

Composition and Metabolic Activity of Columbia River SIZ Microbial Communities in Relation to Habitat and DOC Source

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Hyporheic zone exchange of dissolved organic carbon (DOC) and nutrients between groundwater and river water plays a critical role in biogeochemical cycling within the Columbia River (CR) subsurface interaction zone (SIZ). Fluid exchanges may set limits on absolute rates of bulk carbon and nutrient processing, and impose deterministic selection on attached microbial communities that are adapted to utilize dynamically changing DOC sources. This study examined the response of attached microbial communities that developed *in situ* in three different zones (groundwater, hyporheic, and river water) within the SIZ to “cross-feeding” with DOC and other nutrients in either groundwater (GW) or river water (RW). Our working hypothesis was that *in situ* exposure history would dictate attached microbial community composition and physiological potential, thereby influencing rates of microbial metabolism. Sterile, organic-free Hanford sand was incubated *in situ* in 300 Area Hanford Formation groundwater, in piezometers embedded a few meters into the riverbed, or on the surface of the armored (pebble) layer of the riverbed itself (covered by dark screening to prevent illumination). Following a six-week period of *in situ* incubation and microbial colonization, the sands were retrieved and suspended in RW, GW, or artificial (organic-free) versions of these two fluid sources. Measurements of microbial biomass (ATP content), community structure (16S rRNA gene sequencing), and respiratory activity (aerobic microbial reduction of resazurin to resorufin), were made over a period of six weeks, with weekly exposure of the sands to fresh, sterile-filtered feed solutions. In contrast to expectations, the major observation was that the riverbed incubated sand had much higher biomass and respiratory activity, as well as a very different community structure, compared to the groundwater and piezometer incubated sand. Specifically, 16S rRNA gene analysis of community composition revealed that the most abundant taxa in the riverbed incubated sand were not prokaryotic organisms but rather the chloroplasts of diatoms. These results provide evidence for transport of diatom biomass into the riverbed. Analogous results were obtained in a repeat of the cross-feeding experiment, and in experiments designed to test whether DOC released from riverbed sediments could stimulate metabolism of colonized sand materials. A key implication of these results is that deposition of fresh, labile particulate organic carbon to the river bed leads to relatively high microbial biomass and metabolism by attached microbial communities. It also likely leads to release of DOC which may enter the SIZ via hyporheic zone exchange to stimulate microbial activity therein.