Macroalgal microbiomes; do host associated microbiomes facilitate environmental tolerances?

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Background

Macroalgae are of great environmental and ecological significance, as they are critical components of many marine ecosystems – contributing habitat for many species, as well as playing essential roles in carbon fixation. However, many macroalgae are at risk of habitat loss due to climate change, and this rapid environmental change is often too rapid to allow natural evolutionary response to prevent species decline. Microbes, on the other hand, have much more rapid generation times and may be able to adapt rapidly to climate change. Thus, host-associated microbiomes (the collective microbial community and genes associated with a larger 'macro' organism), present a potential avenue for research into macroalgal community responses to climate change. Microbial communities associated with many macroalgae provide key functions to the host, such as facilitating reproduction, disease resistance, and tolerance to environmental conditions, and thus have the potential to enable rapid adaptation to changing environmental conditions. However, research into microbial based adaptation to climate change is often limited by a lack of baseline knowledge around microbiome composition and function.

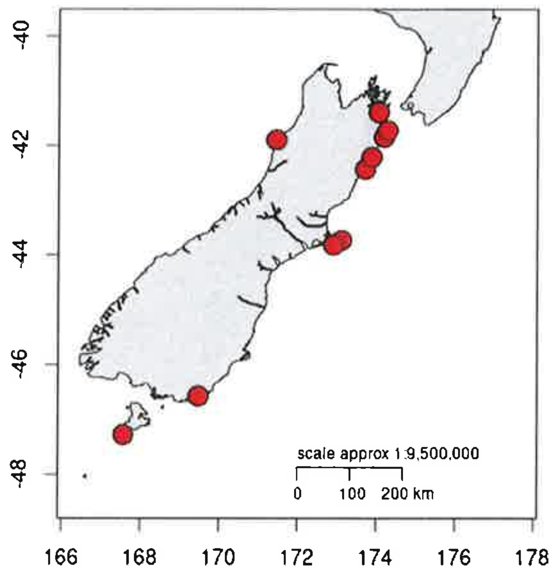
In this project, we examine the microbiome associated with southern bull kelp (*Durvillaea antarctica*), a large macroalga found around New Zealand and the Southern Ocean, in order to provide base line knowledge of microbiome composition across a range of environmental conditions. This work is enabling us to examine the drivers behind community assembly, including environmental and genetic factors, for the microbiome in *Durvillaea*. In this project we collected samples from environmentally diverse locations, across a broad geographic range, enabling us to explicitly examine environmental and genetic factors contributing to microbiome assembly.

Methods

We used GIS mapping to identify optimal sites for sampling in order to maximize the range of environmental conditions our sample hosts are exposed to, focusing primarily on sea surface temperature and wave action. We then sampled 10 individuals from each of 9 populations, collecting both tissue clippings and microbiome samples.

Sample collection

We collected samples from ten sites around the South Island of New Zealand. These sites were selected based on the range of environmental conditions, especially sea surface temperature and wave action.



At each site, we collected tissue clippings and microbial swabs from ten adult individuals. Tissue clippings were stored on silica gel until DNA extraction, and for the swabs we sampled approximately 30cm from the end of a blade. The area to be swabbed was first rinsed with artificial seawater to remove transient microbiomes, and then swabbed by rubbing a Whatman OmniSwab back and forth for 10 seconds over a 25 cm^2 area of the tissue, and then ejecting the tip into a storage solution of sterile DMSO EDTA Saturated Salt solution (DESS solution). Swabs were stored on ice in freezer until sampling was complete, at which point they were transferred to a -80 freezer until DNA extraction.

From each site, we also collected three 2L water samples, and filtered these through a 0.22um polycarbonate filter. In addition, substrate swabs were also collected following the same procedure as for the kelp, by swabbing the bare rock at the base of the host plant.

DNA extraction

DNA was extracted from swabs and filters using the Qiagen PowerSoil kit, following manufacturer's instructions with some modifications. First, all optional incubation steps were included, second, samples were bead beaten for 10 minutes at 25Hz, with sample trays rotated 5 minutes into the process to ensure uniform bead beating. For each batch of extractions, a negative swab was used as an extraction blank.

Samples for microbiome sequencing were submitted to Argonne National Lab (IL, USA) for DNA amplification and sequencing, and the results were then processed using the standard DADA2 pipeline.

Results

While most samples are still undergoing sequencing by Argonne National Lab, data from four sites sampled in 2020 indicate that kelp microbiomes are distinct from both the surrounding seawater and the substrate (Fig 1.).

