

Microbial respiration of oil in surface waters in close proximity to the *Deepwater Horizon* site.

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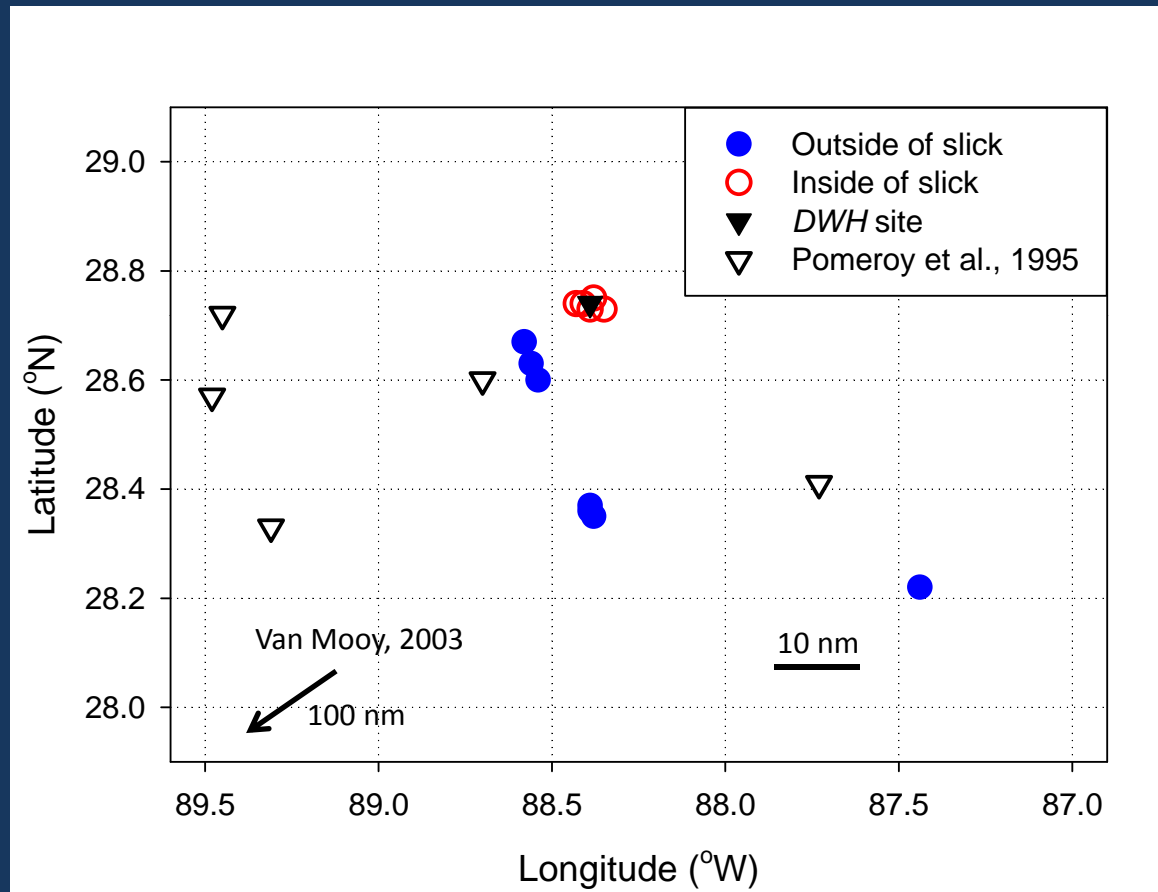
Research supported by NSF-OCE RAPID Awards



See: Edwards et al., (2011) *Environ. Res. Lett.*

Station locations in close proximity to *Deepwater Horizon* site.

