Експериментальні праці

doi: https://doi.org/10.15407/microbiolj80.03.003

UDC 579.266, 502.4, 631.461:633.34

SOIL MICROBIOME OF PRIMEVAL FOREST ECOSYSTEMS IN TRANSCARPATHIA

L. Yu. Symochko¹, A.V. Kalinichenko^{2,3}

¹Uzhhorod National University, 32 Voloshyna str., Uzhhorod, 88000, Ukraine e-mail:lyudmilassem@gmail.com ² University of Opole, 7-9 Dmowskiego Str., Opole, 45-365, Poland ³ Poltava State Agrarian Academy, 1/3 Skovorody str, Poltava, 36021, Ukraine e-mail: akalinichenko@uni.opole.pl

Objective. The aim of this study was to investigate the soil microbiome of primeval forest ecosystems, namely the structure of microbial communities, the number of major ecological-functional groups, functional parameters such as: soil toxicity, as well as enzymatic activity of the soil by the level of catalase and invertase. To analyze the successional processes in the soil microbiocenosis due to the influence of endogenous and exogenous factors. To estimate the integrity of microbial communities in different edaphotopes of primeval forest ecosystems. Methods. Microbiological studies of soil were carried out according to generally accepted methods in soil microbiology. Enzymatic activity of the soil: catalase - was determined by gasometric method and invertase by colorimetric method. Biotesting was used to determine soil toxicity. An assessment of the integrity of microbial communities in different edaphotopes of primeval forest ecosystems was carried out using correlation-regression analysis. Statistical analyses were performed by using Statistica 10 software. Basic descriptive statistics was calculated, that is, the arithmetic mean and standard deviation. Results. Anthropogenic impact: deforestation and soil compaction led to significant negative changes in the structure of soil microbiome. It was observed in the decreasing of the functional biodiversity and the number of microorganisms. In particular, among the disturbed edaphotopes, the level of soil toxicity was increased and also the processes of decomposition of organic matter were slowed down. The level of enzymatic activity of the soil was decreased and the integrity of microbial communities of the soil was violated. Conclusions. The influence of endogenous and exogenous factors in primeval forests ecosystems violates the integrity of the soil microbiome and leads to negative changes of its functional characteristics.

Keywords: microorganisms, soil, primeval forest, ecosystem, enzymatic activity, integrity of communities.

Consequently, the soil resources available for trees and their microbial associates are restricted. In this context, the stability and sustainable development of forest ecosystems rely on the recycling of nutrients contained into the falling leaves, dead roots as well as in the atmospheric deposits or on the weathering of the soil minerals. Soil microorganisms and their interactions play a central role in biogeochemical cycling in forest soils, as major actors for mineral weathering, decomposition and biomineralization of organic matter and pedogenesis. However, due to the high heterogeneity of this ecosystem and its richness, our knowledge on the soil microbiome remains limited [1,2,3]. Soil microorganisms are responsible for most biological transformations and

drive the development of stable and labile pools of carbon, nitrogen and other nutrients, which facilitate the subsequent establishment of plant communities [4]. Over half a century ago, Odum [5] identified mechanistic linkages between the successional dynamics of natural communities and the functioning of natural ecosystems. Specifically, as communities progress through succession, diversity is expected to increase and nutrients will become 'locked-up' in the biota, with consequences for the build-up of soil organic matter and closure of the mineral cycles. More recently, the interplay between aboveground and belowground biodiversity has emerged as a prominent determinant of the successional dynamics in biological communities. However, little is known about how changes in the soil biota contribute to the associated changes in ecosystem functioning [6].

In the complex and dynamic plant root interaction with the microbiome, both biotic and abiotic factors play critical roles for microbiome composition, richness, and diversity. Biotic factors, such as host genotypes, developmental stages and abiotic factors, such as temperature, soil pH, seasonal variation, and the presence of rhizospheric deposits, act as chemical signals for microbes and influence the microbiome community structure and function [7,8]. However, the extent to which both abiotic and biotic factors contribute to microbial communities is not fully understood [9,10]. Primeval forests are ideal ecosystems to study the interaction of bacteria, fungi and archaea with their abiotic environment [11]. Microbial communities can be considered as architects of soils and many ecosystem services that are linked to terrestrial ecosystems, including plant production, safeguarding of drinking water or carbon sequestration, are closely linked to microbial activities and their functional traits [12]. Soil microorganisms have been largely ignored by conservation efforts. However, their role in biogeochemical processes, their diversity and abundance, and their potential as repositories of valuable genetic information and metabolic products make them as important as animals and plants to the biosphere and human welfare. Study of authentic soil microbiota creates the necessary prerequisites for the conservation of microbial diversity and forming the base of the eco-microbiological monitoring.

