## Influence of the bioreactor operating mode and wastewater composition on the structure of microbial communities in activated sludge and abundance and activity of polyphosphate and glycogen accumulating organisms

## Adam Muszyński\*, Monika Załęska-Radziwiłł

Department of Biology, Faculty of Building Services, Hydro and Environmental Engineering, Warsaw University of Technology, Nowowiejska 20, 00-653 Warsaw, Poland, Tel.: +48 22 6212979; emails: adam.muszynski@pw.edu.pl (A. Muszyński), monika.radziwill@pw.edu.pl (M. Załęska-Radziwiłł)

Received 4 January 2023; Accepted 24 February 2023

## ABSTRACT

Two lab-scale sequencing batch reactors were seeded with flocculent activated sludge from a fullscale wastewater treatment plant, fed with synthetic wastewater with acetate as the only source of organic carbon and operated in anoxic-anaerobic-aerobic (SBR1) and truly aerobic modes (SBR2). In SBR1 granules were formed, while flocs in aerobic SBR2 were overgrown with filamentous bacteria. FISH showed that synthetic wastewater equally influenced the abundance of bacterial phyla and classes in both SBRs regardless the operating mode: Proteobacteria increased their abundance and outcompeted Chloroflexi and Actinobacteria, Betaproteobacteria was the dominant class, abundance of Alphaproteobacteria decreased, and Gammaproteobacteria remained stable. The anoxic-anaerobic-aerobic conditions and acetate favoured growth of polyphosphate (PAO) and glycogen accumulating organisms (GAO) in SBR1, whereas the aerobic conditions in SBR2, along with the acetate, triggered growth of filamentous Thiothrix/021N. Despite the large differences between the abundance of PAO/GAO in both SBRs, the composition of their populations did not differ significantly between reactors, and the type of organic substrate was a decisive factor shaping the structure of their communities. Tetrasphaera PAO, predominant in the seed, were outcompeted in both SBRs by Accumulibacter. Clades IA and IIA, C, D together constituted 78% and 73% of the Accumulibacter lineage in SBR1 and SBR2, respectively, whereas 97% of Accumulibacter in the seed was not targeted by any of the clade-specific probes. *Defluviicoccus vanus* cluster 2 was partially replaced by cluster 1 and *Competibacter* in both SBRs. The substantial abundance of PAO in SBR1 was reflected in the high enhanced biological phosphorus removal activity in anaerobic batch tests and high presence of intracellular polyphosphate granules at the end of the aerobic period. Despite the significant abundance of *Accumulibacter*, the biomass from SBR2 neither released P nor took up  $C_{\text{org}}$  in anaerobic batch tests, which indicates that the activity of ecophysiological groups was largely influenced by the mode of operating bioreactors.

*Keywords:* Biodiversity; Ecophysiological groups; Biological nutrients removal; Polyphosphate accumulating organisms; Glycogen accumulating organisms; Filamentous bacteria

\* Corresponding author.