



Bigelow Laboratory for Ocean Sciences

Research Experience for Undergraduates The Gulf of Maine and the World Ocean

**REU Symposium Program & Abstracts
Thursday, August 4, 2016**



Oral Program

10:00 Opening Remarks:

10:15 Sydney Tiemann, Colby College, Waterville, ME
USING THE MARINE PROTOZOAN PARASITE *PERKINSUS* TO INVESTIGATE POTENTIAL MALARIA TREATMENT

Mentors: Dr. José Antonio Fernández Robledo

10:30 Catherine Carlisle¹ University of Virginia, VA, USA
MICROBIAL ASSOCIATIONS WITH BENTHIC INVERTEBRATES: HUNTING FOR THE *ENDOBUGULA SERTULA* SYMBIOSIS IN THE COASTAL GULF OF MAINE

Mentors: Dr. Pete Countway

10:45 Tatiana Barreto, University of Puerto Rico, Río Piedras Campus, Puerto Rico
DOES THE BROWNING OF THE GULF OF MAINE BY HUMIC ACID INPUTS INTERFERE WITH ORGANIC PHOSPHORUS UTILIZATION BY THE TOXIC DINOFLAGELLATE *ALEXANDRIUM FUNDYENSE*?

Mentor: Dr. Cindy Heil

11:00 Abigail Onos, Smith College, Northampton, MA
MIXOTROPHY IN THE COCCOLITHOPHORES, *PLEUROCHRYSID* AND *EMILIANA*

Mentor: Dr. Barney Balch

11:15 **Break (15 minutes)** *****

11:30 Nathaniel Matteson, University of New Mexico, Albuquerque, NM
THE EFFECT OF INCREASED TEMPERATURE AND CO₂ LEVELS ON THE TEMPORAL ABUNDANCE OF KEY MARINE VIRUSES

Mentor: Dr. Joaquín Martínez Martínez, Ilana Gilg

11:45 Darcia Gonzalez, University of Texas Rio Grande Valley, Edinburg, TX
EFFECTS OF OCEAN ACIDIFICATION AND WARMING ON THE DEVELOPMENT AND SURVIVAL OF *HOMARUS AMERICANUS* LARVAE

Mentor: J. Waller and Dr. David Fields

12:00 Yannik Buchi, Colby College, Waterville, ME
THE MICROBIAL ECOLOGY OF *NEREIS (HEDISTE) DIVERSICOLOR*

Mentor: Dr. Dave Emerson, Jarrod Scott

12:15 Megan Harder, Carleton College, Northfield, MN
RATES OF MICROBIALY-MEDIATED IRON OXIDATION IN MARINE WORM BURROWS

Mentor: Drs. Jake Beam, Dave Emerson

12:30 **Break (45 minutes)** *****

1:15 Torrie Renee, McGill University, Montréal, Canada
MODELING PARTICULATE BEHAVIOR IN GEOTRACES TRACE METAL DATA

Mentor: Dr. Ben Twining

1:30 Andrew Hirzel, University of Miami, Coral Gables, FL
CELL BIOVOLUME: TESTING ACROSS SHAPES, SIZES, AND ALGORITHMS

Mentor: Dr. Nicole Poulton, Laura Lubelczyk

- 1:45 Johanna Holman, Husson University, Bangor, ME
THE ECOLOGY AND EVOLUTION OF A RECENTLY DISCOVERED, HIGHLY-DIVERSE, AND GLOBALLY DISTRIBUTED MICROBIAL DARK MATTER ARCHAEAL PHYLUM—
WOESEARCHAEOTA
Mentor: Dr. Pachiadaki M, Becraft ED, Stepanauskas R
- 2:00 Cynthia Michaud, University of Rhode Island, Kingston, RI
GROWTH OF *EMILIANA HUXLEYI* ON PHOSPHONATE
Mentor: Drs. Whitney, Lomas
- 2:15 Break (15 min) *******
- 2:30 Mahallelah Shauer, Humboldt State University, Arcata, CA
ASSESSING THE IMPACTS OF PROJECTED CLIMATE CHANGE ON THE ECOPHYSIOLOGY OF *SACCHARINA LATISSIMA*
Mentor: Dr. Nichole Price
- 2:45 Emma Posega Rappeleye, Carleton College, Northfield, MN
OPERATIONAL FORECASTING OF JELLYFISH IN THE GULF OF MAINE USING CITIZEN REPORTS
Mentor: Dr. Nick Record
- 3:00 Briar Bragdon, Univ of New England, ME
EFFECT OF MYOVIRUS INFECTION ON SYNECHOCOCCUS PHOTOSYNTHETIC EFFICIENCY
Mentor: Drs. Sheri Floge, David Fields
- 3:15 Amanda Herzog, Wheaton College, Norton, MA
LINKING CHEMICAL COMPOSITION TO TOXICITY OF FRESH AND WEATHERED OIL SAMPLES COLLECTED FROM THE 2010 GULF OF MEXICO OIL SPILL
Mentor: Dr. Chris Aepli

Abstracts and Posters

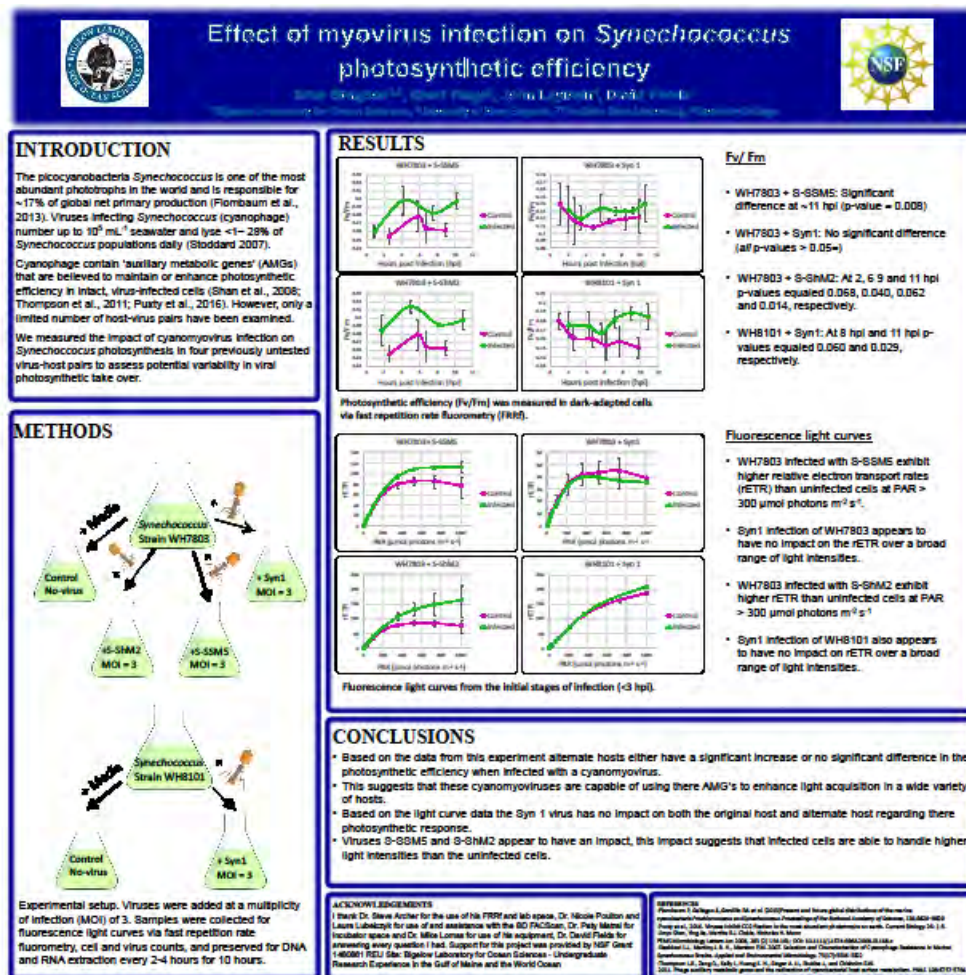
EFFECT OF MYOVIRUS INFECTION ON SYNECHOCOCCUS PHOTOSYNTHETIC EFFICIENCY

Bragdon BL^{1,4}, Flöge SA³, Layman JJ^{1,2}, Fields DM¹, Sullivan MB³¹Bigelow Laboratory for Ocean Sciences, ME²Bowdoin College, ME

³The Ohio State University, OH

⁴The University of New England, ME

Synechococcus is one of the most abundant phototrophs in the world and is responsible for approximately 17% of global net primary production. Cyanophage co-occur with *Synechococcus* and over evolutionary time scales have acquired genes involved in host photosynthetic electron transport. While the ability of cyanophage to utilize ‘photosynthesis genes’ to maintain photosynthetic efficiency has been well-documented in finely tuned host-virus pairs, the effectiveness of such genes in ‘alternative’ hosts remains untested. Using fast repetition rate fluorometry (FRRf) we measured photosynthetic efficiency of virus-infected *Synechococcus* during the first 10 h of infection in one finely tuned host-virus pair (WH8101 and Syn1) and three alternative host-virus pairs (WH7803 and S-SSM5, WH7803 and S-ShM2, WH7803 and Syn1). In all virus-host pairs photosynthetic efficiency (Fv/ Fm) was either equal to or greater than that in control, uninfected cultures. These findings suggest that cyanomyoviruses are capable of enhancing light energy acquisition in diverse hosts.



