

Importance of autotrophy in continental wetlands as revealed by coupled metagenomic and lipid biomarker approaches

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Vegetated continental wetlands are large sinks of carbon dioxide but are also responsible for 20% of the world methane emissions. They encompass diverse ecosystems, from ponds to peatlands. In mid-latitudes, temporary flooded lowlands are very common, due to the seasonal discharge of water from rivers or groundwater but their contribution to the global carbon budget is unknown. These sites experience temporal successions of oxic and anoxic conditions and the impact of these dynamic redox conditions on the microbial communities is poorly understood. The present study was carried out at the Ploemeur-Guidel hydrogeological observatory, in France, where a wetland is created by the seasonal discharge of deep anoxic, iron- and sulfate-rich groundwater. Four one-meter-long cores were recovered along a transect from the inner to the outer side of the wetland. To shed light on the microbial processes at this site, a metagenomic and lipid biomarker analysis was conducted in combination with the analysis of the carbon isotopic composition of the major microbial intact polar lipids. The metagenomic study showed clear taxon stratification with depth. Metagenomic reads at depth were dominated by the archaeal phylum *Candidatus* Bathyarchaeota. *Ca.* Bathyarchaeota is a generalist recognized as a major player of the carbon cycle. It seems to possess genetic capabilities to degrade a wide range of organic compounds but also to grow autotrophically and to perform methanogenesis. A multivariate statistical analysis of the genomic and lipid data sets enabled to identify putative lipid biomarkers for *Ca.* Bathyarchaeota (2G-GDGTs and 1G-BDGTs with up to two rings). Carbon isotopic composition of the biphytanes (BP) derived from these membrane lipids displayed a systematic strong depletion in ¹³C, which indicates a predominantly autotrophic or methanogenic metabolism. Yet, no marker genes for methanogenesis could be detected in the reconstructed metagenome-assembled-genomes. This study evidences, for the first time, the autotrophic metabolism of *Ca.* Bathyarchaeota in anoxic soils and highlights its prominent role in the carbon cycle of wetlands. It also illustrates how detailed present-day biogeochemical studies can help interpret biomarker isotopic records from the past.