Microbial abundance and optical density from micropredator-prey microcosm experiments using sub-estuary seawater from Apalachicola Bay, FL conducted in March of 2025

Website: https://www.bco-dmo.org/dataset/998644
Data Type: experimental
Version: 1
Version Date: 2026-05-14

Project
» Excellence in Research: Assessing the Control by Multiple Micropredators on Bacterial Communities in Estuarine Environments and Characterization of Prey Lysis Products Resulting from Each Predator (Predators of bacteria)

Contributors	Affiliation	Role
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Abstract
The predation ecology of micro-predators on marine microbial consortia in 2025. This dataset includes microbial abundance and optical density from micropredator-prey microcosm experiments using sub-estuary seawater from Apalachicola Bay, FL conducted in February and March of 2025. Study Objective: Micro-predators prey on marine microbial communities and account for much of bacterial mortality. However, the predation efficacy of micro-predators in natural seawater on the autochthonous microbial community is not well understood. This is because many studies on micro-predation have focused on laboratory-grown predators and their common prey. In this study, we examine and compare how natural micro-predators feed on a mixed group of microbes in seawater versus single prey species, emphasizing the role of micro-predators in community dynamics and bacteria activity and survival. Method: The prey in this study consists of cultured bacteria in natural seawater supplemented with 50% marine broth 2216. The mixture was incubated for 48 h and then grown on 50% marine agar 2216 for 48 h. Vibrio parahaemolyticus and Roseobacter spp. used as single prey were grown on seawater yeast extract (SWYE) for 48 h, and then washed twice, and acclimatized for 12 h in sterile seawater. Experimental laboratory microcosms were prepared by selective filtration of natural seawater to select, respectively, for protists, the predatory bacteria (Halobacteriovorax, HBx) and viruses, based on their size difference. The various microcosms were then inoculated with the cultured bacterial communities and the interactions between the bacteria and the HBx, protists and virus micro-predators were monitored. Results: In all treatment groups, protist abundance decreased by 1 log-fold between 12 and 36 h, while levels in seawater control remained constant. The HBx abundance increased first in the consortium (after 36 h of incubation) as compared to 60 h for Roseobacter, with steep declines in OD and Vibrio counts also observed at this time. Optical density (OD) and total Vibrio abundance showed similar trends across microcosms, with the sharpest decline in unfiltered natural seawater (NSW) containing protists, HBx, and viruses, followed by 0.8 µm-filtered seawater (NSW-0.8), 0.1 µm-filtered seawater (NSW-0.1), and sterile seawater (SW). The OD remained stable in the consortium-only microcosm, up to 60 h, while the levels decreased slightly for other microcosms. In all the microcosms, total Vibrio and V. parahaemolyticus remained steady until 60 h, then declined sharply, in the following order; NSW < NSW-0.8 < NSW-0.1. Notably, the decline occurred first in microcosms with V. parahaemolyticus alone than in those with the consortium. Conclusion: In total, HBx showed the largest increase across microcosms (6 log-folds) as compared to the protists (increased by 2 log-folds). This demonstrates the potential effectiveness of HBx and protists in controlling bacterial populations in saltwater systems.

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Coverage

Location: Sub-estuary of Apalachicola Bay at Wolley Park, Florida
Spatial Extent: **Lat**:30.0285 **Lon**:-84.3878
Temporal Extent: 2025-02-25 - 2025-03-08

Methods & Sampling

These methods are required for sample collection, analysis, and establishment of the microcosms.
Sample collection: Seawater samples were collected from the shores of Apalachicola Bay at Wolley Park (30.0285° N, 84.3878° W). Approximately 100 liters of water were collected for laboratory analysis.

Physico-chemical properties: Physico-chemical properties of the water, including temperature, pH, salinity, dissolved solutes, and dissolved oxygen, were measured in situ using a HANNA HI 9829 Multiparameter.

