

Microbiome Manipulation: Using Beneficial Microbes of Coral to Increase Disease Resilience



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The Coral Holobiont

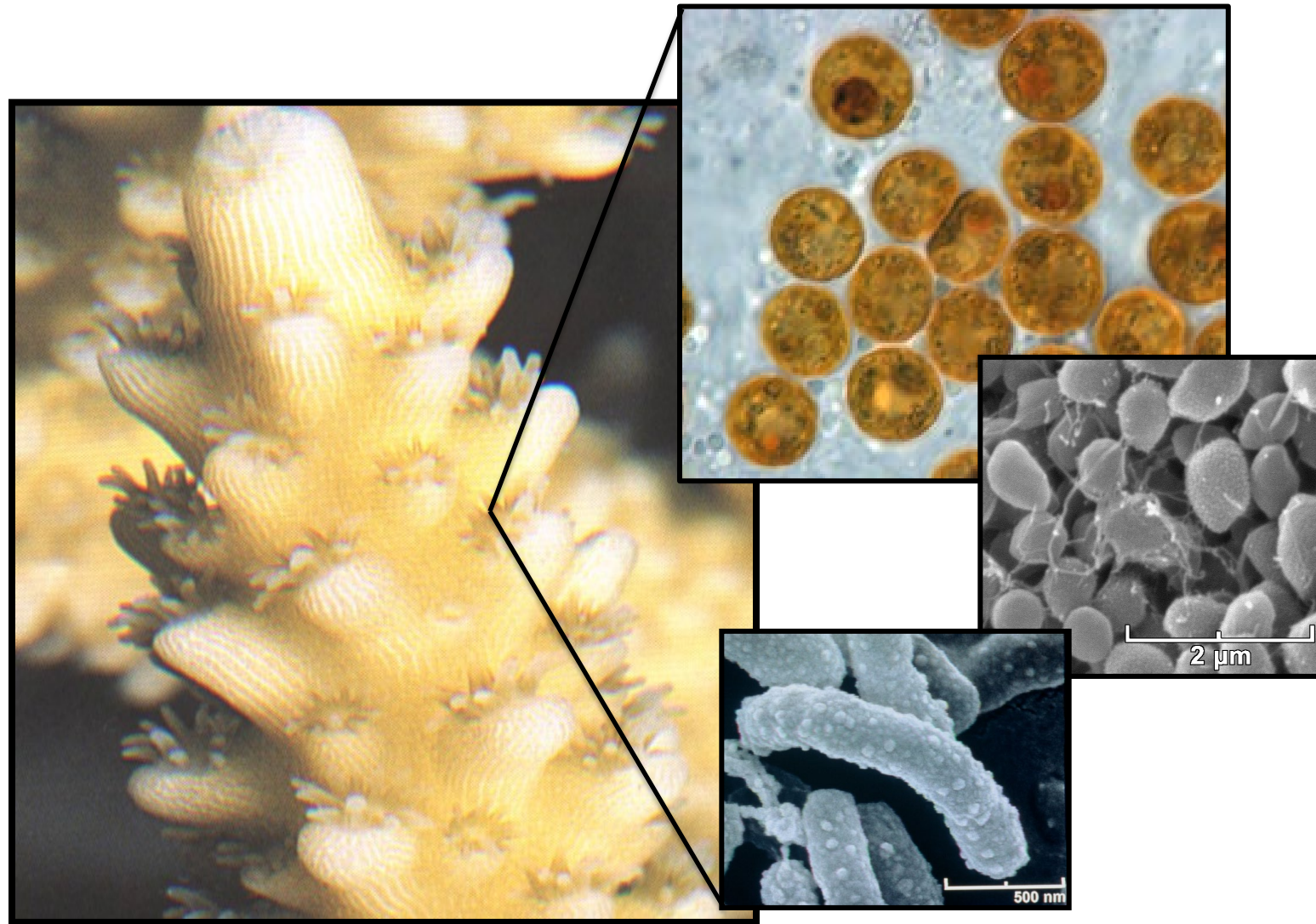
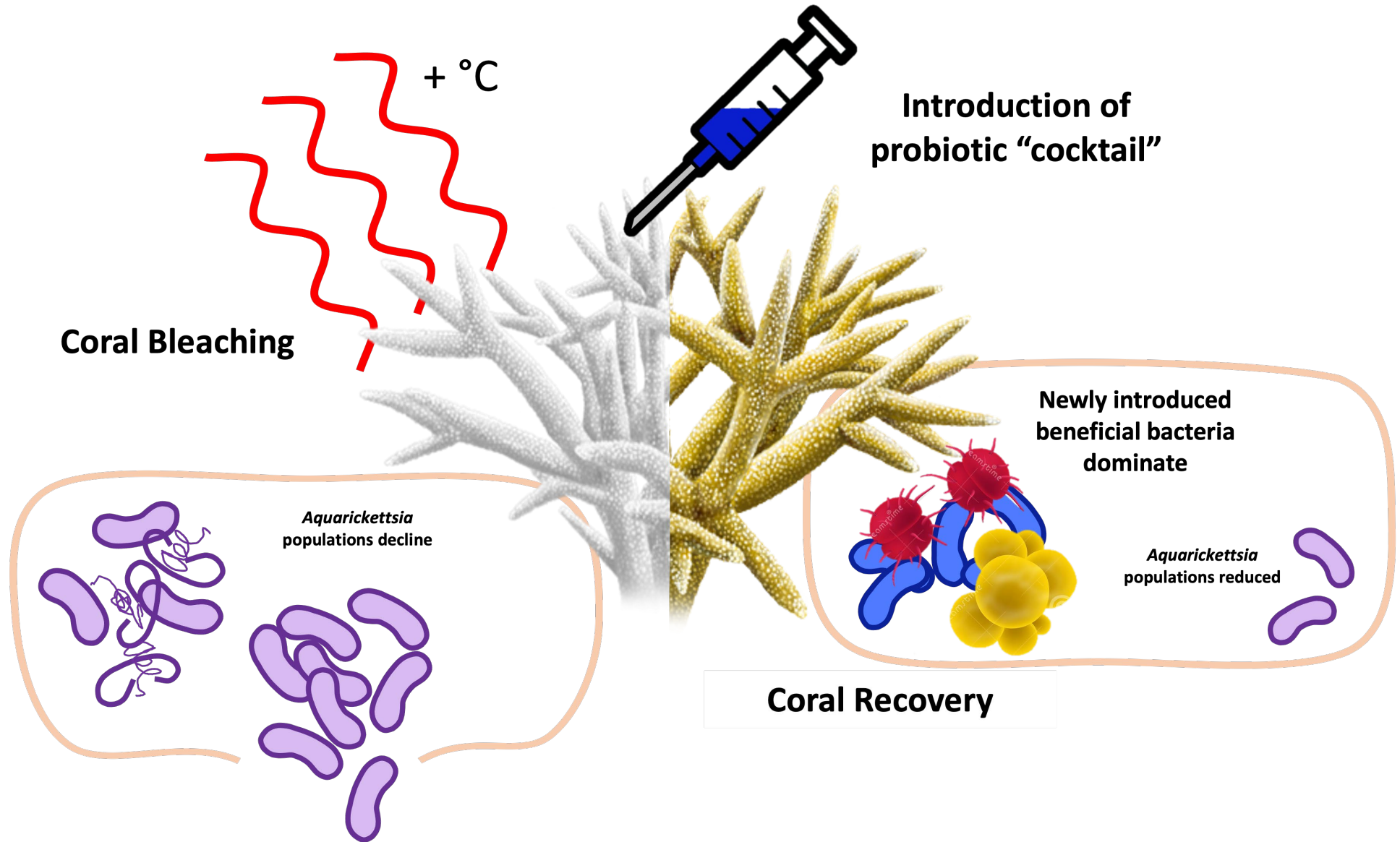


