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Supplementary material

**Vertical distribution of major sulfate-reducing bacteria in a eutrophic shallow meromictic lake**

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Table S1. Oligonucleotide probes used in this study.

<table>
<thead>
<tr>
<th>Probe</th>
<th>Specificity</th>
<th>Probe Sequence (5’ - 3’)</th>
<th>Position</th>
<th>FA (%)**</th>
<th>Probe concentration (fmol µl⁻¹)***</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>EUB338-I</td>
<td>Most Bacteria</td>
<td>GCTGCCTCCCGTAGGAGT</td>
<td>338 - 355</td>
<td>35</td>
<td>28</td>
<td>[1]</td>
</tr>
<tr>
<td>EUB338-II</td>
<td>Supplement to EUB338: order Planctomycetales</td>
<td>GCAGCACCACCGTAGGTTG</td>
<td>338 - 355</td>
<td>35</td>
<td>28</td>
<td>[2]</td>
</tr>
<tr>
<td>EUB338-III</td>
<td>Supplement to EUB338: order Verrucomicrobiales</td>
<td>GCTGCCACCACCGTAGGTTG</td>
<td>338 - 355</td>
<td>35</td>
<td>28</td>
<td>[2]</td>
</tr>
<tr>
<td>NON338</td>
<td>Control probe complementary to EUB338</td>
<td>ACTCCTACGGGAGGCAGC</td>
<td>338 - 355</td>
<td>35</td>
<td>28</td>
<td>[7]</td>
</tr>
<tr>
<td>Delta495a</td>
<td>Most Deltaproteobacteria</td>
<td>AGTTAGCCGCTGCTTCTCT</td>
<td>495 - 512</td>
<td>35</td>
<td>28</td>
<td>[3]</td>
</tr>
<tr>
<td>cDelta495a</td>
<td>Competitor for Delta495a</td>
<td>AGTTAGCCGCTGCTTCTCT</td>
<td>495 - 512</td>
<td>35</td>
<td>28</td>
<td>[4]</td>
</tr>
<tr>
<td>DSS658</td>
<td>Most Desulfobacteraceae</td>
<td>TCCACTTCCCTCCTCCCAT</td>
<td>658 - 675</td>
<td>50</td>
<td>28</td>
<td>[5]</td>
</tr>
<tr>
<td>SEEP1d-468*</td>
<td>Some SEEP SRB-1d bacteria</td>
<td>ACCGTAGTGCAAGATGG</td>
<td>468 - 485</td>
<td>40</td>
<td>60</td>
<td>This study</td>
</tr>
<tr>
<td>SEEP1d-1420*</td>
<td>SEEP-SRB1d group</td>
<td>CAA CTT CTG GTA CAG CCA</td>
<td>1420-1437</td>
<td>40</td>
<td>60</td>
<td>[6]</td>
</tr>
<tr>
<td>cSEEP1d-1420*</td>
<td>Competitor for SEEP1d-1420</td>
<td>CAA CTT CTG GTA CAA CCA</td>
<td>1420-1437</td>
<td>40</td>
<td>60</td>
<td>[6]</td>
</tr>
</tbody>
</table>

* Hybridization and washing temperatures were at 35°C.
** Formamide concentrations in the hybridization buffer (v/v).
*** Probe concentration in the hybridization buffer (v/v).
Supplementary figure captions

Fig. S1  Single cells of sulfate-reducing bacteria in Lake Harutori anoxic water column, visualized by CARD-FISH. A. DSS cells (probe DSS658, green). B. SEEP SRB-1d cells (probe SEEP1d-468, green). DAPI signals are in blue. Scale bars = 10 µm

Fig. S2  Partial alignment of the DSS658 probe targeting position and the sequences of Desulfosarcina-Desulfococcus specific clone library. Base position is according to Eschelichia coli. Identical bases are shown in dots. Sequences in gray-shaded boxes have no mismatch to the probe DSS658.

Fig. S3  A magnified phylogenetic tree based on deduced AprA amino acid sequences of sulfur-oxidizing bacteria retrieved from the water column of Lake Harutori. The tree was constructed based on a distance matrix (119 amino acid positions) by using the neighbor-joining method. Bootstrap value estimation was based on 1000 replicates (only values above 50% are shown). Numbers of sequences obtained in each clone library were indicated in parentheses; 3.0 m, 3.5 m, and 4.0 m in that order. SOP1 and 2 means the different lineages of AprA. The bar represents 5% estimated sequence divergence.

