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Methanogenesis and Associated Prokaryotes in Gut Contents of the Methane-Emitting Earthworm *Eudrilus eugeniae* from Brazil.

K. Schulz¹, S. Hunger¹, G. G. Brown², S. M. Tsai³, C. C. Cerri⁴, H. L. Drake¹;¹Univ. of Bayreuth, Bayreuth, Germany, ²Embrapa Florestas, Colombo, Brazil,³Univ. of São Paulo, São Paulo, Brazil, ⁴Univ. of São Paulo, São Paulo, Brazil

The in situ conditions of the earthworm gut provide an ideal transient habitat for ingested anaerobes. It was recently discovered that the earthworm *Eudrilus eugeniae* emits methane. The objective of this study was to resolve the trophic interactions of methanogens and other prokaryotes in gut contents of *E. eugeniae*. RNA stable isotope probing (RNA SIP) of bacterial 16S rRNA and *mcrA* of methanogens was performed with [¹³C]-glucose as a model substrate in anoxic microcosms of gut contents. Glucose consumption yielded numerous fermentation products, and facultative aerobes (e.g., *Aeromonadaceae*) and obligate anaerobes (e.g., *Lachnospiraceae*, *Veillonellaceae*, and *Ruminococcaceae*) were associated with the diverse fermentations. Methanogenesis was ongoing during incubations but not significantly stimulated by supplemental glucose and fermentation products, indicating that endogenous methanogenic substrates were not limiting relative to methanogenesis. *Methanobacteriaceae* and *Methanoregulaceae* were linked to methanogenesis, and acetogens related to *Peptostreptococcaceae* appeared to participate in the complex anaerobic food web. Methanogenesis was significantly stimulated in enrichment cultures by H₂-CO₂ and methanol but not by formate and acetate. These findings provide insight on the processes and associated taxa potentially linked to the turnover of organic carbon and methanogenesis in the alimentary canal of *E. eugeniae*.