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Crystalline Iron Oxides Stimulate Methanogenesis Under Sulfate Reducing Conditions in the Terrestrial Subsurface

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Microbial methane production is intimately linked to the biogeochemical cycling of iron, sulfur and carbon in sedimentary environments. Sulfate reducing microbes often outcompete methanogens for shared substrates. However, in a prior study we observed the coincidence of sulfate reduction and methanogenesis at our field site, the Oak Ridge Reservation Field Research Center in Bear Creek, TN, in depth horizons (100-150 cm). Interestingly, iron (Fe) oxides of varying crystallinities were also detected in the same samples. Fe oxides are known to act as electron conduits for direct interspecies electron transport (DIET) between syntrophic partners and can connect the metabolisms of methanogens with syntrophic Fe reducing microbes in nature. However, whether the nature of Fe oxides can influence(?)/mediate electron transfer reactions between sulfate reducing microbes and methanogens is less understood. In this study, we hypothesized that more crystalline Fe oxides facilitate the co-existence of sulfate reducers and methanogens. Incubations with microbial communities enriched from these subsurface sediments stimulated methane production in cultures amended with crystalline hematite but not in those amended with the amorphous, short range-ordered ferrihydrite. Furthermore, Synchrotron-based X-ray absorption spectroscopy data Fe reduction occurred only in incubations amended with short-range ordered ferrihydrite indicating how poorly crystalline Fe-oxides potentially contribute to the dynamic redox nature of subsurface sediments. 16S rRNA gene amplicon analysis of microbial communities enriched at different time points during these incubations revealed several taxa commonly associated with iron and sulfate reduction, as well as fermentation and methanogenesis, aligning with our geochemical data. Overall, the results from this work deepen our understanding of the role of Fe oxides in extracellular electron transfer and in mediating anaerobic metabolisms in the terrestrial subsurface environment.