Uncoupling of autotrophy and heterotrophy: effects of the Deepwater Horizon Oil Spill on microbial food webs

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1) General Summary

Oil and its constituents are primarily degraded by native microorganisms with abiotic processes being less important. Oil provides a high amount of organic material available to support bacterioplankton growth, the specific source of carbon, however may select for specific groups of bacterioplankton – shifting the microbial community structure which may alter other biogeochemical cycles as well. The project addressed Category 2 – conduct baseline studies and impact assessments to provide the basis for long term monitoring. Specifically our goal was to provide baseline studies on microbiological and geochemical processes governing the degradation of oil and affected by oil and its constituents in pristine and oil-contaminated Gulf of Mexico near shore environments. Phytoplankton and bacterioplankton are the base of marine food webs; phytoplankton fix inorganic carbon to organic carbon and bacterioplankton are responsible for cycling significant (often 50% or more) of the organic carbon through the microbial loop. Changes in this dynamic have the potential to disrupt microbial biogeochemical cycles and significantly alter microbial food webs with potential cascading effects through higher trophic levels. We focused our efforts on examining how increases in oil affect both primary and heterotrophic production and the linkages between them. During this process we conducted fundamental tests on the toxicity of oil and dispersant (Corexit 9500) to bacterioplankton and phytoplankton as well as how these compounds might shift microbial community structure.

Three locations were sampled; Apalachicola Bay represented an uncontaminated location while Bay Jimmy (Barataria Bay) represented an heavily contaminated site and Pensacola Beach was only episodically contamined with oil from the Deepwater Horizon Oil Spill.

Microcosm experiments were established with waters from each location and the changes in microbial production, carbon transfer, and community production were monitored. Specific results included:

• While carbon released by phytoplankton was not changed by exposure to and or Corexit, the amount of carbon taken up by bacterial shifted away from phytoplankton derived carbon and was replaced by oil carbon
• Both bacterial and phytoplankton production are inhibited by oil in a dose dependent fashion beginning in the ppm range
• Corexit alone has minimal effect on microbial production
• Corexit and oil and Corexit cause the most significant shifts in phytoplankton community structure but with minimal changed in bacterioplankton community structure
• Nutrient replete conditions reduce the effect of oil on both bacterioplankton and phytoplankton
• Stable isotope and DNA sequencing suggests that bacteria in Bay Jimmy are consuming DWH oil
• Oil is much more difficult to work with experimentally than anticipated but Water Accommodated Fractions (WAF) provide and reproducible means to conduct microbial experiments with oil
• These initial experiments were leveraged into later funding and project expansion through participation in GOMRI funded consortia
2) Results and Scientific highlights

- Methods were refined to generate WAFs using sterile seawater. Exposure to sunlight was found to greatly increase the inhibitory properties of the WAF. In all subsequent experiments, a standard protocol of WAF generation (3 days in sunlight) was used.
- Oil plus dispersant is generally more toxic than either component alone, but not in an additive fashion. While oil inhibits phytoplankton primary production more than heterotrophic bacterial production, low concentrations of Corexit have been observed to increase phytoplankton production.
- Toxicity of oil added directly to seawater samples is more complex. In general, while bacterial production is sensitive to added oil in the short term, it quickly recovers to pre-oil levels. In contrast, phytoplankton production is very sensitive to oil, even at levels below 1 ppm (as determined by hydrocarbon analyses). Recovery is minimal. These preliminary results are perplexing and demonstrate that the response to oil and the WAF are different for phytoplankton. Nutrient replete conditions reduced the inhibitory effect of exposure to oil.
- Results from microcosm experiments proved perplexing. While many preliminary experiments identified concentrations of WAF that would inhibit microbial production, when applied during the microcosms experiments the effects were minimal, making broad conclusions more difficult.
- While carbon released by phytoplankton was not changed by exposure to and or Corexit, the amount of carbon taken up by bacterial shifted away from phytoplankton derived carbon and was replaced by oil carbon.
- Corexit and oil and Corexit cause the most significant shifts in phytoplankton community structure but with minimal changed in bacterioplankton community structure.
- Results clearly demonstrate in situ remineralization (i.e. as dissolved organic carbon, DIC) of an older carbon substrate in Barataria Bay relative to that observed in either Pensacola Bay or Apalachicola Bay. Using dual isotope mass balance calculations we estimate as much as 12% of the isotopic signature of Barataria Bay DIC can attributed to oil remineralization.
- Data demonstrates that the bacterial community structure (as determined by DNA sequencing of cloned 16S rDNA genes) at Barataria Bay was distinctly different from that observed at either Pensacola Bay and Apalachicola Bay. Known oil degrading species Actinobacteria and Firmencutes were both present at the most oil impacted site, Barataria Bay, only Firmencutes were present at Pensacola Bay and neither were present in Apalachicola Bay. These isotope tracer and molecular approaches reveal distinct differences in biogeochemical processes and community structure in Barataria Bay where we see $^{14}$C deplete remineralized DIC as well as the presence of oil degrading bacterial. Collectively, these results suggest that Barataria Bay bacterial communities are exposed to and are utilizing oil.
3) Cruises and Field Expeditions

