the microbial community to the application of an individual substrate to the sum of respiration response to all substrates [62].

Microbiological assays were performed on preincubated soil samples (60% of total water capacity, 72 h, 25°C) [39].

Statistical analysis of data. Physical and chemical parameters were evaluated in duplicate, microbiological—in triplicate, the data were calculated on the weight of dry soil (105°C, 8 h). Spatial variation of soil and vegetation properties in forest parks and suburban forests was estimated by the variation coefficient (CV), which was determined as the ratio of the standard deviation to the mean value, expressed as a percentage. The significance of differences in the physical, chemical, and microbial properties of soils between forests and forest parks was assessed by the Student's test modified by Welch (Welch's *t*-test). The principal component analysis (PCA) was performed to generalize and visualize the spatial variation of the studied soil physical, chemical, and microbial properties. Preliminary preparation of experimental data for the PCA included their normalization (taking the logarithm) and centering. To identify the factors of spatial variation of the studied microbial properties, the simple linear regression analysis was performed, before which all experimental data were adjusted to a normal distribution by logarithm transformation. Statistical processing and visualization of experimental data were performed in the R 4.0.4 (R Core Team 2020).

RESULTS

Vegetation cover, physical and chemical properties of the soil. Data on vegetation and physical and chemical properties of soils in urban forest parks and suburban forests are given in Table 1. The density of tree crowns in forest parks turned out to be significantly greater than in natural analogues; however, the projective cover of leaf litter was smaller. In urban forest parks, in contrast to forests, higher values of soil bulk density, pH, and nitrate nitrogen ($N\text{-}NO_3^-$) were noted. The content of ammonium and total N in the soil of the studied forest parks and forests did not differ significantly. It was expected that the content of heavy metals and calcium (Ca) in the soil of the forest parks would be higher compared to that of the suburban forests, and the excess for Ni, Zn, Pb, and Cu averaged 22, 26, 30, and 32%, respectively; for Ca, 12%.

The spatial variability of most of the studied soil properties in forest parks and suburban forests was relatively low (CV was 4–54%, 25% on average), except for the content of $N\text{-}NO_3^-$ in the soil (CV was 79–106%). The PCA made it possible to generalize and visualize the change in vegetation and soil properties relative to one another in the studied forest parks and forests. The first two axes, which were associated with the greatest variation in the experimental data, explained in total only 39% of the total variance for all the studied properties (Fig. 2). The distribution of study points along axis 1 was associated primarily with the variation in the content of heavy metals, Ca, and total C in the soil (correlation with the r^2 axis = 0.53–0.82); along axis 2, with changes in the content of silt and sand, as well as the pH value (r^2 > 0.47–0.59). In general, the ordination of data in the space of two axes demonstrated the absence of a clear differentiation between forest parks and their natural analogues in terms of the studied vegetation and soil properties. However, in forest parks, compared with suburban forests, there was a large variation in the content of heavy metals, Ca, and C in the soil (distribution of points along axis 1).

Thus, the impact of urbanization on changes in the ecological conditions of forest ecosystems was manifested mainly in a decrease in the proportion of forest litter, an increase in soil bulk density, pH value, the content of $N\text{–}NO_3^-$ and heavy metals. In addition, in the forest parks, the spatial heterogeneity of the content of heavy metals, Ca, and total C in the soil increases, which is obviously associated with the uneven anthropogenic load in the forest parks in dependence on the distance from the source of pollution, the intensity of recreation, and the litter extraction activities. These factors can significantly affect the soil microbial community functioning, and hence the intensity and direction of soil processes associated with the cycles of biophilic elements.

Table 1. Vegetation, soil (0–10 cm layer) physical and chemical (available forms) properties and coefficient of their spatial variation (CV) in Moscow forest parks and their natural analogues. Mean ± standard deviation, mean difference is significant at $^{***}p \le 0.01$, **0.05, *0.1 (Welch's t-test).

