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Microbial Community Characterization as an Approach to Addressing Bioremediation Challenges in the Canadian Arctic

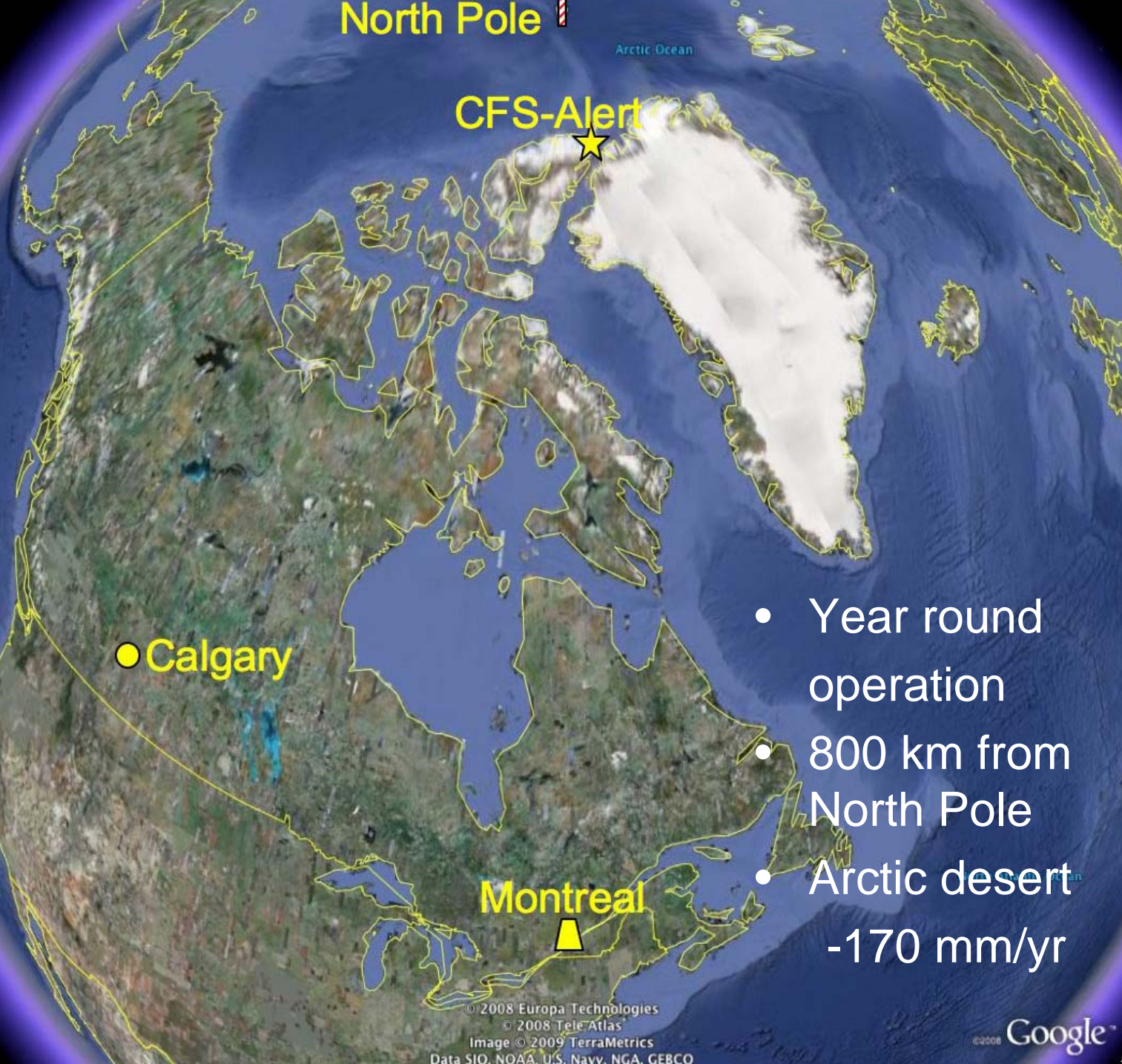
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National Research
Council Canada

Conseil national
de recherches Canada

Canada 



CFS-Alert

- Year round operation
- 800 km from North Pole
- Arctic desert
-170 mm/yr

CFS-Alert, Nunavut





Large berm

LO

LN

Control biopile

Small berm

SO

SN

Excavated areas

Bioremediation Challenges in the Arctic

- Extreme environment (polar desert)
 - temperature
 - water activity
 - nutrient status
- Operational window
 - approximately 2 months/year
- Logistics
 - remoteness
 - simple, practical approaches required

Deciphering the Microbial “Black Box”

The microbial community is responsible for contaminant degradation and genomics tools can help decipher the who, what, where and how

