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TAXONOMIC AND FUNCTIONAL PROFILING OF VINEYARD CHERNOZEM MICROBIOME

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Soil microorganisms are critical functional drivers in plant-soil systems, directly influencing soil health, nutrient cycling, and agricultural sustainability. Traditional soil monitoring has historically focused on quantitative, physical, and chemical parameters, which act as a "black box" and fail to capture the qualitative taxonomic structure and ecological dynamics of the soil microbiome. In alignment with the European Union's Soil Monitoring Law (Soil Directive) incentives to promote soil resilience and quality, this study explores the application of amplicon sequencing (using 16S, 18S and ITS1 markers) to profile the microbiome of chernozem soils under viticulture. The research aims to evaluate amplicon sequencing as a robust diagnostic tool.

Soil samples were collected from an experimental vineyard at the Institute of Genetics, Physiology and Plant Protection of the Moldova State University (Chisinau, Moldova) in October (post-harvest) from the 0–10 cm topsoil layer using a five-point envelope method in triplicate. Total genomic DNA was extracted using the PureLink™ Microbiome DNA Purification Kit (Thermo Scientific, USA), with quality verified via spectrophotometry and agarose gel electrophoresis. High-throughput amplicon sequencing (16S rRNA for bacteria, ITS1 for fungi, and 18S rRNA for eukaryotes) was performed on the Illumina platform by Macrogen Europe (Netherlands). Raw reads were processed through a standardized bioinformatics pipeline to analyze alpha/beta-diversity and predict functional roles using FAPROTAX and FUNGuild.

Alpha- and beta-diversity profiling revealed distinct, scale-dependent structural patterns across soil domains. Total eukaryotic communities (18S) exhibited the highest alpha-diversity (mean: 979 ASVs, Shannon H'_{log2}: 8.54), displaying strong

micro-spatial heterogeneity at the ASV level (mean Bray-Curtis dissimilarity: 0.96) but shifting to high structural homogeneity at the phylum level (mean dissimilarity: 0.12). Fungal communities (ITS) showed intermediate richness (mean: 363 ASVs, Shannon H'_{log2}: 5.91) with moderate spatial stability (mean dissimilarity: 0.37), dominated by perennial decomposers (*Ascomycota, Mucoromycota, Basidiomycota*). Conversely, bacterial communities (16S) possessed lower richness (mean: 83 ASVs, Shannon H'_{log2}: 4.47) but exceptional spatial homogeneity (mean dissimilarity: 0.13); the community was heavily co-dominated by oligotrophic *Acidobacteriota* and predatory *Myxococcota*, which constituted the two most abundant bacterial phyla. Functional profiling highlighted the concurrent presence of these abundant *Myxococcota* alongside a minor but consistent fraction of fungal *Zoopagomycota*, indicating a complex, multi-kingdom trophic structure within the vineyard soil.

In conclusion, the vineyard soil hosts a highly structured ecosystem where high micro-spatial variation among eukaryotic species yields to stable functional configurations, supported by a prominent and well-represented microbial predatory core.

Keywords: vineyard, microbiome, DNA, sequencing.

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