

## Arctic Alder Nodule Microbe: Insights into Nitrogen Fixation

Neslihan Taş,<sup>1,2\*</sup> Nancy Conejo,<sup>1</sup> Hoi-Ying Holman,<sup>2</sup> and Verity G. Samon,<sup>3</sup>

<sup>1</sup>Earth and Environmental Sciences Area, Lawrence Berkeley National Laboratory, Livermore, CA;

<sup>2</sup>Biosciences Area, Lawrence Berkeley National Laboratory, Livermore, CA;

<sup>3</sup>Climate Change Science Institute and Environmental Science Division, Oak Ridge National Laboratory, Oak Ridge, TN

**Contact:** ([ntas@lbl.gov](mailto:ntas@lbl.gov))

**BER Program:** ESS

**Project:** NGEE Arctic

**Project Website:** <https://ngee-arctic.ornl.gov/>

This project uses state of the art sequencing and imaging technologies to resolve complex plant-microbe interactions governing the nitrogen fixation in tundra biomes to better inform efforts to decipher arctic nitrogen cycling. Plant roots are home to beneficial microbes where microorganisms help plants to obtain nutrients, tolerate stress and sustain growth. Nitrogen (N) fixation is a common mutualistic symbiosis where the plant provides the N-fixing bacteria with sugars and a variety of minerals, and the bacteria provide the host with N. We investigated the nodule microbiome of alder *Alnus viridis* spp. *fruticosa*, an N-fixing deciduous shrub and an important contributor to N-cycling in the tundra. *Frankia alni* and its relatives form a symbiotic relationship exclusively with trees in the genus *Alnus* but this actinorrhizal symbiosis is not as well characterized as the agriculturally important legume-rhizobia symbiosis, especially in the Arctic. The Kougarak study site is situated in the Seward Peninsula, AK. At this location alder is found as two distinct communities: short and dispersed alder growing in lowland areas (“alder savanna”) and tall alder growing as dense, tall shrublands along the rocky ridge of the hillslope (“alder shrubland”). We collected root free soil samples next to the plants and nodules from both alder communities. We extracted and sequenced the whole community DNA from both nodules and surrounding soils resulting several hundred metagenome assembled genomes (MAGs) and viral genomes (vMAGs) of bacteriophages. We coupled metagenomics with analysis of intra-nodule biochemistry via synchrotron fourier transform infrared (SR-FTIR) spectral imaging at the Berkeley Infrared Structural Biology beamline of the Advanced Light Source. The alder microbiome was dominated by a novel *Frankia* strain. Both alder communities contained non-*Frankia* bacteria but alder shrubland had a higher bacterial diversity than alder savanna. Non-*Frankia* bacteria in alder nodules contained carbohydrate degradation genes involved in hemicellulose, mannose and xylose degradation. Bacteriophages associated with alder nodule microbiome carry carbohydrate degradation genes specific to cellulose, xylose and pectin degradation. SR-FTIR analysis showed that despite differences in microbiomes alder nodules both communities had similar biochemical in the composition at a functional group level where metabolites involved in N-fixation was dominating the chemical spectra. However, we also observed strong intra-plant and -nodule variation which was decoupled from N-fixation processes. Arctic alder nodules house a diverse microbiome and has a rich biochemical composition. This diversity can be associated with plant growth and inputs of biologically-available N into nutrient-poor Arctic ecosystems.