Indicator	Units	Forest (n = 20)	CV, %	Forest park (n = 30)	CV, %
		Vegetation			
Tree canopy	%	61.8 ± 9.5	15	70.5 ± 14.5***	21
Undergrowth canopy		11.7 ± 12.1	103	17.7 ± 13.1	74
Litter		87.2 ± 15.9	18	76.3 ± 15.1**	20
Grass layer		55.2 ± 27.6	50	43.3 ± 25.5	59
		Physical properties			
Bulk density	g/cm^3	0.82 ± 0.14	16	0.91 ± 0.20*	22
Sand (0.05–2.00 mm)	%	21.8 ± 6.6	30	22.5 ± 6.5	29
Silt (0.002–0.05 mm)		70.5 ± 5.7	8	69.5 ± 5.7	8
		Chemical properties			
pH		5.0 ± 0.2	4	5.2 ± 0.4*	7
C	g/kg	36.3 ± 4.2	11	38.0 ± 8.2	21
N		2.5 ± 0.3	12	2.7 ± 0.6	22
K		15.8 ± 0.6	4	15.8± 1.1	7
C/N		14.6 ± 1.5	10	14.3 ± 1.2	8
$N-NO_3^-$	mg/kg	12.4 ± 13.2	106	26.2 ± 20.7***	79
$N-NH_4^+$		26.2 ± 10.8	41	24.2 ± 12.7	52
P		631.4 ± 152.0	33	696.5 ± 217.1	28
C_{av}	μg/g	167.3 ± 78.9	47	156.5 ± 74.5	48
N_{av}		129.6 ± 72.2	56	97.5 ± 33.4*	34
P_{av}		7.5 ± 1.9	25	9.3 ± 3.9	42
K_{av}		47.6 ± 19.7	41	56.9 ±23.2	40
Mn	mg/kg	1052 ± 358	34	991 ± 249	25
Cu		16.7 ± 2.8	17	24.7 ± 6.2***	25
Pb		17.6 ± 2.7	15	25.1 ± 8.3***	33
Ni		19.9 ± 2.8	14	25.4 ± 6.3***	25
Zn		58.1 ± 6.9	12	78.9 ± 20.8***	26
Ca		4839 ± 417	9	5472 ± 1160***	21

The ratio of CNP in microbial biomass to their content in soil. In the soil of forest parks, a significant decrease in the C_{mic}-SIR and C_{mic} values (by 14 and 25%) was revealed compared to the suburban forests, while no significant changes in N_{mic} and P_{mic} were noted (Table 2). In forest parks, compared to forests, a significant reduction in the share of C_{mic} and N_{mic} in the total soil pool of these elements (C_{mic}/C and N_{mic}/N) was found, which might indicate their lower availability for microorganisms. This conclusion was also confirmed by the significantly lower rate of BR and the specific mineralization of soil C (BR/C) in the soils of forest parks compared to their natural analogues (a decrease by 24–26%). It is important to note that the P availability for microorganisms (P_{mic}/P) in urban soils did not differ compared to forest soil. The specific respiration of microbial biomass (qCO_2) for soils of forest parks and suburban forests did not differ as well.

To generalize and display the spatial variation of the studied microbial properties of soils in forest parks and suburban forests, the PCA were performed (Fig. 3). The first two axes accounted for a total of 66% of the total variance of the experimental data. The distribution of study sites along axis 1 is mainly associated with changes in C_{mic}, N_{mic}, C_{mic}/C, and N_{mic}/N in soils (correlation with the r^2 axis = 0.81–0.89); along axis 2, with BR and BR/C (correlation with the r^2 axis = 0.57–0.69). In general, these parameters were characterized by equally high variation within both suburban

