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BACTERIAL BIOMARKERS IN CAVE CALCITE

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In caves, most chemically precipitated calcite deposits (speleothems) are formed inorganically by the degassing of supersaturated ground-waters. However, a number of forms have been associated with bacterial activity, in particular moonmilk, a porous, plastic calcite deposit commonly composed of elongated and often fibrous crystals. There is currently considerable debate over whether microbes do play a role in moonmilk formation, partly due to the limitations of the techniques used to investigate the question. Here we present the first lipid data from moonmilk deposits, and demonstrate that the samples analysed have a biomarker composition distinctly different from that seen in inorganic cave deposits, with the differences in the lipid signal being predominantly related to bacterially-derived lipid groups. This provides the first biomarker evidence for microbial involvement in cave calcite deposition.

Moonmilk samples were collected from the Italian Alps, and south-eastern Australia, and processed and analysed in line with the protocols described in Blyth et al. (2006). The lipid signals obtained were then compared via Principal Components Analysis to lipid profiles from a range of stalagmites collected from Scotland, Italy, Ethiopia and Australia. Stalagmites are primarily precipitated by inorganic means, but have been shown to contain a bacterial lipid signal from soil and cave microbes (ibid.). However, we hypothesise that the lipid signal recovered from calcite precipitated in close association with bacterial communities should be identifiably distinct in compound abundance and distribution.

The moonmilk from the Italian Alps shows a substantially higher total lipid abundance than is seen in stalagmites (an average of 58μg/g calcite as opposed to 4.8μg/g), and this is primarily driven by significantly higher levels of branched and normal fatty acids with carbon chain lengths of C_{15} and below, and *n*-alkanols with carbon chains of C_{16} and below (which collectively form an average of 63% of the lipid abundance, as opposed to an average of 19% in the stalagmites). These compounds group strongly together in a PCA, and are believed to be of microbial origin. Figure 1 shows the differences in fatty acid distribution between the moonmilk sample and a stalagmite from the same area.

In contrast, the moonmilk from Australia does not show elevated lipid abundances or increased levels of low molecular weight acids and *n*-alkanols. However, it does display a proportionally higher abundance of 3-hydroxy acids, which form 25% of the total lipid signal (as opposed to an average of 4% in the stalagmites), and are associated with the presence of gram negative bacteria.

Together these results demonstrate that there is a strong connection between the studied moonmilk deposits and bacterial communities, of a kind that is not explained by the simple passive presence of cave biofilms. However, it has also been shown that moonmilk samples from different environments do not necessarily show a common lipid signal, indicating that there is not a single mechanism governing the deposition of this speleothem type; as well as separate inorganic and biogenic forms, different deposits may be associated with a variety of different bacteria. This reinforces the need for a multidisciplinary approach to be taken, combining the information offered by textural, chemical and biological analyses.

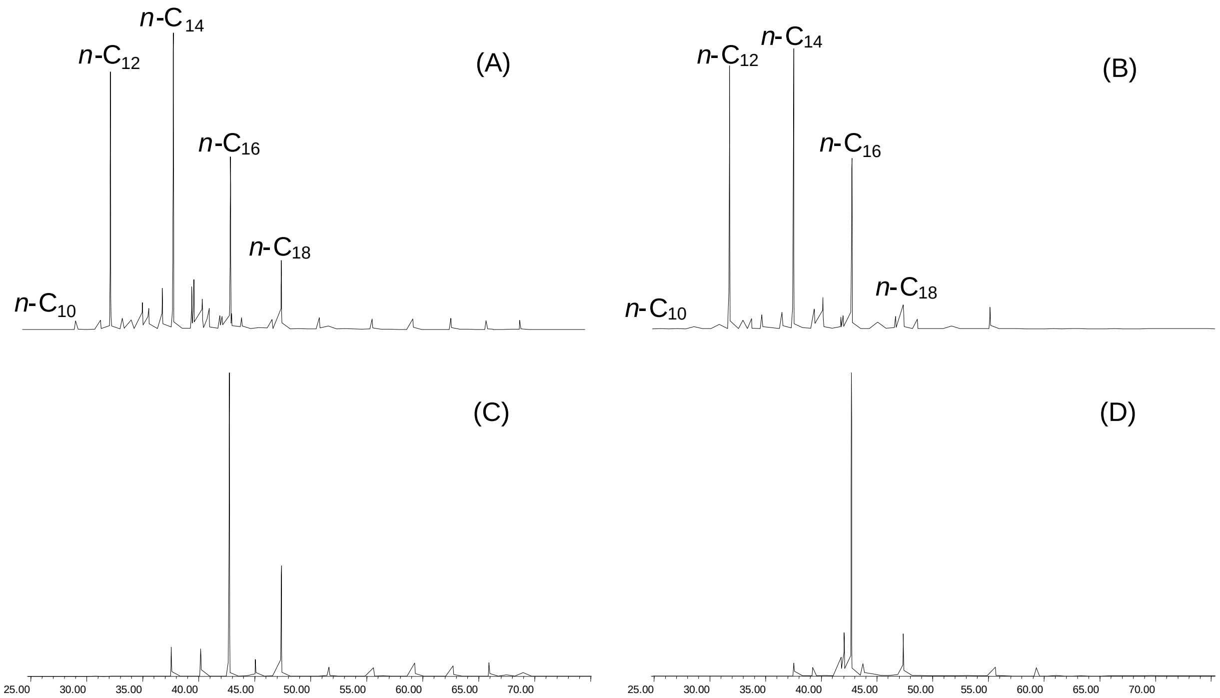

Figure 1. Fatty acids distributions (m/z 74) in A and B) moonmilk from the Italian Alps; C) a stalagmite from the Italian Alps; and D) moonmilk from Australia.

REFERENCES

Blyth, A.J., Farrimond, P., Jones, D.M. 2006. An optimised method for the extraction and analysis of lipid biomarkers from stalagmites. Organic Geochemistry 37, 882-890.