Total Microbial Profiling…
Because Sometimes Bacteria Just Aren’t Enough.

HydroQual Laboratories Ltd
*Alberta Ingenuity r&D Associate
• Athabasca oil sands
• Bioremediation
• Methods
• AIF r&D associateship project
• Results
• Conclusions
The process used to extract oil from the oil sands requires large amounts of water and caustics.

The products of this process are bitumen and tailing slurry.

Creation of lakes and fens are viable reclamation options for mine wastes.

One of the major concerns is the release of naphthenic acids and other potentially toxic substances from the tailings over time.
Natural indigenous microbes have the ability to remediate pollutants

Cooperative effect between the total microbial community

Bacteria, fungi, archaea might be equally important in the process
Conventional techniques

- Culturing techniques e.g. plating, MPN
  - Inherent biases
  - Slow process
  - <1% can grow in lab conditions
Molecular techniques

- Rapid
- More complete
- No cultivation required
- Only small sample sizes required
- We can analyze bacteria, archaea, algae, fungi
- Complement to other analyses
DNA Fingerprinting

- Identify individuals within a population
- CSI, genetic testing, genealogy
- Create a fingerprint of the population
- Monitor population changes
How Do You Fingerprint DNA?

1. Collect sample
2. Extract nucleic acids from sample (DNA, RNA or both)
3. Amplify target gene (e.g., 16S rRNA gene) using PCR
4. Separate PCR products using a denaturing gradient
5. PCR product separation by composition and not size
Limitations of the molecular techniques

- Can’t detect metabolic activities
- It can’t exclude dead bacteria
Total microbial profiling of tailing ponds

- 7 different ecosystems (tailing ponds)
  - Comparing the total microbial profile over time using DNA profiling techniques

- Total microbial community profile includes not only bacteria but also archaea, fungi, cyanobacteria and algae

- Because Sometimes Bacteria Just Aren’t Enough.
Field work

Water and sediment sampling from an experimental tailings pond
Bacterial profile

- The number, precise position and intensity of the bands gives an estimate of the number and relative abundance of dominant species in the samples

- EC: *E. Coli* positive control
- 1-7: Number of tailing pond
- Archaea is also known as extremophiles.
- Live in some of the most extreme environments on the planet.
- Involved in biodegradation of hydrocarbons.
Results

- Seasonal variations in the bacterial and archaeal populations
- No detection of archaea in pond water
- We are in the process of analysing the fungal and cyanobacterial populations in all of our samples collected
Characterization of the community growing in the lab in microcosms

- Duplicates
  - One anaerobic and one aerobic
  - Same pond, same water & sediment
- Stored in dark at 18-22 °C
Microcosms Day 14
2 months later

7 months later
Bacterial profile of Aerobic vs Anaerobic microcosms

AERobic MICROcosms

WATER

SEDIMENT

ANAERobic MICROcosms

WATER

SEDIMENT

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ALBERTA INGENUITY FUND

Golder Associates
Archaeal profile for microcosms

AERobic Microcosms

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Anaerobic Microcosms

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Bacterial profile comparison

I- October field sample, II-May , III-August ,  + Aerobic Microcosm,  - Anaerobic Microcosm, EC & PA  positive control
Conclusions

- Bacterial communities found in the microcosms have not changed from the source material
  - Opportunities for remediation testing
- No archaea detected in test pond water, archaea detected in microcosm water
  - Is it the depth difference?
- Future directions include analysis of fungi and cyanobacteria
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Questions?

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