

Program on Water Quality Community Science for Middle School



Plastic vs. Natural Substrate: Marine Biofouling Experiments

Background:

Although marine biofouling is a term many are unfamiliar with, it is an important and impactful process affecting oceanic systems. It is characterized by marine organisms that accumulate on man-made surfaces like ships, offshore rigs, aquaculture cages, plastic pollution, and other submerged structures.¹ This accumulation occurs naturally and is often referred to as recruitment, meaning that a benthic habitat has been colonized by new organisms.² In addition to organisms growing on, attaching to, and seeking refuge in man-made structures, they also do so on natural surfaces like whales, oysters, crabs, rocks, and shells of various organisms.

Plastics and other marine debris have become a large component of substrates affecting marine life, mainly as a result of the astounding increase in plastic production (up 620% from 1975 to 2012)³ and the minimal amount of that plastic that is recycled (9% of all U.S. plastic in 2012).³ Plastic as well as other human modifications made to natural substrate have caused great impacts on population sizes.⁴ There is some evidence that introducing a new substrate to an estuary may increase the abundance and diversity of subtidal species.⁴ There is also research suggesting that this does not have a negative impact on the diversity of species on nearby natural substrate.⁴ However, seeking refuge on plastic substrates may be detrimental to the health of these organisms as these plastics leach out toxins and harmful chemicals.⁵

Species diversity is important as it is an indicator of how healthy and productive the ecosystem is, and it is essential that we discover the role increasing plastic pollution plays in this. The objective of these experiments is to observe differences in species abundance and diversity associated with colonizing natural or plastic substrates.

Objectives:

Students will learn to:

- Identify types of natural and artificial substrates marine organisms may grow on, attach to, or seek refuge in
- Observe and understand how different substrates promote growth or refuge
- Understand the difference between abundance and diversity
- Calculate abundance and diversity of organisms for each substrate
- Collaborate as a class to assess and compare results for various substrates
- Construct bar graphs illustrating these diversity scores to visually depict the differences among substrate types
- Offer explanations for why different substrates may promote growth or be preferred refuge sites

Duke Marine Lab:

Materials:

- Plastic lids
- Plastic bottles
- Glass tiles
- Granite tiles
- Limestone tiles
- Vexar bags
- Small cable ties to hold vexar bags together
- Thin PVC (0.5") or rebar to keep bags in place
- Rope

Setup:

1. Place the 4 substrates (plastic, glass, granite, limestone) in separate vexar bags. There will be 3 of each substrate (12 bags total).
 - a. Each bag is tied with small cable ties.
 - b. Thin PVC is placed at the bottom of the bags to keep them in place.
2. Vexar bags are dropped from the dock with a rope for 4 weeks to allow for organisms to colonize the substrates.
 - a. Each rope has a brick attached to the bottom to hold the vexar bags down.
 - b. Each bag is zip tied to the bottom of the rope, close to the brick.
 - c. There is 2 feet of separation between each bag.
 - d. Each repetition is conducted with treatments in the same order (i.e. plastic, glass, granite, limestone, plastic, glass...).

Classroom:

Materials:

- Buckets
- Sorting trays
- Watch glasses for sorting
- Dissecting microscopes
- Plastic rulers to look at size distribution
- Data collection worksheet

Setup:

1. Assign groups: Each group will be assigned a different repetition that includes one bag of each substrate.

Methods:

1. Use sorting trays to remove substrate from vexar bags.
 - a. Take great care not to agitate organisms growing on substrate.
2. Observe and record the number of each species on group data sheet.
 - a. This should be completed for each substrate.
3. Calculate the Simpson's Diversity Index scores for your group.
4. Each group report their species totals to entire class, and each person record all findings on class data sheet.
 - a. Each individual calculates total Simpson's Diversity Index score for each substrate.
5. Create bar graphs to compare the diversity of growth on the different substrates (can compare individual group vs. class totals).
 - a. Compare: total number of species found on each substrate.
 - b. Compare: total number of organisms found on each substrate.
 - c. Graph: number of organisms of each species for each substrate.

References:

1. Yang, W.J., Neoh, K.G., Kang, E.T., Teo, S.L.M., Rittschof, D. (2014) Polymer Brush Coatings for Combating Marine Biofouling, *Progress in Polymer Science*, Vol. 39, No. 5, pp. 1017-1042. Available at: <https://www.sciencedirect.com/science/article/pii/S0079670014000227>
2. Scrosati, R.A. and Ellrich, J.A. (2017) Unimodal Relationship Between Small-scale Barnacle Recruitment and the Density of Pre-Existing Barnacle Adults, *PeerJ*, Vol. 5, pp. 1-12. Available at: <https://peerj.com/articles/3444/>
3. Jambeck, J.R., Geyer, R., Wilcox, C., Siegler, T.R., Perryman, M.; et al. (2015) Plastic Waste Inputs From Land Into the Ocean, *Science*, Vol. 347, No. 6223, pp. 768-771. Available at: <http://science.sciencemag.org/content/347/6223/768>
4. Connell, S.D. and Glasby, T.M. (1999) Do Urban Structures Influence Local Abundance and Diversity of Subtidal Epibiota? A Case Study from Sydney Harbour, Australia, *Marine Environmental Research*, Vol. 47, No. 4, pp. 373-387. Available at: <https://www.sciencedirect.com/science/article/pii/S0141113698001263>
5. Safina, C., and J. Perelman. 2016. "Pesky plastic: the true harm of microplastics in the ocean." *National Geographic*. Available at: <https://blog.nationalgeographic.org/2016/04/04/pesky-plastic-the-true-harm-of-microplastics-in-the-oceans/>
6. Hulathduwa, Y.D. Stickle, W.B., Aronhime, B., Brown, K.M. (2011) Differences in Refuge Use are Related to Predation Risk in Estuarine Crabs, *Journal of Shellfish Research*, Vol. 30, No. 3, pp. 949-956. Available at: <http://www.bioone.org/doi/abs/10.2983/035.030.0337?journalCode=shre>

Program on Water Quality
Community Science for Middle School



WORKSHEET 1: Marine Biofouling Experiments Predictions & Methods

Question: Will a greater abundance and diversity of organisms grow on plastic or natural substrate?

