



Identification of novel PCE-dehalorespiring microorganisms in anoxic river sediment by RNA-based stable isotope probing

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Perchloroethene (PCE), a persistent environmental contaminant, can be reductively dechlorinated by anaerobically dehalorespiring bacteria. However, the diversity of dehalorespirers and their environmental distribution is largely unexplored. Here, we identified novel PCE-dehalorespirers by stable isotope probing (SIP), a technique that allows to directly link structure and function of uncultivated microbial populations. River sediment was incubated with PCE at low aqueous concentration (20 μM) at 15°C and dehalogenation activity (40 nmol *cis*-dichloroethene $\text{ml}^{-1} \text{day}^{-1}$) was detected after 8 weeks. The microbial community was probed with ^{13}C -labelled acetate (0.5 mM) as electron donor and carbon source for 4 days. After RNA extraction, „heavy“ and „light“ rRNA were separated by isopycnic centrifugation, and Bacteria-related populations in gradient fractions were characterised by terminal restriction fragment length polymorphism analysis. In heavy gradient fractions of the microcosm with PCE, we detected a prominent 505-bp terminal restriction fragment (T-RF) and a few minor T-RFs, whereas in the control without PCE, bacterial rRNA was restricted to light fractions, and the prominent T-RFs found in the PCE-dechlorinating microcosm were of minor importance. Apparently, label was incorporated more effectively in PCE-respiring microcosms, which suggests the detection of dehalorespiring, ^{13}C -acetate-assimilating populations. In clone libraries of the heavy fractions, a novel clone cluster distantly related to the PCE-dehalorespiring *Dehalococcoides ethenogenes* str. 195 (92-94% sequence identity) was predominant. Other abundant clones grouped with β -proteobacterial perchlorate-respiring *Dechloromonas* and *Dechlorosoma* spp., and the δ -proteobacterial *Desulfobacterium* and *Geobacter* spp.. We postulate that these populations are directly or indirectly involved in PCE dehalorespiration.