33. Does Scale Impact Structure and Function of Microbial Biofilms?

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Project Goals: The overarching goal of the Ecosystems and Networks Integrated with Genes and Molecular Assemblies (ENIGMA) is to understand environmentally-relevant microbial community structure and function through a series of integrated field-to-laboratory campaigns. The described project is designed as an interdisciplinary platform to elucidate the fundamental behavior of microbial biofilms in response to physical scale of the particles to which microbial biofilms interact. Ultimately, we propose to elucidate physical and chemical principals that contribute to the assembly, evolution, and stability of microbial communities.

Sulfate-reducing-bacteria (SRB) occur naturally in a variety of anaerobic environments where sediments are present. In order to investigate the impact of physical surface scale on microbial interactions occurring in anaerobic habitats, attempts were made to standardize the growth of *Desulfovibrio* biofilm on various particle sizes using modified biofilm reactors. The standard coupon holders were modified to contain a mass of particles with continuous access to nutrients and *Desulfovibrio* culture, thus providing a surface for biofilm formation that could be easily removed at the end of the study period. By investigating at this finer resolution, patterns in microbial community structure and composition may be more discernable. The paradigm of "Everything is everywhere" has been commonly invoked, but obviously at a small enough scale 'everything' cannot be 'everywhere'. Does scale of analysis impact observation and interpretation for microbial communities? The reactor systems have used environmental isolates, *Desulfovibrio* RCH1 (Hanford) and *Desulfovibrio* FW1012B (Oak Ridge) to characterize growth on glass beads (30 μm, 425 μm and 3,000 μm). The surface area to volume ratio decreased with increasing bead size, and ranged from 1500 to 58 to 20 cm^{-1}, respectively. The biofilm protein per surface area ($\mu g/cm^2$) was 25-fold and 50-fold greater for the intermediate and largest sized particles, respectively, compared to the smallest. A similar trend was observed for biofilm carbohydrate (17- and 40-fold increased) compared to the smallest bead size. However, the overall biofilm carbohydrate to protein ratio was similar for the tested particle sizes (0.11, 0.07, 0.09, respectively). For the particle sizes tested, the amount of biofilm per unit area decreased with the particle size even as surface area/volume increased. One possible explanation for the counter-intuitive result is that the growth of biofilm significantly alters the porous structure and consequently changes porosity, permeability and dispersivity of the substratum. Because the tested particle sizes are significantly larger than the dimension of cells, we propose that initial colonization of beads is not mass transfer limited. However, as the biofilm formation proceeds on the beads and in the inter-bead space, the porosity of the packed bed changes thereby affecting the kinetics of biofilm growth. The changes are more pronounced for smaller particles and can also lead to heterogenous distribution and/or function of microbial populations. Future analyses include per bead measurements, biomass growth kinetics for different size of particles, and impacts on local diversity in comparison to source diversity.

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