Growth of bacterial species:

Bacterial consortia as potential prey
A consortium of bacteria was grown from natural seawater. Natural seawater (NSW) (100 mL) was mixed with an equal volume of freshly prepared marine broth 2216 (MB) (100 mL) to produce a 200 mL solution containing 50% MB in NSW. To control protist predation, cycloheximide was added at a final concentration of 500 mg/L, following the method described by McCambridge & Mcmeekin (1980). The resulting mixture was incubated at 25°C for 48 hours, with samples collected every 12 hours to monitor bacterial growth via optical density measurements at wavelength (λ) of 600 nm (OD600).
After 48 hours of incubation, 200 µL of the bacterial suspension was spread onto 50% marine agar 2216 (MA) plates (20 polystyrene petri dishes, 150 mm diameter, 143.0 cm^2 growth area) to establish bacterial lawns. Additionally, the bacterial culture was serially diluted to 10^{-4}, and 200 µL of the diluted suspension was spread onto 20 additional 50% MA plates to establish single growing colonies.
All plates were incubated at 28°C for 48 hours for bacterial growth. After incubation, bacteria from all 40 plates were harvested by washing off the MA plates using an L-loop and sterile seawater, pooled together, and centrifuged at 7,000 rpm for 30 minutes. The bacterial pellet was washed twice by re-suspending and homogenizing in sterile seawater, and then re-centrifuging to remove residual media.

Finally, the bacteria were re-suspended in sterile seawater and incubated for 12 hours at 4°C to allow for acclimatization before use in subsequent experiments.

Pure culture prey:
Single colonies of *Vibrio parahaemolyticus*, and *Roseobacter* isolated from seawater were used as potential prey. Pure cultures were maintained on saltwater yeast extract (SWYE) agar. Colonies were cultured on SWYE agar for 48 hours, then harvested and washed twice with sterile seawater. The cells were also acclimated in sterile seawater for 12 hours at 4°C before use.

Preparation of microcosms:
Four seawater treatment groups were established, with each treatment prepared in a final volume of 250 mL. Seawater was first allowed to stand undisturbed for 30 minutes to settle particulates. Subsequently, 250 mL of seawater was transferred into sterile flasks for each treatment.
• Treatment 1: Unfiltered natural seawater containing all native wild microbial predators (protists, predatory bacteria, and viruses).

- Treatment 2: Seawater filtered through 10 µm filter paper (Ahlstrom-Munksjo, Grade 54, 18.5 cm), then through 0.8 µm filter (28mm Corning syringe, Corning, NY) to selectively reduce protist presence while retaining smaller predators such as predatory bacteria and viruses.
- Treatment 3: The 0.8 µm filtered seawater was sequentially filtered through 0.45 µm, 0.2 µm, and 0.1 µm filters using Genesee Scientific Gen Clone vacuum-driver filter system to remove all cells, retaining only viral particles.
- Treatment 4: Seawater subjected to autoclaving at 121°C for 30 minutes to eliminate all biological activity, including viruses.

In each treatment flask, prey bacteria, either from bacterial consortia or single colony isolates, will be inoculated to achieve an initial optical density (OD_{600}) of 0.6 or an OD range (0.1-1 OD) as required. For the prey concentration experiment, the prey concentration was fixed at 1.0 to 0.1 OD. An additional control group consisting of untreated natural seawater without prey inoculation was also included to assess background microbial activity.

Incubation of microcosms

The microcosms established above were incubated at 25°C while shaking at 80 rpm for 120 hours. Samples were retrieved from each microcosm immediately before and after adding the prey/nutrient (at the beginning of the experiment), and at six-hour intervals for 120 hours to monitor the flux in micro-predators and in the Vibrio and total bacteria populations.

Co**llection of samples from microcosms**

Samples were collected and processed into different tubes for different microbial analyses, RT-qPCR, fluorescent microscopy, and flow cytometry. The samples were collected into 2 ml sterile microtubes (MSP brand Cat. No. 62-1008-13). For RT-qPCR, 1 ml of sample was collected into a sterile tube and immediately stored at -20ºC. While for flow cytometry, samples were fixed by mixing 1ml of sample with 100µL of 20 % formalin, then the sample was stored at 4 ºC. For fluorescent microscopy, 1 ml of sample was fixed with 40µL of 1% glutaraldehyde and stored at -20 ºC.

DNA extraction and quantification

The DNA extraction was carried using Qiagen DNeasy®, and PowerLyser® PowersSoil® DNA isolation kit. All centrifugation was done using Eppendorf centrifuge (5417C) at 10,000 rcf, for one minute. The DNA concentration and quality were determined using a Nanodrop Spectrophotometer ND-100, and the DNA was stored at -20ºC for RT-qPCR.

