Morphological Characteristics of Oysters from Predator Experiments at the Dauphin Island Sea Lab, AL, May-July 2019

Website: <u>https://www.bco-dmo.org/dataset/892096</u> Data Type: experimental Version: 1

Version Date: 2023-03-29

Project

» <u>Collaborative Research: Keystone chemicals: Identifying general and universal molecules of fear</u> (Identifying molecules of fear)

| Contributors | Affiliation | Role |
|---------------------------------------|---|------------------------------------|
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Abstract

These data include oyster morphological characteristics after oysters were settled onto shells and either exposed to predator cues (caged blue crabs) or controls of no cues. Oysters were nursed in a flow-through seawater system at the Dauphin Island Sea Lab for two months starting in late May 2019. Afterward, subsets of oysters were assessed for shell strength, size, and dry weight using a Kistler force sensor and drying oven. Predators often produce nonconsumptive effects (NCEs) in their prey in the form of behavioral or morphological changes. Such changes often have larger or equal consequences for population dynamics as the predator directly consumes individual prey. However, it is not well understood how predators feeding across multiple trophic levels cause cascading NCEs that interact across prey trophic levels or how the prey survival benefits from these interactions change across contexts. These data help demonstrate how NCEs can influence population dynamics across space and quantify the strength of these context-dependent interactions. Data were collected by Drs. Benjamin Belgrad, Lee Smee, and Marc Weissburg from the Dauphin Island Sea Lab and Georgia Institute of Technology.

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Coverage

Spatial Extent: Lat:30.248451 Lon:-88.077982 Temporal Extent: 2019-05 - 2019-07

Oyster culturing

Oysters (*Crassostrea virginica*) were cultured as spat-on-shell at the Auburn University Shellfish Laboratory (AUSL) on Dauphin Island, AL starting in late May 2019 using standard techniques (Congrove et al. 2009). Oyster larvae were settled onto sun-bleached oyster shells to create spat-on-shell. After 3 days, when oyster spat were approximately 1.0 millimeters, they were exposed to either exudate from predatory blue crabs or empty cage controls in four flow-through holding tanks (length = 2.4 meters, width = 0.9 meters, water depth = 0.4 meters) supplied with unfiltered seawater pumped directly from the Gulf of Mexico. The number of spat per shell varied from approximately 5 – 40 and we elected to not alter the initial density to mimic natural settlement during the induction period. Oysters were suspended above the tank bottom in oyster aquaculture baskets (64 x 23 x 14 centimeters with 140 spat-covered per shells basket) to prevent sediment buildup from suffocating oysters. Seven oyster baskets were present in each tank (28 total).

Spat were exposed to blue crab predator cues by holding four live caged adult blue crabs (*Callinectes sapidus*) in two of the tanks (8 crabs total), whereas the remaining two tanks contained empty cages (control) to mimic conditions where oysters regularly experience predator cues or are limited in their exposure from cues. Water volumes and crab densities were informed from established procedures (Belgrad et al. 2021). Crabs in each tank were held in four separate cages (32 x 23 x 14 centimeters) to prevent crabs from consuming the experimental oysters or each other. Every crab was fed one adult oyster daily (approximately 5.0 centimeters in length) to maximize predator cue intensity as experimental oysters would be exposed to exudates from predators and damaged conspecifics. This ensured that oysters were exposed to the most natural set of cues indicative of a predation event, which produces a strong response in oysters (Scherer et al. 2016). Crabs were replaced during the experiment as needed due to mortality. Experimental oyster baskets were rotated around the crab cages daily to reduce differences in oyster growth due to proximity to predator cues, and no differences among cages were found. The induction period was 2 months.

Shell morphology measurements

We sampled subsets of oysters to confirm that our predator cue treatments were causing control and induced oysters to exhibit different shell morphologies. Two shells were taken from every basket and three live spat were chosen from each shell for measuring spat shell characteristics after two months (number of individuals = 84 for each cue exposure treatment; 56 shells and 168 spat total). Spat shell properties were assessed by measuring shell size and shell crushing force. Oysters are roughly round at these early life stages, and we measured the shell length from the umbo to the outer shell edge to the nearest 0.01 mm using digital calipers. We took care to only measure individuals that were not bounded by cohorts to reduce any confounding effects on growth due to space limitation.

