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An integrated geochemical approach to microbial biosignatures from a subterranean deep biosphere

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To track organic or inorganic biosignatures of an ancient deep biosphere, an analytical approach combining complementary techniques of high lateral resolution was adopted. This approach included polarization microscopy, time-of-flight secondary ion mass spectrometry (ToF-SIMS), confocal Raman microscopy, and laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS). This instrumental combination enabled a reconstruction and integrated understanding of ancient environmental processes that occurred within the continental deep biosphere, while leaving the integrity of the studied structures virtually intact. Investigations were performed on a minute amount of sample material, namely a 5 mm thick section (2x4 cm^2) exhibiting fracture fillings in a 1.8 Ga old diorite, obtained at -450 m depth in the Äspö Hard Rock Laboratory.

The fracture mineral succession consists of fluorite and calcite phases and a thin (20-100μm), amorphous layer, lining the boundary between the fluorite and the calcite phases. Distinct accumulations of the amorphous material within cavities of the fluorite point to biocorrosion. Analysis of the amorphous layer showed significant accumulations of Si, Al, Mg, Fe, and light rare earth elements (LREE). In the same area, ToF-SIMS imaging revealed numerous, partly functionalized organic ions, e.g. $C_2H_6N^+$, $C_4H_7^+$, $C_3H_5O^+$, and $C_6H_{11}^+$. The presence of such functionalized organic compounds within the amorphous layer was corroborated by extended Raman imaging showing bands characteristic for C-C, C-N, and C-O bonds. According to its organic nature and the abundance of relatively unstable N- and O- heterocompounds, the organic-rich amorphous layer was interpreted to represent the remains of a biofilm that established much later than the initial cooling of the Precambrian host rock. Indeed, δ^{13}C, δ^{18}O and ^{87}Sr/^{86}Sr isotope analyses of the fracture minerals and the host rock pointed to an association of the fossil biofilm with a Pleistocene fracture reactivation event, i.e., in the most recent geological past.