

The background of the entire image is a close-up, high-angle shot of a large pile of multi-colored plastic debris, including small fragments, bottle caps, and unidentifiable pieces of plastic, scattered across a dark, reflective surface. The lighting creates bright highlights and soft shadows on the plastic pieces, emphasizing their texture and the sheer volume of waste. The overall color palette is dominated by the various colors of the plastic (blues, greens, yellows, oranges, reds, purples) against the dark, muted background.

# 7TH ANNUAL TEXAS PLASTIC POLLUTION SYMPOSIUM

**THE HOUSTON ZOO**  
- **APRIL 3, 2025** -

[www.TexasPlasticPollutionSymposium.com](http://www.TexasPlasticPollutionSymposium.com)



# Poster Titles & Presenters

The primary poster session for this symposium is scheduled from 12:45 – 1:45 p.m. on Thursday, April 3, 2025, in the education classrooms within the Brown Education Center at the Houston Zoo. The poster authors will be present during this time to answer questions and discuss their projects.

For those attending the meeting virtually: You will receive an email following the meeting containing PDF copies of the posters that were provided by the authors. Check them out, and feel free to reach out to the authors with any questions or comments you may have.

## **Aerial Invasion: Atmospheric Deposition of Microplastics in Mosquito Lagoon, Florida**

Madison Serrate\*, Tanillesse Gonzalez, Stephanie Fletcher, Paul Sacks, Joshua Fnu, Sara Kim, Lei Zhai, Abby Frey, Julia Kruger, Tara Blanchard, Emily Hays, Linda Walters

## **Analysis of Microplastic Concentrations in Dried Algae Mats and Sediment Collected from Detention Basins in the Edwards Aquifer Recharge Zone**

Paulina Quinonez\*§, Andre Felton, Jeffery Hutchinson

## **Digestion of Polyethylene Terephthalate Fibers by *Zophobas morio* Larvae**

Isabel Li\*§, Dr. Kasia J. Dinkeloo

## **Dynamics of Marine Litter Post-Hurricane Beryl: Assessing the Ultimate Fate of Flotsam**

William Bailey\*§, David Mohrig, Cornel Olariu, Kutalmis Saylam

## **Effects of PET Microfiber Exposure on Mating Behavior, Foraging Behavior, and Problem Solving in *Gambusia affinis***

Adrienne Lihou\*§, Rivers Hartzell\*, and Jing Graber\*

## **Examining the Plastic Degrading Potential of Marine Fungi Found on the Texas Coast**

Jaden Acevedo\*§, Kristen Garsaud\*, Dr. Kasia Dinkeloo

## **Fluorescent Detection of Nile Red-stained Microplastic Uptake in the Roots of *Arabidopsis thaliana***

Kailyn Nonhof\*§, Jing Graber, Kasia Dinkeloo

## **Influence of Microplastics on Sediment Transport Dynamics**

Marufa A. Upoma, Min Y. Pack\*

## **Making Space for Migratory Birds: An Urban Conservation Program Highlight**

Kiara Carrasco\*§, Chloe Dannenfelser, Liz Virgl, Nancy Brown

\* Indicates presenter

§ Indicates student presentation

**Microbial Marvels: Investigating Dubia Roach Microbiota in Relation to Polyethylene Biodegradation**

Roland Quinones\*§, Kasia Dinkeloo

**Non-Plastic Solutions for Oyster Reef Restoration: Efficacy and Environmental Impacts of Novel Restoration Materials**

Cara Womacks\*§, Madison Serrate, Otis Woolfolk, Fnu Joshua, Lei Zhai, Paul Sacks, and Linda Walters.

**Plastic-Free Restored Habitats: Reducing Plastic Pollution in Community-Based Restoration of Oyster Reefs**

Dr. Jennifer Beseres Pollack, Dr. Linda Walters, Dr. Lisa Chambers, Jace Tunnell, Dr. Zhanfei Liu, Dr. Terry Palmer, Natasha Breaux, Erin Hill, Mckenna Reinsch\*

**The Nurdleome: Identification and Characterization of Microbes Found On Gulf Coast Nurdles**

Vibha Annaswamy\*§, Kasia Dinkeloo

**Threads of Change: Zooplankton Community Shifts in Response to Fiber Disturbances**

Caitlyn Lankford, Heaven Thompson, Ashton Fisher, Addison Lehew, Dr. Mary Kay Johnston

\* Indicates presenter

§ Indicates student presentation



## Background

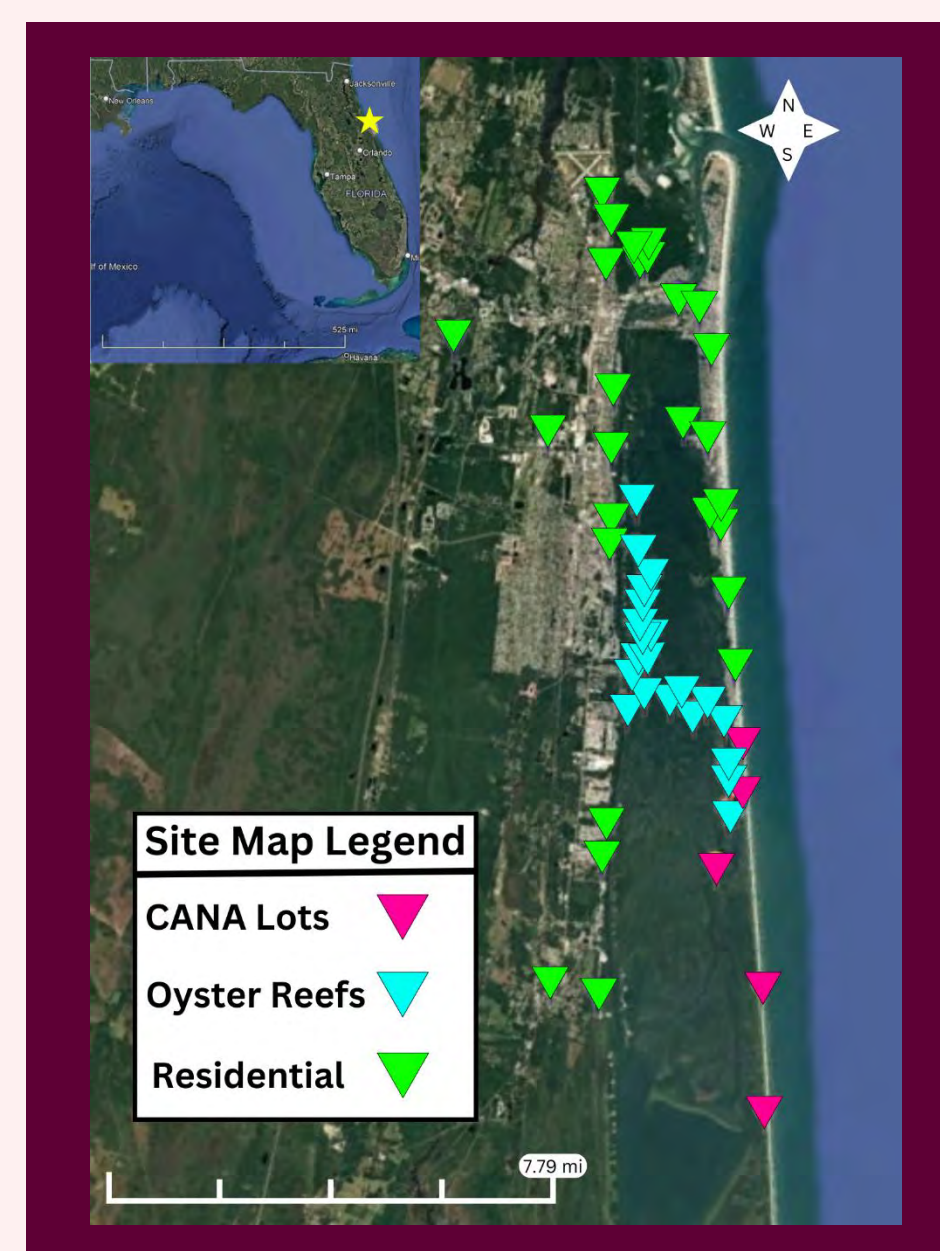
- Microplastics (MP, < 5 mm) have been found in remote regions, suggesting atmospheric deposition (AD) facilitates long-distance transportation<sup>1</sup>
- For example, in the Pyrenees Mountains, 365 ± 69 MP m<sup>-2</sup>d<sup>-1</sup> were found in AD samples<sup>2</sup>
- A 2021 study found that the Indian River Lagoon (IRL) in Florida is a microplastic hotspot, with a mean density (± SE) of 1.47 (± 0.05) MP per liter of lagoon water<sup>3</sup>
- The role of AD in MP abundance around the Indian River Lagoon and the surrounding communities is unknown

## Objectives

- Assess whether MP abundance varies between site types (CANA parking lots, intertidal oyster reefs, residential)
- Evaluate potential differences in MP characteristics between site types

## Field Methods

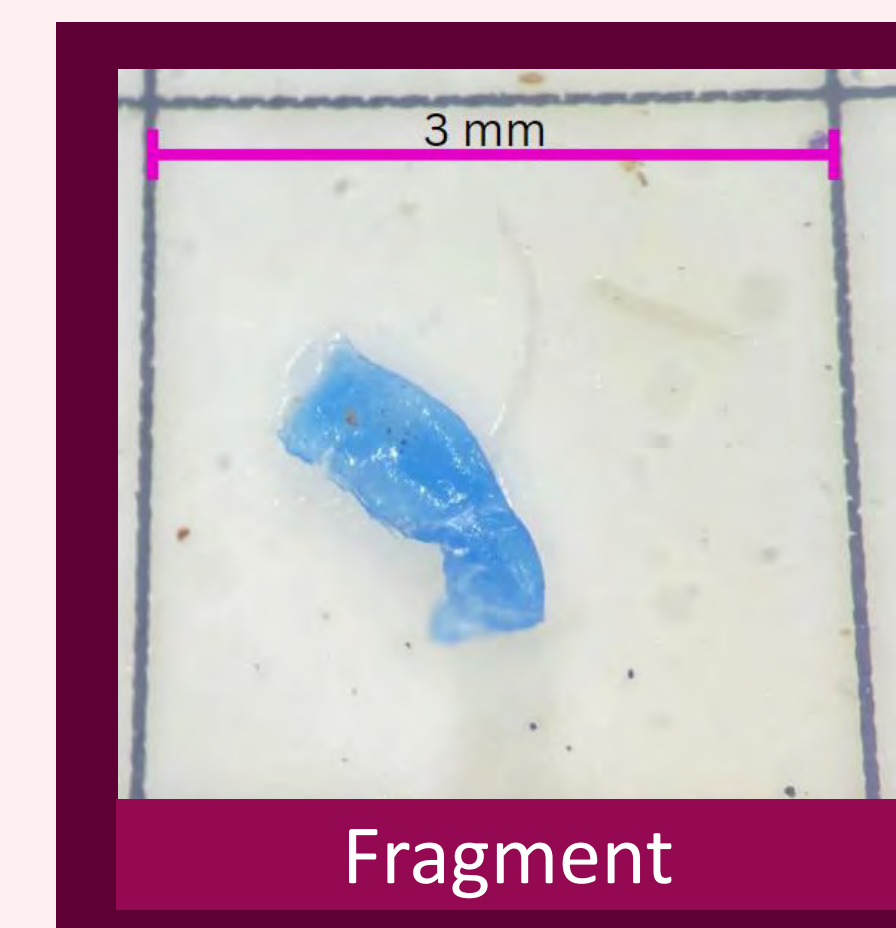
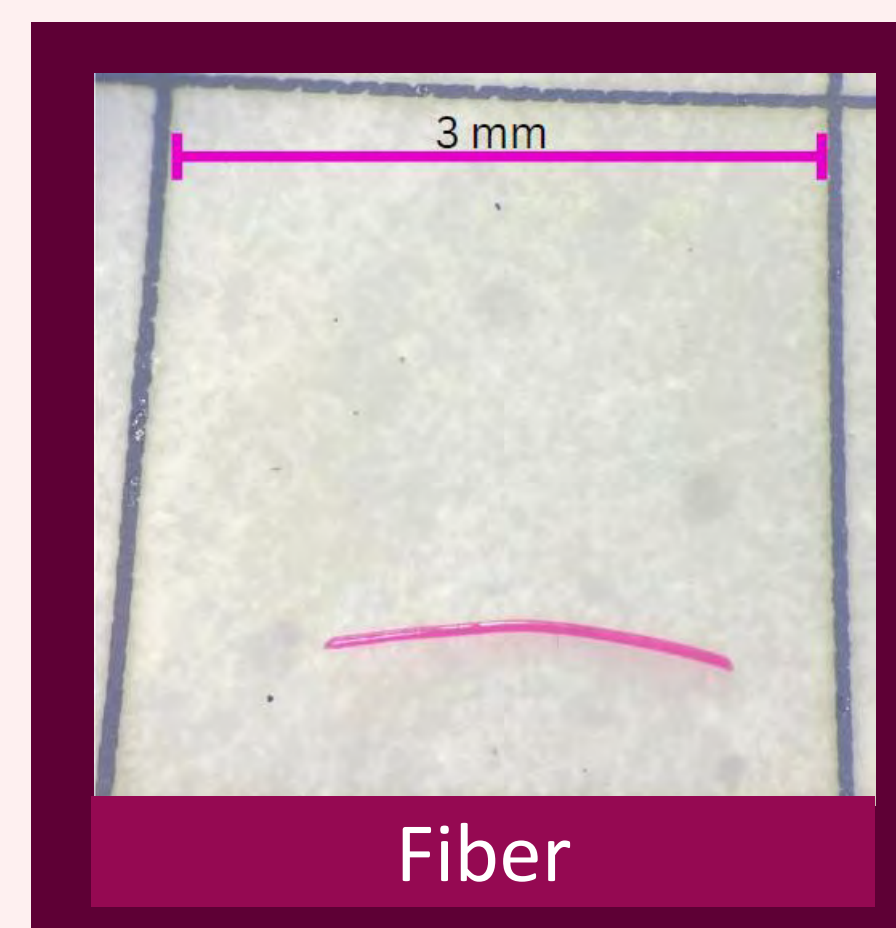
- Collection jars are placed around the IRL at 3 site types: Canaveral National Seashore parking lots (CANA lots), oyster reefs, and residential sites
- Each sample set contains 3 replicate glass jars (1000 mL jar) containing 10 mL of filtered (0.45 µm) deionized (DI) water to trap particle deposits
- Samples are left at the site for 3 hours to collect particle deposits before being sealed and transported back to the lab for processing



## Methods

### Laboratory Methods

- To remove particles from the jars, each was rinsed 3 times with 200 mL filtered (0.45 µm) DI water and filtered three times through a vacuum-pump filtration system using 0.45 µm gridded filter paper
- A 40x microscope is used to examine filter paper contents and the size, color, morphology (fiber, fragment, film, foam, bead, pellet), and suspected material type is recorded for each particle
- To account for aerial contamination, 5 blanks with filter papers dampened with filtered (0.45 µm) DI water are set around each microscope during microscopy processing
- After samples are processed, blanks are examined the same way
- Fourier transform infrared spectroscopy (FTIR) is used for definitive determination of suspected MPs

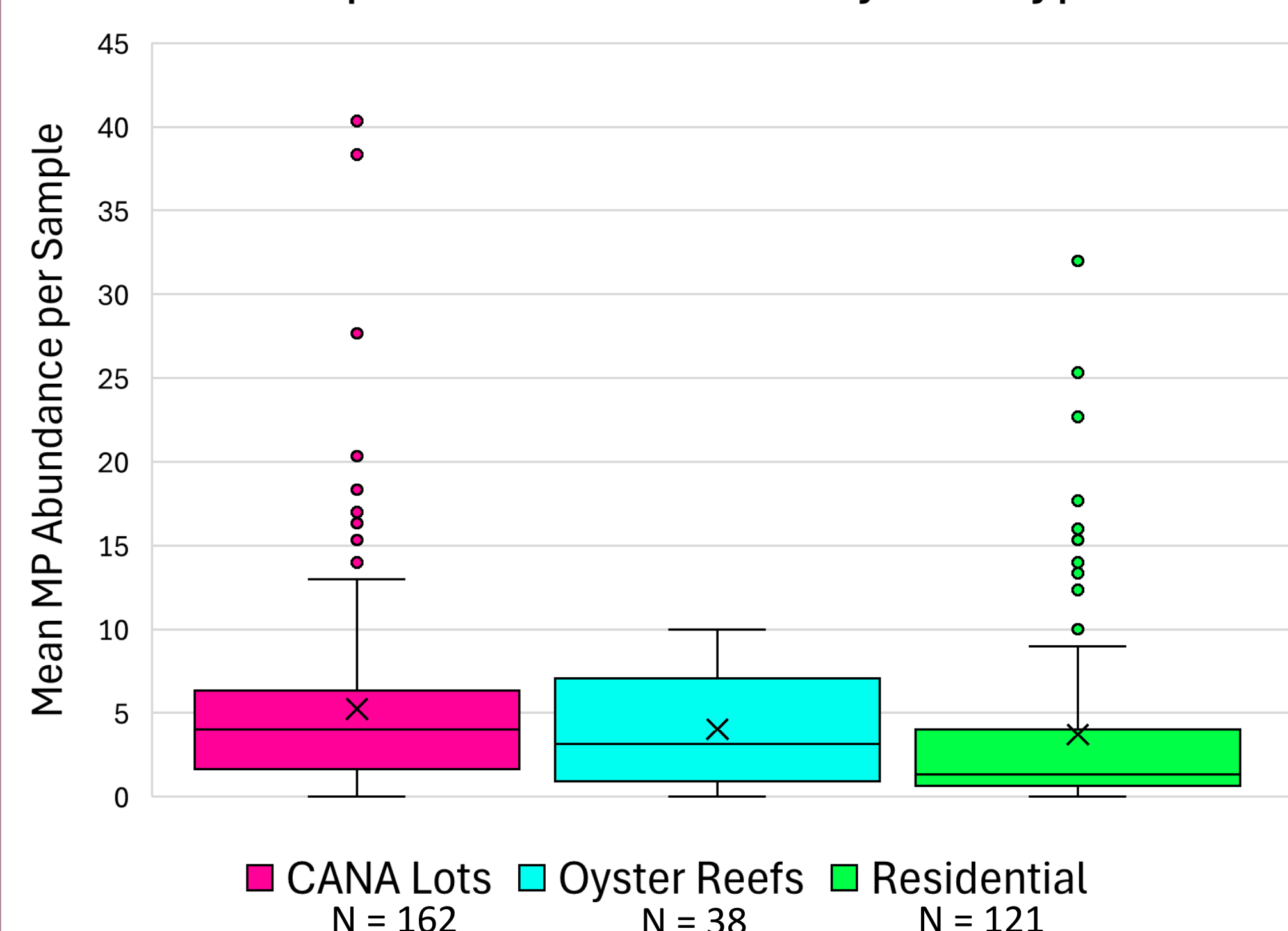


## Discussion

- CANA lot and oyster reef samples are in areas accessible to park visitors, which may contribute to the higher mean MP abundance compared to residential sites
- Several residential samples had very high mean MP abundance, potentially due to nearby home construction
- The mean length (± SE) of MPs across all site types (4.26 ± 8.43 mm) was larger than the mean length of MP found in 2021 in lagoon water (1.94 ± 0.13 mm)<sup>1</sup>, suggesting other MP sources are important in and around the IRL
- The presence of Teflon™ highlights the need to consider MP sources beyond the commonly studied MPs (PET, PP, PE etc.)
- Inhaling MPs serves as a pathway for bioaccumulation, a recent study found MP concentrations of 3345 µg g<sup>-1</sup> in human brain tissue samples<sup>4</sup>
- This study will help policymakers consider strategies that will aid in reducing microplastic pollution by limiting certain single-use plastics (e.g., Styrofoam™)

## Preliminary Results

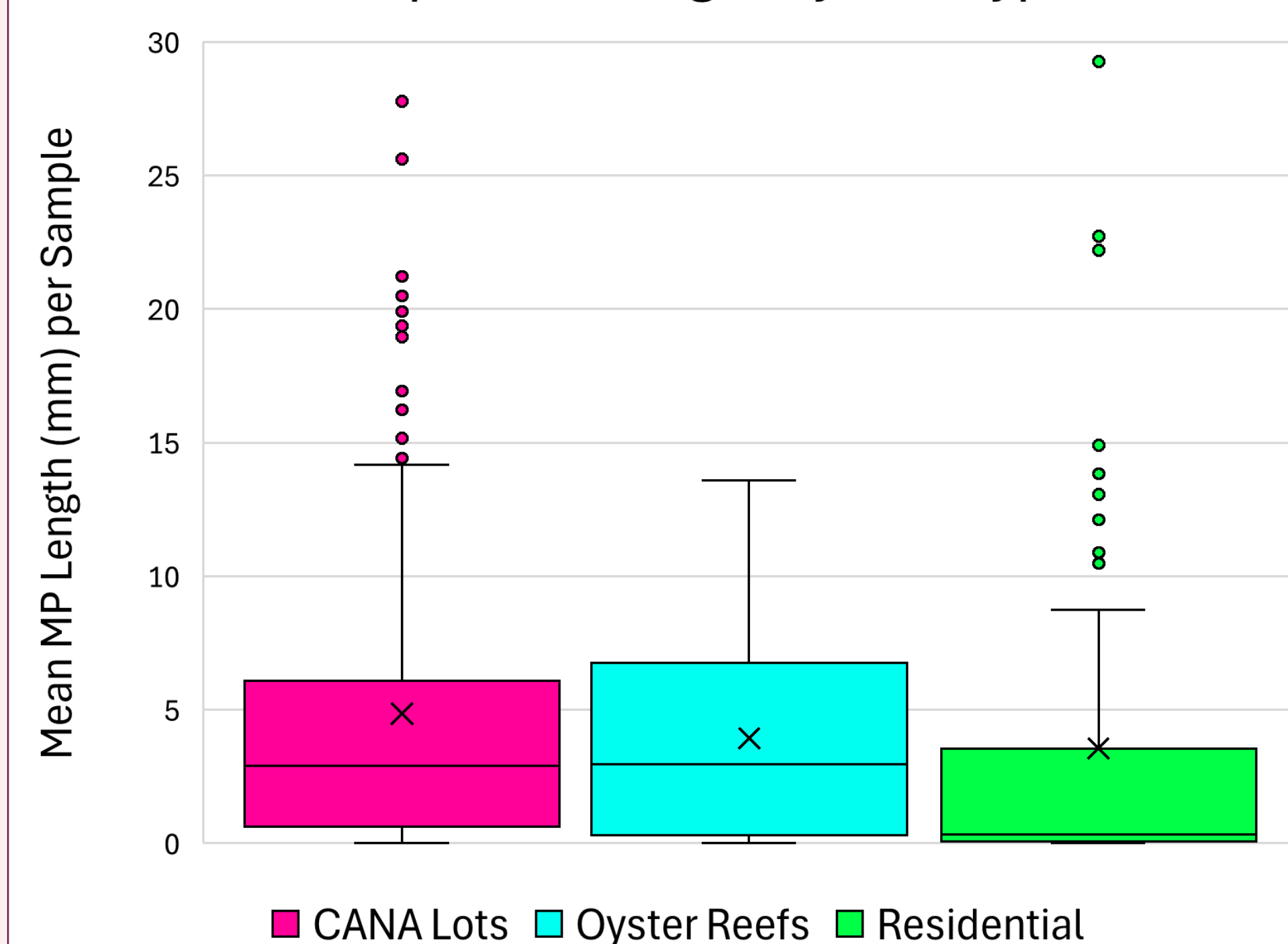
Microplastic Abundance by Site Type



### Abundance

- Kruskal-Wallis Test: There was a significant difference in mean MP abundance between site types ( $p = 1.70 \times 10^{-5}$ )
- CANA lots and oyster reefs had the highest mean MP abundance with a mean of 5.2 and 4.0 MPs per sample, respectively
- Contamination Rate:** Blanks had an aerial MP contamination rate of 0.00006688 MP/minute during processing

Microplastic Length by Site Type



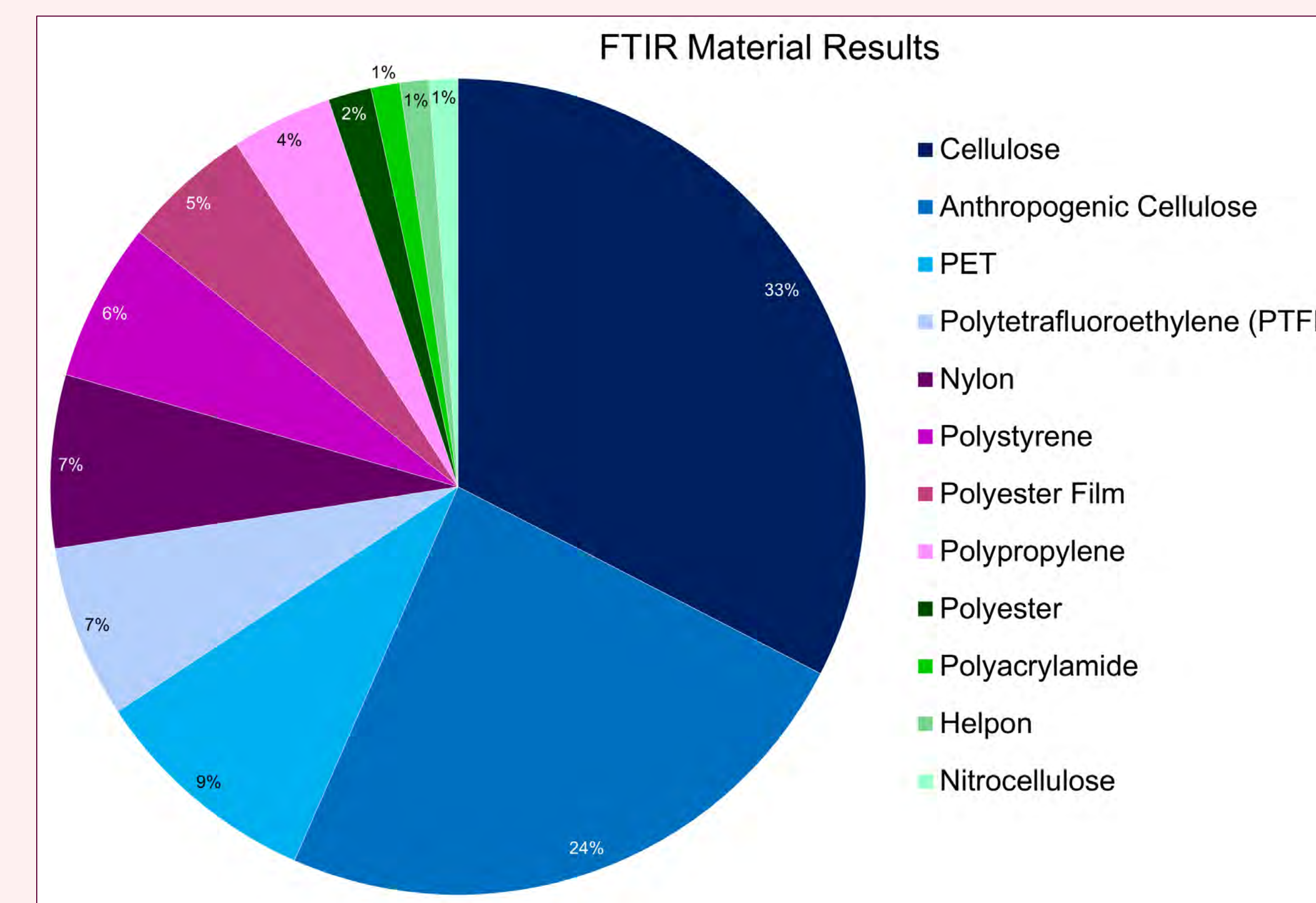
### Characteristics

- Kruskal-Wallis Test: There was a significant difference in mean MP length between site types ( $p = 3.72 \times 10^{-5}$ )
- CANA lots had the longest mean MP length with a mean of 4.86 mm

### Top Three MP Colors and Morphologies

	Clear	Light blue	Black	Fiber	Fragment	Film
CANA Lot	56%	8%	9%	90%	3%	7%
Oyster Reef	65%	8%	8%	93%	3%	3%
Residential	61%	8%	6%	87%	6%	5%

## FTIR Results



- So far, FTIR has been performed on 23% of CANA lot samples, 26% of oyster reef samples, and 13% of residential samples
- 25% of particles tested were materials used in the textile industry, such as nylon, polyester, and PET
- 24% of the materials matched highly similar forms of anthropogenically modified cellulose, such as microcrystalline cellulose, Kayocel 10W80, and powdered cellulose
- Polytetrafluoroethylene (PTFE), commonly known as Teflon™, was matched to 7% of the tested materials
- 6% of particles were polystyrene, which is commonly used to make Styrofoam™ products such as single-use coolers

## Future Directions

- This project will conclude a full year of collecting samples in May 2025
- As concern over the risks associated with inhaling MPs rises, continued research on the AD of MPs is needed worldwide
- Informed policies and regulations are essential to reduce MP pollution and protect vulnerable environments and public health

## Acknowledgments

Thank you to all the citizen scientists and CEElab members who helped collect and process samples. Thank you to MDC for facilitating residential collections, Canaveral National Seashore for site access, and the IRLNEP Small Grant Project for funding.

## Citations

<sup>1</sup>Zhang, Y., et al. (2020). Atmospheric microplastics: A review on current status and Perspectives. *Earth-Science Reviews*, 203 v.2020.103118 <sup>2</sup>Allen, S., et al. Atmospheric transport and deposition of microplastics in a remote mountain catchment. *Nat. Geosci.* 12, 339–344 (2019). <sup>3</sup>Walters, L., et al. (2021). Microplastics, oysters, and the Indian River Lagoon - UCF stars. STARS. <sup>4</sup>Nihart, A., et al. (2025). Bioaccumulation of microplastics in decedent human brains. *Nature Medicine*. <https://doi.org/10.1038/s41591-024-03453-1>





## Analysis of Microplastic Concentrations in Dried Algae Mats and Sediment Collected from Detention Basins in the Edwards Aquifer Recharge Zone

Paulina Quinonez, Andre Felton, Dr. Jeffrey Hutchinson  
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### Introduction

- Plastic pollution is growing exponentially
- Stormwater runoff from roads contains multiple pollutants that end up in surface waters or groundwater
- Detention basins are used to capture, temporarily store, and allow infiltration of stormwater runoff
- Algae might act as the first line of defense for capturing microplastics (MPs) in freshwater systems
- Sediment can tell us more about the movement of MP through detention basins



Roadway runoff

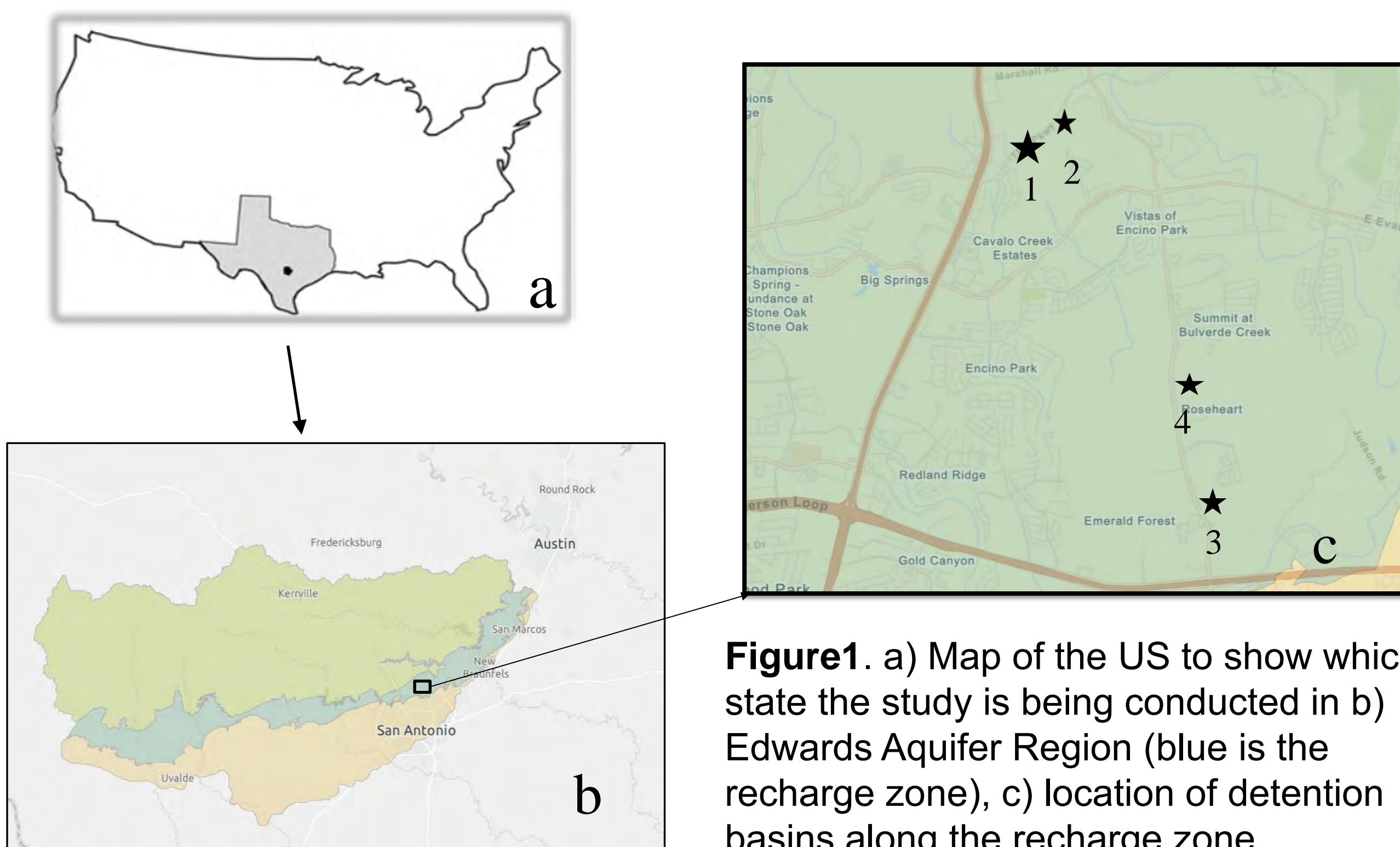


Detention pond holding roadway runoff

### Objectives

- Quantify the amount and types of MPs within algae mats and sediment layers of the basin
- Compare MP concentrations between the upper and lower layers
- Examine differences in MP accumulation across various detention basins.

### Study Area



### Methods

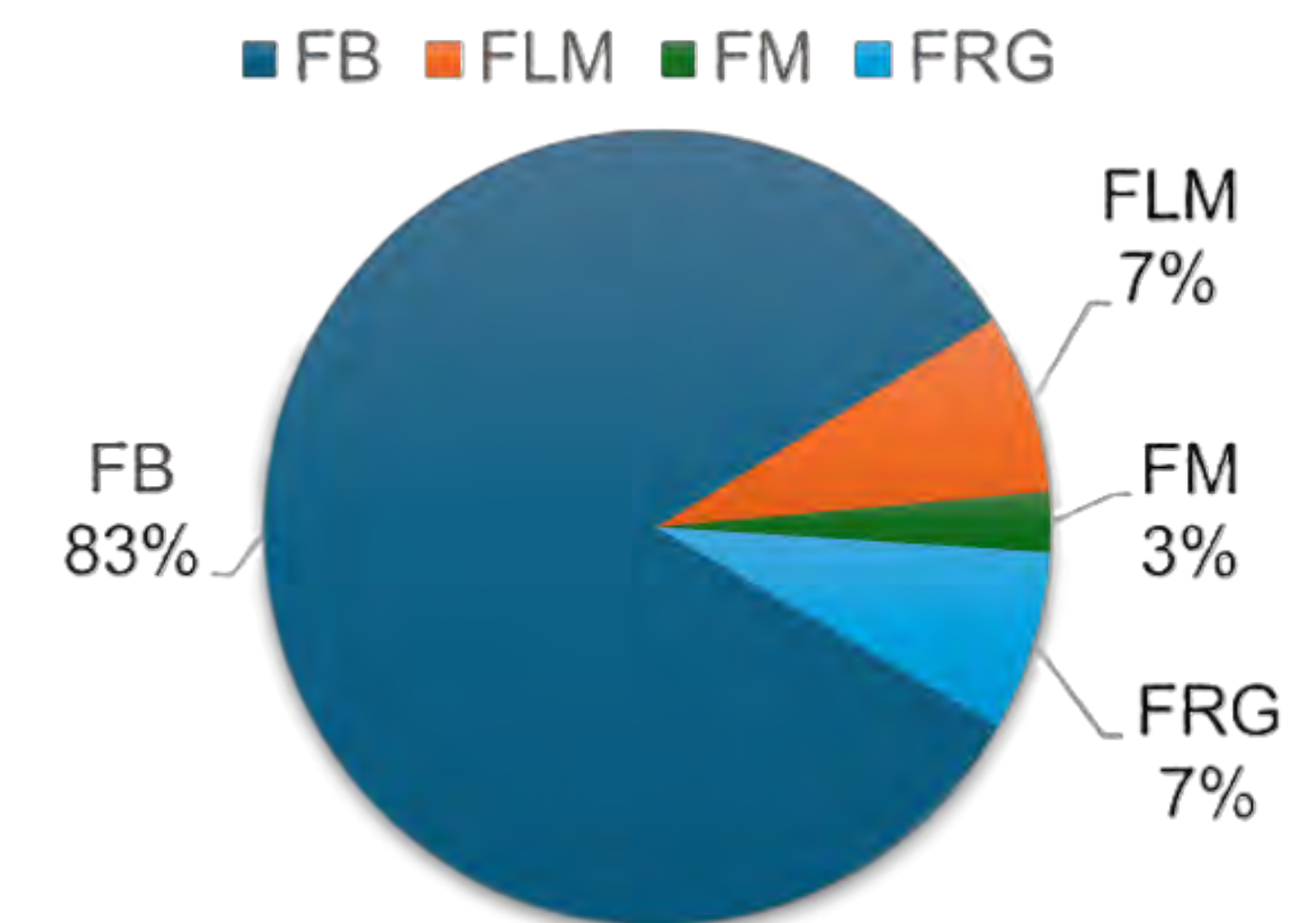
- Sediment and algae mats were processed under similar conditions
- Digestion using peroxide, separation by flotation and vacuum filtration were used to isolate MPs
- MPs were visually identified. Unidentifiable MPs were identified with FTIR



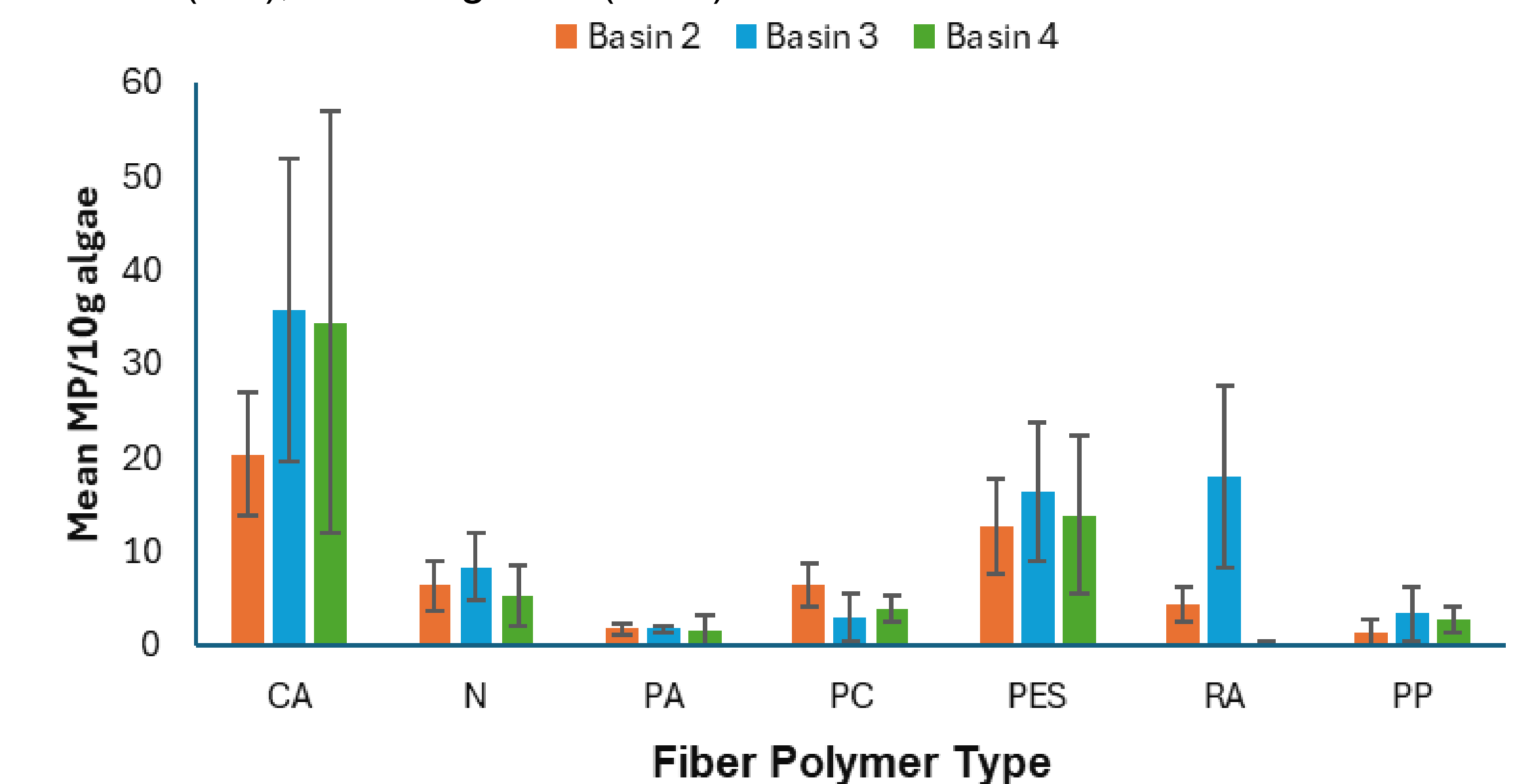
Top Left: Funnel set up, Bottom Left: Student vacuum filtering isolated MP, Right: Bulverde Fire Station Detention Basin with 3 sites where samples were collected.