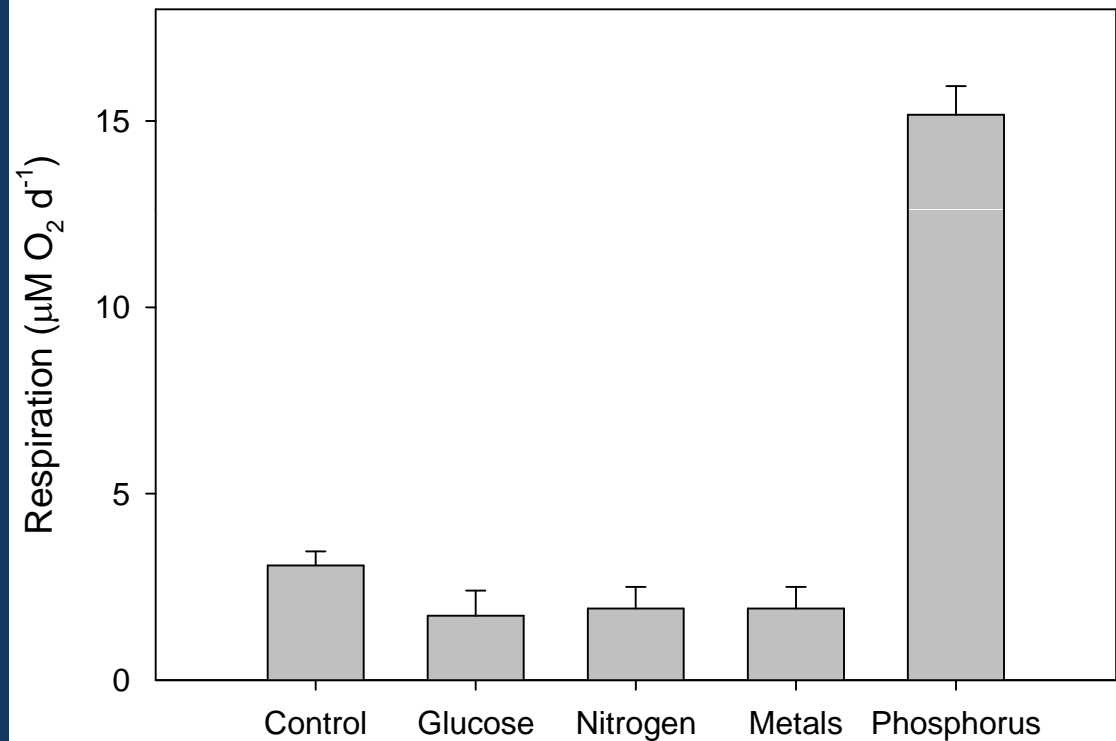
On site June 19 to 28, 2010
aboard R/V *Endeavor*.



Previous evidence of phosphorus limitation of microbial respiration

Right: Results obtained in area of *DWH* in June of 1993 by Pomeroy et al. (1995) *Mar Ecol Prog Ser*

In July of 2002, SRP concentrations of 12 nM and turnover time of 1.0 ± 0.2 hours (Van Mooy, 2003, unpublished data)



Hypotheses and approaches

Hypothesis I: Microbes unable to respond to oil in surface slick

Approach: Compare microbial rates and abundances inside and outside of the surface slick.

- Enzyme assays – standard methods using fluorogenic substrates.
- Respiration – track O₂ in BOD bottles equipped with oxygen optodes
- Hydrocarbon degradation – track hydrocarbons in bottle incubations by GC/FID
- Microbial biomass – intact polar lipid concentrations by HPLC/MS
- Microbial abundance – SYBR Green staining and flow cytometry

Hypothesis II: Microbes limited by inorganic nutrients

Approach: Compare incubations amended with nutrients versus non-amended controls.

- Amendment: 32 μM NH₄NO₃, + 2 μM KH₂PO₄ in HPLC grade water
- Control : HPLC grade water

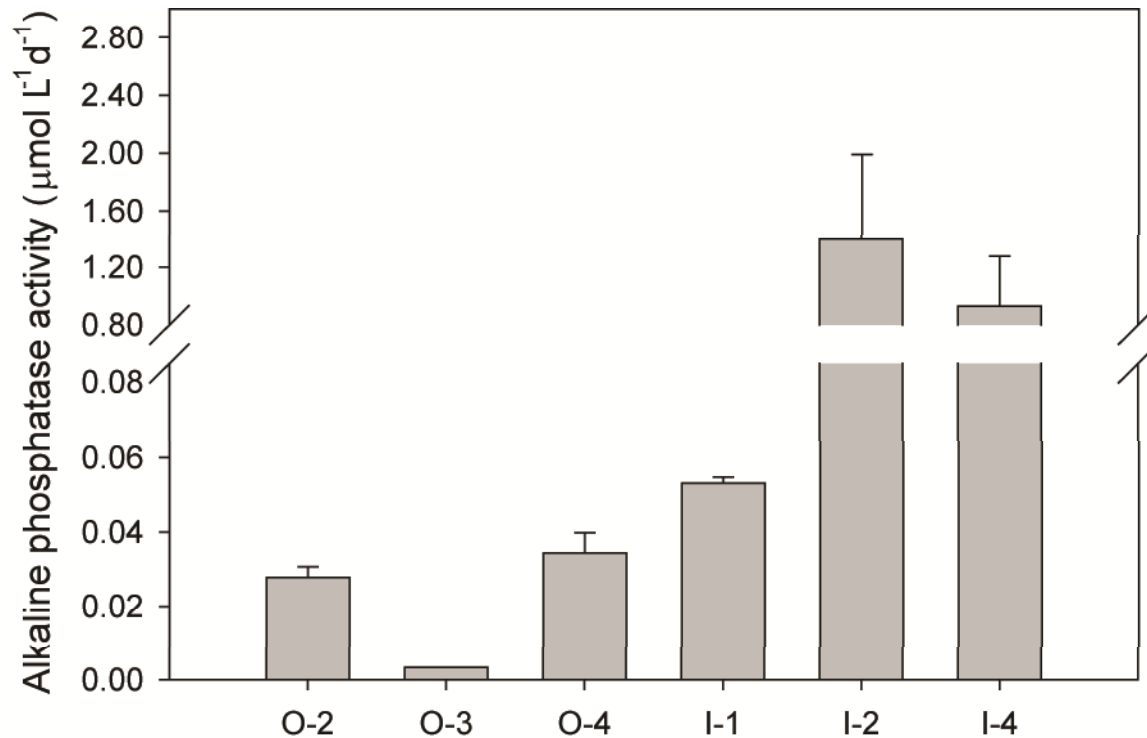
Statistics: Single analyses from triplicate samples. Error bars are magnitude of range. Differences delineated by non-parametric tests.