Our main idea is to study soil microbiome and its functional characteristics of Uzhanskyi National Nature Park. As a model ecosystem we investigated primeval forest. The primeval forests as etalon ecosystems better combine above resistance and stability with high productivity biomass [13]. In the Transcarpathian region of Ukraine (south-west), the Uzhanskyi National Nature Park (Uzhanskyi NNP) offers a unique opportunity for studying the biodiversity and natural processes of primeval forest ecosystems, i.e. forests that have never been significantly modified by human activity.

Materials and methods. Materials of research were soil samples, which had been collected by envelope method from the virgin forests of Uzhanskyi National Nature Park at the deep 0-25cm. Uzhanskyi National Nature Park is located in the western part of Transcarpathia in the basin of the river Uzh and extends from the southwest of the village Zabrod (226 m above sea level) to north-east to Uzhotskyi pass (852 m above sea level). In 2007 primeval beech forests of Uzhanskyi National Nature Park were included into UNESCO World Heritage List "Primeval Beech Forests of the Carpathians and the Ancient

Beech Forests of Germany". The Uzhansky NPP is located in the southwest sub-region of the Atlantic-continental climatic region of the temperate zone, in the low-mountainous zone of moderate relative humidity. Absolute temperature maxima occur most often in July and August, and make up $+34 \circ C - +37 \circ C$, in some years fall on April - May. The absolute minimum temperatures are January-February and are characterized by -28 ° C -32 ° C. The total vegetation period in the region lasts 195 days. The average annual rainfall is 856 - 909 mm. There are mainly brown mountain-forest soils (brown-soils) in the park among them are two subtypes: dark brown and light-brown mountain-forest soils. Researches were conducted during 2012-2017 years. The reaction of the forest soil is slightly acidic. Humus is dominated by full-fatty acids, which are closely linked to the one and a half oxides of iron and aluminum. Sampling was carried out seasonally at different altitudes from 450 m to 650 m. Studies of soils were carried out at the Scientific Research and Educational Center of Molecular Microbiology and the Immunology of Mucous Membranes (Uzhhorod National University), Research Laboratory Monitoring of Water and Terrestrial Ecosystems of department entomology and biodiversity conservation (Uzhhorod National University) and at Laboratory of Department of Phytopathogenic Bacteria, (D.K Zabolotny Institute of Microbiology and Virology of the Academy of Sciences of Ukraine). The research was carried out within the framework of the complex theme "Eco-microbiological monitoring of various types ecosystems of the Carpathian region" №0116U003331 (state registration number).

Microbiological study of soil was performed in sterile conditions following the standard protocol [14]. All soil samples were analyzed within 24 hours. The method of serial dilution was used to obtain the suspension where microorganisms titre were $10^{\text{-}3}$ CFU/ml. - $10^{\text{-}5}$ CFU/ml. 100 μl of the soil suspension was evenly spread on the surface of the medium with a sterile spatula.

For the study we used the following media: Starch-ammonia Agar, Meat peptone Agar, Wilson-Blair Agar, Soil Agar, Agar-Agar, Fedorova Agar, Vinogradsky Agar, Ashbys Agar and Czapek Agar in 4 repetitions. Petri dishes with study material were incubated in the thermostat at 37°C for 48 hours in aerobic and anaerobic (Wilson-Blair Agar, Vinogradsky Agar) conditions. Petri dishes with Czapek agar were incubated in the thermostat at 28°C for 96 hours Toxicity of soil samples was determined by the standard method [15]. Enzymic activity of the soil: catalase - by gasometric method, invertase - by colorimetric method. An assessment of the integrability of soil microbial community in different edaphotops of virgin ecosystems was carried out using correlation-regression analysis [16, 17, 18].

All statistical calculations were performed with Statistica 10 and Excel for Windows-2010.

Results and discussions. Soil microorganism as a part of forest ecosystems plays an important role in sustainable development of forestry. They are highly sensitive to anthropogenic pressure, so changes of qualitative and quantitative composition of the soil microbiota – is an indicator of the environmental changing [19]. Due to the high sensitivity to changes in environment microorganisms serve as a convenient object of observation. They are in a

close contact with habitat and they are characterized by high rate of growth and reproduction. Biocenotic relations of trophic and topical types are decisive in edaphotope shaping of different type of ecosystems [20]. Due to this fact, the purpose of the research was to determine the number of different ecological-functional groups of soil microorganisms. Studies of the soil were taken from primeval ecosystems revealed general regularities of distribution of main ecological-functional groups of microorganisms, their population dynamics in different habitats. The most favourable conditions for the development and functioning of microorganisms were recorded in an edaphotop which was located at an altitude of 450 meters above sea level. It is highly connected to local temperature and water regime, as well as reserves of nutrients (organic origin) in the soil (Table 1).