THE MICROBIAL ECOLOGY OF *NEREIS (HEDISTE) DIVERSICOLOR*

Büchi Y^{1,2}, Emerson, D², Beam J², Scott J²
¹Colby College, ²Bigelow Laboratory for Ocean Sciences

The polychaete worm *Nereis (Hediste) diversicolor* is a bioturbating organism used by toxicologists to monitor sediment and water quality. Scientists are still puzzled by the mechanisms behind its resistance to temperature and salinity changes, high sulfide concentrations, and contaminated environments that make it such a useful tool. Little is known about the interactions between *N. diversicolor* and its environment, making it hard to explain this organism's ecological importance. A few studies have shed light on the high metabolic activity of this species' gut, suggesting it may contain a dynamic microenvironment that could explain *N. diversicolor*'s resilience in a wide variety of environmental conditions. This project investigated the microenvironment inside *N. diversicolor* using whole worms, whole starved worms, and fecal pellets. DNA analysis using an OTU and classification-based approach with a 16S database revealed a high abundance of proteobacteria (44%) and SAR406 (44%) in whole worms and whole starved worms, which differed significantly from the microbial communities found in fecal pellets. These results indicate the presence of a sulfur-cycling bacterial community in the guts of *N. diversicolor*.

The Microbiome of the Common Ragworm *Nereis (Hediste) diversicolor*

Yannik Büchi^{1,2}, Jacob Beam², Jarrod Scott², David Emerson²
¹Colby College, ²Bigelow Laboratory for Ocean Sciences



INTRODUCTION

Nereis (Hediste) diversicolor
 O.F. Müller

• up to 20 cm long

• pigment colors: yellow, green, red, brown

Habitat

- East and Western North Atlantic
- found in mudflats, estuaries, intertidal shallow, brackish water of varying salinity and temperatures



The goal of this project is to generate a detailed analysis of the microbiology inside this organism.

BACKGROUND

Why look inside *N. diversicolor*?

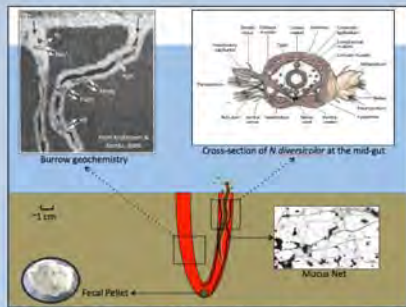
Ecological Agent

- Feeding behavior
- filter-feeder
- carnivore
- detritivore

Geochemical Agent

Environmental Monitor

- resistant to temperature and salinity changes, high sediment sulfide concentrations
- small gut-residence time of sediments
- perfect tool for monitoring water quality & trace metals in sediments



RESULTS

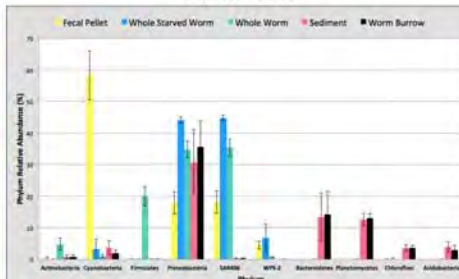


Figure 1: Relative abundance (%) of most prominent phyla across the five sample types: fecal pellets (n=3), whole starved worms (n=3), whole worms (n=3), sediment (n=2) and worm burrow (n=2) from "the Eddy" site. Sediment and worm burrow samples were collected by Jacob Beam.

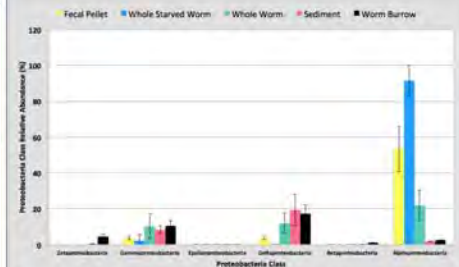


Figure 2: Relative abundance (%) of Proteobacteria classes across the five sample types.

DISCUSSION

- Alphaproteobacteria and SAR406 ("Marinimicrobia") enriched in fecal pellet and worm samples but not in sediment or burrow wall
- High presence of Alphaproteobacteria in fecal pellets and worms
 - Rhodobacterales in fecal pellet and starved worms
 - Rickettsiales in fecal pellets and whole worms

METHODS



DISCUSSION (CONT.)

- Deltaproteobacteria involved in sulfur cycle
- SAR406 may have important implications in the sulfur cycle, sulfur-based energy metabolism, sulfide detoxification?
- Cyanobacteria are elevated in fecal pellets

FUTURE DIRECTIONS

- Isolate and analyze flora of isolated guts
- Sample across environmental gradients
- Study feeding behavior and its effect on gut flora
- Understand metabolic capabilities of SAR406
- Check for endosymbiotic relationship between *N. diversicolor* and guts

ACKNOWLEDGEMENTS

Special thanks to Megan Harder, Eric Huang, Sarabeth George, Matt Wade, the Darling Marine Center, and to all the people behind the 2016 Bigelow REU Summer Program. Support for this project was provided by NSF Grant 1460861 REU Site: Bigelow Laboratory for Ocean Sciences - Undergraduate Research Experience in the Gulf of Maine and the World Ocean

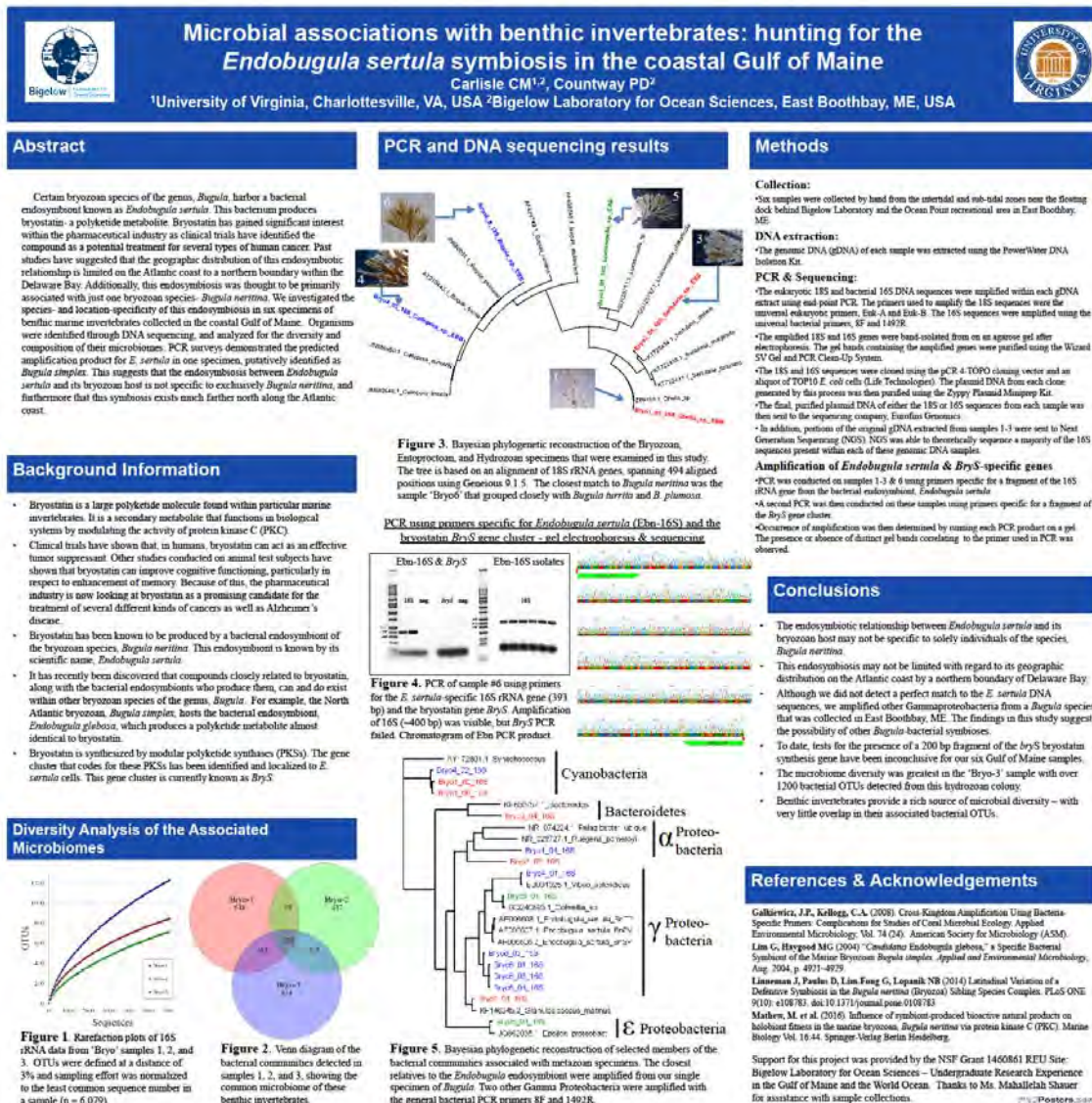
Microbial associations with benthic invertebrates: hunting for the *Endobugula sertula* symbiosis in the coastal Gulf of Maine