General principles of RT-qPCR

A) Instrumentation

Quantitative real-time PCR was performed using a CFX96 Real-Time PCR Detection System (Bio-Rad) coupled with a C1000 Touch Thermal Cycler. To remain within the validated detection range of the assay, all sample DNA concentrations were not below the lowest standard concentration included in the calibration curve.

B) Target genes

All RT-qPCR assays targeted the 16S rRNA gene within genomic DNA, using primers specific for the target bacterial species. The PCR template consisted of genomic DNA extracted from microcosm samples conducted in duplicate (or triplicate, where specified).

Method of quantification of gene product using RT-qPCR

Quantitative PCR (qPCR) assays were performed using the Bio-Rad CFX Real-Time PCR System to quantify target genes from the DNA extracts. Two amplification protocols were implemented. The first protocol was used for the quantification of all the bacterial targets (except *Halobacteriovorax*). It employed a two-step cycling approach consisting of an initial denaturation at 98°C for 2 min, followed by 39 amplification cycles of denaturation at 98°C for 10 s and combined annealing/extension at 60°C for 45 s. The second protocol, used for *HBx* quantification, followed a conventional three-step amplification workflow consisting of an initial denaturation at 94°C for 2 min, followed by 44 cycles of denaturation at 94°C for 30 s, annealing at 55°C for 30 s, and extension at 72°C for 40 s. For both assays, amplification specificity was evaluated using melt curve analysis initiated at 95°C and gradually increased from 65°C to 95°C in 0.5°C increments. Fluorescence data were acquired during amplification cycles, and melt curve profiles were used to confirm the absence of nonspecific amplification and primer-dimer formation.

Standard curve

A genomic DNA (gDNA)-based standard curve was successfully generated using five 10-fold serial dilutions of

a quantified DNA stock solution. The initial genomic DNA concentration (C_0, ng/µL), determined spectrophotometrically, was used to prepare standards corresponding to 10^0, 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, and 10^{-5} dilutions. Each dilution was prepared by transferring 3 µL of the preceding solution into 27 µL of nuclease-free water, producing an exact 10-fold decrease in concentration at each step. All standards were amplified in triplicate using RT-qPCR and the Ct value per DNA concentration was determined.

To convert DNA mass concentration to genome copy number, the following relationship was used:
Copy number/µl = [(DNA concentration (ng/µL) X 6.022 X 10^{23}] / [660 X length (bp) X 10^9]

Using these calculations, the DNA copy number per 10-fold dilution and Cq values were determined for each gene target, and $\log_{10}$(copy number per reaction) was plotted against the corresponding Cq values to generate the standard curve.

The standard curve exhibited strong linearity across five orders of magnitude, with a linear regression equation.

Equation 1:
$C_q = m(\log_{10} \text{DNA copy number}) + b$

where:

Cq (also called Ct) is the *quantification cycle* in quantitative PCR (qPCR).

m is the slope

b is the intercept.

For the test samples, the log of DNA concentration was calculated from Equation 2.

Equation 2:
$\text{Log}_{10} \text{DNA copy number} = (Cq-b)/m$

Population dynamics in the microcosms.
General flux of bacteria
The general flux of bacteria in the microcosms was monitored in two ways;
a) Optical density: The optical density (at λ = 600nm) was monitored at 24-hour interval, for 120 hours using 96-well microtiter plate by an absorbance reader (AccurisTM smartreaderTM 96, model MR9600, USA). The OD600 of natural seawater was used as reference at each point.

According to manufacturer specifications for the **Accuris SmartReader 96 (MR9600)**:

Absorbance range: 0.000 – ~4.000 OD

Photometric resolution: ~0.001 OD

Accuracy: typically ±1% (0–2.0 OD range)

Precision (repeatability): typically, <1%

All OD readings were within the detection limit

Calibration: Blank wells containing sterile culture medium/sterile water were included on each plate and used to identify baseline/background absorbance.

b) Real time quantitative polymerase chain reaction (Rt-qPCR): Rt-qPCR was used to quantify total bacteria, Roseobacter, Flavobacterium, total Vibrio species, and Vibrio parahaemolyticus at six-hour intervals. No template control (NTC) was also included in the experiment.

RT-qPCR for the genes was performed using an initial denaturation at 98 °C for 2 min, followed by 39 cycles of 98 °C for 10 s and 60 °C for 45 s. A melt curve analysis was conducted from 65 °C to 95 °C with 0.5 °C incremental increases to assess amplification specificity.