Table 1 Microbial community composition in soils of primeval forest ecosystems (CFU/gr.ab.d.s.)

| $N_{ar{o}}$ | Biotopes, altitude above msl, m | Ammonifiers*106 | Spore forming bacteria *106 | Micromycetes*10³ | Actinomyces*103 | Bacteria wich are using mineral forms of nitrogen*104 | Anaerobic bacteria*10³ | Aerobic nitrogen fixing bacteria, % | Anaerobic nitrogen fixing bacteria *103 | Oligotrophic bacteria *106 | Oligonitrophic bacteria*104 | Pedotrophic bacteria*106 |
|-------------|---------------------------------|-----------------|-----------------------------|------------------|-----------------|---|------------------------|-------------------------------------|---|----------------------------|-----------------------------|--------------------------|
| 1 | 450 | 5,33 | 2,35 | 430 | 10,78 | 3,35 | 35,20 | 66,51 | 6,70 | 3,45 | 4,45 | 2,34 |
| 2 | 500 | 3,89 | 2,23 | 540 | 12,03 | 3,26 | 41,22 | 49,60 | 9,56 | 3,97 | 3,81 | 2,89 |
| 3 | 650 | 3,54 | 3,07 | 573 | 16,89 | 2,67 | 50,22 | 40,22 | 10,67 | 4,10 | 3,65 | 3,44 |
| 4 | 650.1 | 1,32 | 4,66 | 240 | 32,96 | 1,24 | 94,68 | 17,21 | 16,25 | 7,45 | 1,35 | 1,89 |
| 5 | 650.2 | 4,89 | 3,78 | 380 | 20,34 | 2,66 | 73,82 | 29,73 | 12,78 | 3,24 | 2,97 | 4,78 |
| S | SSD_{05} | 0,24 | 0,16 | 6,28 | 0,37 | 0,44 | 0,21 | 2,18 | 0,64 | 1,20 | 1,77 | 0,72 |

Footnote: 650.1- soil compaction; 650.2-cutting.

The number of ammonifiers at an altitude of 450 m amounted 5.33 million CFU/gr.ab.d.s. and at an altitude of 650 m - 3.01 million CFU/gr.ab.d.s., which indicate a significant enrichment of soil by organic matter of plant origin. Anthropogenic impact, namely soil compaction, was negatively influenced on the structure of soil microbiota. The content of oligotrophic microbiota significantly increased in the samples of this soil, but the number of ammonifiers and pedotrophes microbiota was minimal compared to the other surveyed edaphotopes.

The growth in the number of oligotrophes indicates a decrease in the supply of nutrients in the soil. Similar changes were recorded by us and our colleagues in the study of biodiversity of soil microorganisms in primeval ecosystems of the Carpathian biosphere reserve [21, 22].

With the creation of favourable conditions for competitive species of microbiota we can see changes in microbial cenoses, owing to the active competition of microorganisms. In edaphotopes that were not changed by direct human impact the dominate type of microorganisms was organotrophic microbiota. It should be noted that their percentage in the structure of groups was reduced by 36% with increasing of the height. At the same time the number of oligotrophes also increased with altitude. Significant negative changes in the structure of microbiocenosis of the soil can be the effect of anthropogenic influence. Violation of the integrity of the phytocenosis as a result of deforestation has led to the increase in the content of oligotrophes and pedotrophes, which indicates disruption of the normal flow of microbiological processes in the soil. It also influenced the decline in biodiversity of soil microorganisms. The most negative changes in the structure of soil microbial community were observed due to compaction. Nearly 80% of the studied ecological-functional groups of microorganisms were oligotrophes, which shows a significant deterioration of the ecological state of the soil. Soil microbial communities are intricately linked to ecosystem functioning because they play important roles in carbon and nitrogen cycling, and feedback to plant communities as mutualists and pathogens [22]. Although much research has been done to study the impacts of a range of disturbances on soil microbial communities and their functioning [23], many uncertainties remain about the controls on soil microbial community stability, and the consequences of disturbance-induced changes in microbial communities for their capacity to withstand further disturbances. The influence of biodiversity on ecosystem stability is complex and depends not only on species richness but also on the evenness or composition of the soil microbial community. Resistance and resilience to disturbance might also vary between functional microbial guilds dependent on their levels of functional redundancy.