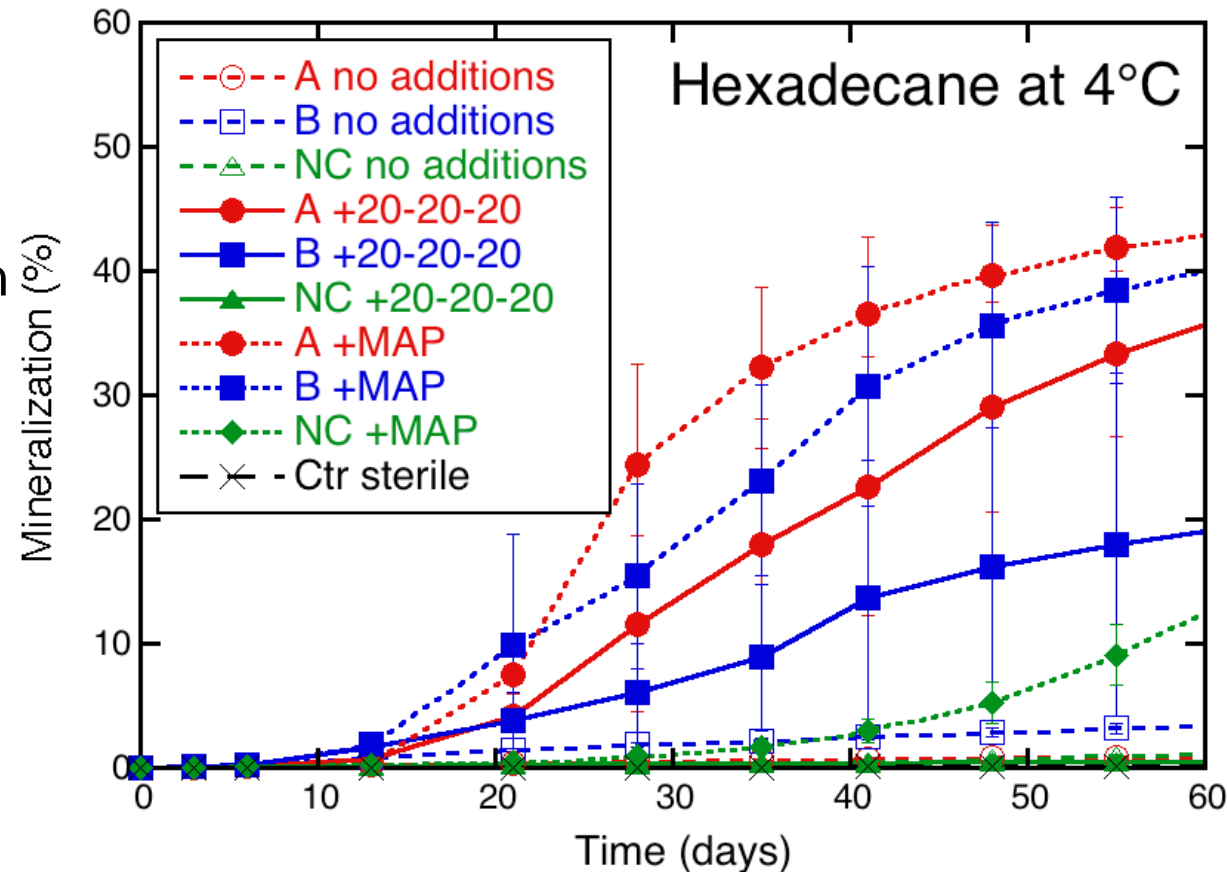
- knowing who's there provides important information on capabilities
- knowing what key genes are present identifies potential
- showing which genes are functional indicates who is active

Initial Laboratory Evaluation

- different fertilizers had a significant effect
- mineralization activity with nutrients good at 4°C
- virtually no activity in unfertilized soil

