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Review of Doctoral Dissertation Entitled "The impact of sulfur transformations on nitrogen removal processes in systems with granular sludge" by Dominika Derwis submitted to the Gdańsky University of Technology Faculty of civil and Environmental Engineering

by

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PURPOSE

The purpose of this report is to provide a review of the doctoral dissertation entitled "The impact of sulfur transformations on nitrogen removal processes in systems with granular sludge" which has been submitted to the Gdańsky University of Technology Faculty of civil and Environmental Engineering by Dominika Derwis. The dissertation is being supervised by Prof. Dr. Hab. Inz. Jacek Makinia and Dr. Inz. Joanna Majtacz. I have been asked to address:

1. Formal evaluation of the dissertation
2. Relevance and appropriateness of the research
3. Assessment of the dissertation merits, including strengths and weaknesses of the objectives, methodology, results, discussion, and conclusions
4. Significance and originality of the research
5. Additional questions for the candidate (if any - normally to be discussed during the defense)
6. Conclusions and final statement

These items will be addressed in order.

FORMAL EVALUATION OF THE DISSERTATION

The research presented in this dissertation addresses the important topic of nitrogen removal and incorporates a variety of known but seldom-used metabolic pathways to accomplish effective biological nitrogen removal (conversion of ammonia-nitrogen to nitrogen gas) while significantly reducing the mass of organic carbon required. This is significant as many wastewaters requiring nitrogen to be removed lack sufficient organic carbon for effective biological nitrogen removal but possess alternate electron acceptor sources, especially sulfur compounds like sulfide, that can be used for denitrification (conversion of oxidized nitrogen species to nitrogen gas). This dissertation also investigates the use of oxidized sulfur compounds like sulfate that can be used as electron donors for the oxidation of ammonia-nitrogen to nitrite- and nitrate-nitrogen which can subsequently be reduced to nitrogen gas. The dissertation investigates the integration of the range of known nitrogen-sulfur-carbon (N-S-C) cycles to reduce the energy (oxygen) and carbon inputs required to achieve effective

biological nitrogen removal from a range of wastewaters, particularly those containing the relevant sulfur species.

The research presented in this dissertation also investigates and demonstrates the use of granular sludge technology in the context of investigating the unique combination of metabolic pathways considered. Granular sludge technology is a recent development that couples biological and physical selection to develop and retain biomass with superior settling characteristics. Superior sludge settling characteristics allow a higher biomass concentration to be maintained, thereby resulting in reduced bioreactor volume. The reduced process oxygen requirement associated with the sulfur-based metabolic pathways investigated in this dissertation can further exploit the improved sludge settling characteristics resulting from development of granular sludge. This occurs because the lowest bioreactor volume achievable is the volume needed to transfer the required process oxygen requirement. Reduced process oxygen requirements associated with use of this combination of metabolic pathways reduces the lowest volume needed for process oxygen transfer and allows maximum benefit of the better sludge settling characteristics to be realized. Granular sludge also provides an enriched source of environmental niches due to diffusion gradients that develop within them, thereby further fostering the ability to create the range of distinct environmental conditions needed to support a variety of biological metabolisms and the organisms responsible for them.

The research presented in this dissertation is also noteworthy in the combined use of a number of complimentary analysis techniques. These include, not only conventional analysis of chemical species, but also coupling these relatively standard methods with molecular methods to assess the microbial composition of the biomass produced in the biological reactors along with stoichiometric analysis of the data to determine the relative contributions of the various metabolisms cultured to overall nitrogen conversions and removal. This coupling of a diverse range of assessment techniques allowed for an extensive characterization of the biological transformations occurring in the biological reactors operated.

The research presented in this dissertation is extensive, representing the successful operation of two bench-scale bioreactors (test and control) for nearly 1-1/2 years. Four published peer-reviewed papers constitute the substance of the dissertation. A total of 15 other publications were produced during the course of the Ph.D. studies that are represented by this dissertation, along with participation in seven conferences. Of course, this was all accomplished during the "Covid Years".