Carlisle CM^{1,2}, Countway PD²

¹University of Virginia, VA, USA

²Bigelow Laboratory for Ocean Sciences, East Boothbay, ME, USA

Certain bryozoan species of the genus, *Bugula*, harbor a bacterial endosymbiont known as *Endobugula sertula*. This bacterium produces bryostatin- a polyketide metabolite. Bryostatin has gained significant interest within the pharmaceutical industry as clinical trials have identified the compound as a potential treatment for several types of human cancer. Past studies have suggested that the geographic distribution of this endosymbiotic relationship is limited on the Atlantic coast to a northern boundary within the Delaware Bay. Additionally, this endosymbiosis was thought to be primarily associated with just one bryozoan species- *Bugula neritina*. We investigated the species- and location-specificity of this endosymbiosis in six specimens of benthic marine invertebrates collected in the coastal Gulf of Maine. Organisms were identified through DNA sequencing, and analyzed for the diversity and composition of their microbiomes. PCR surveys demonstrated the predicted amplification product for *E. sertula* in one specimen, putatively identified as *Bugula simplex*. This suggests that the endosymbiosis between *Endobugula sertula* and its bryozoan host is not specific to exclusively *Bugula neritina*, and furthermore that this symbiosis exists much farther north along the Atlantic coast.

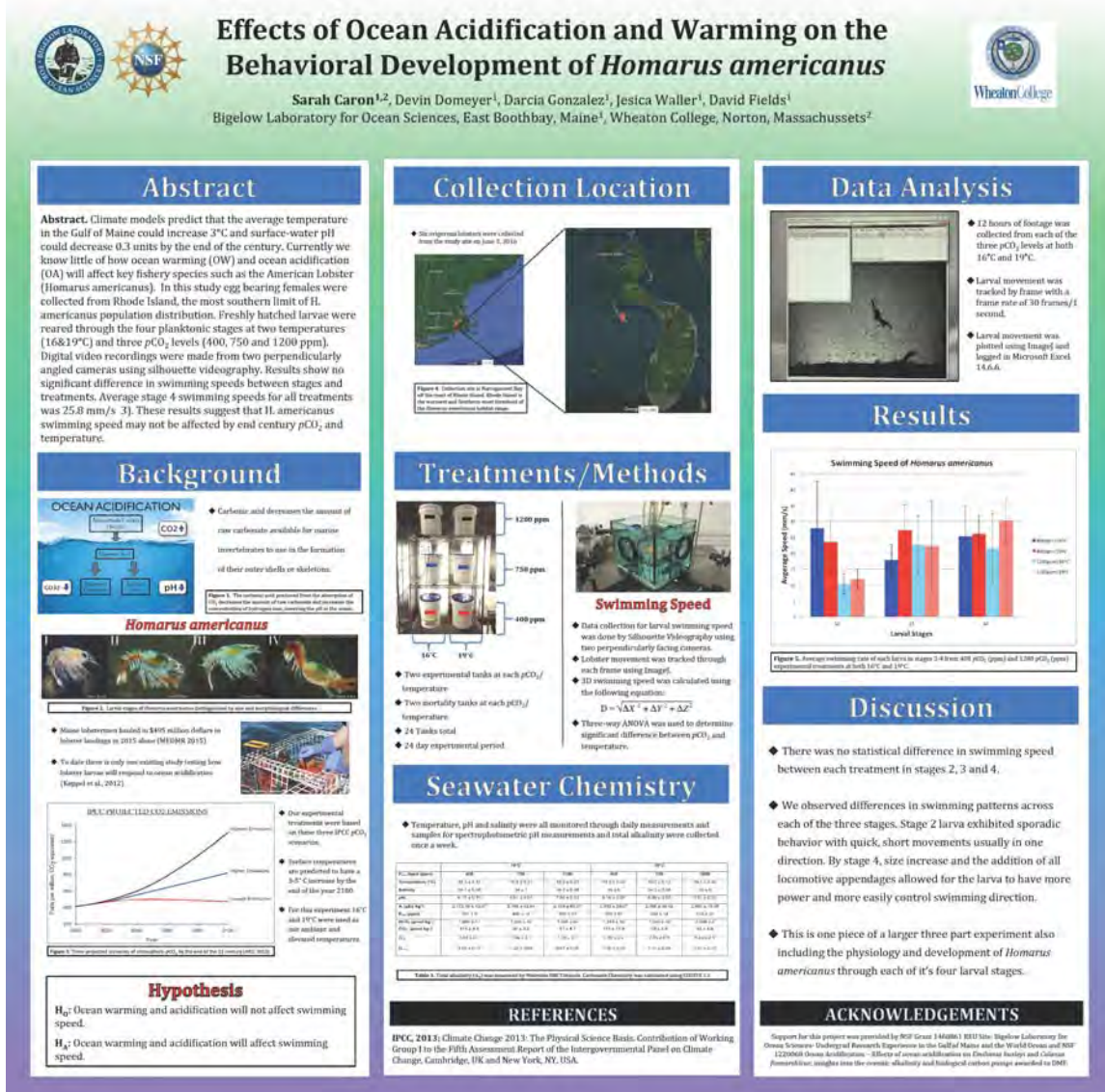


EFFECTS OF OCEAN ACIDIFICATION AND WARMING ON THE BEHAVIORAL DEVELOPMENT OF *HOMARUS AMERICANUS*

Caron S^{1,2}, Domeyer D¹, Gonzalez D¹, Waller J¹, Fields D¹

Bigelow Laboratory for Ocean Sciences, East Boothbay, Maine¹,
Wheaton College, Norton, Massachusetts²

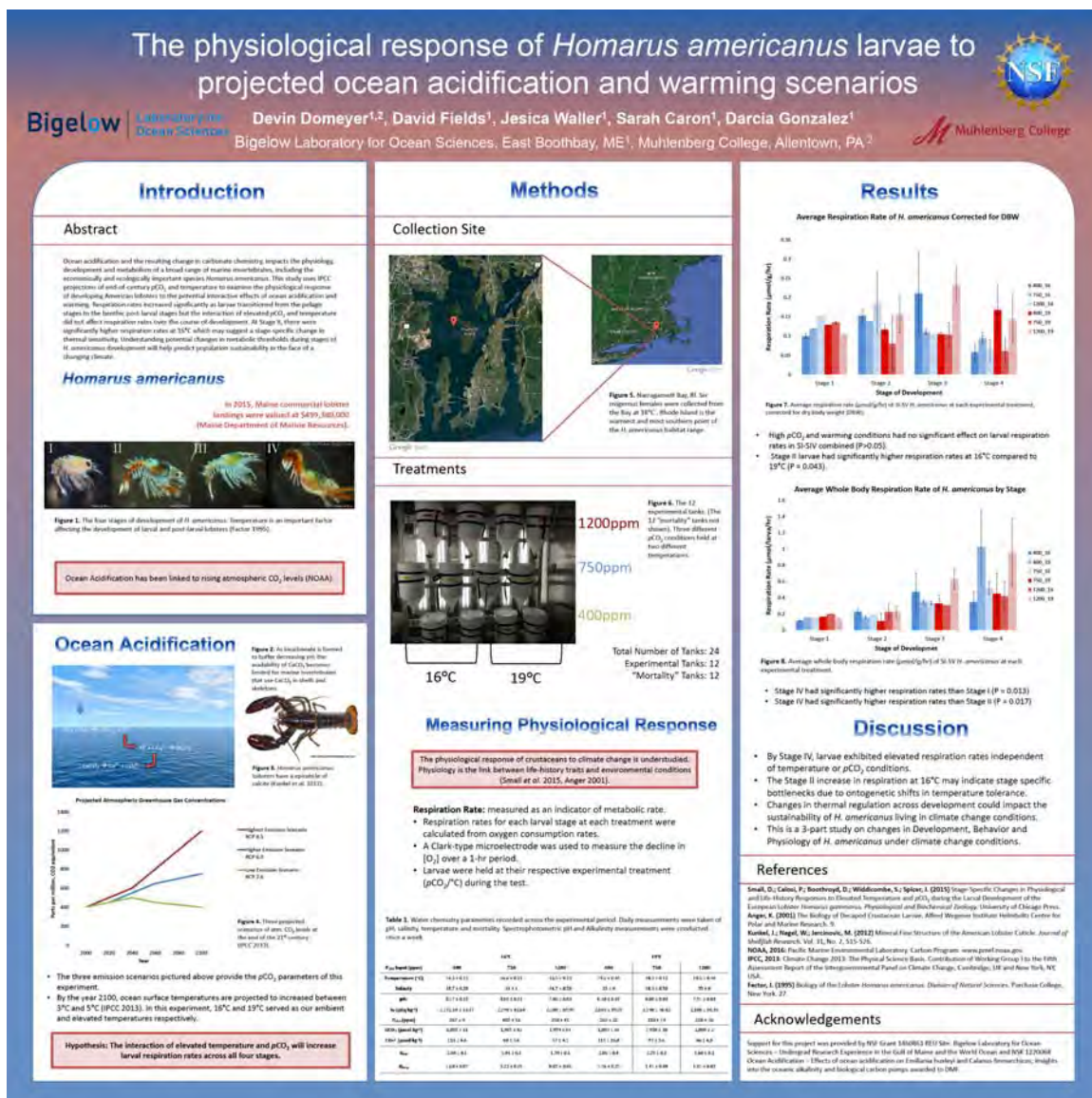
Abstract. Climate models predict that the average temperature in the Gulf of Maine could increase 3°C and surface-water pH could decrease 0.3 units by the end of the century. Currently we know little of how ocean warming (OW) and ocean acidification (OA) will affect key fishery species such as the American Lobster (*Homarus americanus*). In this study egg bearing females were collected from Rhode Island, the most southern limit of *H. americanus* population distribution. Freshly hatched larvae were reared through the four planktonic stages at two temperatures (16&19°C) and three pCO₂ levels (400, 750 and 1200 ppm). Digital video recordings were made from two perpendicularly angled cameras using silhouette videography. Results show no significant difference in swimming speeds between stages and treatments. Average stage 4 swimming speeds for all treatments was 25.8 mm/s (± 3). These results suggest that *H. americanus* swimming speed may not be affected by end century pCO₂ and temperature.