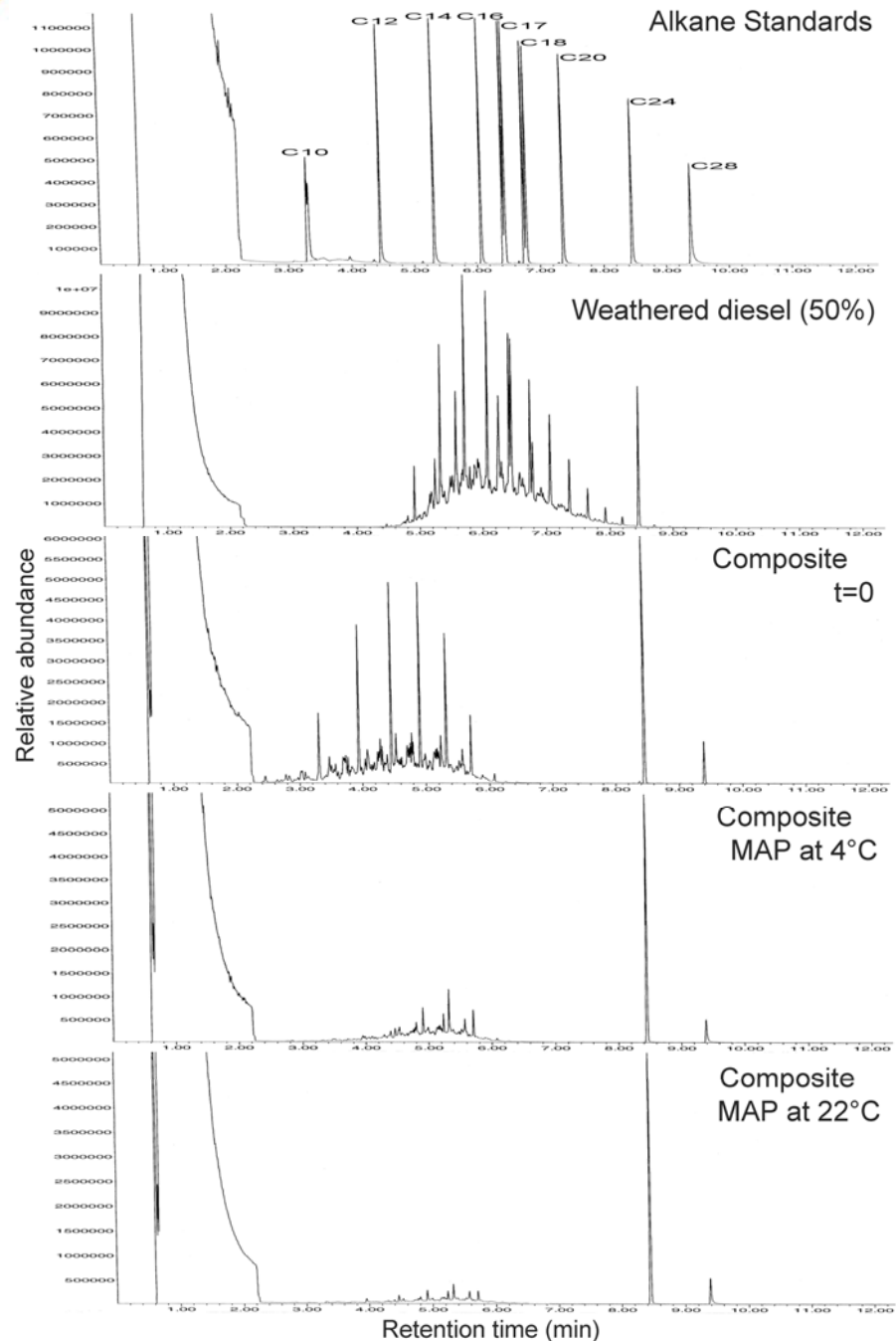
Mineralization

Determines the potential of the indigenous microbial community to degrade a representative hydrocarbon substrate



Diesel Degradation in Laboratory Mesocosms

- extensive degradation with fertilizer addition
- temperature effect with degradation greater at 22°C than at 4°C



2008

Large berm



N

Old
(LO)

New
(LN)