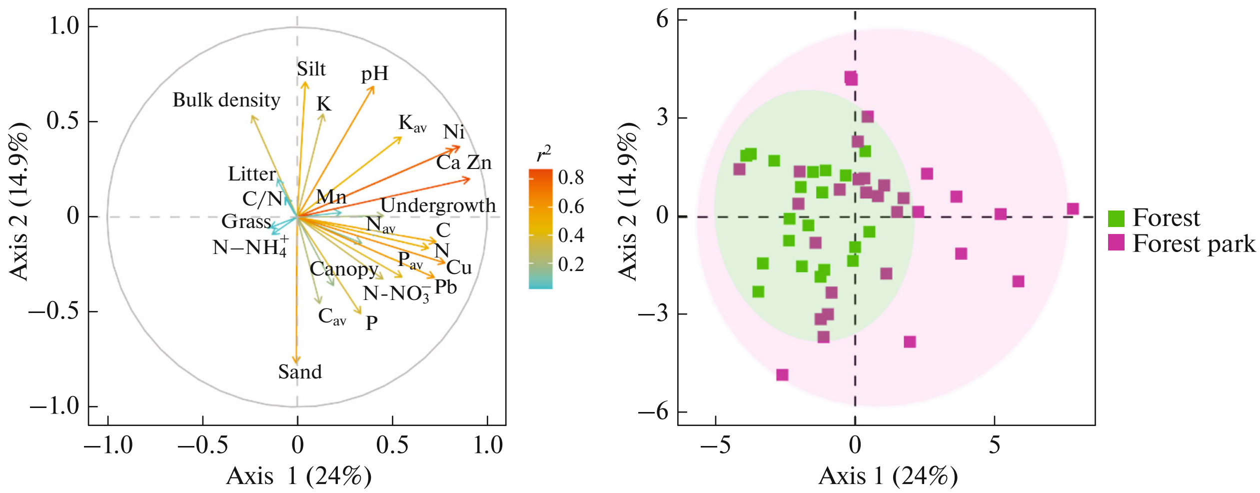

Fig. 2. Results of principal component analysis for vegetation and soil (0–10 cm) properties of the studied forest parks and suburban forests ($n = 50$; on the left, correlation diagram; ordination of study sites is on the right).

forests and forest parks. Therefore, the data ordination did not show a clear differentiation between forest parks and suburban forests.

The regression analysis revealed that the main drivers of spatial variability in soil microbial parameters (C_{mic}, N_{mic}, C_{mic}/C, N_{mic}/N, BR, BR/C) were the abundance of plant litter and the content of available C in the soil (13–35% of the explained dispersion, Table 3). Significant factors were the contents of nutrients (C, N, N_{av}, P), heavy metals (Cu, Pb), and silt in the soil, and the pH value. The negative effect of heavy metals (Cu, Pb) on the rate of soil organic C mineralization (BR/C) and the deterioration of the ecophysiological parameter of the soil microbiome (qCO_2) with an increase in the P content and the acidity values were found.

Thus, the urbanization of forest areas in the studied bioclimatic zone led to a decrease in the C content of soil microbial biomass, its respiration activity, and the availability of soil C and N to microorganisms (C_{mic}/C, N_{mic}/N, BR/C). These changes are mainly associated with a reduction in the input of plant residues into the soil in the form of forest litter and, as a result, a decrease in the content of available forms of C in it. In addition, a decrease in the availability of soil carbon to microorganisms (BR/C) in urban condi-

Table 2. Microbiological parameters (C_{mic}-SIR, microbial biomass carbon according to the substrate-induced respiration method; C_{mic}, N_{mic}, and P_{mic}, microbial elements according to the fumigation–extraction method; BR, basal respiration) and their relationship to the soil content of C, N, and P for the suburban forests and forest parks of Moscow (mean ± standard deviation, the difference between the means is significant at $^*p \le 0.1$, $^{**}p \le 0.05$, $^{***}p \le 0.01$).

Parameter	Units	Forest ($n = 20$)	CV, %	Forest park ($n = 30$)	CV, %
C_{mic}-SIR	μg/g	990 ± 266	27	849 ± 296*	35
C_{mic}		930 ± 366	39	694 ± 249**	36
N_{mic}		135 ± 84	62	97 ± 71[a]	73
P_{mic}		52 ± 31	60	56 ± 35	62
C_{mic}- SIR/C	%	2.72 ± 0.68	25	2.30 ± 0.90*	39
C_{mic}/C		2.54 ± 0.84	33	1.85 ± 0.58***	31
N_{mic}/N		5.40 ± 3.24	60	3.69 ± 2.46**	67
P_{mic}/P		8.34 ± 6.15[b]	74	8.63 ± 6.47	75
BR	μg C/(g h)	1.46 ± 0.44	30	1.11 ± 0.37***	33
BR/C	μg C-CO_2/(g C h)	40 ± 10	25	29 ± 7***	24
qCO_2 (BR/C_{mic}-SIR)	μg C/(mg C_{mic} h)	1.57 ± 0.58	37	1.40 ± 0.42	30

[a]extremely high N_{mic} (564 μg/g) is excluded (point 2, Forest Experimental Dacha); [b]extremely high P_{mic}:P (37.4%) is excluded (point 3, Shishkin forest).