THE PHYSIOLOGICAL RESPONSE OF *HOMARUS AMERICANUS* LARVAE TO THE POTENTIAL INTERACTION OF OCEAN ACIDIFICATION AND WARMING SCENARIOS

Devin Domeyer^{1,2}, David Fields¹, Jessica Waller¹, Darcia Gonzalez¹, Sarah Caron¹
Bigelow Laboratory for Ocean Sciences¹, Muhlenberg College²

Ocean acidification and the resulting change in carbonate chemistry, impacts the physiology, development and metabolism of a broad range of marine invertebrates, including the economically and ecologically important species *Homarus americanus*. This study uses IPCC projections of end-of-century $p\text{CO}_2$ and temperature to examine the physiological response of developing American lobsters to the potential interactive effects of ocean acidification and warming. Respiration rates increased significantly as larvae transitioned from the pelagic stages to the benthic post-larval stages but the interaction of elevated $p\text{CO}_2$ and temperature did not affect respiration rates over the course of development. At Stage II, there were significantly higher respiration rates at 16°C which may suggest a stage-specific change in thermal sensitivity. Understanding potential changes in metabolic thresholds during stages of *H. americanus* development will help predict population sustainability in response to a changing climate.



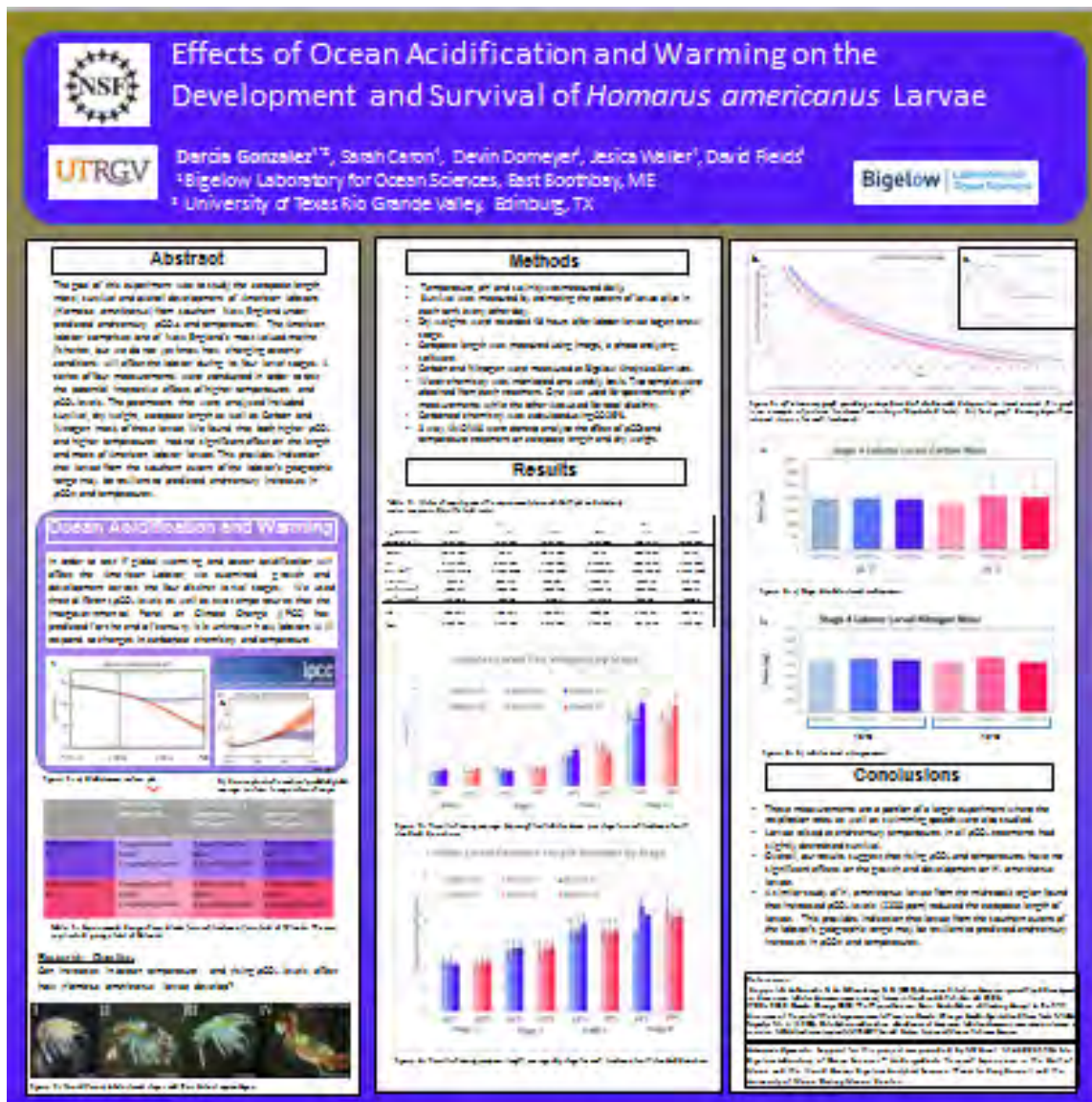
EFFECTS OF OCEAN ACIDIFICATION AND WARMING ON THE CARAPACE LENGTH, MASS AND SURVIVAL OF *HOMARUS AMERICANUS* LARVAE

Gonzalez DN^{1, 2}, Caron S¹, Domeyer D¹, Waller J¹, Fields D¹

¹Bigelow Laboratory for Ocean Sciences, East Boothbay, ME

²University of Texas Rio Grande Valley, Edinburg, TX

The American lobster (*Homarus americanus*) comprises one of New England's most valued marine fisheries, but we do not yet know how changing oceanic conditions will affect the lobster during its four planktonic stages. The goal of this experiment was to study the carapace length, mass, survival and developmental rates of *H. americanus* from southern New England under IPCC predicted end-century $p\text{CO}_2$ and temperatures levels. Our data showed no significant effect of higher $p\text{CO}_2$ and temperature on the length and mass of American lobster larvae. The results of this experiment indicate that larvae from the southern extent of the lobster's geographic range may be resilient to predicted end-century increases in $p\text{CO}_2$ and temperatures.