For complete methods see Edwards et al., (2011) *Environ. Res. Lett.*

Enhanced phosphorus stress inside slick

Alkaline phosphatase activities were higher inside the slick than outside the slick.

Soluble reactive phosphorus (i.e. phosphate) ranged from 5 to 16 nM, but there were no differences between inside and outside of the slick.

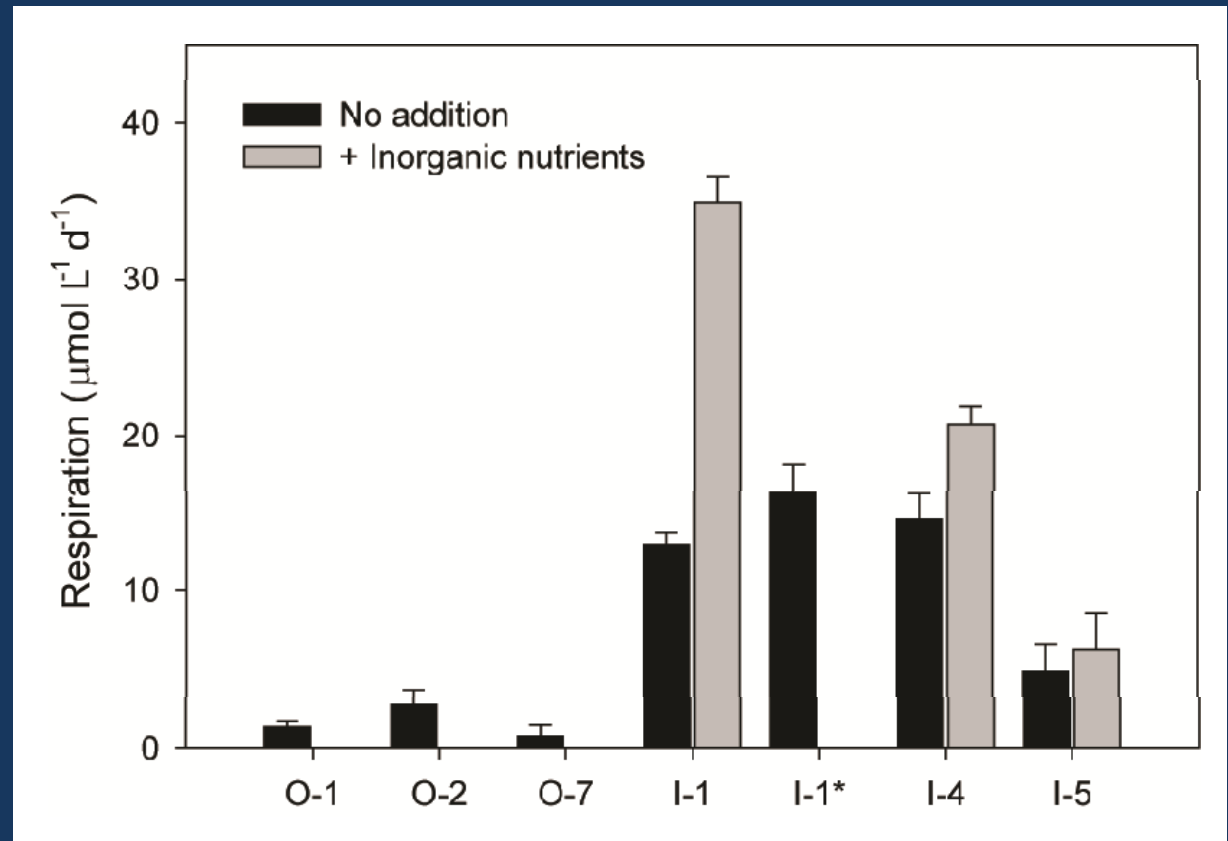


Enhanced respiration inside slick, but nutrient limited?