### Preliminary Results (cont.)

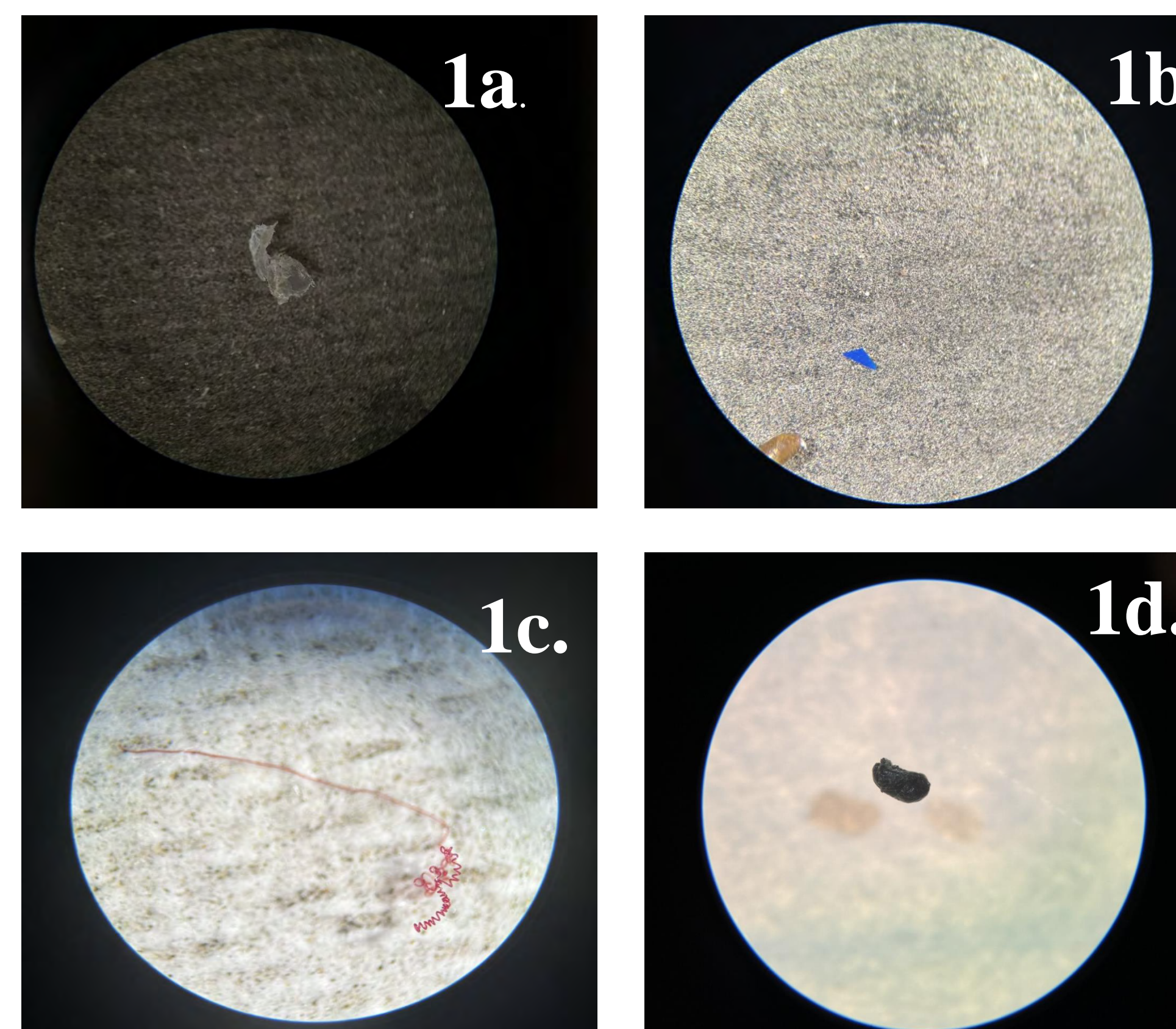


**Figure 3.** Percent morphotype of polymers: Fiber (FB), Film (FLM), Foam (FM), and Fragment (FRG)



**Figure 4.** Mean bar graph of mean MP/10g algae with standard error bars for 3 basins. Most common fiber polymer type are abbreviated as follows: Cellulose Acetate (CA), Nylon (N), Polyamide (PA), Polycarbonate (PC), Polyester (PES), Rayon, (RA), and Polypropylene (PP).

### Preliminary Results



**Figure 2.** Examples of MP found in detention ponds found in the Edwards Aquifer recharge zone; 1a. Teflon, 1b. PMMA, 1c. Polyamide 1.d. Polyurethane foam

### Conclusion

- Fibers are the most abundant throughout the study
- While MP such as Teflon and PMMA are found in low abundance, it should be of concern for potential carcinogens getting into our ground water
- Pharmaceuticals are also present in the basins
- Prelim evidence suggest detention basins are effective at capturing MP
- Data suggest maintenance plan for these basins from the city should be revised

### Acknowledgements

City of San Antonio and associated partners for providing funding









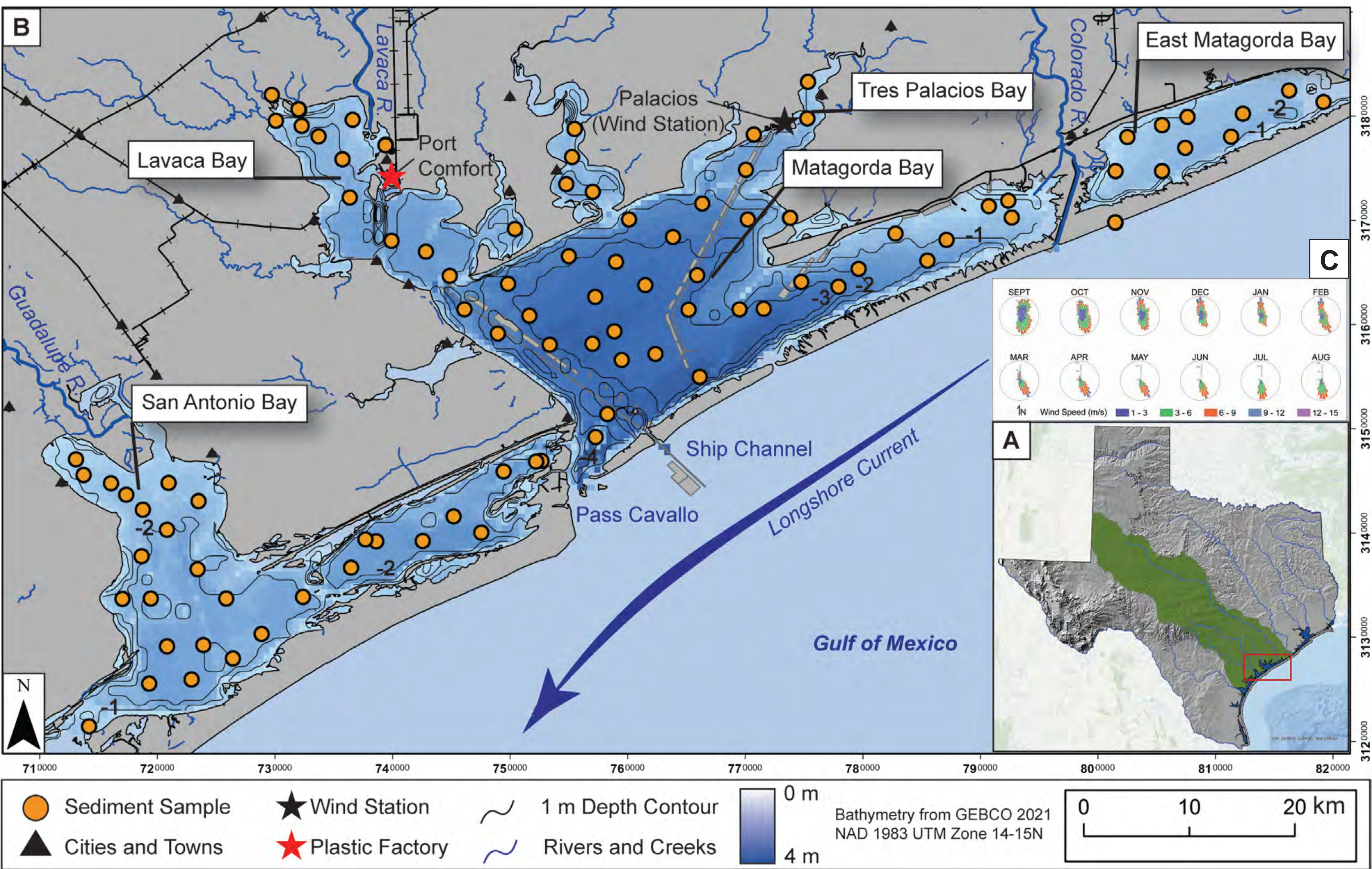
# Dynamics of Marine Litter Post-Hurricane Beryl: Assessing the Ultimate Fate of Flotsam

William Bailey<sup>a</sup>, David Mohrig<sup>a</sup>, Cornel Olariu<sup>a</sup>, Kutalmis Saylam<sup>a,b</sup>

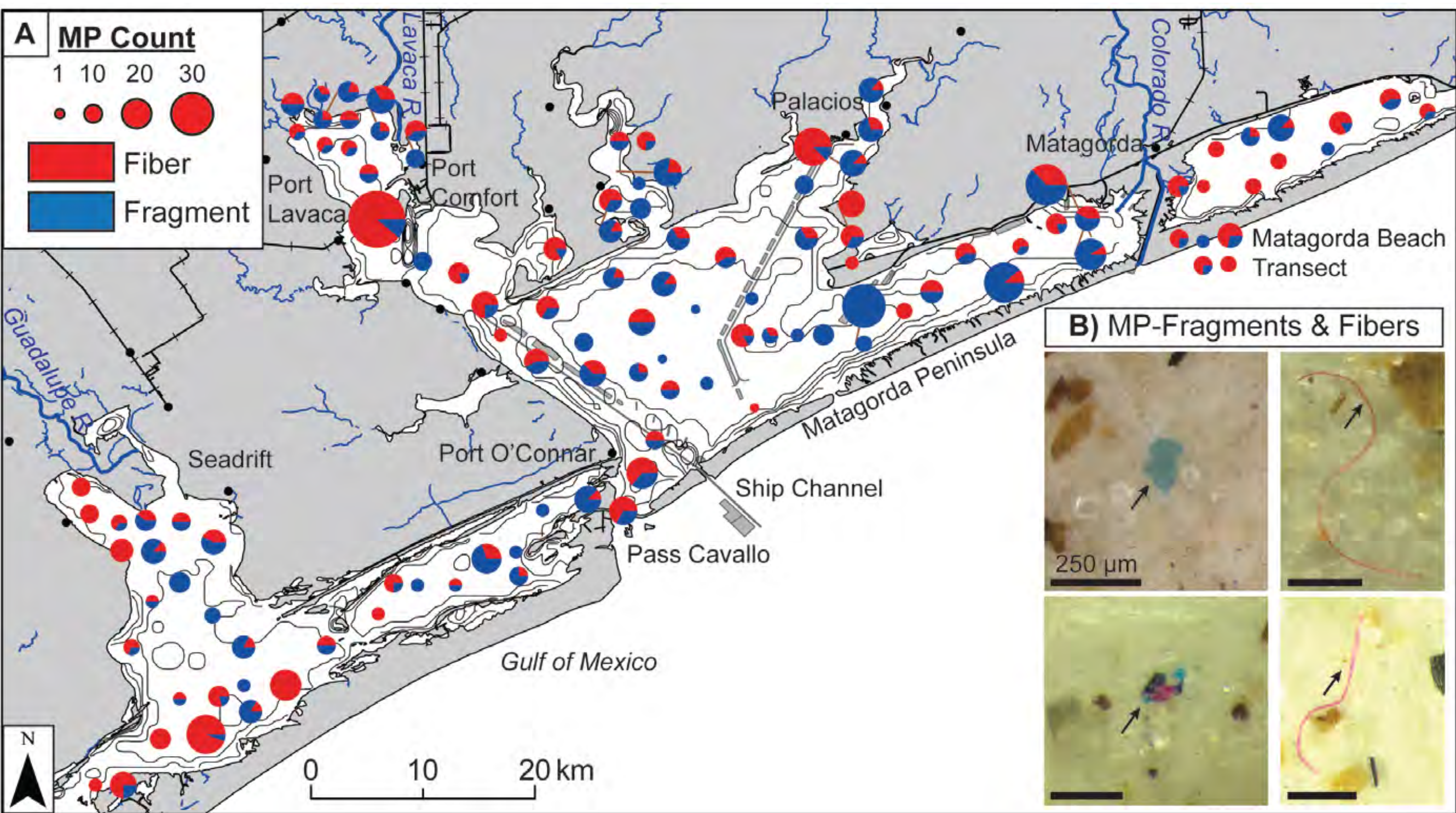
<sup>a</sup>Jackson School of Geosciences | <sup>b</sup>Bureau of Economic Geology | The University of Texas at Austin

## Introduction: Plastics in Coastal Bays and Barriers

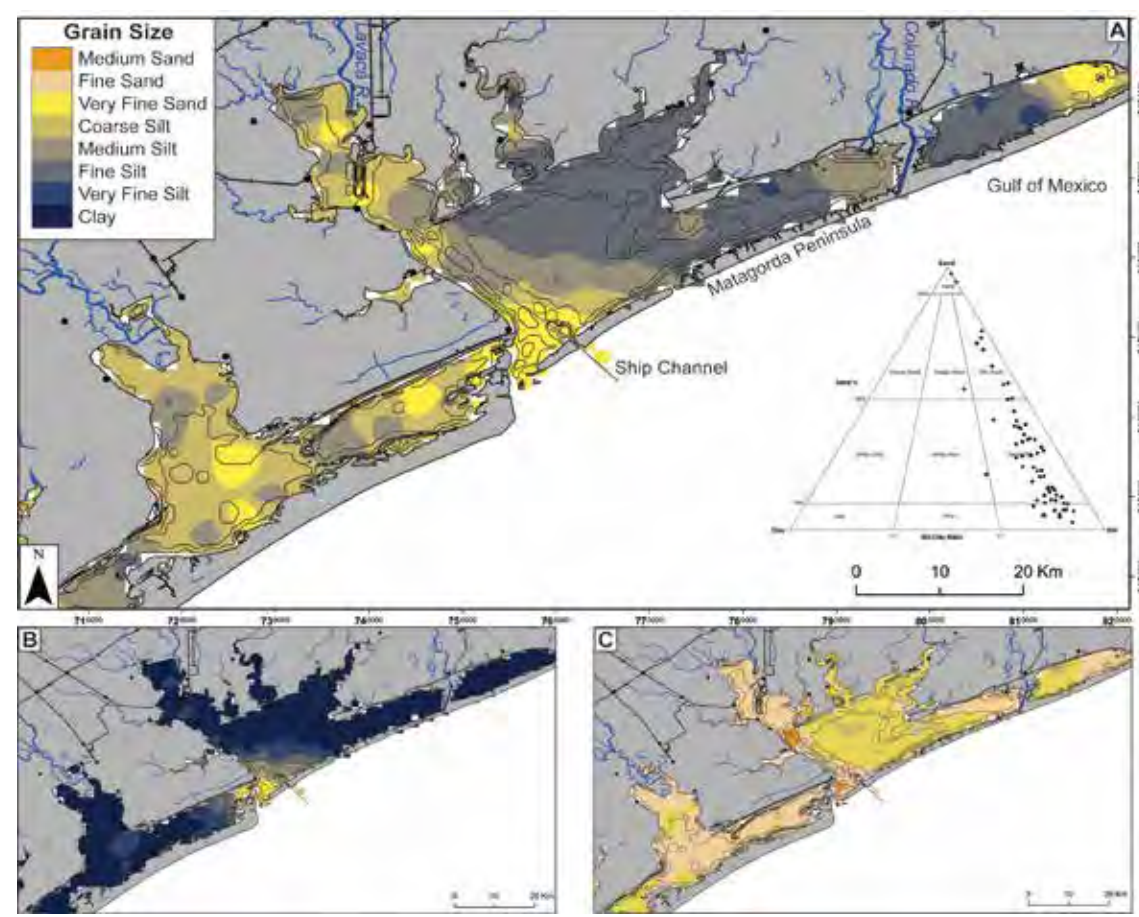
- Research of microplastics along the Texas coast to better understand where these pollutants may form hotspots through transport and depositional processes
- First project targeted Texas bays hypothesized to be depocenters for sediment and microplastics
- Second project targeted the barrier island after Hurricane Beryl to better understand where plastics concentrated during storms



A) Inset map of study area location on Texas Gulf Coast with extent of the river drainage basins discharging into the bays. B) Study area with locations of collected sediment samples. Principal rivers and inlets representing permanent marine exchange points. A red star indicates location of known plastic point source. C) Average monthly wind roses from Palacios, TX.



A) Distribution map of micro-fibers and -fragments, proportional symbols represent total microplastic concentration. B) Microscopic images illustrating MP examples.



Grain size distribution maps: A) D50, B) D10, C) D90. Grain size distribution of sediment vs. MPs. Note most MPs > D50.

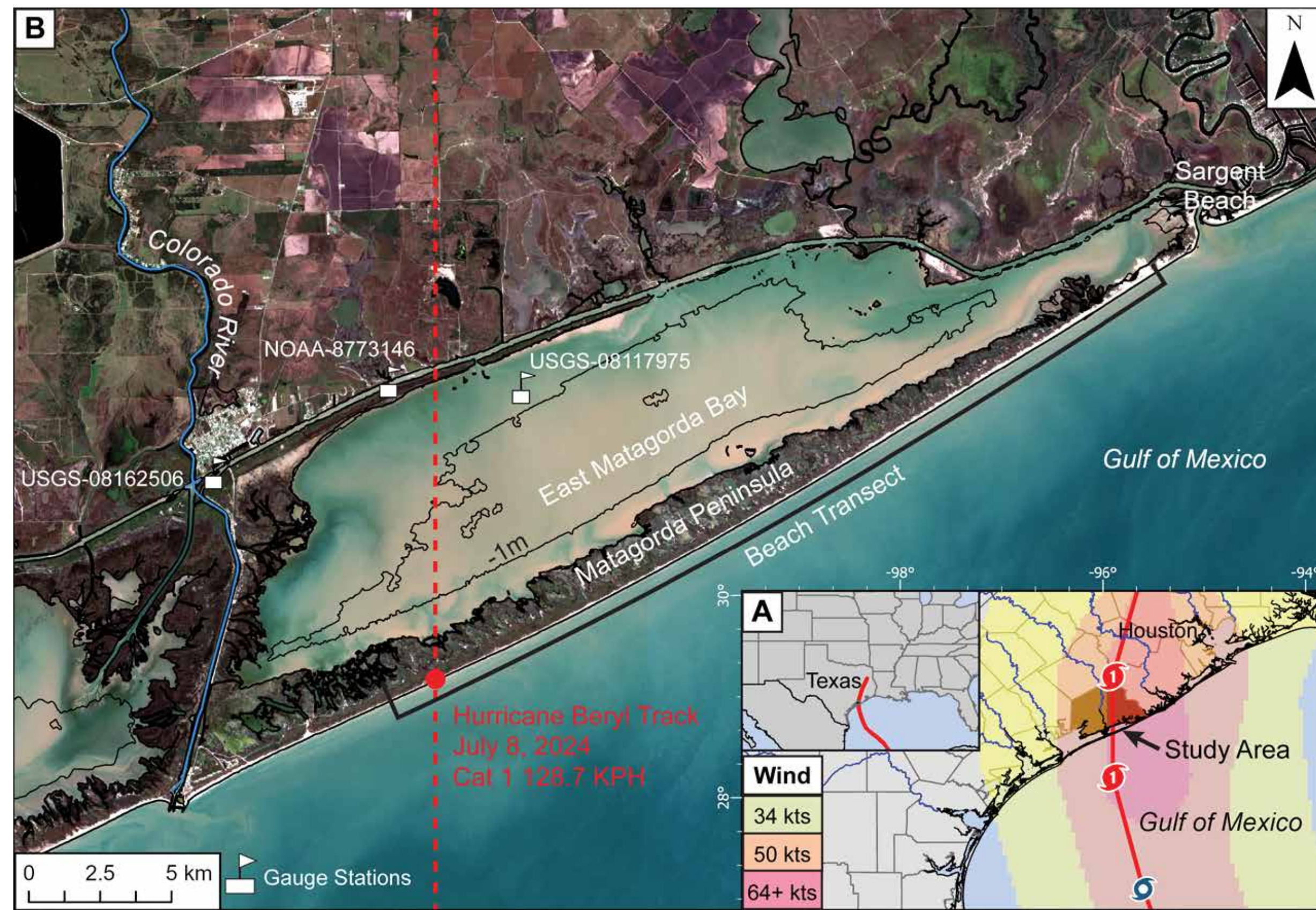
## P1. Bay Microplastics Key Takeaways

- Larger microplastic particles (fragments:  $178 \pm 93 \mu\text{m}$ , fibers:  $0.5\text{--}2.0 \text{ mm}$ ) consistently deposited with finer sediments indicating high transportability.
- The high degree of microplastic resuspension into bay waters has significant implications for limiting microplastic accumulation within bay sediments.
- This work provides a baseline for future studies quantifying the roles of wind on residence time of microplastics in coastal environments.

## Acknowledgments

This work was supported by the Matagorda Mitigation Trust and the Jackson School of Geosciences, University of Texas at Austin. We greatly appreciate the Near Surface Observatory at the Bureau of Economic Geology, UT-Austin, for providing access to the pre- & post-storm lidar. We thank Josh Malone for acquiring the drone imagery and field assistance.

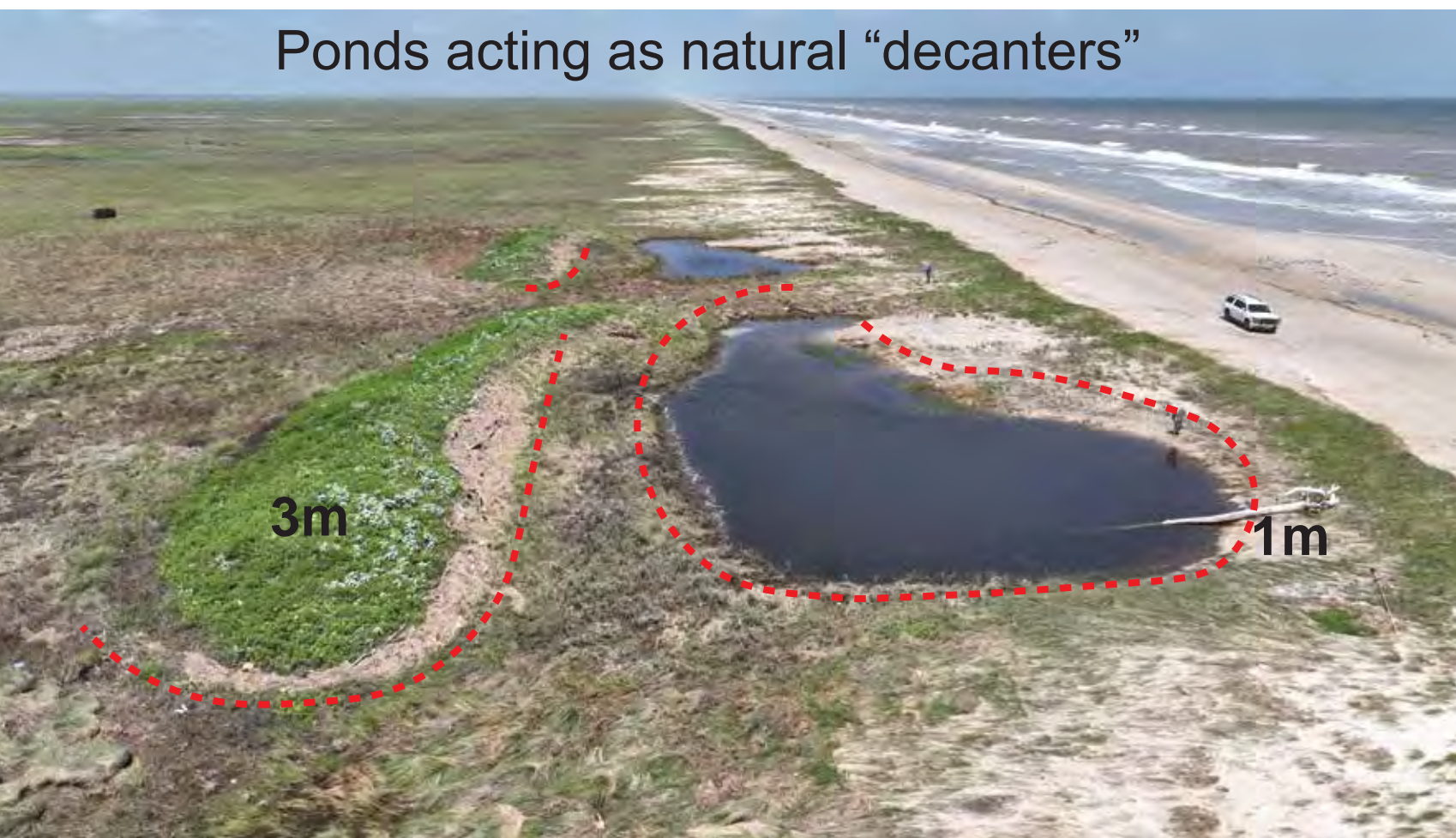
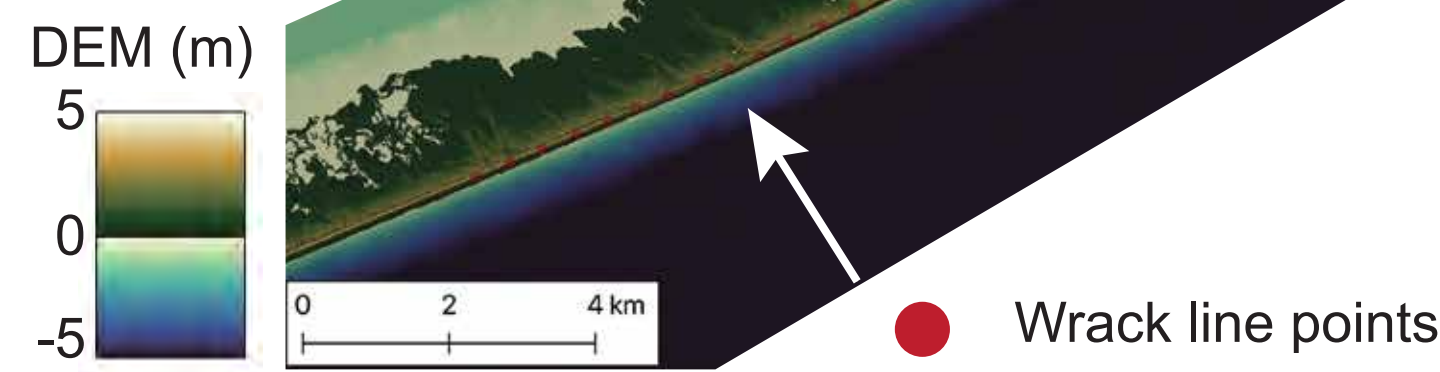
Contact: William Bailey | Email: wsbailey@utexas.edu



- Hurricane Beryl: category 1 storm
- Landfall July 8, 2024 4AM CST Matagorda Peninsula, TX
- Winds: 28 - 40 m/s | Flooding: >1.5 m
- Beach topography: 0 - 3.1 m (increasing East to West)

## Datasets:

- 30 km beach transect measuring plastic-rich deposits & drone surveyed photo-orthomosaics & DEM
- Pre- & post-storm lidar surveys for barrier topographic change

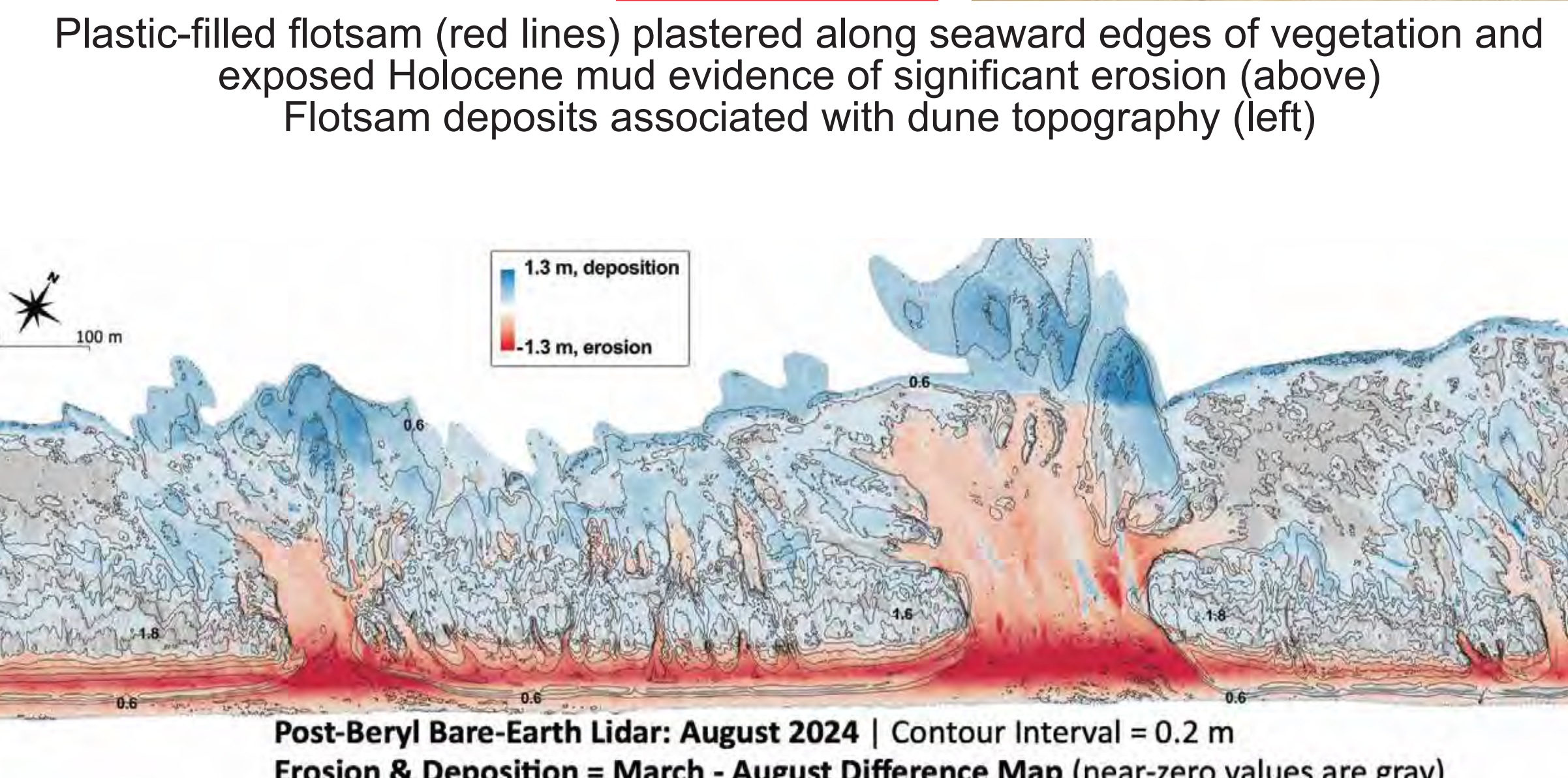
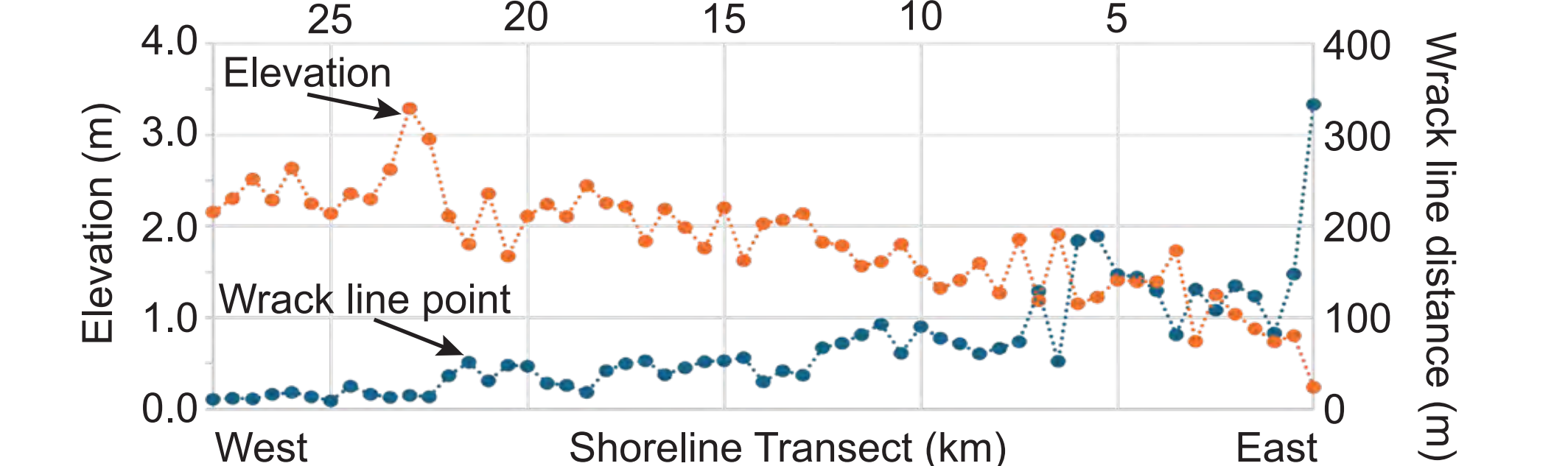
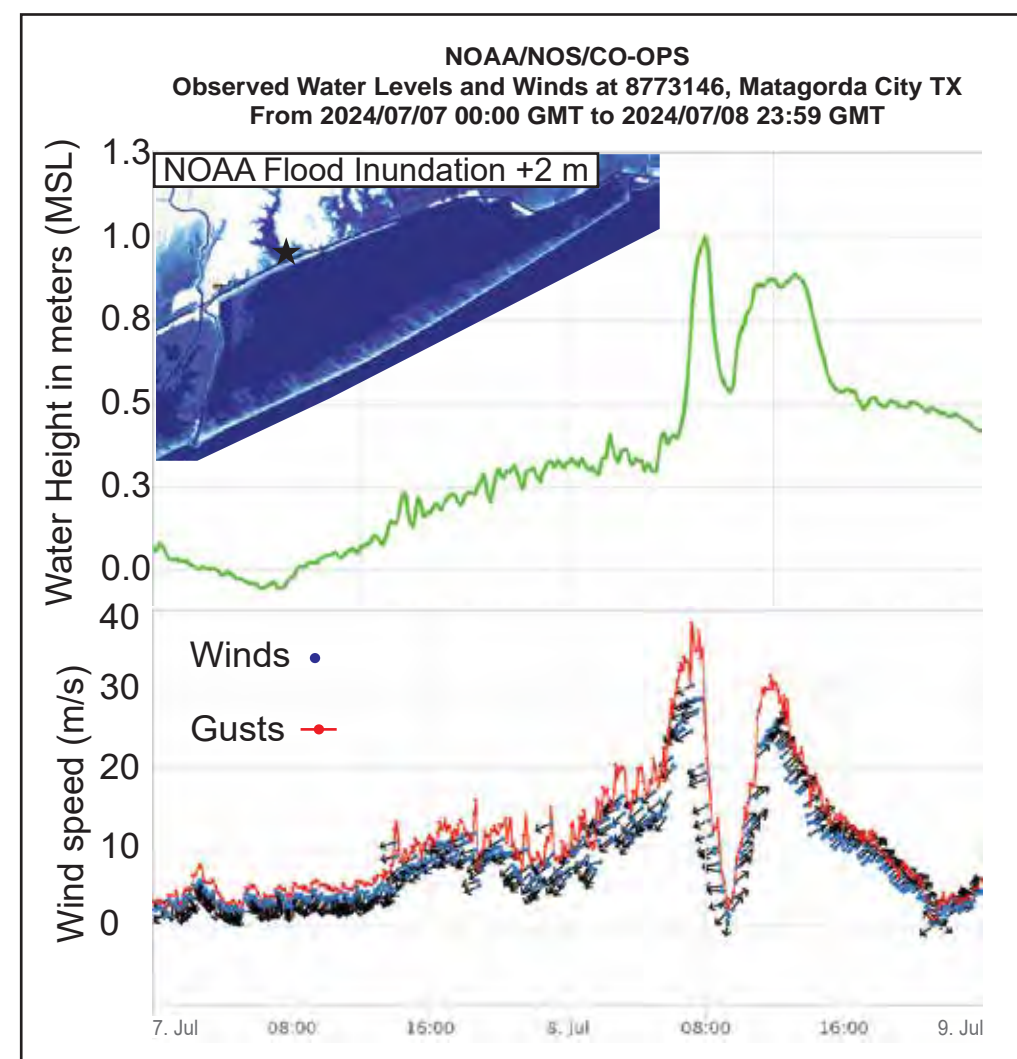


## P2. Results & Discussion

- Flotsam may be quantified using first return lidar surveys to illustrate depositional hotspots after storms.
- New overwash fans indicate large quantities of debris sourced to the back-barrier and bay sediments, as well as return channels exporting material to the Gulf.
- Given the ability of storms to erode material from bays and shorelines, the Gulf of Mexico is likely the ultimate fate for coastal pollutants.