Photo credit: K. Ritchie

Beneficial Microbes of Coral (BMCs)

- Question: Can coral-associated microbes lead to increased resilience in *Acropora cervicornis* used in restoration in the FL Keys?
- Objective 1: *Isolate, identify, and characterize coral-associated strains of bacteria from the resilient genotypes and develop a library of potential isolates for probiotic capabilities.*
- Objective 2: *Introduce potential BMCs to susceptible genotypes of corals to manipulate their microbiomes.*

Beneficial Microbes of Coral (BMCs)



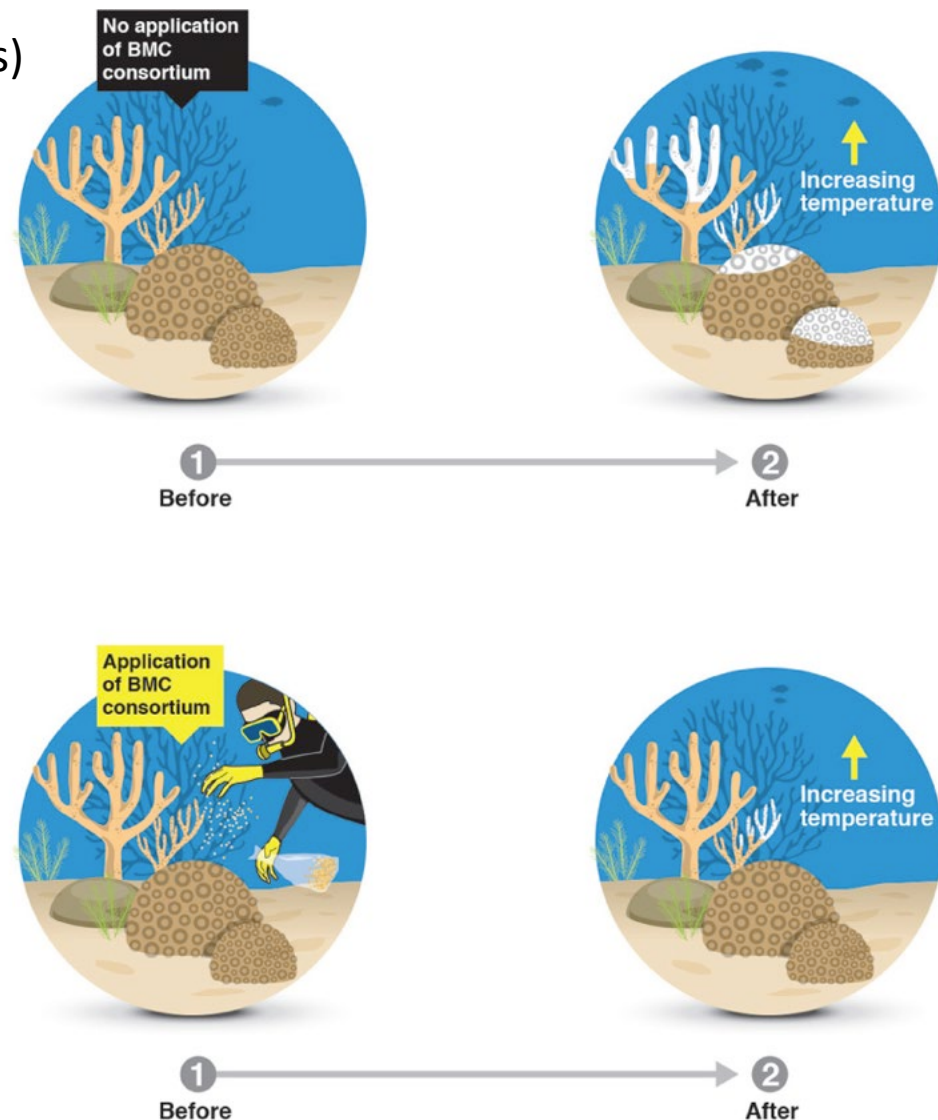
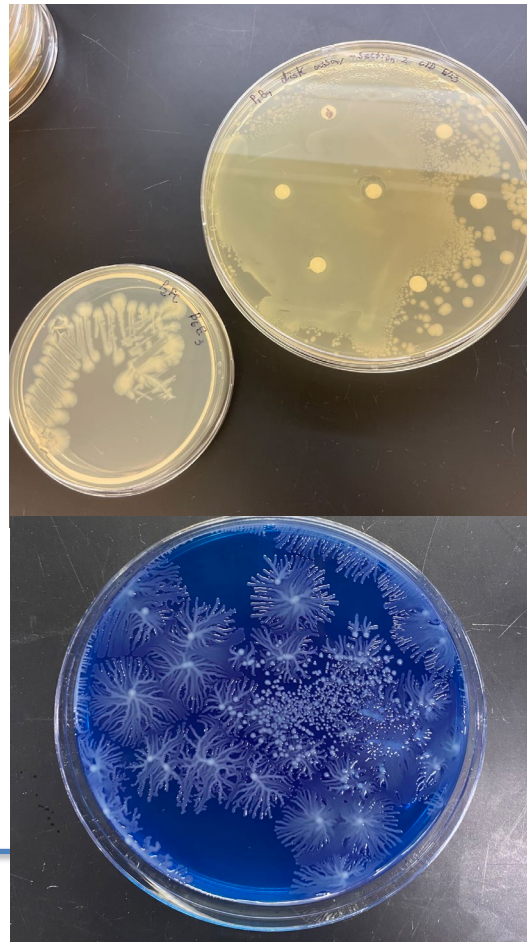


