

Microbial Dynamics and Biofilm Development in Contaminated Aquifers

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Background/Objectives. The performance assessment of monitored natural attenuation (MNA) and Bioremediation technologies in polluted aquifers is generally focused on the planktonic microbial community. Nevertheless, the bioremediation potential of biofilms and processes such as microbial attachment and stability in the attached phase are not fully understood. These processes are relevant in systems such as growing plumes or in studies which usually assume an arbitrary time for the development of a stable biofilm. This presentation illustrates the relevant time scales of microbial attachment and the differences in community structure that develop between planktonic and attached communities in a biofilm. A molecular biological characterization was made of groundwater and aquifer sediment samples collected from a field-scale in situ incubation experiment in a phenol polluted aquifer and corresponding laboratory microcosms constructed with the same materials. The molecular analysis of these samples from both settings was used to identify changes in the community structure of these microbial populations observed during biofilm development in the aquifer.

Approach/Activities. Anaerobic laboratory microcosms were prepared using sterile sediment from the aquifer and a groundwater inoculum sampled 30 meters below ground level (mbgl) at the fringe of the phenol plume using a Multi-Level Sampler (MLS). At field scale the same sediment was incubated in situ at 30 mbgl within the open screen of the MLS, with periodic sampling of the sediment and host groundwater at intervals over 780 days. The planktonic and attached microbial communities in these samples were analyzed using terminal-Restriction Fragment Length Polymorphism (t-RFLP), and Illumina sequencing of 16S rRNA genes, allowing changes in community structure over time in these growth phases to be investigated.

Results/Lessons Learned. The numbers of bacteria in the biofilm increased rapidly to a relatively stable number 3 months after inoculation. This trend was observed in both experimental (lab and field) settings until the end of the monitoring period. The results show that microbial community development and extent of attachment occurs over similar time-scales in both settings. Moreover, t-RFLP analysis allowed the community structure of the biofilm and planktonic communities to be differentiated. In the microcosms the planktonic community structure was temporally stable, whereas the attached community changed progressively in these systems. In contrast, in the in situ incubation experiment the planktonic community structure change over time, whereas the attached community structure was more stable. Therefore, the stability in numbers in the attached phase does not necessarily imply stability in terms of community structure in static environments. Conversely, biofilms could represent a stable community in changeable hydro chemical environments within groundwater plumes. This information is useful for the management of contaminant plumes using engineered interventions such as Pump and Treat (PAT) systems, considering the robustness of the biofilm community across time. It can support optimization of a PAT system to enhance the biodegradation capacity of the attached phase. This information is also important in models used to predict plume development or remediation designs which aim to enhance the biodegradation potential by shifting the community structure via the addition of nutrients and/or electronic acceptors. The bioinformatics analysis of the sequences will identify the groups driving community structure changes, the presence of biodegraders and the downstream design of molecular probes to determine changes in the community composition during biofilm formation in polluted aquifers.