Our results indicate high stability of the studied soil systems and suggest the existence of functional redundancy among soil microorganisms, leading to ecosystem resistance and resilience (Table 2).

A high level of functional redundancy, within a functional community, that is, a high number species performing the same function, might act as a buffer against the effect of biodiversity loss on functioning [23].

However, functional redundancy is likely more limited within specialised rather than global processes.

The results of our studies have shown that there are strong correlation relations between the majorities of functional groups of soil microorganisms in non-disturbed edaphotopes.

Oligotrophic bacteria and pedotrophic bacteria correlated with micromycetes and actinomyces, a correlation coefficients (CC) were 1,0; 0,79 and 0,95; 0,95.

Aerobic nitrogen fixing bacteria positive correlated with ammonifiers (CC=0,98) and bacteria wich are using mineral forms of nitrogen (CC=0,84). anaerobic bacteria high correlated with spore forming bacteria (CC=0,86), micromycetes (CC=0,91) and actinomyces (CC=0,98).

In general, should be noted a high level integrity of soil microbiome, 46 bonds were identified between different groups of microorganisms, which characterized a high level of correlation (CC > 0.75)

The level of integration of soil microbiome in disturbed edaphotopes was lower than in non-disturbed (Table 3).

It was recorded 41 correlation connections between different ecological groups of soil microorganisms with high level of correlation (CC > 0.75).

Strong correlation connections have been observed between anaerobic bacteria and micromycetes (CC=0,91); anaerobic bacteria and actinomyces (CC=0,98). Oligotrophic bacteria strong correlated with micromycetes, actinomyces and anaerobic bacteria

Toxic substances produced by microorganisms enter the plant directly from the soil, and they are concentrated mainly in the overground organs, and almost were not observed in the roots of plants. Soil in virgin forest ecosystems was characterized by relatively low levels of phytotoxic activity (Fig. 1).

Studies have shown that the toxicity of the soil varies seasonally. In non-disturbed edaphotopes the highest level of phytotoxic activity of the

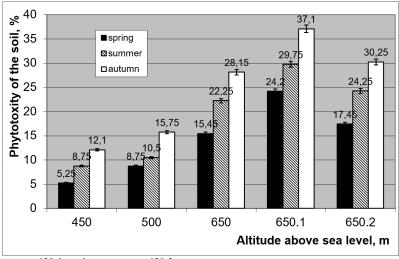
Table 2 Integrity of soil microbial community in non-disturbed edaphotopes

| | Ammonifiers | Spore forming bacteria | Micromycetes | Actinomyces | Bacteria wich are using mineral forms of nitrogen | Anaerobic bacteria | Aerobic nitrogen fixing bacteria | Anaerobic nitrogen fixing bacteria | Oligotrophic bacteria | Oligonitrophic bacteria | Pedotrophic bacteria |
|---|-------------|------------------------|--------------|-------------|---|--------------------|----------------------------------|------------------------------------|-----------------------|-------------------------|----------------------|
| Ammonifiers | _ | -0,55 | -1,00 | -1,00 | 0,74 | -0,90 | 0,98 | -1,00 | -1,00 | 1,00 | -0,94 |
| Spore forming bacteria | -0,55 | _ | 0,58 | 0,95 | -0,97 | 0,86 | -0,68 | 0,62 | 0,55 | -0,55 | 0,79 |
| Micromycetes | -1,00 | 0,58 | _ | 0,81 | -0,76 | 0,91 | -0,99 | 1,00 | 1,00 | -1,00 | 0,95 |
| Actinomyces | -1,00 | 0,95 | 0,81 | _ | -1,00 | 0,98 | -0,88 | 0,84 | 0,79 | -0,79 | 0,95 |
| Bacteria wich are using mineral forms of nitrogen | 0,74 | -0,97 | -0,76 | -1,00 | _ | -0,96 | 0,84 | -0,80 | -0,74 | 0,74 | -0,92 |
| Anaerobic bacteria | -0,90 | 0,86 | 0,91 | 0,98 | -0,96 | _ | -0,96 | 0,93 | 0,90 | -0,90 | 0,99 |
| Aerobic nitrogen fixing bacteria | 0,98 | -0,68 | -0,99 | -0,88 | 0,84 | -0,96 | _ | -1,00 | -0,99 | 0,99 | -0,99 |
| Anaerobic nitrogen fixing bacteria | -1,00 | 0,62 | 1,00 | 0,84 | -0,80 | 0,93 | -1,00 | _ | 1,00 | -1,00 | 0,97 |
| Oligotrophic bacteria | -1,00 | 0,55 | 1,00 | 0,79 | -0,74 | 0,90 | -0,99 | 1,00 | _ | -1,00 | 0,94 |
| Oligonitrophic bacteria | 1,00 | -0,55 | -1,00 | -0,79 | 0,74 | -0,90 | 0,99 | -1,00 | -1,00 | _ | -0,94 |
| Pedotrophic bacteria | -0,94 | 0,79 | 0,95 | 0,95 | -0,92 | 0,99 | -0,99 | 0,97 | 0,94 | -0,94 | _ |

