**Chloroflexus-like bacteria are abundant in anoxic shallow terrestrial subsurface Holocene sediments from the catchment of Lake Kitaura**

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**Introduction**

16S rRNA gene clone library analysis used to characterize diversity and distribution of bacterial communities inhabiting iron- and sulfur-rich anoxic **Holocene sediments** from Lake Kitaura catchment revealed **Chloroflexus-like bacteria (CLB)** to be the dominant group in all three depths (2.1m, 3.6m and 6.1m) under investigation. Here we report the comparative phylogenetic analysis of this Chloroflexus-like bacterial community within the context of geological, geochemical and hydrological information in an attempt to infer in situ microbial ecology of these microorganisms.

**Materials and methods**

Community DNA was directly extracted from sediment core by using MOBIO Ultra Clean Soil kit (Mega Prep) with an additional wash protocol. 16S rDNA were PCR amplified using primers specific for the Domain Bacteria (primers 347F and 907R). PCR products were cloned and sequenced. The 16S rRNA gene sequences of approximately 550bp length were compiled and aligned using the automatic nucleic acid aligner CLUSTAL W software package (EMBL-EBI). Phylogenetic trees were created by using the software package MEGA 2.1 (1).

**Results and Discussion**

None of the sequences had >98% similarity with any cultured representatives. 44% of clones found at 2.1m depth, 54% at 3.6m depth and 48% at 6.1m depth branched deeply within Chloroflexus phylum. Majority of 2.1m depth sequences were closely affiliated to CLB found in habitats contaminated with chlorinated hydrocarbons suggesting heavy contamination by chlorinated aromatic agrochemicals in the region. These bacteria grouped within the most diverse and uncharacterized of the subdivisions (SD), Chloroflexus SD1. Most of the sequences from 3.6 and 6.1m depths were closely affiliated to environmental clones obtained from the deep subsurface Paleosol of DOE Hanford site (2) and grouped within both SD1 and SD2, which also has a cultured representative Dehalococcoides ethenogenes, capable of reductive dechlorination. Two 6.1m depth sequences made a monophylatic clade with SAR202 group of clones initially found in open ocean bacterioplankton (3) and were grouped within SD4. No representatives were grouped within SD3, the most characterized of SDs having majority of cultured thermophilic photoautotrophic members that include C. aurantiacus. Predominance of CLB through the core suggests robustness and the ability to adapt to changing hydrological, oxidation-reduction potential and accompanying changes. They may also be involved in iron- and sulfur-reduction processes.

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