The force required to break each spat shell was quantified by a penetrometer attached to a charge amplifier (Kistler force sensor 9203 and Kistler charge amplifier 5995). The sensor probe was placed in the center of the shell, perpendicular to the shell surface. Gentle, consistent pressure was applied until the shell cracked, and the maximum force applied by the sensor to break the shell (N) was recorded. We divided shell crushing force by shell length to produce a size-standardized metric of shell strength (i.e., standardized crushing force, N per millimeter) because larger individuals naturally have a stronger shell as a byproduct of their larger size (shell thickness).

Oyster shells had their soft tissue removed and were placed in a drying oven for 48 hours at 70 degrees celsius to obtain the shell dry weight.

Data Processing Description

BCO-DMO Processing Description:

- Rounded column "Standard_force" to 2 decimal places

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Data Files

File
Oyster_morphology_2019.csv(Comma Separated Values (.csv), 7.17 KB)
MD5:ab93090926f2d391f084f94776ced38c
Primary data file for dataset 892096, version 1.

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Related Publications

Belgrad, B. A., Combs, E. M., Walton, W. C., & Smee, D. L. (2021). Use of predator cues to bolster oyster resilience for aquaculture and reef restoration. Aquaculture, 538, 736553. https://doi.org/<u>10.1016/j.aquaculture.2021.736553</u> *Methods*

Belgrad, B. A., Smee, D. L., & Weissburg, M. J. (2023). Predator signaling of multiple prey on different trophic levels structures trophic cascades. Ecology, 104(6). Portico. https://doi.org/<u>10.1002/ecy.4050</u> *Results*

Congrove, M. S., Wesson, J. A., & Allen Jr, S. K. (2009). A practical manual for remote setting in Virginia. *Methods*

Scherer, A. E., Lunt, J., Draper, A. M., & Smee, D. L. (2016). Phenotypic plasticity in oysters (Crassostrea virginica) mediated by chemical signals from predators and injured prey. Invertebrate Biology, 135(2), 97–107. Portico. https://doi.org/<u>10.1111/ivb.12120</u> *Methods*

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Related Datasets

IsRelatedTo

Belgrad, B. A., Smee, D. L., Weissburg, M. (2023) **Morphological Characteristics of Oysters from Predator Experiments at the Dauphin Island Sea Lab, AL from July to October 2020.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-03-30 doi:10.26008/1912/bco-dmo.892206.1 [view at BCO-DMO]

Belgrad, B. A., Smee, D. L., Weissburg, M. (2023) **Oyster survival difference experiments in low quality reefs in Mobile Bay, AL in September 2019.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-03-23 doi:10.26008/1912/bco-dmo.892475.1 [view at BCO-DMO]

Belgrad, B. A., Smee, D. L., Weissburg, M. (2023) **Oyster survival differences in high-quality reefs from Skidaway Island, GA from July to October 2019.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-03-23 doi:10.26008/1912/bcodmo.892464.1 [view at BCO-DMO]

Belgrad, B. A., Smee, D. L., Weissburg, M. (2023) **Oyster survival differences in mesocosm experiments at the Dauphin Island Sea Lab, AL between July and August 2019.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-03-29 doi:10.26008/1912/bco-dmo.892425.1 [view at BCO-DMO]

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Parameters

| Parameter | Description | Units |
|------------------|--|-------------------|
| spat_ID | individual spat identification number | unitless |
| shell_ID | individual shell designation that spat were settled upon | unitless |
| tank_ID | designation of tank that spat were raised within | unitless |
| basket_ID | designation of basket that spat were raised within in each tank (7 baskets per tank) | unitless |
| induct_treatment | oyster shells with spat were raised in the hatchery and either continuously exposed to predator cues (cue) or received no predator cues (no cue) | unitless |
| diameter | diameter of individual spat at the conclusion of the nursery period | millimeters (mm) |
| crushing_force_N | crushing force of individual spat at the conclusion of the nursery period | Newtons |
| standard_force | crushing force standardized by the size of individual spat (crushing force per diameter) at the conclusion of the nursery period | Newton/millimeter |
| dry_weight | dry weight of individual spat shell at the conclusion of the nursery period | grams (g) |

