Links between the survival of small vascular fragments and varying conditions in a colonial ascidian

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Background

Invasive ascidians commonly foul vessels, aquaculture, and other structures. They often exhibit rapid growth, have broad environmental tolerances, and may experience limited predation. Vulnerability in ascidians exists at a small scale; mortality may be high for newly settled larvae or minute fragments of the vascular system despite the potential for whole-body regeneration (WBR) as established in studies (1, 2, 4, 6, 8). High mortality rates for small individuals raise the question of ecological relevance of WBR in field populations, with management implications during planned removal operations. Few studies of WBR in the field have been carried out to determine what factors might influence WBR success (1). In the lab, studies showed as few as 100-200 blood cells (a single vascular fragment) may regenerate, but this has not been tested in the field (2). Through a lab and field comparison, this study focuses on the survival of minute fragments and the significant effects of fragment size and environmental variation on survival and WBR.

Large vascular fragments of Botryllus diegensis have successfully regenerated in field in central CA and in lab

Field replicates show less success in comparison to lab replicates

Question

Can minute fragments survive under varying conditions and successfully complete WBR in the field?

Objectives

- Determine the environmental components affecting the survival of minute fragments before WBR occurs, particularly temperature and salinity.
- Determine smallest fragment size of ampullae to pose a significant management threat through regeneration.

Methods

- Six adult B. diegensis colonies collected from local bay area docks.
- Clonal replicates tied to glass slides; 2 week attachment period.
- Surgical zooid removal, isolation of ampullae using a dissecting microscope (Fig. 3).
- First replicates in the SFSU wet lab (14 °C, salinity 33). 2mℓ of Kent Phytoplex fed daily.
- Second replicates deployed in SF Bay in cages (Fig 4) at the Eos Seawall in Tiburon CA.
- Temperature recorded hourly with HOBO logger, single time point temperature and salinity measurements taken daily with YSI across 1 °C through 7 days.
- Morphological changes tracked daily for each treatment using 30X dissecting scope.

Figure 1 (right): Filtering水管(circled) regeneration in the lab in a first experiment with large fragments.

Figure 2 (left): WBR progression in field study by Chow et al., 2012. Day 0: small isolated ampullae (circled) regeneration in the lab in a first experiment with large fragments.

Figure 3 (left): Filtering水管(circled) regeneration in the lab in a first experiment with large fragments.

Figure 4 (left): Multiple clonal replicates tied to glass slides, 2 week attachment period.

Figure 6 (above): Lab colonies show longer persistence than field replicates in all four treatments. All fragments died rather than regenerate though varied temperature and salinity may have accelerated field mortality.

Discussion and Future Directions

- Size was determining factor in survival based on comparison of prior lab and field comparison:
  - Botryllus leachi in lab regenerated from single ampulla in 10-14 days (8).
  - B. diegensis fragments > 10 ampullae regenerated in lab no later than 20 day at 11 °C and as early as 8 days at 16 °C (2).
  - Our preliminary lab experiments of larger B. diegensis fragments (all > 23 ampullae) showed frequent regeneration, with the earliest at 7 days for a large fragment of 83 ampullae. In this study, fragments with < 5 ampullae all died in both lab and field.
  - Survival in this case, apparently influenced in field by varied conditions compared to lab:
    - predation and space competition (Fig.7)
    - fluctuations in temp and salinity
  - Remnants of < 5 ampullae may not pose a reasonable threat in management of invasive botryllids, however temperatures were slightly higher than normal ranging from about 15-19 °C.
  - Ongoing genetic barcoding will assist in identification of this cryptic species, allowing delineation of distribution and native region of B. diegensis to facilitate management.

Acknowledgments

Special thanks to Cassandra Lopez, Bing Huey, Darren Gewant, and Cohen Lab members. The 2018 STEM Teacher and Researcher Program and this project made possible through Chevron (www chevron com), the National Marine Sanctuary Foundation (www marinesanctuary org), the California State University Office of the Chancellor, and California Polytechnic State University, in partnership with the Estuary and Ocean Science Center and Biology Department of SFSU.

References