## Abstract

This work concerns halotolerant and halophilic methanotrophs that have been isolated from seawater, coastal lagoons, and several soda lakes. These organisms use two strategies in order to sustain osmotic equilibrium between cytoplasm and their substrate under high salinity. The first mechanism involves sustaining high concentrations of potassium and chloride ions inside cell. The second strategy concerns the biosynthesis of organic osmotic solutes like sugars (e.g. trehalose), amino acids (e.g. glycine betaine, glutamic acid, L-proline, ectoine, hydroxyectoine) and polyols (e.g. glycerol).

The aim of this study was the recognition the synthesis of ectoine and hydroksyectoine by methanotrophs existing in rocks surroundings of Wieliczka salt deposits.

Methanotrophic microorganisms isolated from studied rocks were identified by NGS using the V3-V4 region of the 16S rRNA gene on the Illumina MiSeq platform (GENOMED S. A.). Released osmolytes was analyzed by GCMS and HPLC technique.

The results indicated that qualitative and quantitative composition of amino acids formed by methanotrophs depends on the salt concentration and temperature. The significant amounts of "intercellular" and "extracellular" amino acids as: ectoine (0,232 mg  $g_{d.w.}^{-1}$ ) and hydroxyectoine (0,657 mg  $g_{d.w.}^{-1}$ ) were found. At the same time trace amounts (<430 ng  $g_{s.m.}^{-1}$ ) of the other amino acids as: GLU, ALA, GLY, VAL, LEU, THR, PRO, ASP, MET, PHE, LYS, TYR, TRP were detected.