soil - 28,15% was observed at an altitude of 650 meters. It should be noted that phytotoxicity of the soil in primeval ecosystems in the spring was twice lower than in the autumn, this pattern was observed in all studied biotopes. Identical changes in the level of soil toxicity were established by us in the forest ecosystems on the territory of the Carpathian Biosphere Reserve [21, 22]. The compaction of soil due to the use of heavy machinery for wood transportation has led to increase the phytotoxicity of the soil. The highest level 37,10±1,81% was in the autumn. Such changes in the level of soil toxicity are associated with succession processes occurring in the structure of microbiocenosis due to the influence of exogenous factors. Another main functional parameter of soil microbiome is enzyme activity - a sensitive indicator of changes in the soil environment. Soil enzymes are natural mediators and catalysts of many important soil processes, such as decomposition of organic matter released into the soil during vegetation, reactions of humus formation and decomposition,

Table 3 Integrity of soil microbial community in disturbed edaphotopes

| | Ammonifiers | Spore forming bacteria | Micromycetes | Actinomyces | Bacteria wich are using mineral forms of nitrogen | Anaerobic bacteria | Aerobic nitrogen fixing bacteria | Anaerobic nitrogen fixing bacteria | Oligotrophic bacteria | Oligonitrophic bacteria | Pedotrophic bacteria |
|--|-------------|------------------------|--------------|-------------|---|--------------------|----------------------------------|------------------------------------|-----------------------|-------------------------|----------------------|
| Ammonifiers | _ | -0,55 | -1,00 | -0,23 | 0,74 | -0,90 | 0,98 | -1,00 | -1,00 | 1,00 | -0,94 |
| Spore forming bacteria | -0,55 | _ | 0,58 | 0,95 | -0,97 | 0,86 | -0,68 | 0,62 | 0,55 | -0,55 | 0,79 |
| Micromycetes | -1,00 | 0,58 | _ | 0,81 | -0,76 | 0,91 | -0,99 | 1,00 | 1,00 | -1,00 | 0,95 |
| Actinomyces | -0,23 | 0,95 | 0,81 | _ | -1,00 | 0,98 | -0,88 | 0,84 | 0,79 | -0,79 | 0,35 |
| Bacteria wich are using mineral forms of nitrogen | 0,74 | -0,97 | -0,76 | -1,00 | _ | -0,96 | 0,84 | -0,80 | -0,74 | 0,74 | -0,92 |
| Anaerobic bacteria | -0,90 | 0,86 | 0,91 | 0,98 | -0,96 | _ | -0,96 | 0,34 | 0,90 | -0,90 | 0,35 |
| Aerobic nitrogen fixing bacteria | 0,98 | -0,68 | -0,99 | -0,88 | 0,84 | -0,96 | - | -1,00 | -0,99 | 0,99 | -0,99 |
| Anaerobic nitrogen fixing bacteria | -1,00 | 0,62 | 1,00 | 0,84 | -0,80 | 0,34 | -1,00 | _ | 1,00 | -1,00 | 0,97 |
| Oligotrophic bacteria | -1,00 | 0,55 | 1,00 | 0,79 | -0,74 | 0,90 | -0,48 | 1,00 | _ | -1,00 | 0,94 |
| Oligonitrophic bacteria | 1,00 | -0,55 | -1,00 | -0,79 | 0,74 | -0,90 | 0,99 | -1,00 | -1,00 | _ | -0,94 |
| Pedotrophic bacteria | -0,94 | 0,79 | 0,95 | 0,35 | -0,92 | 0,35 | -0,99 | 0,97 | 0,94 | -0,94 | _ |



Footnote: 650.1- soil compaction; 650.2-cutting...