## Project 2. Flotsam Storms

- Flotsam: "pieces of debris found on the beach or floating on the sea; anything or anyone that is not wanted or not considered to be important or useful"
- Field campaign to Texas coast after Hurricane Beryl (July 2024)
- Where does plastic concentrate during storms?
- What role does barrier topography and vegetation control flotsam transport and deposition?



## Why Should We Care?

- We dump plastic (and microplastics) into the rivers and coastal waters.
- While our pollutants are seemingly washed away to the ocean, storms reintroduce our unwanted debris (flotsam) to our coasts choking us back.
- The answer to the flotsam problem depends on topography / bathymetry of the coast, frequency of storms, or simply, the sedimentary dynamics of the coast, thus requiring more studies.



# Effects of PET Microfiber Exposure on Mating Behavior, Foraging Behavior, and Cognitive Flexibility in *Gambusia affinis*

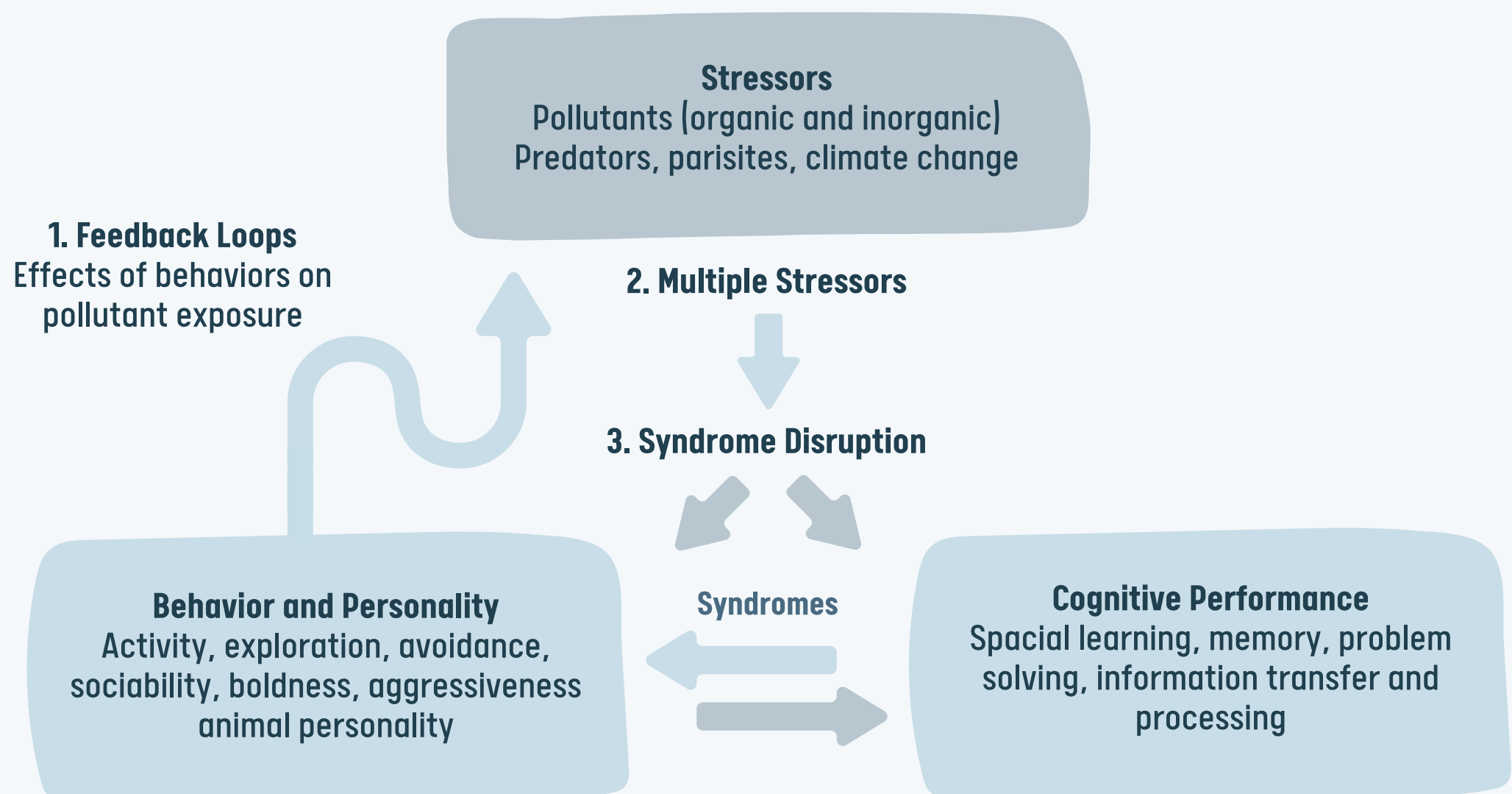
Jingping Graber, Adrienne Lihou, Rivers Hartzell, Maxximus G. Ramsaroop, Molly Cummings  
The University of Texas at Austin



The University of Texas at Austin  
Department of Integrative Biology  
College of Natural Sciences

## Introduction

Plastic production has increased dramatically in the last century, causing many adverse environmental impacts (Li et al., 2024). Although plastics are notable for their longevity, factors such as photodegradation, abrasion, and erosion can lead to fragmentation (Geyer et. al, 2020 & Prüst et. al, 2020). These fragments, known as microplastics (MPs, plastics <5mm in diameter), vary in shape, size, polymer type, and other traits. Polyethylene terephthalate (PET) microfibers are among the most common and persistent forms in aquatic systems (Reblein et al., 2021). Various organisms ingest MPs, which can cause adverse effects on behavior and cognition (Ma et al., 2019 & Jacquin et al., 2020). These behavioral/cognitive impacts can create feedback loops, perpetuating individual exposure and bioaccumulation of plastics and leading to population impacts that traverse evolutionary time spans by creating adaptive or maladaptive responses (Jacquin et al., 2020). Despite their importance, there is a substantial lack of experiments studying environmentally relevant MP concentrations, and PET microfibers are seldom used despite their prevalence (Geyer et. al, 2022 & Cunningham & Sigwart, 2019). To optimize standardization and ecological relevance in this study, *G. affinis* will be exposed to high and environmentally relevant concentrations of 42µm PET MPs.



## Hypothesis

This study aims to identify the effects of PET microfiber exposure on male mating behavior, foraging behavior, and cognitive flexibility in *G. affinis*. We hypothesize that exposure to microplastics will decrease cognitive flexibility, increase feeding motivation, and decrease mating motivation.

## Methods

Figures 2-3: Nile Red-stained MPs under UV and *G. affinis* Illustration

**Microfiber Production:** PET fibers were generated via tissue histology techniques (Knauss et. al., 2021). Microfibers were wound onto a spindle, embedded in paraffin, sectioned via microtome at 42 µm increments, processed with xylene, ethanol, and acetone, then stained with Nile Red for recapture.

**Figure 4: Exposure Treatments & Behavioral Assays**

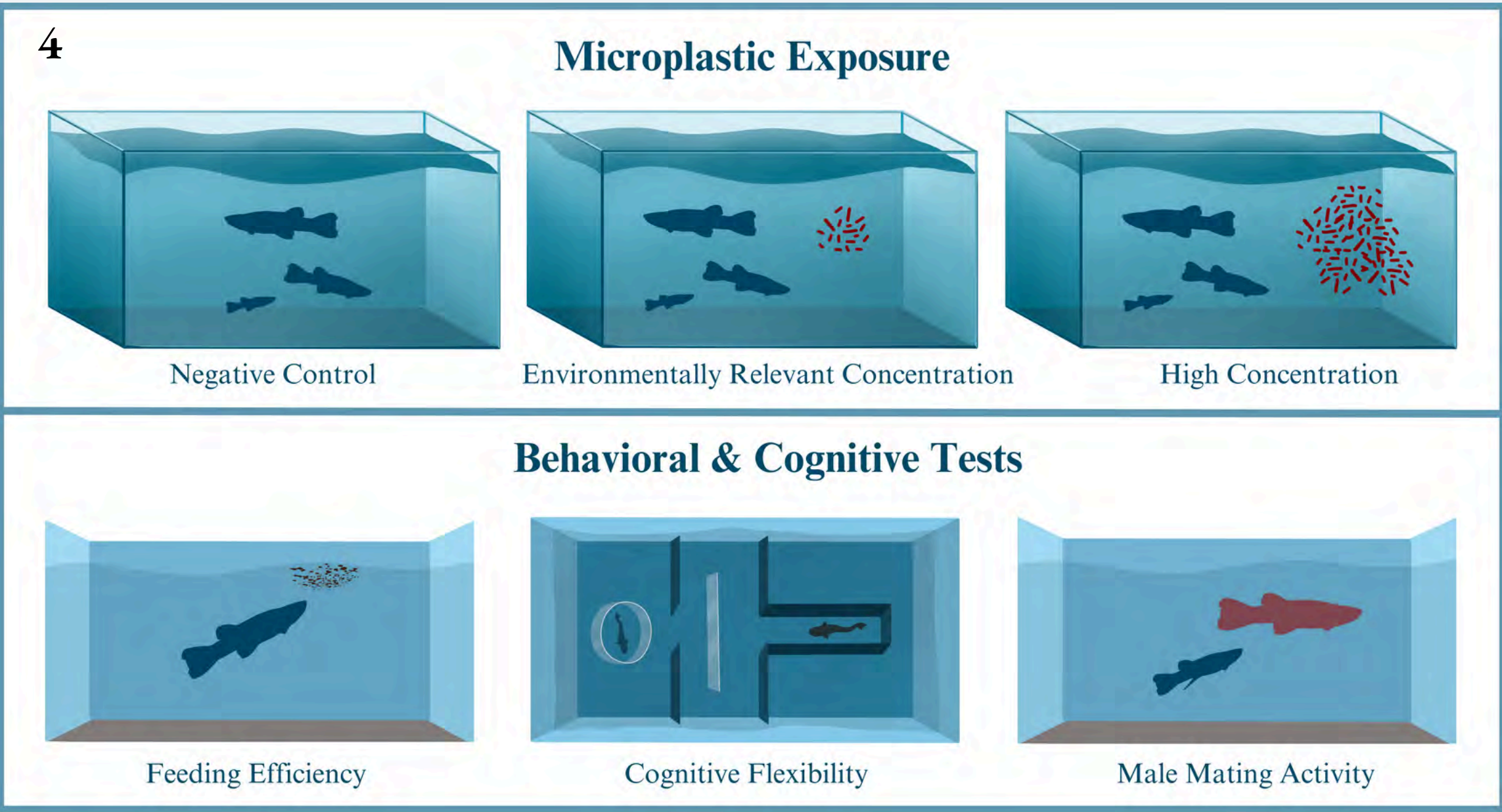
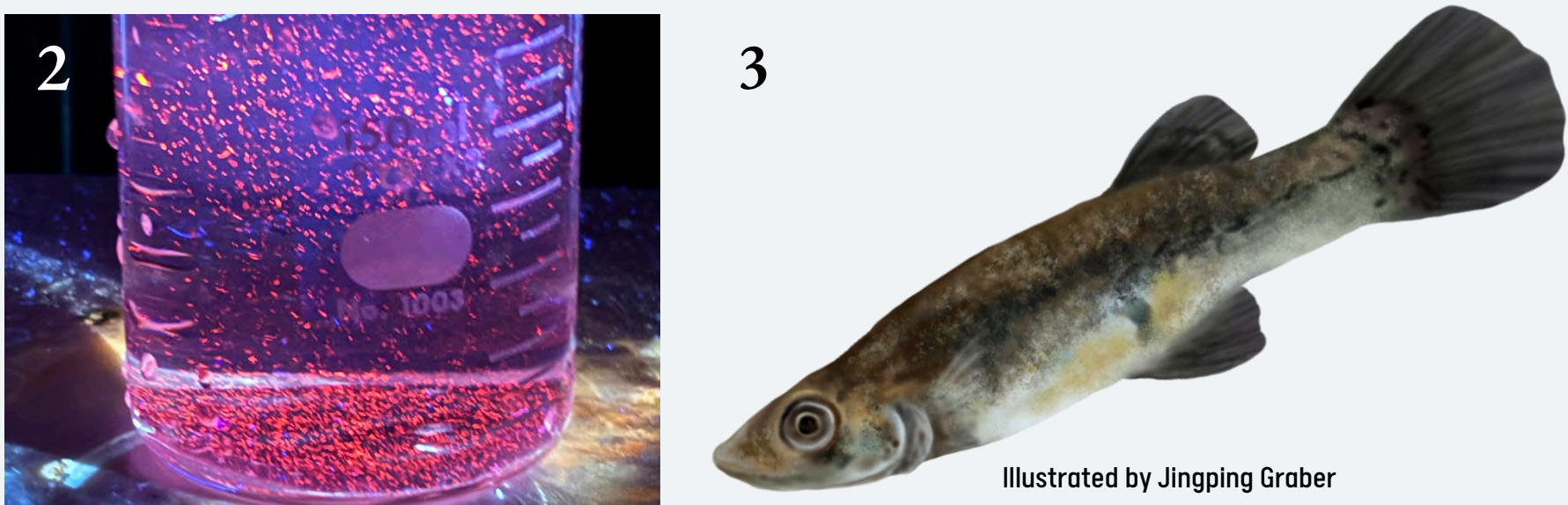
Fish were collected from Brackenridge Field Laboratory, Austin, Texas. Static aquaria were dosed with negative control (0 MPs/L), low (10 MPs/L), or high (1,000 MPs/L) concentrations for 14 days.

**Behavioral & Cognitive Tests:**

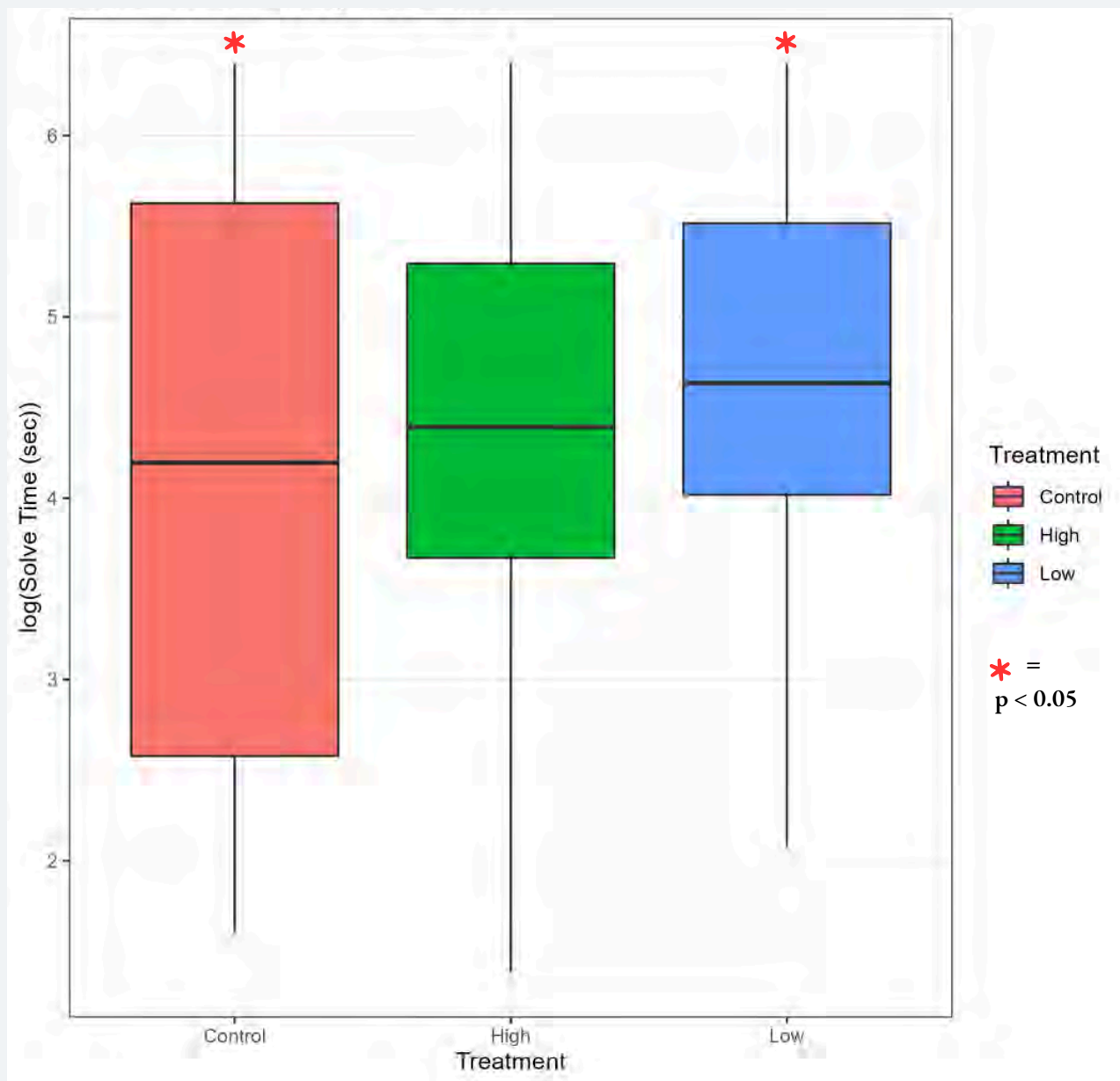
(i) *Foraging Behavior* (n=76): Fish were starved for 24 hours, then provided 0.005g of dry food and recorded.

(ii) *Cognitive Flexibility* (n=124): Fish were placed in an assay tank with a plexiglass obstacle positioned in front of a social reward. After 5 minutes of habituation, navigation of the assay was recorded.

(iii) *Male Mating Activity* (n=44) After 48 hours of isolation, males were introduced to an unfamiliar female. Interactions were recorded.

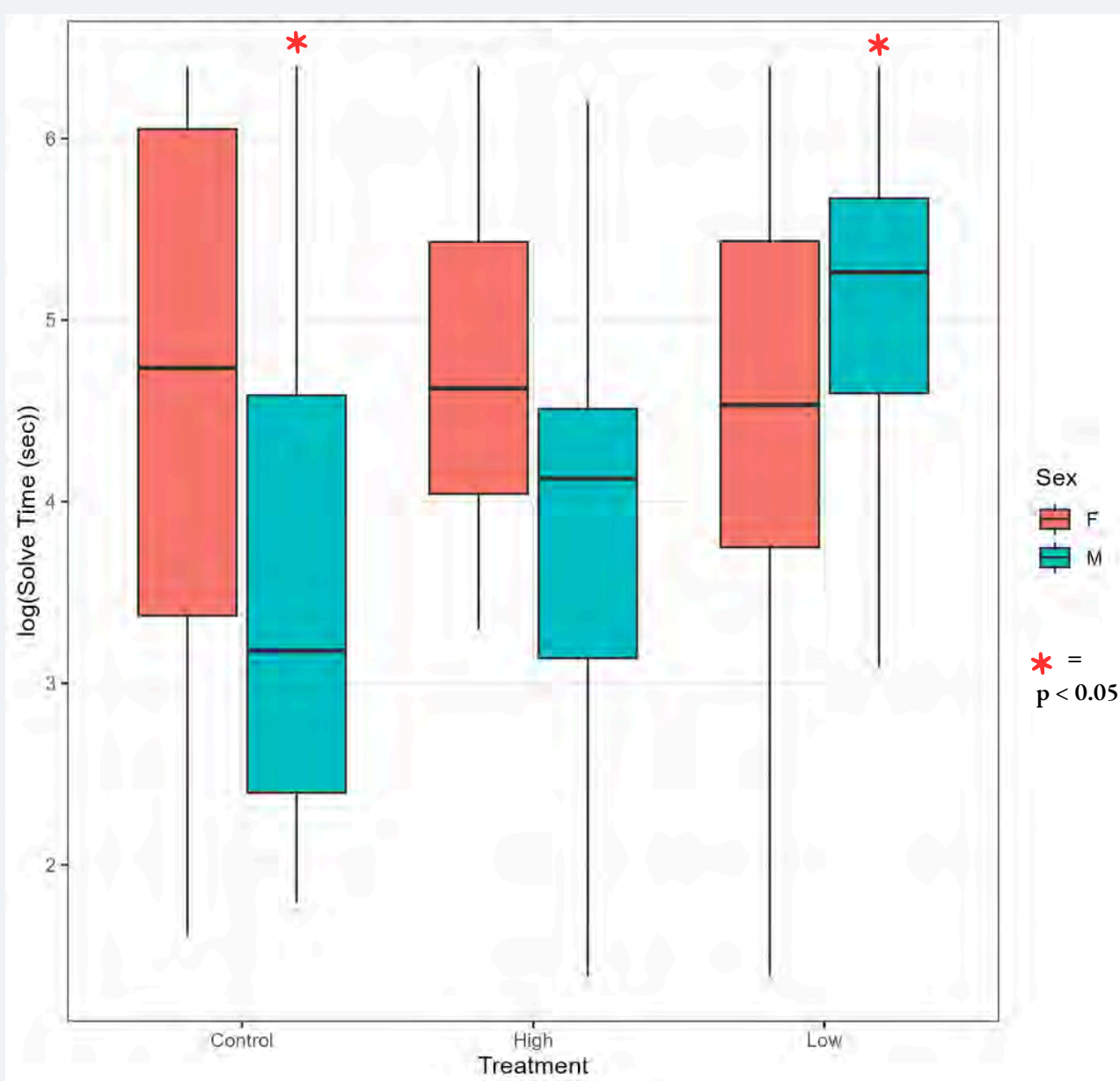


## Results



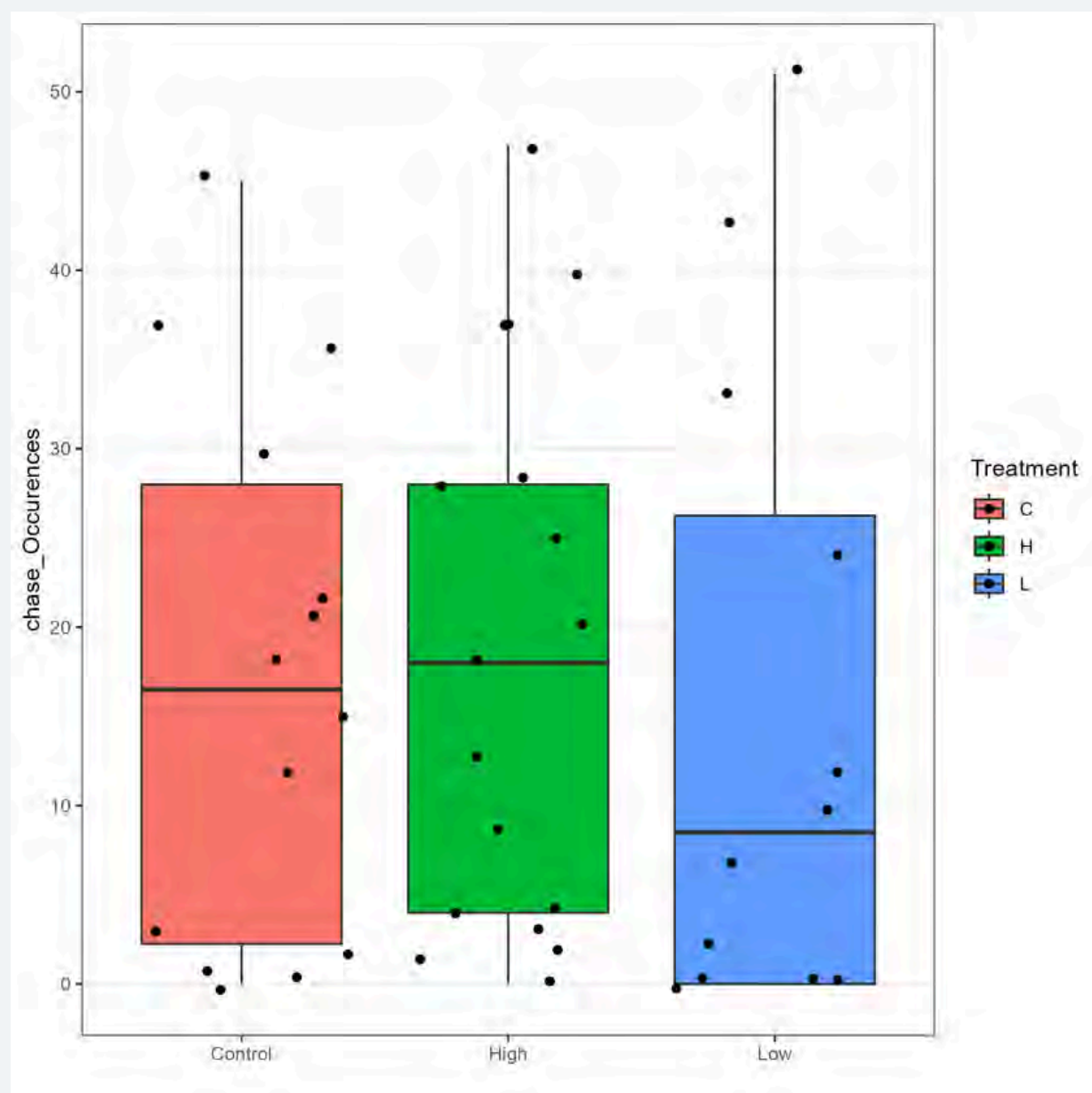
**Figure 5: Cognitive flexibility assay solve time by treatment**

Individuals exposed to low MP concentrations presented significantly greater solve-time than individuals from the control group. ANOVA  $p = 0.022$ ,  $t = 1.460$ ,  $n = 124$



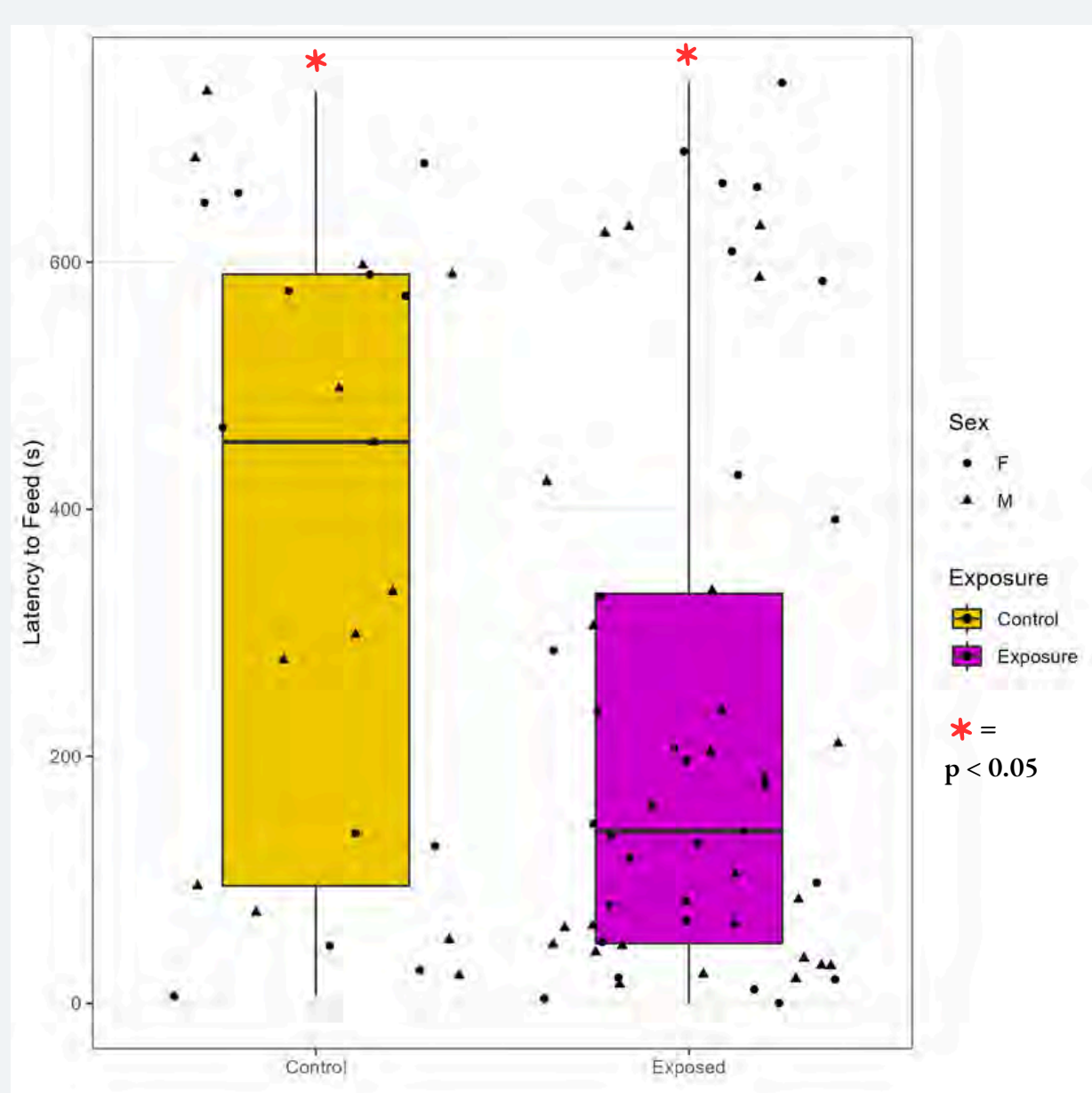
**Figure 6: Cognitive flexibility assay solve time by treatment and sex**

Exposure to low MP concentrations increases solve time in males, and no such effect is seen in females. Post-hoc Tukey test  $p = 0.0391$ ,  $t = 2.545$ ,  $n = 124$ .



**Figure 7: Mating behavior assay chasing occurrences by treatment**

Chasing occurs at the same frequency across all three treatment types. Kruskal-Wallis  $p > 0.05$ ,  $n = 44$ .



**Figure 8: Latency to feed by exposure**

Individuals exposed to MPs present shorter feed latency than individuals without MP exposure. Wilcoxon signed rank test  $p = 0.042$ ,  $w = 883$ ,  $n = 76$ .

## Discussion

**Cognitive Flexibility**

Solve time between control and low groups significantly differed, with low groups solving slower than controls, indicating that MP exposure impacted problem-solving abilities (Figure 5). This is possible because MPs can pass through the blood-brain barrier and affect their behavior (Mattson et al., 2017). Furthermore, MPs can lead to malnutrition, which results in reduced cognitive function and slower swimming speed (Guerrera et al., 2021 & Yin et al., 2018).

**Mating Behavior**

There were no significant differences across groups for chase events occurred, as male mating motivation is innately strong and has little plasticity (Dadda et al., 2008)

**Foraging**

MP-exposed fish began feeding earlier than the control fish (Figure 8). This may be due to malnourishment caused by MP accumulation or damage in the intestinal tract which could result in a higher motivation to feed (Guerrera et al., 2021).

**Future Directions**

Numbers of MPs present across organs will be determined. This data may provide insights into a biological uptake limit of MPs and indicate why cognitive flexibility does not decrease linearly with increasing MP concentrations.

## Acknowledgements

We would like to thank the Brackenridge Field Laboratory for providing fish for our study, Dr. Molly Cummings for her supervision and access to lab resources, and the University of Texas Integrative Biology Department for funding.

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# Examining the Plastic Degrading Potential of Marine Fungi Found on the Texas Coast

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The University of Texas at Austin



## Introduction

12 million tons of plastic enter the ocean every year- this is equal to 80 garbage trucks every hour. The growing rate of plastic pollution requires unique, multi-faceted solutions. This project aims to target one of the faces of the plastic issue: degradation. Nurdles are small, plastic pellets that are used to manufacture plastic goods. When nurdles are transported via ship or train, they are spilled and leaked frequently due to their size and shape, causing them to be the second-largest source of microplastic pollution by weight. Every year, approximately 230,000 tons of plastic nurdles enter the ocean. These microplastics provide a unique angle from which to examine the ability of microbes to degrade plastic. The hard surface and buoyant nature of nurdles make the perfect surface for opportunistic microbes to colonize and form biofilms on. Nurdles create a microenvironment where specialized microbial communities can colonize and potentially evolve enzymatic capabilities to degrade the specific polymer, driven by selective pressures and the abundance of a consistent carbon source. For these reasons, we are investigating if the fungi living on the nurdles are using the plastic as an energy source. If we can identify the species of microbes that have the ability to break down plastic, we can delve into more opportunities for widespread plastic degradation techniques.

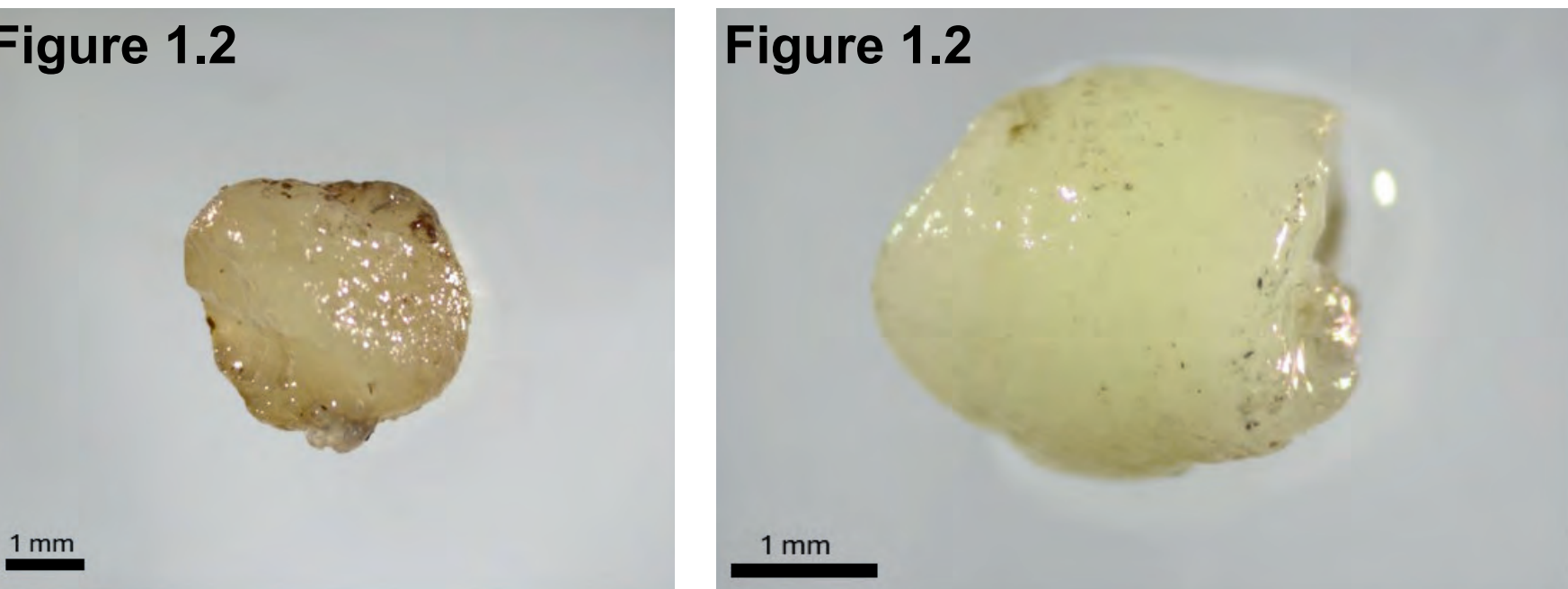
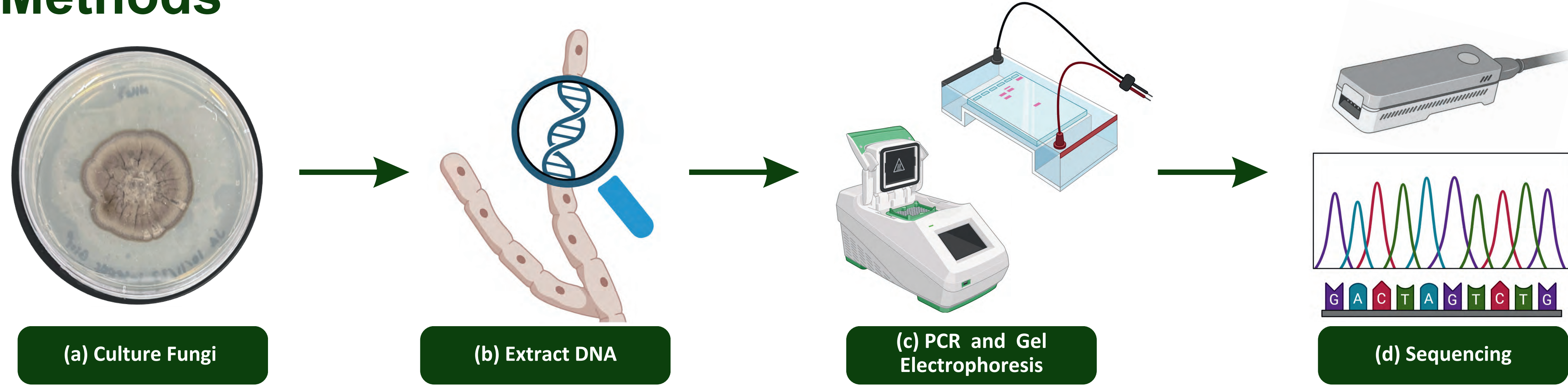


Figure 1.1 shows nurdles being collected from the beach. Figures 1.2 and 1.3 depict nurdles photographed using a stereomicroscope.

