Texas A&M University at Galveston
1001 Texas Clipper Road
Galveston, TX 77554

SEVENTH INTERIM PERFORMANCE REPORT

FEBRUARY 28TH, 2023

Project Title: The Fate and Toxicity of Microplastics and Persistent Pollutants in the Shellfish and Fish of Matagorda Bay

Submitted To:

Matagorda Bay Mitigation Trust

Performing Laboratory:

Texas A&M University on behalf of Texas A&M University at Galveston

Authors:

Ms. Emily Meese (Ph.D. student)

Mr. Asif Mortuza (Ph.D. student)

Mr. Marcus Wharton (Ph.D. student)

Dr. David Hala, Ph.D.

Dr. Karl Kaiser, Ph.D.

Dr. David Wells, Ph.D.

Dr. Lene H. Petersen, Ph.D.

Dr. Antonietta Quigg, Ph.D.

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The Fate and Toxicity of Microplastics and Persistent Pollutants in the Shellfish and Fish of Matagorda Bay

Personnel

Principal Investigators:

Drs. David Hala, Karl Kaiser, David Wells, Lene H. Petersen, Antonietta Quigg

Consulting MBMT Project Coordinator:

Mr. Steven J. Raabe

Location(s):

Texas A&M University at Galveston

Project Duration:

01 June 2021 – 31 August 2024

Objectives:

Objective 1: Quantify the extent of microplastics pollution in the surface waters and biota of Matagorda Bay.

Objective 2: Measure levels of persistent pollutants in surface waters, adsorbed to microplastics, and bioaccumulated in the biota of Matagorda Bay.

Objective 3: Study the toxicity of microplastics and adsorbed pollutants using embryo-larval life stages of sheepshead minnow.

Objective 4: Public educational outreach to local high school students on the science of ecosystem health monitoring.

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1. INTRODUCTION

1.1 Background

This project is studying the extent of microplastics, and persistent pollutant exposure of resident biota (shellfish and fish) sampled from Matagorda Bay and assessing any likely toxicity effects due to exposure. The *new knowledge* gained from the successful completion of this project will contribute to an understanding of the long-term fate and toxicity of microplastics (and adsorbed pollutants) in the Matagorda Bay system.

In this seventh interim report (December 1st, 2022 – February 28th, 2023) we provide a list of key accomplishments as per the third quarter of Year 2 of the project.

2. Key Updates

As of the period encompassing the seventh interim report (December 1st, 2022 – February 28th, 2023), the key achievements associated with each stated objective are detailed below.

Objective 1: Quantify the extent of microplastics pollution in the surface waters and biota of Matagorda Bay.

- The fifth interim report details the levels of various microplastics measured in the surface waters of Matagorda Bay. Plans are in-place for additional dockside sampling at various locations around Matagorda Bay in Spring 2023. Samples from previous collections with a pump system (> 5 µm) and a tow net (> 200 µm) across the bay and along shore have been processed and analysis will be completed by the end of December. Procedural blanks were also performed to determine any contamination issues during collection, processing, and analysis of samples.

- Preliminary results of surface water samples show a significant amount of microplastics to be polyethylene (PE). Polyvinyl chloride (PVC) and nylon-66 (N-66) follow PE in relative abundance (%) (**Table 1** and **Fig. 1**). Polypropylene (PP) was significantly higher at the Port O'Connor Jetty and Mid- Bay near the oyster reef. Additional research and

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analysis will guide the project towards understanding the transport of these plastics throughout the estuary.

Table 1. Microplastic concentrations in surface water samples collected in Matagorda Bay (N-66 = nylon-66, PC=polycarbonate, PE=polyethylene, PET = polyethylene terephthalate, PMMA=Poly(methyl methacrylate), PP=polypropylene, PS = polystyrene, PUR = polyurethane, PVC=polyvinyl chloride, SBR = Styrene Butadiene Rubber).

ng/L	Port O'Connor	Port Lavaca Pier	Palacios Pier	Oyster Reef
N-66	61.88	22.13	22.13	36.72
PC	12.82	16.25	16.25	9.65
PE	138.00	122.52	122.52	223.37
PET	4.36	8.41	8.41	3.30
PMMA	2.05	1.84	1.84	9.48
PP	53.33	0.00	0.00	60.41
PS	1.96	0.00	0.00	3.30
PUR	0.00	0.00	0.00	0.00
PVC	89.12	19.25	19.25	17.94
SBR	20.03	0.00	0.00	8.46
Total	383.55	190.41	190.41	372.62

