



Impact of Salinity on Biological Wastewater Treatment

By

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A thesis submitted for the partial fulfillment of the
PhD Degree

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Statement

This thesis is submitted to Public Works Department, Faculty of Engineering, Ain Shams University in the partial fulfillment of the requirements for the Degree of Doctor of Philosophy in Civil Engineering.

The work in this thesis was carried out in joint supervision between UNESCO-IHE-Delft in The Netherlands and Public Works Department, Faculty of Engineering, Ain Shams University from 2012 to 2015.

No part of this thesis has been submitted for a degree or a qualification at any other university or institution.

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Abstract

Saline wastewaters can be produced from industrial activities, the use of seawater and brackish water in urban environments, or due to saline water infiltration into the sewer system. This can lead to sulfate-rich wastewater that under anaerobic conditions and the presence of electron donors can result in sulfide production. The direct use of saline water as secondary quality water for sanitation is a promising alternative to alleviate fresh water stress. However, sulfate-rich wastewater is produced due to the application of this approach. In the Sulfate reduction, Autotrophic denitrification and Nitrification Integrated (SANI) process, biological carbon and nitrogen removal is efficiently achieved for saline sewage treatment, but does not account for P-removal. The first part of this study focuses on evaluating the sulfide effects on the biological phosphorus removal process which is a key process to prevent eutrophication on surface-water bodies.

In this regard, both anaerobic and aerobic short-term sulfide inhibition tests were performed on an enriched culture of phosphorus-accumulating organisms (PAO) at different pH, and sulfide concentrations. It was found that sulfide had a negative effect on PAO activity, and the effect seemed to be related to un-dissociated H_2S concentration.

50% inhibition of the maximum acetate uptake rate of PAO was observed at around 60 mg $\text{H}_2\text{S}/\text{l}$ regardless the pH. With increasing H_2S concentrations, higher phosphate release to HAc uptake ratios were observed likely due to extra need for additional energy for cell detoxification. P-release for detoxification energy requirements (P_{det}) were estimated relative to the total P-release rate at a zero H_2S level. Increasing H_2S increased P_{det} until a maximum of 50%, while a further increase in H_2S caused a decrease in P_{det} which may have been compensated by higher glycogen utilization. Mathematical expressions have been proposed, which could satisfactorily describe the sulfide effects on acetate consumption and P-release..

The results showed that a reversible inhibition of aerobic P- uptake possibly occurred below a certain threshold concentration between 20 and

25 mg H₂S/L. With the increase of sulfide concentrations a decrease in the aerobic P-uptake was observed, and total cells inactivation likely took place at H₂S concentration ranged from 135 to 150 mg H₂S/L. The residual P uptake activity occurring below the threshold concentration was mathematically modelled and 50% of P uptake activity was reached at sulfide concentration of only 11 mgH₂S/L.

Implementation of these models into activated sludge models, can help to describe the kinetics of PAO when treating wastewater, containing sulfide. Nevertheless, the irreversible inhibition of aerobic PAO activity indicates the high possibility of EBPR failure under sulfide rich conditions. The results suggests that in the presence of sulfide aerobic metabolism of PAO would be the limiting step in the BPR process. Accordingly, the development of a potential SANIP process (SANI plus biological P-removal), could not be a promising solution, however, along the long term exposure of PAO to sulfide, adaptation was thought to be possible, and it was necessary to assess the feasibility of coupling both sulfate reduction and biological phosphorus removal processes, with the focus on the long term effect of sulfide on enriched cultures of phosphorus-accumulating organisms (PAO).

In this study, a quite low concentration of sulfide of 7 mgTS/L was chosen as a start to give a chance for adaptation, however, it was demonstrated that sulfide had detrimental effect on enriched PAO especially anoxically even at that low concentration. Sulfide caused irreversible inhibition to PAO as no recovery of activity was observed after decoupling the reactors. Highly enriched unadapted PAO culture suffered cell inactivation which might hinder the potential coupling of phosphorus removal to SANI process unless, the activated sludge acts differently compared to enriched PAO that was highly sensitive to sulfide exposure.

As a result, the evaluation of the effects of sulfide on the performance of the percentage of (PAO) in the activated sludge was advantageous towards promoting the evolution of the SANIP process. In this regard, short-term sulfide inhibition kinetic tests were performed at different sulfide and MLSS concentrations. It was demonstrated in this study that activated sludge was quite not affected by sulfide under anaerobic and

aerobic conditions and much tolerant to sulfide exposure compared to highly enriched PAO. Which is either due to the adaptation to sulfide shocks in WWTP or the big diversity of microorganisms that live in symbiosis with PAO and are able to use sulfide as electron donors e.g sulfide oxidizing bacteria under aerobic conditions or autotrophic denitrifiers under anoxic conditions. It was concluded that the implementation of SANIP process is a promising solution for nutrient removal when PAO is supported by a big consortium of bacteria as in the activated sludge.

Earlier in this study it was observed that under anaerobic conditions, and stress conditions due to sulfide exposure, there was a shift to GAO metabolism at high sulfide concentrations which agreed with recent experimental observations that indicated that some PAO cultures were able to perform a GAO phenotype under poly-P limiting conditions; i.e. accumulate acetate under anaerobic conditions without involvement of polyphosphate metabolism.

The present study investigated the ability of PAO clade I and II to adopt a GAO phenotype during long-term reactor operation under non-limiting poly-P conditions and during short-term experiments under poly-P limited conditions. Short-term batch tests demonstrated that both *Candidatus Accumulibacter Phosphatis* Clade I and II were able to gradually shift their metabolism from a PAO or mixed PAO-GAO metabolism to a GAO metabolism when the poly-P content decreased. However, under poly-P depleted conditions, the HAc-uptake rate by PAO I was 4 times lower than for PAO II, indicating that PAO II has a strong competitive advantage over PAO I during P-limiting conditions. Thus, from a practical perspective, this study also indicates that the efficiency of P-release processes for biological P-recovery or combined biological and chemical P removal systems, may be significantly affected by the type of PAO that prevails in the system.

Having the highly enriched PAO I culture (99%) in this study, inspired the idea of the last part of this research. Different engineering processes and system configurations were developed to achieve simultaneous

nitrogen and phosphorus removal biologically to favor the growth of DPAOs (PAO I) to take the advantage of using the same COD in the influent for both nitrogen and phosphorus removal to reduce costs, save energy of aeration, and take the advantage of lower sludge yield for less sludge production. Few studies were skeptical about their denitrification capacity of nitrate. However, it was difficult to clearly prove it due to the lack of pure PAO I culture. Hence, experiments were conducted to examine the denitrification capacity of highly enriched pure PAO I culture (>99%) fed with either acetate or propionate before and after acclimatization. When nitrate was the electron acceptor, no P uptake was observed even after acclimatization. On the other hand, simultaneous P and nitrite uptake was found with nitrite as an electron acceptor.

This study suggests that accumulation of nitrite in denitrifying systems is essential for anoxic phosphate removal by denitrifying PAO allowing simultaneous P and N removal over nitrate through the symbiosis of PAO with flanking denitrifying bacteria which denitrify from nitrate to nitrite to be further used by PAOs.

Keywords: SULFIDE; SULFATE SALINITY; SANI; NUTRIENTS REMOVAL, PHOSPHATE ACCUMULATING ORGANISMS; PAO I; ACCUMULIBACTER CLADE I; DENITRIFYING PAO; MODELING.

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