## Research Goals

- Identify fungal samples capable of plastic degradation
- Improve on the process of fungal culture and isolation from coastal nurdles
- Work through genomic analysis of fungal samples using PCR and sequencing

## Methods



## Microscopy

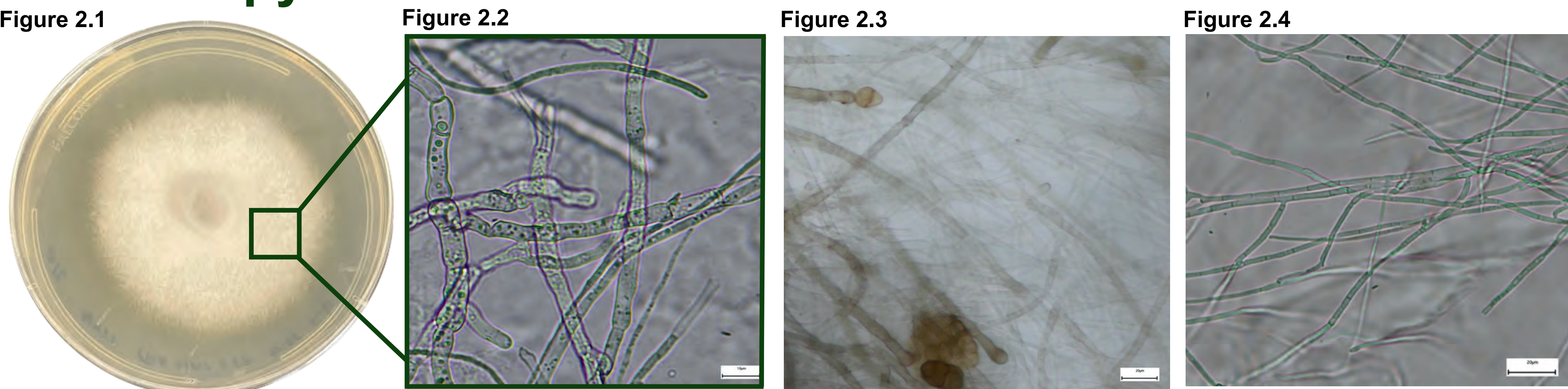


Figure 2.1 shows fungi sample N471, cultured from the nurdles and photographed (figure 2.2) using a compound microscope. Figure 2.3 shows sample number N283, identified as *Phaeosphaeria* sp. Figure 2.4 shows sample N164, which was identified as *Alternaria alternata*. All images were produced using a compound microscope.

## Results

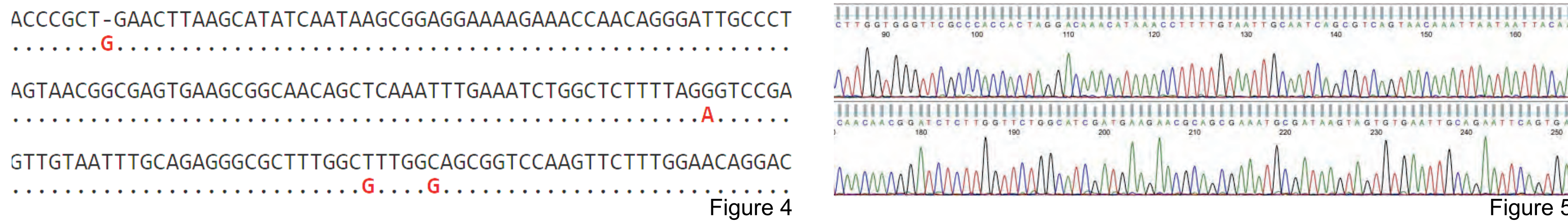
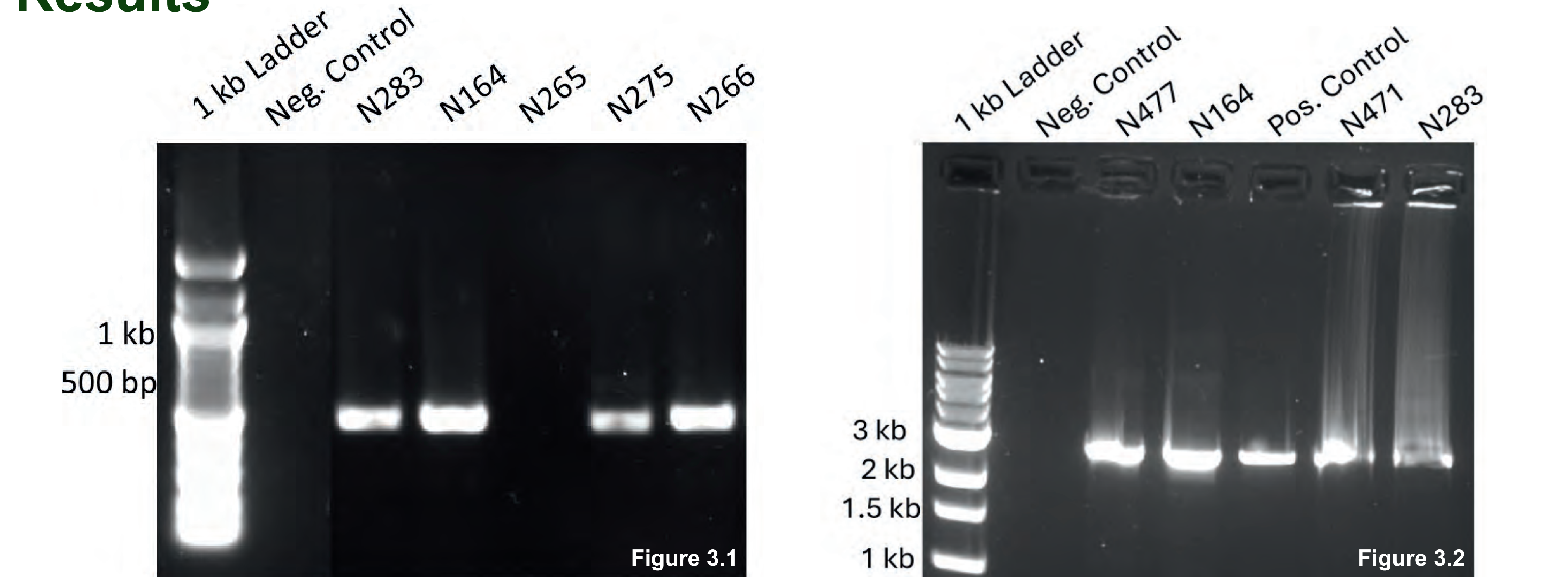


Figure 3.1 shows gel results of amplifying 600 base pairs of the 18S rRNA gene locus, and figure 3.2 is a gel image of a 2.7 kb amplicon of the same locus. Figure 4 shows the Sanger sequencing results from nurdle sample N164, which was identified as *Alternaria alternata* using the Basic Local Alignment Search Tool (BLAST). Figure 5 shows the closest alignment for sample N470. Areas where my sample does not match with the reference species are highlighted in red.

## Analysis and Future Research

Nurdle Sample	Species Identified
N283	<i>Phaeosphaeria</i> sp.
N477	<i>Pleosporales</i> sp.
N471	<i>Pleosporales</i> sp.
N164	<i>Alternaria alternata</i>
N266	<i>Stemphylium lycopersici</i>

*Alternaria alternata* and *Stemphylium lycopersici* are plant pathogens. This raises the question of how they ended up in an aquatic environment and how they were able to colonize a nurdle. Further research is needed to identify more fungal species and to investigate whether any of these species display plastic degradation capabilities. Currently, a carbon source assay is being conducted to quantify these species' plastic degradation capabilities. This assay screens for growth in conditions where plastic is the only carbon source. The end goal of this project is to eventually isolate the enzyme these fungi use to degrade plastic. Once the enzyme(s) that the fungi are creating are identified, we can engineer and improve the enzyme for large-scale plastic degradation.

## Acknowledgements

I would like to acknowledge my Bioprospecting peers for their support in lab; Dr. Kasia Dinkeloo, Dr. Jennifer Brodbelt, and Dr. Andrew Ellington for providing an encouraging research environment; and the Freshman Research Initiative for providing us with this opportunity.

## References

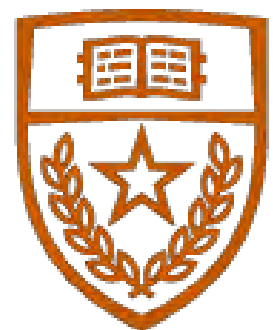
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- Images made with Biorender



# Fluorescent detection of Nile Red-stained microplastic uptake in the roots of *Arabidopsis thaliana*

Kailyn Nonhof<sup>1</sup>, Jing Graber<sup>1</sup>, Kasia Dinkeloo<sup>1</sup>

<sup>1</sup>BioProspecting Lab, Freshman Research Initiative, College of Natural Sciences, The University of Texas at Austin



The University of Texas at Austin  
Freshman Research Initiative  
College of Natural Sciences

## Introduction

With plastic synthesis and distribution having increased dramatically in recent years, a closer look at the side-effects of plastic integration into biological contexts has become vital. More specifically, increasing levels of microplastics (MPs) in the rhizosphere, the portion of soil in direct interaction with plant roots, has sparked an uptake in studies exploring plastic-plant root interactions (1). Microfibers (MFs), a subset of MPs, outnumber other MP types, and polyethylene (PET) MFs are among the most common MFs. However, PET MFs are seldom used in exposure studies, indicating a need for further research (2). One location tracking method of micro-scale plastics is the use of fluorescent staining and imaging. This project seeks to develop a method of fluorescent detection that allows for the visualization of polystyrene (PS) microplastic and PET MF uptake in the roots of *Arabidopsis thaliana*. This method's aim is to inform MP and MF uptake ability and location, providing valuable insight into plastic-plant interactions in the soil.

## Methods

Two different methods were developed to visualize the uptake of Nile Red (NR) stained PS and PET into the roots of *A. thaliana*.

### General workflow

1. Creation of MP Sample
2. Plastic Staining
3. Plastic Sterilization
4. Prepare Square Petri Dishes
5. Growth Period
6. Imaging

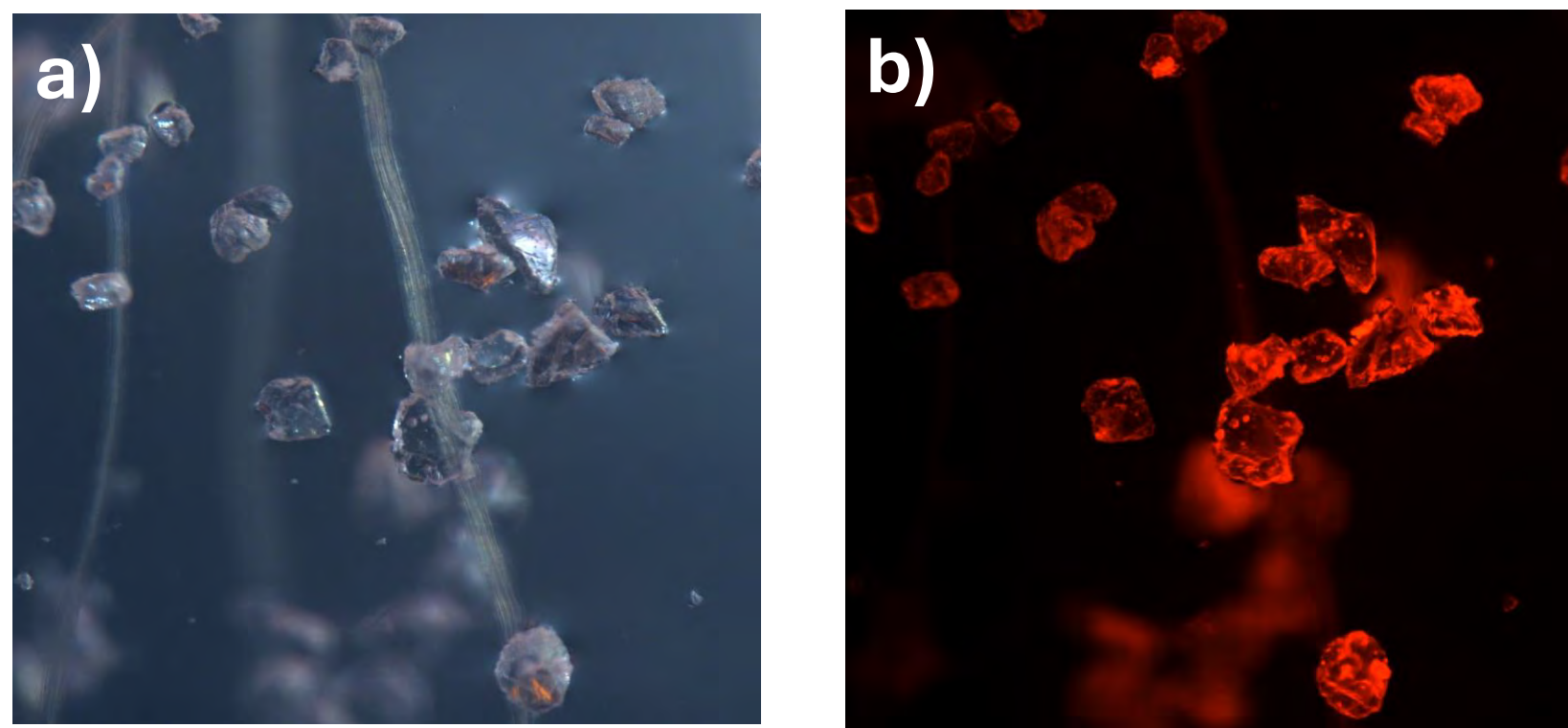
### PS Microplastic Uptake:

Polystyrene pellets were ground into a fine powder with particle size <5 mm using a coffee grinder. The MP sample was then stained with NR dye. *Arabidopsis thaliana* was planted in MS/2 media spiked with the NR-stained PS and allowed to grow for a little over a month in vertical square petri dishes. Root samples were taken from media and stained with DAPI dye, following the method developed by Stanton et. al. (3). Finally, root samples were imaged under NR and DAPI excitation wavelengths using a Nikon SMZ25 fluorescent stereomicroscope.

### PET Microfiber Uptake:

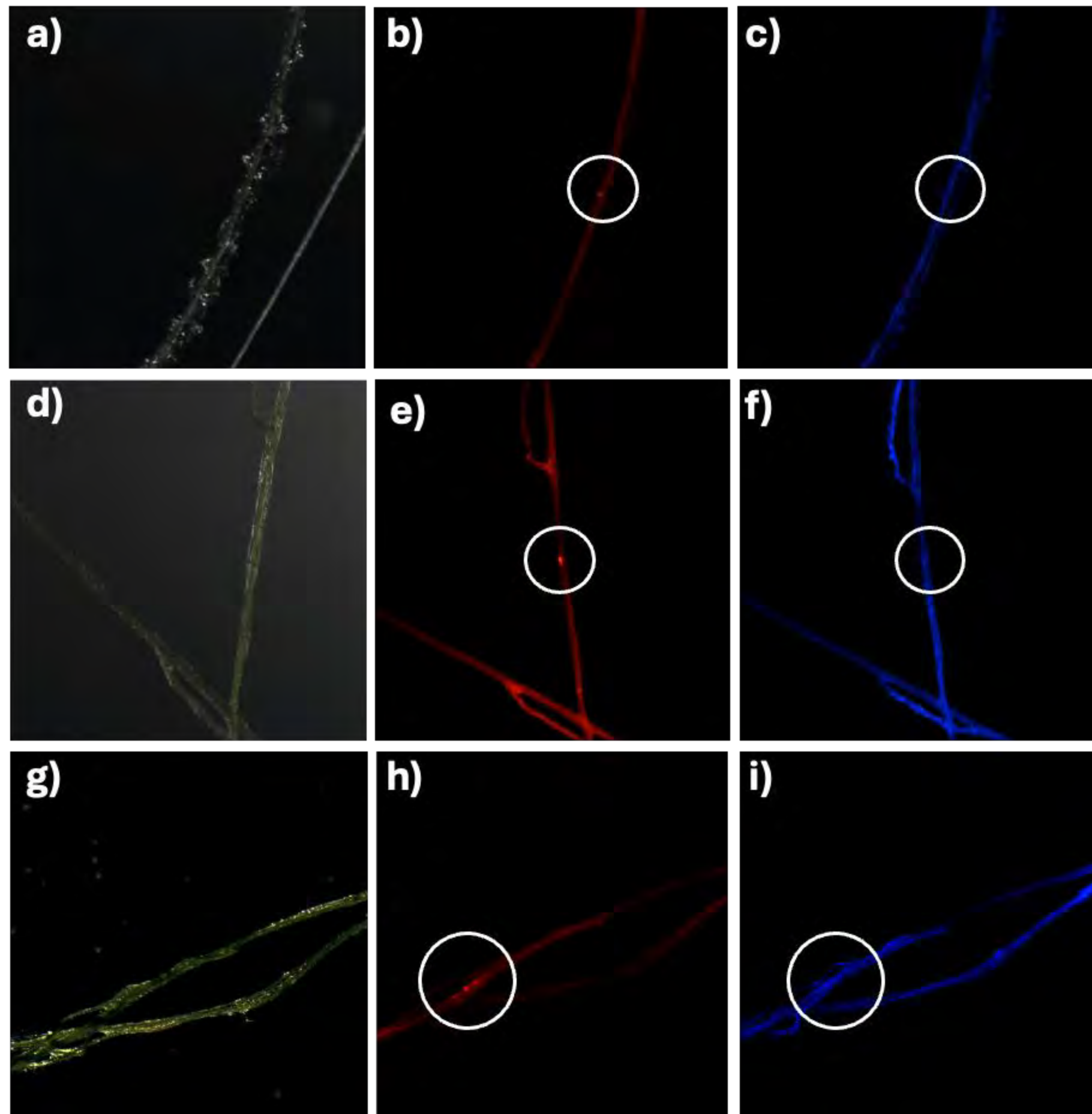
PET fibers with a diameter of 0.014 mm were wound onto a spindle, embedded in paraffin wax, and sectioned with a microtome at various increments under 0.5 mm. Fibers were chemically isolated from paraffin and stained with NR dye. *Arabidopsis thaliana* was planted in MS/2 media spiked with the NR-stained microfibers and allowed to grow for a little over a month in vertical square petri dishes. Root samples were taken from media and imaged under NR and GFP excitation wavelengths using a Nikon SMZ25 fluorescent stereomicroscope.

## Figures and Results



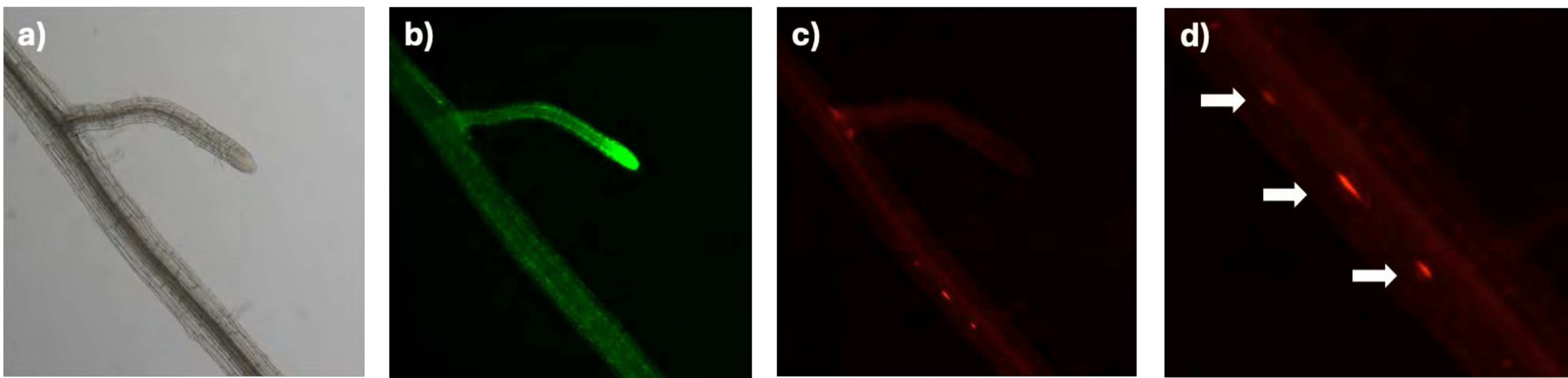
**Fig. 1:** NR-stained PS oriented around plant root: (a) Brightfield (b) NR-emission

**Fig. 1** showcases the growth conditions in the NR-stained PS spiked MS/2 media. A potential affinity of the plastic for the plant root is demonstrated by the orientation of the NR-stained PS about the root. **Fig 1. b.** further validates the viability of the method, in that little to no autofluorescence of the root is observed while the stained MPs fluoresce clearly.



**Fig. 2:** BF and fluorescent DAPI co-stained *A. thaliana* root samples: (a) Root 1 Brightfield (b) Root 1 NR emission (c) Root 1 DAPI emission (d) Root 2 Brightfield (e) Root 2 NR emission (f) Root 2 DAPI emission (g) Root 3 Brightfield (h) Root 3 NR emission (i) Root 3 DAPI emission

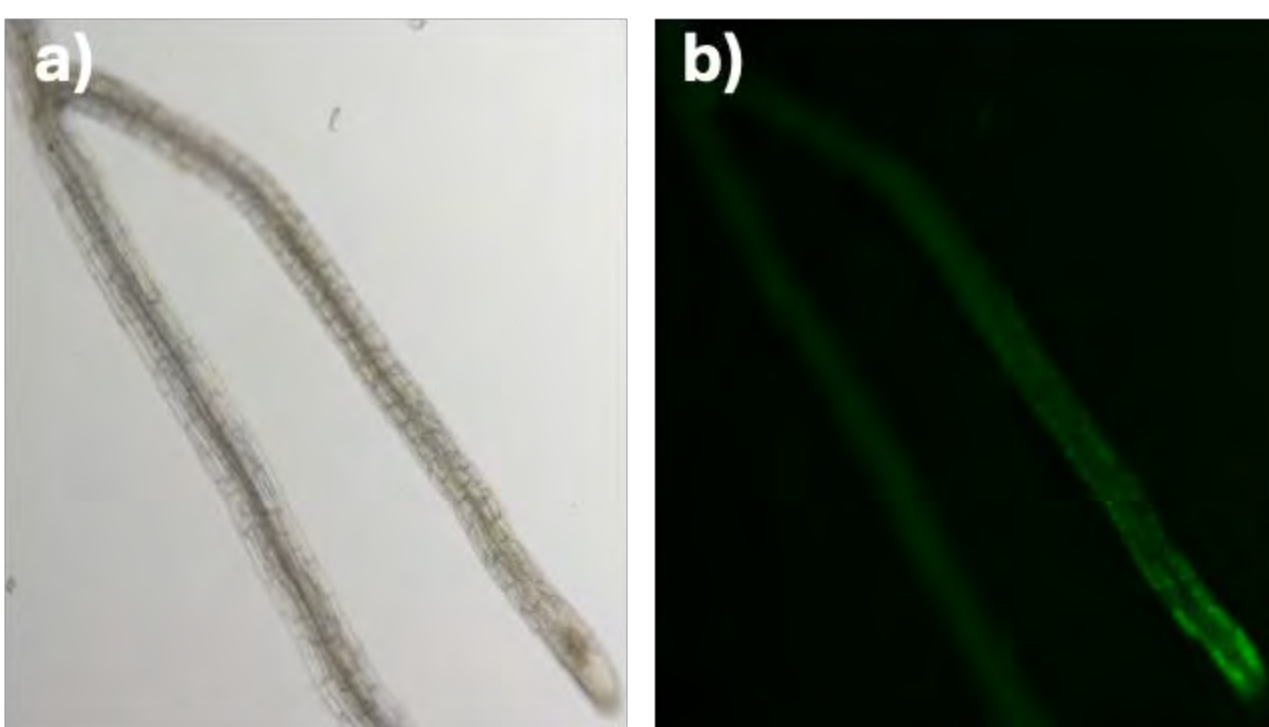
**Fig. 2** demonstrates the identification of possible areas of NR-stained PS microplastic uptake, as visualized by multiple circular regions of intense NR fluorescence that are not similarly intense under DAPI emission. However, due to autofluorescence observed under NR emission, further analytical methods are necessary to produce absolute certainty as to the source of intense fluorescence.



**Fig. 3:** GFP-expressing *A. thaliana* root imaged: (a) Brightfield (b) GFP emission (c) NR emission (d) Enlarged view of PET MF uptake area under NR emission

**Fig. 3** presents a magnified view of an *A. thaliana* root sample grown in NR-stained MF spiked media. **Fig 3. d.** identifies multiple regions of potential MF uptake as indicated by the elongated regions of intense fluorescence under the NR excitation wavelength.

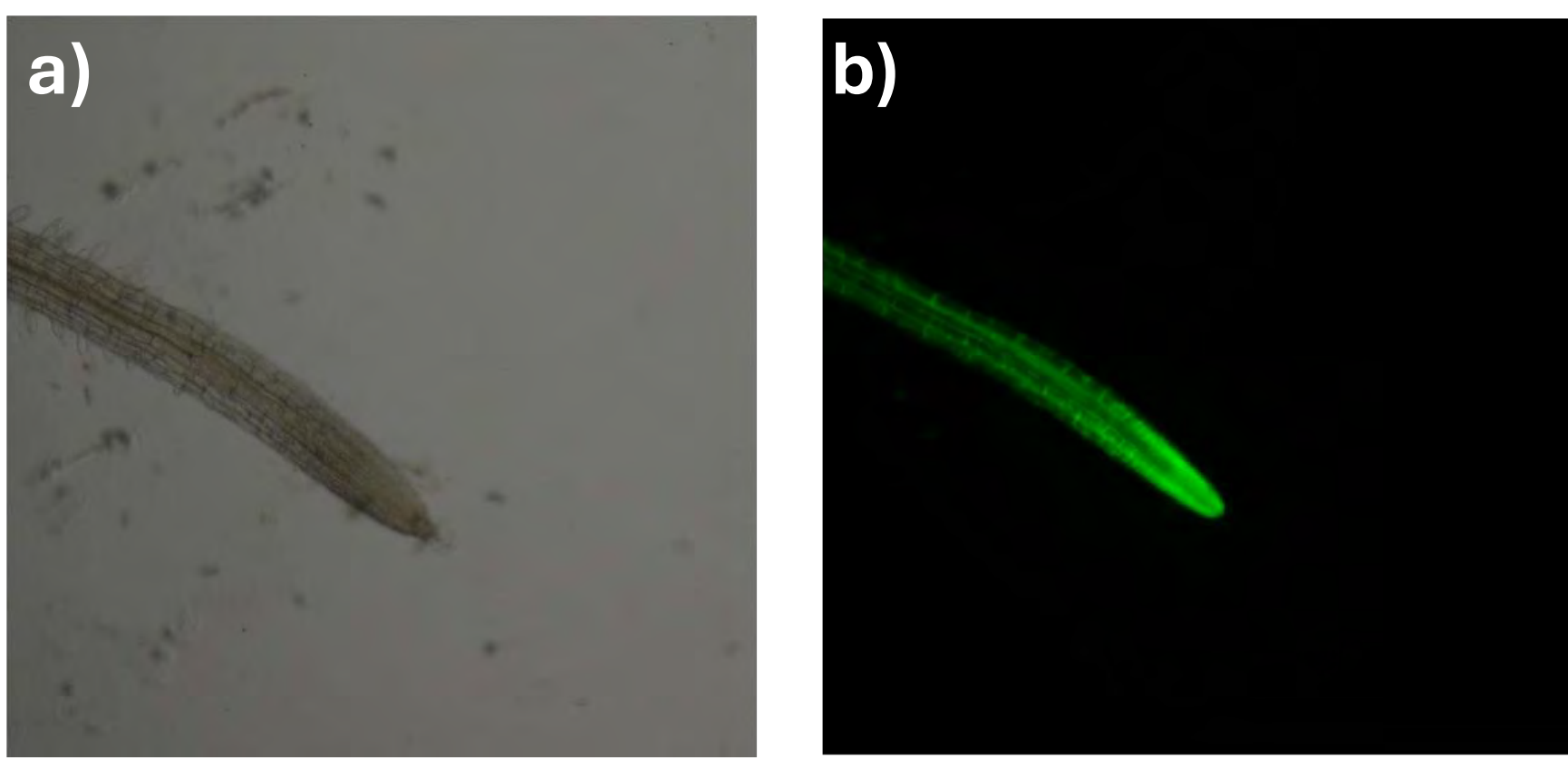
**Fig. 4** shows the GFP fluorescence of a root not exposed to experimentally integrated MPs. No fluorescence was observed under NR emission. GFP expression was evident in all other negative controls planted, though with varying intensity and concentrated fluorescence at root tips.



**Fig. 4:** Negative GFP-expression control root: (a) Brightfield (b) GFP-emission.

## Conclusion + Future Aims

In conclusion, multiple regions of potentially positive MP and MF uptake have been imaged. The introduction of NR-stained PS MPs yielded positive results in the form of multiple areas of circular, high intensity fluorescence in the plant root that were observed only under NR emission. Moreover, when *A. thaliana* was exposed to NR-stained PET MFs, elongated regions of intense fluorescence were observed under NR emission, and not similarly observed under GFP emission. However, due to factors such as the variable autofluorescence of the plant root, further analytical methods are necessary to confidently qualify our results and provide more comprehensive quantitative analysis as to the amount of MP uptake.



**Fig. 5:** Negative control *A. thaliana* root: (a) Brightfield (b) GFP emission

Future aims include expanding on the method of PET MF uptake by employing slightly different experimental conditions that aim to combat uncertainty due to autofluorescence. Major factors in this improvement include imaging roots earlier, in order to retain a high amount of GFP and NR fluorescence, as well as exploring both high concentration and ecologically relevant samples. **Fig. 5** shows preliminary negative controls after approximately 15 days of growth, with clear GFP fluorescence. No fluorescence was observed under the NR emission settings. The improved clarity of the negative controls suggests the viability of the revised method.

## Acknowledgments

We would like to thank Dr. Jennifer Brodbelt and Dr. Andrew Ellington for their support of the BioP lab, as well as the Cummings Lab for providing materials for microfiber production.

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- (3) Stanton, T.; Johnson, M.; Nathanail, P.; Gomes, R. L.; Needham, T.; Burson, A. Exploring the Efficacy of Nile Red in Microplastic Quantification: A Costaining Approach. *Environmental Science & Technology Letters*. **2019**, 6, 606–611. <https://doi.org/10.1021/acs.estlett.9b00499>

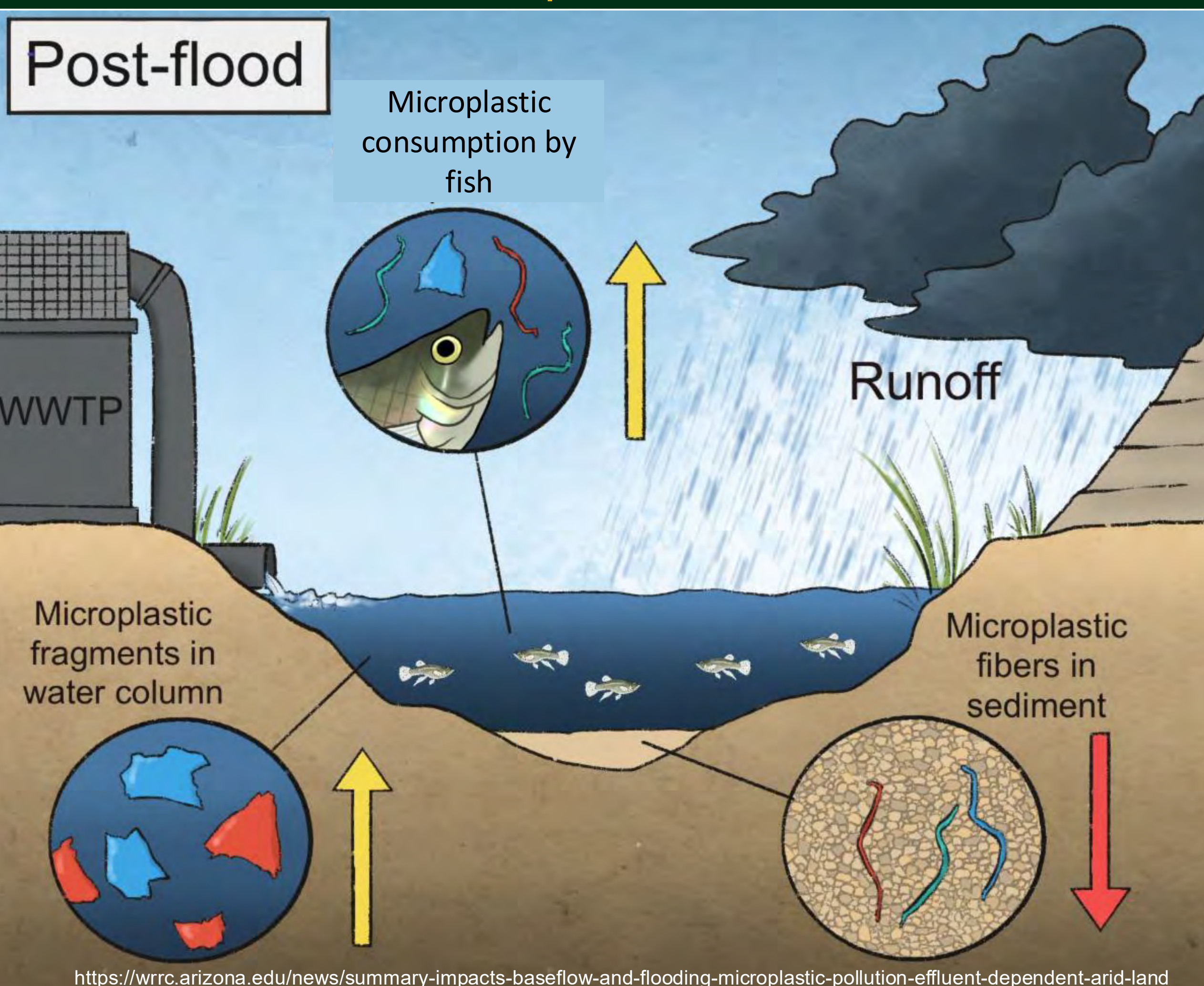


## I. Motivation 1: General sediment transport

Sediment transport affects many natural processes. What are anthropogenic stressors in such systems?



## II. Motivation 2: Microplastics in the environment

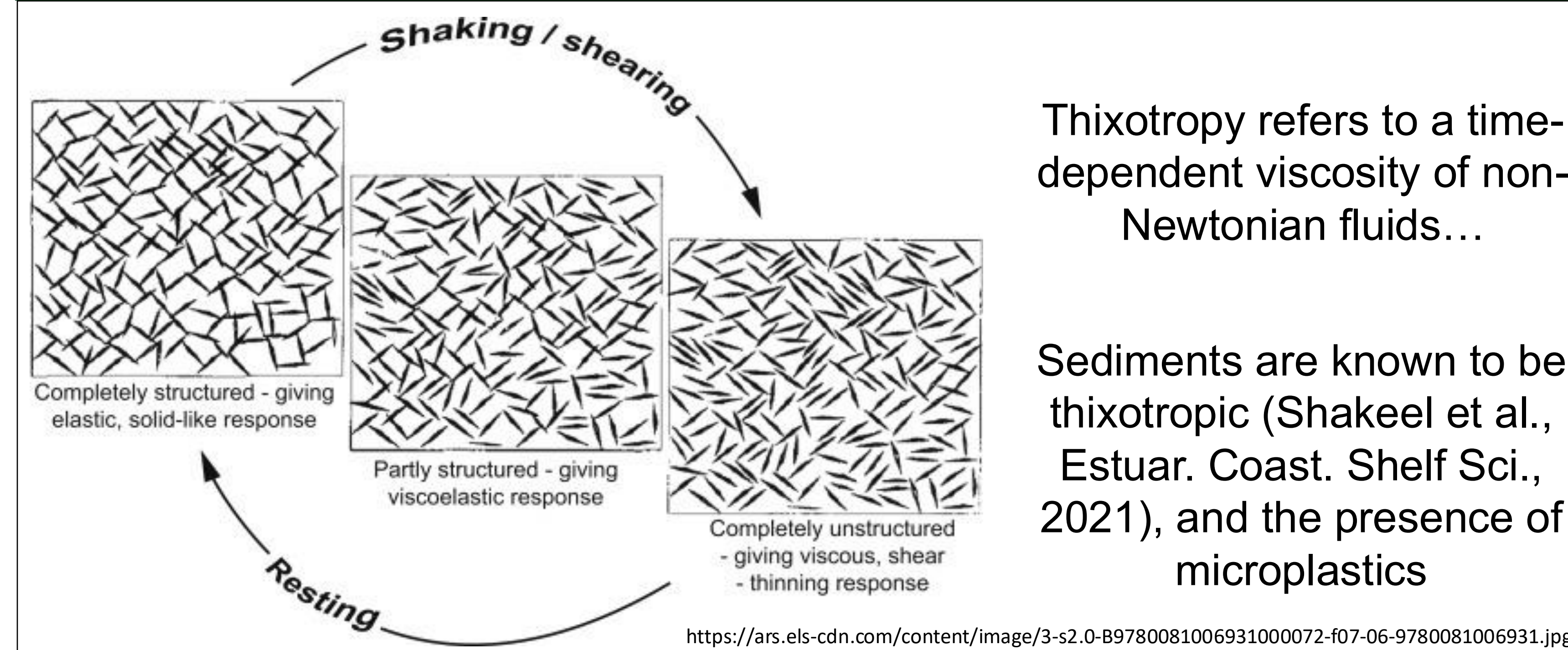


## III. Overview/Burning Question (BQ)

- Wu et al., Environ. Pollut., 2024
  - MPs reduced sediment viscosity and yield stress
  - Potentially increase risk of sediment resuspension
- Wu et al., Sci. Total Environ., 2024
  - Sediment viscosity decreased with MP size and increasing concentration
  - Hypothesized that the reduction in viscosity was due to a decrease in the extracellular polymer substances and the cation exchange capacity of the sediment

BQ: How do MPs affect complex rheological signatures such as thixotropy in sediments?