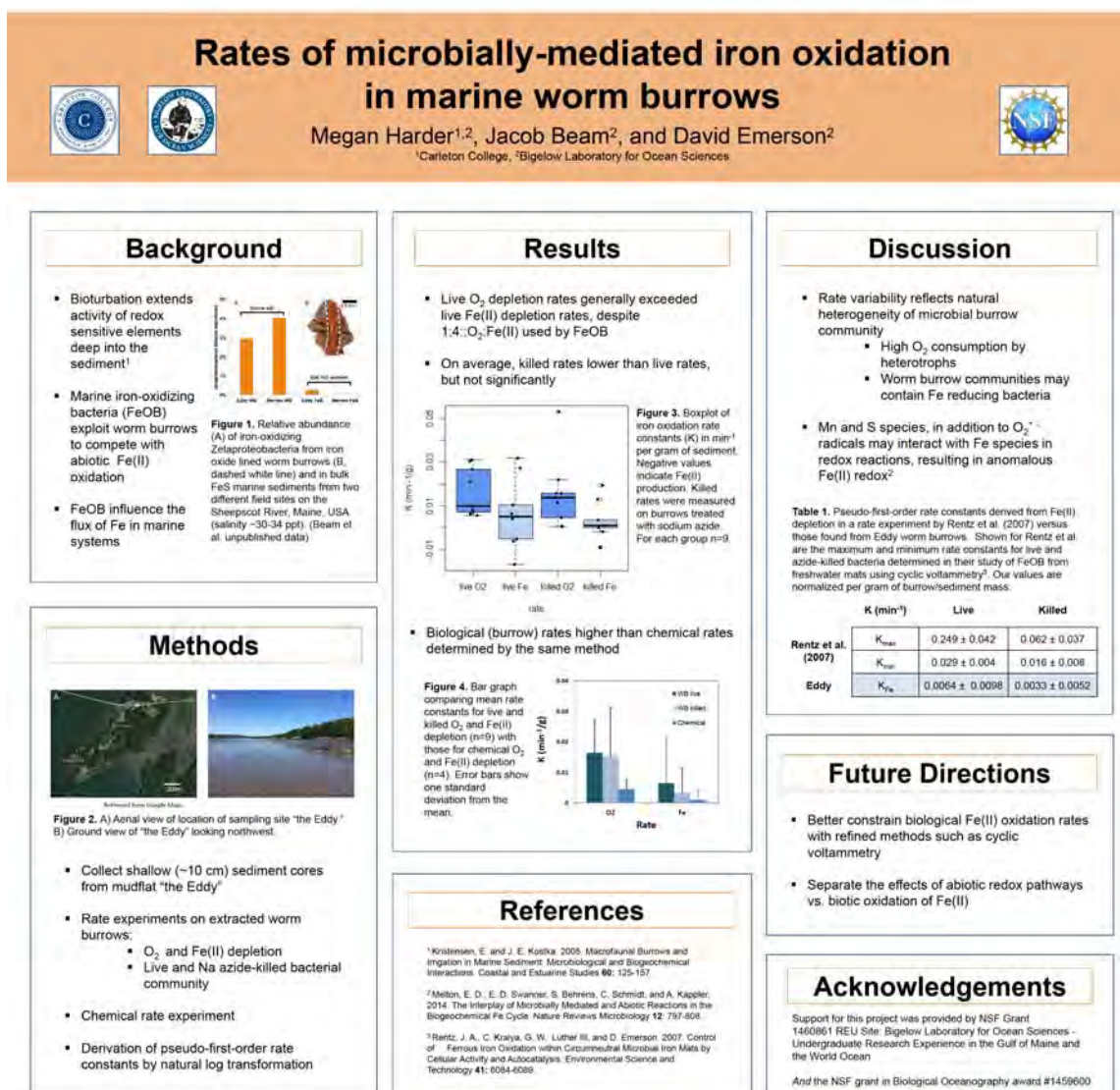
RATES OF MICROBially-MEDIATED IRON OXIDATION IN MARINE WORM BURROWS

Harder MA¹, Beam JP², Emerson D²

¹Carleton College

²Bigelow Laboratory for Ocean Sciences

Bioturbation provides suitable conditions for cycling of redox sensitive elements by extending the sediment water interface, aerating and irrigating the sediment. Microaerobic, iron-oxidizing bacteria exploit this refuge provided by marine worm burrows to compete against abiotic iron oxidation. In a simple rate experiment, worm burrows were extracted from sediment cores taken from a local mudflat and allowed to respire for 30 minutes in artificial seawater in microaerobic conditions. Pseudo-first-order rate constants were derived via natural log transformations of oxygen and Fe(II) depletion data from live and killed treatments. Pseudo-first-order rate constants derived from oxygen depletion data were constrained to $0.016 \pm 0.0068 \text{ min}^{-1} \text{ g}^{-1}$ and $0.015 \pm 0.01 \text{ min}^{-1} \text{ g}^{-1}$, while those derived from Fe(II) depletion were $0.0064 \pm 0.0098 \text{ min}^{-1} \text{ g}^{-1}$ and $0.0033 \pm 0.0052 \text{ min}^{-1} \text{ g}^{-1}$, live and killed, respectively. These rates are higher than rates of chemical Fe(II) oxidation, indicating that iron oxidation was biologically facilitated. The results from this study reflect the natural variability of heterogeneous microbial communities. Further investigation is necessary to constrain biological and chemical contributions to iron oxidation in marine sediments.



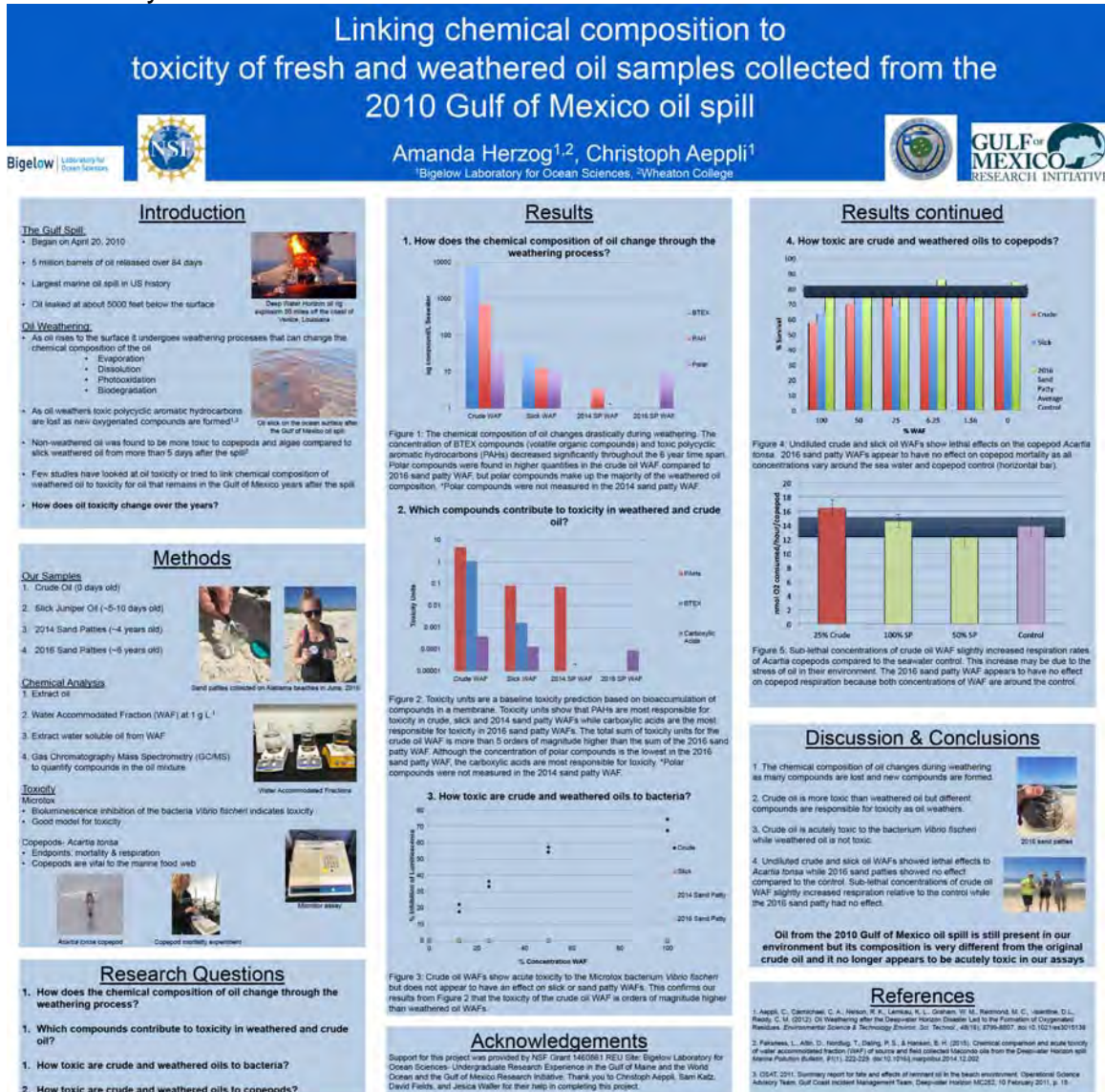
LINKING CHEMICAL COMPOSITION TO TOXICITY OF FRESH AND WEATHERED OIL SAMPLES COLLECTED FROM THE 2010 GULF OF MEXICO OIL SPILL

Herzog AN^{1,2}, Aeppli C¹

1. Bigelow Laboratory for Ocean Sciences, East Boothbay, ME, 04544

2. Wheaton College, Norton, MA, 02766

Oil residues from the 2010 *Deepwater Horizon* oil spill are still found on beaches in the Gulf of Mexico, however few studies have looked at toxicity for such highly weathered oil. To fill this gap, crude and weathered oil samples were collected for chemical and toxic analysis using water-accommodated fractions (WAFs). The chemical composition of oil samples showed that toxic aromatic compounds were almost completely lost in weathered oil and that oxygenated compounds were responsible for predicted toxicity in six-year-old weathered samples. Toxicity tests using luminescent bacteria (*Vibrio fischeri*) showed that crude oil was acutely toxic, while weathered oil had no effect. Copepod (*Acartia tonsa*) assays showed greater mortality in full-strength crude and slick oil (5-30 days old) WAFs compared to highly weathered WAFs, which showed no effect on mortality. Sub-lethal concentrations of crude WAFs showed slight increases in respiration rates, while highly weathered WAFs showed no effect. Although oil from the *Deepwater Horizon* spill still exists in the environment, the chemical composition of weathered oil is much different from crude oil and no longer appears to be acutely toxic.