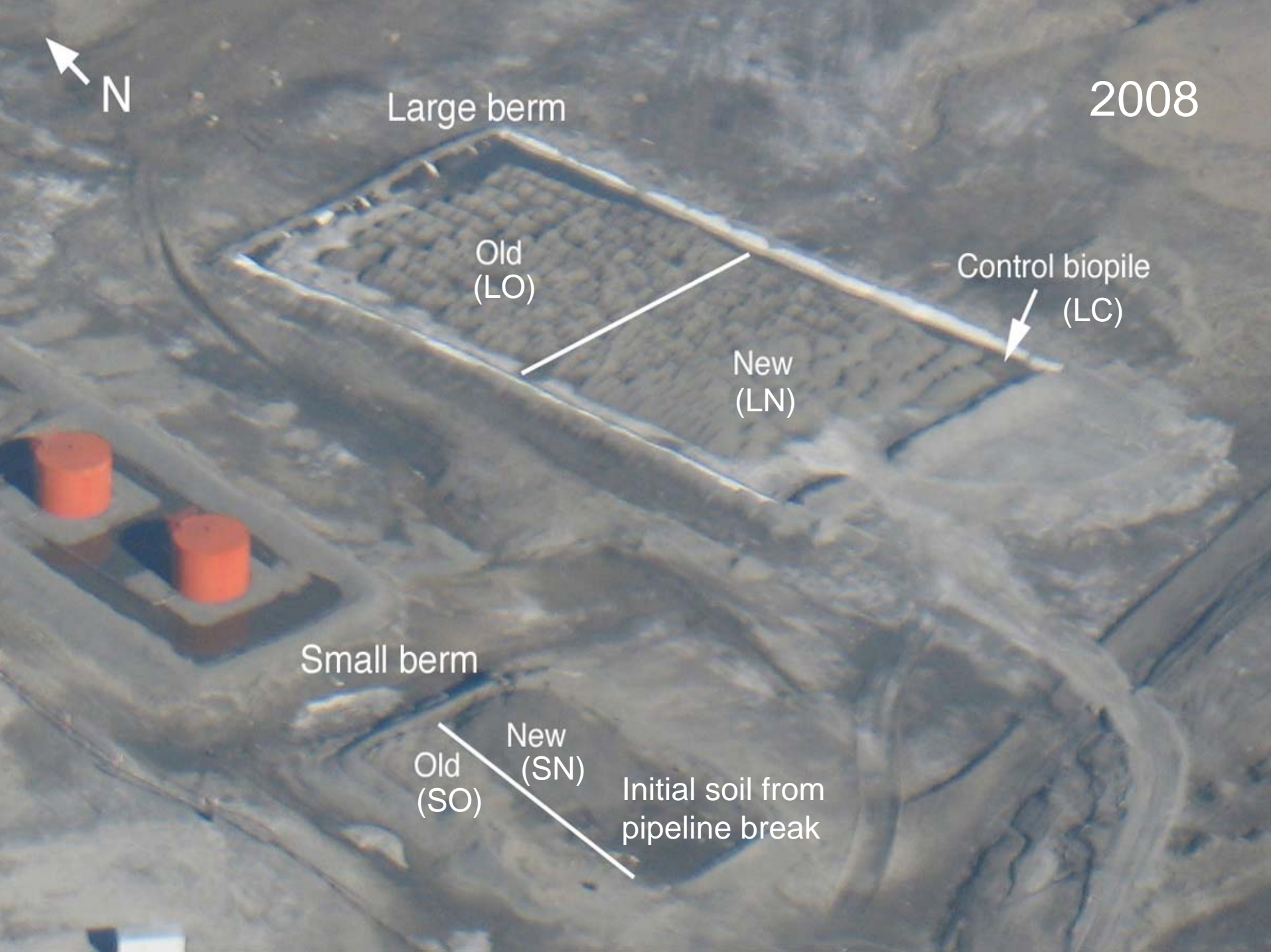
Control biopile
(LC)

Small berm

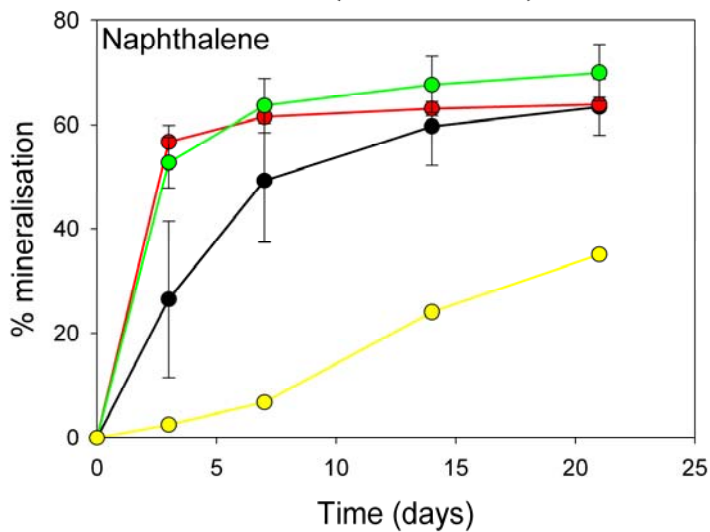
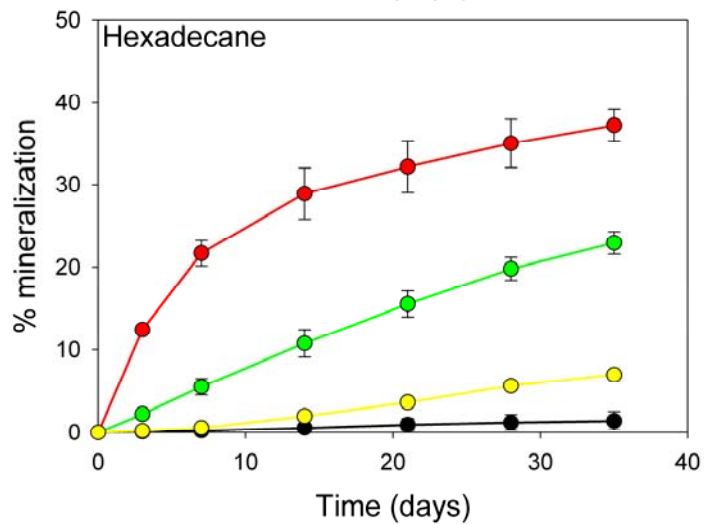
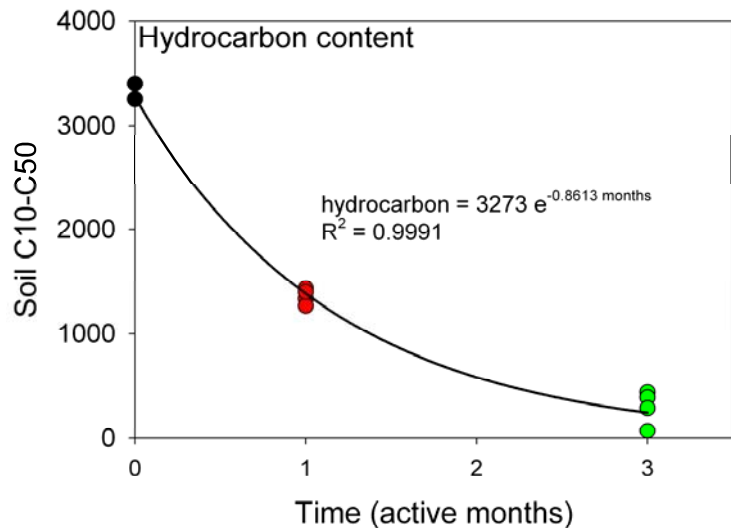
Old
(SO)