Flux of Micro-predators
The micro-predator flux was determined as follows:

a) Halobacteriovorax: The number of HBx in each microcosm vessel was measured using Rt-qPCR to monitor the numbers and population changes of HBx at 12-hour intervals using specific primers.

This RT-qPCR was performed with an initial denaturation at 94 °C for 2 min, followed by 44 cycles of 94 °C for 30 s, 55 °C for 30 s, and 72 °C for 40 s. Melt curve analysis was conducted from 65 °C to 95 °C with 0.5 °C incremental increases to confirm amplification specificity.

See supplemental file (primers_used.csv) for primer details.

b) Protists: Protists were quantified using CytoFLEX Beckman Coulter Life Sciences Headquarters, Indianapolis, IN 46268, USA (Model A00-1-1102) intervals. Specifically, protists were identified based on forward scatter (FSC) and side scatter properties, which provide consistent estimates of total abundance but do not distinguish heterotrophic protists from similarly sized detrital particles or provide taxonomic resolution. While the use of DNA-binding dyes could improve discrimination, our study focused on relative differences in total protist abundance across treatments within the same size class, for which flow cytometric-based counts are robust.

Initial gating was performed on forward scatter area (FSC-A) versus side scatter area (SSC-A) density plots to exclude background noise, electronic debris, and low-scatter particles. A sequential size-based gating strategy was then applied to separate particle populations corresponding to approximately 3 µm, 5 µm, and 10 µm diameter classes. The size of the dominant protist-associated population was identified within the gates, characterized by elevated FSC-A and SSC-A signals relative to other microbial fractions. However, the total events were counted and recorded over 120 s. Additional fluorescence channels, including PE-A and APC-A750-A, were monitored to improve event discrimination and verify population consistency.

Organism identifiers:

Taxon followed by Life Science Identifier (LSID) or DOI
Vibrio,urn:lsid:marinespecies.org:taxname:480248
Vibrio parahaemolyticus,urn:lsid:marinespecies.org:taxname:422811
Roseobacter,urn:lsid:marinespecies.org:taxname:567952
Halobacteriovorax,https://doi.org/10.83108/rn.518680

Data Processing Description

We used log normalization to present the data for each of the microbial population dynamics.

The Total bacteria, *Halobacteriovorax*, total *Vibrio*, *Vibrio parahaemolyticus*, and Roseobacter counts were determined using RT-qPCR. To quantify them, we first generated a standard curve. From each standard curve we obtained equations that determined the relationship between Log_{10} (DNA concentration) and Cq (Equation 1), then we used the equation to calculate DNA concentration (Equation 2).

For protist counts, we calculated the log_{10} DNA concentration of the data obtained from the flow cytometry experiment.

BCO-DMO Processing Description

- Loaded sheet 1 of "20250718. BCO_DMO Micropredator Report.xlsx" with headers from row 2, treating empty strings as missing values, into table "998644_v1_micropredator-microcosm-population-counts." Additional text description provided in row 1 of this sheet was added to the dataset metadata fields.
- Applied metadata (descriptions, standard name IDs, supplied units) to columns: Sample ID, Time (Hours), Total bacterium, OD 600, Halobacteriovorax, Protist, Total Vibrio, V. parahaemolyticus, and Roseobacter
- Renamed columns: Sample ID to sample_id, Time (Hours) to time_hrs, Total bacterium (log(DNA copy #/µL)) to total_bacterium, OD 600 to OD600, Halobacteriovorax (log(DNA copy #/µL)) to Halobacteriovorax, Protist (log(Cell counts #/µL)) to Protist, Total Vibrio (log(DNA copy #/µL)) to Total_Vibrio, V. parahaemolyticus (log(DNA copy #/µL)) to V_parahaemolyticus, and Roseobacter (log(DNA copy #/µL)) to Roseobacter
- Set types: sample_id as string, time_hrs as integer, total_bacterium/OD600/Halobacteriovorax/Protist/Total_Vibrio/V_parahaemolyticus/Roseobacter as number
- Rounded total_bacterium, Halobacteriovorax, Protist, Total_Vibrio, V_parahaemolyticus, and Roseobacter to 4 decimal places with trailing zeros preserved, to address long decimals imported from Excel

