From Genomes to Metabolomes: Studying Mechanisms of Interspecies Interaction Using the Archaeal System *Ignicoccus-Nanoarchaeum*

Richard Giannone[1], Brian Bothner[2], Valérie Copié[2], Robert Hettich[1] and **Mircea Podar**[1,3*](mpodar@utk.edu)

[1]Oak Ridge National Laboratory, Oak Ridge, TN; [2]Montana State University, Bozeman, MT; [3]University of Tennessee, Knoxville, TN.

Few, if any, microbes live in functional or spatial isolation. The nature of the various types of inter-species interactions can be complex, ranging from competition to syntrophy and mutualism.. Such relationships can impact keystone species and play a major role in energy and element cycles at scales that extend past ecosystem boundaries. There is limited understanding of fundamental mechanisms of interspecies recognition and communication, how they impact genome evolution, what genetic regulatory mechanisms control metabolic/energetic coupling between species in response to environmental factors. To address such questions we are using the archaeal system *Ignicoccus hospitalis-Nanoarchaeum equitans*. With a combined genomic complement of less than 2000 genes and an obligate chemolithoautotrophic metabolism, this system represents one of the simplest specific microbial association and allows fundamental system level investigations and modeling of symbiosis. The integrated approach and the results of this research will be applied to investigations of more complex environmental systems.

A laboratory cultivation system for *Ignicoccus-Nanoarchaeum* has been established. The genomes of the two organisms are available and in addition we have sequenced two additional *Ignicoccus* species that do not interact with *N. equitans*. We have also performed an in depth proteomic analysis of *Ignicoccus-Nanoarchaeum* (1). Differences in the relative abundance of >75% of predicted protein-coding genes from both Archaea were measured to identify the specific response of *I. hospitalis* to the presence of *N. equitans* on its surface. A purified *N. equitans* sample was also analyzed for evidence of interspecies protein transfer. The depth of cellular proteome coverage achieved is amongst the highest reported for any organism. Based on changes in the proteome, *I. hospitalis* reacts to N. equitans by curtailing genetic information processing (replication, transcription) in lieu of intensifying its energetic, protein processing and cellular membrane functions. Using the information from initial studies we are now combining cultivation of these archaea under various settings with parallel transcriptomic, proteomic and metabolomic analyses. We are testing the specific hypothesis that the physical interaction between *Ignicoccus hospitalis* and *Nanoarchaeum* in laboratory cultures is induced and controlled by specific temporal gene expression, metabolic events and surface protein-protein interactions. We will identify candidate genes, proteins, and small molecules regulating the metabolic/energetic coupling network shared by the two organisms, which will allow us to establish a model of symbiosis at the genomic and metabolic level. Using comparative and functional genomics in multispecies cultures with archaea that do not serve as hosts for *Nanoarchaeum*, we will independently test the mechanism of interaction between *I. hospitalis* and *Nanoarchaeum* and will derive an evolutionary genomics model for the development of a microbial symbiotic relationship.

References

1. Giannone RJ, Huber H, Karpinets T, Heimerl T, Küper U, Rachel R, Keller M, Hettich RL, Podar M. (2011). Proteomic characterization of cellular and molecular processes that enable the *Nanoarchaeum equitans-Ignicoccus hospitalis* relationship. PLoS One. 2011;6(8):e22942.

This research is funded by the U.S. Department of Energy, Office of Biological and Environmental Research under the Genomic Science Program (DOE-DE-SC0006654).