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# Anaerobic treatment of LCFA-rich wastewater: assessing the bioaugmentation potential of Syntrophomonas zehnderi

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Long-chain fatty acids (LCFA) are commonly present in fatty-wastewaters. Complete LCFA degradation depends on the coordinated activity of syntrophic bacteria, which convert LCFA to acetate and hydrogen, and methanogenic archaea, that utilize these substrates, making the overall conversion energetically possible. LCFA-degrading bacteria are fastidious microorganisms with low predominance in bioreactors. Thus, addition of LCFA-degrading bacteria to anaerobic sludge can possibly improve LCFA biodegradation and enhance methane production.

In this work, a co-culture of *Syntrophomonas zehnderi* and *Methanobacterium formicicum* was added to non-acclimated granular sludge. Two sets of bottles were prepared, with and without sepiolite, a solid microcarrier. Sludge was bioaugmented with co-culture and supplemented with 1 mM oleate. Blanks (without oleate) and controls (with inactivated co-culture) were also prepared. Methane, VFA and LCFA were quantified.

Addition of *S. zehnderi* enhanced LCFA degradation, both in the assays prepared with and without microcarrier. In the bottles containing bioaugmented sludge and no microcarrier, acetate accumulated in the medium indicating a fast LCFA β-oxidation: after 15 days of incubation, maximum acetate concentrations (approx. 5 mM) were attained and 77% of the added oleate could be accounted for the acetate and methane measured. In non-bioaugmented sludge, acetate accumulation started later and, after 15 days of incubation, was not higher than 1.5 mM. In bottles containing microcarrier methane was produced at a higher rate. In this case only residual acetate concentrations were measured, indicating balanced syntrophic relations, maybe due to stimulation of bacteria-archaea relation by the microcarrier. Methane production from oleate was most favored in bottles supplemented with the syntrophic co-culture and containing microcarrier: 71% of the added oleate was recovered as methane after 12 days of incubation and a maximum methane production rate of 1.12 $mMCH_4day^{-1}$ was observed. Bioaugmentation with *S. zehnderi* enhances oleate biodegradation and can be potentially useful for a faster reactor start-up.

Keywords: LCFA, Bioaugmentation, Syntrophomonas zehnderi, Methane