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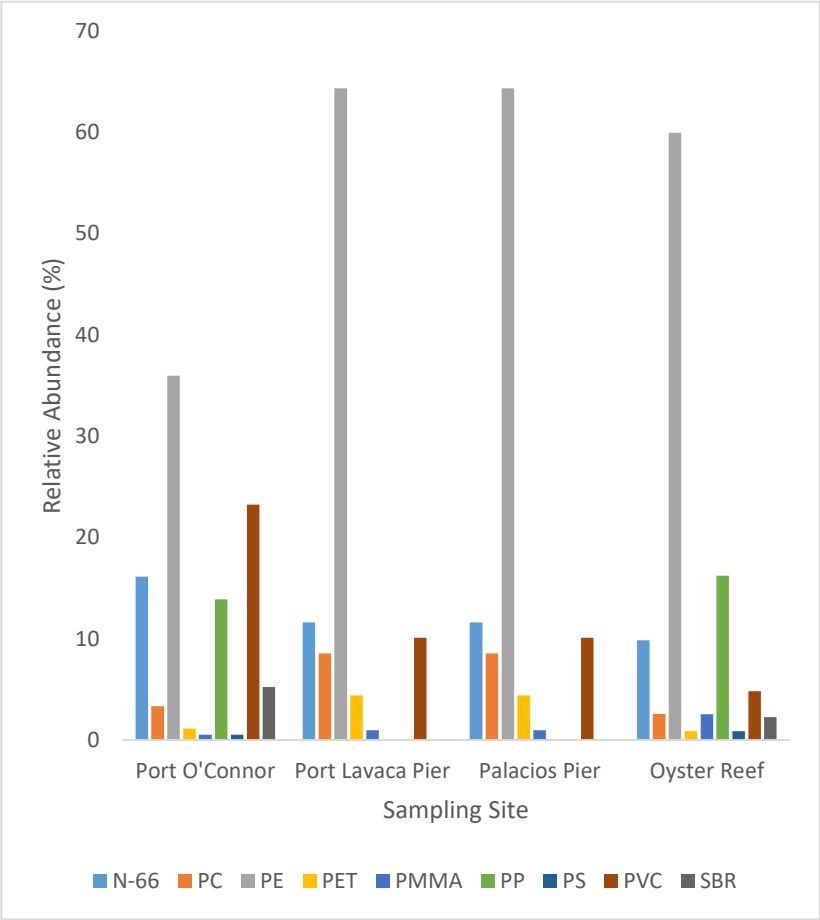

Fig. 1. Relative abundance of microplastic detected in surface water samples around the Matagorda Bay estuary. Please see the legend for **Table 1** for a full list of abbreviations.

Objective 2: Measure levels of persistent pollutants in surface waters, adsorbed to microplastics, and bioaccumulated in the biota of Matagorda Bay.

- Quantification of the levels of PAHs and PCBs in biota from Matagorda Bay has been completed and previously summarized in the previous *sixth* interim report.

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- In addition to the pollutant body-burden analysis, we also performed a risk assessment for human sea food safety and cancer risk (**Fig. 2**). The Food and Drug Administrations (FDAs) cancer risk of consumption of fish and oysters from the two bays was determined to compare consumer safety.

- FDA in association with NOAA and EPA set forth a Level of concern (LOC) for cancer risk after the Deepwater Horizon spill incident to evaluate whether the finfish and oysters in the affected area are safe for consumption (FDA, 2010). It is determined using the formula:

  LOC= (RL x BW x AT x CF)/ (CSF x CR x ED)

- Where, RL= Risk level, BW= Average consumer body weight, AT= Average time, CF= Conversion factor, CSF= Cancer slope or Potency factor of Benzo[a]pyrene (BaP), CR= Consumption rate, ED= Assumed exposure duration, LOC= Level of concern. For finfish and oysters, RL= 0.00001, BW= 80 kg, AT= 78 years, CF= 1000 µg/mg, CSF= 7.3 mg/kg/day, ED= 5 years. The consumption rate for finfish was determined to be 49 g/day and for oyster it is 12 g/day by the regulators. This makes the LOC for finfish to be 0.035 µg/g/BaPE (Benzo[a]pyrene equivalent) and for oysters, 0.142 µg/g/BaPE (FDA, 2010).