In 36-hour incubations, respiration rates inside the slick were about 5 times faster on average than outside the slick.

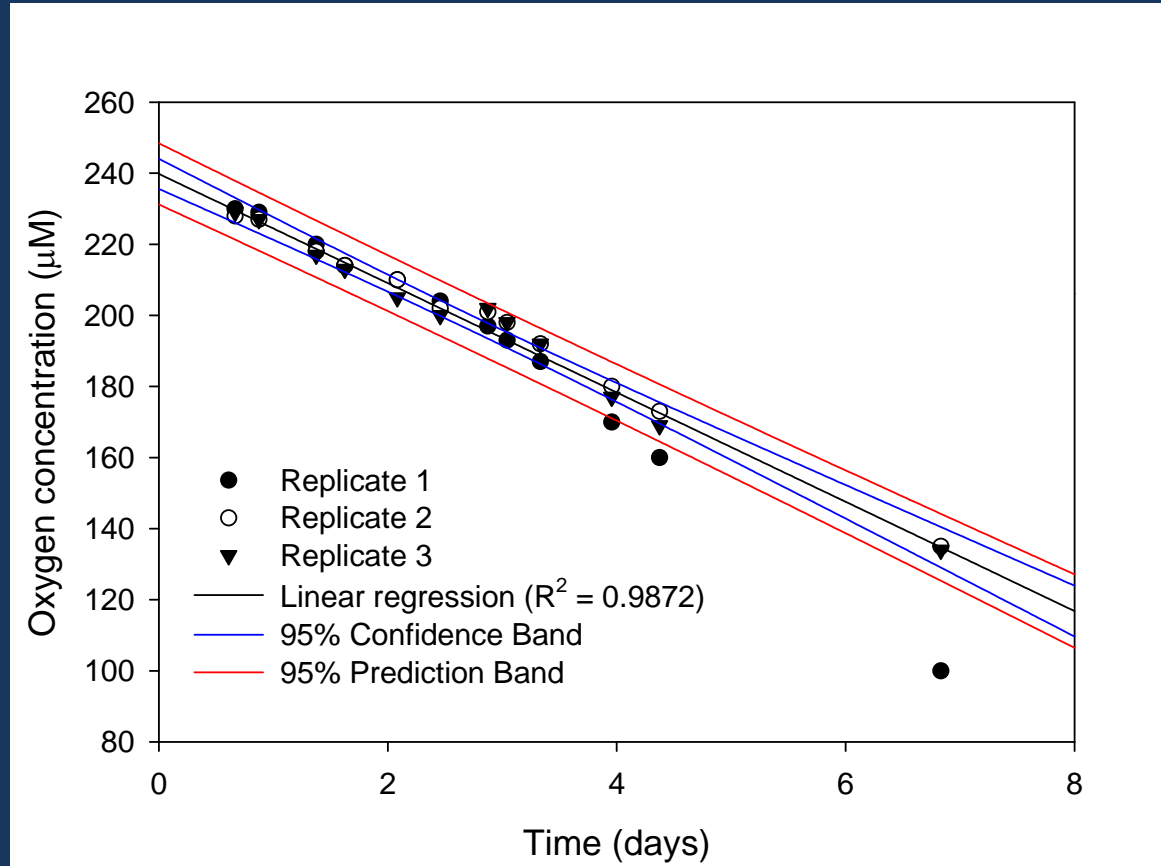
Enhanced by an average of $9.9 \mu\text{M O}_2 \text{ d}^{-1}$.

Nutrient amendments led to a range of responses, from as much as a doubling in the microbial respiration rate to no effect.



Respiration rates were consistent over time.

Longer-term incubation (6 days) showed that respiration was relatively constant for several days.