New
(SN)

Initial soil from
pipeline break

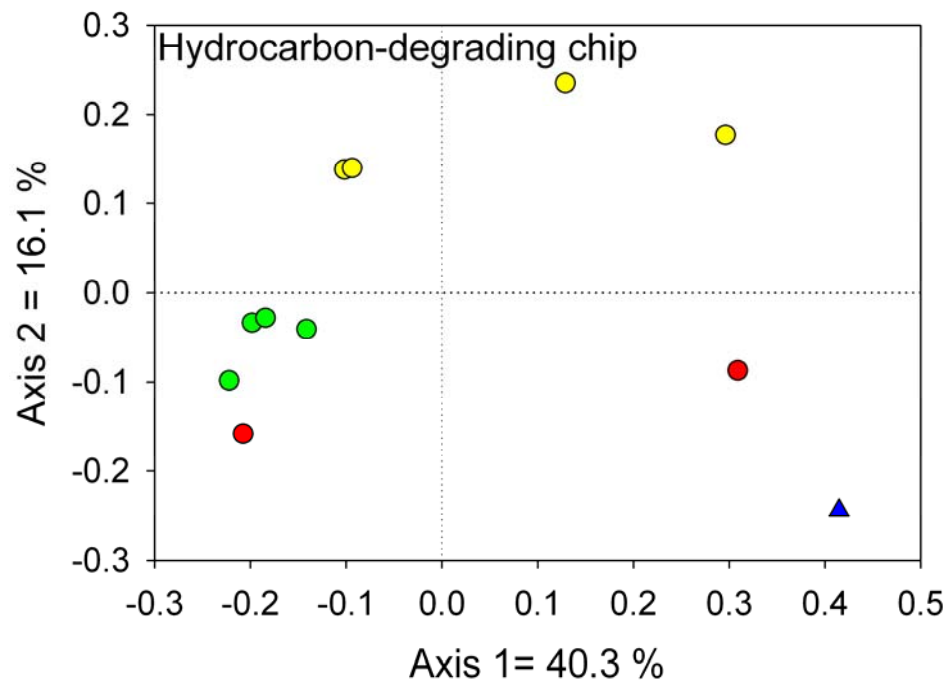
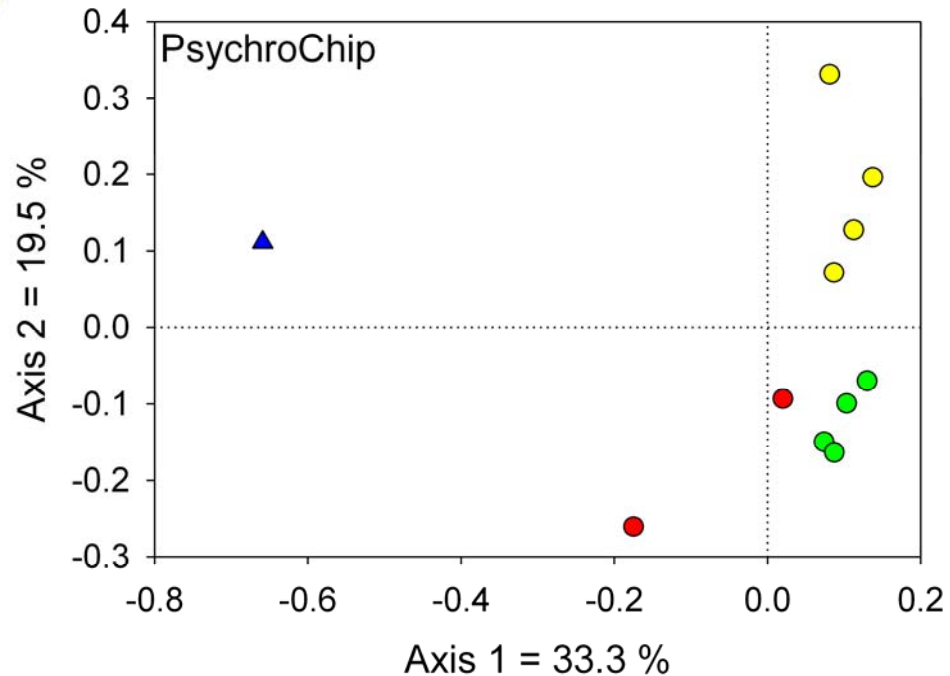