Credit: Penh Alicandro



The Approach in the FL Keys – Summer 2023

Beneficial Microorganisms of Coral (BMCs)



Results from bacterial characterizations

Coral Genotype	Isolates Identified	Swarming Inhibitory	β -galactosidase Inhibitory*	Total Screened (swarming)
#3	80	35	6	95
#7	64	31	0	91

Over 700 isolates in culture from four genotypes

Over 250 isolates identified by 16S rRNA sequencing

Dominated by *Vibrio*, *Photobacterium*, and *Pseudoalteromonas* strains

From the resistant genotypes:

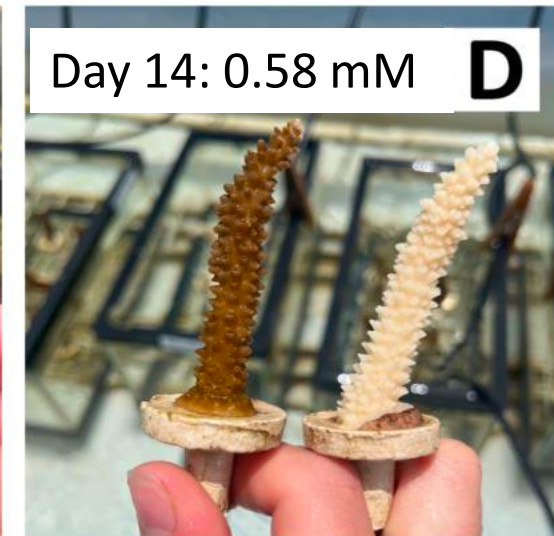
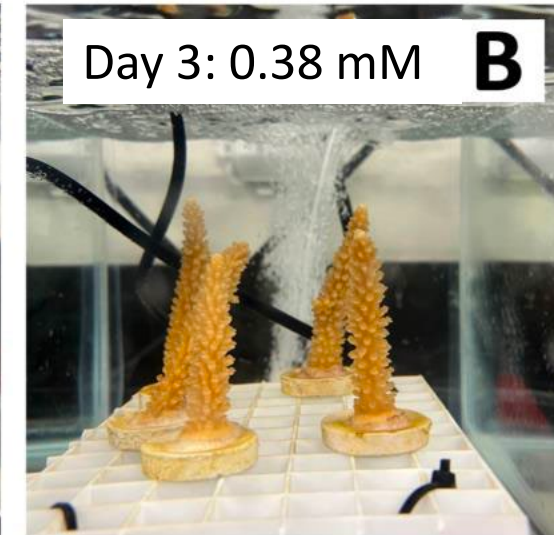
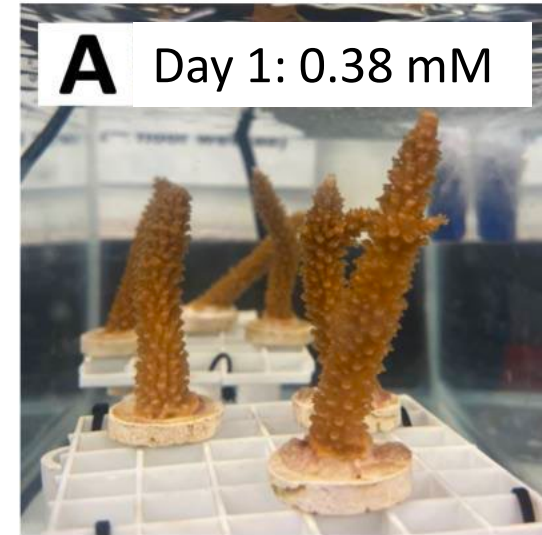
72 isolates show either swarming and/or B-gal* inhibition