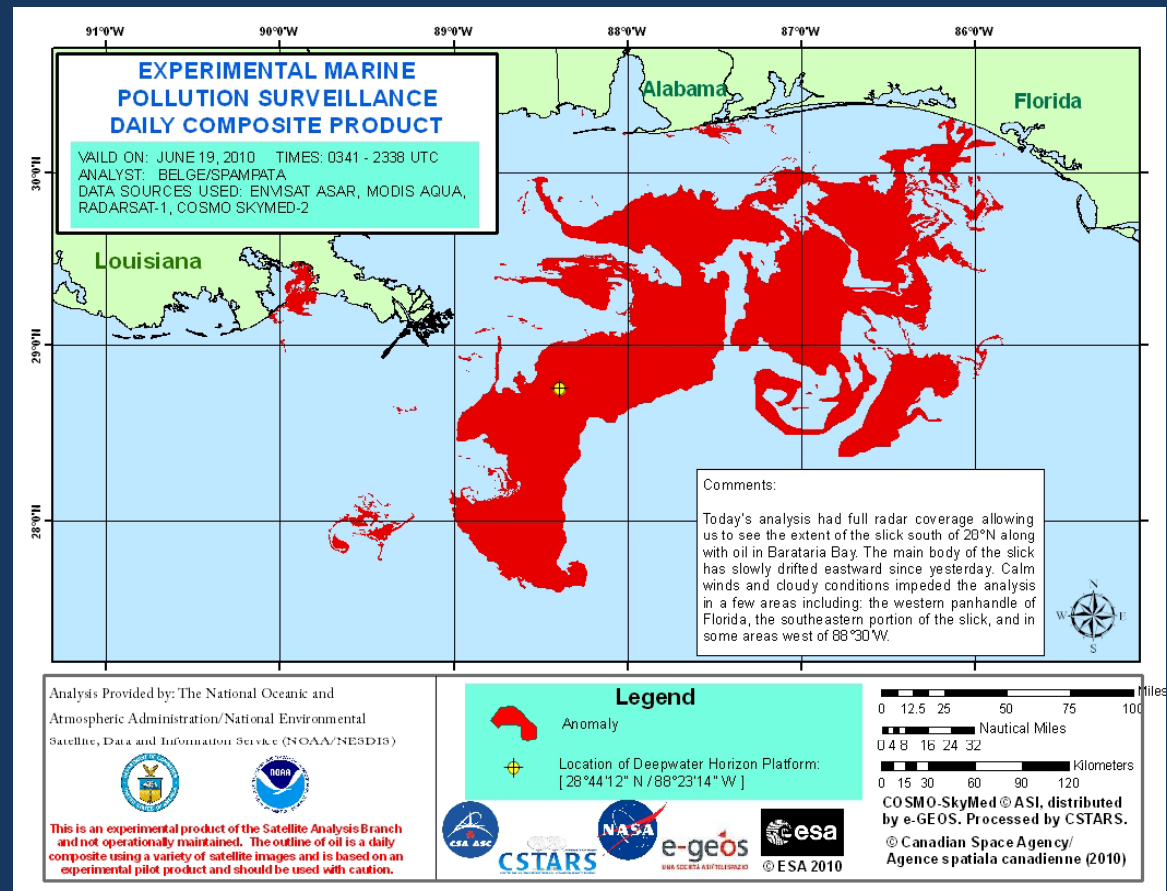


Can we estimate the total impact of microbial respiration?

Experimental Marine Pollution Surveillance Reports: slick 5,600 to 24,000 km² during our cruise.

Extrapolating enhanced respiration over slick area in surface mixed layer of 15 m yielded an *estimated potential* microbial respiration rate of 6×10^8 to 3×10^9 moles C d⁻¹

On the same order as oil leaking from well, which averaged 6×10^8 moles C d⁻¹.

