THE GREAT DIVERSITY OF MICROORGANISMS PRESENT IN NIVAL LAKES ALLOWS THE COMMUNITY TO RAPIDLY ADAPT TO ENVIRONMENTAL CHANGES

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Abstract

Here we present evidence for

- the **enormous bacterial and archaeal diversity** present in cold water ecosystems,
- the presence of **8 new archaeal lineages** (79 to 95% similarity with known 16S rDNA sequences) and
- the presence of **15 new bacterial sequences** (88 to <97% similarity to known 16S rDNA sequences) which are closely related to organisms known to inhabit cold-extreme environments.
- And we show how the **microbial community composition changes** in response to fluctuations of physical (temperature and lake mixing) and nutrient ecosystem determinants.

Fig 1b. High productivity in an oligotrophic ecosystem
Lake bottom at 7m depth with Chironomidae tubes; the rusty brown color on the stone is an iron-oxide layer (picture H. Maag).

Introduction

We have chosen Lake Jöri XIII as our study site because it is characterized by a low nutrient input and active nutrient cycling. It has a well developed, short food-web from prokaryotes to insect larvae (Fig 1b). The small catchment is rich in iron minerals which determine nutrient cycling in the lake. Iron oxide precipitation scavenges phosphate and other dissolved nutrient molecules and retains them in the lake(Fig 1b).

High mountain lake habitats experience extreme and variable environmental conditions. Well-adapted microorganisms which are able to live in these habitats face freezing temperatures in winter to diurnal temperature fluctuations in the summer (Fig 2 and Fig 3), long periods of darkness while the lake is ice covered, strong UV radiation during ice melt in the height of summer, nutrient deprivation during stratification in winter and mesotrophic conditions in late fall (high soluble phosphate concentrations) (Fig. 4).

Results

Physical and chemical variability of Lake Jöri XIII

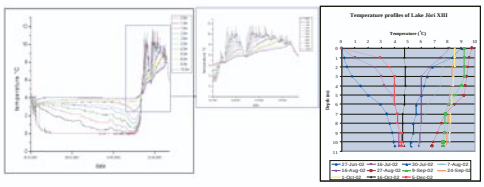


Fig 2. Seasonal temperature fluctuation at different depths of Lake Jöri XIII in 2001-2002 (left) and temperature profiles during ice-covered and ice-free periods in 2002 (right).

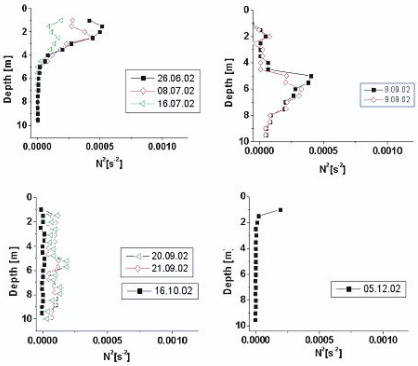
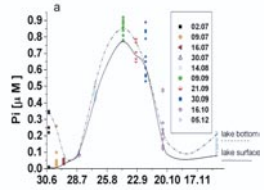


Fig 3. Stability of water masses of Jöri Lake XIII. The dates chosen are those around the sampling dates. An N^2 value of $0s^{-2}$ means an instability in this part of the water column.



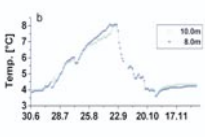


Fig 4. Inorganic soluble phosphate concentrations (left) and temperature fluctuation on the lake ground (10m, and 8m) in Lake Jöri XIII in 2002 (right).

Fig 1a. Lake Jöri XIII (arrow) June 27, 2002

Methods

➢Temperature Gradient Gel Electrophoresis (TGGE) of PCR-amplified 16S-rDNA was used to monitor the microbial populations and their seasonal dynamics at different depths in the water column (Fig 5).

➢Cloning of 16S-rRNA genes from total community DNA and sequencing them allowed us to genotypically identify the clones which revealed dominant TTGE bands (Fig 6).

➢Fluorescent In Situ Hybridization (FISH) and cloning of the 16S rDNA were used to confirm the community changes (Fig 7).

➢Cloning and sequencing analysis of 16S-rRNA genes of the sample from 0.8m above the lake bottom led to the discovery of novel archaeal lineages (Fig 8).

