Identifying potential probiotics to enhance *Pseudodiploria clivosa* (knobby brain coral) recruit growth and survivorship

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Introduction

Coral microbiomes, essential to immune support and overall colony health[1], are established during stages of early development for corals. The extraction of bacteria from healthy microbiomes can potentially assist in coral stressor adaptation through Beneficial Microorganisms for Corals (BMCs)[2]. This technique has been proposed to have implications for benefitting growth, controlling pathogens, and providing settlement cues for larvae, yet no studies have investigated how BMCs can impact survival and growth during the earliest life history stages of corals. By introducing bacteria from the microbiome of *Pseudodiploria clivosa* adult corals to their offspring, this study screens bacteria isolate groups for their potential as probiotics that may improve coral recruit survivorship and growth in the laboratory.

Methods

Coral Reproduction & Recruit Rearing

- *P. clivosa* colonies spawned in the land-based assisted sexual reproduction facility @ UNCW's Center for Marine Science
- Gametes were fertilized and the resulting larvae settled on conditioned ceramic tiles.
- Recruits were monitored for survival (n=692) and growth (n=45).
- Tile cleaning and water changes (50%) occurred weekly.

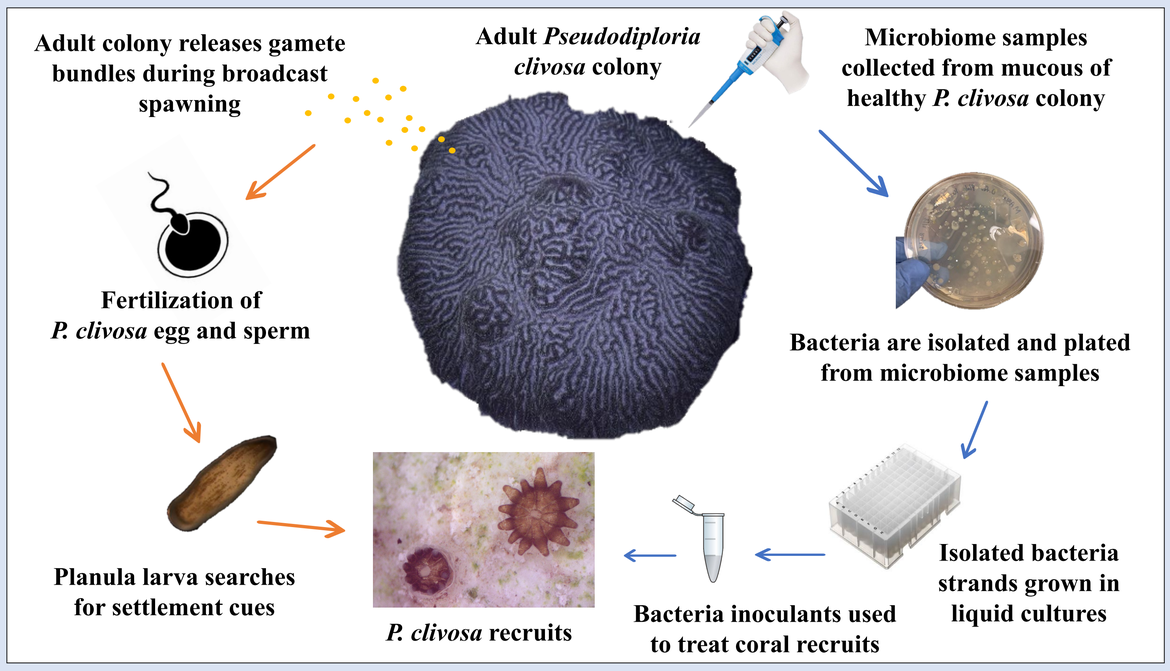

Figure 1. Methodology for recruit rearing and inoculations of bacteria isolated groups cultured from the microbiome assemblage of adult *P. clivosa* reared in the laboratory.

Bacterial Inoculation & Experimental Setup

- 250 bacterial strains isolated from adult *P. clivosa* mucus were divided randomly in 25 groups of 10.
- 18 days post-spawn, recruits were inoculated weekly in glass jars for 24 h.
- Each group was replicated 3 times, while the control of filtered seawater (0.2 µm) was replicated 5 times.
- The experiment lasted 7 weeks.