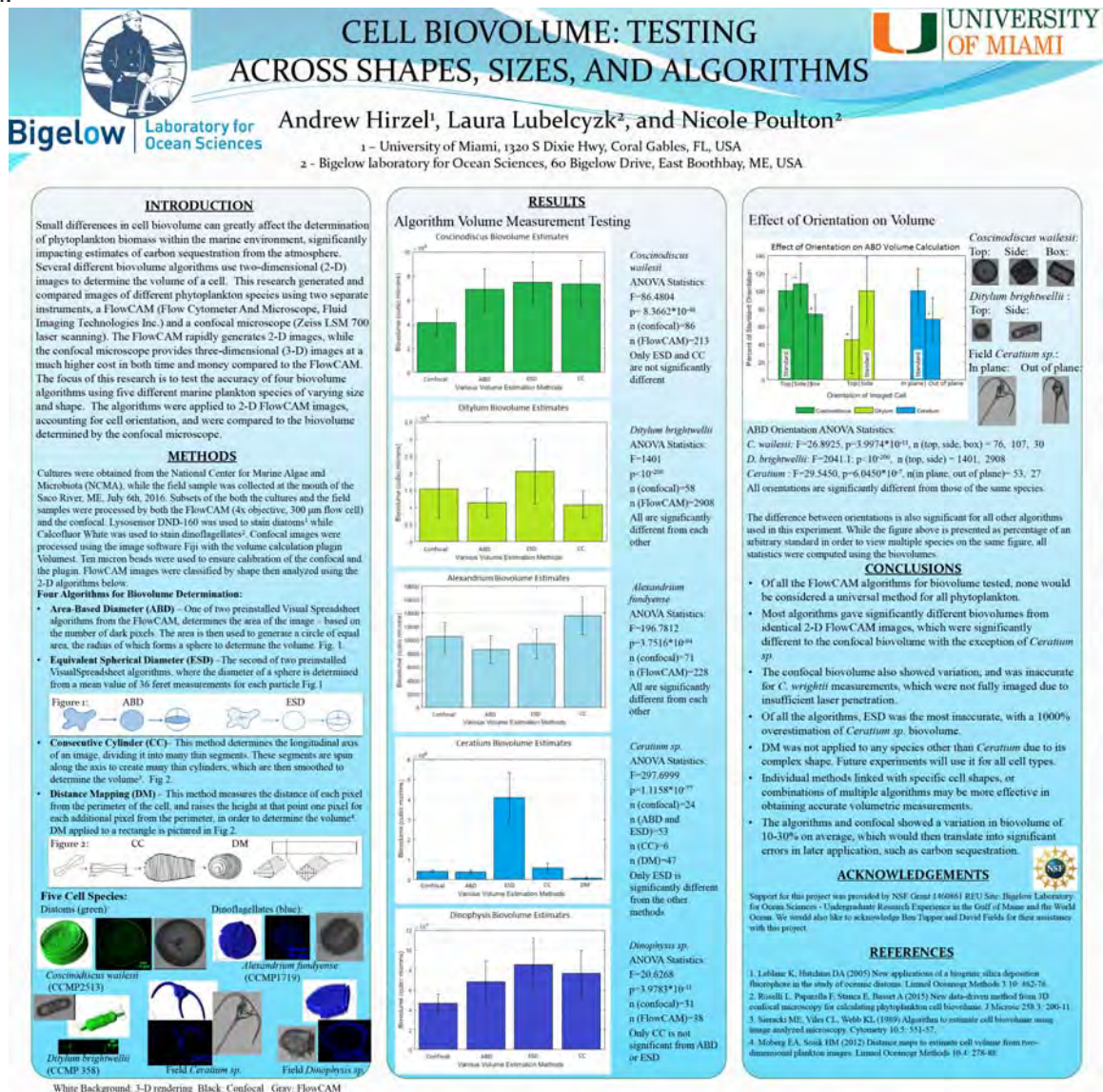
CELL BIOVOLUME: TESTING ACROSS SHAPES, SIZES, AND ALGORITHMS

Andrew Hirzel¹, Laura Lubelczyk², and Nicole Poulton²

¹University of Miami, 1320 S Dixie Hwy, Coral Gables, FL, USA

²Bigelow laboratory for Ocean Sciences, 60 Bigelow Drive, East Boothbay, ME, USA

Small differences in cell biovolume can greatly affect the determination of phytoplankton biomass within the marine environment, significantly impacting estimates of carbon sequestration from the atmosphere. Several different algorithms use two-dimensional (2-D) images to determine the volume of a cell. This research endeavors to test the accuracy of four algorithms; area-based diameter, equivalent spherical diameter (ESD), consecutive cylinder, and distance mapping, on a variety of phytoplankton species. Cultured *Coscinodiscus wailesii*, *Ditylum brightwellii*, *Alexandrium fundyense*, and field *Ceratium* and *Dinophysis* spp. were imaged in 2-D using an imaging cytometer (FlowCAM) and in three dimensions (3-D) using a confocal microscope (Zeiss LSM700). The algorithms were applied to 2-D images, taking into account the orientation of the organisms, and compared to the biovolume determined by the confocal microscope. ESD was found to be the least reliable of the algorithms, overestimating *Ceratium* sp. biovolume by 1000%. No method was accurate for all cell types, (including confocal microscopy) showing varying accuracy for different cell shapes. The biovolume methods showed an average variation in biovolume of 10-30%, which would then translate into significant errors in later application.



THE ECOLOGY AND EVOLUTION OF A RECENTLY DISCOVERED, HIGHLY-DIVERSE, AND GLOBALLY DISTRIBUTED MICROBIAL DARK MATTER ARCHAEAL PHYLUM—WOESEARCHAEOTA

Holman JM¹, Pachiadaki M², Becraft ED², Stepanauskas R²

¹Husson University
²Bigelow Laboratory for Ocean Science

Woesearchaeota is a recently discovered, poorly understood, candidate Archaea phylum. To elucidate the distribution, evolutionary history and metabolic capabilities of the Woesearchaeota phylum, samples were taken from various sites around the world, single cells were sorted, and whole genome sequencing was performed to obtain partial genomes from 24 potential Woesearchaeota.

Phylogenetic analysis showed that 14 of 24 single amplified genomes belong to the Woesearchaeota. We report, for the first time, the presence of this phylum in marine pelagic waters and hydrothermal vents. Average nucleotide identity and 16S rRNA gene identity revealed low genetic relatedness between the SAGs, indicating that Woesearchaeota may not be a single phylum. Analysis of the metabolism of the genomes revealed flagellar motility, potential for degradation of organic matter and near-complete glycolytic pathway and TCA cycle. No lipid synthesis genes were annotated in any of the genomes. Hypothetical proteins comprise up to half of each of the genomes. Some of them appear to encode for putative conserved domains associated with adhesion and antibiotic/toxin production. We hypothesize that Woesearchaeota degrade organic matter and might be pathogenic to bacteria.

The ecology and evolution of a recently discovered, highly-diverse, and globally distributed microbial dark matter Archaea—Woesearchaeota

Bigelow | Laboratory for Ocean Sciences

Holman JM¹, Pachiadaki M², Becraft ED², Stepanauskas R²



ABSTRACT

Woesearchaeota is a recently discovered, poorly understood, candidate Archaea phylum. To elucidate the distribution, evolutionary history and metabolic capabilities of the Woesearchaeota phylum, samples were taken from various sites around the world, single cells were sorted, and whole genome sequencing was performed to obtain partial genomes from 24 potential Woesearchaeota.

Phylogenetic analysis showed that 14 of 24 single amplified genomes belong to the Woesearchaeota. We report, for the first time, the presence of this phylum in marine pelagic waters and hydrothermal vents. Average nucleotide identity and 16S rRNA gene identity revealed low genetic relatedness between the SAGs, indicating that Woesearchaeota may not be a single phylum. Analysis of the metabolism of the genomes revealed flagellar motility, potential for degradation of organic matter and near-complete glycolytic pathway and TCA cycle. No lipid synthesis genes were annotated in any of the genomes. Hypothetical proteins comprise up to half of each of the genomes. Some of them appear to encode for putative conserved domains associated with adhesion and antibiotic/toxin production. We hypothesize that Woesearchaeota degrade organic matter and might be pathogenic to bacteria.

BACKGROUND

Archaea are important drivers of geochemical cycles, are globally distributed and are evolutionarily important as our closest prokaryotic relatives.

Castelle et al., 2015, published a paper introducing Woesearchaeota as a candidate phylum¹.

Since Castelle et al. published their paper, Woesearchaeota have been found in a variety of environments¹.

