Introduction

Prochlorococcus is a numerically dominant photosynthetic prokaryote residing in subtropical and tropical oceans. It plays a key role in global carbon and energy cycles. Although members of the Prochlorococcus lineage are closely related, they have evolved significant genetic differences. The question our laboratory is addressing involves how this genetic diversity translates into variation in photosynthetic capacity and the ability to survive environmental stress.

In order to extend our understanding from the laboratory to the open ocean, we conducted a metagenomic analysis of the Sargasso Sea, where Prochlorococcus is often predominant among bacterioplankton. We aim to characterize the Sargasso Sea microbiome and understand how environmental selection shapes Prochlorococcus populations at different depths in the water column. We hypothesize that the partitioning of bacterioplankton (and specifically Prochlorococcus) populations and functions will exhibit significant differences between near-surface (40 m) and deeper (100 m) waters. These differences should illuminate micro- and macro-scale heterogeneity in key physico-chemical properties and biological interactions in the open ocean. We will attempt to answer these questions through comparative taxonomic and functional analysis along with contrasting cultured and uncultured genomes.

Metagenomics

Metagenomics is the application of genomics to uncultured assemblies of organisms in the natural environment. It is a fairly recent field; the first paper on marine metagenomics was published in 2004 (1). During August of 2009, the Ting Lab conducted field work in the Sargasso Sea (31° 40.00’N, 64° 10.00’W) on board the R/V Ting. The psychologist was an idea and Claire Ting - Department of Biology, Williams College

We hypothesized that the composition of bacterial phytoplankton at 40 m and 100 m will differ significantly. In order to describe the taxonomic distribution of our environmental samples, we identified genes in each scaffold and then assigned them to operational taxonomic units (OTU’s) in a process called binning. Gene calling was carried out by comparing each scaffold to the NCBI reference database. Genes were subsequently binned into OTUs with the least common ancestor (LCA) algorithm (Fig. 2).

Notably, we discovered that the 40 m microbiome is dominated by Cyanobacteria and, in particular, Prochlorococcus. The 100 m microbiome is extremely diverse.

12-strain Comparison

In order to illuminate the microdiversity between cultured and wild-type Prochlorococcus, the genes of all 12 cultured strains were compared to the environmental scaffolds. The number of subsequences producing significant alignments (hits, with significance determined by e-score) with these genomes were totaled.

Although the Prochlorococcus population decreases in abundance with depth, lineages and ecotypes trade dominance throughout the water column.

Functional Characterization

We expect the functional profiles of the water column to reflect micro- and macro-scale physico-chemical and biological characteristics. To describe the functional landscape of our metagenomes, we categorized proteins into hierarchical subsystems. Subsystems are a generalization of the term “pathway.” Protein-coding genes were queried against the SEED database for subsystem classification (6) (Fig. 4).

• The functional landscapes of the 40 m and 100 m bacterial communities are strongly correlated (r = .962) indicating conservation of broad community functions.
• Prochlorococcus at 40 m has a similar functional landscape as its community (d = .89) but Prochlorococcus at 100 m is dissimilar to its community (d = .307). This is consistent with Prochlorococcus’s dominance of the 40 m sample.
• In both communities, Prochlorococcus accounts for the majority of photosynthetic potential.
• Interestingly, the number of stress response genes associated with Prochlorococcus is higher at 100 m (Million vs. All). Communities lose their photosynthetic potential with depth.

Signature Analysis

To learn more about uncultured, environmental Prochlorococcus at 40 m and 100 m, we used sequence signatures to examine inter-genus differentiation (7). We computed tetranucleotide profiles for Prochlorococcus-specific scaffolds with a 1-bp sliding window and summed pairs of reverse complementary tetranucleotides. Principal components analysis revealed distinct partitioning of Prochlorococcus gene signatures at 100 m. The smaller group (cluster 1) is closely related to MIT 9303, while the larger group (cluster 2) contains sequences that bear similarity to a broad mix of cultured and uncultured strains.

The two clusters have similar functional profiles but differ in several ways. For example, cluster 2 has a greater proportion of photosynthesis genes (not shown).

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