

Structural and Functional Insights from Microbial Communities of the Soudan Iron Mine

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Although deep subsurface environments are isolated from Earth's surface processes, diverse and active microbial communities thrived in these unique environments. Embedded deep within northern Minnesota's expansive Iron Range, the Soudan Iron Mine transects massive veins of hematite and Archaean (2.7 Gy) banded iron formations, reaching a depth of 713 m (2,341 ft) below the surface. On its lowest level, abandoned exploratory boreholes act as low-flow conduits for anoxic, calcium chloride brines with ionic strengths up to three times saltier than seawater, low oxidation-reduction potentials, circumneutral pH, and low concentrations of organic electron donors. Here, we describe microbial communities living in harsh conditions such as low temperatures, high brines, and limited energy sources. Amplicon sequencing shows these borehole communities are low in diversity and dominated by Firmicute and Proteobacteria phyla. Despite carbon limitation, anoxic brines contain reduced metals and continuously evolve methane gas which is driven by methanogenesis such as *Methanobus* and suggests cycling of methylated compounds. Furthermore, the high concentration of sulfate in brines suggests that sulfur cycling may occur through both possible sulfate-reducing and thiosulfate-oxidizing microorganisms detected. In summary, metagenomic sequence data provided insight into the structure and possible function of microbial communities in these deeply isolated systems.