Sizing up the Deep-sea Vent Subseafloor Biosphere

Production, Standing Stock, and Turnover

Stefan Sievert
Biology
Diffuse-Flow or Warm-Water Vent:
Mixing of hydrothermal fluids with seawater below the seafloor

Providing conditions conducive for chemoautotrophs growing as biofilms in cracks and crevices of ocean crust

Hentscher and Bach, 2012
**Ambient Deep-Sea Water:**
- 2°C
- 2.3 mM DIC
- 115 µM O₂
- 40 µM NO₃⁻
- 0 µM S²⁻, H₂
- 10⁴ cells/ml

**Crab Spa Diffuse-Flow Fluid:**
- 14:1 ADSW:HT
- 25°C
- 8.2 mM DIC
- < 3.6 µM O₂
- < 6 µM NO₃⁻
- ~12 µM NH₄⁺
- ~ 200 µM S²⁻
- < 2 µM H₂
- 2-5 *10⁵ cells/ml

**Hydrothermal Fluid:**
- >275°C
- 85 mM DIC
- 7.7 mM S²⁻
- 410 µM H₂
- 0 µM O₂
- 0 µM NO₃⁻
- 0 Cells

Based on McNichol et al., 2016, DSR-I; 2018, PNAS
Sievert – Fluid Underground Mtg, Nov 5, 2019
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Stewart et al., 2019, ISME J

Sievert – Fluid Underground Mtg, Nov 5, 2019
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Incubations under simulated *in situ* conditions using isobaric gas tight samplers (IGTs)

Based on McNichol *et al.*, 2016, DSR-I; 2018, PNAS

Sievert – Fluid Underground Mtg, Nov 5, 2019
Primary productivity below the seafloor at deep-sea hot springs

Jesse McNichol\textsuperscript{b,1,2}, Hryhoriy Stryhanyuk\textsuperscript{b}, Sean P. Sylva\textsuperscript{c}, François Thomas\textsuperscript{b,3}, Niculina Musat\textsuperscript{b}, Jeffrey S. Seewald\textsuperscript{c}, and Stefan M. Sievert\textsuperscript{a,1}

\textsuperscript{a}Biology Department, Woods Hole Oceanographic Institution, Woods Hole, MA 02543; \textsuperscript{b}Department of Isotope Biogeochemistry, Helmholtz Centre for Environmental Research – Umweltforschungszentrum (UFZ), 04318 Leipzig, Germany; \textsuperscript{3}Marine Chemistry and Geochemistry Department, Woods Hole Oceanographic Institution, Woods Hole, MA 02543

Edited by David M. Karl, University of Hawaii, Honolulu, HI, and approved May 16, 2018 (received for review March 13, 2018)

Sample Fluids \quad \rightarrow \quad \text{Return under pressure} \quad \rightarrow \quad \text{Add substrates} \quad \rightarrow \quad \text{Monitor growth}

\textsuperscript{13}C-CO\textsubscript{2}

SHIPBOARD INCUBATIONS WITH HYDROTHERMAL FLUIDS UNDER SIMULATED \textit{IN SITU} CONDITIONS

Sievert – Fluid Underground Mtg, Nov 5, 2019
Primary Productivity Inferred from Incubations at *In Situ* Temperature and Pressure Using by HISH-SIMS

<table>
<thead>
<tr>
<th>Treatment</th>
<th>µg C fixed · L⁻¹ · day⁻¹</th>
<th>CARD-FISH: 80-100% <em>Campylobacteria</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (n=3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>H₂ only (n=3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NO₃⁻ only (n=3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>O₂ (80µM) (n=2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>O₂ (110µM) (n=2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NO₃/H₂ (n=3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NO₃/H₂, 50°C (n=2)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**A**

- *Campylobacteria* dominate carbon fixation!
- Amendments increase carbon fixation

**McNichol et al., 2018, PNAS**

**Sievert – Fluid Underground Mtg, Nov 5, 2019**
Fraction of Electrons from Dissimilatory Reactions Used for Carbon Fixation: Chemosynthetic Growth Efficiency (CGE)