Fig. 1. Phytotoxical activity of the soil in virgin beech forests of Uzhanskyi National Nature Park

production of mineral nutrient forms available for plants, nitrogen fixation, as well as the flow of carbon, nitrogen and other basic elements of the biochemical cycle. Determination of enzyme activity along with regulating factors are indispensable to characterize the metabolic potential, soil fertility and are useful in soil assessments with regard to soil microbiome. At the same time, they can be used to study soil biochemical processes and to evaluate soil quality The process of transformation of organic matter in to the soil takes place with the participation of soil microorganisms and enzymes [25, 26, 27]. Species composition within forest tree stands determines the diversity of microorganisms along with their enzymatic activity [28]. Tree species [29] and altitude above sea level affect soil pH. The pH value has a significant effect on the activity of microorganisms in the soil, enzymes are highly susceptible to soil reaction. The results of the present study indicate that enzyme activity varies considerably within the studied forest soils (Table 4).

Table 4
Enzyme activity of soil in virgin beech forests of Uzhanskyi National
Nature Park

| № | Altitude above sea level, | рН | Catalase, sm ³ O ₂ /gr. soil | Invertase, mg.glucose/ | | |
|---|---------------------------|----------|--|------------------------|--|--|
| | m | pm | per 1 min. | gr.soil. | | |
| 1 | 450 | 5,7±0,15 | 9,3±0,22 | $16,12\pm0,35$ | | |
| 2 | 500 | 6,0±0,24 | 9,5±0,14 | $17,73\pm0,28$ | | |
| 3 | 650 | 6,2±0,15 | 9,8±0,18 | 22,10± 0,60 | | |
| 4 | 650.1 | 5,0±0,20 | 4,20±0,15 | 14,46± 0,45 | | |
| 5 | 650.2 | 5,5±0,17 | 5,3±0,10 | $20,23\pm0,35$ | | |

Footnote: 650.1- soil compaction; 650.2-cutting.

The lowest activity of catalase $(4,20\pm0,15 \text{ sm}^3\text{O}_2/\text{gr. soil per 1 min.})$ was recorded in the in compressed soils. The activity of invertase also was the lowest $(14,46\pm0,45 \text{ mg.glucose/gr.soil})$ on this territory.

The highest activities of catalase $(9.8\pm0.18 \text{ sm}^3\text{O}_2/\text{gr. soil})$ per 1 min) and invertase $(22.10\pm0.60 \text{ mg.glucose/gr.soil})$ were determined in the soils at the altitude 650 meters above sea level in non-disturbed edaphotopes (table 4).

The observed higher enzyme activity of forest soils in non-disturbed biotopes (Table 1) can be attributed to the fact that these soils contain more fungal biomass in comparison to disturbed biotopes. Fungal organisms play an important role in the first stages of breakdown of large molecule compounds, such as lignin and cellulose.

Conclusions. The obtained results of the study soil microbiome and its functional characteristics of Uzhanskyi National Nature Park confirmed that microorganisms are sensitive reagents to the influence of external factors and can be used as good indicators of ecosystem condition. It was established that the ratio of different ecological-functional groups of soil microorganisms varies depending on the height of the habitat above sea level, what is caused by the influence of abiotic factors. Violation of the integrity of the phytocenosis in the result of deforestation and soil compaction led to significant negative changes in the structure of soil microbiome. It was observed the decreasing in the functional biodiversity of the authentic groups of microorganisms where dominated groups of microorganisms were oligotrophes and pedotrophes. Negative changes have been observed in enzyme activity of soil, level of catalase and invertase decreased. The integrity of soil microbial community was violated.

Conflict of interest. Authors declare no potential conflicts.

Acknowledgements. We express our sincere appreciation to our colleagues: Professor Nadiya Boyko - Director of Scientific Research and Educational Center of Molecular Microbiology and the Immunology of Mucous Membranes (Uzhhorod National University) and to Professor, Academician of NAASU Volodymyr Patyka — Chair of Department of Phytopathogenic Bacteria, (D.K Zabolotny Institute of Microbiology and Virology of the Academy of Sciences of Ukraine) for their support in organizing these studies.

