Oil & Natural Gas Technology

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Quarterly Progress Report

October 2011 – December 2011

ASSESSING THE EFFICACY OF THE AEROBIC METHANOTROPHIC BIOFILTER IN METHANE HYDRATE ENVIRONMENTS

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Prepared for:
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EXECUTIVE SUMMARY

In October 2008 the University of California at Santa Barbara (UCSB) initiated investigations of water column methane oxidation in methane hydrate environments, through a project funded by the National Energy Technology Laboratory (NETL) entitled: assessing the efficacy of the aerobic methanotrophic biofilter in methane hydrate environments. The twelfth quarter of this project was dedicated to the continued work-up of data resulting from a series of cruises to the Santa Barbara and Santa Monica Basins, and the Gulf of Mexico, to the revision and acceptance of manuscripts resulting from expeditions to study the methane plume originating from MC 252 and methane in the basins offshore southern California.

During this period project personnel focused on the revision of two manuscripts for publication in a special issue of the Proceedings of the National Academy of Sciences on the Deepwater Horizon incident, on the revision of an article previously submitted to Limnology and Oceanography, and to the revision of an article previously submitted to Continental Shelf Research. All four were accepted for publication during this period, and fifth previously-accepted paper was published in the Proceedings of the National Academy of Sciences. We continue to polish a manuscript on methane oxidation in ice covered Arctic lakes and plan submission in the near future. We also continue to analyze the methanotrophic bloom from the Deepwater Horizon event, and are planning additional laboratory analyses in the next period. Project personnel also demobilized from a cruise aboard the R/V Atlantis to the borderland basins off Southern California.

PROGRESS, RESULTS AND DISCUSSION

Task 1 - Project Management Plan (PMP)
This task was completed during the first quarter of this award.

Task 2 - Field Sampling of Microbial Mats
- **Subtask 2.1 - Coal Oil Point Sampling**
- **Subtask 2.2 - Santa Monica Basin Sampling**

Task 2 was completed during a previous reporting period.

Task 3 - Turnover Rates for Methane Oxidation in Microbial Mats
- **Subtask 3.1 - Turnover Rates for Coal Oil Point Samples**
- **Subtask 3.2 - Turnover Rates for Santa Monica Basin Samples**

Task 3 was completed during a previous reporting period.

Task 4 - Molecular Analyses of Methanotrophs
We continue to expand our approach for molecular analysis of methanotrophs and during this reporting period one of our manuscripts was published in the Proceedings of the National Academy of Sciences that highlights two of our approaches: stable isotope probing, and comparative community analysis by combined T-RFLP and clone libraries. We also continued our collaboration with the laboratory of V. Orphan (Caltech) to analyze mmo using alternative approaches including an intergenic spacer analysis and quantitative PCR. We also continue manuscript preparation.

Task 5 - Stable Isotope Probing
- **Subtask 5.1 - Stable Isotope Probing of Coal Oil Point Samples**
- **Subtask 5.2 - Stable Isotope Probing of Santa Monica Basin Samples**
- **Subtask 5.3 - Stable Isotope Probing of Gulf of Mexico Water Samples**

Task 5 was completed in a previous reporting period.

Task 6 - Field Measurements in the Santa Barbara Basin
- **Subtask 6.1 - Shallow Water Sampling and Measurements, Santa Barbara Basin**
- **Subtask 6.2 - Deep and Bottom Water Sampling and Measurements, Santa Barbara Basin**
- **Subtask 6.3 - Repeat Sampling, Santa Barbara Basin**

Task 6 was completed in a previous reporting period.

Task 7 – Analysis of Methane Oxidation Rates and Methane Turnover Times Throughout the Santa Barbara Basin
- **Subtask 7.1 - Shallow Water**
- **Subtask 7.2 - Interior Water**
- **Subtask 7.3 –Targeted Measurements**

We continue with data analysis of methane oxidation rates and turnover times for methane in the Santa Barbara Basin. These data are being prepared for publication in the peer reviewed literature.

Task 8 - Analysis of Current Velocity Data

Subtask 8.1 – Current Velocity Analysis for the Shallow Santa Barbara Basin

Subtask 8.2 - Current Velocity Analysis for the Deep Santa Barbara Basin

Task 8 was completed in a previous reporting period

Task 9 - Development of a methane budget for the Santa Barbara Basin

We continue to develop a draft methane budget for the Santa Barbara Basin. We also had a manuscript on the budget of the northern Santa Barbara Basin accepted with Continental Shelf Research.

Task 10 - Field Sampling of Waters

Subtask 10.1 - Santa Barbara Basin Water Sampling

Subtask 10.2 - Southern California Margin Water Sampling

Subtask 10.3 - Targeted Water Sampling

Subtask 10.4 – Gulf of Mexico Water Sampling

This task was completed in a previous reporting period.

Task 11 - Sensitivity Testing of Methane Oxidation Rates

This task was completed during a previous reporting period.

