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The microbiome of the canopy-forming kelps, Nereocystis and Macrocystis, from the outer Olympic Coast to the Puget Sound

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The microbiome of the canopy-forming kelps, *Nereocystis* and *Macrocystis*, from the outer Olympic Coast to the Puget Sound

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Canopy-forming kelps in the Salish Sea

Macrocystis pyrifera
(Perennial)

Nereocystis luetkeana
(Annual)
Epiphytic microbial communities on *Nereocystis* kelp blades

Alphaproteobacteria
Gammaproteobacteria

Microscopy Image Credit: Jessica Mark Welch
Part I: Which microbes live on the blade surfaces of the canopy forming kelps *Nereocystis* and *Macrocystis*, and do they vary across a large spatial gradient?
Kelp forest spatial sampling

**Green** = *Nereocystis* only
**Pink** = *Nereocystis* + *Macrocystis*
Kelp forest spatial sampling

At each of the 13 sites:

• Sampled *Nereocystis* \((n = 6)\) and *Macrocystis* \((n = 6)\) blade tissues

• Sampled seawater \((n = 3)\) microbial communities: filtered 1.0 L through a 0.22 µm filter to collect microbes

• Collected environmental data (water temperature, salinity, etc.)

Green = *Nereocystis* only
Pink = *Nereocystis* + *Macrocystis*
Characterizing kelp and seawater microbial communities

Next-generation DNA sequencing:
- 16S rRNA gene
- bacterial/archaeal primers
  (515 F - 806 R)
- Average # of bacterial sequences per sample = 28,000
- Classified as different microbial taxa with the Green Genes database, and clustered into bacterial species with QIIME2
Results
Nereocystis and Macrocystis host significantly different microbial communities

Pairwise PERMANOVA:
pseudo-F = 10.32
$P = 0.001$
Macrocystis microbiome has greater bacterial diversity & evenness of taxa

Average # bacterial species per sample

Macrocystis = 164
Nereocystis = 62
Seawater = 321
Nereocystis and Macrocystis host some distinct microbes, share others.

Nereocystis shares ~50% of unique bacterial taxa with Macrocystis.

Macrocystis shares ~30% of unique bacterial taxa with Nereocystis.

Seawater 7925

Nereocystis 313

Macrocystis 688

Nereocystis shares 100

Macrocystis 107

Nereocystis shares 317

Macrocystis 358
Nereocystis microbial communities have a unique composition at certain sites.
Nereocystis microbial communities have a unique composition at certain sites in Southern Puget Sound.
*Nereocystis* microbial communities have a unique composition at certain sites.
Spatial variation in the *Nereocystis* microbiome

Significant spatial variation among sites
(PERMANOVA, pseudo-F = 2.40, P = 0.001)
Spatial variation in the *Nereocystis* microbiome

Abundance of *Saprospiraceae* bacteria on *Nereocystis*

Abundance of *Saprospiraceae* bacteria in seawater
Spatial variation in the *Nereocystis* microbiome

- *Saprospiraceae* (phylum *Bacteroidetes*) are significantly more abundant on the outer coast of WA.
- *Saprospiraceae* are known to degrade complex carbon substrates in marine environments (McIlroy and Nielsen 2014).
Spatial variation in the *Nereocystis* microbiome

Abundance of *Hyphomonadaceae* bacteria on *Nereocystis*

Abundance of *Hyphomonadaceae* bacteria in seawater
Spatial variation in the *Nereocystis* microbiome

- **Family Hyphomonadaceae** (class *Alphaproteobacteria*) are more abundant in southern Puget Sound

- *Hyphomonadaceae* are aerobic, heterotrophic, stalked bacteria that often live in oligotrophic waters (Abraham and Rohde 2014)
Part II: how do kelp and their microbes interact, and why should we care?
Kelp provide an abundant carbon resource for microbes

• On average, **14 – 40%** of the carbon fixed by kelp is leaked into the surrounding seawater as dissolved organic carbon (Abdullah and Fredriksen 2004, Wada et al. 2007, Reed et al. 2015)

• This process has never been measured in *Nereocystis*
Nereocystis blade dissolved organic carbon (DOC) production using $^{13}$Bicarbonate

$H^{13}CO_3^-$

$^{13}$DOC
Nereocystis blades exude 16% of total fixed carbon as dissolved organic carbon.

**Kelp**

- **Carbon Fixation**
  - $50.7 \text{ µmol L}^{-1} \text{ hr}^{-1}$

- **$^{13}$DOC Production**
  - $7.9 \text{ µmol L}^{-1} \text{ hr}^{-1}$

During an 8 hour long experiment, kelp fixed & released $^{13}$C.

16% of total fixed carbon is exuded as dissolved organic carbon.
Future directions: understand the links between the kelp microbiome, dissolved carbon exudation & nutrient cycling in kelp forests.
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  - University of Chicago Committee on Evolutionary Biology
EXTRA SLIDES...
(for questions)
Nereocystis vs. Seawater Spatial Variation

Nereocystis Samples

Seawater Samples

Bacterial Class
Spatial variation in the *Macrocystis* microbiome

Significant spatial variation among sites (PERMANOVA, pseudo-$F = 1.98$, $P = 0.001$)
PERMANOVA:
Cape Johnson and Destruction Island microbial communities are the same ($P = 0.16$), all other sites are significantly different ($P < 0.05$)