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Zależności ekologiczne bakterii przemian związków azotowych w osadzie czynnym sekwencyjnego reaktora biologicznego podczas prowadzenia procesu anammox

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Abstract

Both in artificial and natural ecosystems biochemical processes involved in the nitrogen compounds transformations are related and nitrogen removal bacterial groups such as: anammox bacteria, ammonia oxidizing bacteria (AOB), nitrite oxidizing bacteria (NOB) and denitrifies often coexist with each other's. Among the nitrogen removal processes applied in wastewater treatment technology, the anammox (*anaerobic ammonium oxidation*) process is recognised to be the most energy-efficient and environmentally friendly process, especially for high strength ammonia wastewater.

The presented doctoral dissertation consists of four publications describing the ecological interactions among the particular nitrogen removal bacterial groups presented in the anammox *Sequencing Batch Reactors* (SBRs). The selection of experiments' parameters and the configuration of SBRs allowed for the study of the ecophysiology of microorganisms and anammox process efficiency, depending on the feeding medium type, temperature and immobilization of the tested biomass in the alginate carriers. Due to the most of nitrogen cycle bacteria are uncultivable in the laboratory *via* traditional microbiological methods, the molecular biology tools such as *Polymerase Chain Reaction - Denaturing Gradient Gel Electrophoresis* (PCR-DGGE), *quantitative PCR* (qPCR), *Fluorescent in Situ Hybridization* (FISH) and *Next Generation Sequencing* (NGS) were applied in order to: (I) present an insight into the microbial community dynamics and microbial ecophysiology in SBRs, (II) monitor the dominant species and genotypes as well as the relative number of functional genes of nitrogen transforming bacteria, (III) study of the spatial distribution of individual groups of bacteria in the anammox biomass, (IV) test the adaptability of anammox bacteria to the variable process conditions.

One of the limitations of the large-scale use of the anammox process is slow anammox bacteria growth, which extends an effective technological start-up. Therefore, the subject of the research described in publication 1 was the ecophysiology and dynamics of nitrogen removal bacteria during the implementation of the anammox process in the SBR. In publication 2 proportion of particular nitrogen removal bacterial groups and changes in the microbial community structure were examined in the relation to the feeding strategy in the SBR, while feeding medium was shifted from synthetic wastewater to real wastewater - landfill leachate.

Except for the slow growth rate of anammox bacteria, implementation of anammox process in mainstream wastewater treatment plants is limited by its relatively high optimal temperature (30°C). The promising approach to cultivate slow-growing bacteria and to develop a stable and high-rate nitrogen removal anammox system is the immobilization of anammox

sludge in the gel carriers, which was the aim of study presented in publications 3 and 4. The anammox process below its optimal temperature (so called: cold anammox) was enhanced by entrapping anammox sludge in the sodium alginate with reduced graphene oxide carriers which was used as a nanomaterial supporting the anammox process at low temperatures (publication 3) and combined carriers with sodium alginate and polyvinyl alcohol (publication 4). The process efficiency and microbial interactions inside the beads were monitored during temperature decrease in SBR reactors to 15°C, what is temperature close to the temperature of mainstream of wastewater entering the wastewater treatment plant.

The results presented in publication 3 and 4 pointed that there was a slower decrease in nitrogen removal efficiency during temperature changes in the case of immobilized biomass, what may suggest that immobilization, especially in the sodium alginate and polyvinyl alcohol carriers, is temperature shock-resistant during long-term operation. The protective properties of anammox sludge immobilization may open a way to a wider application of anammox process in the mainstream of the wastewater treatment plant, and may enhance the anammox sludge retention in the operational systems.

The results of the research carried out as a part of this doctoral dissertation confirmed that anammox bacteria, AOB, NOB and denitrifies coexist in the activated sludge in SBRs, even under conditions favourable to only one of the studied bacterial groups. The studies show that, especially in activated sludge systems, adaptation to changing environmental conditions should be considered as the adaptation of the entire community of microorganisms, along with the interactions of individual groups and the dominance of specific species and changes in the. The nitrogen removal bacteria in the investigated SBRs present mutual relationships, both in the suspended biomass and in the immobilized biomass, inside the carriers. The applied technology may promote the growth of a specific bacterial group, but molecular biology analyses indicate that remaining bacteria are present in the bioreactors. Physicochemical parameters affect the proportions of the presence of nitrifiers, denitrifies and anammox bacteria, but do not eliminate any group from the system.