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Instruments

| Dataset- specific Instrument Name | digital calipers |
|--|---|
| Generic Instrument Name | calipers |
| Generic Instrument Description | A caliper (or "pair of calipers") is a device used to measure the distance between two opposite sides of an object. Many types of calipers permit reading out a measurement on a ruled scale, a dial, or a digital display. |

| Dataset-specific Instrument Name | |
|---------------------------------------|-----------------------------|
| Generic Instrument Name | Drying Oven |
| Generic Instrument Description | a heated chamber for drying |

| Dataset-specific Instrument Name | Kinlan orce sensor 9203 |
|-------------------------------------|--|
| Generic Instrument Name | Force sensor |
| Generic Instrument Description | Instrument that measures forces such as dynamic and quasistatic tensile and compression forces. Units commonly as Newtons (N). |

| Dataset-specific Instrument Name | Kistler charge amplifier 5995 |
|-------------------------------------|--|
| Generic Instrument Name | Power Amplifier |
| Generic Instrument Description | An electronic device designed to take an input signal and output an increased signal strength. |

Project Information

Collaborative Research: Keystone chemicals: Identifying general and universal molecules of fear (Identifying molecules of fear)

Coverage: Wassaw Sound, GA, US and Dauphin Island, AL

NSF abstract:

Many prey species use chemicals released in predator urine to detect imminent danger and respond appropriately, but the identity of these 'molecules of fear' remains largely unknown. This proposal examines whether prey detect different estuarine predators using the same chemical or whether the identity of the chemical signals varies. Experiments focus on common and important estuarine prey, mud crabs and oysters, and their predators including fishes, crustaceans and marine snails. Bioactive molecules are being collected from predators and prey and characterized. The goal is to determine if there are predictive relationships between either the composition of prey flesh or the predator taxon and the signal molecule. Understanding the molecular nature of these cues can determine if there are general rules governing likely signal molecules. Once identified, investigators will have the ability to precisely manipulate or control these molecules in ecological or other types of studies. Ovsters are critical to estuarine health, and they are important social, cultural and economic resources. Broader impacts of the project include training of undergraduate and graduate students from diverse backgrounds and working with aquaculture facilities and conservation managers to improve growth and survival of oysters. One response to predator cues involves creating stronger shells to deter predation. Determining the identity of cues used by oysters to detect predators can provide management options to produce oysters that either grow faster or are more resistant to predators. Project personnel is working with ovstermen to increase yields of farmed ovsters by managing chemical cues.

For marine prey, waterborne chemical cues are important sources of information regarding the threat of predation, thus, modulating non-consumptive effects of predation in many systems. Often such cues are produced when the predators consume the flesh of that prey. In nearly all cases, the specific bioactive molecules responsible for modulating these interactions are unknown, raising the question whether there is a universal molecule of fear that prey respond to. Thus, the focus of the project is to determine the generality of fear-inducing metabolites released by predators and prey in estuarine food webs. The project combines metabolomics analysis of diet-derived urinary metabolites with bioassays to identify the bioactive molecules producing responses in two prey species from different taxonomic groups and trophic levels (oysters, mud crabs). Metabolites are sampled from three types of predators, fish, gastropods or crustaceans. This project aims to: 1) identify bioactive molecules produced by several common estuarine predators from different taxa; 2) compare cues from predators that induce defenses in prey vs. changes in prey behavior; and 3) contrast the identities and effects of predator-released cues with fear-inducing molecules from injured conspecifics. By identifying and contrasting the effects of waterborne molecules that induce prey responses from six predators and injured prey, this project is yielding insights into the mechanisms that mediate non-lethal predator effects, while addressing long-standing questions related to predator-prev interactions. In addition to the search of a universal molecule of fear, the experiments are exploring the role of complementary and distinct chemical information on the specificity of prey responses to different types of predators.

This award reflects NSF's statutory mission and has been deemed worthy of support through evaluation using the Foundation's intellectual merit and broader impacts review criteria.

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Funding

| Funding Source | Award |
|--|-------------|
| NSF Division of Ocean Sciences (NSF OCE) | OCE-1948423 |
| NSF Division of Ocean Sciences (NSF OCE) | OCE-1948441 |

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