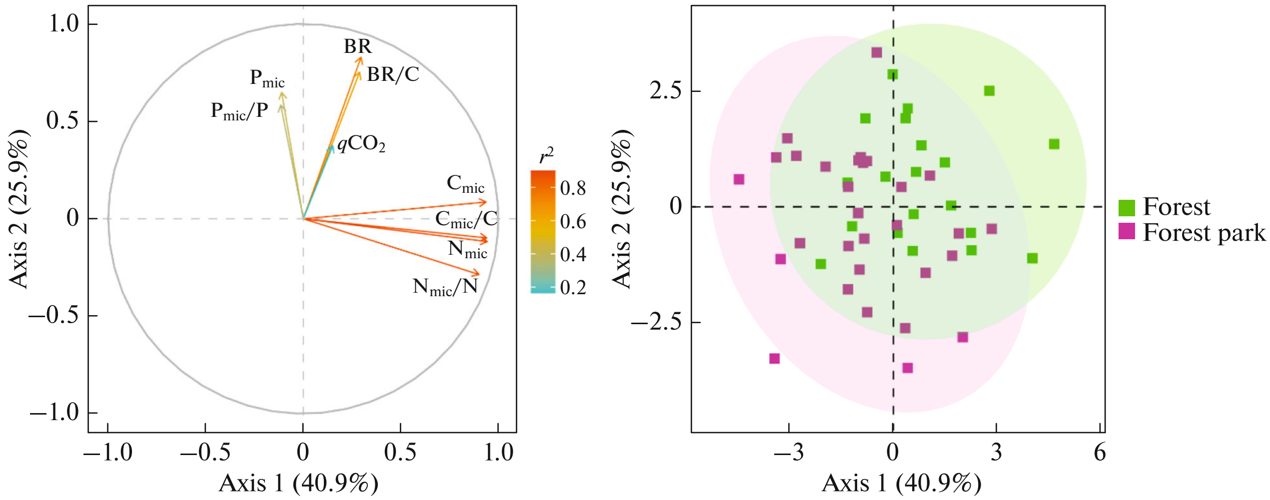

Fig. 3. Results of the principal components analysis for the soil microbial properties (0–10 cm layer) of the studied forest parks and suburban forests ($n = 50$; the correlation diagram is on the left; the ordination of study sites is on the right).

tions was associated with the negative effect of an increased concentration of heavy metals (Cu, Pb). It should be noted that all the detected changes of soil microbial properties in forest parks compared to suburban forests were mainly associated with their carbon and nitrogen cycles, while no significant changes were found for the phosphorus cycle (P_{mic}, P_{mic}/P).

Catabolic activity of the soil microbiome. The respiration response of microorganisms to the application of C-containing substrates in the soil of forest parks turned out to be comparable with that of suburban forests, which confirmed our second hypothesis (Figs. 4a–4f). The highest microbial response was noted for the carbohydrates (median was 13.6 and 12.0 μg C/(g h) for forests and forest parks) and carboxylic acids (15.2 and 14.2 μg C/(g h); the lowest response was observed for amino acids (5.1 and 5.2 μg C/(g h), phenolic acids (3.0 and 3.4 μg C/(g h), and amino sugars (4.6 and 5.5 μg C/(g h). Soil microbiome functional diversity index (H_{CLPP}) indicating the uniformity of utilization of the studied range of organic compounds by microorganisms did not differ significantly between forest parks and suburban forests (Fig. 4f).

The regression analysis showed that the general drivers of the microbial response to the addition of carbohydrates, amino acids, and amino sugars into the soil are the contents of K_{av} and Ca (12–32% of the explained variance, Table 4). Soil pH and the content of heavy metals (Ni, Zn) were additional factors of the microbial response to carbohydrates and amino sugars. In general, the identified patterns were characterized by a directly proportional relationship (with an

Table 3. Variation explained (%) in the studied soil microbial properties of forest parks and suburban forests ($n = 50$; significant factors at $p \leq 0.01$ are in bold italics; simple linear regression).

