



The microbial ecology in the brines of two Antarctic frozen lakes (Boulder Clay, Northern Victoria Land)

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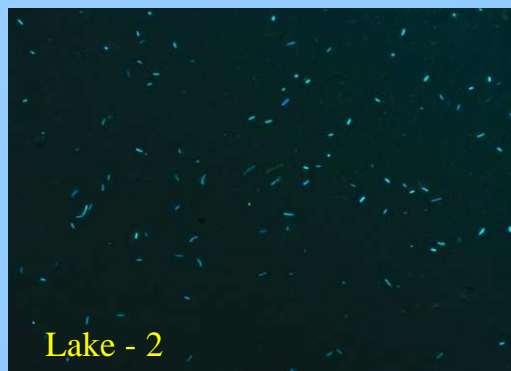
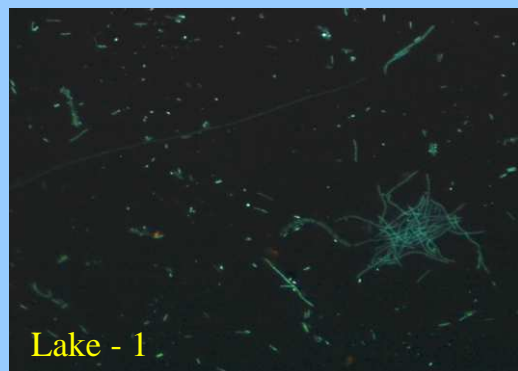


Research Background Perennial glacial lakes of Antarctica sometimes encapsulate lenses of brines, representing an extreme cryogenic ecosystem. The study of these unique environments allows insights on the geochemical processes affecting the microbial life in analogues terrestrial cryoecosystems and increases our perceptions on possible traces of life that must be sought beyond Earth.

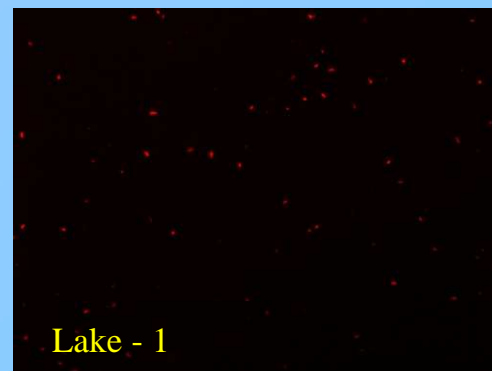
Objectives In order to find new extreme refugia for microbial life, in October-November 2014 several frozen lakes in the Northern Victoria Land were investigated through a ground probing radar (GPR). The microbial assemblage and metabolism of brines were investigated in two lenses of liquid brines found at different depths in two adjacent Boulder Clay lakes (Lake-1: 2.5 m; Lake-2: 2.0).

Methods Brines were aseptically sampled and analysed to assess: prokaryotic abundance and biomass, cell-shape and cell-volumes by microscopic image-analysis; respiring cells (CTC+); virus abundance by flow-cytometry. Moreover, the physiological profiles by Biolog-Ecoplate™ were determined as well as the potential ectoenzymatic activities rates (leucine-aminopeptidase, LAP; β -glucosidase, GLU; alkaline-phosphatase, AP).

