

# Testate Amoebae (Protozoa) and Microbial Communities as Bioindicators in Wetlands Impacted by Oil Sands Processed Materials (OSPM)

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## Summary

Microbial communities in wetlands are the foundation on which all higher trophic levels in the ecosystem depend. Generally, bacteria, fungi, and algae are regarded as the predominant components of wetland microbial communities. Among the different groups of protozoa occurring in wetlands, testate amoebae have been shown to represent up to 25% of total microbial biomass in wetlands, especially peatlands. Testate amoebae have the added advantage over most other microbial groups in that they can be readily identified to the species level, even after the death of the organism, and there is a growing knowledge base of their ecology in northern wetlands. Recent studies have shown them to be sensitive to hydrology and hydrogeochemistry. There have been no previous investigations of testate amoebae in OSPM wetlands.

Testate amoebae from OSPM wetlands show differences in species composition and abundance in natural (peatlands and marshes) and created wetlands (marshes and open water wetlands), including both OSPM-affected and non-OSPM affected wetlands. Testate amoebae abundance appears to be limited in OSPM-affected wetlands compared to non-OSPM sites. Genera such as *Diffflugia* may be attributed to OSPM-affected mineral wetlands while genera such as *Nebela* may be indicative of non-OSPM mineral wetlands. Total microbial biomass for bacteria, fungi and testate amoebae are greatest for peatland sites compared to mineral wetland sites; biomass numbers may be slightly greater in non-OSPM wetlands compared to OSPM wetlands but needs further confirmation. Testate amoebae show considerable potential as bioindicators given their limited occurrence in OSPM wetlands and greater occurrence in non-OSPM wetlands, especially in comparison to other more dominant components of the microbial community such as the bacteria and fungi.

Studies on bacteria revealed clear functional and genetic differences respectively between OSPM and non-OSPM wetlands. Community level physiological profiling (CLPP) with BIOLOG™ Ecoplates revealed a trend in which communities in OSPM sites were most similar to each other, followed by those in natural sites, and communities in reference wetlands were variable. The vegetated areas of a wetland displayed increased functional richness and diversity compared to non-vegetated areas. Analyses using the genetic approach, denaturing gradient gel electrophoresis (DGGE), applied to the total bacterial community, revealed the highest number of operational taxonomic units in OSPM samples. When DGGE was applied to the bacterial subgroups,  $\gamma$ -Proteobacteria and Actinomycetes, the distinction between site type was more evident than when a total bacterial approach was taken. Within OSPM wetlands,  $\gamma$ -Proteobacteria populations were varied while Actinomycetes were similar across sites. Given that CLPP and DGGE could distinguish between communities based on site type, both may be

useful for monitoring microbial communities in Athabasca wetlands throughout reclamation. In particular, BIOLOG™ is a simple technique that might be a practical tool for monitoring microbial communities and group-specific DGGE analyses are recommended for future research. This was the first study of testate amoebae and the first to attempt to characterize and quantify biomass of components of the microbial community in OSPM wetlands; further work on a larger field data set is needed to confirm the trends and implications of the results revealed in this study. We focused primarily on the total bacteria, fungi and testate amoebae components of the microbial community; future studies should include other components such as the micro-algae, ciliates, archaea, as well as other specific bacterial groups such as cyanobacteria.

### **Acknowledgements**

We are grateful to the Cumulative Environmental Management Association (CEMA) and NSERC for financial support.