Parameter	C_{av}	N_{av}	P_{av}	C_{av}/C	N_{av}/N	P_{av}/P	BR	BR/C	qCO_2
Litter	***21.5***	8.9	9.2	***12.6***	4.0	9.9	***21.6***	***14.8***	0.0
C	10.4	1.3	6.4	3.0*	1.5*	1.6	***25.2***	0.0*	3.8
N	3.4	0.0	5.4	6.8*	7.8*	1.2	***26.3***	0.5	8.5
P	0.1	0.1	0.0*	1.4*	0.0	***13.8****	1.7	0.0*	***25.1***
C_{av}	***31.3***	***35.2***	0.4	***22.9***	***27.9***	0.0	1.2	0.0	6.0
N_{av}	11.0	2.9	2.6	2.9	0.2	1.0	***18.6***	8.7	6.0
pH	0.0*	0.1*	0.8	0.6*	1.0*	2.8	1.3	0.5	***28.7****
Cu	0.6*	0.2	0.2	7.8*	0.2*	0.4*	2.2*	***16.7****	0.7
Pb	0.0*	2.1	0.0	4.1*	0.7	0.8*	3.5*	***19.0****	0.1
Ca	2.9	0.3	7.9	2.4*	3.0*	4.5	***13.6***	0.2	4.0*
Silt	0.3*	4.8	9.2	0.2*	4.7*	***12.6***	0.6	1.2	9.2*

*Negative regression dependence.

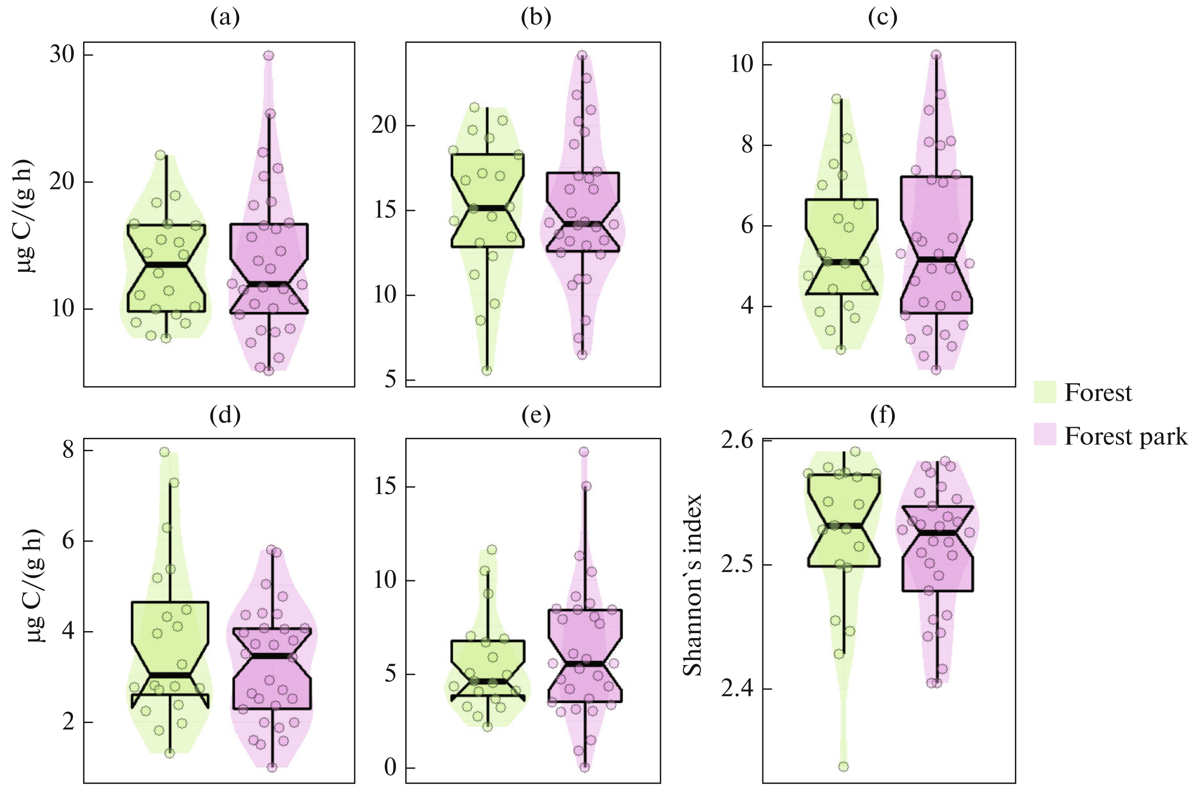

Fig. 4. Distribution of the respiration response of the soil microbial community in urban forest parks and suburban forests (averaged value) to the input of organic substrates: carbohydrates (a), carboxylic acids (b), amino acids (c), phenolic acids (d), amino sugars (e); Shannon`s index (f).

increase in the value of the factor, the microbial response increases). It should be noted that no significant drivers ($p \leq 0.01$) for the microbial response to carboxylic and phenolic acids were identified.