МІКРОБІОМ ҐРУНТУ ПРАЛІСОВИХ ЕКОСИСТЕМ ЗАКАРПАТТЯ

¹ Ужгородський Національний Університет, вул. Волошина, 32, Ужгород, 88000, Україна ² Опольський університет, вул. Дмовського, 7-9, м. Ополє, 45-365, Польща ³ Полтавська державна аграрна академія, вул. Сковороди, 1/3, Полтава, 36021, Україна

Резюме

Мета. Дослідити мікробіом грунту пралісових екосистем, зокрема, його структуру, чисельність основних еколого-фізіологічних груп, функціональні параметри, такі як: токсичність грунту, ферментативну активність грунту за рівнем каталази і інвертази. Проаналізувати сукцесійні процеси, що відбуваються в мікробіоцено- зі грунту внаслідок впливу ендогенних і екзогенних факторів. Оцінити інтегрова-

ність мікробних угруповань у різних едафотопах пралісових екосистем. Методи. Мікробіологічні дослідження ґрунту проводили за загальноприйнятими у ґрунтовій мікробіології методиками. Ферментативну активність ґрунту (каталазну, інвертазну) визначали газометричним методом та колориметричним методом відповідно. Для визначення токсичності ґрунту використовували біотестування. Оцінку інтегрованості мікробних угруповань у різних едафотопах пралісових екосистем здійснювали за допомогою кореляційно-регресійного аналізу. Статистичну обробку експериментальних даних проводили з використанням програми Statistica 10. Результати Антропогенний вплив – ущільнення грунту, санітарні рубки – призвів до змін в угрупованнях грунтових мікроорганізмів, зниження їх чисельності і функціонального різноманіття. Зокрема, в порушених едафотопах зростав рівень токсичності ґрунту, уповільнювались процеси розкладу органічної речовини, знижувався рівень ферментативної активності ґрунту, порушувалась інтегрованість мікробних угруповань ґрунту. Висновки Вплив ендогенних і екзогенних факторів у пралісових екосистемах порушує інтегрованість мікробіому ґрунту та призводить до негативних змін його функціональних характеристик.

Ключві слова: мікроорганізми, грунт, праліси, екосистема, ензиматична активність, інтегрованість, угруповання.

МИКРОБИОМ ПОЧВЫ ПРАЛЕСОВЫХ ЭКОСИСТЕМ ЗАКАРПАТЬЯ

 Π .Ю. Симочко 1 , А.В. Калиниченко 2,3

¹ Ужгородский Национальный Университет, ул. Волошина, 32, Ужгород, 88000, Украина ² Опольский университет, ул. Дмовского, 7-9, г. Ополе, 45-365, Польша ³ Полтавская государственная аграрная академия, ул. Сковороды, 1/3, Полтава, 36021, Украина

Резюме

Цель. Исследовать микробиом почвы пралесовых экосистем, в частности, его структуру, численность основных эколого-физиологических групп, функциональные параметры, такие как: токсичность почвы, ферментативную активность почвы по уровню каталазы и инвертазы. Проанализировать сукцессионные процессы, происходящие в микробиоценозе почвы вследствие влияния эндогенных и экзогенных факторов. Оценить интегрированность микробных сообществ в различных едафотопах пралесовых экосистем. Методы. Микробиологические исследования почвы проводили по общепринятым в почвенной микробиологии методикам. Ферментативную активность почвы (каталазную, инвертазную) определяли газометрическим и колориметрическим методами соответственно. Для определения токсичности почвы использовали биотестирование. Оценку интегрированности микробных сообществ в различных едафотопах пралесовых экосистем осуществляли с помощью корреляционно-регрессионного анализа. Статистическую обработку экспериментальных данных проводили с использованием программы Statistica 10. Результаты Антропогенное воздействие – уплотнение почвы, санитарные рубки – привело к изменениям в сообществах почвенных микроорганизмов, снижению их численности и функционального разнообразия. В частности, в нарушенных едафотопах возрастал уровень токсичности почвы, замедлялись процессы разложения органического вещества, снижался уровень ферментативной активности почвы, нарушалась интегрированность сообществ микроорганизмов почвы. Выводы Влияние эндогенных и экзогенных факторов в пралесовых экосистемах нарушает интегрированность микробиома почвы и приводит к негативным изменениям его функциональных характеристик.

Ключевые слова: микроорганизмы, почва, пралеса, экосистема, энзиматическая активность, интегрированность, сообщества.

