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Microbes in geothermally heated marine sediments - community structure and metabolism

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Devoid of both light and allochthonous organic material, microbial communities in subsurface marine sediments rely on chemolithoautotrophy for primary production. In the hydrothermal system of the Aeolian Archipelago, Italy, the reduction of oxygen, sulfate, and nitrate are strongly exergonic processes [1] and thus, potential energy sources for microbial metabolism. Anoxic sediment incubations at 90 °C have shown that the hyperthermophilic microbial community metabolizes a variety of naturally occurring organic compounds via fermentative and sulfate reducing pathways [2].

In sediments collected at hydrothermal sites of the Aeolian Islands, the structure of the microbial communities was quantified by fluorescence in situ hybridization (FISH). Archaea were 2-3 times as abundant as bacteria, and about half of the detected archaea belonged to the Crenarchaeota. The euryarchaeal order Thermococcales, exclusively heterotrophs that grow on various organic compounds by fermentation or sulfur respiration, represented 12-22 % of the archaea. Another 9-22 % of the archaea belonged to the order Archaeoglobales - strict anaerobes that use oxidized sulfur compounds, iron, or nitrate as electron acceptors. Most of the detected bacteria belonged to the order Aquificales or to the genus *Thermus*. Both groups typically use oxygen in their auto- or heterotrophic metabolism, so that highest abundances are expected where hydrothermal fluids get in contact with oxic seawater.

Twelve metabolically defined subpopulations were studied in enrichment cultures, using geothermally heated sediment as inoculum. Chemolithoautotrophic cultures grew notably. Growth was not improved by the addition of nitrate and/or fatty acids. Upon addition of sulfate and/or glucose, increased growth rates and changes in community structure were observed. Our presentation will compare the FISH results of these enrichments with the in situ community structure. Turnover rates of fermentative and respiratory metabolism will be estimated from time series of fatty acid and sulfate concentrations measured during exponential growth of the enrichment cultures.

References

- [1] Amend J.P., Rogers K.L., Shock E.L., Gurrieri S., and Inguaggiato S. (2003) *Geobiology* **1**, 37-58.
- [2] Tor J.M., Amend J.P., and Lovley D.R. (2003) *Env. Microbiol.* **5**, 583-591.

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Microbial biofilm processes in deep granitic groundwater

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The underground MICROBE laboratory

A container laboratory, denoted MICROBE, was installed underground at the 450 m level in the Äspö hard rock laboratory tunnel, Sweden. Three boreholes were drilled in the rock adjacent to MICROBE. A discrete fracture in each of three boreholes was packed off with metal free packers and connected to temperature controlled circulation systems in the lab. Biofilms were grown on surfaces installed in laminar flow cells (figure) under *in situ* pressure (24-32 kg cm⁻²) and chemistry at a flow rate of about 200 µm sec⁻¹. The chemistry and the gas composition were analysed. All components were constructed of PVDF or PEEK plastics. Metals were not exposed to the groundwater.

Numbers, trace element sorption and activity

Biofilms developed slowly to about 10⁷ cells per cm². An amorphous matrix formed and covered the surface. The biofilms were exposed to radionuclides such as cobalt-60 and promethium-147. More trace elements were sorbed on the biofilms compared to non-grown control surfaces. Subsurface biofilms seem to promote retention of migrating trace elements. Addition of a carbon source and an electron acceptor, lactate and nitrate, stimulated growth activity of the studied biofilm. This is in agreement with results obtained previously under atmospheric pressure.



Figure: Pressure resistant laminar flow cell for investigations of *in situ* biofilm formation. Three flow diffusers are installed before and after the pile of laminar surfaces for growth.