Thus, the transformation of natural forests into urban forest parks did not lead to significant changes in the respiration response of soil microorganisms to the input of various available C sources. The results obtained confirmed the earlier conclusion about the key role of soil C and its available form, which determined the mineralization of OM and the immobilization of biophilic elements in the microbial biomass.

DISCUSSION

Soil biophilic elements and their availability to microorganisms under urbanization. No differences in the content of total pools of biophilic elements in the soils of urban forest parks and suburban forests were found, which did not confirm our assumptions about their imbalance under urbanization conditions (Table 1). In urban forest parks, there was a decrease in their availability to soil microorganisms, i.e., a significant decrease in the C_{mic}/C and N_{mic}/N ratios. Such changes were obviously associated with a smaller

Table 4. Variation explained (%) in the rate of microbial utilization of various groups of organic compounds in the soil of the studied forest parks and suburban forests ($n = 50$; significant factors at $p \leq 0.01$ are in bold italics; simple linear regression)

Parameter	Carbohydrates	Carboxylic acids	Amino acids	Amino sugars	Phenolic acids
Litter	3.1	1.6	8.3	3.3	8.0
C	5.9	0.5	8.6	***14.0***	0.0
K_{av}	***17.9***	4.5	***12.7***	***22.9***	0.0*
pH	***18.3***	1.1	7.3	***24.5***	3.5*
Ni	***14.3***	3.7	7.1	***18.2***	1.4*
Zn	9.4	1.7	3.2	***16.8***	1.1*
Ca	***22.5***	2.9	***12.3***	***31.8***	0.7*

*Negative regression dependence.

amount of forest litter in urban conditions (Table 3) because of recreational loads (compaction, systematic litter exclusion, etc.) and a regular decrease in the content of soluble forms of C—important source of available nutrients and energy for soil microorganisms—in the soil [40]. The content of available C and N compounds in the soil is a certain balance between their mineralization and immobilization in microbial biomass [62]. Therefore, periodic enrichment of the soil with an easily accessible organic substrate, such as root exudates or decomposition products of plant residues (for example, when leached out from the forest litter), leads to an increase in the microorganisms' abundance [41]. The resulting priming effect enhances the microbial mineralization of more complex soil OM [53], which, in turn, can lead to an increase in the proportion of C_{mic} and N_{mic} in it. Therefore, a greater input of plant residues into the soil of the studied suburban forests also contributes to a more accelerated mineralization of their soil OM. Thus, decreasing natural enrichment of soil by OM in Moscow forest parks due to the litter exclusion is the main "trigger" for a decrease in soil biogenicity (an abundance of microbial biomass) and the intensity of changes in C and N soil cycles. However, the reverse trend—intensification of soil cycles of biogenic elements—can also be observed during urbanization, as evidenced by the results of numerous studies [31, 38, 60]. Thus, for the urban forest parks of Florida (USA), an increased content of soil microbial biomass and its mineralization activity were noted compared to those of natural forests [38]. Similar trends were also identified for urban forest parks in China [31, 60]. Moreover, in [60], the key role of soluble pools of soil OM in changing the content of its microbial biomass (C_{mic}; N_{mic}) in urban forest parks was shown, which is consistent with the results of our study. We did not find a significant change in the soil properties of urban forest parks associated with the P cycle, although an increase in its content in the urban soils of Moscow was expected. Phosphorus can enter urban ecosystems with food residues and various anthropogenic wastes [65], and its excessive content can become a source of urban water pollution. Previously, we have shown that an additional input of P is typical for residential and industrial areas of Moscow, which is associated with the use of mineral fertilizers and organic substrates containing a large amount of biogenic elements [47, 48]. However, the practice of maintaining forest park zones in this city, apparently, does not imply such an impact [13].