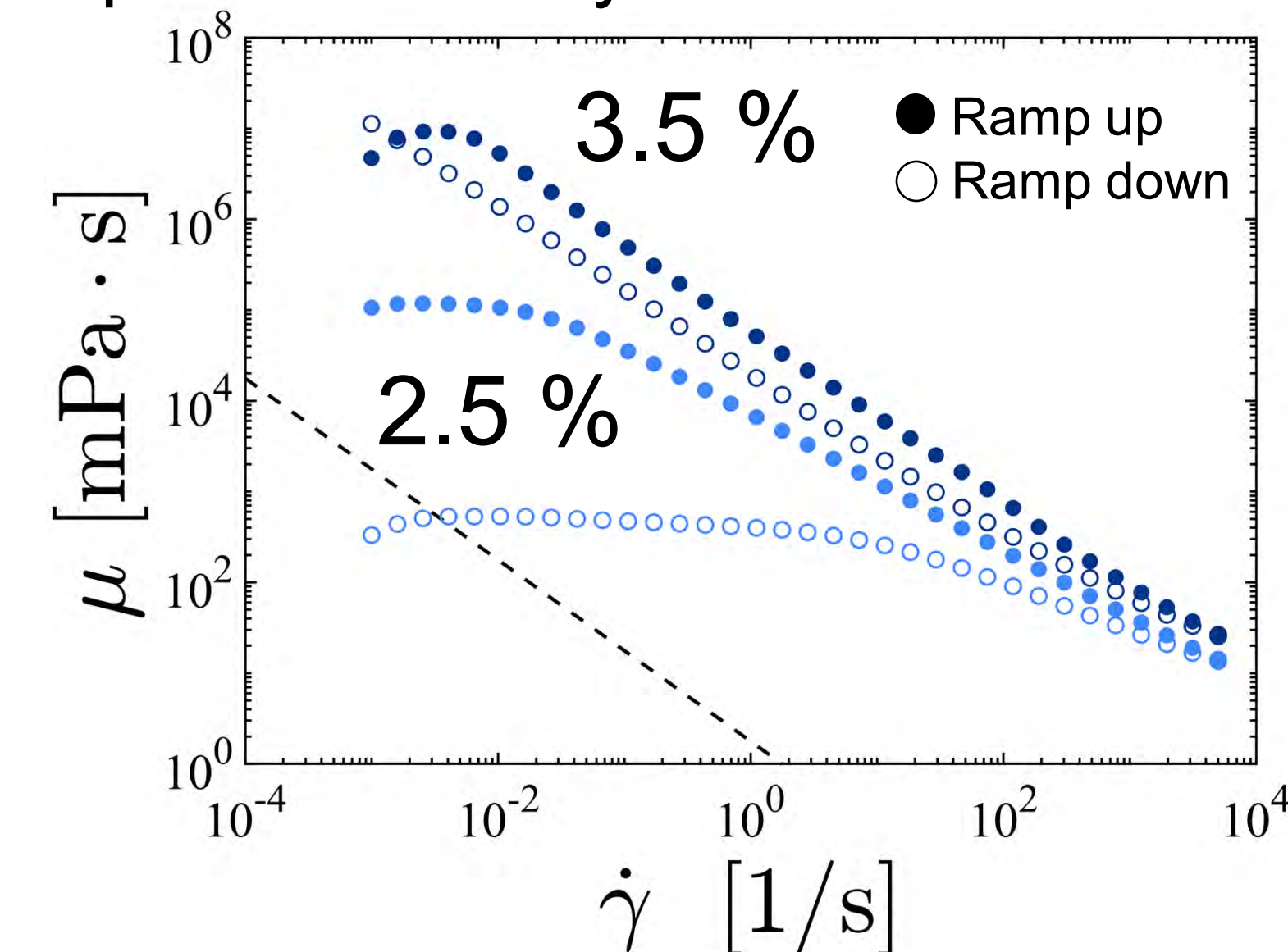
## IV. Background



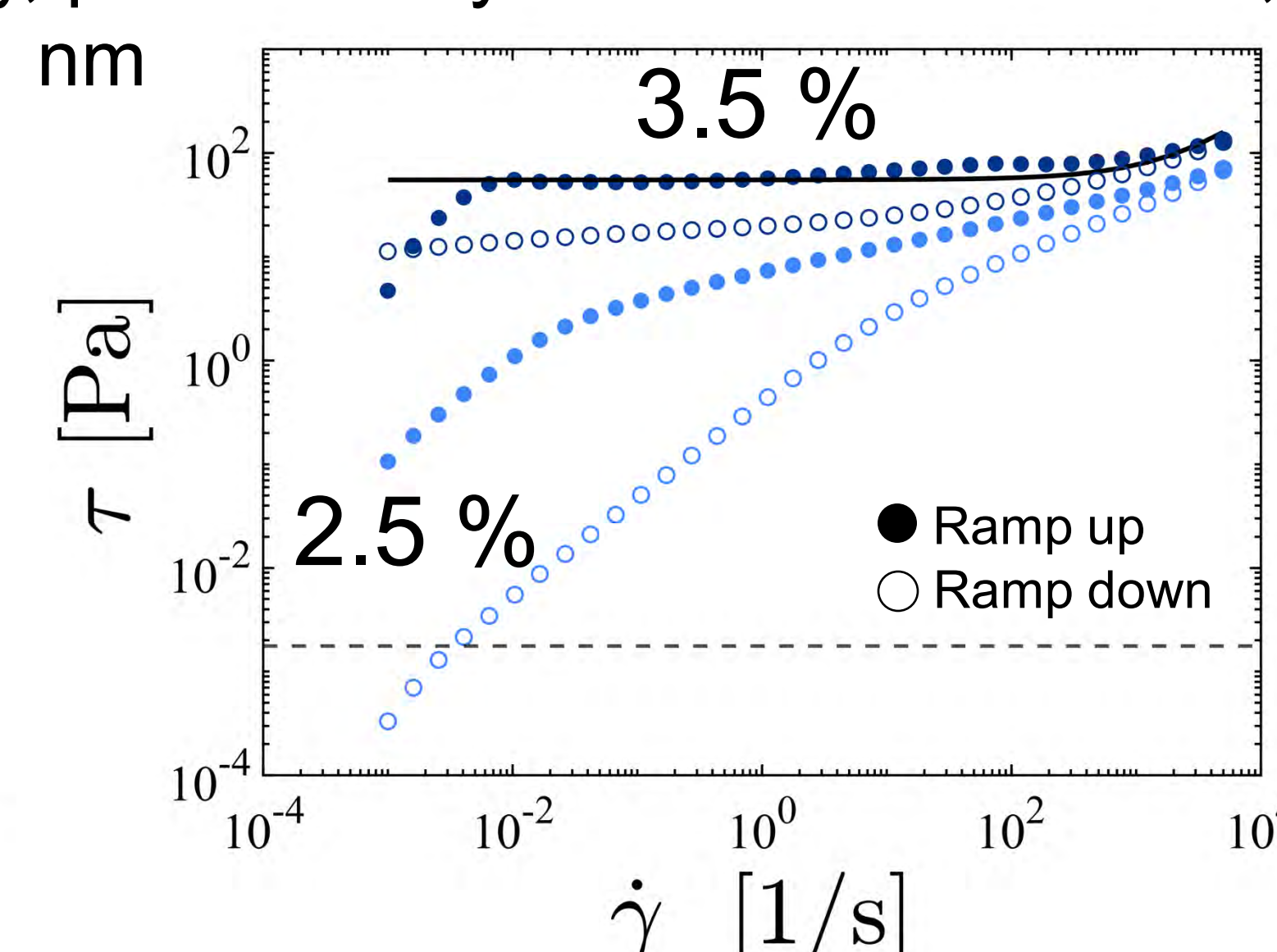
**Hypothesis: MPs may affect the aggregation times of sediments influencing their transport dynamics in nature**

## VI. Methods/Results: Pure Laponite

Laponite RD – Synthetic smectite clay, plate-like crystals w/  $d = 20\text{--}25\text{ nm}$ ,  $h = 1\text{ nm}$



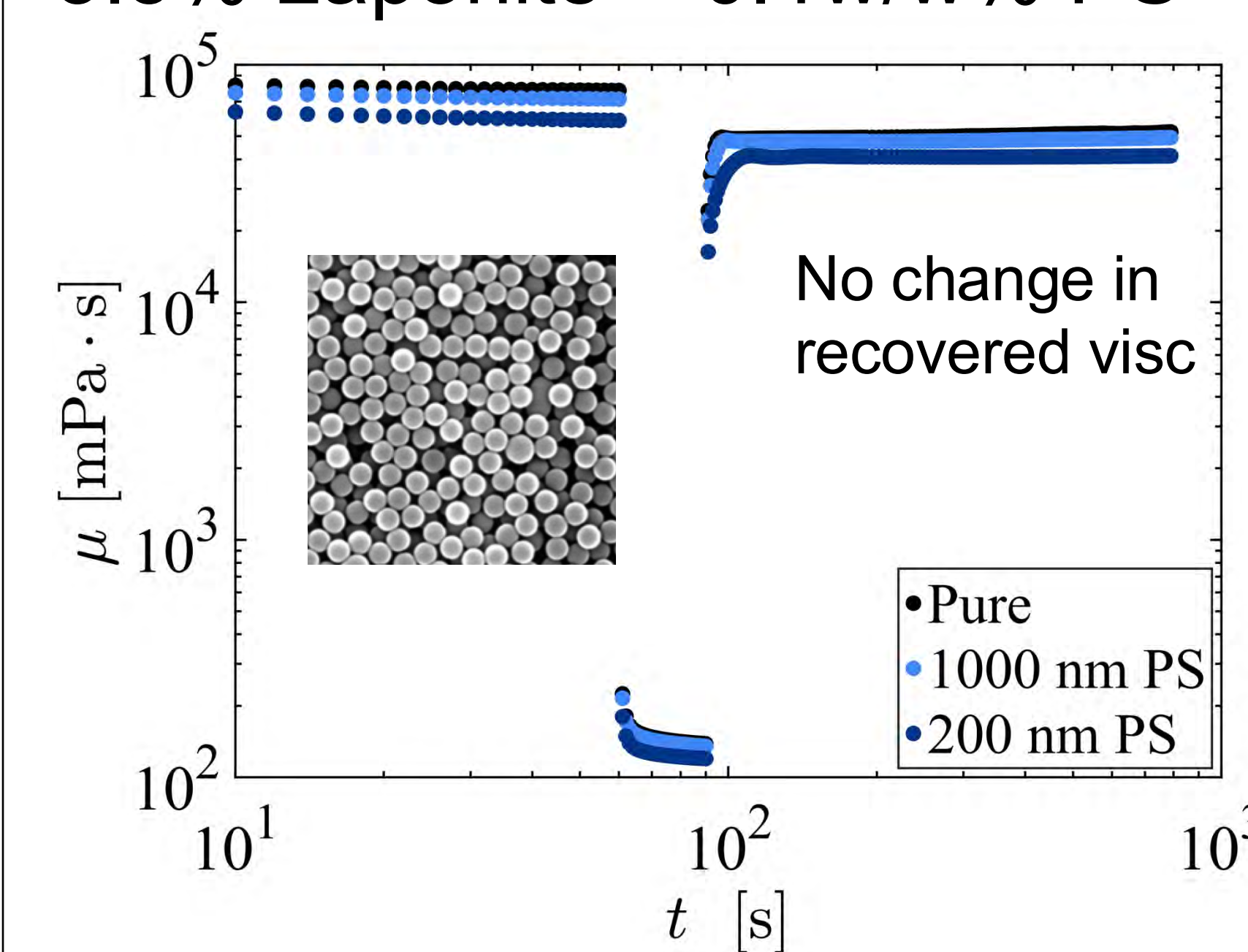
Rheometer - Anton Paar 302e + Cone&Plate geometry (1/10 particle size to gap thickness required)



Solutions prepared with ultrapure DI water and Laponite received from BYK and bentonite from Millipore Sigma

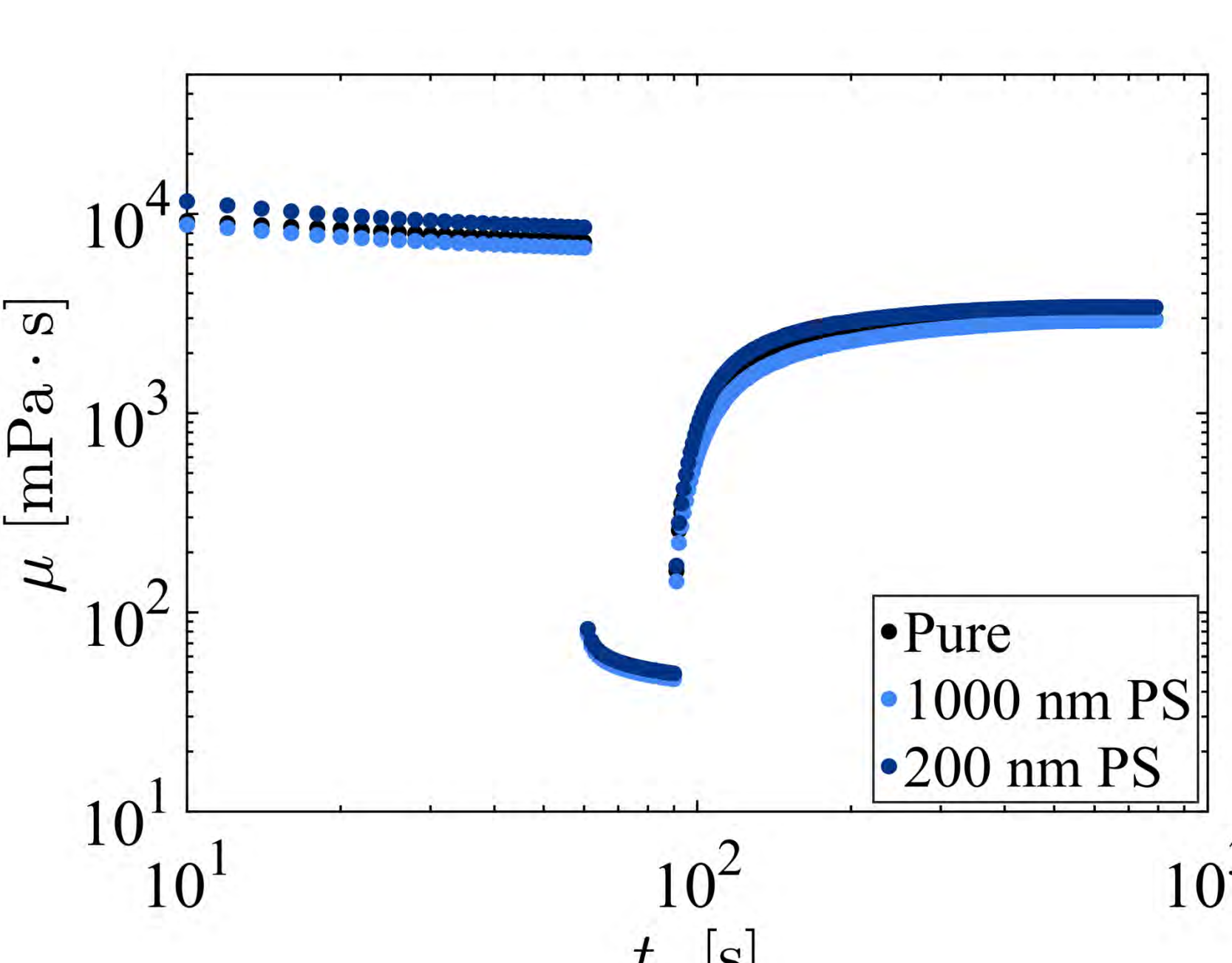
## VII. Results: Clay & Polystyrene (PS) particles

3.5% Laponite + 0.1w/w% PS



- ↓ 5% in  $\mu$  w/ 0.1% 1000 nm PS
- ↓ 19.5% in  $\mu$  w/ 0.1% 200 nm PS

7.0% Bentonite + 0.1w/w% PS

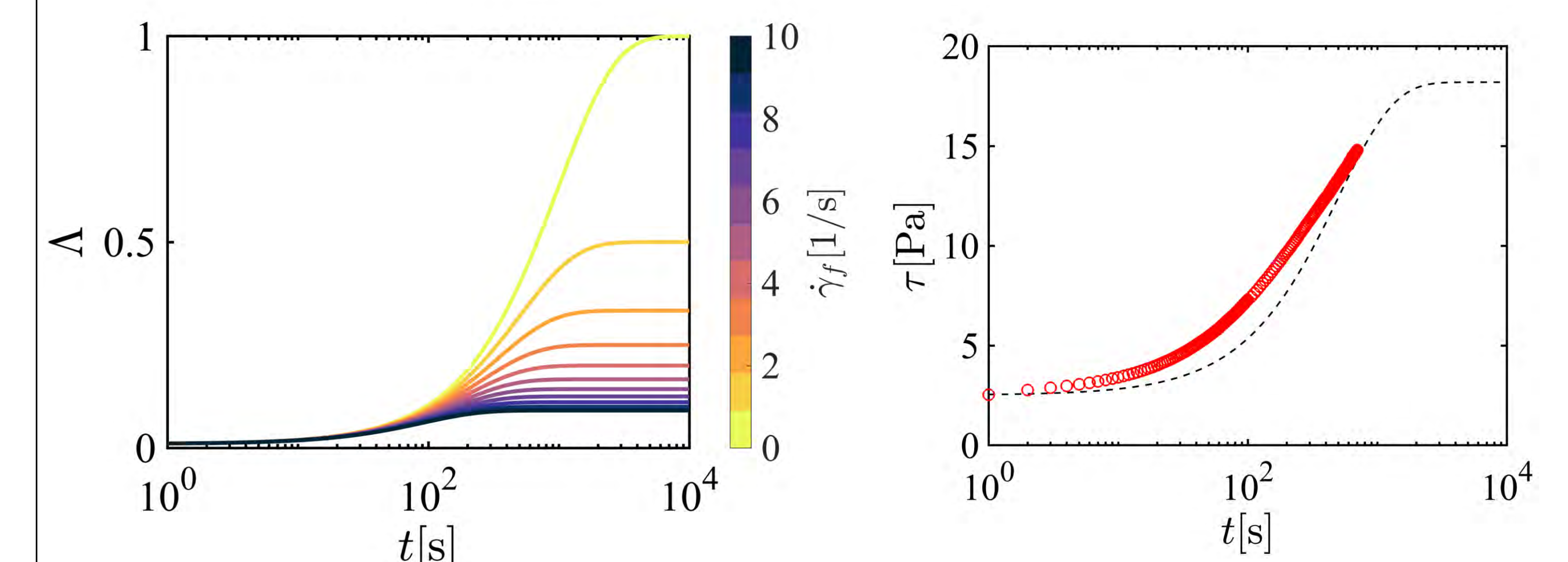


- No change w/ 0.1% 1000 nm PS
- ↑ 30% in  $\mu$  w/ 0.1% 200 nm PS

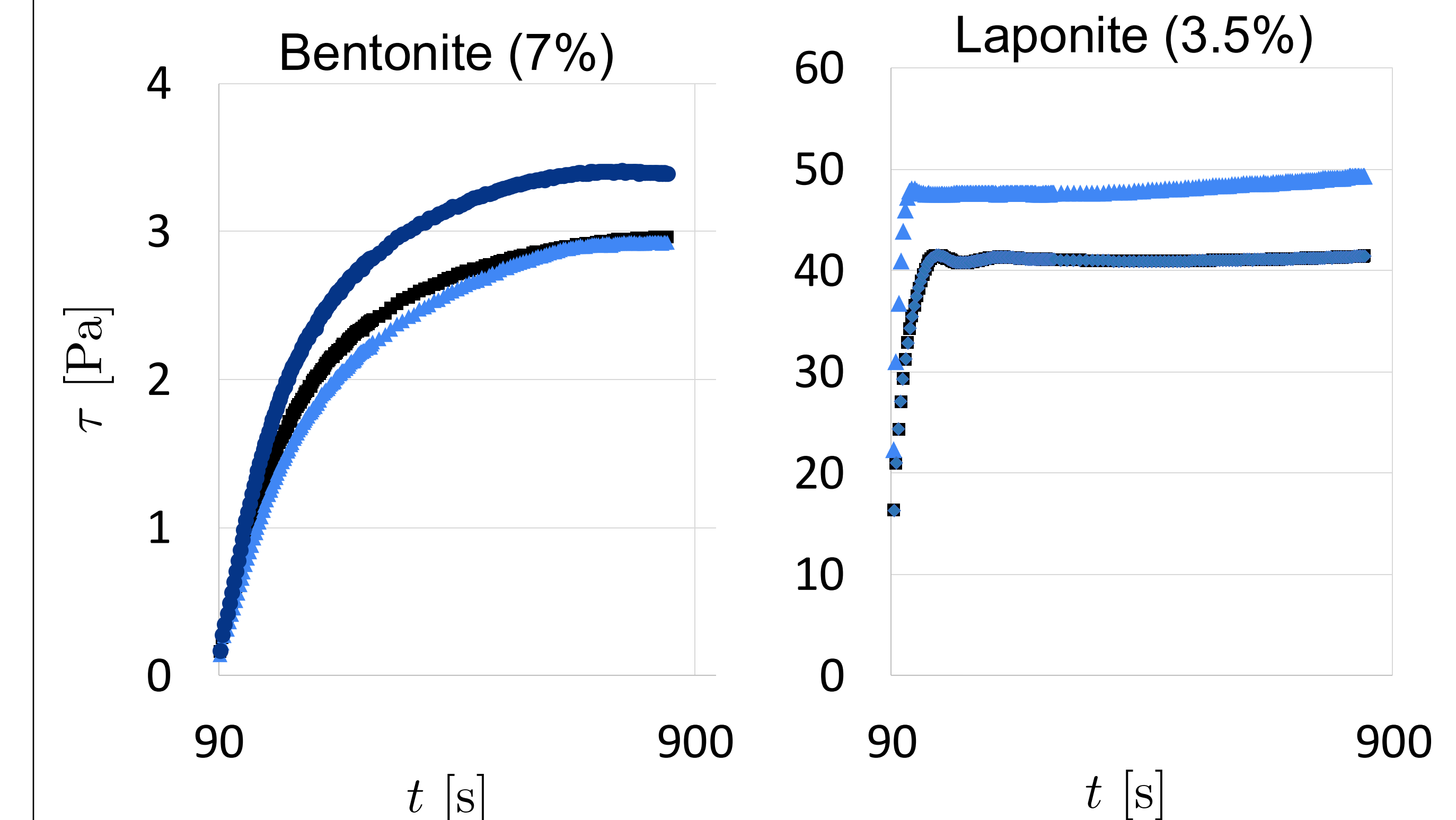
## VIII. Results: Thixotropic recovery & MPs

### Thixotropic kinetic

$$\Lambda(t) = k_A \lambda_T + (\Lambda_{ss} - k_A \lambda_T) \exp(-t/\lambda_T)$$



Shear-induced structure breakdown leads to structure recovery and is limited by the final shear rate



The thixotropic recovery is faster for the 1 micron particle for bentonite but 200 nm for Laponite.

## VIII. Summary

- MPs have a significant influence on the zero-shear viscosity changes to clays in non-trivial ways
- Viscosity dec. with dec. particle size w/ laponite yet viscosity inc. with dec. particle size with bentonite
- The thixotropic recovery rate is greatest for the 1  $\mu\text{m}$  particle case for Laponite
- Future work will probe the thixotropic effects across a large range of particle sizes

## IX. Acknowledgements

- Authors acknowledge Baylor University startup funds for the funding of this work as well as Dr. Christie Sayes, Taiwo Ayorinde, Debora Berti for their help with the microplastics, DLS, and SEM



# Making Space for Migratory Birds: An Urban Conservation Program Highlight

Kiara Carrasco<sup>1</sup>, Chloe Dannenfelser<sup>1</sup>, Liz Virgl<sup>1</sup>, and Nancy Brown<sup>2</sup>

<sup>1</sup>American Bird Conservancy, <sup>2</sup>U.S. Fish and Wildlife Service



## BACKGROUND

Due to its unique position between two major migratory bird flyways and its diverse range of ecosystems, the Houston-Galveston region plays a crucial role in supporting migratory birds. The region faces challenges from a growing human population, with one major concern being the accumulation of trash. This pollution degrades habitats, poses ingestion and entanglement risks, and threatens the overall health of coastal and urban ecosystems. With over 7 million residents in the greater metropolitan area, it is essential to create and maintain bird-friendly spaces to support the millions of migratory birds that pass through each year.



## URBAN BIRD TREATY PROGRAM

The **Urban Bird Treaty** is a collaborative partnership program between U.S. cities and the U.S. Fish and Wildlife Service with the goal of protecting urban bird populations.

The treaty was signed in Houston on April 18, 2003.

The four main program focus areas include:

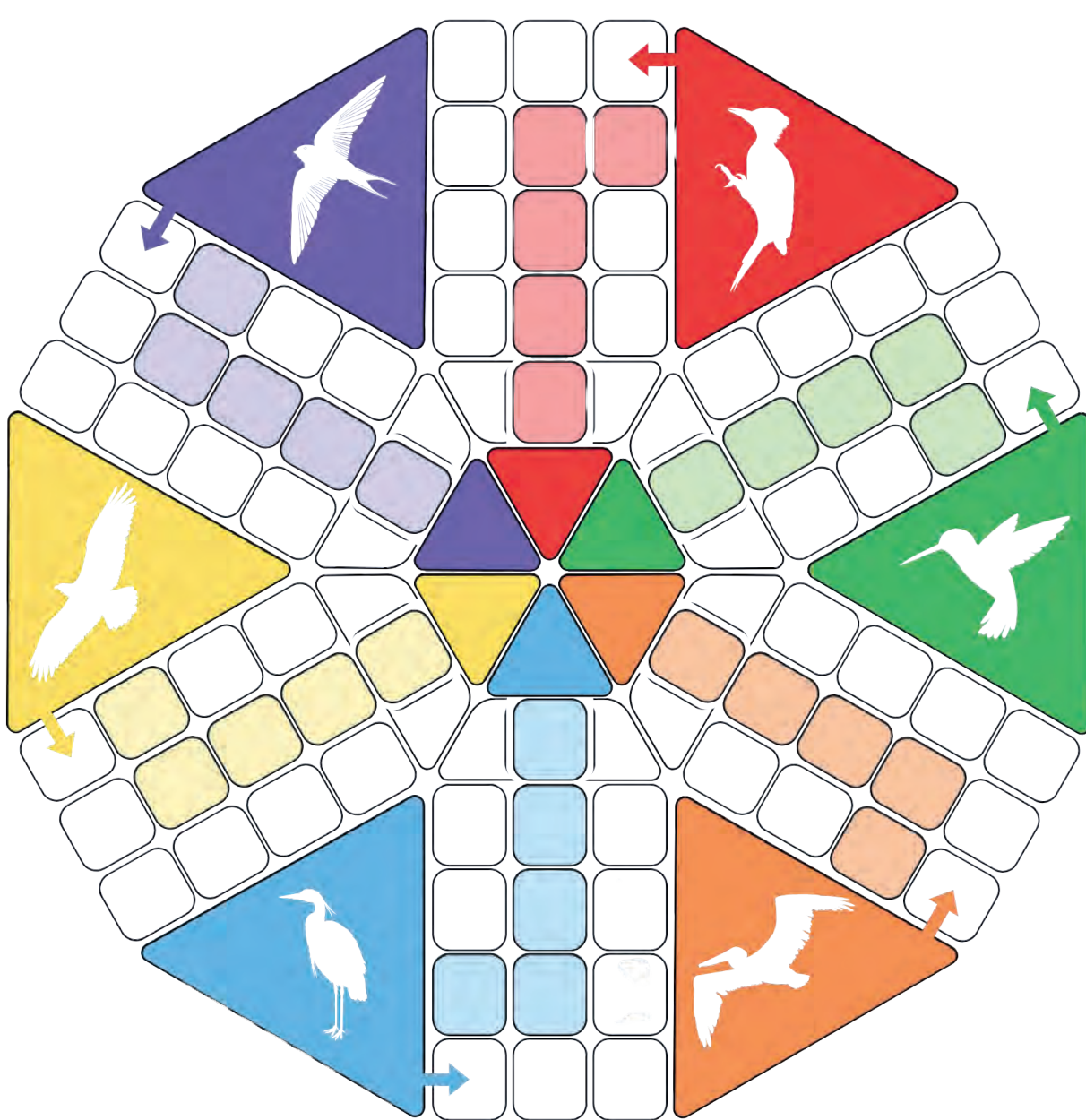
- Habitat Conservation**
- Community Engagement**
- Hazard Reduction**
- Grant Program**

American Bird Conservancy received funding in 2023 through the NFWF Five Star and Urban Waters Restoration Program.



## CURRICULUM HIGHLIGHT

### Migratory Bird Board Game



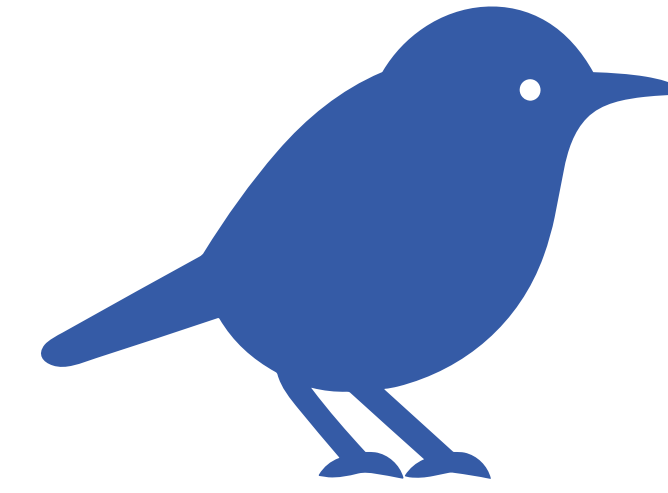
## ABC PROJECT OBJECTIVES AND CURRENT OUTCOMES



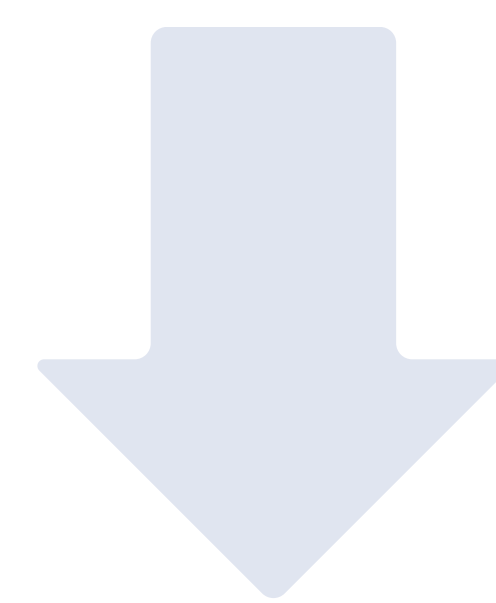
- Hire Conservation Fellow for migratory bird-related education and outreach programming.



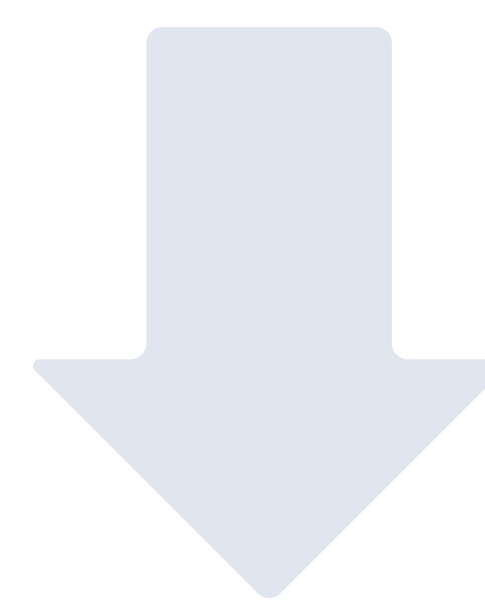
- Host beach and bayou cleanups to remove trash and restore habitat.



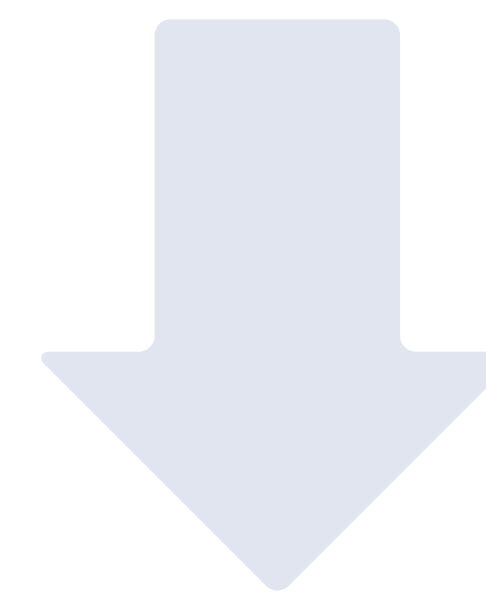
- Coordinate fall and spring bioblitz.
- Develop bird-focused curriculum for students.



- 76 cleanup volunteers.
- 138 students engaged.
- 404 reached from outreach events.



- 2383 lbs of trash removed.
- 38 acres of habitat restored.



- Upcoming bioblitz on May 10, 2025 in celebration of World Migratory Bird Day.

## LEARN MORE

Urban Bird Treaty Houston



Education



Cleanups



Bioblitz



Stay connected to SPLASH



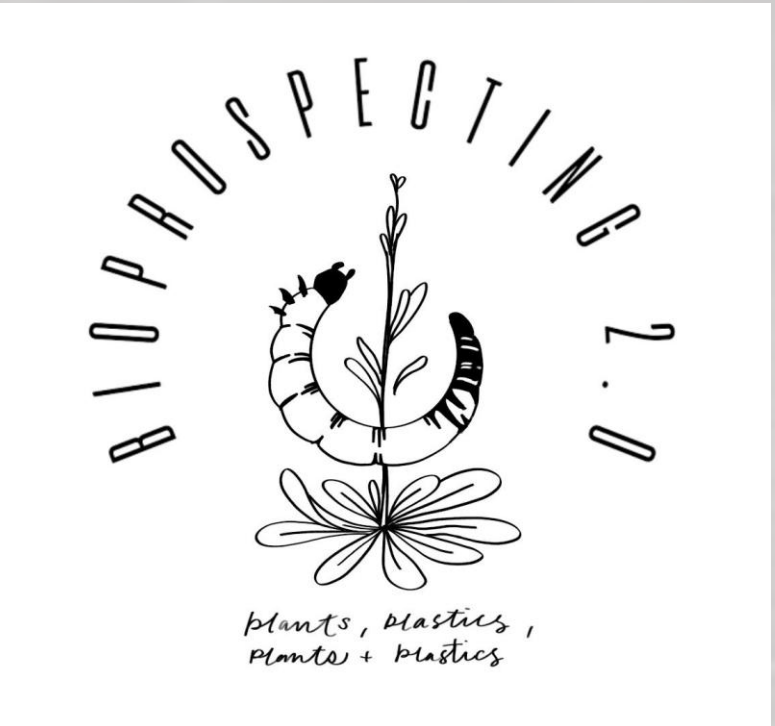
## ACKNOWLEDGEMENTS

This work was supported by the National Fish and Wildlife Foundation Five Star and Urban Waters grant program with guidance from U.S. Fish and Wildlife Service. The authors gratefully acknowledge the contributions of the American Bird Conservancy and Gulf Coast Bird Observatory SPLASH program staff.



# Microbial Marvels: Investigating Dubia Roach Microbiota in Relation to Polyethylene Biodegradation

Roland Quinones, Kasia Dinkeloo  
BioProspecting 2.0 Stream, Freshman Research Initiative, The University of Texas at Austin



## Background

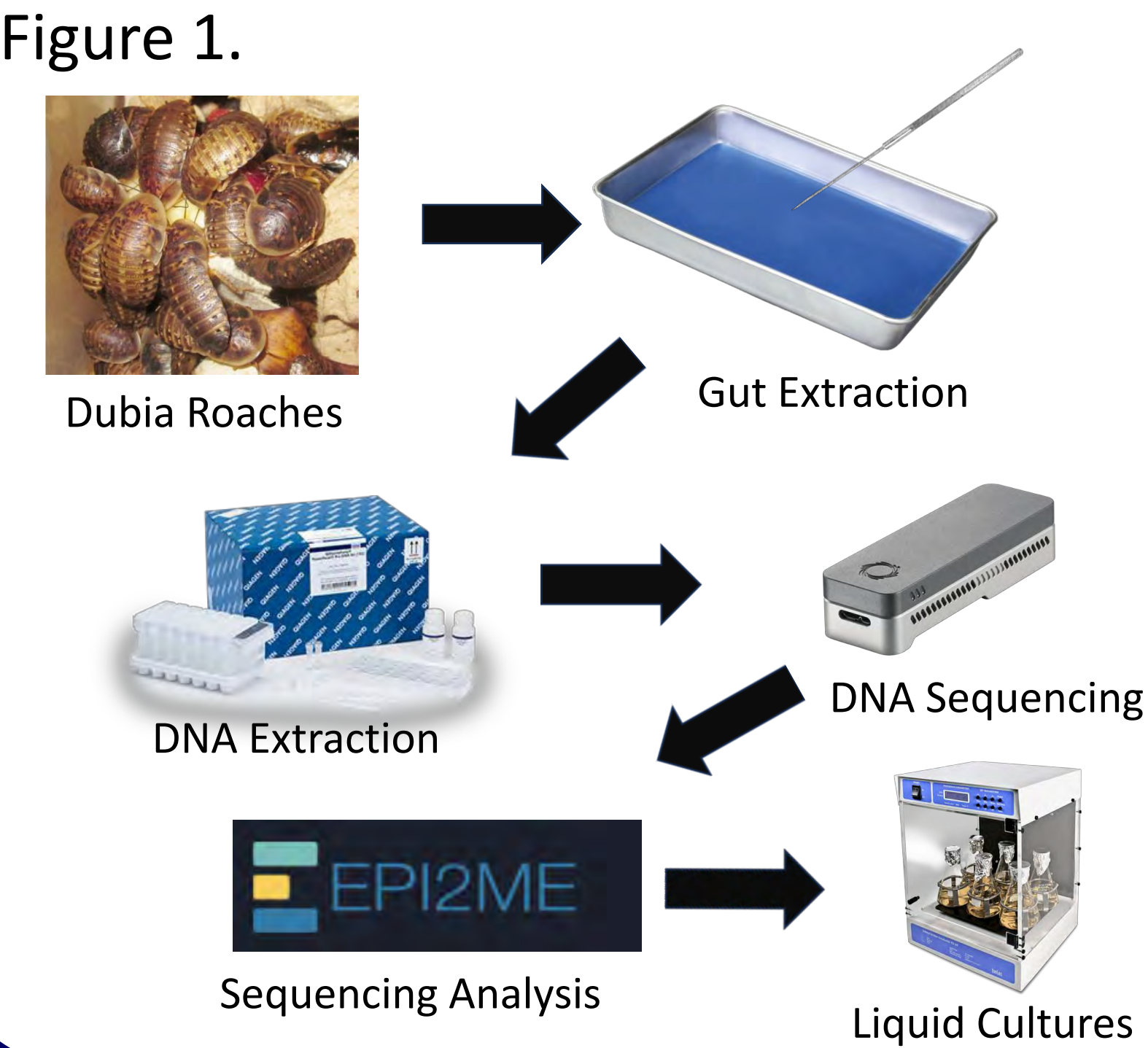
Our planet is being embalmed in plastic, and nature is suffocating under its overwhelming weight. To date, over 8.3 billion metric tons of plastic have been produced, but less than 10% of this amount has actually been recycled.<sup>2</sup> As plastic waste continues to infiltrate Earth's ecosystems, researchers are increasingly focusing on biologically mediated methods of plastic degradation as a potential solution to this growing crisis.