Fig. S4  Alignment of 263 deduced AprA amino acid sequences used in this study. The amino acid sequences were aligned using ClustalW implemented in MEGA5.05 software.
Fig. S1
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*Sequence HADCC-54 (Accession No. AB894641) in OTU1 has a single terminal mismatch to the probe DSS658.
**SOP1**

- Janssand intertidal sediment clone A24 (German Wadden Sea, ADW77087)
- Janssand intertidal sediment clone A102 (German Wadden Sea, ADW77077)
- Deep-sea hydrothermal vent of the Logatchev field clone aprE9 (CBH30954)
- OTU A11 (4-0-0)
  - "Candidatus Pelagibacter ubique" HTCC1002 (EAS84493)
  - Thiococcus pffennigi (ABV80070)
  - Thermochromatium tepidum (ABV80058)
  - Allochromatium wamingii (ABV80048)
  - Thiorhodovibrio winogradskyi (ABV80076)
  - Allochromatium minutissimum (ABV800107)
  - Lamprocystis roseopersicina (ABV80064)
  - Allochromatium vinosum (ADC62066)
  - Rhabdochromatium marinum (ABV80078)
  - Thiohalocapsa halophila (ABV80050)
  - Thiolamprovum pedioforme (ABV80068)
  - Sideroxydans lithotrophicus ES-1 (ADE11948)
- OTU A21 (1-0-0)
  - Freshwater Lake Okotanpe sediment clone Oko34 (BAN17852)
  - Freshwater Lake Maruwan O-Ike sediment clone MW0-2 3 (Antarctica BAN18021)
  - Filament of *Thioploca* sp. Lake Biwa clone aBiw47 (BAK52632)
  - Freshwater Lake Biwa sediment clone Biwa23 (BAN17815)
  - Freshwater Lake Okotanpe sediment clone Oko9 (BAN17854)
  - Metaliferous peat soils clone S6CL3 (ABJ90325)
  - Sponge symbiont (AFK76411)
- OTU A22 (1-0-0)
  - Freshwater Lake Oyako Ike sediment clone Oya78 (Antarctica, BAN17951)
  - *Sulfuricella denitrificans* (BAI66427)
  - Filament of *Thioploca* sp. in Lake Okotanpe clone aOko5 (BAK52553)
  - Freshwater Lake Maruwan O-Ike sediment clone MW2-4 13 (Antarctica, BAN18068)
  - OTU A10 (4-0-0)
    - Filament of *Thioploca* sp. in Lake Biwa clone aBiw12 (BAK52550)
  - OTU A40 (1-0-0)
  - OTU A39 (1-0-0)
  - *Thiobacillus plumbophilus* (ABV80021)
  - Terrestrial mud volcano in Azerbaijan clone BJ C1 (AFV48096)
  - Filament of *Thioploca* sp. in Lake Biwa clone aBiw20 (BAK52559)
  - Freshwater Lake Okoike like sediment clone Oya19 (Antarctica, BAN17962)
- OTU A42 (1-0-0)
  - *Thiobacillus thioparus* (ABV80027)
  - Filament of *Thioploca* sp. in Lake Biwa clone aBiw45 (BAK52555)
  - OTU A5 (4-4-0)
    - *Thiobacillus denitrificans* (ABV80035)
  - OTU A41 (1-0-0)
  - Freshwater Lake Okoike like sediment clone Oya16 (Antarctica, BAN17966)
  - OTU A23 (0-1-0)
  - Thiodictyon sp f4 (ABV80029)
  - Gammaproteobacterium symbiont of *Thyasira* sp. Eastern Mediterranean (CBX41648)
  - OTU A43 (1-0-0)
  - Terrestrial mud volcano in Azerbaijan clone B E1 (AFV48102)
- OTU A46 (1-0-0)
  - *Thiotrix* sp. CT3 (ABV80023)
  - *Sulfur-oxidizing symbiont of Oasiis* sp. (ABV80105)
  - *Symbiont of *Ifremeria nautilei* (ABV80045)
  - Bacteria endosymbiont of *Lucinoma* aff. kazani (CAJ85653)
  - *Thiobacillus aquaesulis* (ABV80019)
  - *Thiocapsa pendens* (ABV80015)
  - *Thiocytsa gelatnosa* (ABV80009)
  - *Thiorhodococcus minor* (ABV80013)
  - *Thiocystis violacea* (ABV80011)
  - *Thiocapsa roseopersicina* (ABV80001)
  - *Thiodictyon bacillosum* (ABV80017)
- OTU A45 (1-0-0)
  - Uncultured *Thiobacillus* sp. (ABH01152)
  - Meromictic Lake Pavin clone apsA40m41 (ADD84894)
  - OTU A44 (1-0-0)
  - *Thiobacillus* sp. (ABH01156)
  - District heating system biofilm clone DH R123 (ABV90490)
  - Mouse gastrointestinal tract clone APS2 (AAF16941)
  - *Sulfuritalea hydrogenivorans* (BAJ14737)

**SOP2**

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Fig. S3
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References