- Replaced extended ASCII µ character with "u" in sample_id column for interoperability
- Sample ID column which contained a combination of treatment description and sample ID was renamed Sample_Name and a dedicated field for Sample_ID was added (matches Sample_ID in supplemental treatment_condition_metadata.csv table).
- Output final table as "998644_v1_micropredator-microcosm-population-counts.csv"

Problem Description

NA

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Data Files

File	
998644_v1_micropredator-microcosm-population-counts.csv	(Comma Separated Values (.csv), 15.87 KB) MD5:c2f5add2280ae552c023264549c7f2a0
Primary data file for dataset ID 998644, version 1. See the "Parameters" section for detailed column information. This table shows the total bacteria counts, OD600, Halobacteriovorax, Protist, Total Vibrio and Vibrio parahaemolyticus counts of each treatment. Three different prey types; consortuium (Co), V. parahemolyticus (Vp), and Roseobacter (Ro) were used. The following is a description of each microcosm, indicating the filter size used for seawater (SW) filtration and the micropredators remaining after filtration. Co (1–4), Vp (1–4), and Ro (1–4) represent treatments with different microbial groups and filtration levels: Co = Consortium, Vp = Vibrio parahaemolyticus, Ro = Roseobacter spp. The numbers indicate filtration conditions: 1 = unfiltered seawater, 2 = 0.8 µm filtered seawater, 3 = 0.1 µm filtered seawater, 4 = sterile seawater. NSW = Natural Seawater. Results are presented in log (counts/µL).	

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Supplemental Files

File	
Experimental_design.png Graphic showing experimental design.	(Portable Network Graphics (.png), 236.35 KB) MD5:22777acb5f15651be461e34fed0cc521
Preparation_of_prey.png Graphic showing the procedure for the preparation of prey.	(Portable Network Graphics (.png), 295.80 KB) MD5:8fdd9e42cf5d58c776feac7c4b5c98b9
primers_used.csv Table of primers used. Columns include: Primer_Name, Primer name (e.g. 519F) Sequence, Sequence (e.g. "5'-CAGCAGCCGCGGTAATAC-3'") Role, How it was used (e.g. "PCR amplification of HBx")	(Comma Separated Values (.csv), 942 bytes) MD5:11e0cc92fbf01ae1f9aa49436026f2a4
treatment_condition_metadata.csv This table contains a description of the treatment groups used during the experiment Columns: Sample_ID, sample identifier Filtration_treatment, Treatment description Prey_type, Description of prey type in the treatment Initial_OD600, Indication of whether prey bacteria were inoculated to achieve an initial OD600 of 0.6	(Comma Separated Values (.csv), 710 bytes) MD5:38e7b23a68235349cc70a34b9370b477

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Related Publications

McCambridge, J., & McMeekin, T. A. (1980). Relative effects of bacterial and protozoan predators on survival of Escherichia coli in estuarine water samples. Applied and Environmental Microbiology, 40(5), 907–911. https://doi.org/10.1128/aem.40.5.907-911.1980
Methods

Williams, H. N., Lymperopoulou, D. S., Athar, R., Chauhan, A., Dickerson, T. L., Chen, H., Laws, E., Berhane, T.-K., Flowers, A. R., Bradley, N., Young, S., Blackwood, D., Murray, J., Mustapha, O., Blackwell, C., Tung, Y., & Noble, R. T. (2015). Halobacteriovorax, an underestimated predator on bacteria: potential impact relative to viruses on bacterial mortality. The ISME Journal, 10(2), 491–499. https://doi.org/10.1038/ismej.2015.129
Related Research

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Parameters

Parameter	Description	Units
sample_id	Sample identification for which treatments were used in the microcosm, where filter size for seawater and micropredators are listed	unitless
time_hrs	Time in hours of the incubation	Hours
total_bacterium	Concentration of total bacteria present in the different treatment groups during the experiment (from Rt-qPCR analysis).	log (DNA copy number/uL)
OD600	Turbidity of the sample, an indicator of the microbial biomass.	unitless
Halobacteriovorax	Concentration of Halobacteriovorax (predatory bacteria) present in the different treatment groups during the experiment (from Rt-qPCR analysis).	log (DNA copy number/uL)
Protist	Concentration of protists (eukaryotic micro-predators) present in the different treatment groups during the experiment (from flow cytometry analysis).	log (cell counts/uL)
Total_Vibrio	Concentration of total Vibrio (prey susceptible to all micro-predators) present in the different treatment groups during the experiment (from Rt-qPCR analysis).	log (DNA copy number/uL)
V_parahaemolyticus	Concentration of Vibrio parahaemolyticus (A species of Vibrio prey susceptible to all micro-predators) present in the different treatment groups during the experiment (from Rt-qPCR analysis).	log (DNA copy number/uL)
Roseobacter	Concentration of Roseobacter (A gram-negative bacteria abundant in coastal seawater, potentially susceptible to all micro-predators) present in the different treatment groups during the experiment (from Rt-qPCR analysis).	log (DNA copy number/uL)