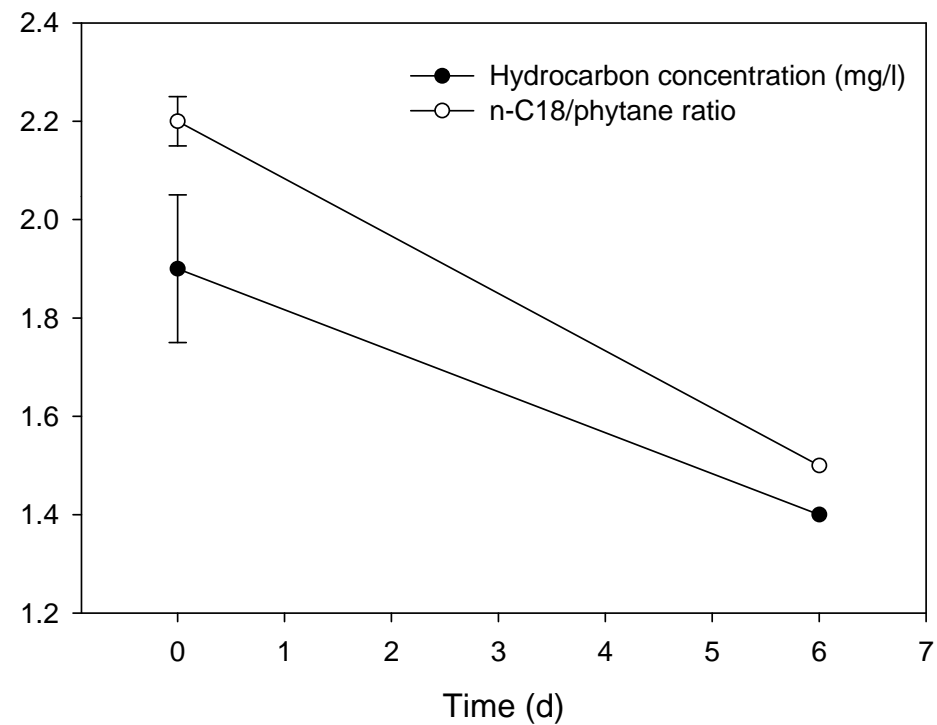


Respiration in slick was fueled by hydrocarbon degradation

Hydrocarbon degradation equivalent to $7.1 \mu\text{M O}_2 \text{ d}^{-1}$, which is 71% of observed enhanced respiration rate of $9.9 \mu\text{M O}_2 \text{ d}^{-1}$.

First-order rate law yields hydrocarbon degradation rate of about 4 to 6% per day.

Decrease in ratio of *n*C18/phytane is consistent with microbial degradation.

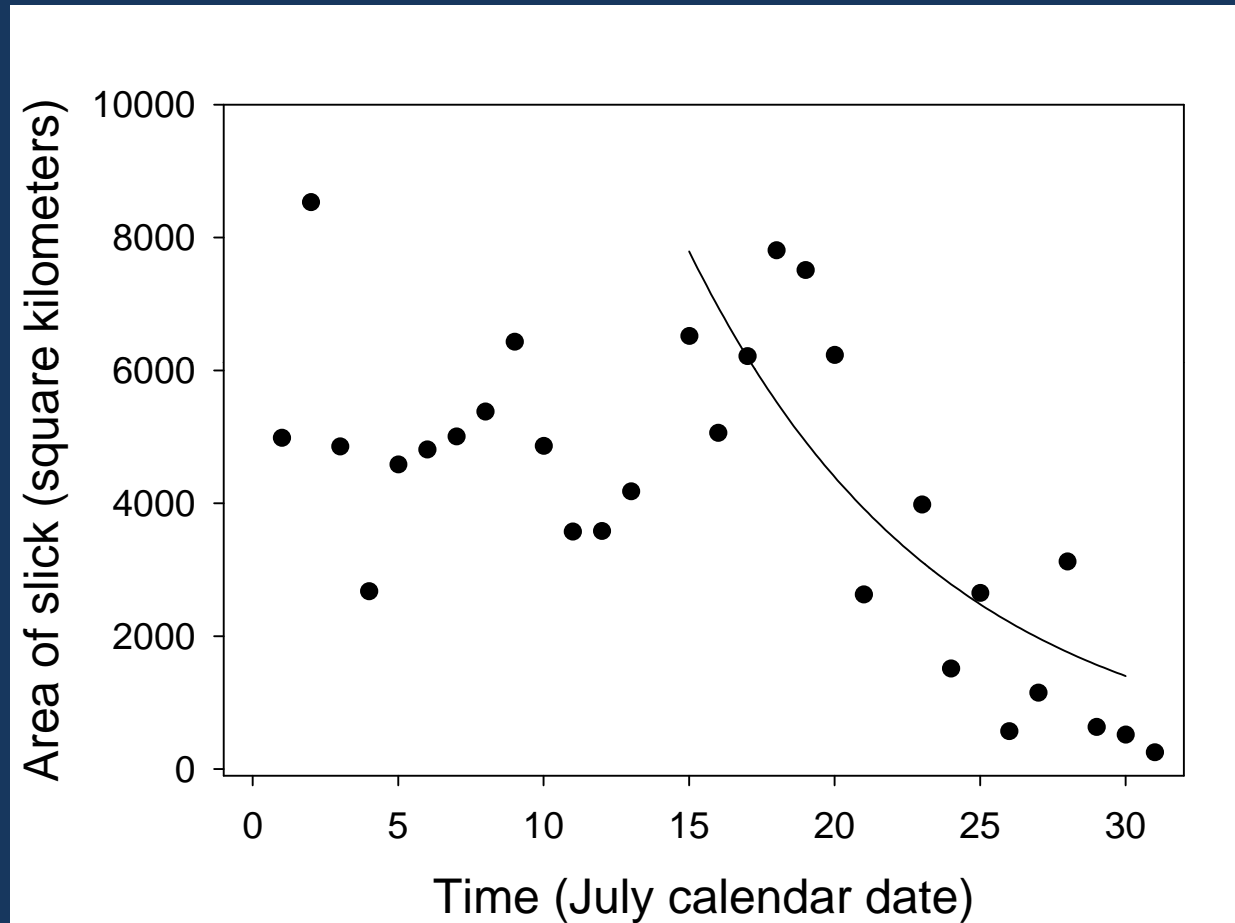


How to our observed hydrocarbon degradation rates compare?