- Saijai S. et al. Analysis of microbial community and nitrogen transition with enriched nitrifying soil microbes for organic hydroponics. Biosci Biotechnol Biochem. 2016; 27:1–8.
- Dimitriu PA, Grayston SJ. Relationship between soil properties and patterns of bacterial b-diversity across reclaimed and natural boreal forest soils. Microb. Ecol. 2010; 59:563–573.
- 3. Högberg MN, Högberg P, Myrold DD. Is microbial community composition in boreal forest soils determined by pH, C-to-N ratio, the trees, or all three. Oecologia. 2007; 150:590–599. doi: 10.1007/s00442-006-0562-5.
- 4. Banning N.C., Gleeson D.B. Grigg A.H., Grant C.D., Andersen G.L., Brodie E.L., Murphy D.V. Soil microbial community successional patterns during forest ecosystem restoration. Applied and environmental microbiology. 2011; 77(17):58–64.
- 5. Odum E. P. Strategy of ecosystem development. Science. 1969; 164:262–270.
- 6. Walker L.R., Walker J., Hobbs, R.J. Linking restoration and ecological succession. Springer; 2007.
- 7. Rout ME., Southworth D. The root microbiome influences scales from molecules to ecosystems: the unseen majority. Am J Bot. 2013; 100: 1689–1691.
- 8. Minz D, Ofek M, Hadar Y. Plant rhizosphere microbial communities. The Prokaryotes: Prokaryotic Communities and Ecophysiology. Berlin: Springer-Verlag; 2013. p. 57–84.
- 9. Turner TR., James EK., Poole PS. The plant microbiome. Genome Biol. 2013; 14:209–219.
- 10. Zolla G, Badria DV., Bakker MG., Manter DK., Vivanco JM. Soil microbiomes vary in their ability to confer drought tolerance to Arabidopsis. Appl Soil Ecol. 2013; 68:1–9.
- 11. Grayston S.J., Rennenberg H. Assessing effects of forest management on microbial community structure in a central European beech forest. Canadian Journal of Forest Research. 2006; 2595–2604.
- 12. Hillebrand H., Matthiessen B. Biodiversity in a complex world: consolidation and progress in functional biodiversity research. Ecol Lett. 2009; 12:1405–1419.
- 13. Magurran A. Measuring biological diversity. African Journal of Aquatic Sciences. 2004; 29:285-286.
- 14. Goldman E., Green, L. H. Practical Handbook of Microbiology. 2-th ed. New-York: CRC Press; 2012.
- 15. Bitton G., Rossel D. Soil Ecotoxicology. Boca Raton: CRC Lewis; 1997.
- 16. Volkogon V.V., Nadkernichna O.V., Tokmakova L.M. ta in. Eksperymental'na gruntova mikrobiologiya: monografiya. Kyiv: Agrarna Nauka; 2010. Ukrainian.

- 17. Guan S.M. Soil enzyme and its research method. Beijing: Agriculture Press; 1986.
- 18. Dick R.P. Soil enzyme activities as indicators of soil quality. Defining Soil Quality for a Sustainable Environment. SSSA Special Publication. 1994; 35:107–124.
- 19. CAO Rui, WU Fu-zhon, YANG Wan-qin, TAN Bo, WANG Bin, LI Jun, CHANG Chen-hui. Effects of altitudes on soil microbial biomass and enzyme activity in alpinegorge regions. Chinese Journal of Applied Ecology. 2016; 27(4):1257–1264.
- Reynolds HL, Packer A., Bever JD., Clay K. Grassroots ecology: plant-microbe-soil interactions as drivers of plant community structure and dynamics. Ecology. 2003; 84:2281–2291.
- 21. Symochko L., Patyka V., Symochko V., Kalinichenko A. Soil Microbial Activity and Functional Diversity in Primeval Beech Forests. Journal of Earth Science and Engineering. 2015; 5(6):363–371.
- 22. Symochko L., Hamuda H.B. Microbial monitoring of soil as additional tools for conservation biology. Obuda University e-Bulletin. 2015; 5(1):177–185.
- 23. Van der Heijden M.G.A., Bardgett R.D., Van Straalen N.M. The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. Ecol. Lett. 2008; 11:296–310.
- 24. Griffiths B.S., Philippot L. Insights into the resistance and resilience of the soil microbial community. FEMS Microbiol. Rev. 2013; 37:112–129.
- 25. Schimel J.P., Bennett J. Nitrogen mineralization: challenges of a changing paradigm. Ecology. 2004; 85:591–602.
- 26. Dick R.P. Soil enzyme activities as integrative indicators of soil health. Biological indicators of soil health. Oxford University Press. 1997; 121–156.
- 27. Błońska et al. The relationship between soil properties, enzyme activity and land use. Forest Research Papers. 2017; 78 (1): 39–44.
- 28. Baldrian P. Distribution of extracellular enzymes in soils: spatial heterogeneity and determining factors AT various scales. Soil Science Society of American Journal. 2014; 78: 11–18.
- 29. Błońska E., Lasota J., Gruba P. Effect of temperate forest tree species on soil dehydrogenase and urease activities in relation to Rother properties of soil derived from less and galciofluvial sand. Ecological Research. 2016; 31(5): 655–664.

Отримано 22.02.2018