Among the many organisms being studied for their role in plastic biodegradation, several insect species have emerged as promising candidates, with *Zophobas morio* leading much of the current research.<sup>5</sup> Although this species has shown significant potential, studies involving other hexapod invertebrates remain limited. This is particularly true for species in the order *Blattodea*, which includes both cockroaches and termites.

This project aims to highlight the biodegradation potential of *Blaptica dubia*. A recent study suggests that these cockroaches might have the ability to degrade polystyrene, but beyond this, very little is known about their role in the breakdown of plastics in general. The goal of this research is to demonstrate that *Blaptica dubia* can ingest and biodegrade polyethylene, as well as to discover how this impacts the gut microbiota composition of the species.



## Methods



## Dubia Roaches

*Blaptica dubia*, or more popularly known as the “dubia roach,” is a medium-sized roach species native to several countries in South America. This species is very popular in the exotic pet trade and has become a staple feeder insect due to its hardiness, nutrition, and how prolific it is.<sup>1</sup> Going through incomplete metamorphosis, nymphs of this species go through several instars until eventually reaching sexual maturity. The species itself is ovoviviparous, with adults being between 1-2 inches in length. Sexual dimorphism is also observed in dubia roaches, as males exhibit long wings, a characteristic not seen in females.<sup>1</sup>

Widely overlooked in terms of their biodegradation capabilities, one recent study suggests that these roaches harbor a unique gut microbiome capable of degrading plastic. Although that study focused entirely on the biodegradation of polystyrene, it does serve to lay a foundation for the bioprospecting research for this species as a whole.<sup>4</sup>



## Data

Figure 2.

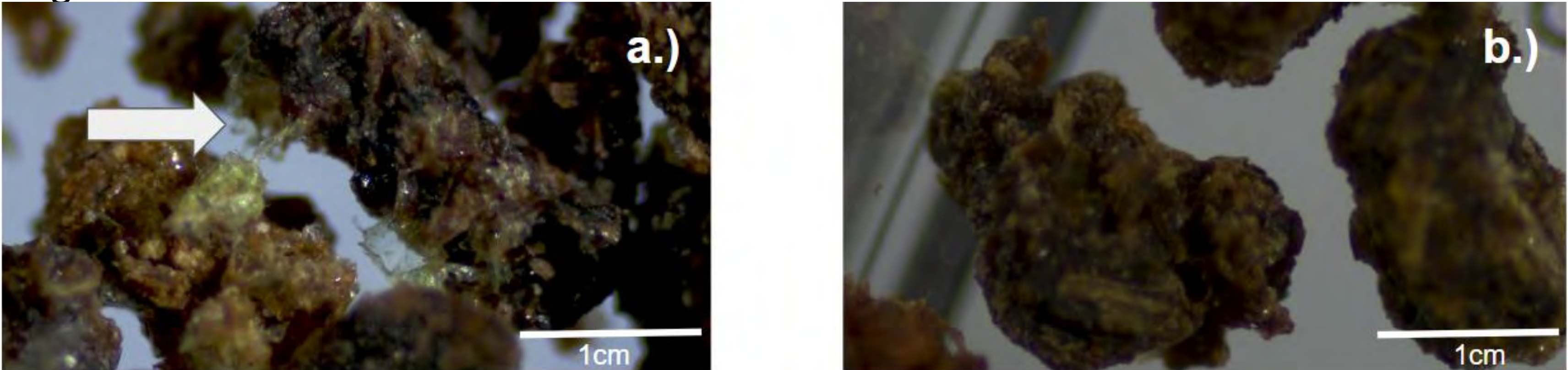


Figure 3.

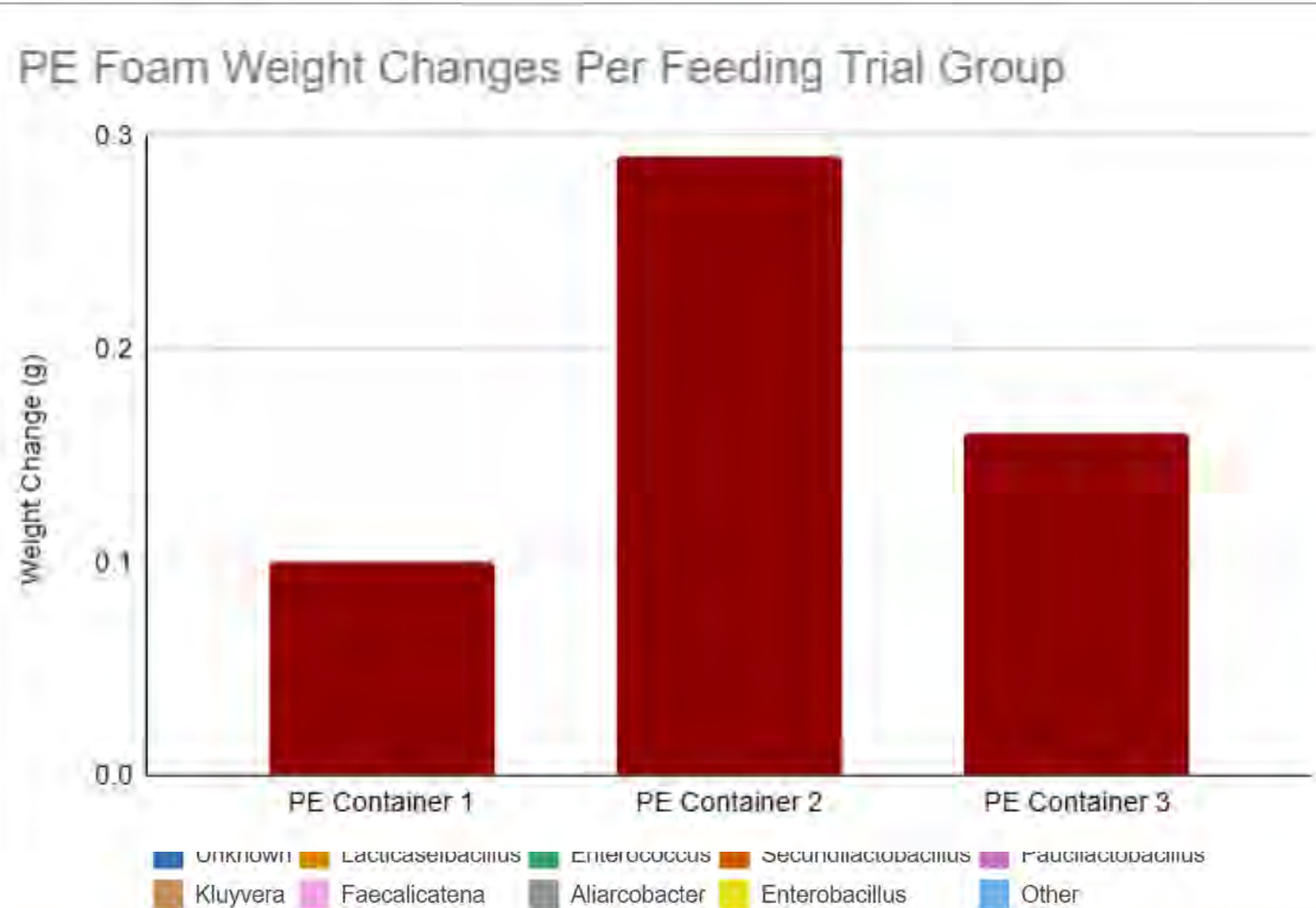


Figure 4.

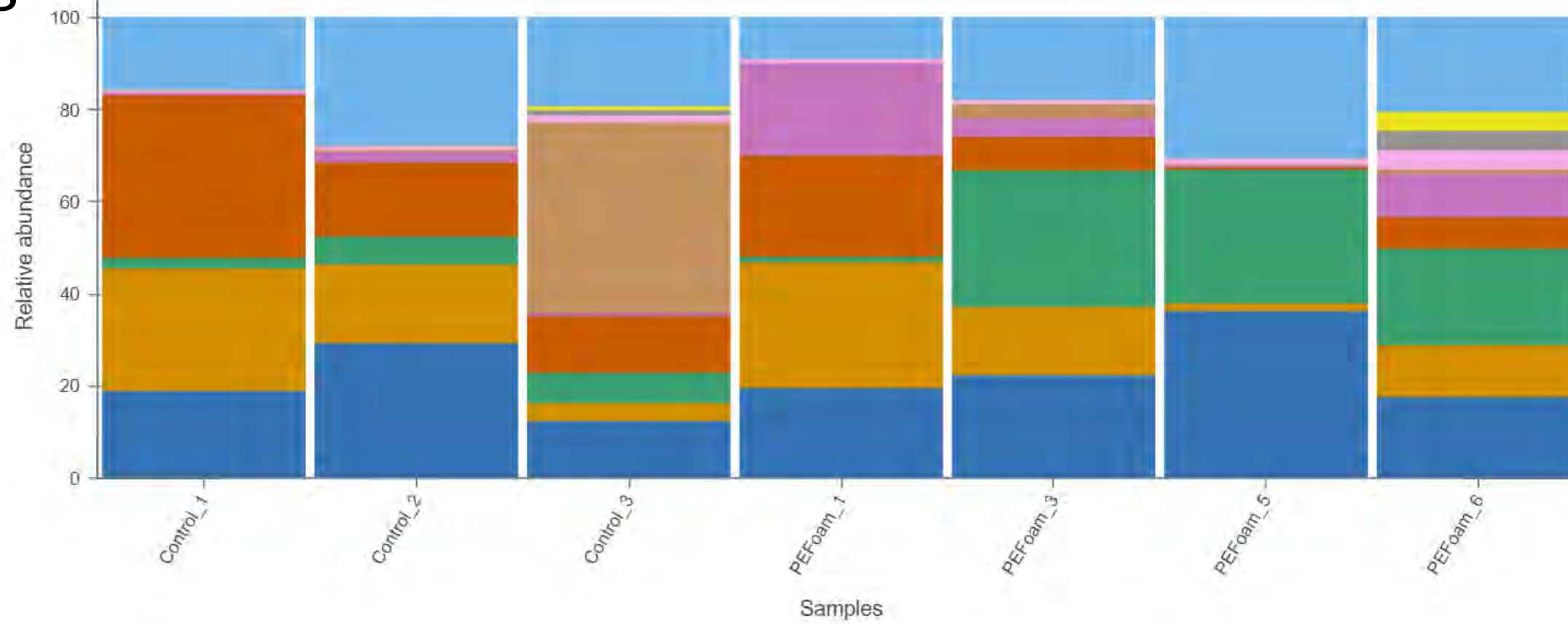
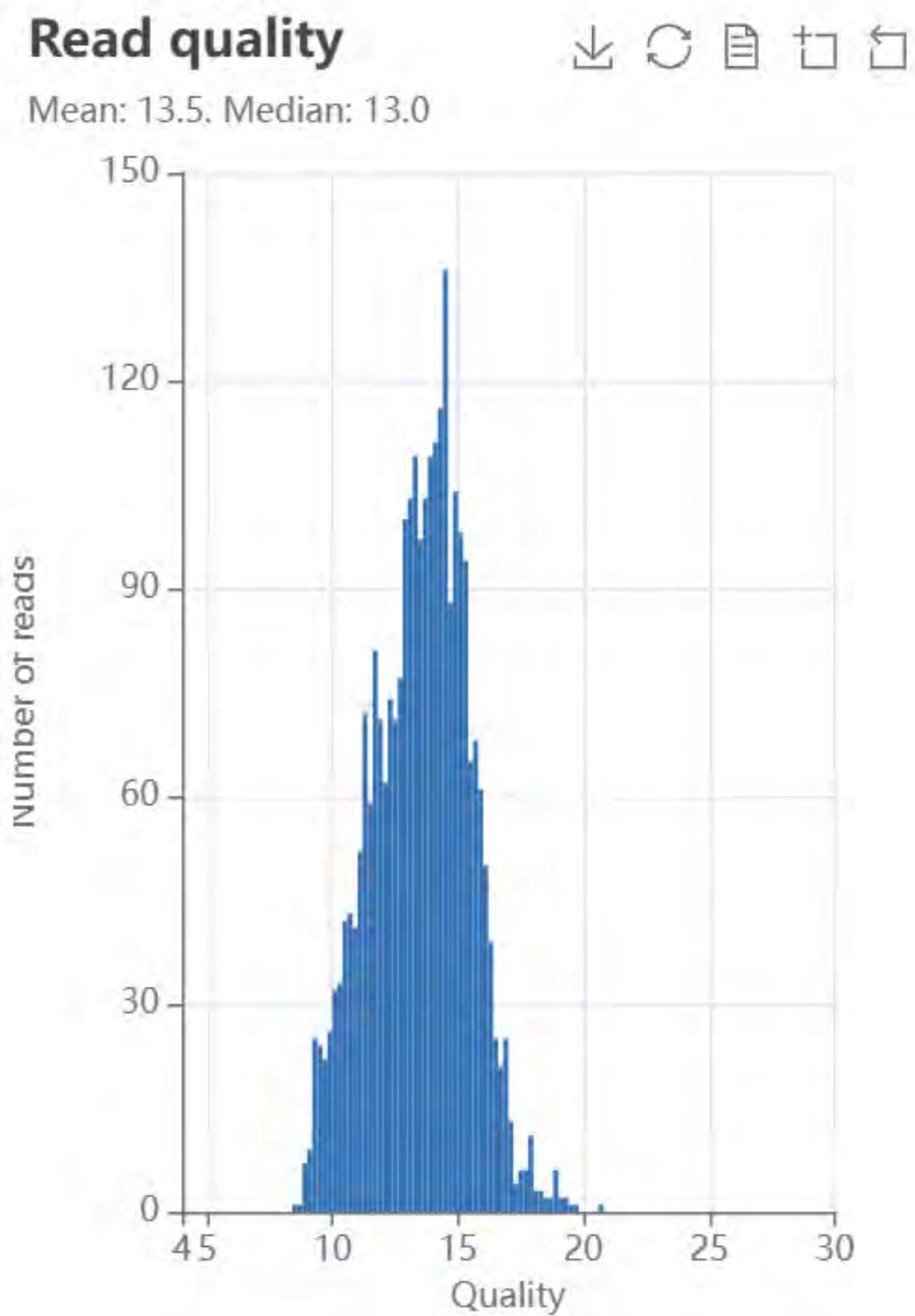


Figure 5.



## Analysis and Future Direction

Once the guts were extracted from the dubia roaches used in the feeding trials, the DNA was extracted and sequenced using Oxford Nanopore Tech's Minion and EPI2ME software (Figure 1). In our analysis, we observed that the cockroach frass contained pieces of a foam-like material (seen in figure 2a.). This indicates the ingestion of polyethylene by said roaches. This is a unique characteristic that is not seen in normal frass (control frass pictured in figure 2b.). From this data, as well as the weight changes observed in the PE foam pre- vs. post-feeding trials (figure 3), we can indeed conclude that these roaches will ingest polyethylene foam.

To analyze the impact of such a diet on these roaches, we ran metagenomic sequencing using Oxford Nanopore Tech's Minlon sequencer and analyzed the data through EPI2ME software. Through sequencing, and as seen in figure 4, we found that the representation of *Enterococcus spp.* increased by 25% in pre- vs. post-feeding trials. This is an interesting observation, as some species in this genus have previously been studied for their ability to thrive in polyethylene-rich environments.<sup>3</sup> Considering this and the moderately decent mean 13.5 Q-score (figure 5), this finding suggests that the PE-based diet of the cockroaches may be setting up the conditions for the growth of these specific microbes.

As a future direction for this study, we plan to isolate and characterize strains of microbes from the guts of these roaches. This will be accomplished through selective LDPE liquid cultures. By isolating these bacteria, the microbes can be studied under more controlled conditions. From here, a much more thorough analysis of the microbes themselves will be performed. Furthermore, it would also prove useful to repeat the entire experiment under the same conditions to confirm the consistency of the results. This approach would serve to validate the conclusions from this study and provide a foundation for future research into this species as a whole.

## Acknowledgements

I would like to thank HEB for making this research possible. I would also like to thank user *OxDionysus* from Arachnoboards.com for providing the post that gave inspiration for this study.



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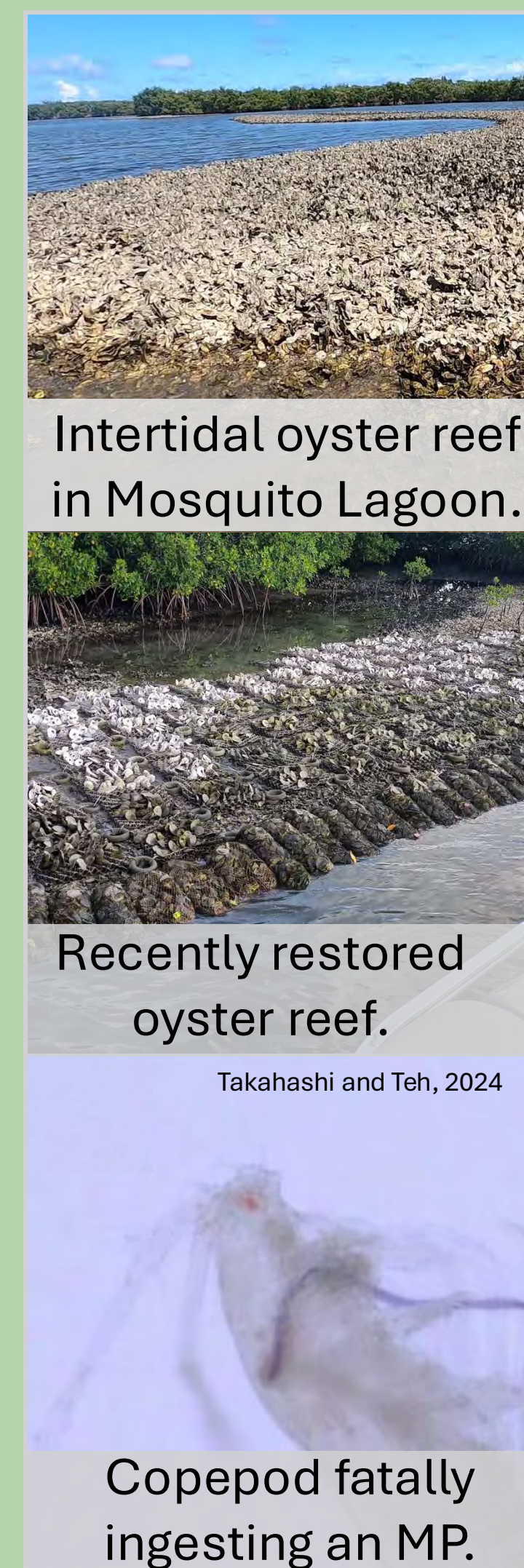


## Introduction

- Oyster reefs are declining globally<sup>1</sup> with 62.6% lost in Mosquito Lagoon, Florida (ML) since 1943.<sup>2</sup>
- Plastic restoration materials may shed microplastics (MP) into waterways, leading to interest in non-plastic alternatives.<sup>3</sup>
- MPs that are ingested can be fatal to small organisms by blocking their digestive tract.<sup>4</sup>
- Efficacy and microparticle shedding in non-plastic restoration materials is poorly understood.

### Research Questions

1. Do non-plastic restoration materials shed microparticles?
2. How durable are non-plastic restoration materials?



## Methods

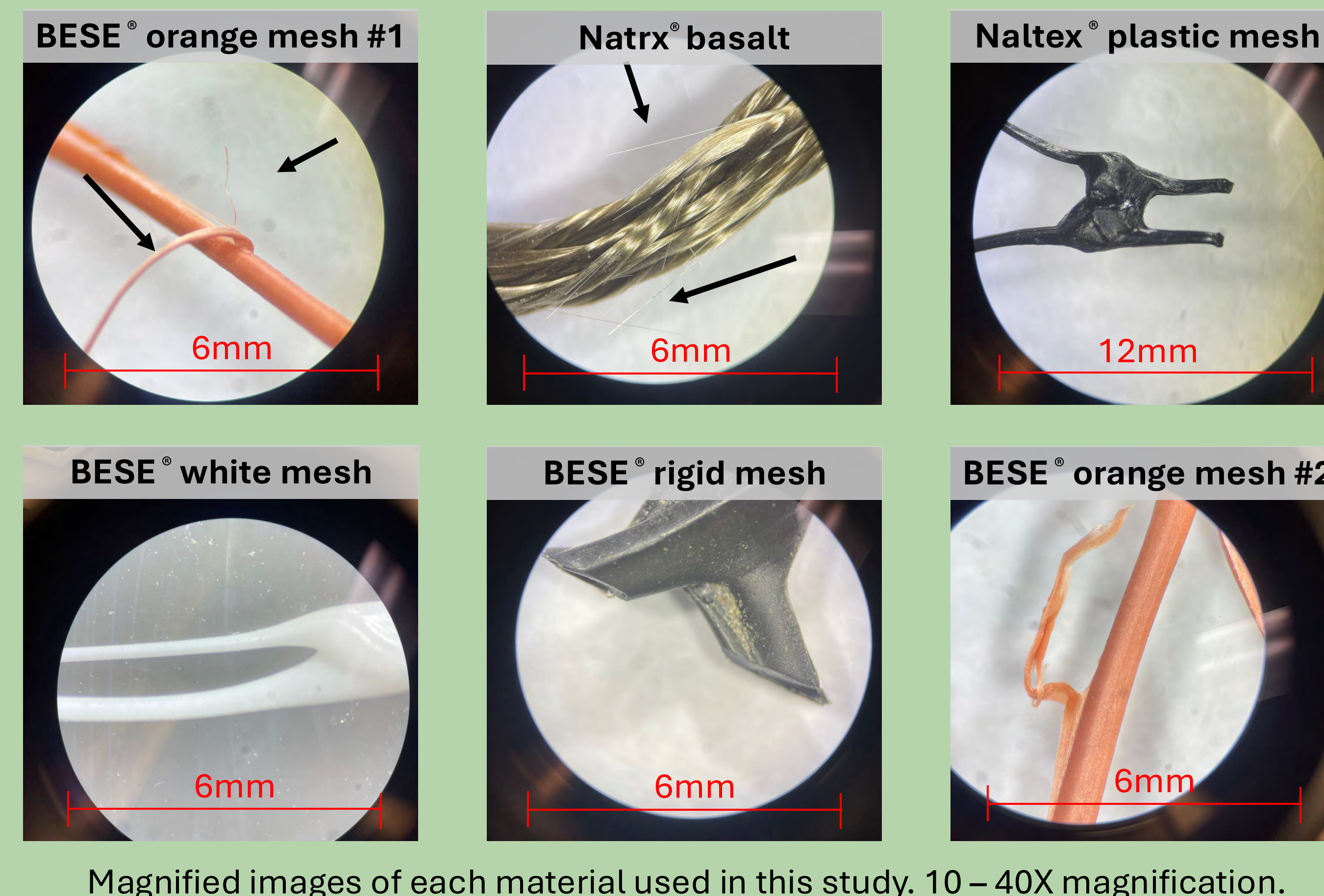
### Laboratory Experiment

We tested four types of BESE<sup>®</sup> biopolymer mesh and pre-soaked and unsoaked Natrx<sup>®</sup> basalt bags, using Naltex<sup>®</sup> plastic mesh and flasks containing only water as controls.

1. Squares (5 x 5 cm) of each material were placed in 150mL Erlenmeyer flasks of 30 ppt artificial seawater on a shaker table to simulate field conditions, with five replicates for each material.
2. Contents of the flasks were vacuum filtered every four weeks. Flasks were then refilled with new water.
3. Microparticles were counted and measured on gridded filter paper using a microscope at 40x magnification.
4. Differences between treatments were assessed using Kruskal-Wallis tests and Wilcoxon signed-rank tests.

### Field Data Collection

We deployed the above materials on oyster reefs in May 2024 and examined them after seven months to measure durability in materials by measuring number and dimensions of tears.

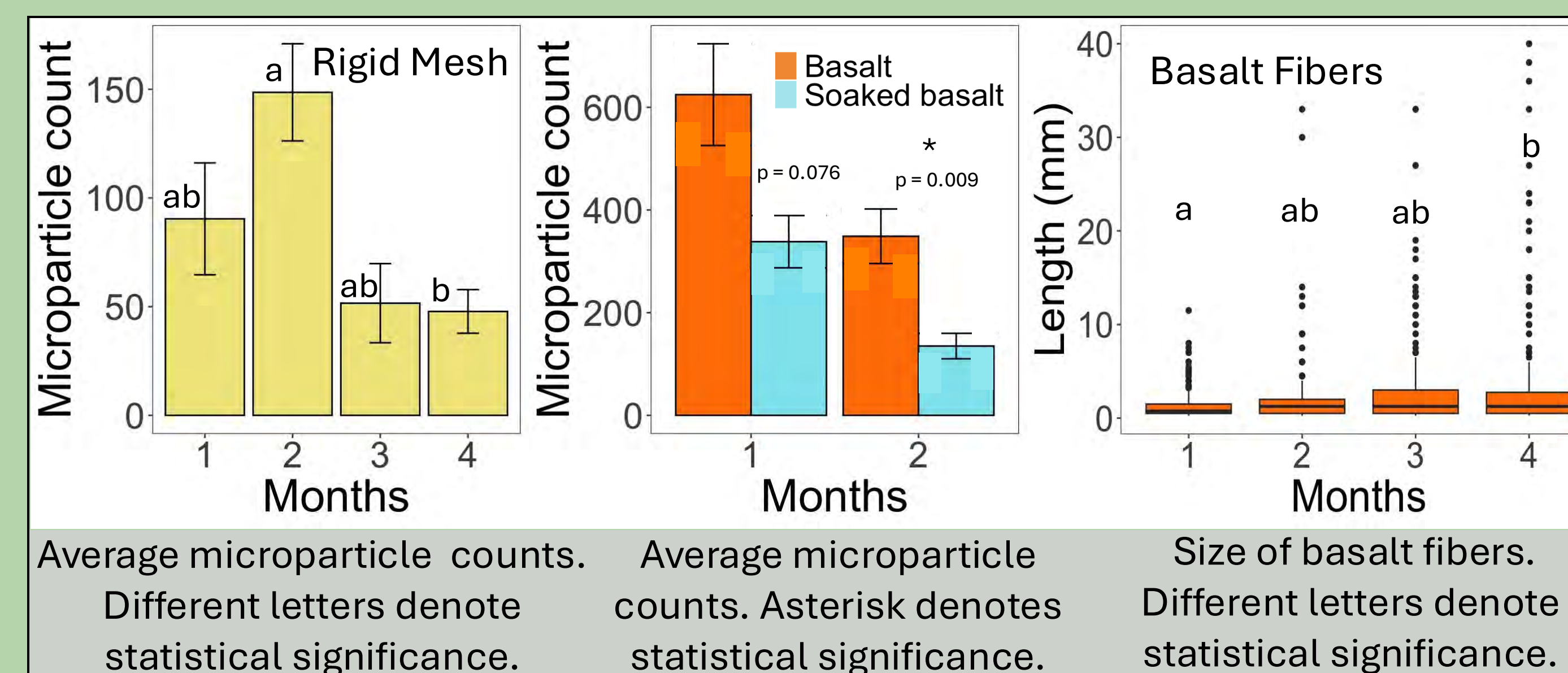


Magnified images of each material used in this study. 10 – 40X magnification.

## Preliminary Results

### Laboratory Results

- Most materials stopped shedding particles after one month, but Natrx<sup>®</sup> basalt bags and BESE<sup>®</sup> rigid mesh continued to shed throughout the study.
- Basalt microfibers significantly increased in length over time.



### Field Results

- All materials experienced some damage after deployment.
- BESE<sup>®</sup> white mesh and BESE<sup>®</sup> rigid mesh had the largest tears.

Summary of damage per shell bag after seven months in field. Numbers presented are mean ( $\pm$  SE). White mesh bags completely disintegrated after seven months.

Material	Number of tears	Tear length (mm)
Orange mesh 1 (n = 30)	4.6 $\pm$ 0.50	90.5 $\pm$ 18.55
Orange mesh 2 (n = 30)	3.6 $\pm$ 0.43	87 $\pm$ 17.83
White mesh (n = 30)	NA	NA
Rigid mesh (n = 5)	0.8 $\pm$ 0.15	268.3 $\pm$ 112.12
Basalt (n = 25)	1.4 $\pm$ 0.37	56.1 $\pm$ 7.34
Plastic mesh (n = 30)	1.8 $\pm$ 0.28	31.9 $\pm$ 4.63

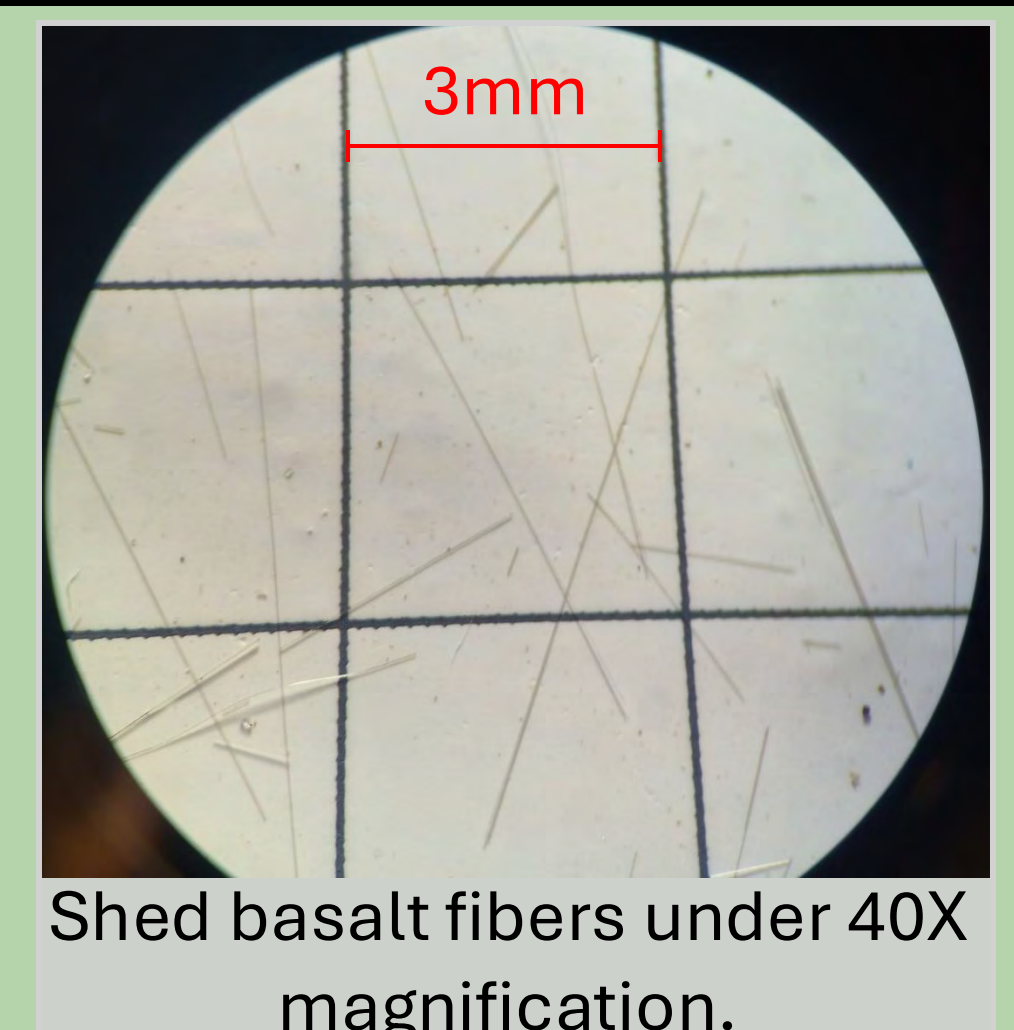
## Discussion

- Materials that shed microparticles should be carefully studied to prevent replacing plastic with another harmful substance.
- White mesh performed well in laboratory trials but failed in field deployment. **This result emphasizes the importance of examining both environmental impacts and material integrity in novel restoration materials.**

Material	Free of Microparticles?	Durable in field?
Orange mesh 1	Yes	Yes
Orange mesh 2	Yes	Yes
White mesh	Yes	No
Rigid mesh	No	No
Basalt	No	Yes

### Natrx<sup>®</sup> Basalt Bags:

- Fibers were shed in high volumes throughout the lab experiment.
- Basalt fibers are not understood and may persist in waterways.
- The consequences of basalt microfiber ingestion by aquatic organisms are unknown.



**Data are preliminary and will continue to be collected for 12 months before final analysis. Additional chemical analyses are also underway.**

## Acknowledgements

We would like to thank all restoration partners and volunteers. This project is funded by the NOAA/SeaGrant Marine Debris Challenge grant and UCF.