Results

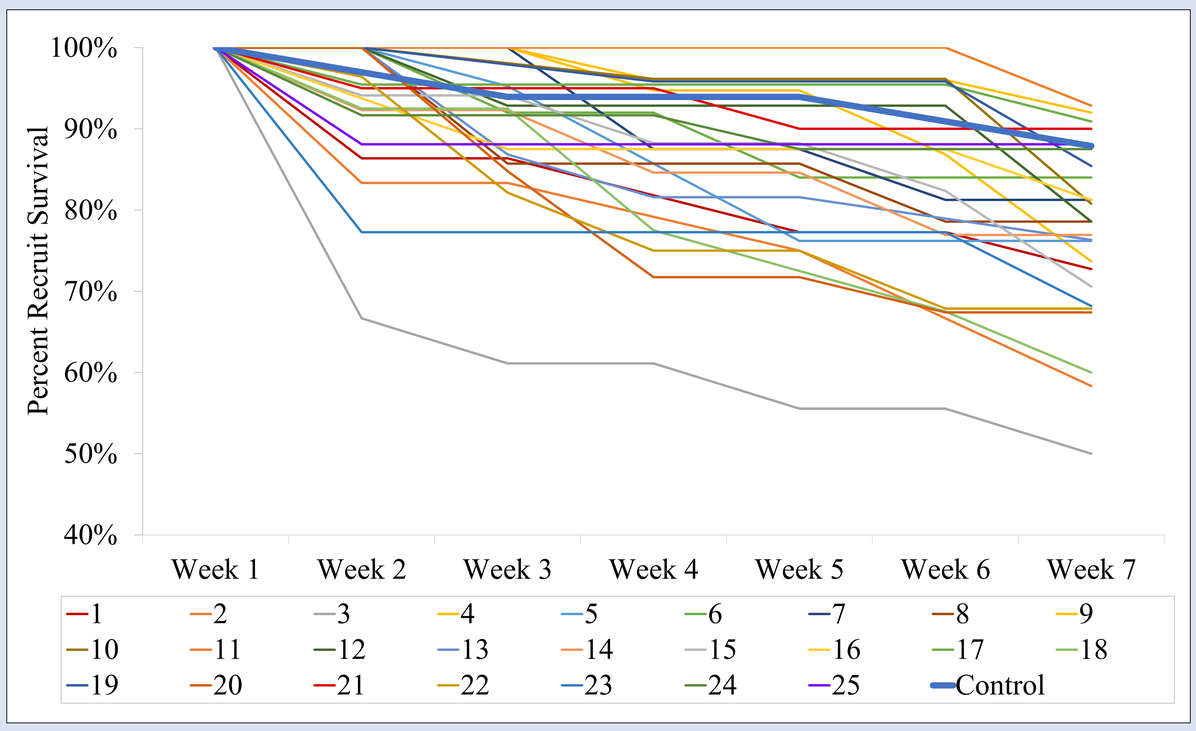

Figure 2. *P. clivosa* recruit survival over time for 25 bacterial isolate groups and controls based on weekly survivorship counts beginning 16-18 days post-spawning.

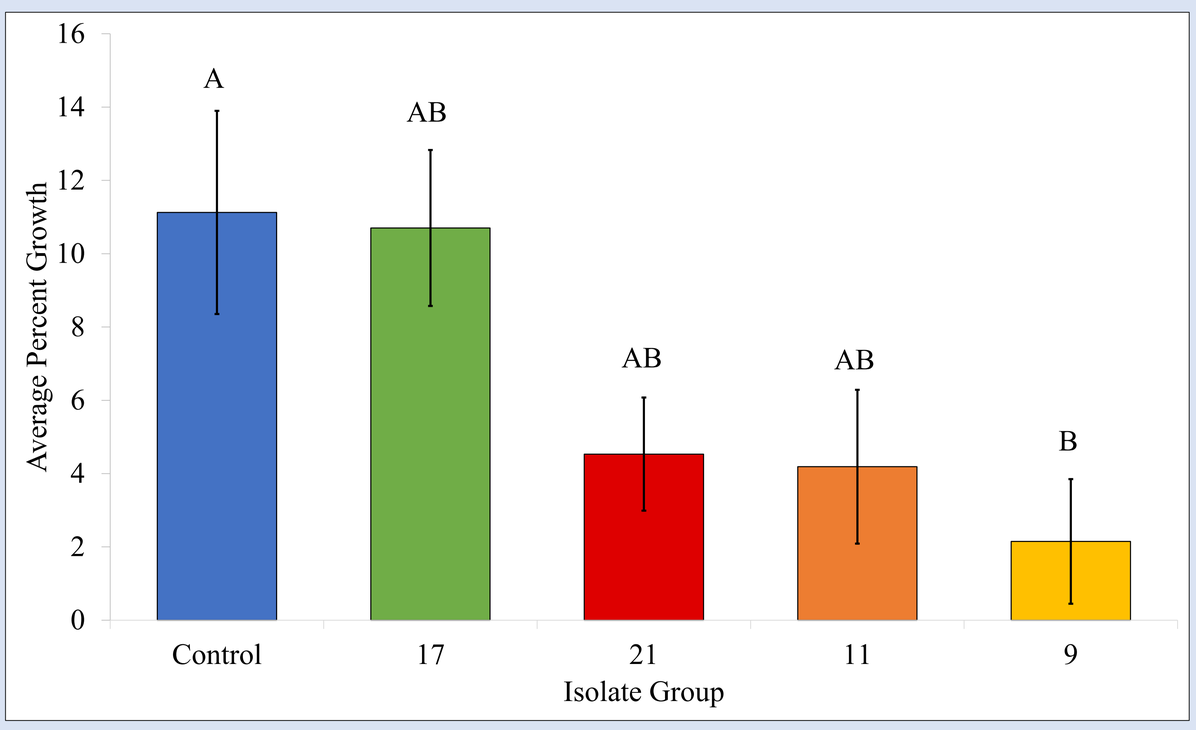

Figure 3. *P. clivosa* recruit growth after 7 weeks of bacteria inoculations for the 4 isolate groups with the highest survival. Different letter denote significant differences (ANOVA, p = 0.0172).

- Of the 25 isolate groups, 4 enhanced survivorship by 2-5% compared to the control; however, this increase was not significantly different from the control (log-rank survival analysis p>0.05, Fig. 2).
- The average percent growth for recruits was not significant between isolate groups, and only group 9 significantly impacted recruits compared to the control, though adversely (Fig. 3).

Conclusions

- Instead of significantly enhancing coral recruit survival and growth, most of the tested bacterial groups reduced recruit survival and growth.
- Analyzing the growth data for the remaining 21 isolates may reveal a bacterial group strain which increases growth.
  - However, it is possible that probiotics or the right bacterial strain for this application have yet to be isolated.

References

1. Damjanovic, K., van Oppen, et al. (2019). Experimental inoculation of coral recruits with marine bacteria indicates scope for microbiome manipulation in *Acropora tenuis* and *Platygyra daedalea*. *Frontiers in microbiology*, 1702.
2. Peixoto, R. S., Sweet, M., et al. (2021). Coral probiotics: premise, promise, prospects. Annual review of animal biosciences, 9, 265-288.

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