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Redox stratified biofilms to support completely autotrophic nitrogen removal: Principles and results

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After 10 years of pilot and full-scale studies, completely autotrophic nitrogen via coupled aerobic and anaerobic ammonium oxidation is now firmly established in the wastewater treatment community. The reasons for the popularization of the technology are numerous, but the most attractive are probably the savings in energy consumption due to reduced aeration, the possibility of running the process without addition of an external carbon source and the low amounts of sludge generated.

Membrane-aerated biofilm reactors (MABRs) have been used in practice for more than 20 years in order to treat concentrated wastewaters. This technology consists of the installation of aeration membrane modules in completely mixed reactors. The purpose of the installed modules is not only to supply the electron acceptor required for completing a certain biochemical reaction, but also to serve as substratum for biofilm development. This unique configuration allows higher oxygen transfer efficiencies than conventional aerators. The resulting biofilms differ from conventional co-diffusion biofilms found in MBBRs or RBCs in the fact that the oxic biofilm region is in contact with the biofilm substratum and not with the bulk liquid. If operated properly, MABRs yield compact and homogeneous redox-stratified biofilms capable of hosting side-by-side aerobic and anaerobic microbial communities.

We have recently demonstrated that completely autotrophic nitrogen removal is feasible in MABRs at nitrogen removal rates as high as 5.5 g-N/m²/day in lab-scale reactors loaded with synthetic wastewater (8 g-N/m²/day) and compressed air (1.6 atm) with N₂O emissions 100-fold lower than conventional co-diffusion systems. A detailed study of the microbial community with Fluorescence In-Situ Hybridization revealed a very stratified biofilm structure with aerobic ammonium oxidizing bacteria dominating the areas of the biofilm closer to the membrane surface while anaerobic oxidizing bacteria populated mainly the outer biofilm region. The biofilm was approximately 500 µm thick and featured both aerobic and anaerobic ammonium oxidizing bacteria in compact reaction zones about 100 µm thick separated by an intermediate zone with low or null metabolic activity. Both identified microbial communities showed a very low diversity and were dominated by halophilic and halotolerant *Nitrosomonas sp.* and *Candidatus Brocadia anammoxidans*.

The continuous and sustained inoculation of metabolically active anaerobic oxidizing bacteria from a biofilm reactor placed in the recirculation line of our MABRs showed to shorten considerably the onset of autotrophic nitrogen removal. However, the main hurdle keeping MABRs from attaining high removal efficiencies was the presence of active nitrite oxidizing bacteria in the aerobic regions of the biofilm. Reactor conditions like operation at high pH and NH₄⁺ concentrations, low DO concentrations, and temperatures between 30-33 °C can favor the growth of aerobic ammonium oxidizing bacteria over nitrite oxidizing bacteria, but not in MABR biofilms because their presence at the biofilm base, once established, provides spatial protection. During our study we could demonstrate that intermittent aeration of MABRs for completely autotrophic nitrogen removal (even at high pulsing frequencies) prevented the outgrowth of nitrite oxidizing bacteria communities.

Autotrophic nitrogen removal is already revealing itself as a cheap alternative to treat concentrated nitrogen streams. However, we believe that this concept can be taken a step further in MABRs and become an even more cost-effective, compact and environmentally friendly solution.