CGE = \frac{Eq_{CFIX}}{(Eq_{CFIX} + Eq_{DISS})}

McNichol et al., 2018, PNAS

Sievert – Fluid Underground Mtg, Nov 5, 2019
Seawater (SW): $O_2$, $NO_3^-$, $SO_4^{2-}$ (3°C)

Hydrothermal Fluid (HF): $H_2S$, $H_2$, $CH_4$ (>275°C)

14:1 mix (SW:HF)

Basalt 14:1 mix (SW:HF)

Subseafloor Biosphere

Exiting Fluids depleted in $H_2$, $H_2S$, $NO_3^-$, $O_2$ relative to conservative mixing; enriched in cells

Depletions in diffuse-flow fluids + CGE => *in situ* primary productivity!

Based on McNichol *et al.*, 2016, DSR-I; 2018, PNAS

Sievert – Fluid Underground Mtg, Nov 5, 2019
Determination of Rates of Various Chemosynthetic Reactions

Summary of rates of change in chemical concentrations and cell densities.

<table>
<thead>
<tr>
<th>Incubation condition</th>
<th>Control (24 °C)</th>
<th>H₂ addition (24 °C)</th>
<th>NO₃⁻ addition (24 °C)</th>
<th>O₂ (80 μM) (24 °C)</th>
<th>O₂ (110 μM) (24 °C)</th>
<th>NO₃⁻/H₂ (24 °C)</th>
<th>NO₃⁻/H₂ (50 °C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>H₂S consumption (fmol cell⁻¹ d⁻¹)</td>
<td>475.8</td>
<td>434.3</td>
<td>508.4</td>
<td>527.1</td>
<td>510.6</td>
<td>312.5</td>
<td></td>
</tr>
<tr>
<td>H₂S production (fmol cell⁻¹ d⁻¹)</td>
<td>797.1</td>
<td>681.6</td>
<td>506.7</td>
<td>483.2</td>
<td>494.5</td>
<td>523.3</td>
<td>458.1</td>
</tr>
<tr>
<td>H₂ consumption (fmol cell⁻¹ d⁻¹)</td>
<td>642.7</td>
<td>238.6</td>
<td>481.7</td>
<td>776.5</td>
<td>N.A.</td>
<td>N.A.</td>
<td></td>
</tr>
<tr>
<td>O₂ consumption (fmol cell⁻¹ d⁻¹)</td>
<td>N.A.</td>
<td>77.8</td>
<td>51.7</td>
<td>51.7</td>
<td>418.1</td>
<td>351.8</td>
<td>1535.2</td>
</tr>
<tr>
<td>NO₃⁻ consumption (fmol cell⁻¹ d⁻¹)</td>
<td>49.6</td>
<td>30.6</td>
<td>30.6</td>
<td>30.6</td>
<td>418.1</td>
<td>351.8</td>
<td>1535.2</td>
</tr>
<tr>
<td>NH₄⁺ production (fmol cell⁻¹ d⁻¹)</td>
<td>N.R.</td>
<td>N.R.</td>
<td>N.R.</td>
<td>N.R.</td>
<td>N.R.</td>
<td>N.R.</td>
<td>N.R.</td>
</tr>
<tr>
<td>% NO₃⁻ to DNRA at tₜend</td>
<td>57.5</td>
<td>65.0</td>
<td>65.0</td>
<td>65.0</td>
<td>193.0</td>
<td>654.7</td>
<td>187.7</td>
</tr>
<tr>
<td>Initial cell density (x 10⁵ cells mL⁻¹)</td>
<td>2.3</td>
<td>4.1</td>
<td>4.1</td>
<td>4.1</td>
<td>4.1</td>
<td>4.1</td>
<td>4.1</td>
</tr>
<tr>
<td>Maximum cell density (x 10⁵ cells mL⁻¹)</td>
<td>6.8</td>
<td>5.0</td>
<td>7.1</td>
<td>8.2</td>
<td>9.2</td>
<td>11.2</td>
<td>7.5</td>
</tr>
</tbody>
</table>

- Using in situ consumption and rates per cell allows calculation of standing stock
- Biomass residence time can be inferred from CGE-derived productivity and standing stock assuming steady state


