ENRICHMENT AND ISOLATION OF NOVEL POLYSACCHARIDE-DEGRADING ANAEROBES FROM ABYSSAL BLACK SEA SEDIMENT

D.M. VAN VLIET[1,2] (DAAN.VANVLIET@WUR.NL),
A.J.M. STAMS[1,2,3], L. VILLANUEVA[2,4], I. SÁNCHEZ-ANDREA[1, 2]

[1] Laboratory of Microbiology, Wageningen University, Wageningen, The Netherlands.
[2] Soehngen Institute of Anaerobic Microbiology (SIAM), Nijmegen, The Netherlands
[3] Centre of Biological Engineering, University of Minho, Braga, Portugal
[4] Department of Marine Organic Biogeochemistry, Royal Netherlands Institute for Sea Research (NIOZ), 't Horntje, Texel, The Netherlands

Marine anaerobes play an important role in the global carbon cycle. This includes the degradation of organic matter by hydrolytic and fermentative marine anaerobes, of which ecologically relevant isolates are lacking. This lack could stem from the use of monomeric substrates in anaerobic cultivation rather than the recalcitrant polymeric substrates available to marine anaerobes *in situ*. We tested this hypothesis by using environmentally representative cultivation conditions with polysaccharides as substrates. Enrichment cultures were set up with mineral marine medium and low concentrations (2.5 mM monomer equivalent) of one of several polysaccharides. The cultures were inoculated with anoxic Black Sea sediment from the abyssal plain at 2100 m depth and incubated at 15°C (*in situ* T = 8°C). Various microbial clades without cultured representatives were enriched, indicating a potential hydrolytic and/or fermentative metabolism. We isolated hydrolytic strains of *Kiritimatiellaeota* (formerly *Verrucomicrobia* subdivision five) clade R76-B128, which is ubiquitous in anoxic marine waters, including the Black Sea. The only isolate of this clade is *Kiritimatiella glycovorans*, which is a halophilic bacterium that does not grow on polysaccharides. The 16S rRNA genes of our isolates form two phylogenetic clusters that share 94% identity with each other, and 83-84% identity with the 16S rRNA gene of *K. glycovorans*. According to taxonomic threshold values, this is strong evidence that our isolates represent two novel genera within a novel family of the *Kiritimatiellaeota*. Ongoing work on these strains includes characterization, genomics, and comparative proteomics to identify hydrolytic pathways.