- To assess the cancer risk of consuming biota from Matagorda and Galveston Bays, the BaP concentration in biota was monitored (Collins et al., 1991). Similar, high molecular weight PAH compounds are standardized to the BaP equivalent concentrations using their toxicity equivalent factor (TEF). The BaP equivalent concentration or BaPE is then determined by adding the concentration of BaP with the other BaP equivalent PAHs (Benzo[a]anthracene, Chrysene, Benzo[b]fluoranthene, Benzo[k]fluoranthene, Indeno[123-cd]pyrene, Dibenzo[a,h]anthracene), after multiplying them with their toxicity equivalent factor or TEF. The formula is as follows:

  BaPE concentration = BaP concentration + (sum of other PAH concentrations x TEF)

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- In this calculation, the concentrations are reported in µg/g of wet weight (ww) BaPE. To test for significant differences in BaPE between each of the species from each bay, a Kruskal-Wallis test was performed followed by Dunn's post hoc to determine significant differences in cancer risk between the species (**Fig. 2**).

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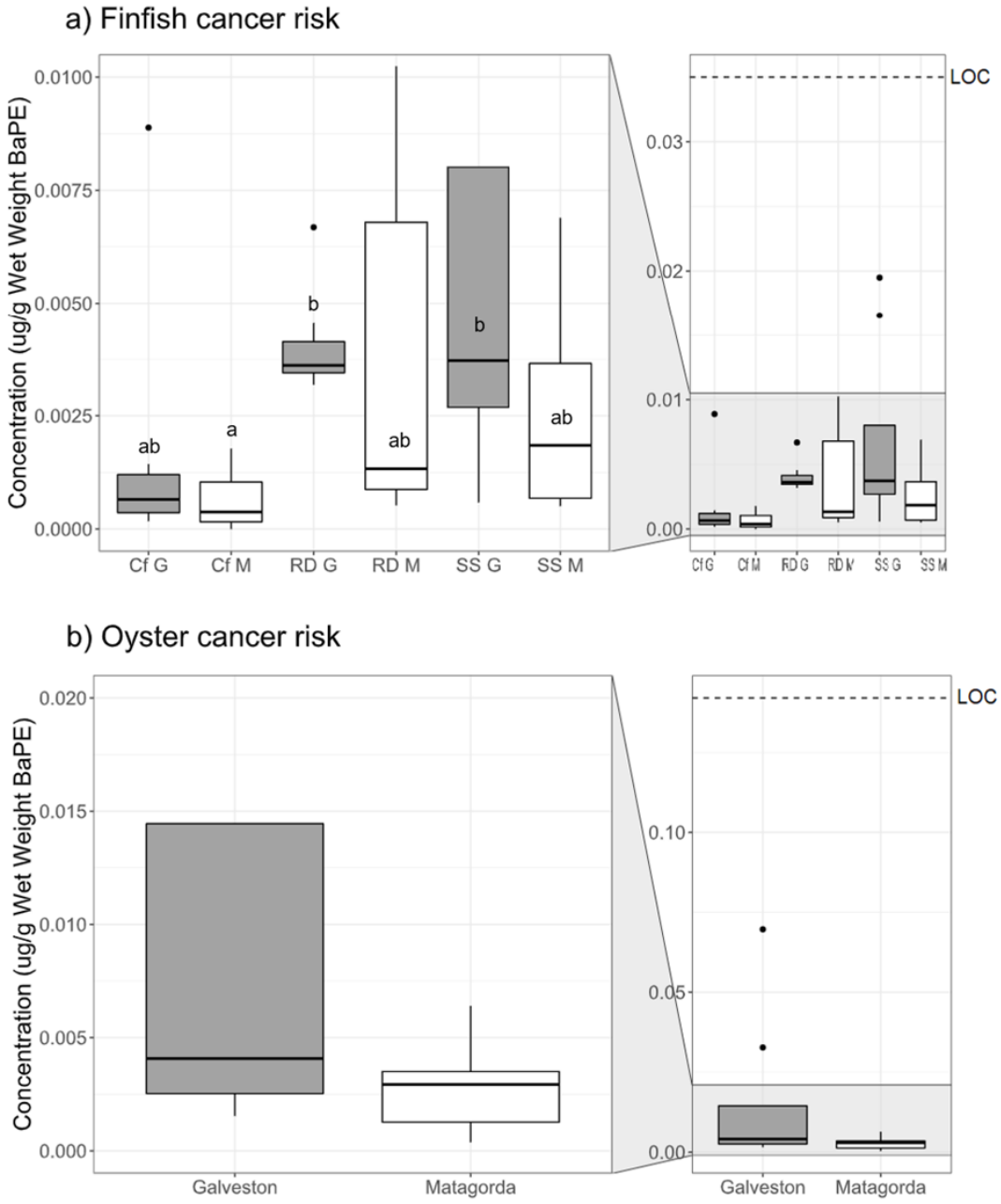

