



Deliberations on microbial life in the subglacial Lake Vostok, East Antarctica

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The objective was to estimate microbial contents of accretion (lake originating) ice from the Lake Vostok buried beneath 4-km thick East Antarctic ice sheet with the ultimate goal to discover microbial life in this extreme icy environment featured by no light, close to freezing point temperature, ultra-low DOC contents, and an excess of oxygen. The PCR based bacterial and archaeal 16S ribosomal RNA gene sequencing constrained by Forensic Biology and Ancient DNA research criteria was used as a main approach. Epifluorescent and confocal microscopies as well as flow cytometry were implemented.

DNA study showed that the accretion ice is essentially bacteria- and archaea-free. Up to now, the only accretion ice type 1 featured by mica-clay sediments presence and namely one horizon of four studied (3607m) allowed the recovery a few bacterial phylotypes. This unexpectedly included the chemolithoautotrophic thermophile *Hydrogenophilus thermoluteolus* and two more unclassified phylotypes all passing numerous contaminant controls (1). In contrast, the deeper and cleaner accretion ice 2 (three cores) with no sediments presence and near detection limit gas contents gave no reliable signals.

The microbes detected in accretion ice 1 are unbelievable to resist an excess of oxygen in the lake water body (700 - 1300 mg O₂/l). They are supposed to be thriving in rather warm anoxic sediments in deep faults at the lake bottom and sporadically flushing out along with sediments to the lake veins in a shallow depth bay due to a seismotectonic activity likely operating in the lake environment. A few geophysical

and geological evidences support this scenario. In the bay the presence of mica-clay sediments, higher accretion rate due to relief rise and likely oxygen-depleted upper layer of water can provide microbes with a chance to escape the high oxygen tension by the rapid entrapment into accretion ice 1. Sediment-free accretion ice 2, which forms above a deeper part of the lake, shows no evidence for reasonable source for microbe contribution given highly oxygenated lake water environment.

Microscopy and flow cytometry trials on strictly decontaminated ice samples gave supporting results. While microscopy failed to reveal cells because the local concentrations were below the detection limit, the flow cytometry succeeded in a preliminary estimate of 9 and 24 cells/ml for accretion 1 (3561m) and control glacial (2054m) ice samples, respectively. However, given the ratio contaminants to indigenous cells is about 10:1 (from PCR results), the genuine microbial contents for both accretion and glacial ice samples is expected to be as low as 1 cell/ml what practically means "sterile" conditions.

Thus, the accretion ice from Lake Vostok contains the very low unevenly distributed biomass indicating that the water body (at least upper layer) should also be hosting a highly sparse life, if any. By this, the Lake Vostok for the first time could present the big natural "sterile" water body on Earth providing a unique test area for searching for life on icy moons and planets. The search for life in Lake Vostok is constrained by a high chance of forward-contamination which can be minimized by using of stringent decontamination procedures and comprehensive biological controls.

(1) Bulat, S.A., Alekhina, I.A., Blot, M., Petit, J.-R., de Angelis, M., Wagenbach, D., Lipenkov, V.Ya., Vasilyeva, L.P., Wloch, D.M., Raynaud, D. and V.V. Lukin (2004) DNA signature of thermophilic bacteria from the aged accretion ice of Lake Vostok, Antarctica: implications for searching for life in extreme icy environments. *Int J Astrobiology* 3(1):1-12