BMCs of Disease-Resistant *A. cervicornis*

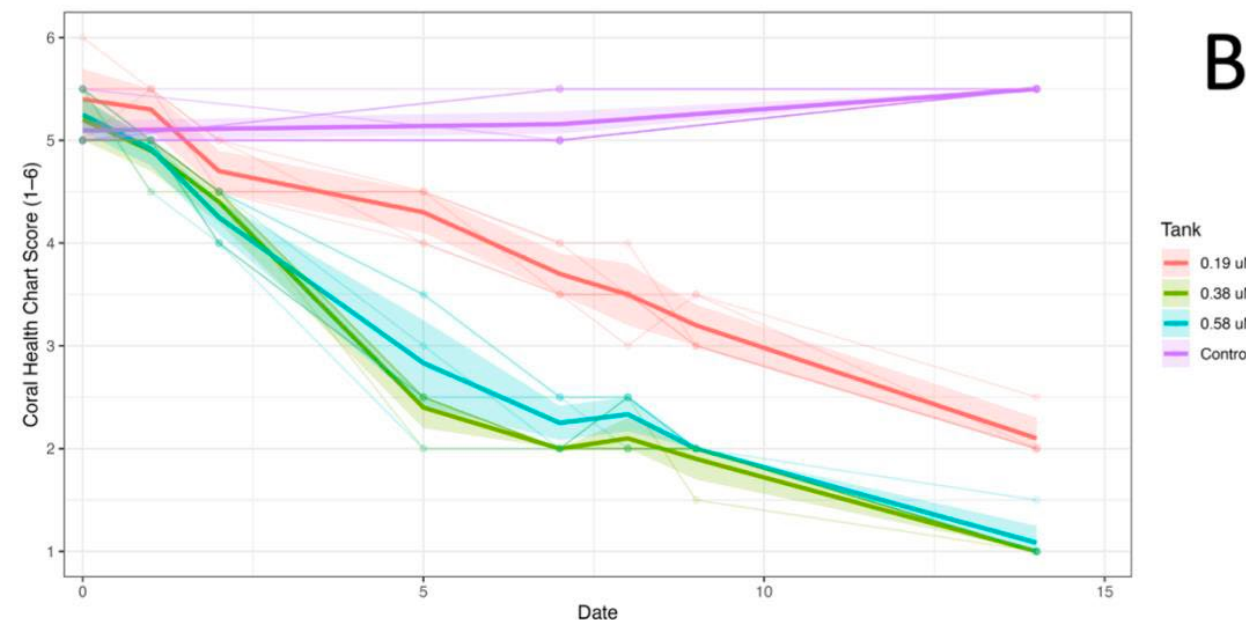
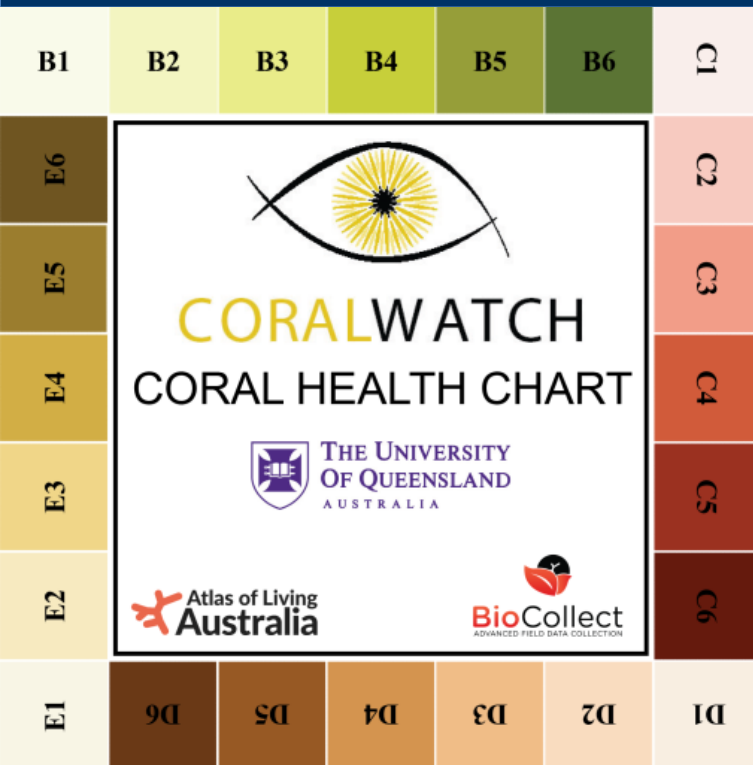
Strain Name	Best BLAST hit with 16S rRNA gene
P1A3	<i>Alteromonas macleodii</i>
P1B9	<i>Vibrio rotiferianus</i>
P1D7	<i>Vibrio alginolyticus</i> / <i>Vibrio owensii</i>
P1G7	<i>Vibrio harveyi</i>
P2B9	<i>Pseudoaltermonas shioyasakiensis</i>
P2C11	<i>Mariomonas</i> spp.
P2D4	<i>Vibrio coralliilyticus</i>
P2E2	<i>Vibrio ponticus</i>
P2F1	<i>Shewanella</i> sp. 49WBP
P2F6	<i>Vibrio orientalis</i>

Menthol-Induced Bleaching of *A. cervicornis*

- Menthol-Induced Bleaching Protocol
 - Three doses:
 - 0.19 mM, 0.38 mM, 0.58 mM
 - Incubation for 6 hours daily for up to two weeks
- Thermal-Induced Bleaching
 - 31°C after ramp from 27°C