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# Plastic-free restored habitats: Reducing plastic pollution in community-based restoration of oyster reefs

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<sup>3</sup> University of Texas Marine Science Institute



## Importance

- Unsustainable harvest practices and environmental changes have reduced oyster reef distribution globally
- Habitat restoration plays a key role in rebuilding degraded reefs, often by deploying substrate into coastal waters to facilitate larval recruitment of oysters



Figure 1. Placing bagged shell at a community-based restoration event (Photo Credit: TAMU-CC MarComm)

## Problem

- Small scale restoration efforts often rely on materials that contribute to plastic pollution
- Plastic mesh offers affordability, versatility, and durability

## Project Goals

1. Determine the efficacy of using plastic-free alternatives to restore intertidal oyster reefs
2. Quantify the unintended consequences of using plastic materials in restoration
3. Conduct community outreach, education, and dissemination of project findings

## Approach

### Materials

- Natrx basalt mesh
- BESE biopolymer mesh
- Cement-infused jute rings
- Polyethylene plastic mesh

### Deployment

- Fill bags with 12 L of recycled oyster shell
- 6 reefs deployed in both TX and FL
- 10 reps of each material per reef
- Quarterly sampling for 2 years
- Decomposition trials in triplicate adjacent to each reef



Figure 2. Litterbag with float to assess decomposition (Photo Credit: Lisa Chambers)

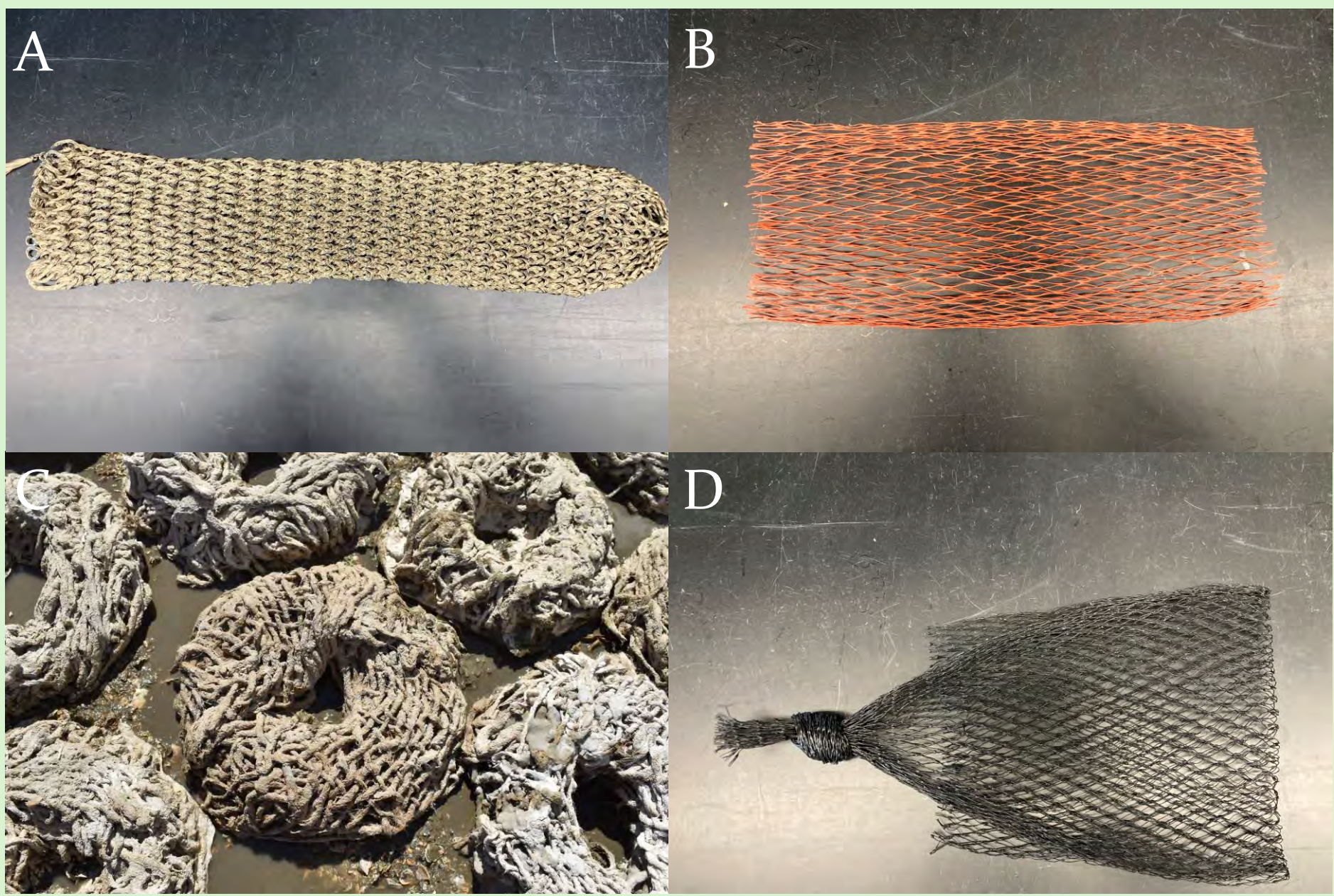


Figure 3. Restoration materials (A) Natrx basalt mesh, (B) BESE biopolymer mesh, (C) cement-infused jute ring, and (D) polyethylene plastic mesh (Photo Credit: TAMU-CC staff, Lisa Chambers)



Figure 4. Creating cement-infused jute rings (Photo Credit: Linda Walters)

## Novel materials tested in both Texas and Florida to assess usability and longevity

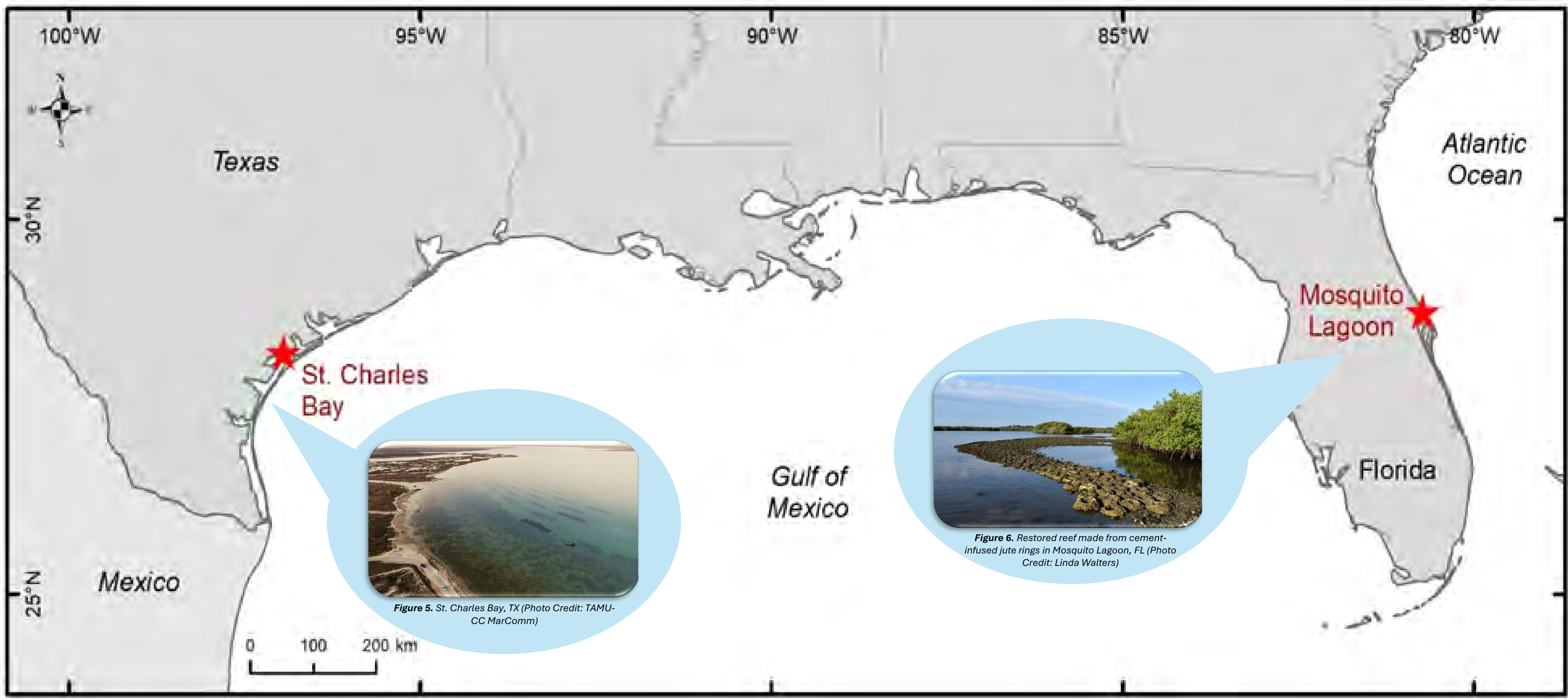


Figure 7. Project Locations

## Monitoring

### Restoration Efficacy

- Oyster abundance and size
- Faunal community
  - Encrusting fauna
  - Motile fauna
- Bag rips and tears
- Benefit-cost ratios

### Material Degradation

- Decomposition trials
- Chemical analysis
  - Carbon
  - Nitrogen
  - Phosphorous
  - Heavy metals
- Micro- and nanoplastics

### Community Engagement

- Engage with community groups
  - Unidos en STEM Teen Cafes
  - Voices of the Colonias
  - The Arc of Indian River County
- Community restoration events
- Material volunteer-friendliness



Figure 8. Photos from previous community engagement by the project team: (A) Voices of the Colonias, (B) Unidos en STEM Teen Cafes, (C) The Arc of Indian River County, and (D) School outreach event (Photo Credit: TAMU-CC staff, Linda Walters)

## Anticipated Benefits and Impacts

- Determine best practices
- Reduce plastic debris
- Community engagement
- Informed management
- Enhance ecosystem resilience



Figure 9. Volunteers at shell bagging event (Photo Credit: Kiese and Co)

## Acknowledgements

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# THE NURDLEOME

## Identification and characterization of microbes found on gulf coast nurdles

Author: Vibha Annaswamy | Research Educator: Dr. Kasia Dinkeloo  
University of Texas at Austin | Freshman Research Initiative | Bioprospecting 2.0

FIND THE MICROBES

FIND THE DEGRADERS

FIND THE SOLUTIONS

### Why Nurdles?

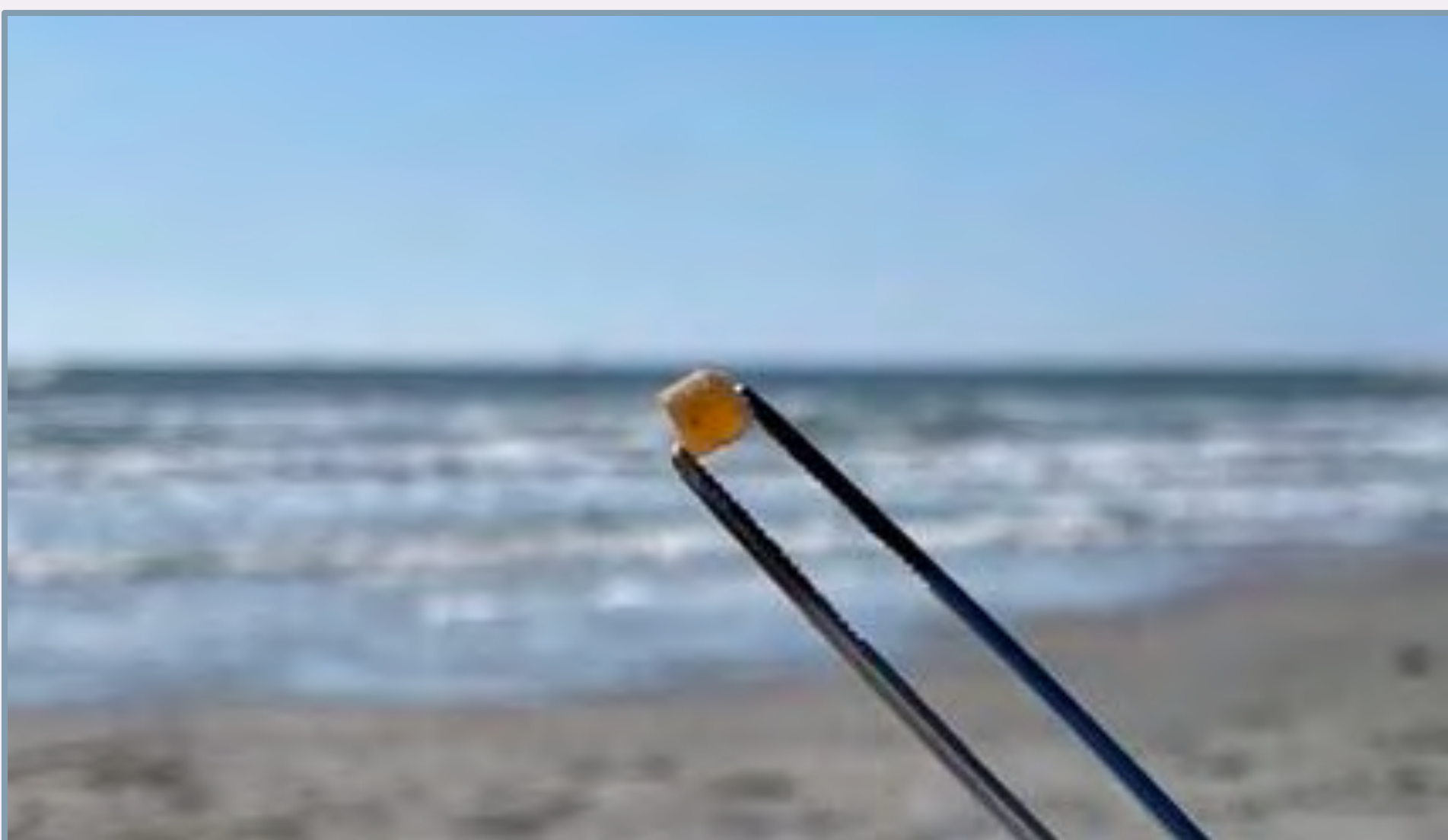
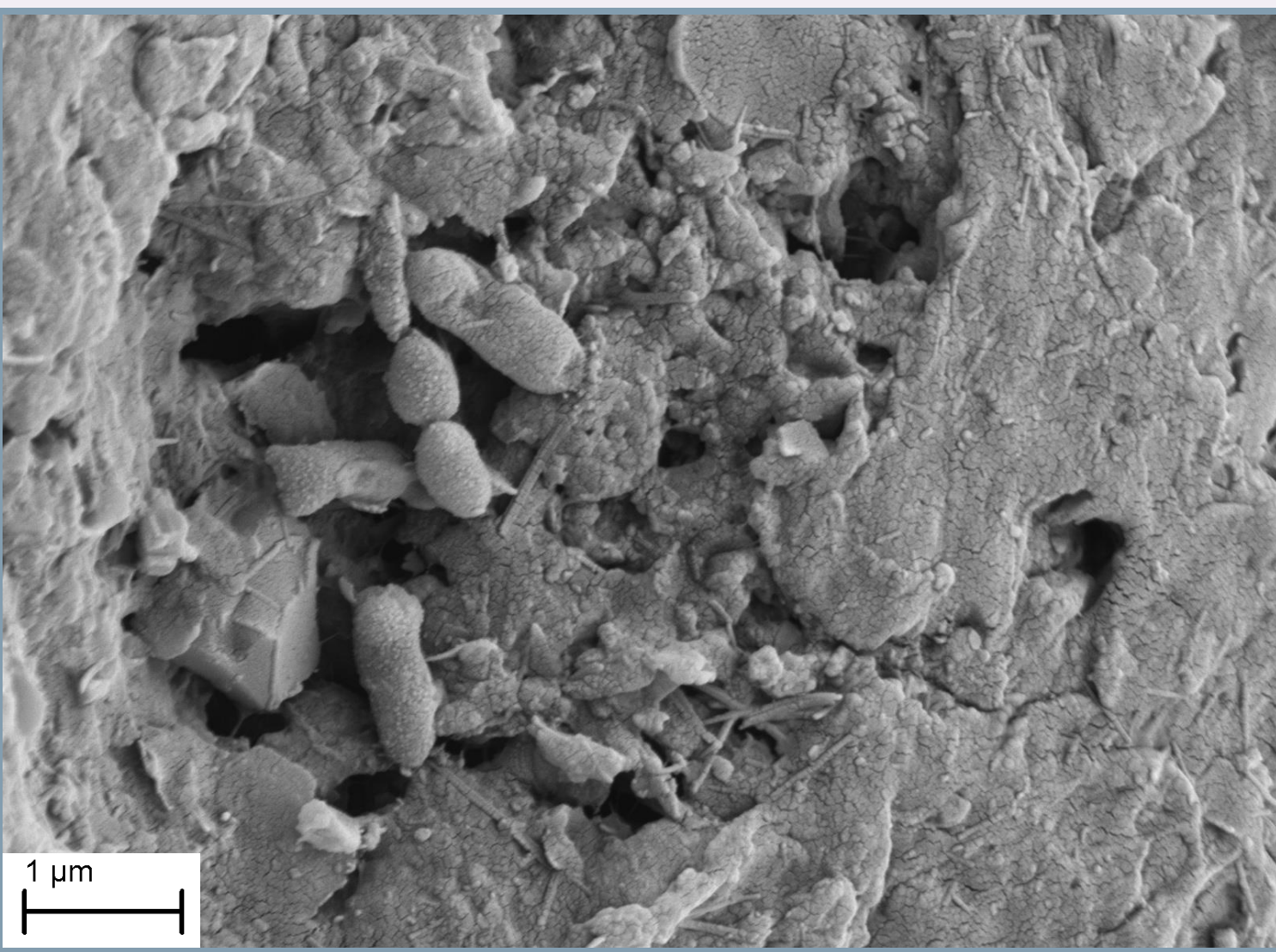
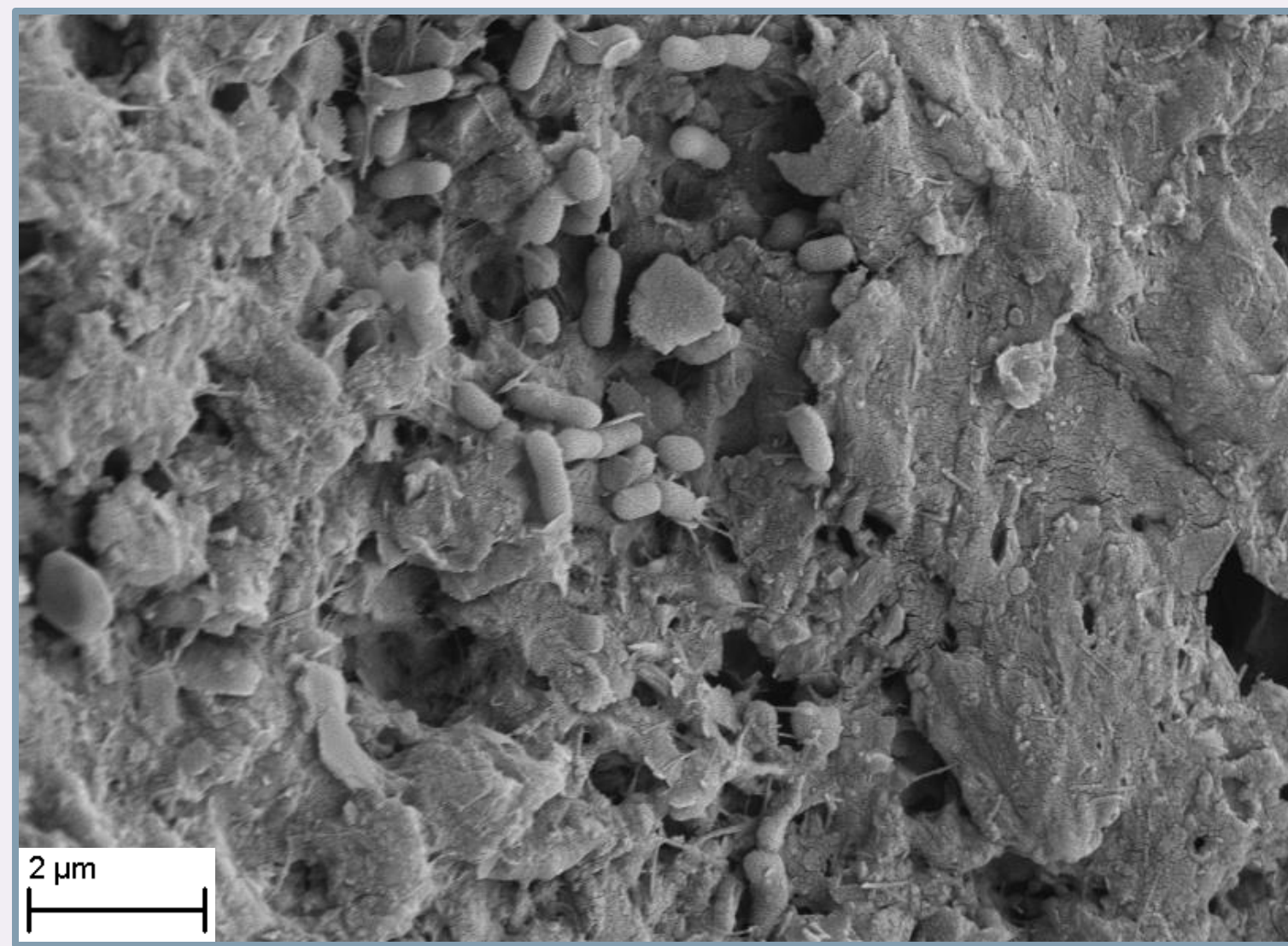


Figure 1: Photograph of a nurdle taken at Bob Hall Pier.



Figure 2: Stereomicroscope image of bulk nurdles collected from Port Aransas.



Figures 3-4: SEM images of coastal nurdles using enhanced prep to preserve bacteria.

### Targeted Metagenomic Sequencing

#### Methods

Nurdles collected from Bob Hall Pier in the spring of 2024 were processed using targeted metagenomic sequencing. For this process, we extracted DNA directly from bulk collections of nurdles to limit interference with their original microbiome. We also extracted DNA from a sand sample in order to compare the bacterial diversity associated with nurdles to that of the surrounding environment. Through library prep and sequencing, we used metagenomics to study the entire composition of microbial species present in samples of bulk nurdles while minimizing potential culture bias.

#### Results

Figures 6-7 display the most abundant genera identified in DNA samples extracted from bulk collections of coastal nurdles and from the sand. It is clear that the microbes residing on nurdles are distinct from those in the surrounding environment, revealing that the nurdle microbiome is unique to these plastics.

Genus	Number of Reads in Nurdles	Number of Reads in Sand
<i>Sulfitobacter</i>	5704	688
<i>Planococcus</i>	5104	9
<i>Psychrobacter</i>	4771	9
<i>Salinimonas</i>	4733	6
<i>Limnobacter</i>	110	16108

Figure 6: Average number of sequencing reads across nurdle samples as compared to the sand. The most abundant genera have been included in this table.

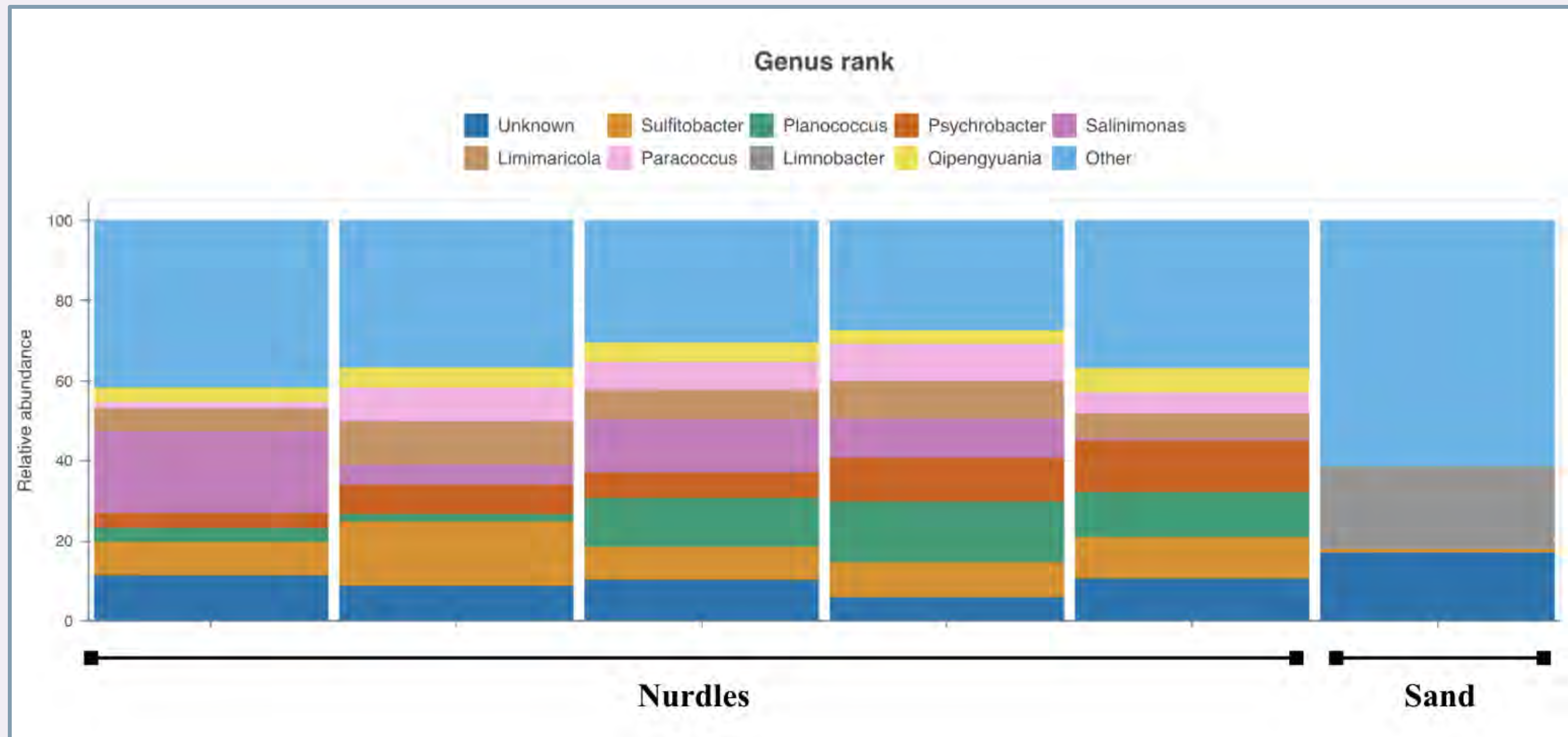


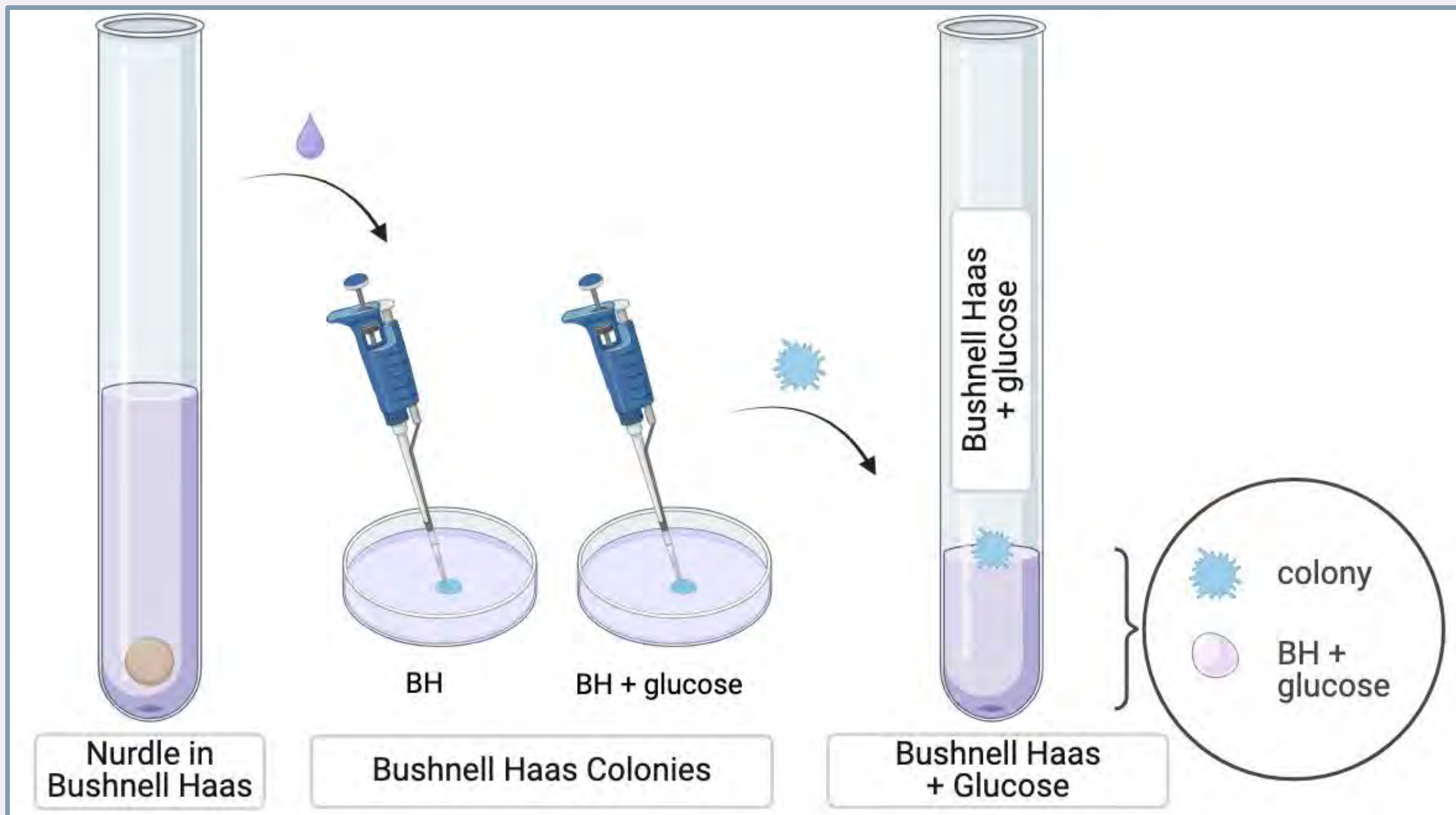
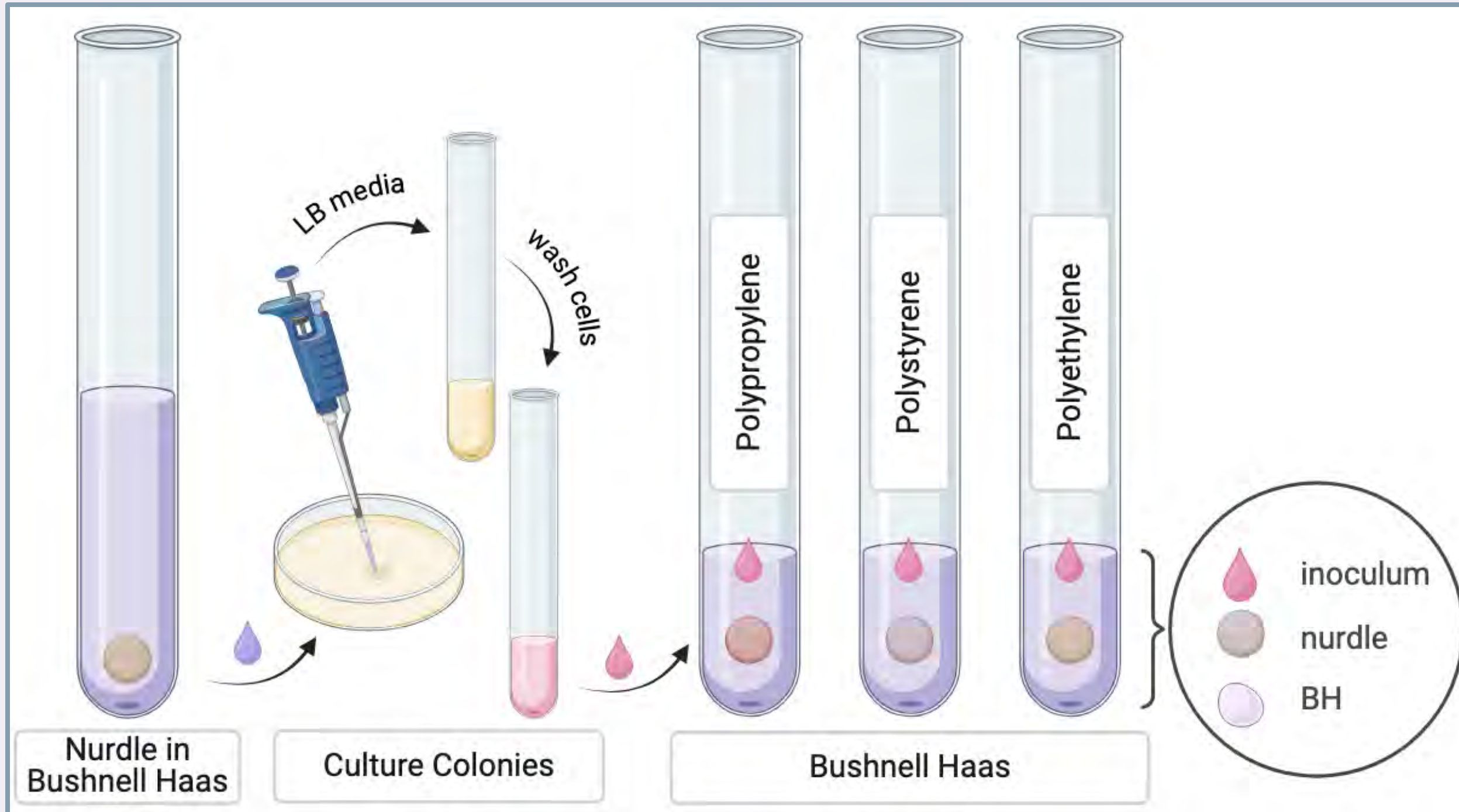
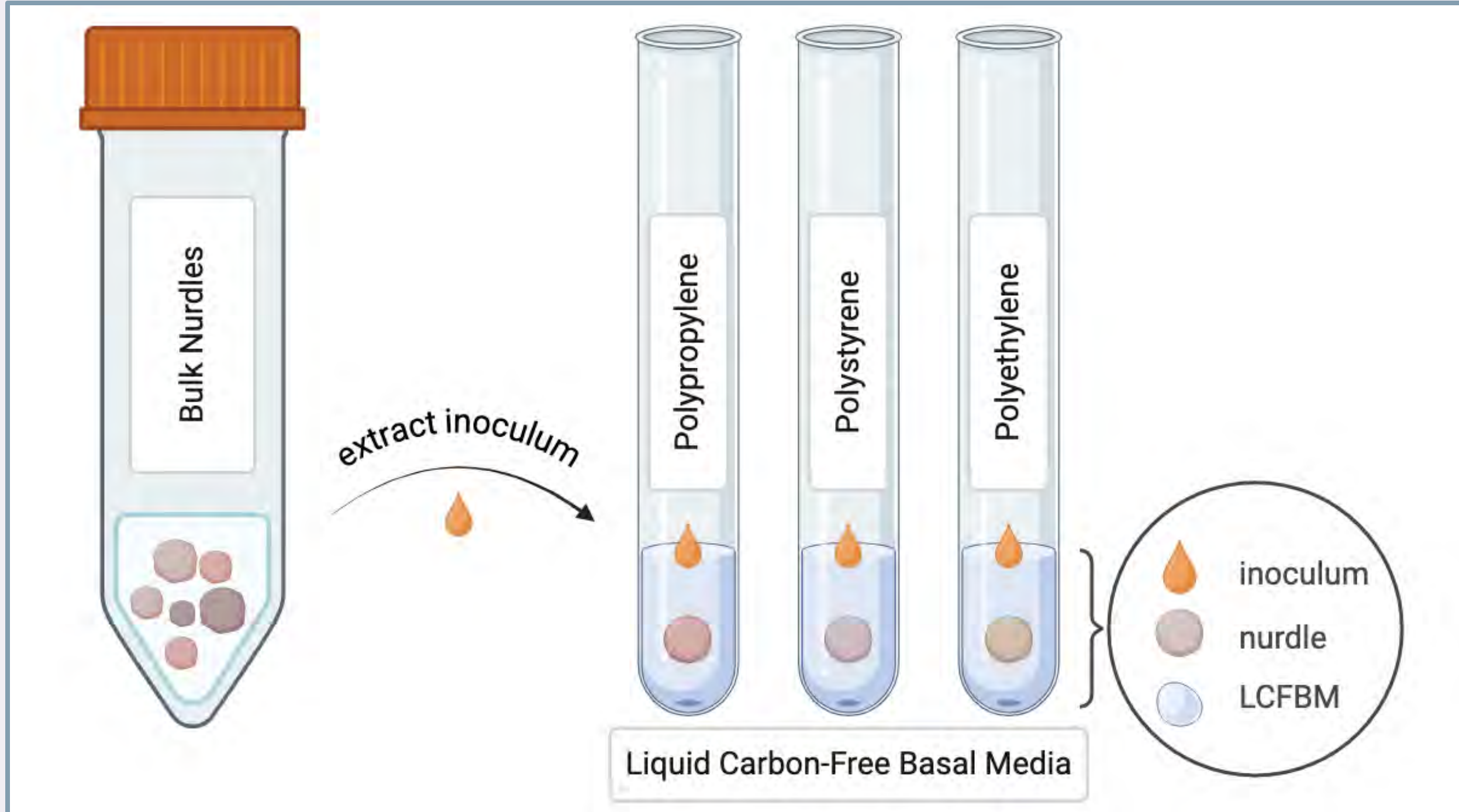
Figure 7: Relative abundances of genera identified in DNA extracted from coastal nurdles and the surrounding sand. Sequencing data was analyzed using Minimap2.

Sample	DNA Concentration (ng/μL)
Nurdle Sample 1	13.6
Nurdle Sample 2	12.2
Nurdle Sample 3	11.8
Nurdle Sample 4	4.90
Nurdle Sample 5	9.42
Sand Sample	9.74

Figure 5: DNA concentrations after barcoding PCR and purification. Samples were quantified using a Qubit.

### Selective Culturing

#### Methods



Figures 8-10: Methods for selective culturing.

Figure 11 and figure 12 show genera identified from the OC (coastal nurdle cultured in Bushnell Haas media) through isolation either in LB media or on Bushnell Haas plates. From LB, the samples identified as *Bacillus* and *Halopseudomonas* were used as inoculum for plastic cultures. All colonies isolated and identified from Bushnell Haas plates were also used as inoculum for Bushnell Haas liquid cultures. The genera that have been identified thus far are promising candidates for plastic degradation. *Bacillus* and *Pseudomonas* have been commonly identified and are microbes that have previously been shown to degrade certain types of plastic.<sup>3</sup> Additionally, *Halomonas* has been found to degrade a species of red seaweed, breaking down its cell wall, proteins, lipids, and carbohydrates.<sup>4</sup> This microbe produces enzymes including lipase, protease, cellulase, and amylase, thereby carrying the hypothetical machinery required for plastic degradation.

### Acknowledgements

Thank you to the Freshman Research Initiative and our stream's research educator, Dr. Kasia Dinkeloo. Thank you to UT Austin's Office of Undergraduate Research awarding fellowship funds to this project. Additional thanks to electron microscopy specialist, Michelle Mikes, for assisting with the use of SEM. And of course, I would like to acknowledge the nurdles that so graciously host microbes for us to study.

### Citations

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We are using selective culturing methods to target species of interest for plastic degradation. Figures 8-10 illustrate three different experimental set-ups that are currently being utilized.

In figure 8, inoculum for the cultures was extracted directly from bulk samples of nurdles. Nurdles from the coast were agitated in 1X PBS buffer and centrifuged repeatedly to form microbial pellets. The pellets were resuspended in PBS and used as the inoculum for three types of plastic cultures. Liquid carbon-free basal media was used for each culture with either a sterilized polypropylene nurdle, polystyrene nurdle, or polyethylene nurdle added as the sole carbon source. This will allow us to investigate whether certain microbial species are better at degrading specific types of plastic polymers.

In figure 9, the original culture (OC) consisted of a coastal nurdle as the sole carbon source in another carbon-free liquid called Bushnell Haas media. Growth from the OC was isolated on LB media plates and then cultured in LB liquid media. These isolated cultures were washed and then used as inoculum for plastic cultures in Bushnell Haas. We also extracted DNA from the isolated cultures and conducted PCR and sequencing to identify the isolated microbes (figure 11).

In figure 10, the OC was used to isolate colonies on Bushnell Haas agar plates with and without 0.5% glucose. These colonies were added to Bushnell Haas liquid media with 0.5% glucose to further grow the isolated colonies. We also conducted colony PCRs to directly identify species that grew in the OC and were isolated on the Bushnell Haas plates. Identified genera are listed in figure 12.

### Results

Sample	Genus Identified	Query Cover
LB Isolated 1	<i>Bacillus</i> <i>Neobacillus</i>	99% 99%
LB Isolated 2	<i>Bacillus</i>	99%
LB Isolated 3	<i>Niallia</i>	100%
LB Isolated 4	<i>Halopseudomonas</i> <i>Pseudomonas</i>	100% 100%

Figure 11: Genera identified through 16S rRNA gene sequencing of DNA samples extracted from OC cultures that were isolated in LB media.

Colony	Genus Identified	Query Cover
BH Isolated 1 (with 0.5% gluc)	<i>Halopseudomonas</i>	100%
BH Isolated 2 (with 0.5% gluc)	<i>Burkholderia</i> <i>Pseudomonas</i>	100% 100%
BH Isolated 3 (with 0.5% gluc)	<i>Halomonas</i>	100%
BH Isolated 4 (no gluc)	<i>Burkholderia</i> <i>Pseudomonas</i> <i>Halopseudomonas</i>	100% 100% 100%

Figure 12: Genera identified through 16S rRNA gene sequencing of OC colonies that were isolated on Bushnell Haas plates.

### Future Questions

Do certain microbes degrade specific types of plastic polymers? What enzymatic pathways allow them to do so? Can these mechanisms be applied to large-scale plastic degradation?



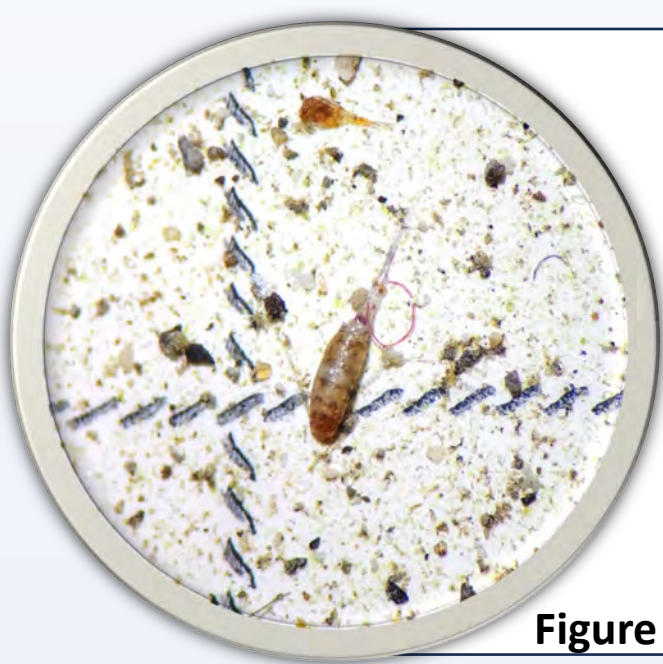


Figure 1: Copepod with microfibers

# Threads of Change: Zooplankton Community Shifts in Response to Fiber Disturbances

Authors: Caitlyn Lankford, Heaven Thompson, Ashton Fisher, Addison Lehew, Dr. Mary Kay Johnston



## Introduction



Fig 2: Shoal Creek sampling site between César Chávez (1<sup>st</sup>) and 2<sup>nd</sup> Street.