Prokaryotic cells stained by DAPI observed by microscopic image-analysis

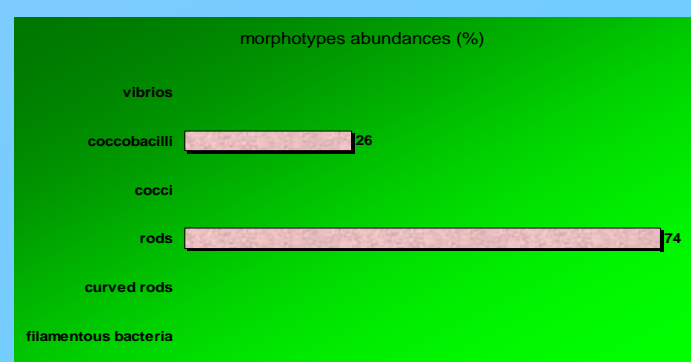
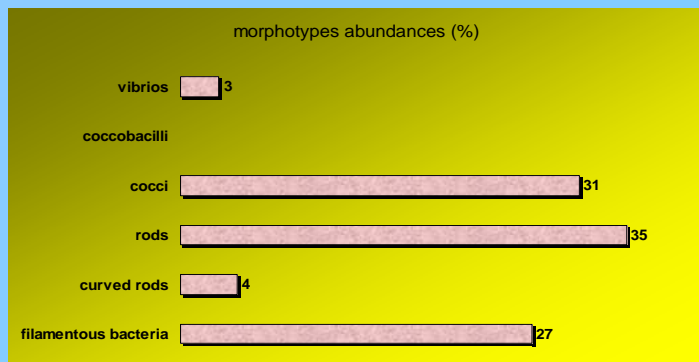
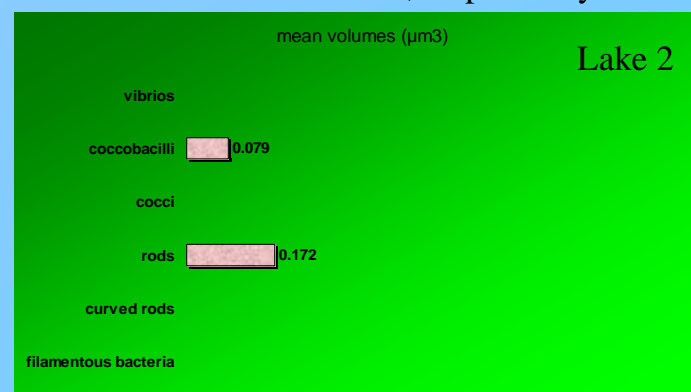
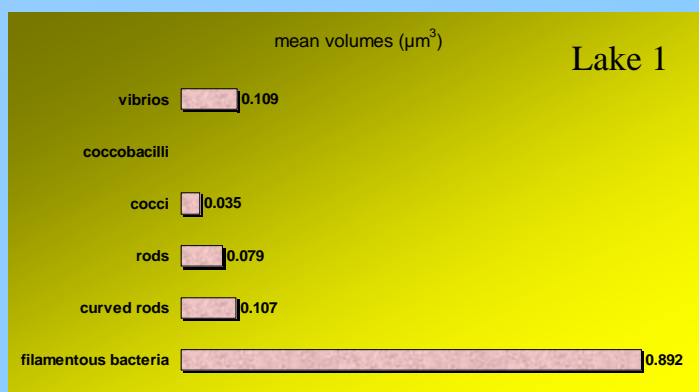