Project personnel focused their primary efforts on data analysis and manuscript publication. Significant efforts were needed to demobilize from the SEEPS 11 cruise, which ended Sep 29. We further analyzed samples using alternative approaches for analysis of mmo, and are moving forward with restriction fragmentation, quantitative PCR and single cell genomics to address questions of methanotrophic ecology.

Conclusion

The current period saw the continued analysis of data, the acceptance of several manuscripts and the publication of one manuscript in the Proceedings of the National Academy of Sciences. We continue to transition toward data analysis and publication as this award draws to a close.

COST STATUS

There are no subcontracts to this award. All funds are being expended by UCSB. Financial report under separate cover.

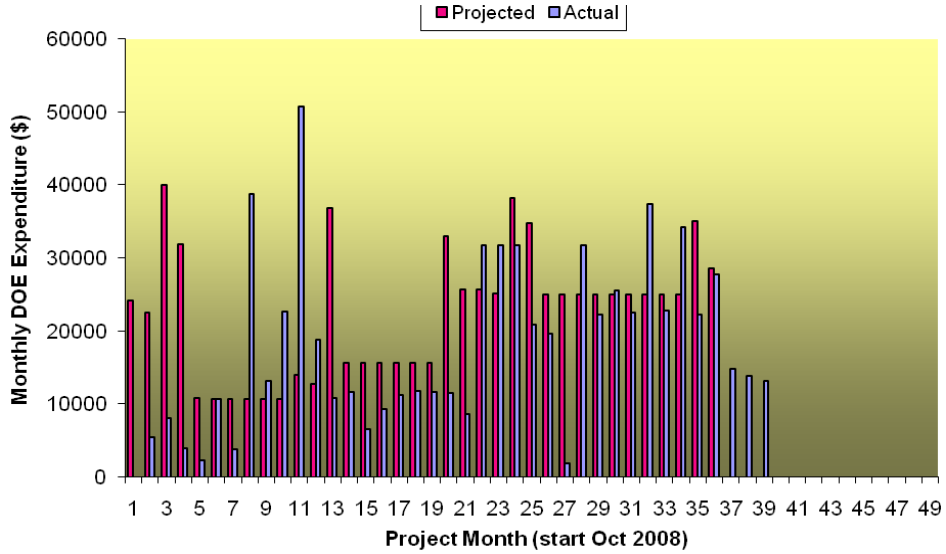

Figure 1. Project costing profile

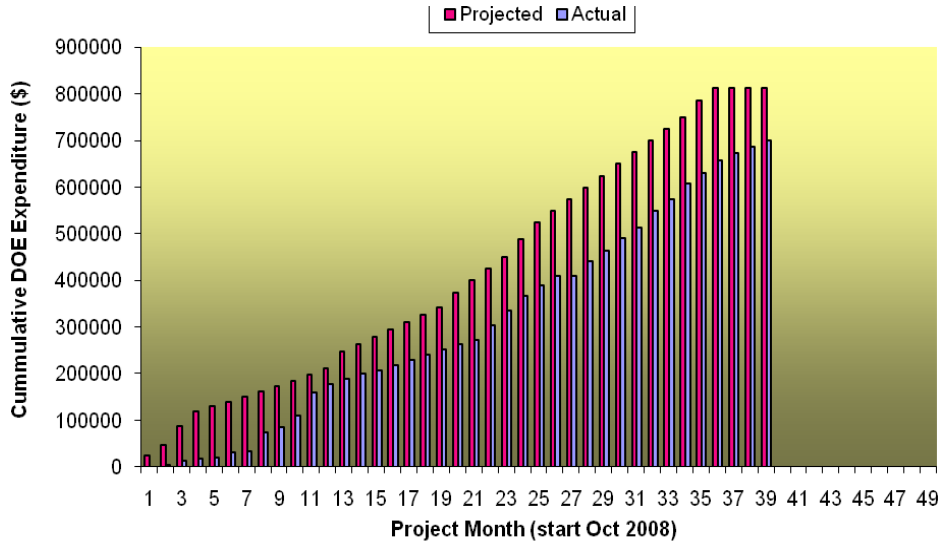

Figure 2. Project cummulative costs

MILESTONE STATUS

Milestone 1: Successful installation and sea trial of the CTD rosette system and ADCP. This milestone relates closely to Tasks 6.1, 6.3, 10.1, and 10.3, and must be reached to enable sampling in support of tasks 7.1, 7.3, 8.1, and 11. The estimated completion date for this milestone is 4/1/09, but may be pushed back until June/July, 2009 on account of missing the fall, 2008 weather window.

Status: This milestone was completed during a previous reporting period.

Milestone 2: Confirmation of $^{3}H\text{-}CH_4$ oxidation and $^{13}C\text{-}CH_4$ uptake by benthic microbial mats from Coal Oil Point seeps. This milestone relates directly to Tasks 2.1, 3.1, and 5.1 and will further facilitate the completion of tasks 4, and 5.2. The estimated completion date for this milestone is 7/1/09.

Status: This milestone was completed during a previous reporting period.

