Clues on methane's biogeochemistry and associated heterotrophic microbes in marine sediments from AOM enrichment cultures

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Enrichment cultures of consortia of anaerobic methaneoxidizing archaea (ANME) and sulfate-reducing bacteria (SRB) help to investigate the physiological potential and ecological role of these microbes in the marine environment. Their activity during the anaerobic oxidation of methane (AOM) shapes the stable carbon isotopic composition of methane and affects the abundance of associated microbial community members. The former has been shown to be enigmatic because AOM often depletes methane in its heavy isotopes rather than enriching it. Using a thermophilic ANME-1 enrichment culture from the Guaymas Basin, we shed light on this effect and its physiological underpinnings [1]. Low sulfate concentration was identified as the driving factor of residual methane being substantially depleted in ¹³C. In contrast, high sulfate concentration lead to a ¹³C-enrichment, as classically expected. Based on a metabolicisotopic model, we could explain the sulfate dependence through the thermodynamic drive along the line of the intracellular reactions. The laboratory enrichments of investigation are, however, not devoid of microbes unrelated to AOM. To understand their metabolism and dependence on the AOM consortia, we used a lipid stable isotope probing approach and amended the enrichment cultures with L-leucine-3-13C, hence tracking heterotrophic potentials [2]. The ¹³C-label was predominantly transferred into bacterial fatty acids, e.g. methylbranched $C_{15:0}$ and $C_{17:0}$ as well as unsaturated $C_{18:1\omega9}$ and C18:107. Such fatty acids are frequently detected in natural methane-rich environments and often show carbon istope values being more negative than fatty acids from AOM consortia members. This strongly suggest production by ancillary bacteria that grow on ¹³C-depleted necromass or cell exudates/lysates of the AOM core communities. Candidates that likely benefit are Spirochaetes and Anaerolineae, known to produce abundant branched fatty acids and present in all AOM enrichment cultures. These latest advances using enrichment cultures indicate that environmental factors shape methane's biogeochemistry and that degradation of AOM biomass feeds additional microbial community members in ocean-floor ecosystems.

[1] Wegener, Gropp, Taubner, Halevy & Elvert (2021),

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[2] Zhu, Wegener, Hinrichs & Elvert (2022), Frontiers in Microbiology 13, 1–12.