Prokaryotic cells staining with CTC+



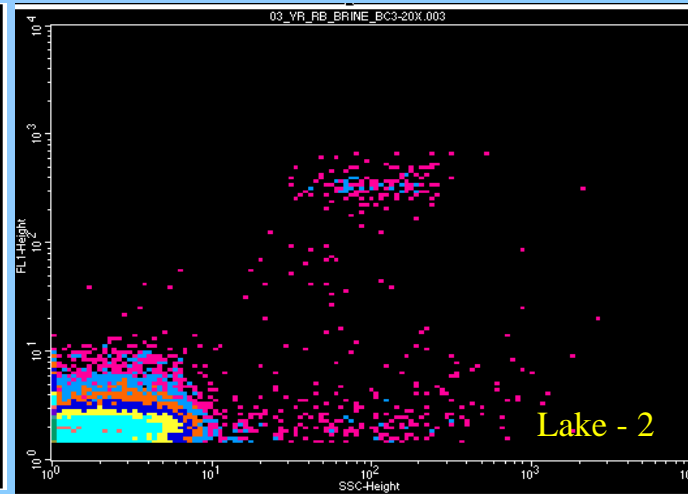
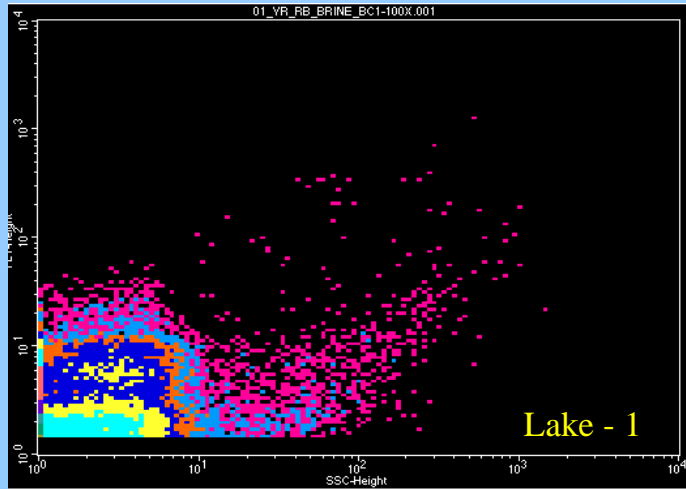
Prokaryotic abundances by microscopic image analysis were in the order of 10^8 cells L^{-1} in Lake-2 and 10^9 cells L^{-1} in Lake-1, respectively.

CTC+ ranged between 10^7 cell L^{-1} in Lake-2 and 10^8 cell L^{-1} in Lake-1. The highly respiring cells amounted to the 4 and 7 % of total cells in Lake-2 and Lake-1, respectively.



The **prokaryotic cell-volumes** were widely different between the two studied lakes, ranging - on average - between $0.242 \pm 0.456 \mu m^3$ (Lake-1) and 0.138 ± 0.177 (Lake-2). In Lake-1, cocci and rods were the most common morphotypes together with the filamentous forms, distributed in long or short chain rods. Few curved rods and vibrios were detected. Differently in Lake-2 two morphotypes only occurred: rods - that dominated the entire data set - and coccobacilli.

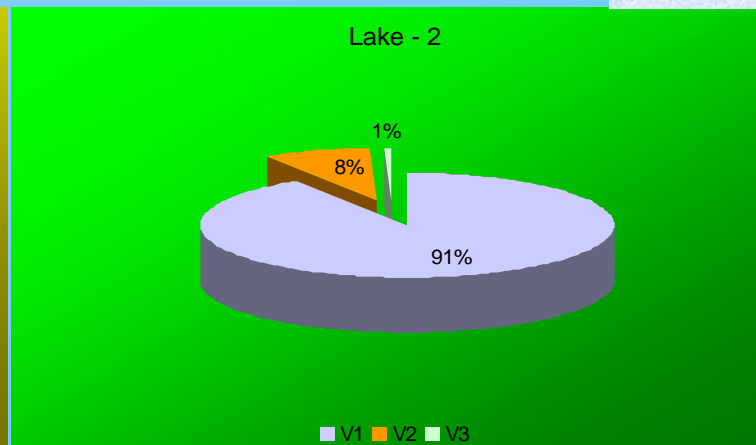
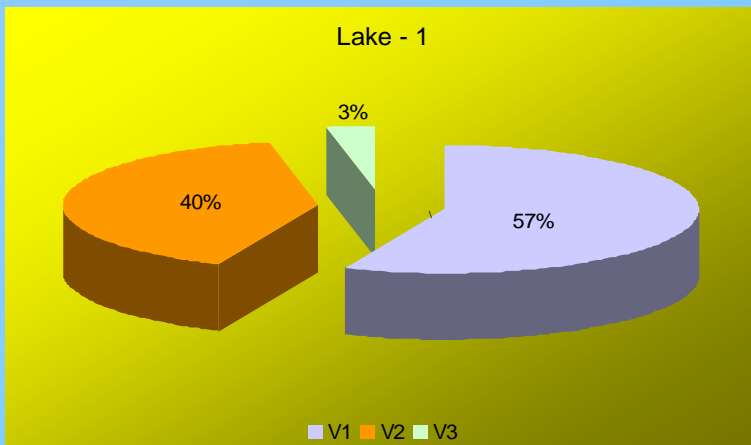
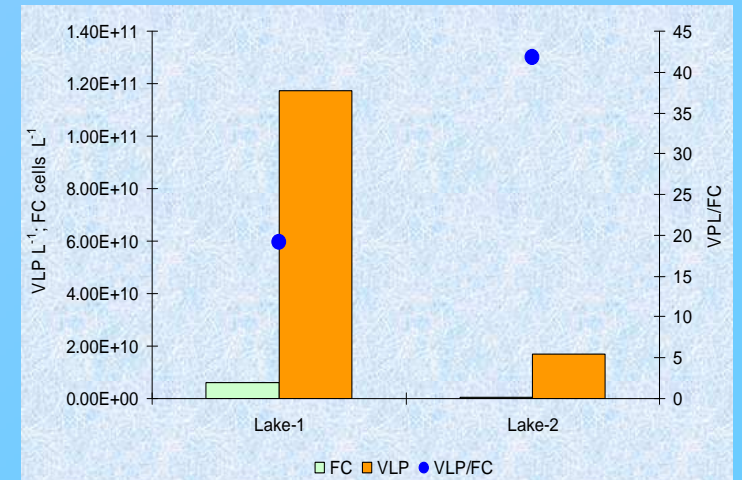
Flow cytometric cytograms of the VPL in the samples Lake-1 and Lake-2



