Potential dolphin pathogens detected at highly variable levels across individual bivalves around San Diego Bay

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Summary

- Bivalves were utilized to examine the relationship between pollutants in the San Diego Bay and pathogens harmful to dolphins.
- Pathogens harmful to dolphins were detected, specifically *P. sordellii* and *C. perfringens*.

Abstract

The health of dolphins in an ecosystem relies heavily on its environment due to various factors like natural feeding cycles, pollution, and climate change. This project aims to find a correlation between ocean pollution and harmful bacteria in bivalves, as well as their potential effects on dolphins through biomagnification. Using DNA extraction and sequencing, two harmful dolphin pathogens were found in the mussels from San Diego Bay, suggesting a biological threat to the dolphins.

Introduction

Hypothesis

H_a: Bivalves in polluted San Diego Bay sites have dolphin pathogens in their gut microbiota.

Pollutants found in San Diego Bay[1]:

- Polychlorinated biphenyls (PCBs)
- Polycyclic aromatic hydrocarbons (PAHs)
- Metals (Mercury, Lead, Cadmium)

Using Bivalves to Study Water Quality:

- Mussels and oysters filter food from the water (Figure 1)
- Bioaccumulate contaminants from the environment[2], and can cause biomagnification[3]

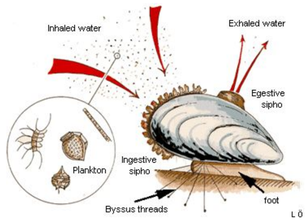

Figure 1. Diagram of a mussel's filter feeding process[4]

Goals:

- Observe the effect of pollution on the pathogens in San Diego Bay
- Investigate potential threats to dolphin health

Materials & Methods

Sample Collection

- 15 mussels from 5 sites (three replicates per) were collected from the U.S. Navy Marine Mammal Program Facility in the San Diego Bay
- 8 oysters were collected from 4 sites (two replicates per) around San Diego Bay

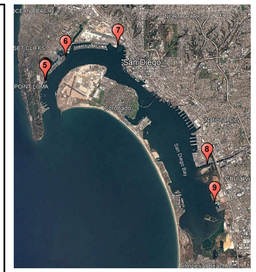

Figure 2. Google Earth image of the 9 sampling sites. Sites 1-5 are clustered together under the Site 5 label.

Sample Processing

- Mussels and oysters were dissected to isolate digestive organs
- Saponin and Turbo DNase were used to deplete host (mussel and oyster) DNA

Chemical Analysis

- Chemicals will be extracted from composite integrative passive samplers (CIPS) to detect chemical pollutants

DNA Extraction

- Powerfood™ Microbial DNA Isolation Kit

Quality Check

- Polymerase Chain Reaction (PCR) followed by agarose gel electrophoresis

Nanopore 16S Library Preparation and Sequencing

- PCR to amplify the 16S rRNA gene V4 region
- MinION sequencer

EPI2ME R Studio®

Data Analysis

- Sequencing reads were processed with EPI2ME
- Bacterial data were processed in RStudio

Results

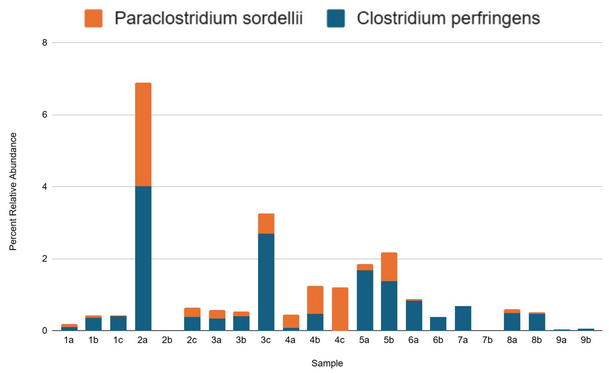

Figure 3. Relative abundance of *Clostridium perfringens* and *Paraclostridium sordellii* across all samples.

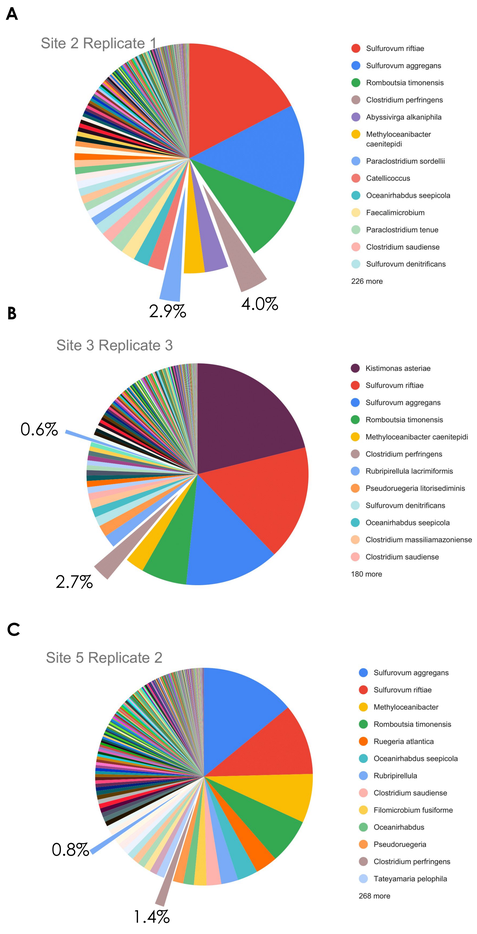

Figure 4. The three mussel samples with the highest abundance of *C. perfringens and P. sordelli.* Mussels were collected from three different locations: (a) Site 2, (b) Site 3, (c) Site 5.

Conclusion & Discussion

Dolphin pathogens were detected across most of the sampled bivalves (Figure 3).

- *Clostridium perfringens*
  - If it enters the bloodstream through cuts, animals can get gas gangrene, a condition where gas builds up inside tissues[6,7,8]
  - Produce toxins that cause death of blood cells and tissues[7]
- *Paraclostridium sordellii* (formerly known as *Clostridium sordellii*)
  - Releases toxins that can induce toxic shock syndrome[9]
  - Contributes to septicemic infections (blood poisoning)[10]
- The highest relative abundance of *Clostridium perfringens* and *Paraclostridium sordellii* was detected at Site 2 (Figure 4).
  - The pathogen abundance levels are relatively low compared to other bacteria

Study Limitations and Future Directions

- Water at the same sites was sampled using Composite Integrative Passive Samplers (CIPS)
  - Both hydrophobic and hydrophilic compounds bind to CIPS, which enables the quantification of organic pollutants
  - Chemical analysis could help identify associations between pollution and bacterial levels
- Sampling was only performed during one season; therefore, seasonal variation in pollution patterns was not accounted for.
  - Routine sampling is recommended to effectively monitor the dynamic coastal water environment.
- Similarly, sampling bivalves across a wider range outside of the San Diego Bay area could reveal site-specific variations in bacterial composition

References

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