Milestone 3: Completion of the SEEPS 09 cruise. The SEEPS 09 cruise presents an unrivaled level of access to recently discovered methane hydrate sites in the Santa Monica Basin and to water column sites throughout the Southern California margin including the deep Santa Barbara Basin. The cruise and associated sampling relate closely to Tasks 2.2, 6.2, and 10.2, and will facilitate completion of tasks 3.2, 4, 5.2, 8.2, 9, and 11. The estimated completion date for this milestone is 1/1/10, but the timing will necessarily depend on the UNOLS scheduling of this (already approved) cruise.

Status: This milestone was completed during a previous reporting period.

Milestone 4: Completion of the Gulf of Mexico (GoM) cruise. The GoM cruise presents an unprecedented opportunity to track the fate of methane from a massive methane plume. During this cruise aboard the *R/V Cape Hatteras* samples will be collected for methane concentration, methane oxidation rates, methane stable isotopes, microbial cells, and large volume filtrates for DNA. The estimated completion of this milestone is 6/30/10 and is associated with tasks 5.3 and 10.4.

Status: This milestone was completed during a previous reporting period.

Milestone 5: Complete a preliminary analysis of current velocity data and oxidation rate data from the SEEPS 09 cruise. This milestone must be achieved to address Tasks 6.3, 7.3 and 11. The estimated completion date for this milestone is 10/1/10.

Status: This milestone was completed during a previous reporting period.

Milestone 6: Conduct a preliminary analysis for mmo and 16SrRNA gene sequences for putative methanotrophs from the Santa Monica Basin, and compare to se-

quences from Coal Oil Point seeps. This milestone relates directly to Tasks 4, 5.1, and 5.2, and will determine the approach taken in completing Tasks 4 and 5. The estimated completion date for this milestone is 12/1/10.

Status: This milestone was completed during a previous reporting period.

Milestone 7: Complete the ocean-going sampling program, and perform preliminary analysis of all physical and chemical data to ensure sufficient data for further analysis. This milestone relates directly to Tasks 6.3, 7.3, and 10.3 and will facilitate the completion of Tasks 9 and 11. The estimated completion date for this milestone is 4/1/11.

Status: This milestone was completed during a previous reporting period.

ACCOMPLISHMENTS

→ Demobilized from the SEEPS 11 research cruise aboard the R/V Atlantis, which included conducting repeat methane oxidation rate measurements in both the Santa Barbara and Santa Monica Basins.
→ Published a paper in the journal Proceedings of the National Academy of Sciences (Redmond and Valentine, 2011) highlighting the changes in microbial community structure that followed the irruption of hydrocarbon gases into the ocean following the Deepwater Horizon event.
→ Submitted a final revised manuscript to the journal Proceedings of the National Academy of Sciences (Valentine et al in press) developing a coupled physical oceanographic and microbial metabolic model to explain the patterns, timing and impacts of hydrocarbon release to the deep ocean during the Deepwater Horizon event. The manuscript was accepted and is in-press.
→ Submitted a re-revised manuscript to Limnology and Oceanography (Heintz et al, in press) on methanotrophy in the Basins offshore southern California. The manuscript was accepted and is now in-press.
→ Submitted a re-revised manuscript to Continental Shelf Research highlighting a methane budget along the Northern margin of the Santa Barbara Channel. The manuscript was accepted and is now in-press.
→ Submitted a revised manuscript to the Proceedings of the National Academy of Sciences (Ryerson et al, in-press) describing the magnitude and fate of hydrocarbon emissions from the Deepwater Horizon event. This manuscript was accepted and is now in-press.

PROBLEMS OR DELAYS

None

PRODUCTS

→ Twelfth Quarterly Report Submitted

→ Published **Redmond MC and DL Valentine (2011) Natural gas and temperature structured a microbial community response to the Deepwater Horizon oil spill. Proceedings of the National Academy of Sciences, USA. doi/10.1073/pnas.1108756108**

→ Accepted for publication: **Valentine DL, I Mezić, S. Maćešić, N. Črnjarić-Žic, S. Ivić, P Hogan, V Fonoberov and S Loire (in-press) Dynamic auto-inoculation and the microbial ecology of a deep water hydrocarbon irruption Submitted to the Proceedings of the National Academy of Sciences, USA.**

→ Accepted for publication: **Heintz M, S Mau and DL Valentine (in-press) Oceanographic Control on Methanotrophic Potential in the Santa Monica Basin, Southern CA Borderland. Limnology and Oceanography.**

→ Accepted for publication: **Mau S, M Heintz and DL Valentine (in-press) Quantification of CH_4 loss and transport in dissolved plumes of the Santa Barbara Channel, California. Continental Shelf Research**

→ Accepted for publication: **Ryerson T, Camilli R, Kessler J, Kujawinski EB, Reddy CM, Valentine DL et al. (in-press) Chemical composition measurements quantify Deepwater Horizon hydrocarbon emissions and distribution in the marine environment. Proceedings of the National Academy of Sciences, USA, doi/10.1073/pnas.1110564109.**

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