**TITLE:** Molecular and computational strategies for recovering metagenome-assembled genomes from Eastern Amazonian soils and sediments

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## ABSTRACT

The recovery of metagenome-assembled genomes (MAGs) allows the taxonomic and functional characterization of previously unknown microorganisms. Therefore, this approach can be used to study complex environments, such as Amazonian soils and sediments. Changes in land use have been placing this region as the focus of several studies due to their impacts on the soil microbiota, including the microbial communities related to the cycling of the greenhouse gas methane (CH<sub>4</sub>). In this study, we demonstrate how we are recovering archaeal and bacterial MAGs from Eastern Amazonian soils and sediments using culture-dependent and -independent strategies followed by computational methods in the KBase platform. Forest and pasture soils were subjected to cultivation with CH<sub>4</sub> and a microcosm experiment with different moisture levels. Samples from floodplain sediments were also directly evaluated. DNA samples were shotgun-sequenced on an Illumina platform and, using KBase apps, sequence processing (FastQC and Trimmomatic), assembly (MetaSpades and MEGAHIT), and binning (MetaBAT2, MaxBin2, CONCOCT, and DAS Tool) were performed for each group of samples, followed by quality checking (CheckM), taxonomic classification (GTDB-Tk and MiGA), and annotation of bins (DRAM). Understanding the microbial communities present in Amazonian soils and sediments is a great challenge, especially considering their diversity and complex interactions. Our results revealed the recovery of 16 medium- and high-quality MAGs from cultivation, 41 MAGs from the microcosm experiment, and 45 MAGs from floodplain sediments, belonging to 18 archaeal and bacterial phyla, including Actinobacteriota, Proteobacteria (cultivation), Binatota, Halobacterota (microcosm experiment), Crenarchaeota, and Euryarchaeota (floodplain sediments). Novel genomes from both domains have been detected, including taxa associated with the production and consumption of CH<sub>4</sub>, in addition to nitrogen and sulfur cycles. These MAGs have been the subject of in-depth studies in the laboratory and are expanding our view on the diversity and functioning of Amazonian soil microbial communities, recognized for their importance in biogeochemical processes on a local and global scale.

**Keywords:** Soil microbiology; metagenomics; MAGs; KBase; **Development Agencies:** FAPESP, CNPq.