

## **Evaluating the cost of accuracy of sequencing approaches**

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Water samples from British Columbia's Sakinaw Lake were used to help evaluate accurate phylogenetic resolution of microbial communities using iTags and PhyloTags. Credit: UBC Science, Flickr CC BY-NC-SA 2.0

Researchers use synthetic and natural microbial lake communities to compare the microbial community profiles generated from high throughput short-read sequencing and high throughput long-read sequencing approaches.

Microbes play key roles in maintaining the planet's <u>biogeochemical</u> <u>cycles</u>, but only a fraction of them have been characterized. Accurately profiling microbial communities provides researchers with information about the interactions and metabolic pathways in play, and provide insights into those gaps on the microbial tree of life belonging to



uncultured and unknown organisms.

A recent study estimated that the Earth could be home to as many as a trillion species, with the majority comprised of microbes. Characterizing and classifying these species makes use of 16S rRNA gene sequences, which represent a phylogenetic marker commonly used to assign all life into a particular classification. Characterizing the planet's microbial diversity provides researchers with more information about the roles these microscopic organisms play in maintaining the planet's biogeochemical cycles, and provides the U.S. Department of Energy (DOE) with insights into strategies for remediating environmental contamination. In the last decade, the ascension of high throughput, short-read sequencing platforms over the more costly but tried-and- true Sanger sequencing platform has allowed researchers to characterize tens of thousands of microbes, but at the cost of generating microbial community profiles at lower taxonomic resolution.

In a study that appeared online February 9, 2016 in The *ISME Journal*, researchers from the DOE Joint Genome Institute (JGI), a DOE Office of Science User Facility, describe an approach called "PhyloTags" that harnesses the long reads generated by the Pacific Biosciences sequencing platform. PhyloTags are then evaluated against the iTags generated from Illumina's high-throughput, short-read sequencing platform. They found PhyloTags provide a reliable adjunct or alternative to cost-effective iTags, enabling more accurate phylogenetic resolution of microbial communities and predictions on their metabolic potential.

Specifically, some microbial genera involved in nitrogen and methane cycling across the Lake's water column could only be resolved with PhyloTags, being missed by the short-fragment iTags. The team describes their work as the first benchmark study to offer a comprehensive comparison between these two sequencing approaches, allowing other researchers to see how using one platform over another



impacts the characterization of a microbial community. Each approach has advantages but each comes with costs as well. This work clarifies the benefits of each, enabling scientists to select the approach best suited to their experimental needs.

**More information:** Esther Singer et al. High-resolution phylogenetic microbial community profiling, *The ISME Journal* (2016). DOI: 10.1038/ismej.2015.249

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