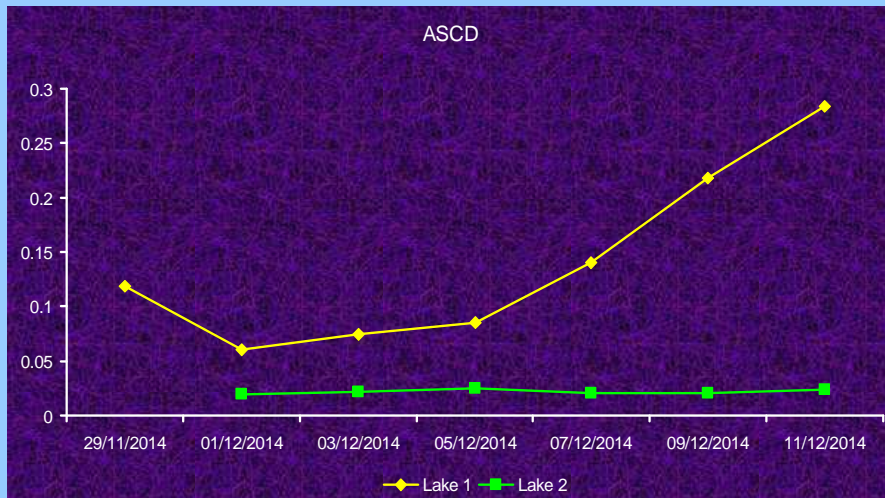
Prokaryotic abundances by flow cytometry (FC) were in the order of 10^8 cells L^{-1} in Lake-2 and 10^9 cells L^{-1} in Lake-1, respectively.

Total Virus like particles abundance (VLP) accounted for 10^{10} L^{-1} in Lake-2 and 10^{11} L^{-1} in Lake-1. Different percentage of VPL subpopulations were observed.

