Microbial processes at the oxycline of humic lakes

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Humic lakes

- Typical lake type in boreal region
- Small, but numerous
Humic lakes

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- Small, but numerous

> 170 000

Produced by Sam Carana based on Peel et al. doi:10.5194/hess-11-1633-2007
Humic lakes

- Receive high loads of organic matter from the catchment
  - High concentration of dissolved organic carbon (DOC)
  - Dark water colour
  - Steep oxygen and nutrient gradient from surface to bottom
Metabolism in humic lakes

• Primary production limited to surface layer

• High heterotrophic production

Net heterotrophy

Important sources of greenhouse gases, especially methane
Previously freshwater microbiology has focused on the oxic water compartment.
Freshwater microbes

- Proportion of unclassified 16S genes is low in oxic freshwater samples

Freshwater microbes

- Database coverage decreases with decreasing oxygen concentration
Freshwater microbes

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Freshwater microbes

- Amplicons give only information on diversity for the environments of poor database coverage
Microbial communities in humic lakes – 16S

Peura et al. 2012, ISME J.
Microbial communities in humic lakes – 16S

- Hypolimnetic community is more diverse but also more stable than epilimnetic
Functional diversity - metagenomes

COG = Cluster of Orthologous Groups
Functional diversity

Proportion of genomes with the COG

COG = Cluster of Orthologous Groups
Proportion of genomes with the COG

Information storage and processing
Cellular processes and signaling
Energy production and conversion
Carbohydrate transport and metabolism
Amino acid transport and metabolism
Nucleotide transport and metabolism
Coenzyme transport and metabolism
Lipid transport and metabolism
Inorganic ion transport and metabolism
Secondary metabolites biosynthesis, transport and catabolism
Poorly characterized
Multiple classes
Degradation (carbohydrates)

• No differences between layers

• Full pathways for starch degradation in all samples and for glycogen in all but one sample, only partial for cellulose
Aerobic and anaerobic respiration coexist in the hypolimnion
Respiration in the anoxic layer

- Missing ubiquinol-cytochrome c reductase in one lake
Electron acceptors in the anoxic layer

- Chemical analyses suggest that possible acceptors include nitrate, nitrite and sulfate

<table>
<thead>
<tr>
<th>Layer</th>
<th>Dissimilatory NO\textsubscript{3} reduction</th>
<th>NO\textsubscript{2} reduction</th>
<th>Dissimilatory SO\textsubscript{4} reduction</th>
<th>Sulfur reduction</th>
<th>Iron reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epilimnion</td>
<td>3/3</td>
<td>2/3</td>
<td>0/3</td>
<td>3/3</td>
<td>2/3</td>
</tr>
<tr>
<td>Metalimnion</td>
<td>2/3</td>
<td>1/3</td>
<td>2/3</td>
<td>3/3</td>
<td>0/3</td>
</tr>
<tr>
<td>Hypolimnion</td>
<td>2/3</td>
<td>2/3</td>
<td>2/3</td>
<td>2/3</td>
<td>0/3</td>
</tr>
</tbody>
</table>
Methane

- Majority of methane production is expected to happen in the sediment, although it has been shown to occur also in the water column (Grossart et al., 2011, PNAS)

- Methane oxidation expected to be significant contributor to the total lake metabolism (Bastviken et al., 2003, Ecology)

- Highest oxidation rates measured from the oxygen transition zone (Kankaala et al., 2006, Limnol. Oceanogr.)

- Isotopic evidence of methane oxidation in the anoxic/suboxic layer (Peura et al., 2012, ISME J.)
Methane

**a** *Methylomirabilis oxyfera*

Nitrate reducers

\[ 2\text{NO}_3^- \rightarrow 2\text{NO}_2^- \rightarrow 2\text{NO} \rightarrow \uparrow \text{N}_2 \rightarrow \text{O}_2 \rightarrow 2\text{NO}_2 \rightarrow \text{CH}_4 \]

**b** Methanogenesis

Complex organics

Anaerobic food web

Fermentation products

\[ 4\text{H}_2 + \text{CO}_2 \rightarrow \text{CH}_4 + 2\text{H}_2\text{O} \]

Methanogenic archaea

**c** Aerobic oxidation of methane

\[ \text{O}_2 \quad \text{Oxic} \quad \text{CH}_4 + 2\text{O}_2 \rightarrow \text{CO}_2 + 2\text{H}_2\text{O} \]

**d** Reverse methanogenesis (AOM)

\[ \text{SRB} \
\text{SO}_4^{2-} + 2\text{H}^+ \rightarrow \text{H}_2\text{S} \rightarrow \text{ANME} \rightarrow [?] \rightarrow \text{CO}_2 + 2\text{H}_2\text{O} \]
Methane

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Nitrate reducers

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\begin{align*}
2\text{NO}_3^- & \rightarrow 2\text{NO}_2^- \\
\text{N}_2 & \rightarrow 2\text{NO} \\
\text{O}_2 & \rightarrow \text{CO}_2 + \text{H}_2\text{O}
\end{align*}
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- Anaerobic food web
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\text{CH}_4 \\
\begin{align*}
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\text{Methylococcus capsulatus}
\end{align*}
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\]

Oremland, 2010, Nature
Methane

5-methyltetrahydrosarcinapterin:corrinoid/iron-sulfur protein Co-methyltransferase

Methyltransferase

Acetogenic

Oremland, 2010, Nature
Methane

**Acetogenic**

**Hydrogenotrophic**

**Tetrahymethanopterin S-methyltransferase**

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Oremland, 2010, Nature
Methane

a. *Methylophilis oxyfera*
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*Methylococcus capsulatus*

d. Reverse methanogenesis (AOM)

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Oremland, 2010, Nature
Methane

**Methylomirabilis oxyfera**
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- $2\text{NO}_2^{-} \rightarrow 2\text{NO} \rightarrow \uparrow \text{N}_2 \rightarrow \text{O}_2 \rightarrow \text{CO}_2 + \text{H}_2\text{O}$
- $\text{M. oxyfera} \rightarrow \text{CH}_4$

**Methanogenesis**
- Complex organics
- Anaerobic food web
- Fermentation products
- $4\text{H}_2 + \text{CO}_2 \rightarrow \text{CH}_4 + 2\text{H}_2\text{O}$
- Methanogenic archaea

**Aerobic oxidation of methane**
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- Oxic
- Anoxic
- $\text{CH}_4 + 2\text{O}_2 \rightarrow \text{CO}_2 + 2\text{H}_2\text{O}$

**Reverse methanogenesis (AOM)**
- $\text{SO}_4^{2-} + 2\text{H}^+$
- SRB
- $\text{H}_2\text{S} \rightarrow \text{ANME} \rightarrow \text{CH}_4 \rightarrow [?] \rightarrow \text{CO}_2 + 2\text{H}_2\text{O}$

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Methylococcus capsulatus

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Oremland, 2010, Nature
Conclusions

It can be expected that unknown community members have a significant contribution to elemental cycles in humic lakes.

Metagenomes confirm the capacity for most predicted functions.

Possible “new”/non-matching enzymes in some predicted/expected pathways.

Future perspectives
  • Compiling composite genomes for metabolic analyses
  • Incubation experiments
  • Enrichments of anoxic organisms
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