Treatments:

- Plastic substrate (plastic lids, plastic bottles)
- Natural substrate (glass, granite, limestone)

Hypotheses:

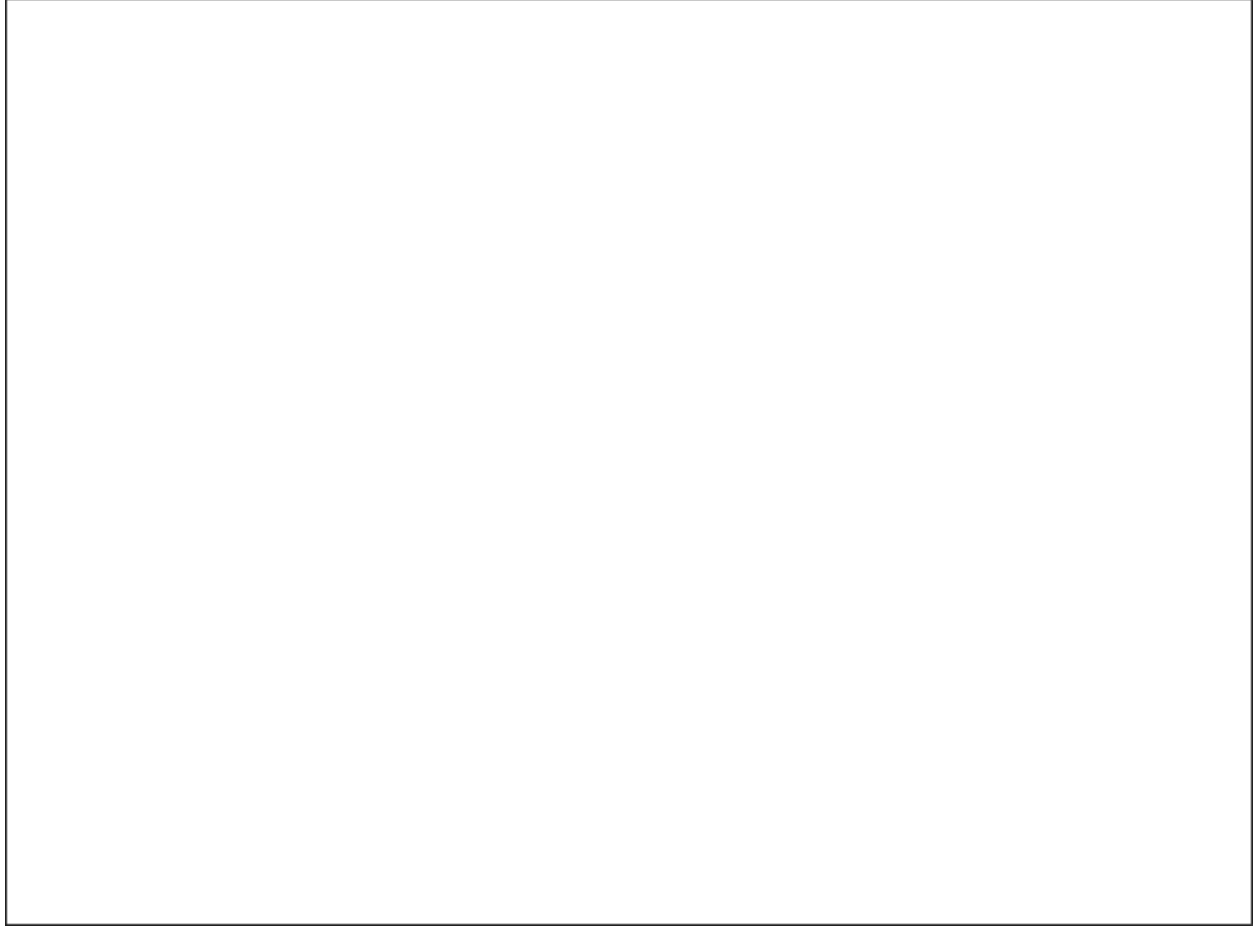
- Which material do you think will support a greater abundance of organisms? Why?
- Which material do you think will support a greater diversity of organisms? Why?

Predictions:

- If there is a greater abundance of organisms on _____ substrate, then _____
- If there is a greater variety of organisms on _____ substrate, then _____

Graphically draw your predictions for your experiment:

- What will be on the y-axis? Units? [# of species, # of organisms, size of organisms?]
- What will be on the x-axis? Units?



Do you think there will be a difference among treatments? Why or why not?

Scientist: _____

WORKSHEET 2: Marine Biofouling Experiments Data collection

Substrate Type	Species	Number (n)	n(n-1)

Equations:

Simpson's Diversity Index:

$$D = 1 - (\sum n(n-1) / N(N-1))$$

n = total number of organisms of a particular species

N = total number of organisms of all species