Hydrocarbon degradation and mineralization

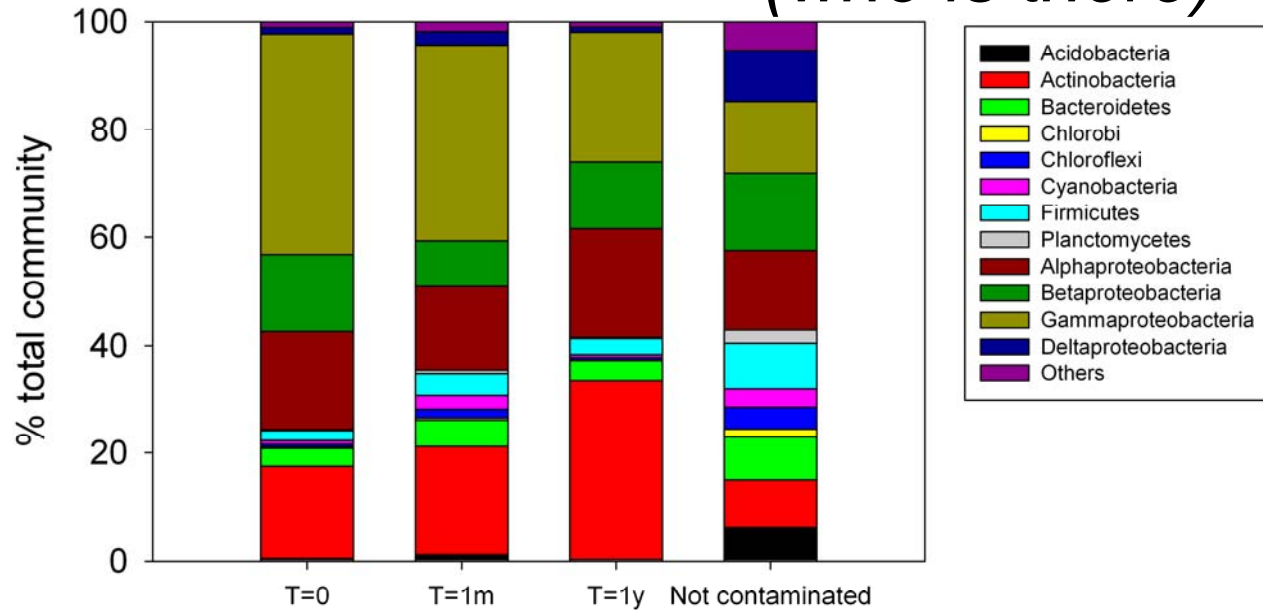


Microarray Analysis

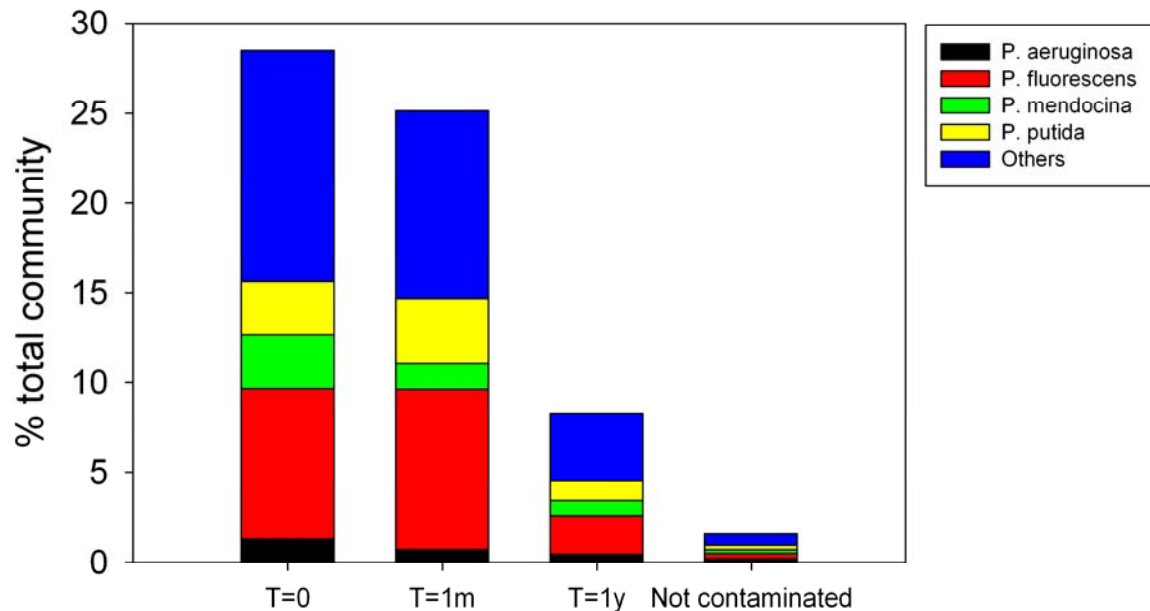
- Samples from different times clustered together
- Microbial community changed over time
- Which bacteria are responsible for hydrocarbon degradation activity?