TTGE patterns of total community DNA

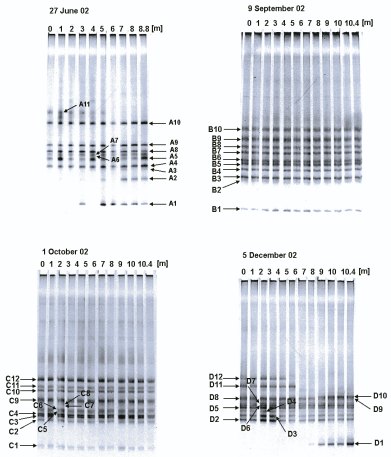


Fig 5. TGGE patterns of total community DNA of Lake Jöri XIII in 2002. Letters and arrows indicate excised bands for RFLP. Designations correspond to sequenced clones.

Who are they?

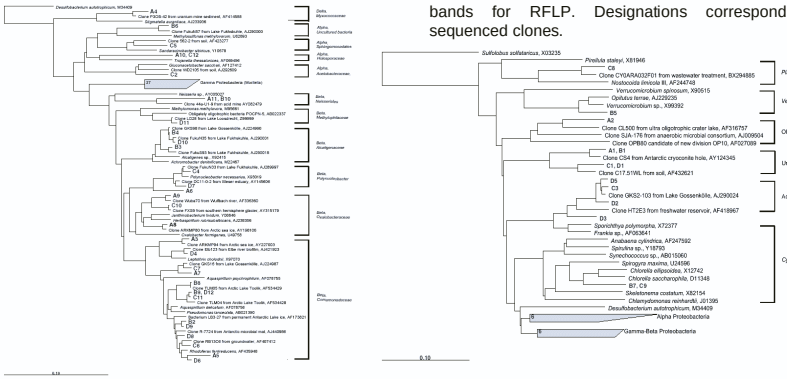
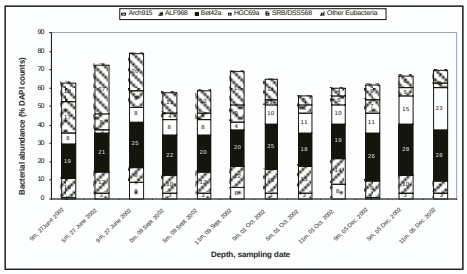


Fig 6a. Phylogenetic affiliation of predominant Proteobacteria. A sulfate reducing bacterium *Desulfobacterium autotrophicum*, M34409 is used as an outgroup (left).
Fig 6b. Phylogenetic affiliation of predominant communities of Proteobacteria. A thermophilic, sulfur oxidizing archaeum *Sulfolobus sulfataricus*, X03235 is used as an outgroup (right).

FISH analysis confirms the seasonal and spatial dynamics of the microbial community composition, including the novel archaeal lineages



DAPI-stained cells

CARD-FISH
with Arch915 probe

Percentage of hybridized cells in relation to total DAPI counts. Autofluorescent cells and NON EUB338 cells were substracted from the total number of fluorescent cells. 'Other bacteria' refers to those cells which hybridized with EUB338 but which cannot be counted as either ALF968, Bet42a, HGC69, or SRB/DSS568.

Fig 7. FISH analysis of prokaryotic communities (bacteria and archaea) in the water column (left). DAPI and Arch915-stained cells from water sample 0.8 m above the sediment (right).

Novel archaeal lineages from Lake Jöri XIII

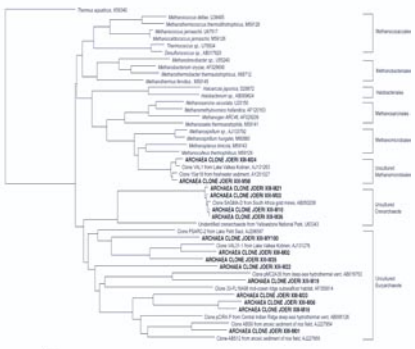


Fig 8. Phylogenetic affiliation of novel archaea from Lake Jöri XIII. A thermophilic bacterium, *Thermus aquaticus* is used as an outgroup.

Conclusions

- The large number of unique TGGE patterns indicate phylogenetically very diverse microbial populations in Lake Jöri XIII. They belong to the α, β, δ groups of the Proteobacteria, the Actinobacteria, the OP10, the Cyanobacteria (organelles), the Planctomycetes, the Verrucomicrobia and to a number of uncultured bacterial groups.
- The bacterial community composition changes rapidly in response to environmental fluctuations.
- We discovered novel Archaea lineages which belong to the uncultured Euryarchaeota, the Crenarchaeota, and the Methanomicrobiales.