The dissertation is of very high quality. The work is extensive, well described and documented, the analysis is excellent, and the conclusions well-supported. The quality of the writing is also excellent and would be considered to be excellent, even for a native English speaker.

The outputs from the results presented in this dissertation are excellent. The review paper that constitutes the first major contribution is extensive and will serve as a long-lasting reference for others. The three data papers progress logically through a sequence of increasingly complex bioreactor influent feeding conditions that build on each other and successively create

important knowledge and learnings that will be of value to both the scientific and practice communities. Taken together, these four papers, and their synthesis in this dissertation, provide an excellent roadmap for both further research but, perhaps most importantly, a guide to practitioners who seek to exploit the advantages of the combination of microbial pathways investigated in this research.

RELEVANCE AND APPROPRIATENESS OF THE RESEARCH

The topic of biological nitrogen removal, as addressed in this dissertation, is of high relevance. Scientific knowledge concerning the biological conversions of nitrogen species continues to grow, and the results of this dissertation contribute significantly to this growing knowledge. Even more important for an engineering dissertation, it contributes important knowledge that can (and will) advance the practice of removing nitrogen from wastewaters more effectively and with reduced input of other resources (energy and carbon). Nitrogen discharges represent one of the most significant impacts on water quality and water resource availability on a global basis. Nitrogen discharges result in oxygen depletion creating dissolved oxygen (DO) deficient hot spots along nearly all coastlines around the world, along with compromising the quality of inland water bodies. The research presented in this dissertation is highly relevant to advancing technologies and approaches to reduce nitrogen discharges. While the focus of this research is on the treatment of wastewaters to reduce nitrogen discharges, the results are more broadly applicable. The insights gained are also applicable to further understanding and optimizing reductions in discharges from non-point sources, such as agriculture. They are also applicable to further understand the cycling of nitrogen in the natural environment, especially in the deeper waters where low-DO conditions leading to hot spots are initiated. Research of this type is not only relevant and appropriate for an engineering faculty but especially relevant and appropriate for environmental engineering.

MERITS

Merits for this dissertation include:

- An extraordinary amount of work was accomplished. The volume of work completed, and completed in an exemplary manner by the way, is more than sufficient for a Ph.D. dissertation.
- This dissertation addresses one of the most important topics relative to the restoration and preservation of water quality on a global basis. Nitrogen pollution is near or at the top of the list in terms of factors adversely impacting water quality on a global basis. DO-deficient hot spots occur along essentially any coastline, and these are the locations where the majority of humanity live. While wastewater is not the only source contributing to these hot spots, it continues to be an important one. Moreover, the scientific knowledge produced from the research presented in this dissertation contributes to understanding methods to reduce nitrogen discharges from other sources, such as agricultural run-off.

- This dissertation demonstrates an integrated methodology for analyzing and better understanding the metabolic conversions occurring in biological systems. The coupling of traditional analysis of chemical species along with molecular biological analysis and fundamental process engineering analysis characterizing process stoichiometry provides an outstanding basis for fully extracting useful information for analysis and understanding. Moreover, the stoichiometric analysis can be further extended in practice as a very useful tool for analyzing existing systems and for system design. Thus, the basis is provided for use of the techniques integrated in this research for future extension into practice.
- This dissertation demonstrates that granular sludge can be developed in largely autotrophic and in mixotrophic biological systems, as studied here. Demonstrating that granular sludge can be developed in these systems will encourage others, including practitioners, to further pursue this option. The results also provide useful information and guidance concerning how to realize granular sludge. The known practical advantages of granular sludge and the fact that it can be developed for biological systems using these metabolisms will further encourage practitioners to pursue these highly resource (energy and carbon) efficient options for a wider variety of wastewaters.
- The significant contributions this work is making to expanded knowledge in the environmental engineering field. Not only do four peer-reviewed journal publications form the basis for this dissertation, but an additional 15 publications were authored during this Ph.D. program. This is an extraordinary number of publications and represents a significant contribution to knowledge.
- The research presented in this dissertation significantly extends knowledge on how additional microbial processes and metabolisms can be incorporated into full-scale treatment plants to reduce energy and carbon requirements. This represents a significant contribution to reducing the cost for wastewater treatment for wastewaters where applicable. Moreover, the cost reductions, coupled with reduced energy and carbon requirements, result in the advancement of more sustainable water management options.