Castelle et al. used metagenomic sequencing and assembly, which results in composite genomes that represent all of the genetic potential within a population.

Single cell genomics is representative of one cell, and can thus improve genomic and metabolic resolution.



PURPOSE/METHODS

Purpose: to determine the evolutionary relatedness of 24 potential Woesearchaeota genomes and analyze their metabolic capabilities.

Samples for single cell analysis were collected from deep subseafloor aquifers, goldmines, and for the first time marine environments.

Cells were sorted using Fluorescent Activated Cell Sorting. DNA was amplified using Whole Genome Amplification, and genomes were sequenced with Illumina technology.

16S rRNA genes were aligned, maximum likelihood phylogeny was generated, and all genomes annotated.



EVOLUTION

16S maximum likelihood phylogeny identified 14 Woesearchaeota, 2 unknown candidate phyla containing 5 genomes, 1 Paecearchaeota, and 3 Micraearchaeota. Average nucleotide identity and 16S rRNA identity analysis showed low relatedness between Woesearchaeota.

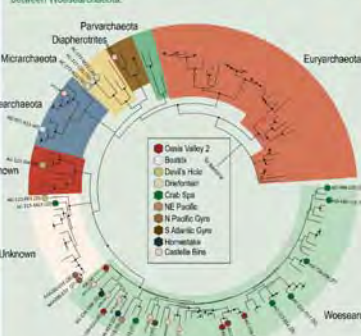


Figure 2. Maximum likelihood phylogeny of 16S rRNA genes. Undifferentiated lineages represent 16S sequences from GenBank.

Discussion

Low relatedness between Woesearchaeota genomes and the high-level of genetic diversity contained within the phylum may indicate the potential for a super-phylum (i.e. multiple phyla).

METABOLISM

Results

Woesearchaeota were found to have small genomes. Up to half of genomes were comprised of hypothetical genes, some with potential adhesive and antibiotic domains. No genes for lipid biosynthesis were detected. Genes for toxin synthesis were found, as well as genes coding for extracellular peptidases.



Figure 3. Composite metabolic diagram of 14 Woesearchaeota SAGs. Legend indicates total number of SAGs from each site. Figure squares indicate total number of SAGs from sites that have at least one representative gene of the pathway.

Discussion

Presence of partial pathways for glycolysis, pyruvate, TCA cycle, and pentose phosphate metabolism and oxidative phosphorylation in each of the genomes indicates an ability to oxidize organic carbon and produce ATP. Presence of aldehyde and alcohol dehydrogenases in most of the genomes may be indicative of fermentative lifestyles.

The coding potential for flagella, toxins, and adhesion molecules, and the absence of lipid synthesis may indicate pathogenicity to bacteria.

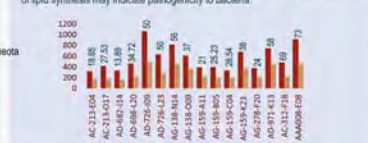


Figure 4. Number of hypothetical genes to total number of gene coding sequences. Numbers at top of bars indicate genome completeness.

ACKNOWLEDGEMENTS

Support for this project was provided by NSF Grant 1408811 REU Site: Bigelow Laboratory for Ocean Sciences - Undergraduate Research Experience in the Gulf of Maine and the World Ocean. NSF grants OCS-12392, OCS-1441717, and OCS-1136488.

Special thanks to our collaborators and the REU program coordinators.

REFERENCES

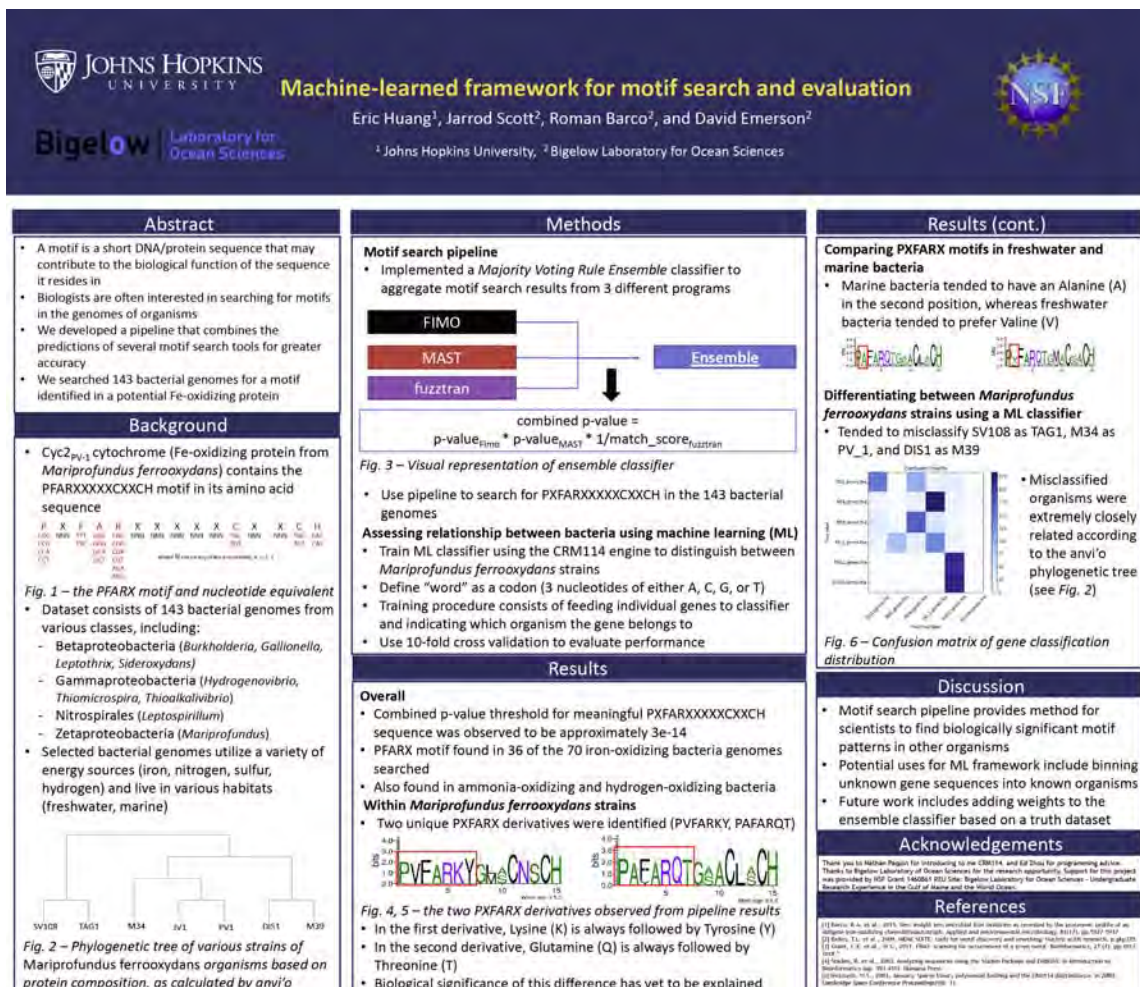
- Castelle CJ, Wrighton KC, Thomas BC, et al. Genomic exploration of domain archaea highlights roles for organisms from new phyla in anaerobic carbon cycling. *Cell* 2015;256:690-701. doi:10.1016/j.cell.2015.01.014.
- Ortiz-Alvarez R, Casamayor EO. High occurrence of Paecearchaeota and Woesearchaeota (Archaea superphylum DPANN) in the surface waters of oligotrophic high-altitude lakes. *Environ Microbiol Rep* 2015;8(2):210-217. doi:10.1111/1751-2225.12370.

UPGRADING MOTIF SEARCH TOOLS WITH ENSEMBLE LEARNING

Huang E1, Scott J2, Emerson D2

1Johns Hopkins University, 2Bigelow Laboratory for Ocean Sciences

A motif is a short and recurring DNA or protein sequence that may determine the biological function of the sequence it resides in. Once a motif is identified in one organism's genome, biologists may be interested in finding its occurrences in other organisms' genomes. Many tools have been developed to search for occurrences of a given motif in gene and protein sequences. However, if used alone, a search tool may miss motif occurrences in a sequence or erroneously identify a motif in a sequence where one does not exist. To reduce the errors from individual tools, we developed a pipeline that combines the predictions of three motif search tools (FIMO, MAST, and fuzztran) via a majority vote ensemble learning method. As a proof-of-concept, we searched 193 different bacterial genomes for an amino-acid motif (PXFARXXXXCXCH) from a protein that is thought to play a role in the iron oxidizing processes of *Acidithiobacillus ferrooxidans*. We discovered that the motif was found in 34 of 70 iron-oxidizing bacteria genomes and was also present in some species of ammonia-oxidizing bacteria.



