Microbial Dark Matter Phase II: Stepping deeper into unknown territory

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JGI JOINT GENOME INSTITUTE DEPARTMENT OF ENERGY

Abstract

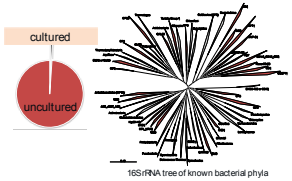
Currently available microbial genomes are of limited phylogenetic breadth due to our historical inability to cultivate most microorganisms in the laboratory. The first phase of the Microbial Dark Matter project used single-cell genomics to sequence 201 single cells from uncultivated lineages, and was able to resolve new superphyla and reveal novel metabolic features in bacteria and archaea. However, many fundamental questions about the evolution and function of microbes remain unanswered, and many candidate phyla remain uncharacterized. Phase II of the Microbial Dark Matter project will target candidate phyla with no sequenced representatives at a variety of new sites using a combination of single-cell sequencing and shotgun metagenomics approaches.

Key Questions

- How did bacterial and archaeal domains evolve?
- Is the early evolution and diversification of bacteria and archaea linked to adaptations to novel environments?
- What functional roles are candidate phyla playing in the environment?
- Are there detectable co-occurrence patterns of micro-organisms?
- What is the phylogenetic distribution of key metabolic functions?
- How variable is the use of genetic codes by bacteria and archaea?
- Are there novel phylum-level branches not present in reference databases?

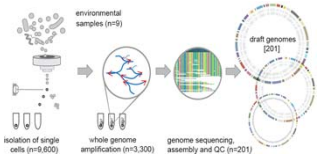
Microbial Dark Matter Phase I

RATIONALE



- The majority of microbial diversity remains unexplored
- Many branches of the tree of life have no cultured representatives or sequenced genomes

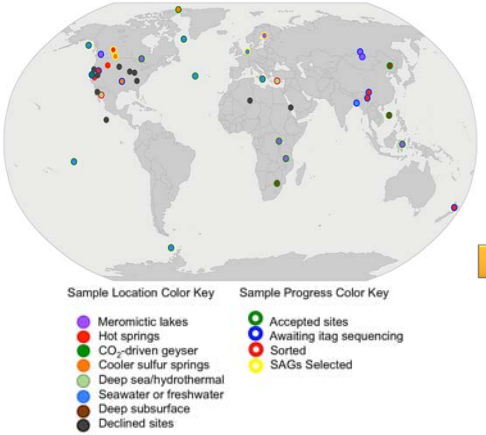
METHODOLOGY



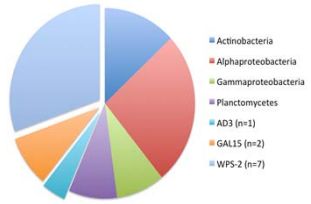
- Environmental samples are screened with 16S rRNA itags
- Samples enriched in target lineages are selected for sorting and single cell sequencing

FINDINGS

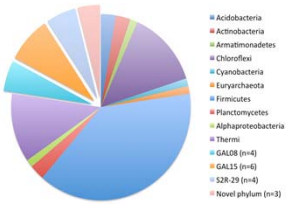
Phase II: New Sites, New Phyla



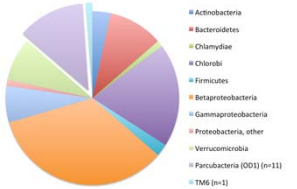
- Recruited new collaborators by searching literature for data sets with a high abundance (~40%) of candidate phyla
- Four samples sorted so far, others in various stages of progress
- Highly promising SAGs found in sorted samples, including a potentially novel phylum at Dewar Creek



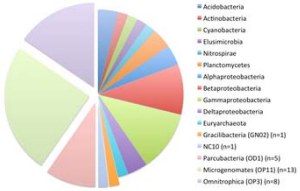
The Paint Pots
British Columbia, Canada
Peter Dunfield



Dewar Creek
British Columbia, Canada
Peter Dunfield



Lake Alinen Mustajärvi
Finland
Sari Peura



Unchlorinated drinking water
The Netherlands
Paul Van der Weilen

Next Steps

- Continue 16S rRNA itag screening and single cell sequencing
- Shotgun metagenomics on replicates of sorted samples, with multiple samples sequenced to enable binning by differential coverage (where available)

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