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Instruments

Dataset-specific Instrument Name	Eppendorf centrifuge (5417C, 5430R)
Generic Instrument Name	Centrifuge
Dataset-specific Description	Eppendorf centrifuge (5417C), Eppendorf centrifuge (5430R)
Generic Instrument Description	A machine with a rapidly rotating container that applies centrifugal force to its contents, typically to separate fluids of different densities (e.g., cream from milk) or liquids from solids.

Dataset-specific Instrument Name	CytoFLEX Flow Cytometer
Generic Instrument Name	Flow Cytometer
Dataset-specific Description	CytoFLEX Flow Cytometer (Beckman Coulter Life Sciences Headquarters, Indianapolis, IN 46268, USA) (Model A00-1-1102)
Generic Instrument Description	Flow cytometers (FC or FCM) are automated instruments that quantitate properties of single cells, one cell at a time. They can measure cell size, cell granularity, the amounts of cell components such as total DNA, newly synthesized DNA, gene expression as the amount messenger RNA for a particular gene, amounts of specific surface receptors, amounts of intracellular proteins, or transient signalling events in living cells. (from: http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm)

Dataset-specific Instrument Name	Incubator-shaker
Generic Instrument Name	Incubator
Dataset-specific Description	New Brunswick Scientific Excella E24 incubator Shaker Series
Generic Instrument Description	A device in which environmental conditions (light, photoperiod, temperature, humidity, etc.) can be controlled. Note: we have more specific terms for shipboard incubators (https://www.bco-dmo.org/instrument/629001) and in-situ incubators (https://www.bco-dmo.org/instrument/494).

Dataset-specific Instrument Name	HANNA HI 9829 Multiparameter
Generic Instrument Name	Multi Parameter Portable Meter
Dataset-specific Description	HANNA HI 9829 Multiparameter
Generic Instrument Description	An analytical instrument that can measure multiple parameters, such as pH, EC, TDS, DO and temperature with one device and is portable or hand-held.

Dataset-specific Instrument Name	Smartreader
Generic Instrument Name	plate reader
Dataset-specific Description	Eppendorf centrifuge (5430R) AccurisTM smartreaderTM 96, model MR9600, USA
Generic Instrument Description	Plate readers (also known as microplate readers) are laboratory instruments designed to detect biological, chemical or physical events of samples in microtiter plates. They are widely used in research, drug discovery, bioassay validation, quality control and manufacturing processes in the pharmaceutical and biotechnological industry and academic organizations. Sample reactions can be assayed in 6-1536 well format microtiter plates. The most common microplate format used in academic research laboratories or clinical diagnostic laboratories is 96-well (8 by 12 matrix) with a typical reaction volume between 100 and 200 uL per well. Higher density microplates (384- or 1536-well microplates) are typically used for screening applications, when throughput (number of samples per day processed) and assay cost per sample become critical parameters, with a typical assay volume between 5 and 50 µL per well. Common detection modes for microplate assays are absorbance, fluorescence intensity, luminescence, time-resolved fluorescence, and fluorescence polarization. From: http://en.wikipedia.org/wiki/Plate_reader, 2014-09-0-23.

Dataset-specific Instrument Name	Absorbance Microplate Reader Q4
Generic Instrument Name	plate reader
Dataset-specific Description	Absorbance Microplate Reader Q4 (BIO-TEK Instruments Inc., USA)
Generic Instrument Description	Plate readers (also known as microplate readers) are laboratory instruments designed to detect biological, chemical or physical events of samples in microtiter plates. They are widely used in research, drug discovery, bioassay validation, quality control and manufacturing processes in the pharmaceutical and biotechnological industry and academic organizations. Sample reactions can be assayed in 6-1536 well format microtiter plates. The most common microplate format used in academic research laboratories or clinical diagnostic laboratories is 96-well (8 by 12 matrix) with a typical reaction volume between 100 and 200 uL per well. Higher density microplates (384- or 1536-well microplates) are typically used for screening applications, when throughput (number of samples per day processed) and assay cost per sample become critical parameters, with a typical assay volume between 5 and 50 µL per well. Common detection modes for microplate assays are absorbance, fluorescence intensity, luminescence, time-resolved fluorescence, and fluorescence polarization. From: http://en.wikipedia.org/wiki/Plate_reader, 2014-09-0-23.