Respiration activity of soil microbiome and factors of urbanization. As expected, the rate of basal soil respiration (mineralization activity) was significantly lower in the forest parks of Moscow compared to the suburban forests, which was consistent with a decrease in the CN content in microbial biomass (Table 2). We assumed that the additional input of NP into the soil of urban forest parks would be the main driver of a decrease its microbial mineralization activity. Indeed, in the soil of urban forest parks, an increased content of nitrate nitrogen was revealed compared to the suburban forests (Table 1), which we attributed to its additional input from plants and vehicles. This fact was also noted for other cities in the temperate and tropical climatic zones [11, 34]. The effect of additional nitrogen input on the dynamics of soil C is discussed in scientific literature [34, 67]. Soil enrichment with nitrates in the city of the tropical zone contributed to a decrease in the activity of hydrolytic enzymes [34], which may be the reason for the slowdown in the respiration activity of the microbiome, which was noted in various studies [23, 61]. There are data on the absence of the effect of additional nitrogen input on soil microbial activity [17, 24]. In our study, the decrease in the respiration activity of the microbial community in the forest parks of Moscow was largely associated with a decrease in the amount of plant litter (Tables 1, 3) and, as a result, a decrease in the share of the labile pool of soil OM. At the same time, the input of various readily available C sources into forest park soils stimulated the activity of the microbiome, which corresponded to that of natural forest soils (Fig. 4). This indicated a high potential of the microbial community in the soils of urban forest parks to the mineralization of soil OM, provided there is a sufficient input of available forms of carbon, for example, as a result of the decomposition of leaf litter or root exudates. The availability of soil OM, which was estimated based on the ratio of respiration activity to the soil C content (BR/C), was significantly lower in the forest parks than in the suburban forests (Table 2), which was determined by the increased content of heavy metals, Cu and Pb (Table 3). The effect patterns of heavy metals on the microbial activity of urban soils is ambiguous in the scientific literature [4, 28], since it is determined by their concentration, the proportion of bioavailable forms, the duration of pollution, the stability of the microbial community, and other factors [43, 44]. In a short-term laboratory experiment, it was shown that the input of heavy metals (Cd, Cu, Pb) into Technosols in amounts from 0 to 200 of their approximate permissible concentration (Hygienic Standard 2.1.7.2511-09) contributed to an increase in microbial respiration and respiration per unit C of microbial biomass [4]. Other authors, on the contrary, noted a decrease in microbial respiration per unit soil C contaminated with heavy metals in urban environment [28]. In polluted soils, the microbiome is believed to adapt to stress conditions [20], which requires certain energy costs. Consequently, the energy for the growth of microorganisms and the synthesis of enzymes for anabolic and catabolic processes is spent more economically [28, 50]. Therefore, there is reason to believe that C removal (the main source of energy) with leaf litter in urban forests can significantly reduce the resistance of their soil microbial community to heavy metal pollution [52].

Thus, our first hypothesis about the decrease in respiration activity and the share of CN of microbial biomass in the total pool of these elements in the soil of forest parks was confirmed. However, this change was associated with a decrease in the amount of forest litter and the available C content in the soil and was not related to an additional input of N and P, as was assumed. The key role of C in the functioning of urban soils was also confirmed by the activation of the microbial community after its additional input as available organic substrates, which fully confirmed our second hypothesis.

CONCLUSIONS

Forest parks are a key component of urban green infrastructure. Their ecological and environmental significance is determined by the larger area and less significant anthropogenic transformations in comparison with other urban green areas. Occupying an intermediate position between urban and natural ecosystems, forest parks are traditionally used for environmental monitoring and assessment of the state of vegetation and soils. Recreational load, measures for the maintenance and care of urban areas lead to changes in the fluxes of matter and energy in the ecosystems of forest parks, which significantly affects the content of biophilic elements and their microbial availability.

A comprehensive comparative analysis of the soils of Moscow forest parks and the corresponding suburban forests revealed close values of many traditionally measured physicochemical parameters (particle size distribution, C/N, P and K contents), which in urban conditions is accompanied by a significant (by 30–60%) reduction in basal respiration and microbial availability of C and N. At the same time, the ability of the soil microbial community of the studied ecosystems to decompose certain organic compounds, expressed by its respiration response, did not differ significantly, and the main limiting factor of this ability was a decrease in the input of litter and the content of available C in forest parks. Thus, the impact of urbanization on forest ecosystems, first of all, led to a change in the microbial indicators of the soil cycle of C and N, but not P. The revealed patterns are determined, apparently, by the existing practice of maintaining and caring for green spaces, which regulates, in particular, the need to clean up and dispose fallen leaves. The results obtained cast doubt on the application of this approach in urban forest parks. In the context of the current topic of maintaining the carbon balance and achieving carbon neutrality, modern approaches based on the preservation of the natural processes of input, mineralization, and microbial absorption of biophilic elements seem to be more promising.

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CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

SUPPLEMENTARY INFORMATION

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Table S1. Geographic coordinates and characteristics of wood stand and herb layers in forest parks of Moscow (North, South, East, West) and background areas (2020).

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