

Metagenomics analysis of the pyloric caeca (gut) microbiome of the sea star, *Patiria miniata*

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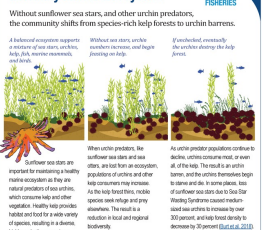
Abstract

Sea stars are keystone species in marine ecosystems, playing a critical environmental role by cleaning the ocean floor through controlling prey populations like mussels and sea urchins. Sea star wasting disease (SSWD) affects multiple sea star species, and causes formation of deadly skin lesions, leading to rapid deterioration and death. While the direct cause of SSWD remains unknown, scientists hypothesize a connection to microbial dysbiosis; since microbiomes affect host health in organisms from insects to humans. Published research indicates spirochetes comprise echinoderm microbiomes and unpublished data the Onorato Lab suggest this is also true for the sea star species, *Patiria miniata*. Therefore, we isolated DNA from pyloric caeca (i.e. gut) of *P. miniata* animals that were visibly wasting and those that did not appear to be wasting. Next generation sequencing was performed to determine microbiome composition using a 16S rRNA analysis. Our preliminary results showed low percentages of Spirochaetae, but higher percentages of Vibrionales in wasting animals. These data, when compared to our previous data from 2014 and 2015, suggest an inverse relationship between Spirochaetae and Vibrionales in the pyloric caeca microbiome of *P. miniata*.

Introduction

Sea stars, also referred to as starfish or asteroids, are star-shaped echinoderms that belong to the class Asteroidea.

Sunflower Sea Stars are Important for Maintaining a Healthy Marine Ecosystem



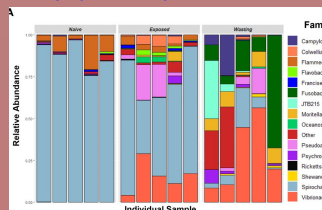
Patiria miniata are detritivores & scavengers help keep ocean healthy by collecting algae & dead animals off ocean floor in addition to eating bivalves

Sea star wasting disease (SSWD) is described as a series of symptoms found in sea stars that typically leads to death including "lethargy and disinterest in nearby prey, pronounced body wall lesions, and arm autotomy" (Oulhen et al., 2022).

Although the first reported observation of SSWD occurred in 1898, the most extreme documented example of this disease occurred in 2013, on the Pacific coast of North America (Oulhen et al., 2022).

Similarly, microbial diversity varies in the different regions of a sea star. Microbiomes are communities of microorganisms that contribute to the health of the host.

Understanding the microbiomes present in different sea stars is incredibly important to fully understand SSWD.



Microbiomes of the sunflower sea star, *Pycnopoda helianthoides*, before (None) and during (Exposed and Wasting) the initial outbreak in Southeast Alaska. Front. Mar. Sci., 16 March 2023 Sec. Marine Biology Volume 10 - 2023 doi.org/10.3389/fmars.2023.1130912

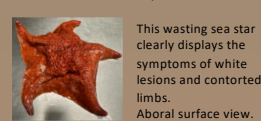
DNA Isolation



March 2024 isolation (3 animals sequenced)
Top images = aboral side; bottom images = oral side; animal order 3PC1, 3PC3, 3PC4 (left to right). 3PC4 appears to be more advanced in SSWD



One of the sea stars sequenced in 2023

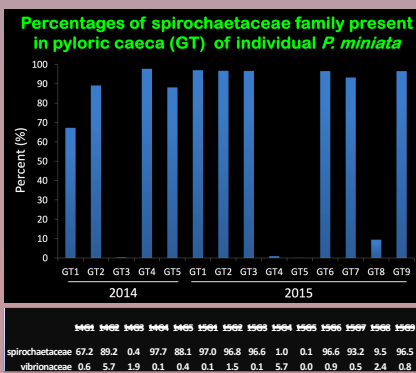
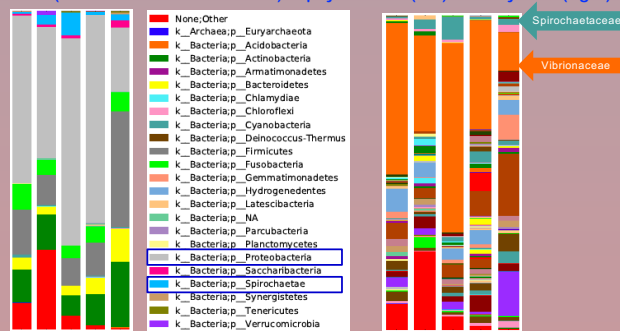


This wasting sea star clearly displays the symptoms of white lesions and contorted limbs. Aboral surface view.

Image of isolated pyloric caeca used DNA isolation

Results

Pyloric caeca microbiome composition of 5 individual wasting *P. miniata* sea stars (animals collected Fall 2022) at phylum level (left) & family level (right)

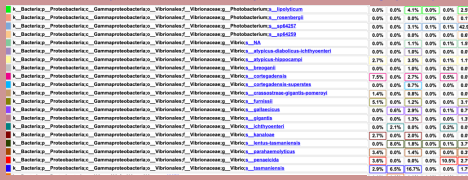


Pyloric caeca microbiome composition at family level of 6 sea stars sequenced in March (3PC) & February (2PC) 2024



Animal 3PC1 looked healthy but may have been in early stages of SSWD

Pyloric caeca microbiome composition at species level of 6 sea stars sequenced in March (3PC) & February (2PC) 2024



Pyloric caeca microbiome vibrio (top table) & spirochete (bottom table) composition at species level of 5 sea stars sequenced in 2023

Tables showing species-level composition for Vibrio and Spirochaetae across 5 sea stars.

Discussion

- Our results from 2014 and 2015 indicate that spirochetes are a major constituent of the pyloric caeca (PC) microbiome of *Patiria miniata*, commonly known as the bat star.
- Our recent data from 2023 and 2024 indicate that *P. miniata* undergoing SSWD have low levels of spirochetes present in the PC.
- Wasting *P. miniata* have higher levels of vibrio bacteria in the PC microbiome than healthy animals (comparison of data from 2014-15 with 2023-24).
- Data suggests an inverse relationship between spirochetes & vibrio in the PC microbiomes of healthy and wasting *P. miniata*.
- Our findings are in accordance with those found for sunflower sea stars (see introduction section in the poster).
- SSWD in *P. miniata* appears to correlate with microbial dysbiosis of the pyloric caeca that involves contrasting changes in spirochetes and vibrio bacteria.
- Future experiments will be conducted to elucidate the relationship between spirochetes and vibrio in the PC microbiome and its impact on SSWD in *P. miniata*

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Acknowledgements

- Pinkerton Foundation
- Zymo Research for their generous philanthropy in providing the isolation kits and sequencing of samples used in 2023 and 2024.
- The faculty of the Urban Barcode Research Program & DNA Learning Center, especially Dr. Allison Mayle
- Wessel Lab at Brown University for providing us with the animals.
- Dr. Thomas M. Onorato, our mentor
- Special thanks to Angela Lee for taking lab photos and assisting in lab activities.

