

CULTURED METHANOGENIC SPECIES AND MOLECULAR DIVERSITY OF ARCHAEA IN AN AMAZONIAN RIVER SEDIMENT

Theme: Metagenomics and biodiversity

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In recent years, many research groups are devoted to study the Amazon region, including the relation of this area with climate change. In studies of greenhouse gas emissions, a high positive flux of methane from the Amazonian region to the atmosphere has been detected. Methane is the second most important greenhouse gas and is mainly produced by microorganisms belonging to the *Archaea* domain. These methanogenic archaea are responsible for approximately 70% of the total methane emitted to the atmosphere annually. However, studies involving archaea in natural environments are relatively recent and still scarce in Brazil. The objective of this work was to culture methanogenic strains from the Madeira river sediment and to characterize the *Archaea* diversity using 16S rRNA gene library. Most sequences obtained in the library (76%) were related to uncultured groups from the *Crenarchaeota* domain and almost half of the sequences presented less than 97% of similarity when compared to sequences available in databases, revealing the existence of archaea groups yet to be described in the literature. The enrichment cultures for methanogenic archaea in Madeira river sample exhibited a high production rate of methane (33% in 16 days) revealing this sediment harbor active methanogenic microorganisms. It was possible to detect cells belonging to the *Methanosarcinaceae* and *Methanobacteriaceae* families through the use of RNA fluorescent probes. These cells were cultured and identified as strains of *Methanosarcina* sp. and *Methanobacterium* sp., and are both being maintained alive under laboratory conditions, though not as pure cultures. Only one genus of bacteria was detected in the *Methanosarcina* culture; the bacteria belongs to the family *Veillonellaceae*, class *Clostridiales*, but had only 92% of similarity with other sequences in the databases. The use of culture media and substrates in concentrations standardized for the study of samples from organic rich environments can explain the preferential growth of these organisms and not of the uncultured ones detected in the libraries. The great diversity of uncultured Archaea found in this study emphasizes the need to broaden studies involving this group, focusing mainly in culturing and physiological assays that could help understand the ecological roles of these microorganisms in the Amazonian environment. Keywords: *Archaea*, methanogens, river sediment, microbial diversity.