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Isotopic Fingerprinting of Methane and CO_2 Formation from Aliphatic and Aromatic Hydrocarbons

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We investigated the stable carbon and hydrogen isotopic signature of methane, CO_2 and water during microbial formation of methane from mineral oil related compounds in order to determine the variability of methane carbon and hydrogen isotopic signatures. The isotopic discrimination for carbon and hydrogen between substrate and methane was calculated and resulted in $\epsilon C_{DIC-CH4}$ 26 - 60‰, $\epsilon C_{substrate-CH4}$ 16 - 33‰, $\epsilon H_{H2O-CH4}$ 257 - 336‰, and $\epsilon H_{substrate-CH4}$ 174 - 318‰, respectively. These carbon and hydrogen isotope signatures fell in a relatively narrow range, suggesting a coupling of fermentation with acetoclastic and CO_2 reducing methanogenesis. In order to characterize the microbial consortia involved in the methanogenic degradation of hexadecane, a methanogenic enrichment culture was incubated with 1^{-13}C-hexadecane and its biomass was analyzed for the pattern and isotopic signature of carboxylic acids. The highest labelling was detected in n-C^{17} fatty acid with methyl groups at carbon atom 4, presumably indicative of Syntrophus sp. To determine if the isotope composition of methane can be used as an indicator for methanogenesis during growth with oil related compounds in field studies, we analyzed the isotope composition of methane in a confined mineral oil contaminated aquifer. The variability of carbon and hydrogen isotope composition was almost identical to the values obtained from enrichment cultures, thereby providing a tool for screening for microbial methane formation during hydrocarbon exploration.