Analysis of Plastics in American Oysters (*Haematopus Palliatus*)

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Abstract

- General about plastics
- MP in oysters
- General methods
- Major results
    - Total MP and MF
    - Which sample had the most MP
    - Trends?
- Why is matters

Introduction

- Oysters are a great bioindicator for monitoring plastic pollution because they are filter feeders.
    - Sentinel species: they will always be affected by the pollution. Therefore, oysters are sensitive.
- About Snow Island Oysters
    - 2 farms locations
- The Snow Island Oysters give us a good gauge about the concentration of microplastics in the Quahog Bay.

Methods

Instruments used

Four 250 mL beakers
Four safety goggles
Latex Gloves
INCU-Shaker Mini
Rocker 300 Vacuum Pump
1 liter vacuum trap
SAS Positive Pressure room (clean room)
Two pyrex glass bowls
1000 mL flask
Metal clamp
53 µM Sieve
Vacuum filter reservoir
Vacuum filter funnel
Air Science Purair Ductless Fume Hood (negative pressure chemical hood)

Knife
1.0 M sodium hydroxide
Four Cytiva Whatman GF/F 47 mm filters
Zeiss Stemi 305 Microscope- brand
Petri dishes
Tin foil
Four 150 mm sterile Petri Dishes with covers
Stainless Steel dissection tray
Ohaus Adventurer Scale

Collection of Oyster Samples

The American Oysters were harvested on Dog's Head, North of Orr's Island in Harpswell, ME (Figure 1). Four oysters were selected ranging in size: small, medium, large, and jumbo. All oyster samples (1-4) were put on a small steel dissection tray, brought to the lab, put in the clean room, where they were shucked (Figure 2).

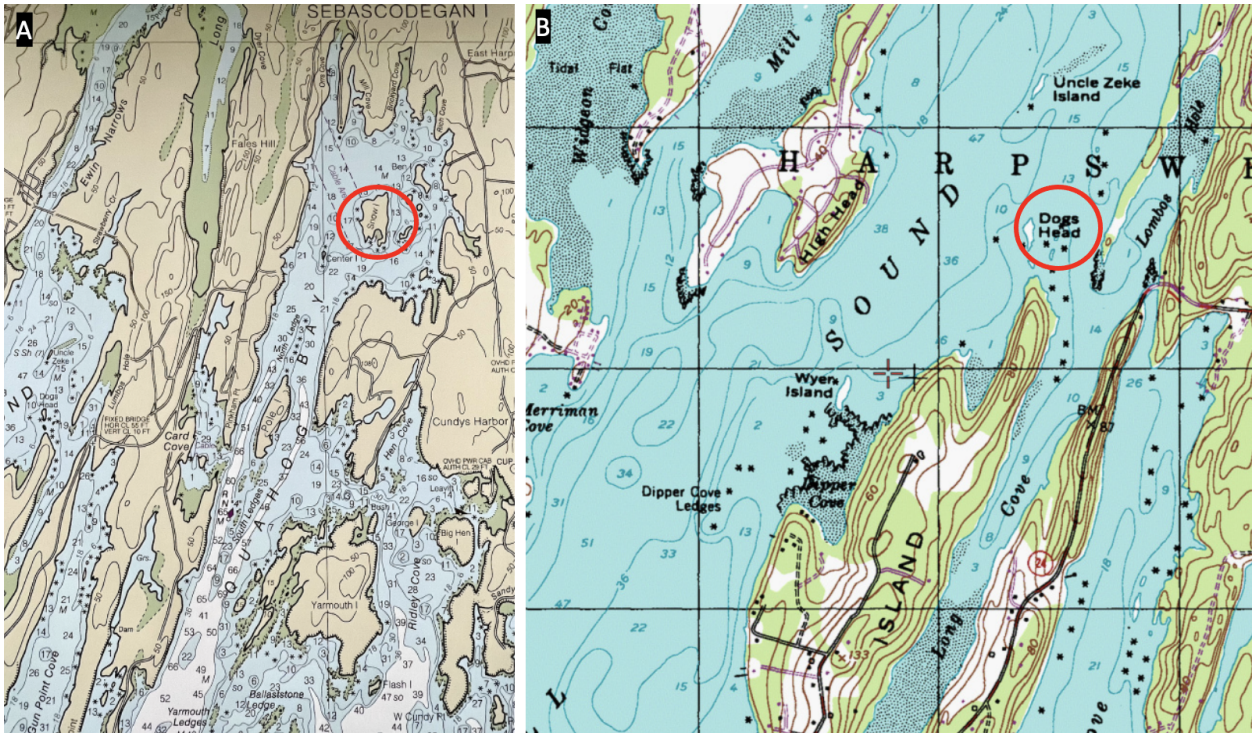

Figure 1: Topographic map of Quahog Bay (A) including Snow Island (Snow I, circled in red) and Harpswell Sound (B) showcasing Dogs Head (circled in red).