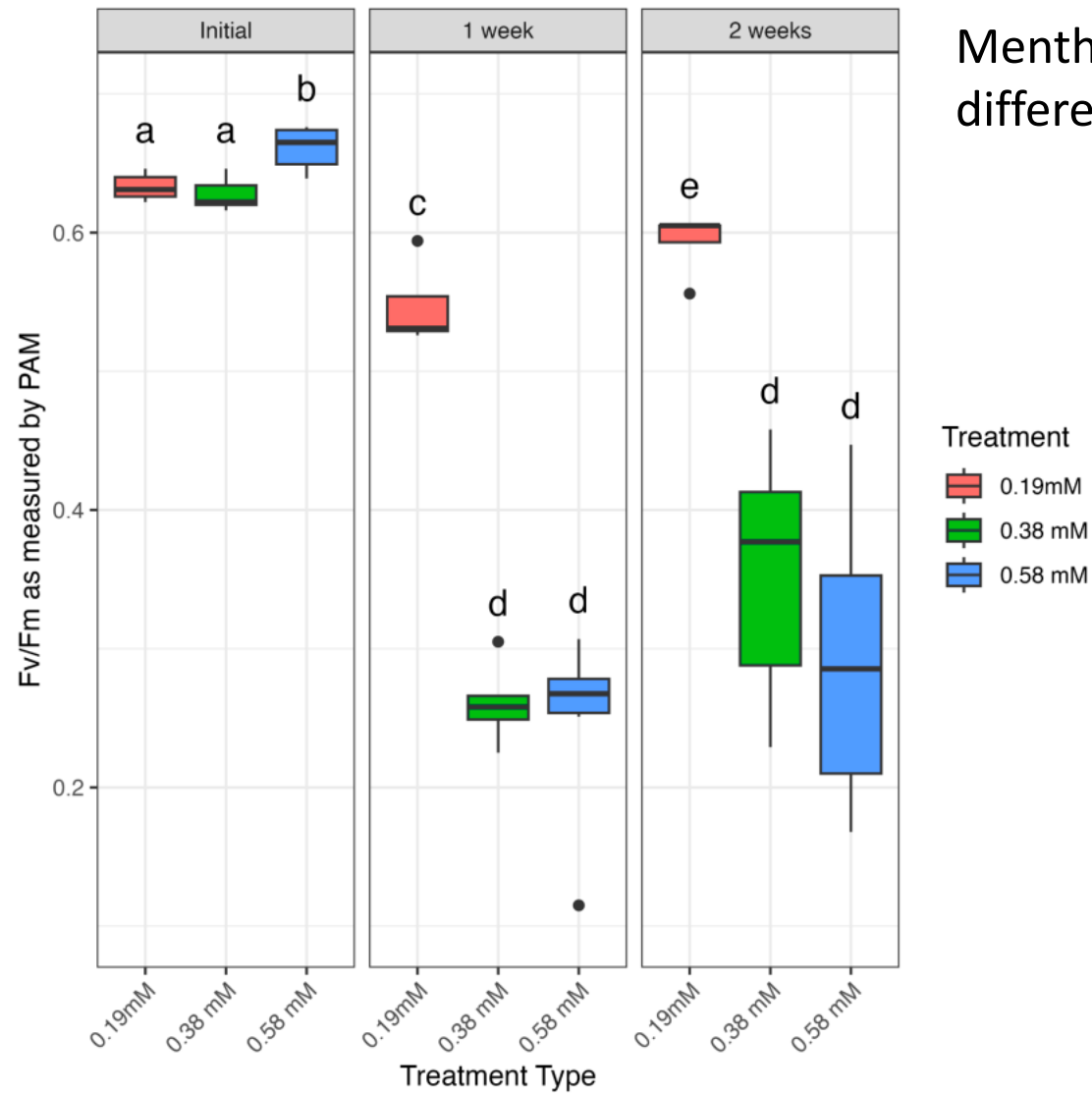


Monitoring Bleaching by CoralWatch Coral Health Chart

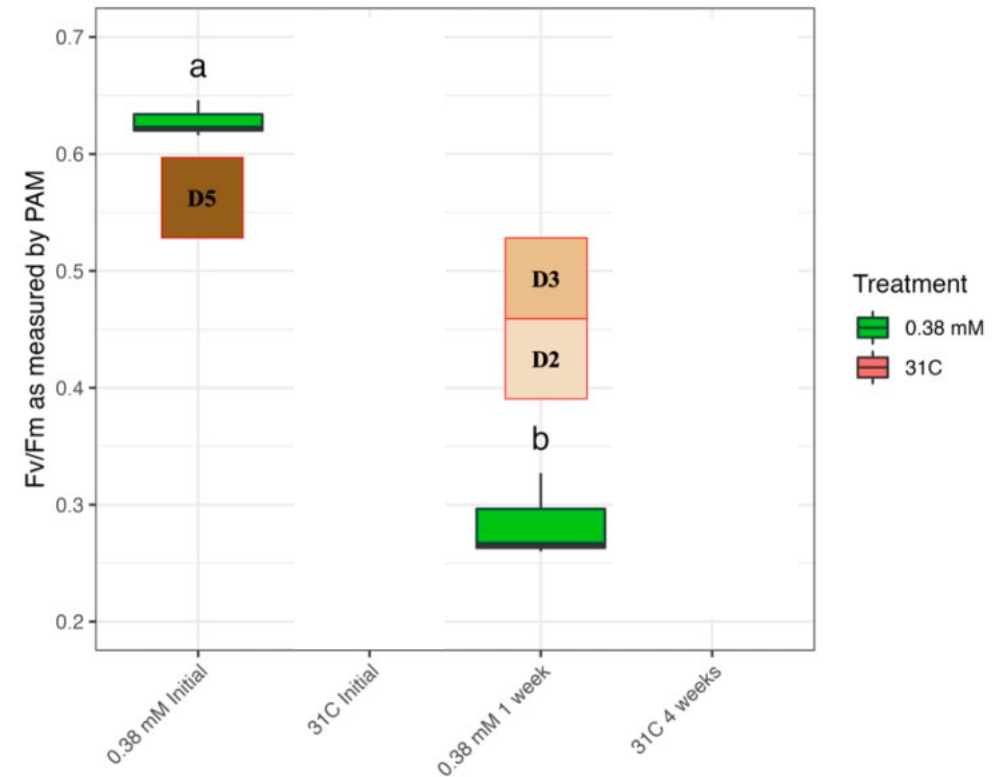


<https://coralwatch.org/product/coral-health-chart/>

Monitoring Bleaching by Photophysiology (Fv/Fm)