<table>
<thead>
<tr>
<th>Ship or Platform Name</th>
<th>Class (if applicable)</th>
<th>Chief Scientist</th>
<th>Objectives</th>
<th>Dates</th>
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<tr>
<td>WEATHERBIRD II</td>
<td></td>
<td>Cherrier</td>
<td>Microbial community structure and stable isotope analyses of carbon cycling in the DeSoto Canyon and DWH vicinity</td>
<td>5 Days May 2011</td>
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4) Peer Reviewed Publications

The project should result in multiple manuscripts, all of which are in the very early stages of preparation. It is anticipated that papers on the following topics will be completed:

(i) Changes in Microbial carbon transfer between phytoplankton and bacterioplankton after exposure to oil and or Corexit
(ii) Changes in microbial community structure after exposure to oil and or Corexit
(iii) Evidence for microbial degradation in contaminated vs non-contaminated sites
(iv) PAH concentrations in the sediments of the Northern Gulf of Mexico after the Deepwater Horizon Oil Spill

5) Presentations and Posters


INVITED TALKS


**Jeffrey, W.H.** 2012. The oil spill and microbes – it ain’t all just biodegradation. The University of West Florida, March 16.


Jeffrey, W.H. 2013. The effects of the Deepwater Horizon Oil Spill on Microbial Production: there’s more to it than biodegradation. January 10.


6) Other Deliverables
None

7) Data
The type of data generated during this project does not lend itself to easy archiving (for instance, there is no cruise metadata collected specifically for this project). Data are maintained in the PI’s notebooks and computers.

PARTICIPANTS AND COLLABORATORS

8) Project Participants

<table>
<thead>
<tr>
<th>First Name</th>
<th>Last Name</th>
<th>Role in Project</th>
<th>Institution</th>
<th>Email</th>
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<tbody>
<tr>
<td>Wade</td>
<td>Jeffrey</td>
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<td><a href="mailto:wjeffrey@uwf.edu">wjeffrey@uwf.edu</a></td>
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9) Student and Post-doctoral participants

<table>
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<tr>
<th>First Name</th>
<th>Last Name</th>
<th>Post-doc / PhD / MS / BS</th>
<th>Thesis or research topic</th>
<th>Institution</th>
<th>Supervisor</th>
<th>Expected Completion year</th>
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<tr>
<td>Jessie</td>
<td>Rosanbalm</td>
<td>MS</td>
<td>Effects of oil and dispersant on phytoplankton communities of the northern Gulf of Mexico</td>
<td>UWF</td>
<td>Jeffrey</td>
<td>2012</td>
</tr>
<tr>
<td>Tiffany</td>
<td>Baskerville</td>
<td>PhD</td>
<td>Changes in microbial community structure after exposure to oil and evidence for microbial degradation of oil</td>
<td>FAMU</td>
<td>Cherrier/Chauhan</td>
<td>2014</td>
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10) Student publications
It is anticipated that Jessie Rosanbalm’s MS thesis will result in at least 1 or 2 publications and Tiffany Baskerville’s dissertation at least 2 or more publications.

11) Student Presentations


13) Continuing Funding
PI Jeffrey is now a Co-PI on the C-IMAGE GOMRI funded consortium in which his project is predominantly examining spatial and temporal variability in microbial (bacteria, archaea, ciliates, and foraminifera) community structure in the sediments of the Northeastern Gulf of Mexico

PI Jeffrey is also a co-Investigator on the DEEP-C GOMRI funded consortium in which the primary objective is to look at microbial and oceanographic processes on the Northwest Florida Shelf water column.