

## Final report VISTA 2012

### Metagenomics and metaproteomics of deep arctic hydrothermal systems

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Project duration:	<u>01.03.09 – 29.02.12</u>
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Project number:	6501

Summary: The main goal of this project was to use cutting-edge metagenomic and metaproteomic technologies on deep arctic hydrothermal systems and cold methane seeps to characterize the diversity, physiological potential and adaptive features of archaea and bacteria in these habitats. The main habitat studied during the project was cold methane-enriched sediments from Nyegga focusing on the G11 pockmark and the CNO3 area. We found a dominance of anaerobic methanotrophic archaea (ANME) which is a group of microorganisms that play a vital role globally by reducing the emission of the methane seeping from deep reservoirs into the overlying water body and the atmosphere. New understanding on the stratification of ANME dominated communities below a white microbial mat within Pingo structures in the G11 pockmark Nyegga was generated. Furthermore, how methane seepage rates influence the vertical variation in community structure was studied by comparing the stratification observed for the G11 pockmark with that in the sediments in the CN03 micro-seeping area. Our data indicate that ANME populations in less active seepage areas at Nyegga have the potential to increase in cell number and act as seed communities when the geochemical environment is altered. Details about the *in situ* metabolism and molecular adaptations of an ANME-1 dominated community in the G11 pockmark were obtained by a coupled metagenomic and metaproteomic approach. The obtained data are currently being compared with data obtained from the Loki's Castel hydrothermal Vent Field which is rich in methane. The data obtained through the project have thus provided Statoil with context data in naturally occurring hydrocarbon seepage areas found in cold, arctic and/or pristine environments. Finally, initial surveys of the metagenomic data indicate a reservoir of biotechnologically relevant enzymes.

Deviations from targets: In the original project plan we intended to perform more metagenomics of microbial mats from the Loki's Castle vent field as well as from additional sediments horizons at Nyegga. Initial surveys of the biotechnological potential in these data have been performed. However, the data obtained are complex and it is time-consuming to analyze and interpret the data. Manuscripts are in preparation but publication of the data was impossible within the time frame of the project. However, we are planning to publish data obtained through the project in 2013.

#### Publications:

Doctoral thesis Irene Roalkvam: Diversity, stratification and *in situ* metabolism of anaerobic methanotrophic archaea in Nyegga cold seeps. The three following papers were included in the thesis.

1. Roalkvam I., Jørgensen S-L., Chen Y., Stokke R., Dahle H, Hocking W-P, Lanzen A., Haflidason H., and IH Steen. (2011). New insight into stratification of anaerobic methanotrophs in cold seep sediments. FEMS Microbiology Ecology 78:233-243

2. Stokke R., Roalkvam I., Lanzen A., Haflidason H., and IH Steen (2012). Integrated metagenomic and metaproteomic analyses of an ANME-1 dominated community in marine cold seep sediments. *Environmental microbiology* 14: 1333–1346
3. Roalkvam, I., Dahle, H., Chen, Y., Jørgensen, S. L., Haflidason, H. & Steen, I. H (2012). Fine-scale community structure analysis of ANME in Nyegga sediments with high and low methane flux. *Frontiers in Extreme Microbiology*. 3:216.

In addition, Roalkvam has during her PhD-project contributed to the following manuscript.

4. Dahle, H., Roalkvam, I., Thorseth, I., Pedersen, R. B., and IH Steen. The versatile *in situ* gene expression of an Epsilonproteobacteria dominated biofilm from a hydrothermal chimney. In revision *Environmental Microbiology Reports*

Continuation of the project: The expertise in the use of cutting edge metagenomics and metaproteomics approaches obtained through the project is valuable for Statoil. Knowledge transfer to Statoil to solve their biotechnological and environmental relevant issues is important. Possible focus in a continuing project could be to perform a baseline study of a high number of Statoil samples (reservoir, sediments, biofilms on various constructions etc) using pyrosequencing of PCR-amplicons, qPCR and analysis of geochemical context data. Then these data could be compared with environmental data obtained through this VISTA-project 6501 and the data that we already have from the arctic hydrothermal vent fields studied at Centre for Geobiology in Bergen. A focus could be on hydrocarbon-degrading microorganisms.