Dataset-specific Instrument Name	CFX96 Touch Deep Well Real-Time PCR Detection System
Generic Instrument Name	qPCR Thermal Cycler
Dataset-specific Description	CFX96 Real-Time PCR Detection System (Bio-Rad) coupled with a C1000 Touch Thermal Cycler. (Bio-Rad, Hercules, CA 94547, USA)
Generic Instrument Description	An instrument for quantitative polymerase chain reaction (qPCR), also known as real-time polymerase chain reaction (Real-Time PCR).

Dataset-specific Instrument Name	NanoDrop ND-1000 UV spectrophotometer
Generic Instrument Name	Spectrophotometer
Dataset-specific Description	NanoDrop ND-1000 UV spectrophotometer (Thermo Scientific, Wilmington, USA)
Generic Instrument Description	An instrument used to measure the relative absorption of electromagnetic radiation of different wavelengths in the near infra-red, visible and ultraviolet wavebands by samples.

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Project Information

Excellence in Research: Assessing the Control by Multiple Micropredators on Bacterial Communities in Estuarine Environments and Characterization of Prey Lysis Products Resulting from Each Predator (Predators of bacteria)

NSF Award Abstract:

Microbes are the most abundant organisms on Earth and play an important role as degraders, cycling nutrients in the environment. Too many or too few bacteria may disrupt a sensitive ecological balance and proper functioning of environmental processes such as carbon, nitrogen and phosphorus cycles. The abundance of bacteria populations in any given environment is controlled by various biological, chemical and physical mechanisms. Among the biological agents are microscopic predators, or micropredators, of bacteria. The most studied of these are protists, viruses that infect bacteria, and a group of bacteria collectively known as the Bdellovibrio and like organisms (BALOs). These micropredators prey upon certain bacteria to obtain required nutrients or other cellular material for their replication. In the process, cellular products from the prey bacteria are released into the environment and utilized as nutrients by other microbes. Although the micropredators co-occur, and likely interact, in nature, most experimental studies have investigated their activities individually, rather than collectively. As a result, little is known about their collective role in controlling bacteria populations and the cycling of nutrients. The goal of the proposed research is to address this gap in knowledge by investigating all three as a collective group under simulated natural conditions representing a range of temperature, salinity and abundance of prey. This project is conducted at two Historically Black Universities (HBCUs) with strong records of training and mentoring students and postdocs from underrepresented populations in science. The project benefits up to 100 students by providing unique and meaningful educational and research training experiences at the undergraduate and graduate student levels and for early-career scientist. Specific activities include courses on scientific writing and presenting results at annual project workshops as well as national and international scientific meetings. Graduate students are being trained in modern advanced methodologies in chemistry and microbiology. There is an ongoing assessment module to document education and training outcomes.

Up to now, the two mainly accepted mechanisms of mortality in bacterial populations are heterotrophic protist grazing and viral infection. Increasingly, it has become evident that an understudied group of predatory bacteria, BALOs, can also contribute to bacterial mortality. Yet, the mechanisms underlying the dynamics of BALO-prey interactions are poorly understood, as are the interactions among the micropredators, BALOs, protists and bacterial viruses. Ultimately, these processes may have contrasting influences on the structure and functioning of the microbial loop, including impacting higher trophic levels and biogeochemical cycles. The investigators hypothesize that environmental factors significantly influence how mortality in bacterial populations is partitioned among the micropredators. To test this hypothesis researchers are (1) investigating the interactions amongst the micropredators, (2) examining the molecular-level composition and dynamics of dissolved organic matter as the result of the different mortality processes by the NMR/ FT-ICR mass spectrometry (MS) hybrid approach, and (3) modeling these tri-trophic dynamics. Intellectual Merit: Results from this research will define a new mechanistic understanding of mortality dynamics that influence the microbial loop and oceanic biogeochemical cycles.

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-1948758

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