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Direct identification of lactate-metabolizing microbial populations in lake sediment by stable isotope probing

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Organic matter turnover in lake sediments is driven by the activity of microorganisms, but the specific identity of the microorganisms involved in situ is largely unknown. In this study, we identified 13C-lactate-metabolizing microorganisms in littoral sediment of lake Constance by stable isotope probing (SIP) of rRNA. Sediment samples were incubated anaerobically in the presence of fully labelled 13C-lactate. Turnover of 13C-lactate started immediately as measured by the formation of 13C-carbondioxide and 13C-methane. After 23, 28, and 48 h of incubation, rRNA was extracted from samples and fractionated by density using isopycnic centrifugation. Terminal restriction fragment length polymorphism (T-RFLP) PCR analysis of reversely transcribed 16S rRNA molecules revealed a large diversity of Bacteria in gradient fractions containing "light" (12C-) rRNA; "heavy" (13C-) rRNA showed fewer terminal restriction fragments, indicating a specific labeling of populations from 13C-lactate. Specifically active populations were identified by comparative sequence analysis of clones in libraries of "heavy" rRNA fractions. Over time, the composition of predominant microbial populations in the most heavy fractions shifted from delta-Proteobacteria (23 h), to gamma- and beta-Proteobacteria (28 h), and members of the Clostridiales (48 h). Our results indicate that lake sediments inhabit a diverse and highly responsive lactate-metabolizing anaerobic microbiota.