Microbial Community Structure (who is there)



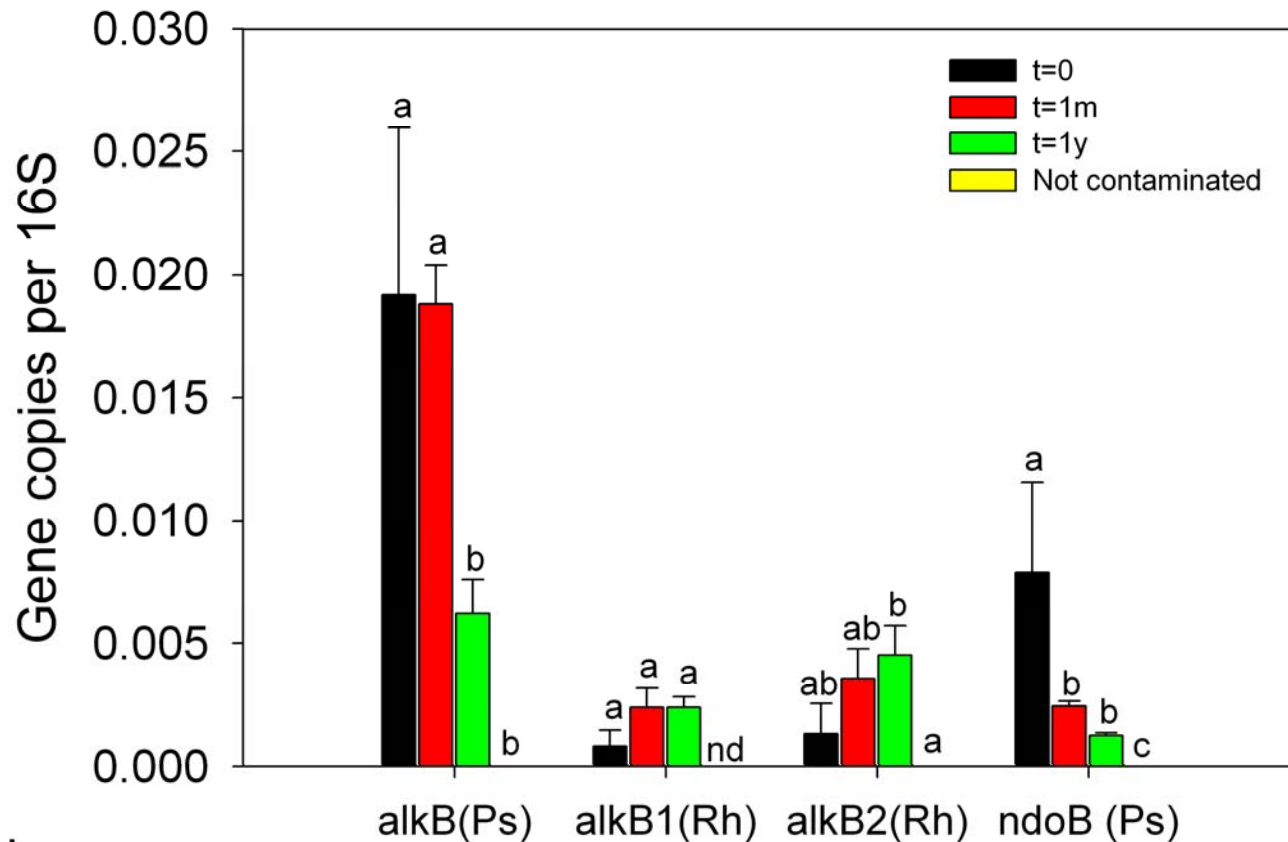
- decrease in diversity upon contamination
- high initial numbers of *Gammaproteobacteria*, decreased over time
- increase over time in *Actinobacteria*