Menthol at 0.38 mM and 0.58 mM were not significantly different, but did bleach significantly more than 0.19 mM.



Inoculation of BMCs onto Acer Fragments

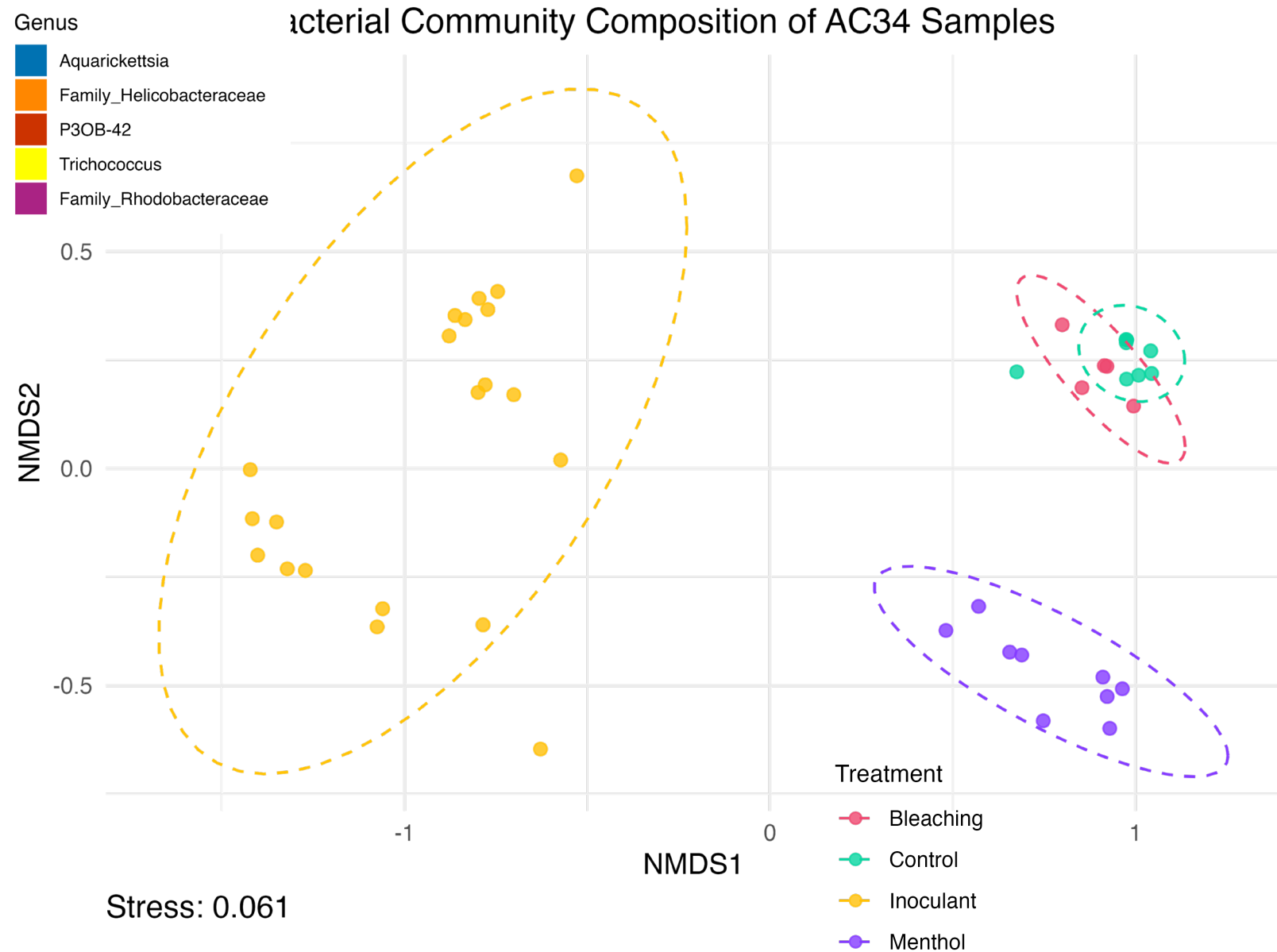
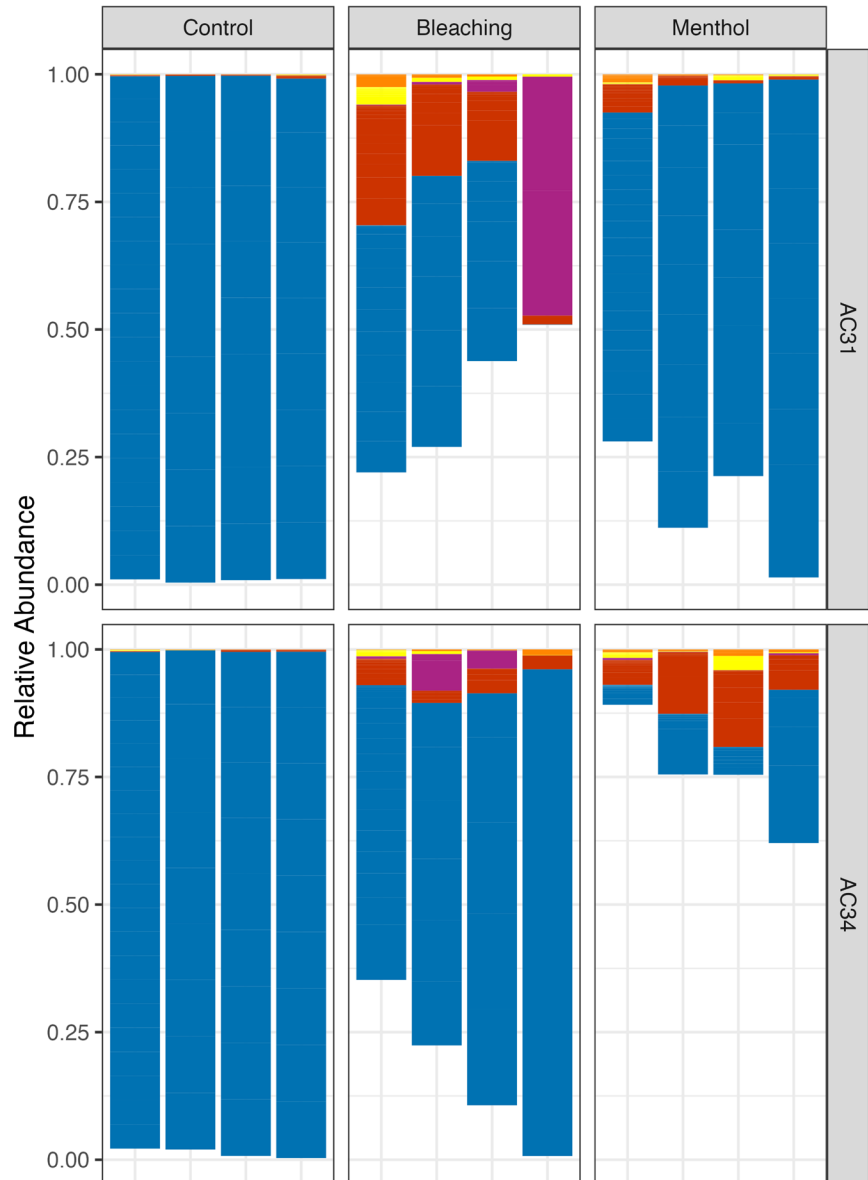


