Merging fungal and bacterial community profiles via an internal control

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Project Goals

1) Develop a synthetic marker to serve as an "internal control" that can be used to scale bacterial and fungal profiles from next generation sequencing of environmental samples.
2) Implement this internal control to calculate the fungal to bacterial ratio, a metric which can reveal ecosystem functioning.

Abstract

In soil communities, the ratio of fungal to bacterial taxa in a community can predict responses to environmental change and potential impacts on ecosystem function, facilitating both carbon management as well as modeling. As such, these processes and the underlying microbial taxa are a relevant focus of the LANL Terrestrial Microbial Carbon Cycling SFA program. While members of a microbial community can be measured with shotgun metagenomics, the cost of this approach severely limits the number of samples that can be examined. Targeted metagenomic methods are less expensive, and therefore more practical for large-scale studies involving hundreds to thousands of samples; however, these approaches require separate measurements of domains (e.g. bacteria versus fungi) due to the lack of a universal genetic marker. In an effort to mitigate the inequities of targeted approaches, we present the addition of a synthetic internal control marker.Compatible with kingdom-specific PCR primers for both the bacterial small ribosomal subunit (SSU) V3-V4 region, the fungal large subunit (LSU) D2 region, and the fungal ITS (internal transcribed spacer) region, this marker can serve as a scaling factor to aggregate fungal and bacterial taxonomic profiles for the quantitative analysis of targeted approaches. We demonstrate the utility of this universal marker on soil communities

with known and unknown composition, using next-generation sequencing and quantitative PCR. For environmental samples of unknown composition, our internal marker predicts F:B ratios that are consistent with qPCR. Additionally, the marker outperforms qPCR in terms of producing F:B values that are closer to the actual values of a defined mock community. Furthermore, the internal marker allows for a more streamlined approach than qPCR because it is quantified in tandem with standard next-generation sequencing.

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