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Interactive Comment

Interactive comment on "Impact of flow velocity on biochemical processes – a laboratory experiment" *by* A. Boisson et al.

Anonymous Referee #2

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This paper presents laboratory experiments on denitrification of water from the Ploemeur site (France) using PVC tubes both as substitute for a porous medium and as source of an unknown electron donor. While diffuse contamination by nitrate is a widespread threat to groundwater and thus deserves detailed studies, I must admit that I am not overly impressed by the presented work. The experiments presented by the authors are incomplete, the model is rudimentary at best, and a mechanistic, process-based analysis of the data is missing. The authors conclude that the flow velocity has an impact on denitrification rates. But this does not come to any surprise considering that the system is controlled by mass-transfer kinetics between the PVC tubes and the water flowing therein. In order to gain an in-depth understanding of the underlying processes, it is not sufficient to present data that mainly consist of nitrate losses between

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Interactive Discussion

Discussion Paper

the in- and outlet. I don't quite see what the readers learn by reading this paper beyond noticing that this particular set of experiments has been done. The material presented is not enough for a scientific paper in an international peer-reviewed journal.

While the authors repeatedly emphasize that the experimental setup is convenient, they don't discuss whether it is representative for any natural system. Plastic tubes are deliberately manufactured in such a way that the inner surface is as smooth as possible, whereas surfaces in real porous or fractured media are rough, which can have a dramatic impact on the formation of biofilms. Thus, it is difficult to transfer the laboratory experiments to in-situ conditions at the Ploemeur site.

The authors do not present a carbon balance. Some unknown additives within the PVC tubes are released into the water. Their chemical composition is not determined. Also, the authors miss quantifying the release rate of DOC from the tubes under different flow rates (which would have required additional experiments using sterile water rather than water from the site). It remains completely unclear which fraction of the released DOC is can be assimilated by the bacteria, whether these compounds are in any way comparable to DOC released by the matrix of organic-rich natural porous media, or whether the release rate of electron donors is comparable to conditions found in nature. The complete balance of carbon would include the release rate from the PVC tubes (and a mechanistic model explaining it), the fraction incorporated in the biomass, the fraction mineralized to inorganic carbon, the fraction leaving the system without modification, and the fraction metabolized to other organic compounds. All of that is missing.

The authors also do not present a real nitrogen balance. They state that a full balance is complicated because of small flow rates and simply measure the difference of the nitrate concentration between the in- and outflowing water. They cannot say whether denitrification is complete, neither do they quantify the nitrogen within the biomass. The authors make courageous assumptions about the fate of the nitrogen, which are based on assuming that molecular nitrogen is not formed at all and that the specific yield for the denitrifying bacteria in the Ploemeur water equals a single literature value, even though the microbiological literature has reported a wide range of specific-yield values for denitrifying bacteria, implying that every experiment requires a new estimation of the specific yield from accurate measurements. The only measurement that the authors really offer is the concentration difference between the in- and outflow as function of time and flow velocity. This is not enough to inform a mechanistic, process-based model that can be transfered to other experimental conditions.

As a final shortcoming of the experiments, the authors also don't quantify biomass directly. Based on their assessment, they come up with an effective thickness of the biofilms, but they don't measure the bacteria washed out of the system (which could easily be done by flow cytometry), neither have they performed a sufficient number of parallel experiments to sacrifice the tubes at different time points in order to quantify the biomass attached to the tube walls. With modern molecular-biological techniques becoming more and more effordable, a complete study would have included a genetic characterization of the microbial community in the natural inoculum, a genetic comparison with the bacteria in the biofilms and in the cells in the outflow, amended by transcriptomic and metabolomic analyses to quantify the denitrification activity. I don't want to say that "-omic" tools are always leading to quantitative results that can easily be used in models, but the microbiology in this paper is missing altogether.

Given the limited data, the authors present a "model" that does not aim at quantitatively describing mechanistic processes governing microbial kinetics and reactive turnover of nitrate and DOC in the system, but is restricted to book keeping. Interesting approaches on modeling the mechanisms of bioavailability at the pore scale have for instance been presented by Falk Hesse at UFZ (Hesse et al., Adv. Water Resour. 32(8): 1336-1351, 2009; Environ. Sci. Technol. 44(6): 2064-2071, 2010; - I am not a coauthor of these papers). A complete model would require: the advection-dispersion equation for longitudinal transport, a mechanistic description of the DOC release, mass-transfer from bulk water to the tube wall, dual-Monod growth and turnover kinetics, and most likely terms expressing biomass mobilization (such as growth-mediated transport). I can imagine many interesting aspects of microbial kinetics that could be addressed by fitting sophisticated models to a sufficing data set gained from an experimental system like the one presented by the authors. However, the current paper does not even aim at that.

In conclusion, I don't think that this study is anyway near to being sufficient for publication in HESS or another peer-reviewed journal. I retain my long list of editorial comments (a large fraction of which addressing the correct use of prepositions in English) because I don't want to give the impression that a superficial revision would do.

Interactive comment on Hydrol. Earth Syst. Sci. Discuss., 11, 9829, 2014.