SIGNIFICANCE AND ORIGINALITY

The research presented in this dissertation is quite original. Others have observed the occurrence of the various nitrogen transformation metabolisms evaluated in this research, but generally only when it happened to occur and then to explain unexpected results and the overall transformations observed. In contrast, this research systematically and progressively evaluates the contributions that individual metabolisms and their combination can contribute to more resource-efficient (energy and carbon) biological nitrogen removal. Moreover, this research couples the systematic investigation of these metabolisms with the development of granular sludge which, itself, offers numerous advantages. The research presented in this dissertation also integrates a variety of complimentary analysis techniques to fully characterize the transformations occurring, the organisms responsible for them, and the environmental

conditions that create the environmental conditions resulting in growth of the organisms responsible for the various transformations and the extent of the transformations realized.

This research makes a wider range of options available to remove nitrogen from wastewaters, along with means to reduce the resources needed (smaller bioreactors due to granulation, less energy and carbon). This is very significant due to the continuing and ubiquitous nature of nitrogen pollution. Not only do the results of the research presented in this dissertation contribute to cost reductions for wastewater treatment, they also contribute to the availability of more sustainable solutions to this wide-spread water quality problem. The results also inform the full range of transformations which can occur in natural systems, including for use to control nitrogen discharges from non-point sources such as agriculture and in the receiving environment.

ADDITIONAL QUESTIONS FOR THE CANDIDATE

1. What applications appear most beneficial to apply the knowledge gained from this research?
    a. Why do you say this?
    b. What opportunities would you seek to pursue these applications?
2. What are the benefits you expect when the results of this research are applied in practice?
    a. What is the order of magnitude value of the benefits?
3. Can you envision application of the learnings from this research to improved control of nitrogen discharges from non-point sources?
4. How do the results of this research inform a better understanding of the processing of nitrogen compounds in the natural environment?
5. Can these results be incorporated into biological process models, such as the International Water Association (IWA) Activated Sludge Models (ASMs) and the supporting software platforms such as SUMO?
    a. How would you proceed to do so?

CONCUSIONS AND FINAL STATEMENT

In conclusion, my evaluation indicates that the subject dissertation is extremely well conducted, contributes significantly to advancing our ability to control nitrogen discharges to the aquatic environment in a more resource efficient fashion, and is highly original. It integrates the N-S-C cycles and provides original, relevant, and significant insights concerning how to use this improved integration to create improved nitrogen removal processes. The research has also demonstrated the integration of a variety of analysis and assessment techniques to achieve an integrated characterization of the microbial processes occurring, not only in the systems studied but in other complex biological systems. This represents another important contribution.

The research is conducted with high quality and is very well documented in both the open peer-reviewed literature and in the dissertation itself. The document is very well written.

Overall, I would conclude that this dissertation is one of the best I have had the opportunity to review. It should receive the highest grade possible. I have also reviewed the Doctoral Dissertation Distinction Rules for the Gdańsk University of Technology Department of Environmental Engineering, Mining and Energy and find that, from my perspective, it clearly meets the criteria for being designated as a distinguished dissertation. I hereby provide my strongest support for the dissertation to be submitted to the Council of the Scientific and Research Discipline with the highest possible recommendation for designation as a Distinguished Dissertation.

Respectfully submitted:

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