- *A. cervicornis* fragments in aquaria were bleached using menthol or thermal stress.
- Coral food was inoculated with a cocktail of BMCs and then fed to the coral fragments during the first week of the experiment.



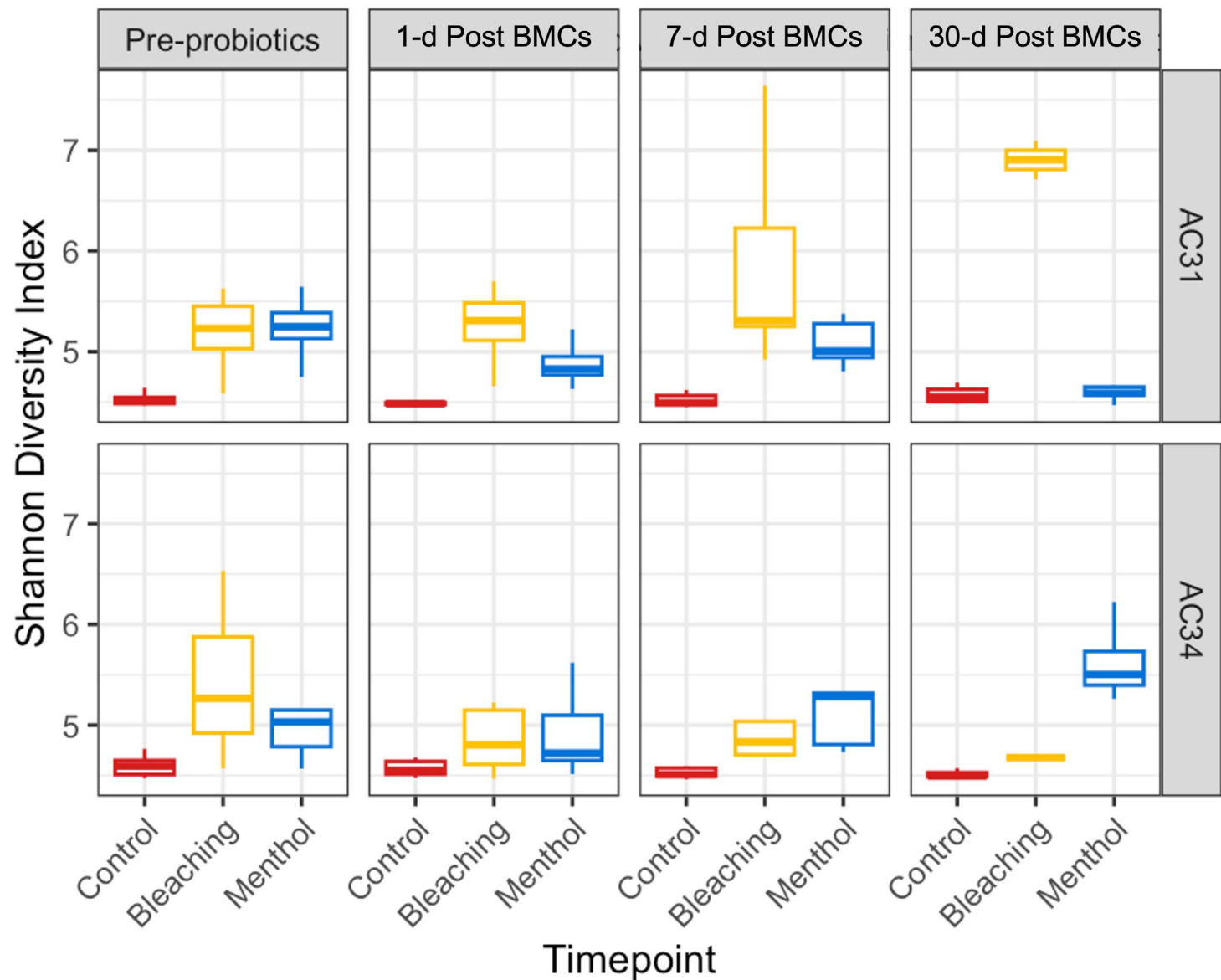
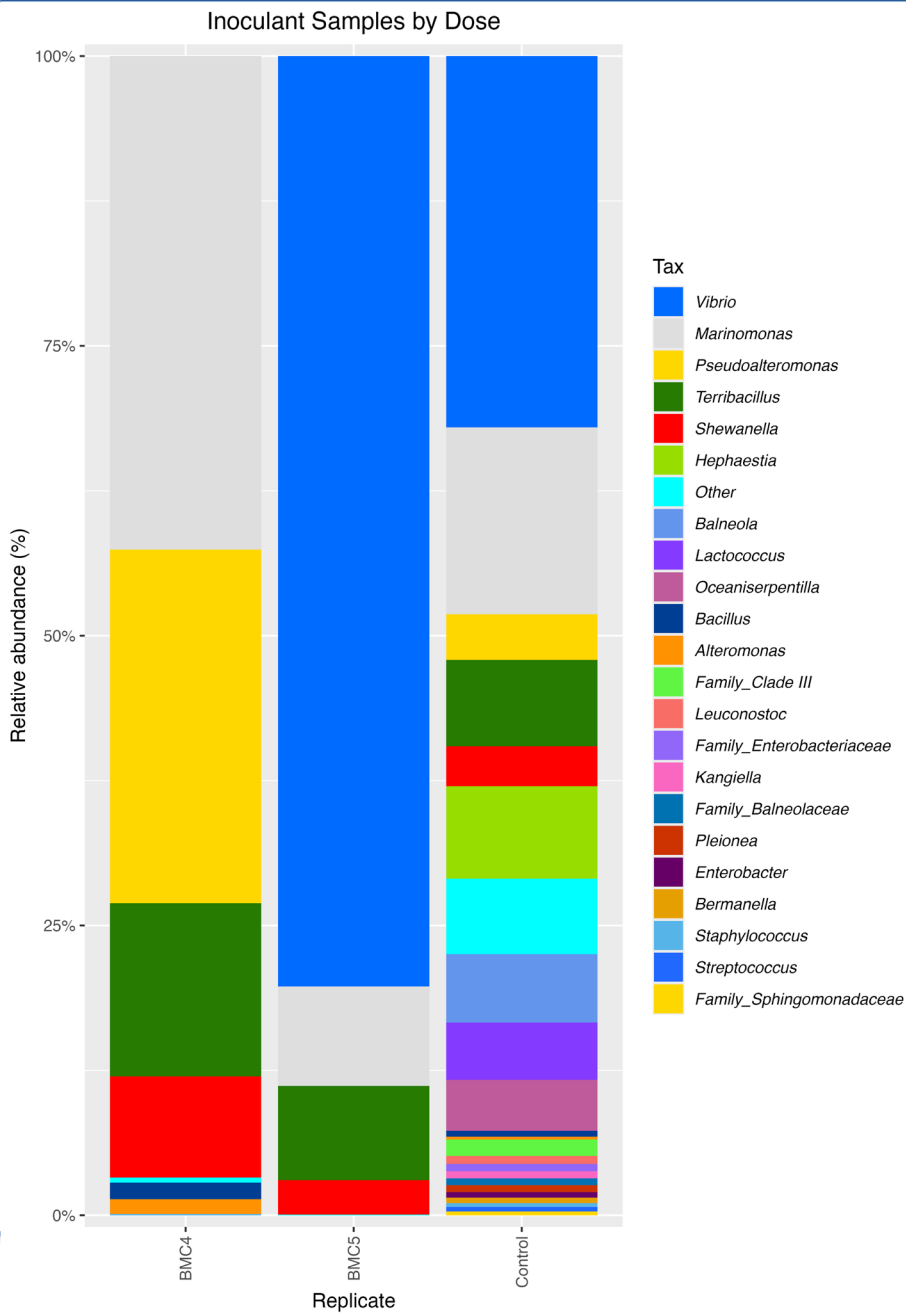
- 16S sequences were obtained using Illumina Sequencing from samples pre-inoculation, and 1 day, 7 days, and 30 days post-inoculation.

Effect of Bleaching and BMCs on Acer Microbiomes



Time →

Effect of Bleaching and BMCs on Acer Microbiomes



Some Final Thoughts...

- Menthol-induced bleaching is a quick and reliable method to bleach genotypes of *Acer* to reduce both algal and bacterial symbionts.
- *Acer* fragments did not robustly take up BMCs in this study.
- Beneficial microorganisms of corals (BMCs) could help to increase resilience of corals to current and future environmental stressors
- Next steps:
 - Optimize application of BMCs on recent sexual recruits
 - Current project underway at Mote's IC2R3

Questions?