Experimental Marine Pollution Surveillance Reports show a rapid decrease in the area of the slick.

After July 15th, the first-order disappearance rate was about 8 to 12 % per day ($R^2=0.7020$)

Compare to hydrocarbon degradation rate from bottles of 4 to 6% per day.

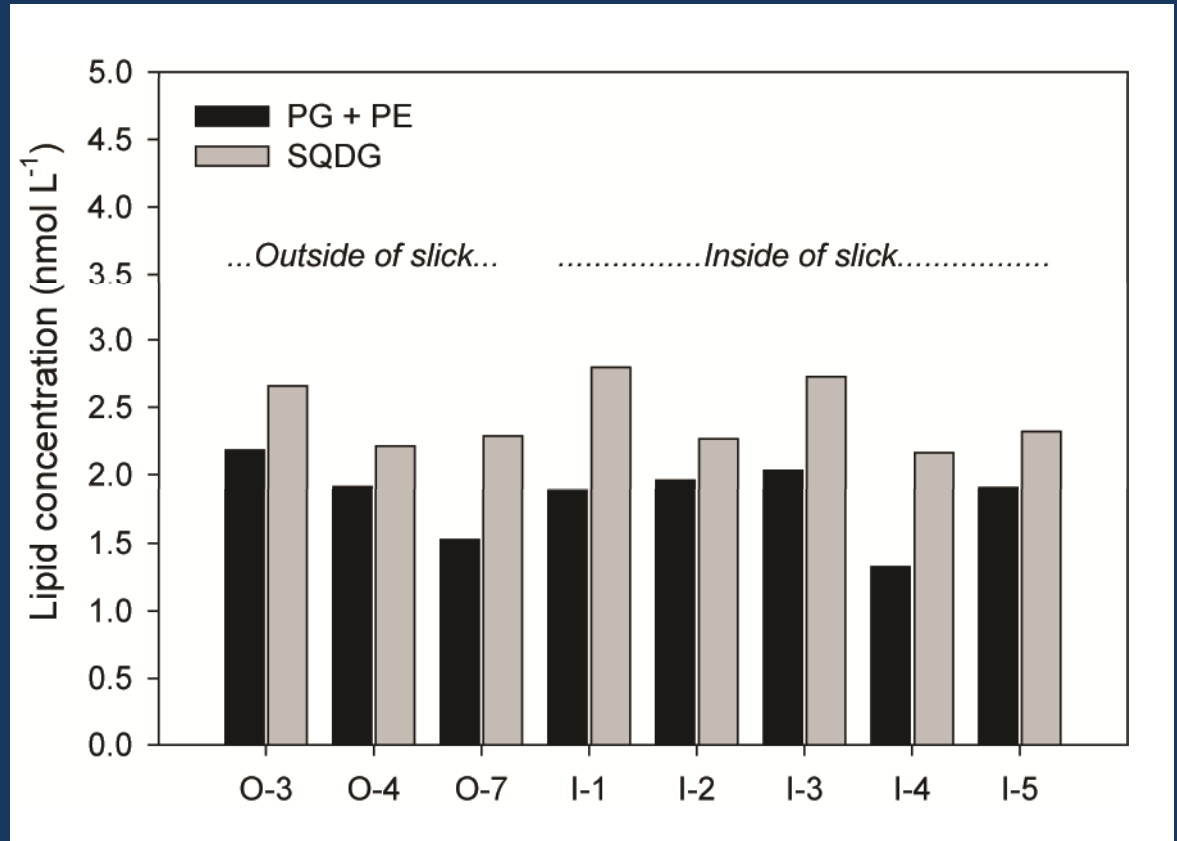


Enhanced respiration in slick, but no increase in microbial biomass

Phosphatidylglycerol (PG) and phosphatidylethanolamine (PE) are common membrane phospholipids in marine heterotrophic bacteria.

Sulfoquinovosyldiacylglycerol (SQDG) abundant in picocyanobacteria.