Microplastics, particularly synthetic fibers like polyester, are a growing concern in urban waterways. They can disrupt aquatic ecosystems by altering zooplankton feeding, reproduction, community dynamics, or destabilizing food webs and nutrient cycling (Rochman et al., 2015; Setälä et al., 2013). This study explores microplastic impacts through two approaches: controlled microcosm experiments

assessing zooplankton responses to synthetic and natural fibers, and a field survey of microplastic concentrations across two urban creeks in Austin, Texas. We hypothesized that polyester fibers would negatively affect zooplankton species richness, diversity, and community composition, with greater impacts in fishless systems due to altered predation dynamics. We expected higher microplastic concentrations downstream in urban creeks, reflecting urban runoff accumulation.

## Methods

### Microcosm Experiment

Zooplankton from fish and fishless ecosystems were exposed to polyester fibers, cotton fibers, or control conditions across 48 microcosms. Fibers (36–80  $\mu\text{m}$ ) were introduced at  $9 \times 10^6$  fibers/mL. Microcosms were maintained under a 12-hour light cycle for three weeks and regularly fed *chlorella vulgaris*. We assessed species richness, diversity, and community composition.

Table 1: Microcosm experimental design

	Control	Cotton	Polyester
Fish	4	4	4
Fishless	4	4	4

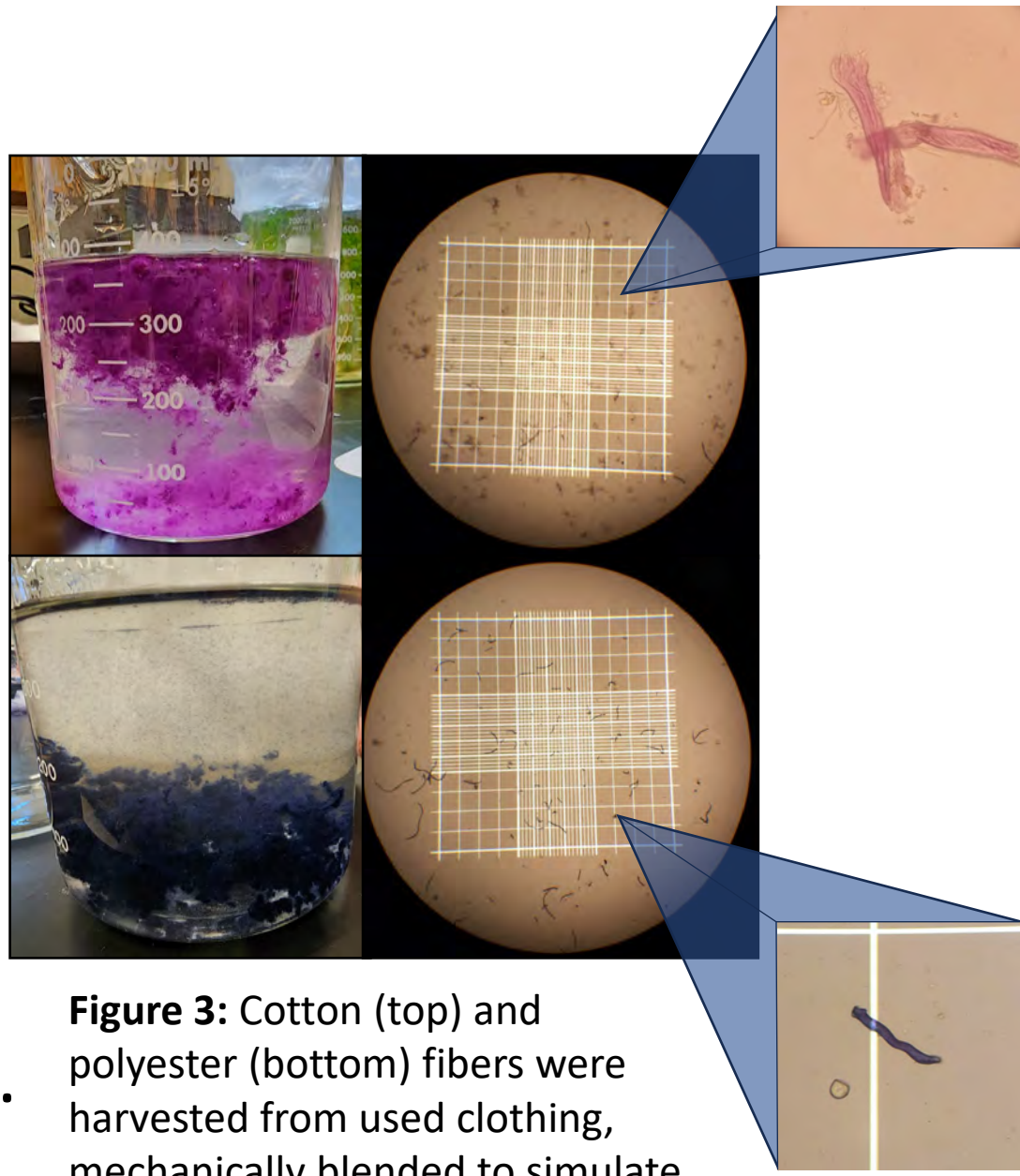


Figure 3: Cotton (top) and polyester (bottom) fibers were harvested from used clothing, mechanically blended to simulate breakdown, and quantified using a Neubauer hemocytometer. Callouts show close-up views of each fiber type.

### Microplastic Survey

Vertical samples of the water column were collected from four sites along Shoal Creek and Waller Creek, aligned with the City of Austin's Environmental Integrity Index (EII) locations. At each site, three 200 mL samples were vacuum-filtered through a 0.45  $\mu\text{m}$  membrane filter.

Microplastics were visually identified under a dissecting microscope and categorized by type (fiber or fragment) and color (red, white, blue, green, black).

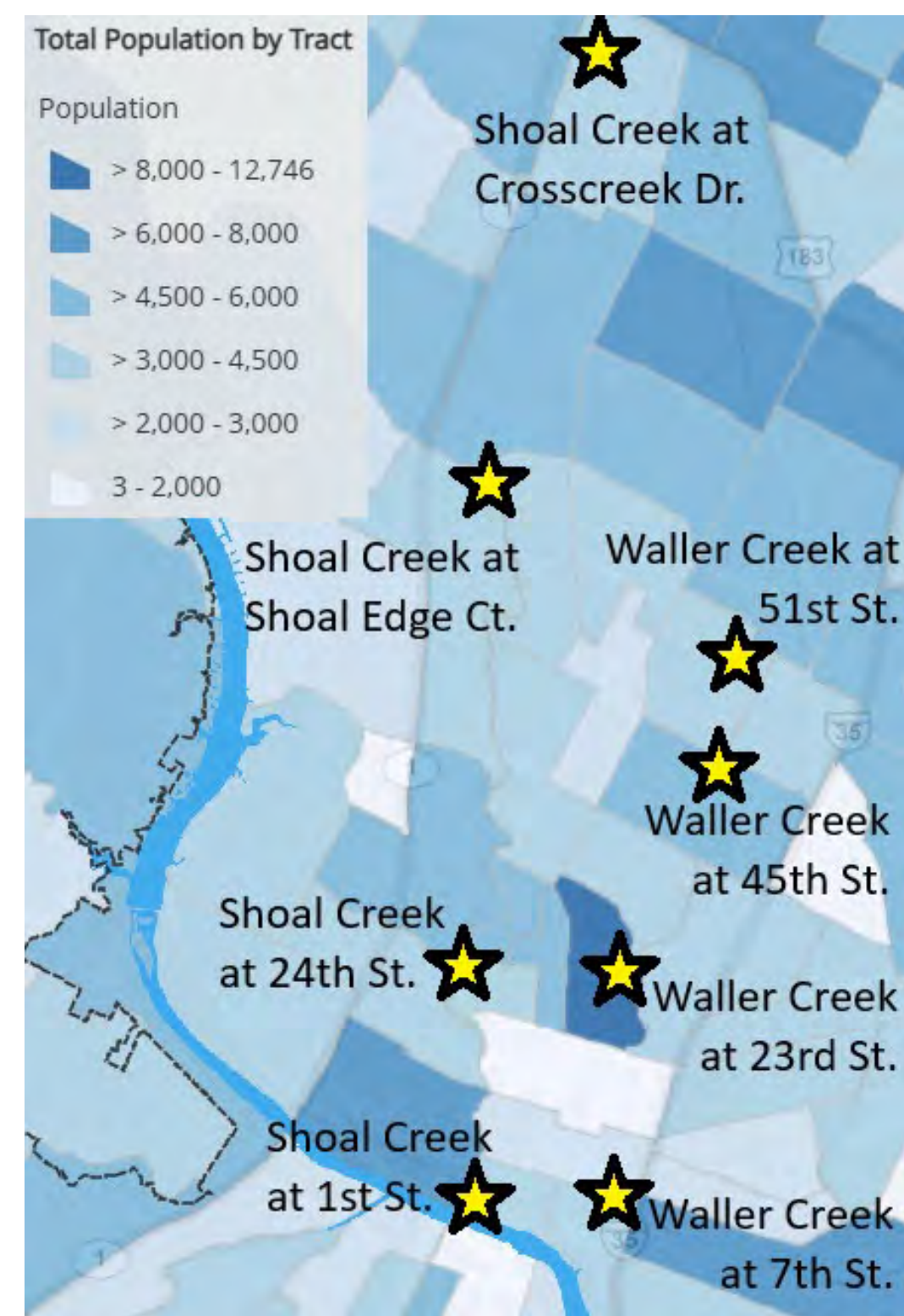


Figure 4: Population density by census tract in Austin, TX, overlaid with microplastic sampling sites along Shoal Creek and Waller Creek. Darker shades indicate higher population density. Data source: City of Austin (n.d.), Austin Demographics Hub..

### Microcosm Experiment

Zooplankton communities were primarily shaped by ecosystem type, with fishless systems supporting higher abundance and diversity. Fiber presence had subtle, taxon-specific effects. Copepods increased in cotton treatments and decreased with polyester. In contrast, rotifers remained unaffected by fiber type, thriving in fishless environments where predation was absent. These findings highlight that while overall community structure remained stable, specific taxa like copepods may act as indicators of microplastic impacts due to their adaptable feeding strategies and ecological importance.

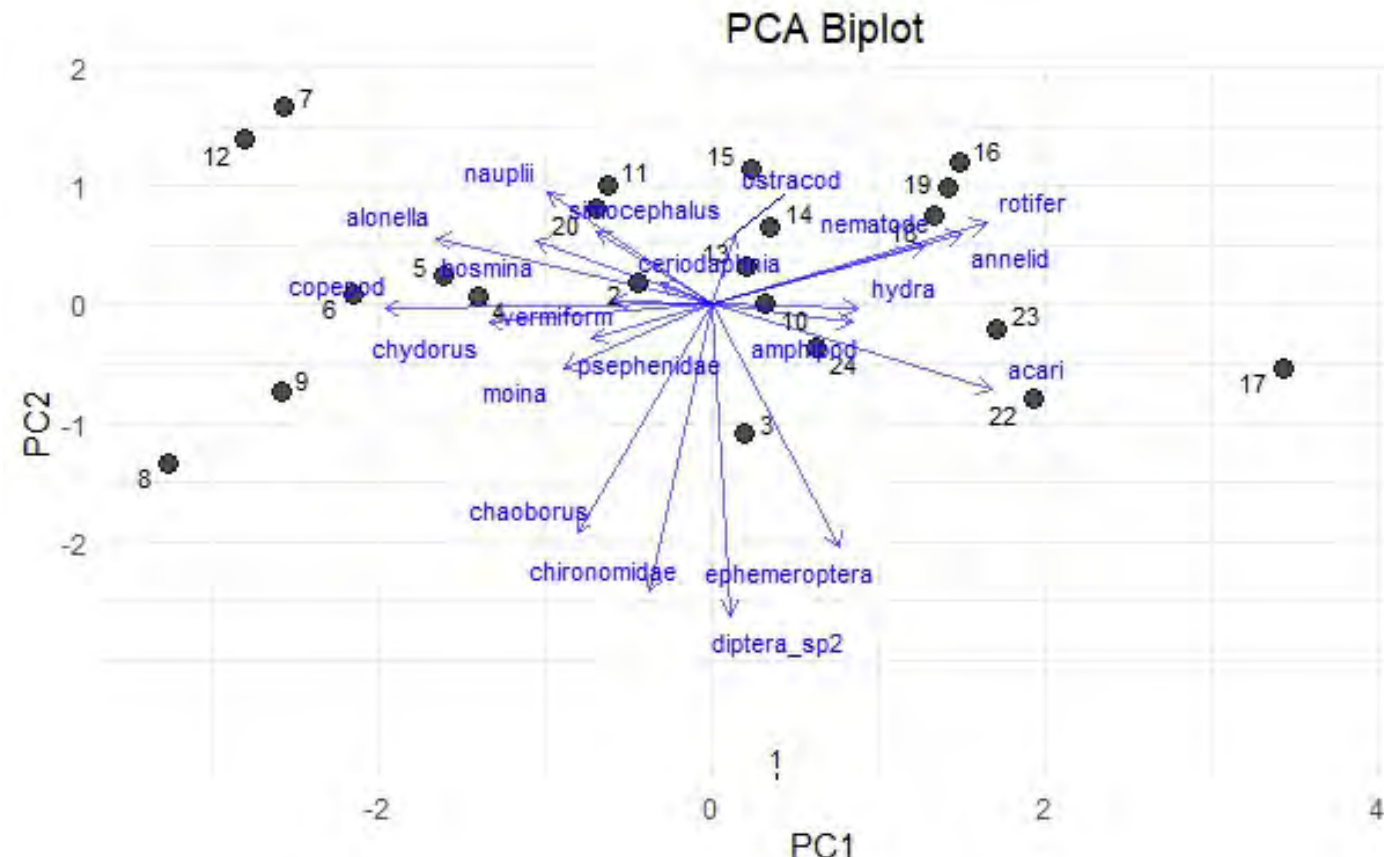


Figure 5: PCA biplot showing zooplankton community variation across fiber treatments and ecosystem types. PC1 explains 18.3% and PC2 12.9% of the variance. Copepods, chironomids, and rotifers were key drivers. Ecosystem type significantly influenced PC1 ( $F_{1,18} = 28.90$ ,  $p < 0.001$ ), while fiber effects were context-dependent ( $F_{2,18} = 3.35$ ,  $p = 0.058$ ).

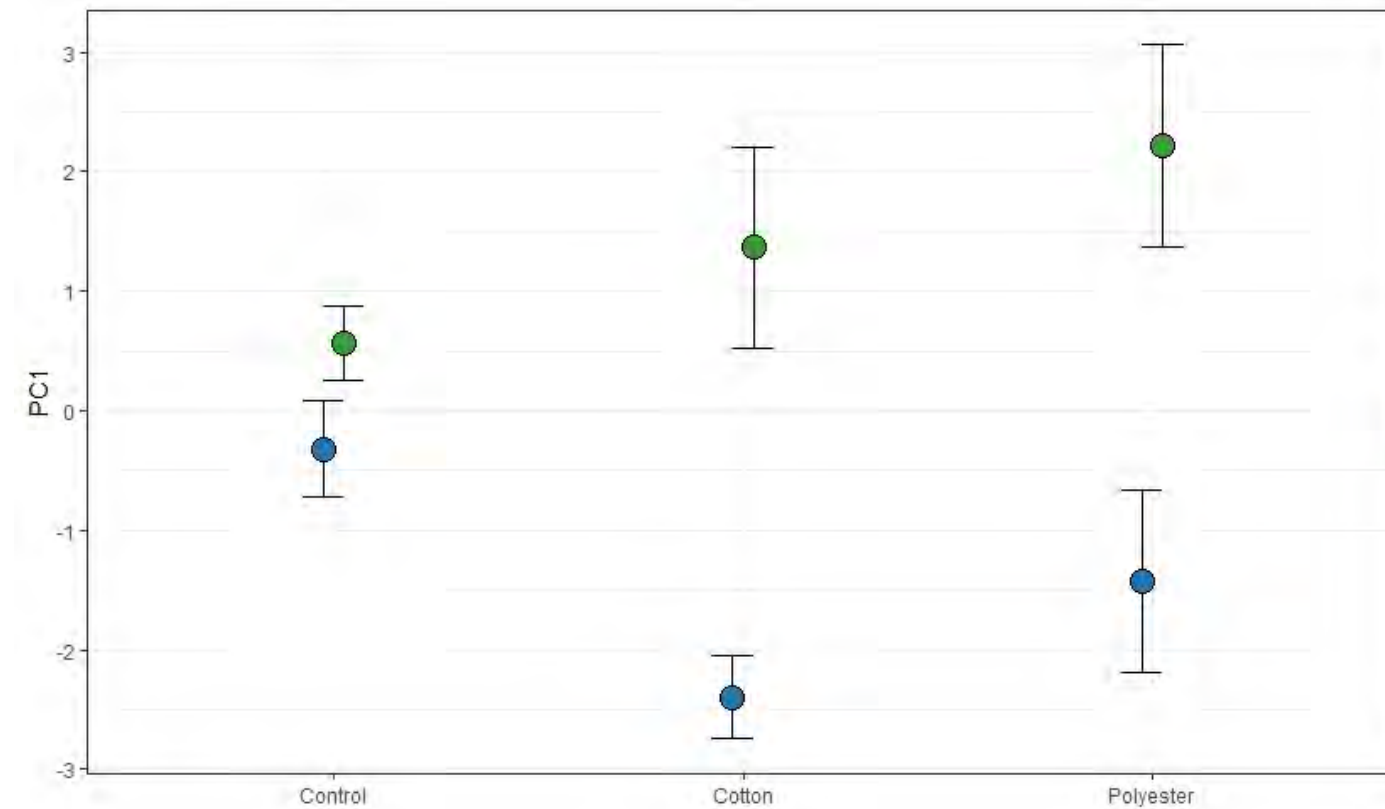
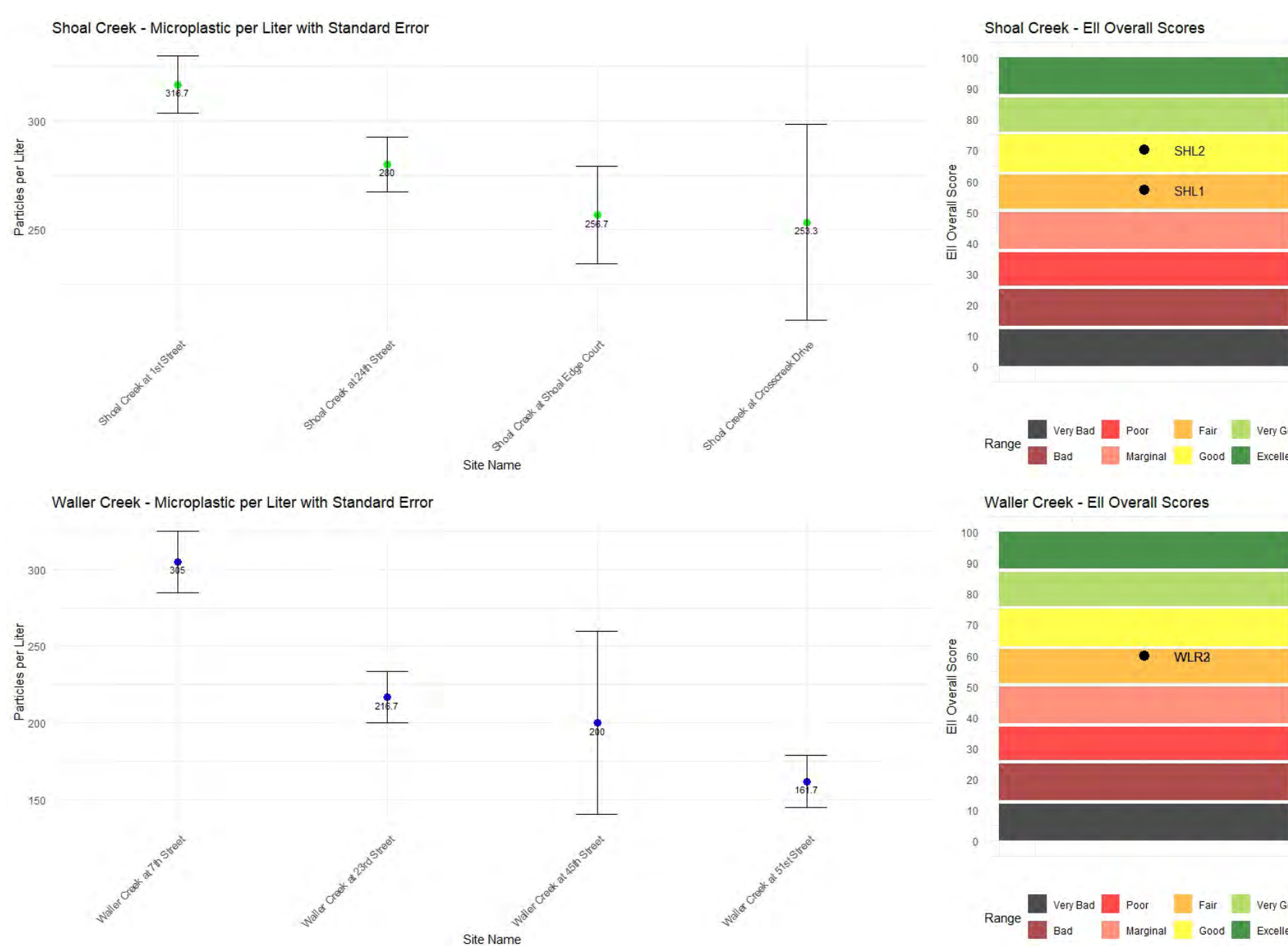


Figure 6: PC1 scores of aquatic communities under different fiber treatments (Control, Cotton, Polyester) for Fish (blue) and Fishless (green) ecosystems. Error bars show standard error. A significant interaction between fiber and community type ( $F_{2,18} = 5.82$ ,  $p = 0.011$ ) was observed. Polyester caused the largest divergence, with fishless systems consistently scoring higher on PC1.

### Microplastic Survey

Microplastics were found at all sites, with fibers consistently more abundant than fragments (Figure 12). Both creeks showed increasing concentrations of microplastics downstream. Contrary to expectations, Shoal Creek, despite active conservation efforts, recorded higher microplastic levels than Waller Creek. The highest average microplastic concentrations of both streams were observed before entering Lady Bird Lake.

Black and white fibers dominated in both creeks, indicating common urban sources like textiles or construction runoff. Microplastic distribution patterns generally aligned with the streams' EII scores, reinforcing the connection between urbanization and pollutant load.



Figures 10 & 11: Microplastic concentrations and Environmental Integrity Index (EII) scores for Shoal Creek and Waller Creek. (Left Panels): Microplastic concentrations (particles/L) with standard error across four sites. Shoal Creek peaked at 1st St. (316.5 particles/L), while Waller Creek peaked at 7th St. (305 particles/L). (Right Panels): EII scores indicate "Fair" to "Good" conditions in Shoal Creek (SHL1, SHL2) and "Fair" for Waller Creek (WLR3).

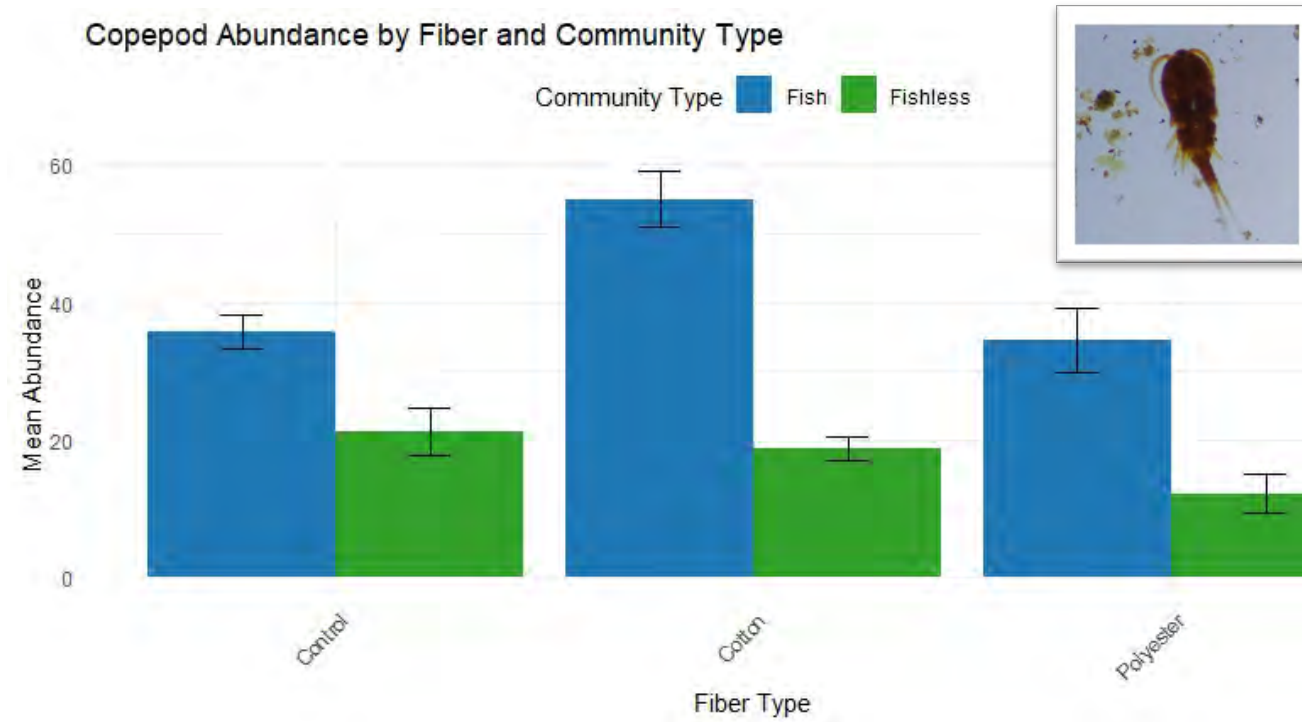


Figure 7: Mean copepod abundance across fiber treatments and community types. Error bars indicate standard error. Both fiber type ( $F_{2,18} = 9.35$ ,  $p = 0.0028$ ) and community type ( $F_{1,18} = 53.33$ ,  $p < 0.001$ ) significantly affected abundance. Cotton increased the abundance of copepods in fish systems, while polyester reduced counts overall. Inset: Copepod image.

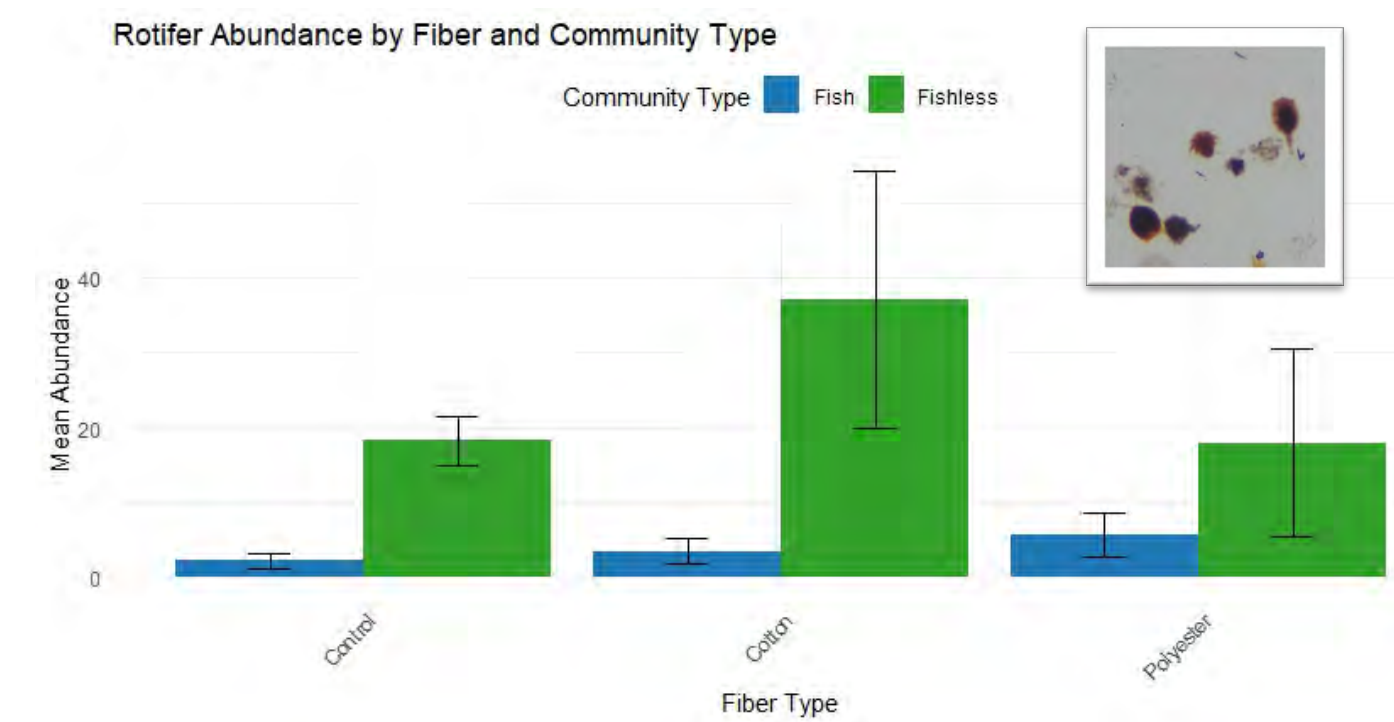


Figure 8: Mean rotifer abundance across fiber treatments and community types. Error bars show standard error. Rotifers thrived in fishless ecosystems, with the highest variability in cotton treatments. Fiber type had no significant effect; community type was the main driver. Inset: Rotifer image.

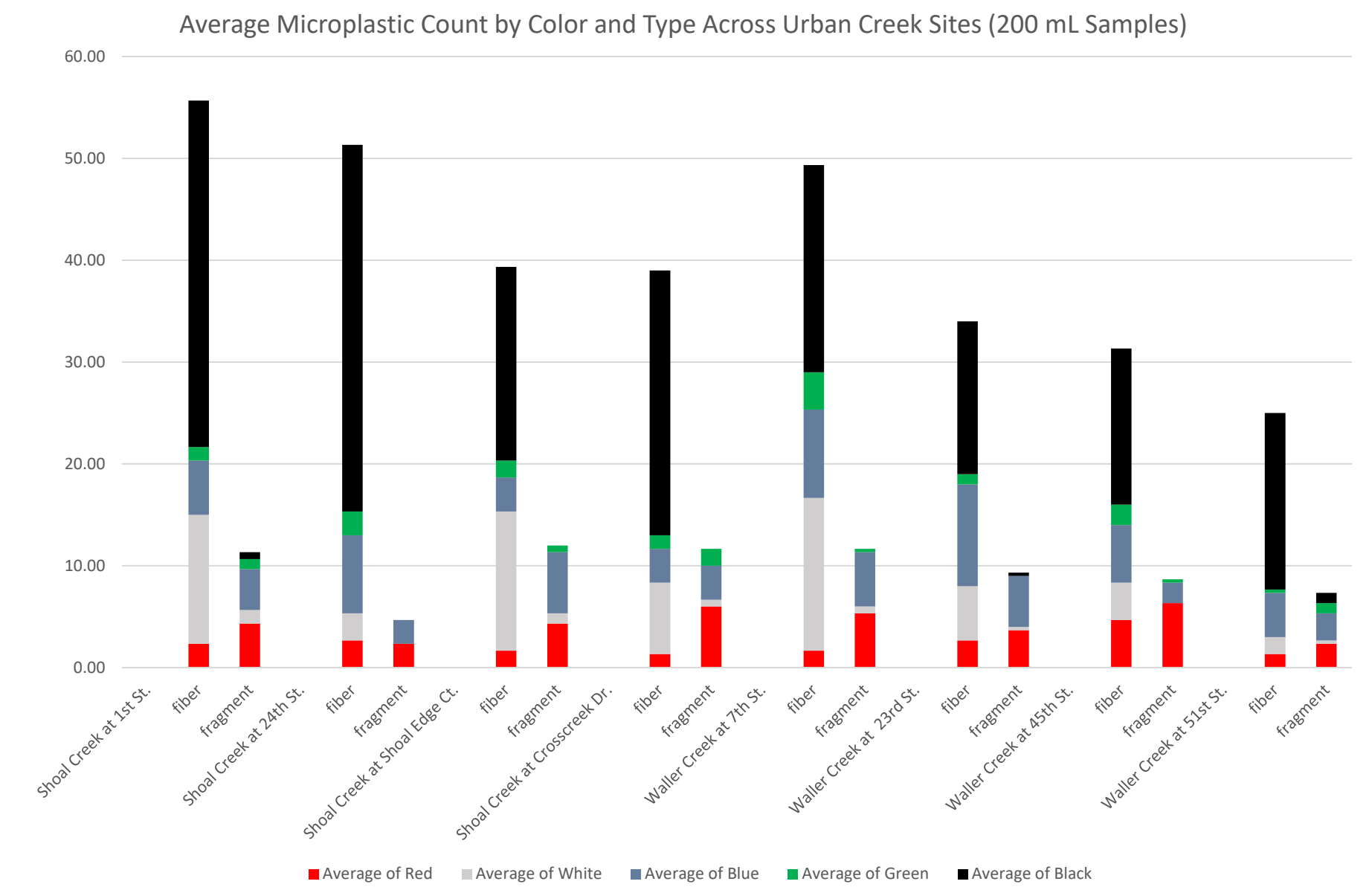


Figure 12: Stacked bar graph showing average microplastic counts per 200 mL water sample across Shoal Creek and Waller Creek sites, separated by fiber and fragment types. Colors represent microplastic types (Red, White, Blue, Green, Black), with black fibers consistently most abundant. Data were averaged from replicates filtered through 0.45  $\mu\text{m}$  filters.

## Discussion

Microplastic impacts are context-dependent, shaped by ecosystem dynamics and pollutant types. The microcosm experiment demonstrated that copepods responded to fiber type, suggesting that microplastic exposure may influence certain taxa. However, broader community patterns remained stable, indicating that short-term exposure at tested concentrations may not cause immediate shifts in species composition.

Field data showed the expected downstream accumulation of microplastics. Population density mapping suggests a link between urbanization and microplastic concentrations (Figure 4). While urban density may contribute to these trends, further investigation is needed to determine how factors like stormwater management, infrastructure, and localized pollution sources interact to influence microplastic distribution.

Future studies should integrate urban planning perspectives to assess how population density, waste management, and city infrastructure impact microplastic pollution. Additionally, long-term monitoring is needed to evaluate potential ecological consequences, particularly for taxa sensitive to fiber exposure.

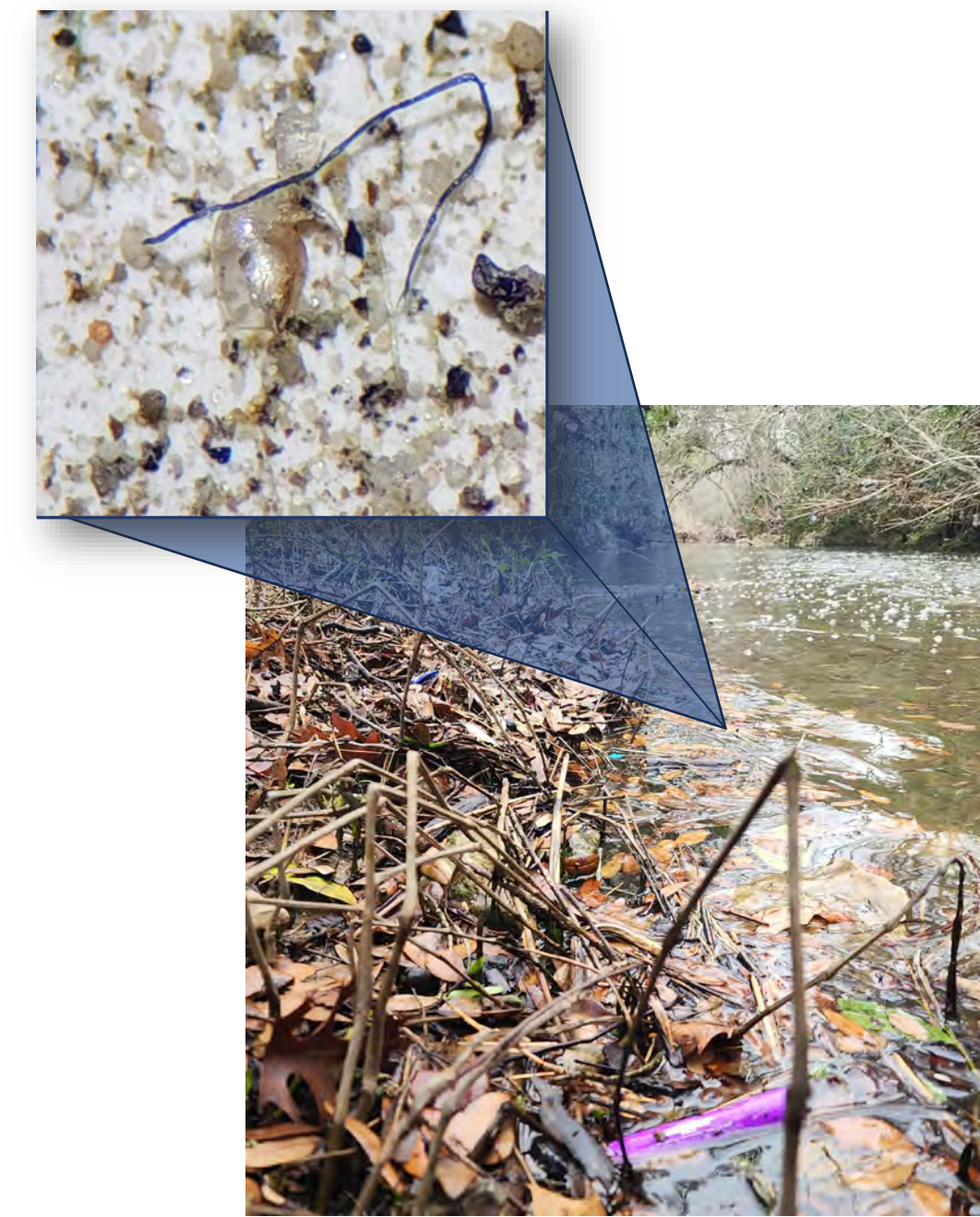


Figure 13: Image of Shoal Creek sampling site with a callout showing a magnified view of a *Bosmina* with a blue microfiber on the membrane filter.

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