

## **The role of fluid residence time and colonization substrate in deep subsurface microbial community structure**

H. M. SAPERS<sup>1,2,3\*</sup>, R. BHARTIA<sup>3</sup>, G. WANGER<sup>1,2,3</sup>,  
J. P. AMEND<sup>2</sup>, B. KRUGER<sup>4</sup>, C. CASAR<sup>5</sup>, M. OSBURN<sup>5</sup>,  
V. J. ORPHAN<sup>1</sup>

<sup>1</sup>California Institute of Technology, CA 91125, USA  
(\*correspondence: [hsapers@caltech.edu](mailto:hsapers@caltech.edu))

<sup>2</sup>University of Southern California, CA 90007 USA

<sup>3</sup>NASA Jet Propulsion Laboratory, California Institute of  
Technology, CA 91109 USA

<sup>4</sup>Desert Research Institute, NV 89119 USA

<sup>5</sup>Northwestern University, IL 60208 USA

Assessing the influence of fluid residence time and mineralogy on the structure of subsurface microbial communities will lead to a better understanding of the factors that constrain their colonization and persistence. The Sanford Underground Research Facility (SURF), SD, USA, provides a natural laboratory for longitudinal study of microbial communities hosted within a warm (~30°C), deeply sourced, reducing aquifer transecting Early Proterozoic metabasalt and associated carbonate-facies iron formation. A series of four *in situ* environmental incubation experiments were designed to assess the effects of fluid residence time and substrate on the substrate-associated microbial community. Each deployment consisted of two parallel flow-through incubation systems in place for 3-5 months: a manifold (fast recharge, short residence time), and carboy (slow recharge, long residence time) both containing cartridges filled with either glass beads or locally-sourced iron formation as colonization substrates. There was a significant difference between the microbial communities colonizing the manifold and carboy cartridges in all but one deployment despite a significant difference in the overall microbial community between each deployment. The microbial community only differed significantly between the glass bead and iron formation colonization substrates in the longest duration (5-month) manifold deployment. These results suggest that fluid residence time has a significant effect on community structure in the deep subsurface over short time scales and that the colonizing substrate influences community structure over longer time scales when fluid residence time is short. These results have implications for deconvoluting the roles of transport and dispersion and environmental selectivity in the establishment and differentiation of endemic subsurface communities.