The silicification of extremophilic biofilms: Abiotic vs. biotic

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The identification of microbes in the fossil record is problematic due to the poor preservation potential of cellular material. Experimental studies of microbial fabric preservation during silicification exist, but most focus upon sheathed cyanobacteria. Few studies have addressed the preservation of microbes created by non-photosynthetic extremophiles. In this study, a terrestrial hot spring is simulated where silicification of a non-photosynthetic biofilm occurs.

The simulated hot spring consists of spring fluid (distilled water or Wairakei separated geothermal water), a HPLC pump, an oven to depolymerise silica, an incubator containing a tray with experimental substrates (standard microscope slides), an exit pump, and a nutrient feed pump.

At pH 7 and 60^{o}C, a biofilm did not form when distilled water was passed through the tray. However, a continuous biofilm formed over the surface of the sample tray after addition of bacterial nutrient (TSB). Sub-culturing suggests that this is a pure culture. 16S rRNA gene sequencing shows 92% similarity to *Geobacillus stearothermophilius*, possibly representing a new genus within the family *Bacillaceae*. In a second experiment, using Wairakei water (570 mg/kg SiO_2), the biofilm became heavily encrusted in silica forming a hard sheet over the tray. Silica was entirely *extracellular* and replaced the EPS between cells. These results, in addition to field studies, suggest that the growth of silica ledges is the result of competition between biofilm growth and abiotic silica precipitation at the air-water interface.

At pH 4 and 60^{o}C, a continuous biofilm also formed when nutrient was added. Upon switching to Wairakei water, the biofilm remained pliable and silica mineralisation was almost entirely *intracellular*. The dominant species in this biofilm also contained inclusion bodies that remained resistant to silicification. The composition of these bodies is not known but has implications for the preservation of molecular biomarkers. 16S rRNA gene sequencing is underway to identify this species.

Further experiments have been undertaken using the thermophile *Anoyxbacillus flavithermus* to search for biochemical recognition of silica. The total cellular proteins of *Anoxybacillus flavithermus* were analysed before and after silica addition, using one-dimensional SDS-PAGE, however, results were inconclusive.