Figure 2: Four oyster sampling ranging in size from small to jumbo on the dissection tray, in the clean room with deionized water squirter, and four 250 mL beakers (A). Shucking the four oysters in the clean room and placing them in their respective beakers (B).

Process of Disintegrating Oyster Samples

Before putting the samples into the 250 ml beakers, first the beakers were labeled 1-4, and the dry weight of each beaker was weighed. Gloves were worn for all processes to reduce contamination of microplastics and fibers in the samples. Once dry weight was weighed, the oyster samples were placed into the labeled beakers and weighed to determine the sample wet weight in grams. Once the dry beaker weights and sample wet weights were collected, the wet weight was subtracted from the dry weight to determine the calculated sample weights. Sample 1 weighed 5.724g, sample 2 weighed 7.499g, sample 3 weighed 14.724g, and sample 4 weighed in at 20.583g (Table 1). Samples were then brought into the chemical hood. 150ml of 1.0 mol sodium hydroxide was poured into all four of the 250ml beakers that the samples were in. Samples were then put into the INCU-Shaker Mini at 60℃ at 125 rpm for 3 days.

Table 1: Measured wet weight of each sample and dry weight of beaker to determine the calculated sample weight (g).

Filtration of Oyster Samples- edit out Tuna parts

Once the samples dissolved for 72.5 hours, each sample was taken out of the shaker and put into the SAS Positive Pressure Room to prepare for filtration. All glassware and tools were rinsed with filtered deionized (DI) water prior to experiment. Each sample was individually poured into a 55 sieve with a bowl underneath to filter out all organic matter and other unnecessary juices. The sieve was then turned over to be backwashed with DI water into a separate bowl. Once the sieve was completely backwashed and all material was into the bowl, each individual sample was carefully poured into a new, sterilized 250ml beaker. Samples were

then poured into the vacuum filter reservoir, with a 47mm filter between the reservoir and the vacuum filter funnel connected by a metal clamp, which flowed into a 1000ml Erlenmeyer Flask. The Rocker 300 vacuum pump was connected to a 1 liter jug to collect all excess water from the flask by a tube. The vacuum pump was also connected to the flask itself by using two tubes. Once all samples were filtered, the filters were carefully placed into small tin holders with tweezers and put in labeled, sterile protection cases.

Microscopic Counting of Microplastics and Fibers

To view microplastics and microfibers, each sample was individually taken out of its case and placed in a petri dish to be put under the Stemi 305 Microscope. 9 to 12 images were taken of each sample filter to locate all MP and MF. All microscope images were analyzed on our laptops. To identify and count MF, all thin hair-like pieces throughout all the images were considered. For MP, they were considered to be any relatively dark or shiny reflective blob. Only the pieces that had body to them were counted, not the nanometer sized dots. Additionally, the brownish-green pieces were not counted as MP due to them most likely being organic matter that did not have enough time to dissolve in the sodium hydroxide. Some fish bones and algae were identified in the samples as well which were not counted.

Results

- Sample 1 microfibers ranging from 3 mm - 22 mm in size.
- Sample 2 MFs ranging 3 mm - 45 mm
- Sample MFs ranging 3 mm - 10mm

Oyster Size	Sample Number	Number of Microfiber	Number of Microplastics	Average Microfiber Length (mm)	Average Microplastic Length (mm)
Small	1				
Medium	2				
Large	3				
Jumbo	4				

Table 2: Number of microplastics and microfibers in each sample (1-4) with an estimated average length for each.

Oyster Size	Sample Number	Microfibers per gram	Microplastics per gram
Small	1	7.86163522	31.27183788
Medium	2	4.667288972	9.601280171
Large	3	0.7470795979	6.112469438

Jumbo	4	0.6315891755	1.846183744

Table 3: Calculated number of microfibers and microplastics per gram. The number of microplastics in each sample was divided by the sample weight in grams.

Figure 3: Microplastics images from Semi 305 Microscope on 4.0 zoom from samples 1 (A) and 2 (B & C).

Discussion

Conclusion

References

https://www.topoquest.com/map.php?lat=43.79178&lon=-69.95969&datum=nad83&zoom=2&map=auto&coord=d&mode=zoomin&size=m&cross=on (HS topographic map)