Sievert – Fluid Underground Mtg, Nov 5, 2019
## Constraints on Subseafloor Productivity, Standing Stock, and Turnover from Measurements of CGE

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Lower bound</th>
<th>Upper bound</th>
<th>Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absolute carbon fixation rates$^1$</td>
<td>17.3</td>
<td>321.4</td>
<td>μg C ◦ L$^{-1}$ ◦ day$^{-1}$</td>
</tr>
<tr>
<td>Chemosynthetic growth efficiency$^1$</td>
<td>0.06</td>
<td>0.13</td>
<td>Fraction electron equivalents to Carbon fixation</td>
</tr>
<tr>
<td>Estimated in situ carbon fixation$^2$</td>
<td>104</td>
<td>253</td>
<td>μg C ◦ L$^{-1}$</td>
</tr>
<tr>
<td>(per L Crab Spa mixed fluid):</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(per L Crab Spa end-member fluid):</td>
<td>1.4×10$^3$</td>
<td>3.5×10$^3$</td>
<td></td>
</tr>
<tr>
<td>Estimated annual productivity$^3$ of:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Crab Spa vent$^4$</td>
<td>6.1×10$^3$</td>
<td>1.5×10$^4$</td>
<td>g C ◦ y$^{-1}$</td>
</tr>
<tr>
<td>Surrounding vent field$^5$</td>
<td>3.8×10$^6$</td>
<td>9.3×10$^6$</td>
<td></td>
</tr>
<tr>
<td>Global diffuse-flow vents$^6$</td>
<td>4.5×10$^{10}$</td>
<td>1.4×10$^{12}$</td>
<td></td>
</tr>
<tr>
<td>Standing stock$^7$, Crab Spa</td>
<td>28.6</td>
<td>NA</td>
<td>g C</td>
</tr>
<tr>
<td>Biomass residence time$^8$, Crab Spa</td>
<td>17</td>
<td>41</td>
<td>hours</td>
</tr>
<tr>
<td>Global standing stock$^6$</td>
<td>1.4×10$^9$</td>
<td>2.7×10$^9$</td>
<td>g C</td>
</tr>
</tbody>
</table>

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McNichol et al., 2019, PNAS

Sievert – Fluid Underground Mtg, Nov 5, 2019
Synthesis

**Ambient Deep-Sea Water:**
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- 0 µM S²⁻, H₂
- ~10⁴ cells/ml
- ~0% *Campylobacteria*

**Diffuse-Flow Fluid:**
- 25°C
- 8.2 mM DIC
- < 3.6 µM O₂
- < 6 µM NO₃⁻
- ~12 µM NH₄⁺
- ~ 200 µM S²⁻
- < 2 µM H₂
- ~5*10⁵ cells/ml
- ~80% *Campylobacteria*

**Subseafloor productivity rivals above seafloor production by symbiotic associations!**

**Hydrothermal Fluid:**
- >275°C
- 85 mM DIC
- 7.7 mM S²⁻
- 410 µM H₂
- 0 µM O₂
- 0 µM NO₃⁻
- 0 Cells

**Productivity:** 40 gC d⁻¹
**Standing stock:** 29 g C
**Biomass residence time:** 17-41 h

Based on McNichol *et al.*, 2016, DSR-I; 2018, PNAS

**Sievert – Fluid Underground Mtg, Nov 5, 2019**
Fluid geochemistry, local hydrology, and metabolic activity define methanogen community size and composition in deep-sea hydrothermal vents

- Fluid Residence Time: 29-33 h
- $10^{11}$ methanogens occupying as little as 2 m$^3$ pf ocean crust needed to create CH$_4$ anomalies
- Small, but very active subseafloor biosphere

Sievert – Fluid Underground Mtg, Nov 5, 2019
Outstanding Questions

Mode of growth in subseafloor?
*In situ* rates?
Flow path and plumbing?
Permeability of ocean crust?
Fluid mixing?
Fluid residence time?
Fluid volume?
Flow rate?

Sievert – Fluid Underground Meeting, Nov 5, 2019