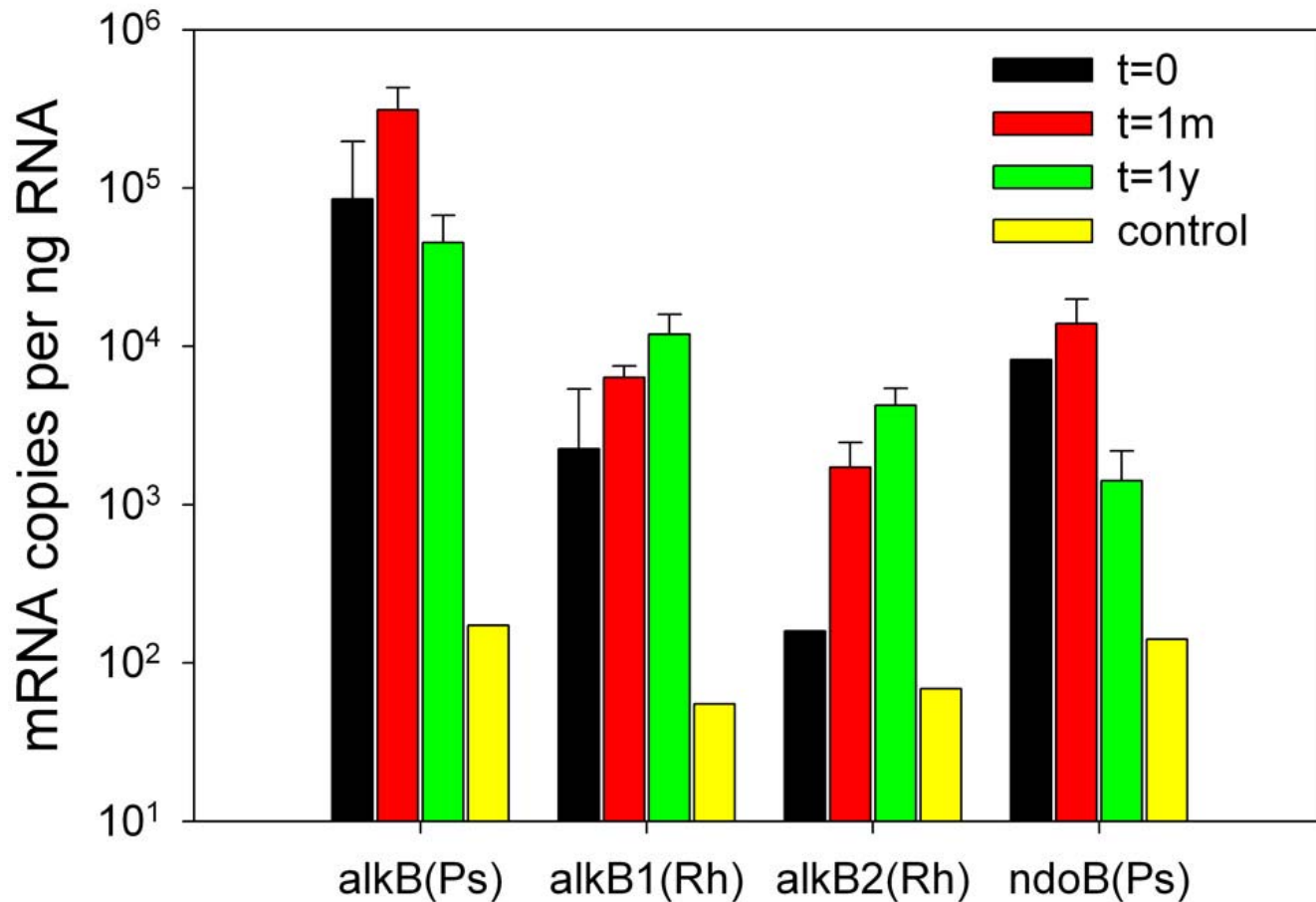
- main *Gammaproteobacteria* are *Pseudomonas* species
- low numbers of *Pseudomonas* in pristine soil
- increased dramatically following contamination

Quantitative PCR (who has the degradation genes)

- *alkB* (Ps)-soil hydrocarbon: $r = 0.738$, $P=0.00945$
- *alkB1* (Rh)-soil hydrocarbon: $r = -0.703$, $P=0.0234$



Quantitative RT-PCR (who is actively degrading)



Conclusions

- The microbial community structure changed dramatically in response to contamination and later during remediation
- Fertilizer addition had a positive impact on hydrocarbon biodegradation
- Key bacteria (eg. *Pseudomonas*), known to be hydrocarbon degraders, responded positively to fertilizer addition
- Known hydrocarbon degradation genes (*alkB*, *ndoB*) showed increased numbers corresponding to increased degradation activity
- Predominant hydrocarbon degraders were *Pseudomonas* spp. while nutrients available: other bacteria, such as *Rhodococcus* spp. became more dominant when degradation activity and nutrient concentrations decreased

Next Steps

- Optimize field conditions for *Pseudomonas* spp. or bioaugment field sites with *Pseudomonas* spp. and monitor performance/activity
- Isolation and characterization of Arctic *Pseudomonas* responsible for high degradation activity at low temperature
- Design molecular tools to track the degradation activity of specific *Pseudomonas* spp.

Soil Bioaugmentation and Aeration (2008)



Acknowledgements

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