METAGENOMIC 16S rRNA INVESTIGATION OF THE BLACK SEA MICROBIAL DIVERSITY IN THE REGION OF ZMIINIY ISLAND

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Zmiiniy is a small island situated in the northern-western part of the Black Sea. The seawater around it and its marine ecosystem are under great impact of the outside factors such as: Danube river flow that brings nutrients with allochtonic microorganisms from mainland and development of ship activity in this region. All these factors lead to forming of unique marine ecosystem of the island where microorganisms are the integral part of it. The aim of the study was to determine the marine microbial biodiversity of the Zmiiniy island coastal seawatr with the help of metagenomic 16S rRNA analysis.

The surface water for analysis was collected in July 2014. Water was filtered and the following total DNA extraction was performed from the membrane filters. The dual-indexing primers were used to amplify v4 variable region of the 16S rRNA gene and construct the clone library. Sequencing was performed on Illumina MiSeq platform with custom primers Read1, Read2 and Index. Obtained raw nucleotide sequences were analyzed in QIIME (Caporaso et al, 2010) and SILVAngs (Quast et al, 2013) pipelines.

In the result of metagenomics 16S rRNA analysis there were obtained 80206 raw sequences and detected about 3200 distinct Operational Taxonomic Units. The main revealed phyla among Domain Bacteria were: Proteobacteria, Bacteroidetes, Cyanobacteria, Actinobacteria, Verrucomicrobiota, Planctomycetes, Tenericutes, Fusobacteria, Firmicutes and candidate divisions of uncultivated prokaryotic representatives: SR1, BD1-5, BHI80-139, OP3, OD1, WS3, NPL-UPA2, SHA-109, SM2F11and TM6. The most abundant classes of the Proteobacteria were Gamma- and Alpha-Proteobacteria. The bioinformatics analysis allowed identification up to genus level. The dominating genera among studied samples were: Marivita, Loktanella, Paracoccus, Pseudoalteromonas, Vibrio, Coraliomargarita, Acholeplasma, Pseudomonas, Phaeobacter, Neptunomonas, Allivibrio, Oceaniserpentilla, Oceanospirillum, Robiginitalea, Acinetobacter, Haliea and others.

The provided metagenomic 16S rRNA analysis allowed evaluating of rich taxonomic prokaryotic communities in Zmiiniy island region of the Black Sea. This research determined microbial representatives that are poorly investigated and yet are not capable of cultivation. The majorities of identified bacterial species are known as typical inhabitants of marine environments and have been described previously in similar metagenomic studies.