# Microbial Trait Diversity Across Time in the North Pacific Subtropical Gyre

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## Introduction

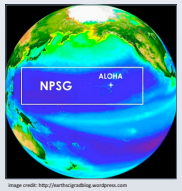
Microbes are a central component of marine biogeochemical cycles. Yet we still do not understand the extent to which microbial communities change across time or the environmental factors driving such changes, especially in relatively constant environments. Recent advancements in DNA sequencing technologies have given scientists an unprecedented window into the microbial world. In particular, shotgun metagenomic methods (community genome sequencing) provide a genetic 'parts list' of the traits microbes utilize to persist in their environment.

Here we leveraged both a 16s rDNA marker gene and shotgun metagenomic approach to study temporal variation in marine microbial communities in the North Pacific Subtropical Gyre. First, we investigate whether these two very different sampling approaches yield phylogenetic and trait based diversity measurements that are similar. Then, we investigate potential environmental drivers of trait diversity.

## Methods

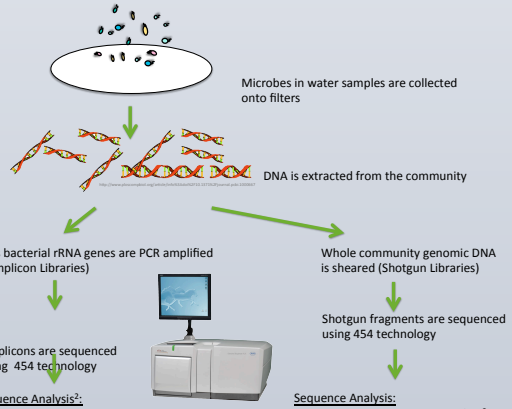
### Study Site & Sampling

Our study took place at Station ALOHA located north of Hawaii in the North Pacific Subtropical Gyre. Surface water at station ALOHA is highly oligotrophic year round and contains a permanently stratified mixed layer. Station ALOHA experiences mild but measurable seasonal climate variation.




Marine microbes were collected at roughly monthly intervals from August 2007 through September 2009, at 25m ocean depths. Microbial sampling took place alongside the ongoing long-term Hawaiian Ocean Time Series study[1], which regularly measures physical, chemical and biotic attributes across the water column.

### Characterizing Microbial Communities




## Results

### Trait Diversity Correlates with Phylogenetic Diversity

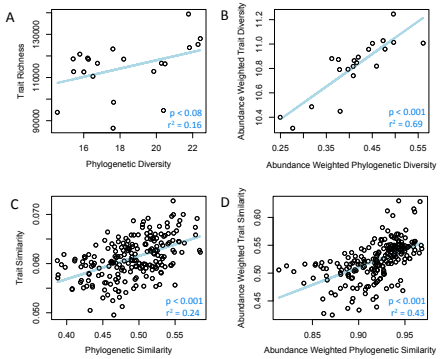



Figure 2. Diversity across microbial communities measured using PCR amplified bacterial 16s rRNA genes (x-axes) and bacterial protein clusters extracted from shotgun data (y axes). A) Phylogenetic diversity is the sum of branch lengths in a phylogenetic tree connecting all members of that sample[4]. Trait diversity is the number of unique bacterial protein clusters identified in each sample. B) Abundance weighted phylogenetic diversity is calculated by weighting phylogenetic branch length based on number of individuals at the tips of the phylogeny[5]. Abundance weighted trait diversity is calculated using the Shannon Index. C & D) Comparisons of the trait and phylogenetic composition of all possible combinations of two samples. Phylogenetic similarity and abundance weighted phylogenetic similarity are based on the fraction of branch lengths leading to members of both samples divided by the combined branch lengths of both samples[7]. Trait similarity is calculated using Jaccard and Bray-Curtis indices. Significance values calculated with linear regression (A & B) or mantel tests (C & D).

### Wind Driven Mixing Potentially Drives Variation in Trait Richness

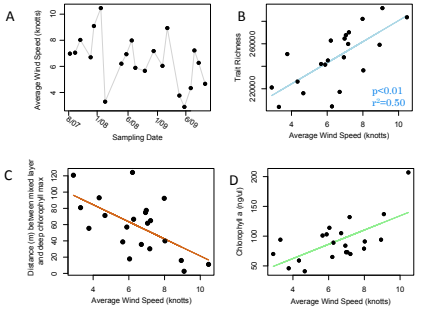



Figure 3. Evidence suggesting wind driven mixing events are a dominant driver of trait diversity at 25 m depth. Average wind speed is the average of data collected approximately every 10 seconds across the 4 days leading up to sampling[8]. A) Average wind speed of sampling dates. B) The relationship between average wind speed and trait richness. C) The relationship between the distance from bottom of the mixed layer to the deep chlorophyll max and wind speed. The depth at which chlorophyll concentrations at station ALOHA peak (the deep chlorophyll max) is variable and generally occurs below the mixed layer. D) Chlorophyll concentrations at 25 m correlate with average wind speed.

### Microbial Communities Segregate by Solar Radiation

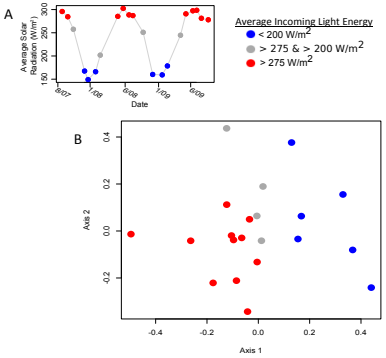



Figure 4. Solar radiation values are the average of shortwave (0.3-3µm) radiation measurements collected approximately every 10 seconds over the 30 days prior to sampling at the surface of the ocean[8].
A) Average solar radiation during sampling dates. B) Non-metric multidimensional scaling ordination of samples in trait (protein cluster) space.

## Discussion/Conclusions

1. **Phylogenetic and trait-based diversity measures of microbial communities sampled using 16s rDNA marker gene and shotgun metagenomic sequencing, respectively were correlated despite differing inherent biases in these two sequencing technologies and the subsequent diversity estimations. Our results are notable as our samples span a much smaller environmental gradient than other studies.**

2. **The correlation between wind and microbial trait diversity suggests that wind is a dominant driver of variation in trait diversity across 25 m samples. It is nearly impossible to determine underlying mechanisms from observational data, but the relationship between wind, chlorophyll concentrations and the deep chlorophyll max relative to the mixed layer supports mediation by wind-driven mixing.**

3. **Microbial trait composition correlates with average incoming solar radiation. As solar radiation is largely influenced by day length and sun angle, this observation demonstrates a seasonality to the traits of the microbes within communities at 25 m, despite the muted seasonal variation at the North Pacific Subtropical Gyre relative to other environments.**

## References

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