## Linking Ecosystem Function to Methanogen Community Structure in Peatland Soils

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### Microbial ecology: a core question

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 Does spatial and temporal variation in microbial diversity and activity contribute to the control of key biogeochemical processes?

## Methanogens: a small community, an essential function

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 Two distinct biochemical pathways that are phylogenetically distributed within the methanogens, and can be quantified independently via radioisotope labeling Figure courtesy of Dr. Jason Keller, Chapman University



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 $CH_3COOH \rightarrow CH_4 + CO_2$ 

$$CO_2 + 4 H_2 \rightarrow CH_4 + 2H_2O$$



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$$^{14}\text{CO}_2$$
 + 4 H<sub>2</sub>  $\rightarrow$   $^{14}\text{CH}_4$  + 2H<sub>2</sub>O

Addition of radioactive substrate allows direct quantification of each pathway!

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 Vulnerable to large increases in mean annual temperature in the coming decades

рΗ

hydrology

methane pathway



#### ombrotrophic



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## **Study Sites**



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## ombrotrophic Study Sites

- Bog
- Bog
- Poor Fen
- Intermediate Fen
- Cedar Swamp
- Rich Fen













## **Study Sites**

#### Sampling:

- 5 events (two in 2009, three in 2010)
- 5 replicate samples from each site
- Samples incubated at average in situ temperature with <sup>14</sup>C-labeled bicarbonate tracer











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 as expected, there was a significant shift in dominant pathway across the gradient, with acetoclastic methanogenesis becoming increasingly dominant in more minerotrophic sites





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• the increase in total methane was instead driven by an explosive increase in hydrogenotrophic methanogenesis, which increased significantly in all sites in the summer and fall!

• what could account for this interannual variability in hydrogenotrophic methanogenesis?



Water table depth appears to strongly effect hydrogenotrophic methanogenesis



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- Hydrogenotrophic methanogenesis, on the other hand, varied tremendously between 2009 and 2010, becoming dominant in nearly every site in the latter year, which was unusually wet
- Could the substantially different spatial and temporal dynamics of the two pathways be explained by the phylogenetically distinct methanogens carrying them out?

# Community Structure Analysis: *mcrA*, functional gene marker about town

- *mcrA* is a gene that codes the alpha-subunit of methyl coenzyme-M reductase (MCR), which catalyzes the terminal step of methanogenesis in *all methanogens*
- Only one copy per genome, simplifying quantification



## Preliminary Community Data

 DNA was extracted from one core from each site taken in May 2010

 mcrA was amplified from each core using PCR, and the PCR amplicons were cloned and sequenced via Sanger sequencing

 the resulting sequence libraries were trimmed, aligned, and binned into operational taxonomic units (putative genera) using the MOTHUR microbial genomics software package







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- All communities dominated by putative hydrogenotrophs

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- The interannual variability in methanogenesis within the peatland sites appears to be driven by changes in hydrogenotrophic methanogenesis rates
- Putative hydrogenotrophs are dominant in all May 2010 community samples
- Acetoclastic methanogenesis correlate very strongly to gradient position, while hydrogenotrophic methanogenesis does not

### Hypotheses

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- H2: Hydrogenotrophic methanogenesis in northern peatlands is primarily driven by dynamic or opportunistic changes in hydrogenotroph community activity by season, resulting in a strong temporal pattern of rate variation

• Thoroughly sample the *mcrA* DNA of each site during all seasons, to determine if the total communities remain stable throughout the growing season (H1) using high-throughput sequencing (454 pyrosequencing)

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- Use SEM and NMS to compare effects of community and hydrogeomorphic context on the two pathways (H1)
- Thoroughly sample *mcrA* mRNA from each site and sampling event, to determine if the transcriptional activity of functional methanogen groups fluctuates in sync with the rates of their associated pathways (H2)

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