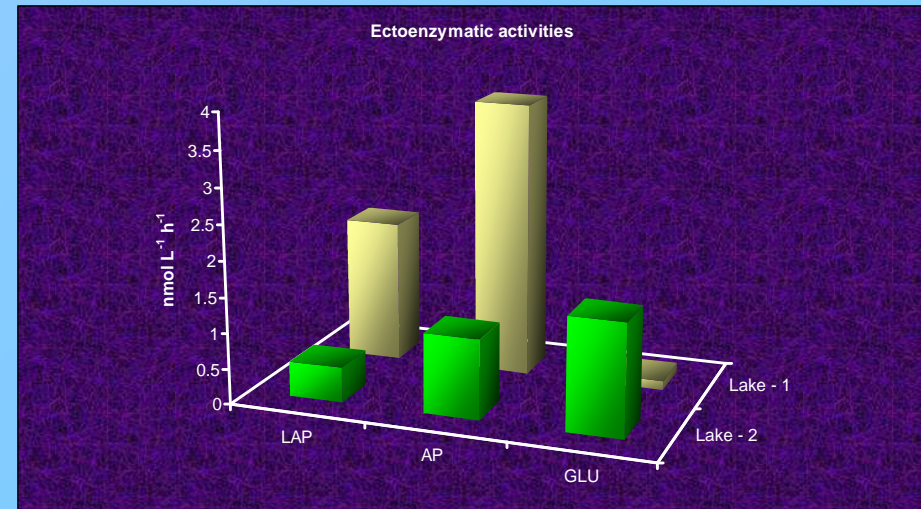
The VPL/FC ratio was significantly higher in Lake-2 than Lake-1.



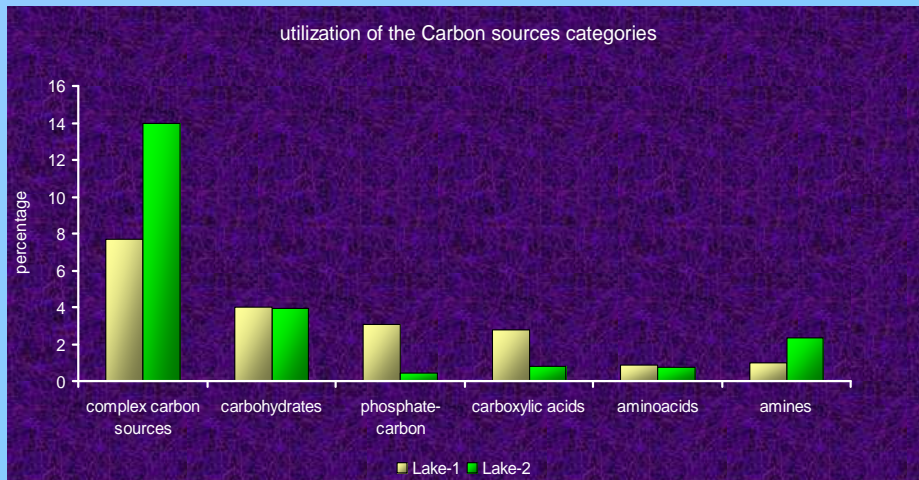
Kinetic curves of ASCD



Physiological profiles: Kinetic curves of averaged substrate colour development (ASCD) highlighted a different ability of the microbial communities to utilize the substrates: high and increasing with time in Lake-1 and very low and constant in Lake-2.



Differences in the **potential ectoenzymatic activities rates** were observed in the two lakes. In the brine of Lake-1, higher values of leucine-aminopeptidase and alkaline-phosphatase than in Lake-2 were detected, suggesting the presence of cells more able to decompose proteins and organic phosphates; β -glucosidase activity on polysaccharides was negligible in Lake-1 and high in Lake-2.



Physiological profiles: Lake-2 was characterised by higher utilisation of polymers and amines and lower of phosphate-carbon and carboxylic acid than Lake-1. Carbohydrates and amino acids were similarly utilized in both lakes.

REMARKS

In spite of their spatial proximity, the two studied brines of Boulder Clay lakes appear to be microbiologically diverse.

This first screening highlights different characteristics in terms of both abundance and metabolism of the prokaryotic assemblages.

In particular:

Lake-1 harboured microbial populations characterized by:

- ✓ **higher prokaryotic abundances, volumes, biomass and virus like particles;**
- ✓ **higher numbers of respiring cells, of rates of ectoenzymatic activities and of carbon sources utilization**

than Lake-2. To understand which factors are responsible of the microbial differences in the two brines, climatic and environmental forces should be related to our findings.