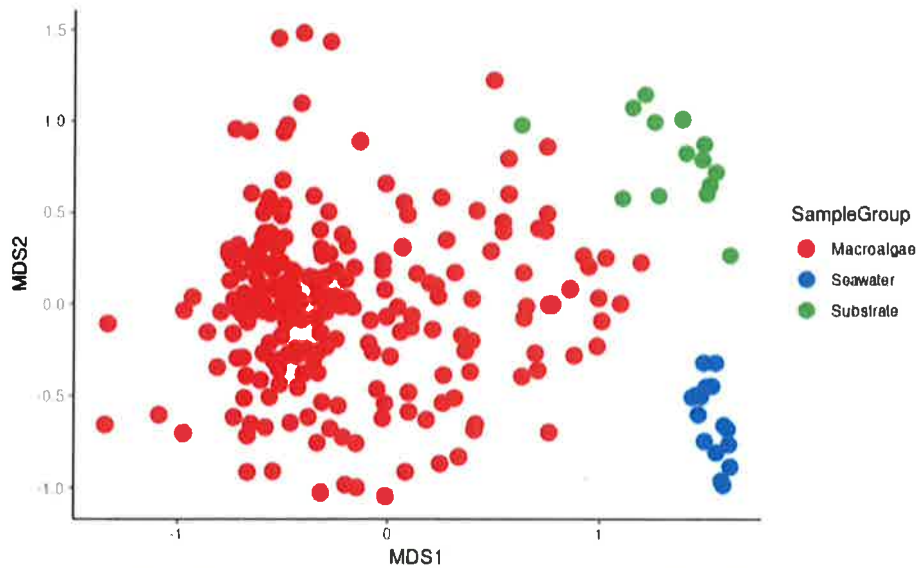


Figure 1 Multidimensional scaling analysis of microbiome samples collected from Durvillaea antarctica. *Seawater (in blue) and substrate (in green) samples cluster tightly and separate from* Durvillaea *(in red).*

The goal of the 2020 sampling was to simultaneously characterise the baseline microbial community of *Durvillaea,* identifying the core microbiome, as well as to understand the effects of the 2016 Kaikoura earthquake on the microbiome.

In communities flanking the uplifted zone we found significant overlap between microbial communities. Conversely, in communities within the uplifted zone we found much greater beta diversity of microbial communities (Fig. 2). Further analyses of these communities suggested that ecological drift (random changes in the abundance or presence of a microbial taxa) was a much greater driver for community assembly in uplifted and largely extirpated host communities, while selection (variance in reproductive success of different microbial taxa due to biotic [i.e., host] and abiotic [i.e., environmental] factors) appeared to be a greater driver in established flanking communities.

