Identifying chemosymbiosis through geological time by stable isotope analysis of shell organics

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Biomineralizing organisms use organic templates during shell formation, and this shell-bound organic matter (SBOM) records the isotopic composition of an animal's diet [1]. We have obtained SBOM with a novel method using ion-exchange resin, and aim to investigate the fossil occurrence of chemosymbiosis. This is an unusual nutritional strategy whereby invertebrate animals obtain their nutrition from symbiotic bacteria, that oxidize sulphur (thiotrophy), methane (methanotrophy) or both (dual symbiosis). Chemosymbiosis allows bivalves to thrive at inhospitable ecosystems of the deep sea (cold seeps and hydrothermal vents), but whether or not ancient seep dwellers were chemosymbiotic is unknown.

In modern bivalve and brachiopod taxa nutritional strategies can be distinguished using $\delta^{15}N$, $\delta^{34}S$ and $\delta^{13}C$ SBOM values. $\delta^{34}S$ values of thiotrophic bivalves (-12.3 to -2.5‰, n=7) are distinct from heterotrophic species (-3.0 to 11.9‰, n=28), due to their use of depleted sulfide. SBOM $\delta^{13}C$ values distinguish methanotrophic/dual symbiosis (-64.4 to -33.3‰, n=16), because ^{13}C depleted methane is used as a carbon source (other strategies: -37.0 to -16.6‰, n=88). SBOM $\delta^{15}N$ is lighter for all chemosymbiotic taxa (0.9 to 4.3‰, n=12), than for other strategies (5.6 to 14.6‰, n=36).

SBOM has successfully been isolated from fossil cold seep specimens (Cretaceous to Pleistocene), and preliminary results allow positive identification of thiotrophy in fossil seep clams ($\delta^{15}N$ and $\delta^{34}S$ values) and seep mussels ($\delta^{13}C$). These results hold the promise of unravelling the previously unknown evolutionary history of chemosymbiosis.

[1] Dreier et al., (2012) *FEMS Microbiol Ecol*, **81**: p. 480-493.