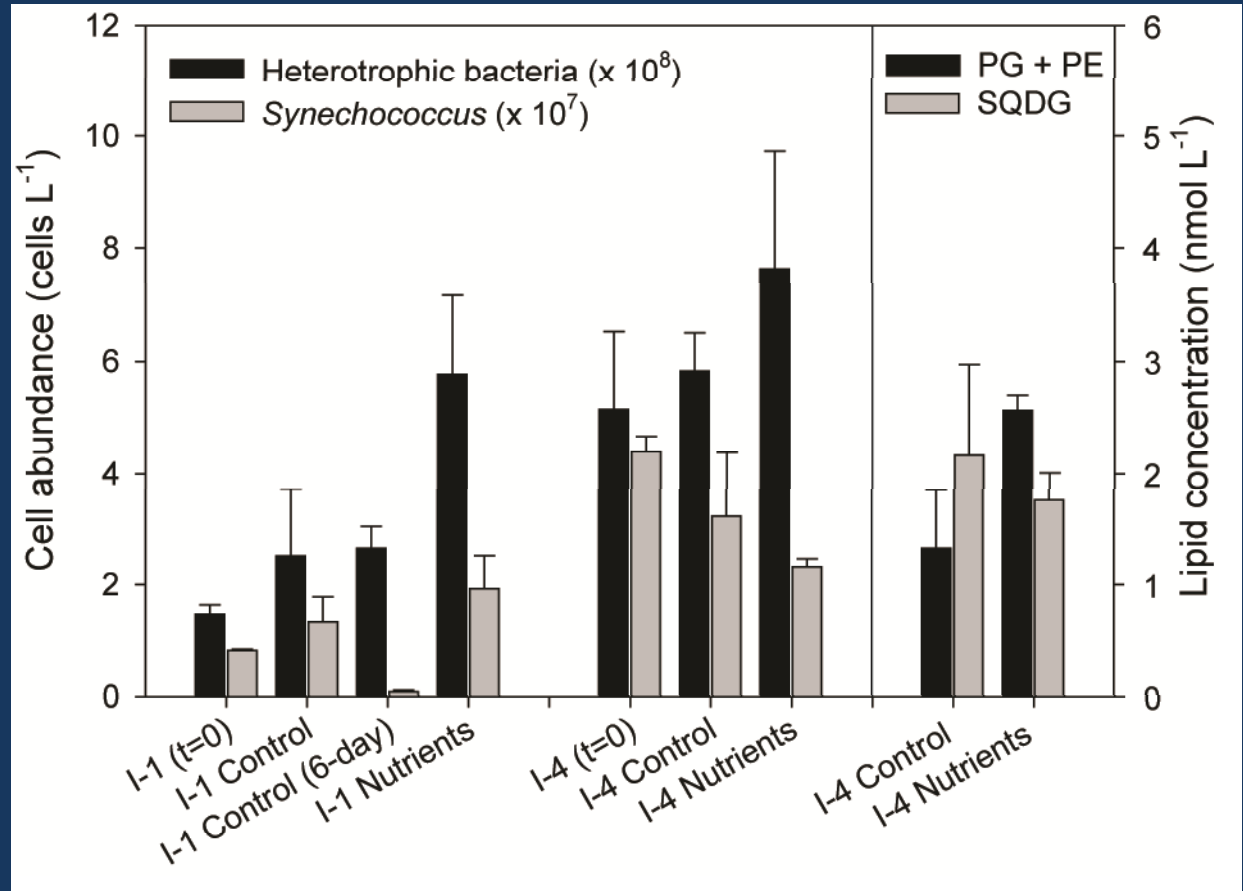
Cell abundances by flow cytometry show fewer microbes in slick (not shown); sampling artifact?



Van Mooy et al., (2006), *PNAS*; Van Mooy et al., (2009) *Nature*; Van Mooy and Fredricks (2010) *Geochim. Cosmochim. Acta*; Pendorf et al. (2011) *Org. Geochem.*

Microbial growth limited by nutrients.

Microbial abundances and biomass increase in response to nutrient amendments, in some cases by as much as a factor of two.



Conclusions

Hypothesis I: Microbes unable to respond to oil in surface slick → not supported.

Oil in slicks led to anomalously high rates of microbial respiration.

Respiration rates of $9.9 \mu\text{M O}_2 \text{ d}^{-1}$ extrapolated across the slick indicated that microbes may have had the potential to remove all of the oil from the surface slicks. *This potential was could NOT have been realized.*

Hydrocarbon degradation proceeded at a rate of about 4 to 6% per day, which based on our crude analysis of satellite data products suggests microbial degradation was responsible for between 30 to 80% of slick disappearance.

Hypothesis II: Microbes limited by inorganic nutrients → some support.

Microbial biomass was similar inside and outside of the slick suggesting that growth was limited by nutrients.

Nutrient amendments stimulated both respiration and growth.

Thank you.