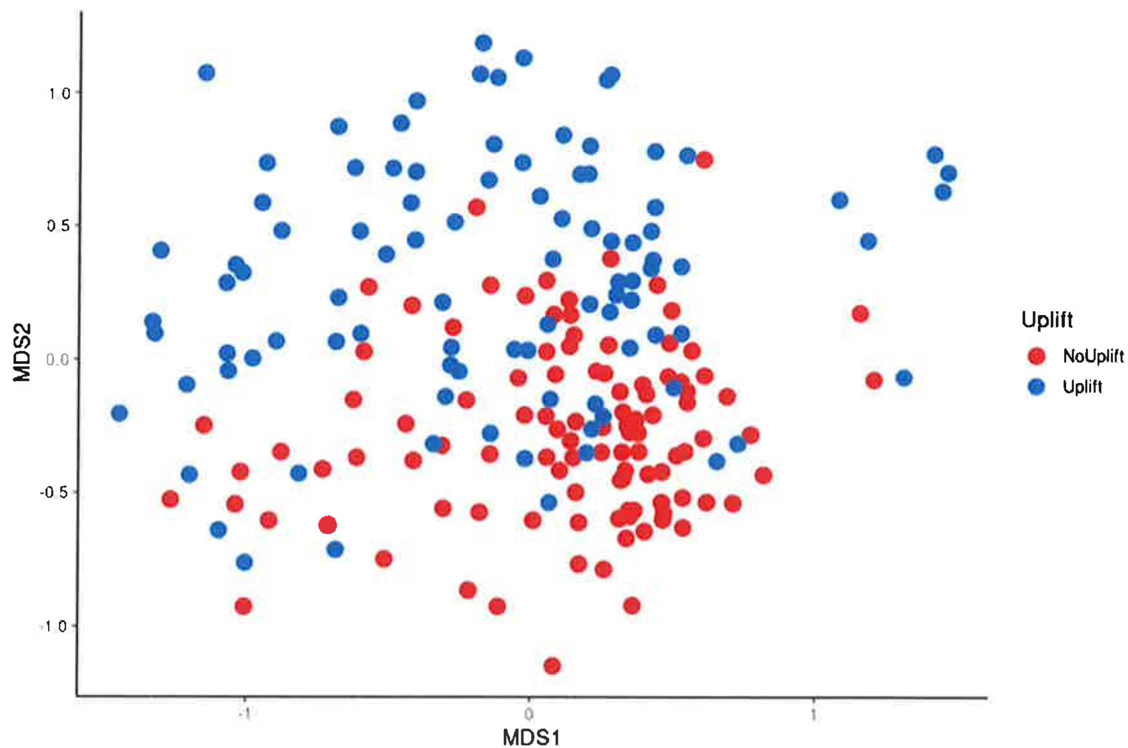


Figure 2 Multidimensional scaling of Durvillaea *associated microbiomes for hosts affected by the 2016 Kaikoura earthquake. Kelp from uplifted locations (in red) have greater dispersion than non-uplifted hosts, and also cluster separately to each other.*

While the current data are unable to determine environment-microbiome relationships, we find that four microbial taxa are present in over 90% of samples. Our data suggests that these 'core' microbiomes differ between two species of *Durvillaea*, and core microbes tend to be significantly more abundant in the host than in either seawater or the surrounding substrate (Fig. 3). This finding suggests that there is an element of host selection for the microbiome, which will be further examined with the samples currently being processed.

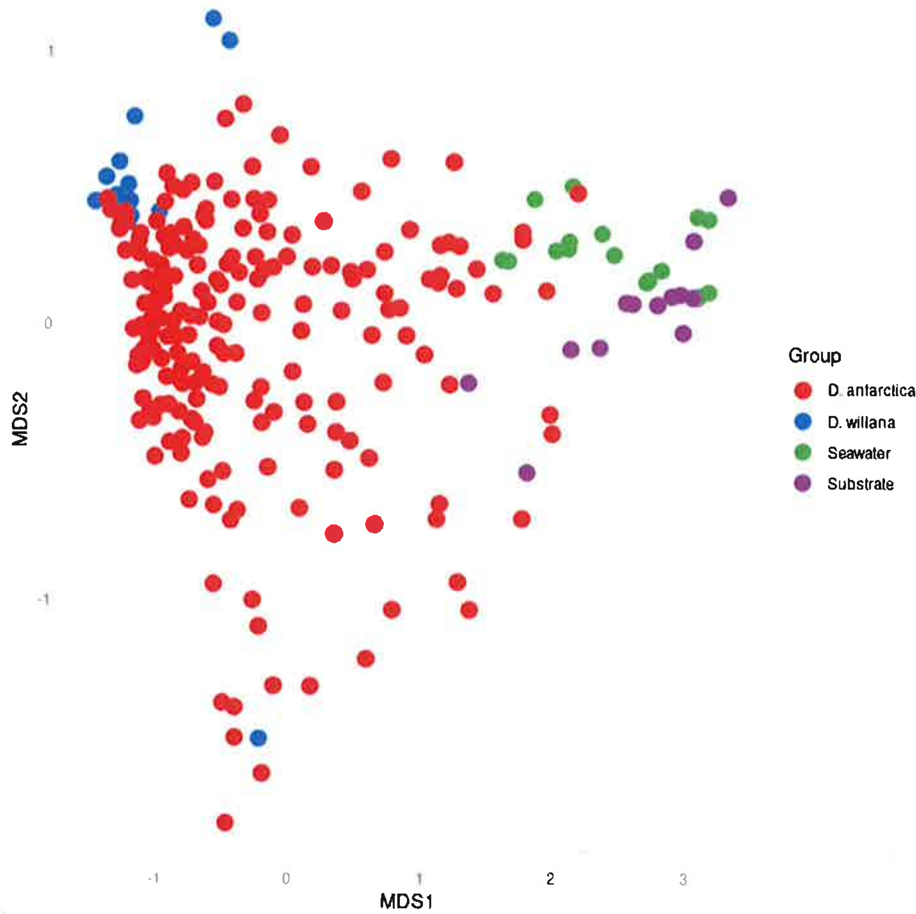


Figure 3 Multi-dimensional scaling analysis of the 'core' microbiome of Durvillaea antarctica. *Distinct clustering is observed between species* (D. willana vs D. antarctica*) and environment (seawater vs substrate).*

Use of funds

The majority of the $1,500 awarded was used to cover field work associated costs, production of experiment set ups, and DNA extraction consumables

Use of funds:

Item	Cost ($1,500) *Costs beyond the $1,500 were used from institutional and RSNZ funding.*
DNA extraction consumables	$421.59
Fieldwork costs (groceries, petrol, accommodation)	$718.69
Experiment set ups	$464.65

Remaining objectives and status:

At present, all samples for this project have been collected, and the DNA extracted. These samples are currently being processed for sequencing, and we anticipate all data to be generated within the next three months.

Signed: William Pearman **Supervisor: Ceridwen Fraser**