



## **Life detection strategy for Jovian's icy moons: Lessons from subglacial Lake Vostok exploration**

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The objective was to estimate the microbial content of accretion ice originating from the subglacial Lake Vostok buried beneath 4-km thick East Antarctic ice sheet with the ultimate goal to discover microbial life in this extreme icy environment. The DNA study constrained by Ancient DNA research criteria was used as a main approach. The flow cytometry was implemented in cell enumerating. As a result, both approaches showed that the accretion ice contains the very low unevenly distributed biomass indicating that the water body should also be hosting a highly sparse life. Up to now, the only accretion ice featured by mica-clay sediments presence allowed the recovery a pair of bacterial phylotypes. This unexpectedly included the chemolithoautotrophic thermophile *Hydrogenophilus thermoluteolus* and one more unclassified phylotype both passing numerous contaminant controls. In contrast, the deeper and cleaner accretion ice with no sediments presence and near detection limit gas content gave no reliable signals. Thus, the results obtained testify that the search for life in the Lake Vostok is constrained by a high chance of forward-contamination. The subglacial Lake Vostok seems to represent the only extremely clean giant aquatic system on the Earth providing a unique test area for searching for life on icy worlds. The life detection strategy for (sub)glacial environments elsewhere (e.g., Jovian's Europa) should be based on stringent decontamination procedures in clean-room facilities, establishment of on-site contaminant library, implementation of appropriate methods to reach detection level for signal as low as possible, verification of findings through ecological settings of a given environment and repetition at an independent laboratory within the specialized laboratory network.