Fig. 2. Benzo[a]pyrene equivalent toxicity (BaPE) concentrations of finfish a) and oysters b) collected from Galveston and Matagorda Bay in comparison to FDA Level of Concern (LOC) for cancer risk set for finfish and oysters respectively. The concentrations are reported in µg/g of wet weight BaPE. G= Galveston Bay (gray), M= Matagorda Bay (white), Cf= Catfish, RD= Red drum, SS= Spotted Sea trout. Different letters above the plot denote significant differences between the plots (p<0.05).

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- We found that, finfish BaPE was ~5- 56x less than the level of concern for cancer risk.

- Oyster BaPE was ~10- 53x less than the level of concern for cancer risk of oyster consumption.

- Matagorda Hardhead Catfish cancer risk of consumption ($p<0.05$) was:
    - ~11x less than Galveston Spotted Seatrout.
    - ~6x less than Galveston Red Drum.

- All biota BaPE concentrations were lower than FDA level of concern for cancer risk.

Objective 3: Study the toxicity of microplastics and adsorbed pollutants using embryo-larval life stages of sheepshead minnow.

- This objective will be engaged with starting in Fall 2023 and onwards.

- An Animal Use Protocol (AUP) to perform *in vivo* experimentation with early life-stages of embryo-larval sheepshead minnows (*Cyprinodon variegatus*) has already been approved by the A&M Institutional Animal Care and Use Committee (IACUC).

Objective 4: Public educational outreach to local high school students on the science of ecosystem health monitoring.

- Educational outreach engagement will be pursued in collaboration with the TAMUG Sea Camp program in Summer 2023. A previous such outreach activity was reported in the *sixth* interim report.

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3. FURTHER WORK

Planned work for completion over the duration of the seventh interim report (Year 2) are as follows:

1) Prepare a manuscript for publication by Summer 2023 on the PAH and PCB data generated as part of the research performed in this project (comparing pollutant levels in Matagorda vs. Galveston Bays).
2) Commence microplastics analysis in water and biota samples. Prepare a manuscript describing microplastics analysis methods (Summer 2023).
3) Plan the initiation of toxicological studies on the effects of microplastics and PAH/PCB mixtures on embryo-larval life stages of fish (Fall 2023).

4. REFERENCES

Collins, J., Brown, J., Dawson, S., & Marty, M. (1991). Risk assessment for benzo [a] pyrene. Regulatory toxicology and pharmacology, 13(2), 170-184.

FDA. (2010). Protocol for Interpretation and Use of Sensory Testing and Analytical Chemistry Results for Re-Opening Oil-Impacted Areas Closed to Seafood Harvesting Due to The Deepwater Horizon Oil Spill. FDA. https://www.fda.gov/food/food-safety-during-emergencies/protocol-interpretation-and-use-sensory-testing-and-analytical-chemistry-results-re-opening-oil

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Reviewed by:

2/28/2023

_____ Date: _____

Dr. David Hala, TAMUG, P.I.

Approved by:

_____ Date: 3/1/2023

Mr. Steven J. Raabe, Trustee