

3-21

In situ identification of estrone-degrading microorganisms belonging to Betaproteobacteria in activated sludge

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In last 20 years, a variety of EDCs has been observed in sewage effluents, the further toxicity evaluation of EDCs in sewage effluents suggested that natural estrogens are the main contributors to the estrogenic potency of sewage effluents. To date, a number of bacterial strains have been isolated from activated sludge or soil, which can utilize estrone (E1), 17 β -estradiol, estriol or even 17 α -ethynylestradiol as carbon sources. However, who are the key members responsible for estrogen degradation in activated sludge are still unclear. The aim of this study was to in situ identify the key bacteria active in E1 degradation in activated sludge. MAR-FISH (microautoradiography combined fluorescence in situ hybridization) analysis was applied with a set of hierarchic FISH probes to target the key E1-degrading bacteria in more and more specific phylogenetic level. Based on the MAR-FISH screening results and the retrieved 16S rRNA gene information, probes Inc1197 and SHA822 were designed to target several genera in Incertae sedis-group and the cloned sequences associated to genus *Sphaerotilus*, respectively. MAR-FISH results with newly designed oligonucleotide probes revealed that the key in situ E1-degrading bacteria in three activated sludge samples affiliated with the sheathed bacteria *Sphaerotilus*. Moreover, *Sphaerotilus*-like E1-degrading bacteria were phylogenetically different from all the previously reported E1-degrading bacteria isolated from activated sludge. *Sphaerotilus* related E1-degrading bacteria contributed around 60 to 80 % of [³H]E1-assimilating cells in E1 degradations, as well as 78% of tritium-assimilating cells in E1-3-sulfate conjugate degradation in the studied activated sludge samples.

3-22

Effect of the addition of 3,5-dichlorophenol and bio degradable plastic on activity of activated sludge

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The activated sludge process is frequently exposed to various toxic pollutants, which have negative effects on growth of activated sludge microorganisms. One of these toxic pollutants is 3,5-dichlorophenol (3,5-DCP), an intermediate metabolite of the chlorine-based pesticide. 3,5-DCP has inhibitory effects on respiratory energy production, which are known as uncoupling effects. Recently, it has been shown that 3,5-DCP is adsorbed by biodegradable polyesters such as poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV). This study was undertaken to investigate the effects of the addition of 3,5-DCP and PHBV on microbial community structure and activity of activated sludge. Sequencing-batch activated sludge reactors were operated at a BOD loading rate of 0.4 g L⁻¹ d⁻¹ and with a hydraulic retention time of 2 days. Every week the sludge concentration was adjusted to 3,000 mg L⁻¹. Immediately upon the addition of 3,5-DCP, the total organic carbon removal rate decreased to 60%, but increased again to around 90% during 3 days of operation from the DCP addition. The addition of 3,5-DCP also resulted in the reduction of excess sludge. However, these effects were significantly reduced by the addition of PHBV. A 16S rRNA gene-targeted PCR-DGGE analysis showed that population shifts took place in the DCP-added reactor.

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