

Microorganisms and their enzymes play essential roles in the functioning of the soil environment because they take part in the formation of the soil structure, the transformation of soil organic matter, thereby making nutrients available for plants and degrading contaminants. However, most studies of microbiological and biochemical soil properties, especially in soils that are in agricultural use, focus on the upper soil horizons (up to 30 cm) because this layer has the highest concentration of soil microbial communities and the highest enzymatic activity. In turn, although it is well known that microbes and enzymes take part in the biogeochemical processes that occur in the deeper soil horizons, the microbiological and biochemical properties in the deeper soil horizons are little understood. That is why limiting research to the topsoil not only decreases the possibility to identify the structure of these microbial communities, their functional diversity and changes in soil enzymatic activity but also the understanding of the common processes of the transformation of soil organic matter. The soil microbial structure and enzymatic activity throughout the soil profile depend on various physico-chemical factors such as the organic matter content, the concentration of available carbon and nutrients, soil reaction, soil moisture and oxygenation as well as its temperature and texture. Additionally, microbial and biochemical soil properties depend on the distribution of plant root systems, which change with depth.

The aim of the planned research is to perform a complex analysis of soil enzymatic activity as well as to determine the functional and genetic diversity of microorganisms against its physico-chemical properties within genetic horizons of soil profiles as related to the soil type and type of agricultural soil management. We will determine the influence of different agricultural management systems on the studied properties in the same soil types as well as the influence of different types of pedogenic processes in soils that are under the same agriculture use on soil properties. In this way, we will obtain information about how the enzymatic activity, functional and genetic diversity and other soil properties are influenced by pedogenic processes in the different genetic horizons of the soil profile and to what extent they are affected by the various plants that are cultivated. The novelty of the project will be the use of modern methods in molecular biology to assess the functional and genetic diversity of microorganisms across the soil depth profiles of soil that is used agriculturally. A few studies of this type have primarily focused on the surface soil horizons and were related only to bacterial communities. Using the molecular techniques will make it possible to obtain information concerning the (97-99%) group of microorganisms that are VBNC (*Viable But Not Culturable*) that to date is unavailable.

The study will include the excavation of 24 profiles of the mineral soil types that most frequently occur in the area of the Polish Lowlands (Luvisol, Brunic Arenosols, Cambisols, Phaeozems), which are under various types of agricultural management such as arable soil (rape and/or cereal) and the cultivation (alfalfa or clover) over the past few years (first year of the study), permanent plantations (hop fields, vineyards) (second year of the study) and permanent grasslands and orchards (third year of the study). Soil samples will be collected at different times depending on the nature of the type of soil management (various crops), but August and/or September will be the preferred period for the soil sampling. Soil samples will be collected from each soil genetic horizon and within three years of the study, approximately 120 samples will be analyzed, with the exception of the Next Generation Sequencing analysis (NGS), which will be assayed in 40 selected samples only.

The physico-chemical properties of properly prepared soil samples will be determined (e.g. bulk density; particle size analysis; active, exchangeable and hydrolytic acidity; different forms of carbon and nitrogen; fractional composition of organic matter in the soil samples with an organic C content equal 5 g/kg (or higher); CEC; exchangeable cations (Na, K, Ca, Mg); available forms of P, K and Mg); enzymatic activity of C, N and P transformation (e.g. cellulases, α - and β -glucosidases, xylanase, invertase, peroxidase, phenoloxidase, N-acetylo- β -D-glucosaminidase (NAG) urease, nitroreductase, proteases, acid and alkaline phosphatase, dehydrogenases). Moreover, the concentration of microbial biomass C, N and P and soil respiration activity will also be determined. To determine the activity and biodiversity of the soil microbial communities the BIOLOG system will be used along with Ecoplate-type biochemical tests. The diversity and composition of fungal and bacterial communities will be determined by using the method based on Next Generation Sequencing (NGS) after DNA isolation and ITS1/16S rRNA gene amplification. Since plants are one of the sources of enzymes in soil, the morphology of plant root systems will be performed using a scanner and the appropriate software.

The research will contribute to the development of complex and little known area of research by identifying the changes in the functional and structural diversity of the various microbial communities that inhabit the deeper soil horizons as well as the enzymes that they secrete. The data, which will provide new information, could significantly increase the quality of research in the fields of environmental microbiology and enzymology and soil ecology. Such multi-aspect research of the soil properties throughout the soil profile have not yet been carried out in Poland.