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Visualizing biofilms within porous media using contrast-enhancing staining agents

Tuesday, 31 May 2022 17:00 (15 minutes)

Bacteria colonize almost every habitat, including porous media such as rocks, sediments and soils. They are usually attached to the surface and agglomerated in biofilms. The location, extent and composition of the biofilm depend on the environmental conditions and chemical and physical characteristics of the material (Miller et al., 2012). They affect the material properties and influence fluid transport by obstructing the pore space, reducing the permeability and hydraulic conductivity (Baveye et al., 1998). Their influence is investigated for numerous industrial fields as they could contribute to wastewater treatment (di Biase et al., 2019), bioremediation of groundwater (Meckenstock et al., 2015) and carbon dioxide sequestration (Ebigbo et al., 2010).

It is important to visualize biofilms within a porous rock, know their location, and understand their effect on fluid flow inside the pore system. Within porous media, this could be achieved by X-ray micro-computed tomography (μ CT). However, distinguishing biofilms from the pore fluid, such as water, is hard due to a similar X-ray attenuation coefficient. Contrast-enhancing staining agents (CESAs) could enhance the X-ray attenuation, and a few CESAs such as particulate BaSO₄ (Davit et al., 2011), silver-coated microspheres (Iltis et al., 2011), 1-chloronaphthalene (Rolland du Roscoat et al., 2014) proved to be successful. However, these CESAs have some drawbacks, such as sedimentation, heterogeneous distribution of the particles and the fact that they change the pore fluid properties, such as the viscosity and wettability (Carrel et al., 2017). FeSO₄ could overcome these drawbacks (Carrel et al., 2017), and other CESAs, like Mono-WD POM and Hf-WD POM, could be interesting as they proved to be powerful staining agents for tissues (de Bournonville et al., 2020).

Within this research, numerous CESAs (KBr, FeSO₄, BaCl₂, Hexabrix, CA₄₊, Mono-WD POM, Hf-WD POM and Hexabrix) were tested that bind to the biofilm, and which could afterwards be replaced by the original pore fluid. These CESAs were screened for their potential to stain bacterial biofilms in between sand grains and on stones. The biofilms were imaged by HECTOR at the Centre for X-Ray Tomography (UGCT) of Ghent University (Masschaele et al., 2013).

In our experiments, most CESAs had a limited effect on the X-ray attenuation of the biofilms. However, Hf-WD POM and isotonic lugol were very promising CESAs for biofilm visualization using μ CT. Both were able to visualize cyanobacterial biofilms on rocks. Isotonic lugol led even to the visualization of (bundles of) filaments, and provides opportunities for future 3D microbial mat visualization. Moreover, it was possible to image and quantify the spatial distribution of biofilms inside a sand column.

Hf-WD POM and isotonic lugol create thus new possibilities to increase our understanding of the effect of biofilms on the pore scale. It could be possible to directly visualize their effect on flow paths, flow velocities and pressure gradients during a dynamic μ CT experiment. Moreover, μ CT could link the presence or absence of biofilms with changes in the pore system, including dissolution or precipitation.

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Time Block Preference

Time Block A (09:00-12:00 CET)

Participation

In person

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