QUANTIFYING THE MARINE VIRAL SHUNT: CARBON FLOW ACROSS TROPHIC LEVELS

Layman JJ^{1,2}, Fløge, SA³, Bragdon, BL⁴, Fields, DM¹, Sullivan MB³

¹Bigelow Laboratory for Ocean Sciences, ME

²Bowdoin College, ME

³The Ohio State University, OH

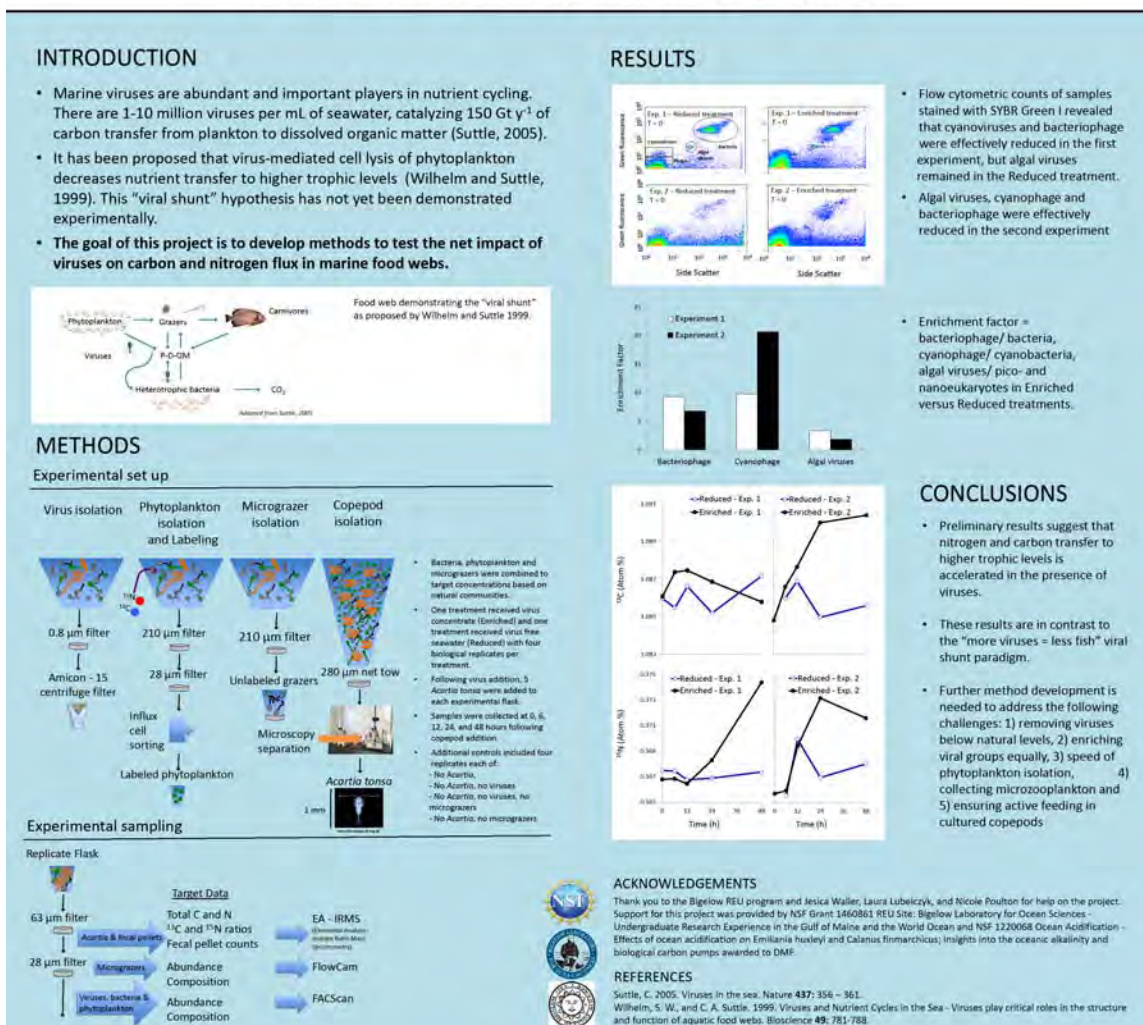
⁴The University of New England, ME

Marine viruses are abundant and important players in nutrient cycling, transferring an estimated 150 gigatons of carbon per year from phytoplankton to dissolved organic matter. However, quantifying virus-mediated carbon flux has remained challenging due to technical limitations. We have applied a combination of fluorescence activated cell sorting (FACS) and isotope ratio mass spectrometry (IRMS) to quantify viral impacts on carbon and nitrogen flux in complex planktonic food webs collected from coastal waters of West Boothbay, Maine. Our goal was to preserve predator-prey interactions among bacteria, archaea, phytoplankton and micrograzers while generating treatments with reduced and enriched virus abundance, relative to natural waters. Using ¹³C- and ¹⁵N-labeled phytoplankton we observed enhanced transfer of carbon and nitrogen to the copepod *Acartia tonsa* in treatments enriched in natural virus populations. These findings are in contrast to hypothetical models that assume trophic transfer is dependent upon prey density, and thus negatively impacted by viral infection of phytoplankton and bacteria.

Nutrient flow across trophic levels: testing the viral shunt hypothesis

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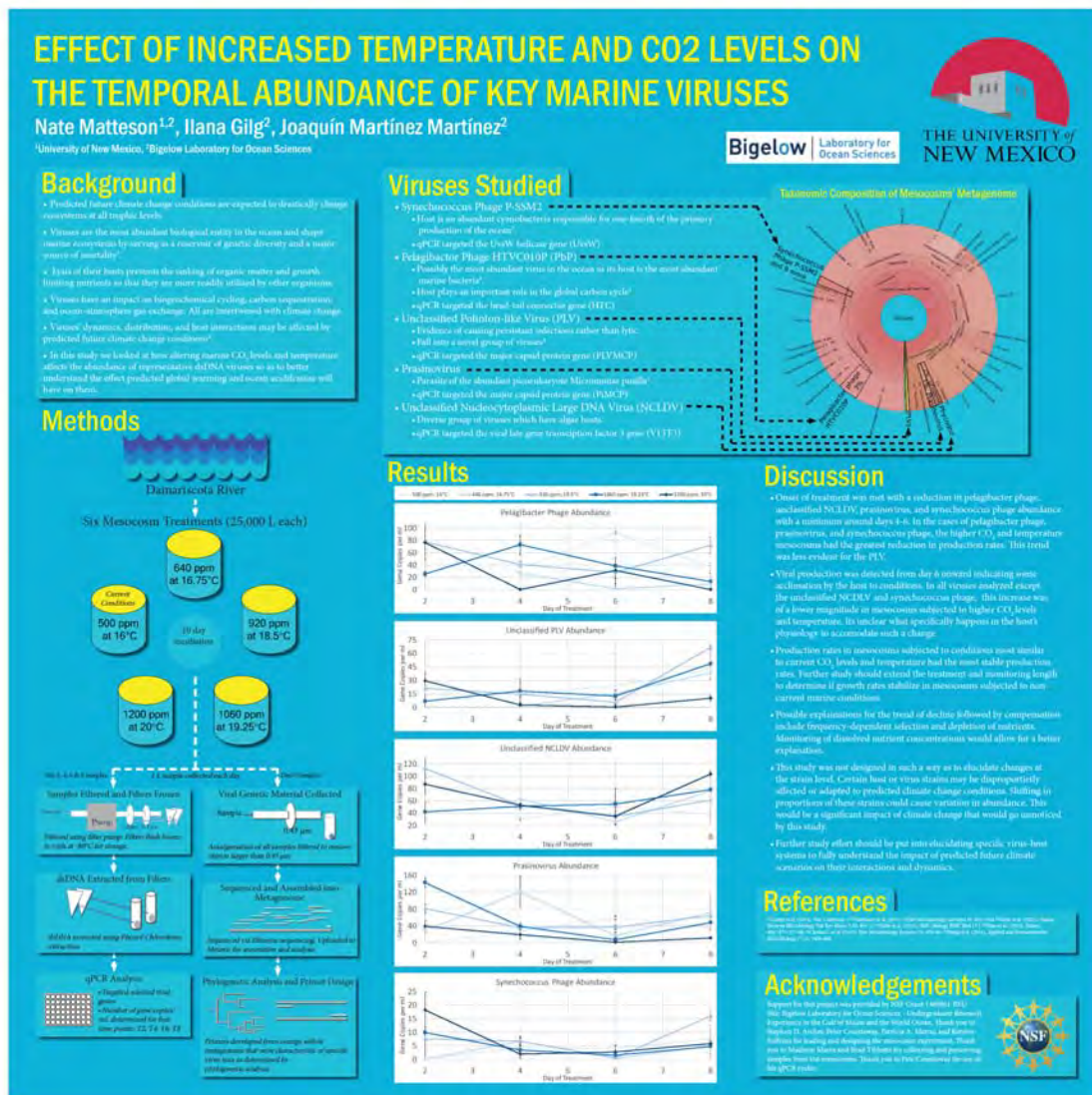


THE EFFECT OF INCREASED TEMPERATURE AND CO₂ LEVELS ON THE TEMPORAL ABUNDANCE OF KEY MARINE VIRUSES.

Nate Matteson^{1,2}, Ilana Gilg², Joaquín Martínez Martínez²

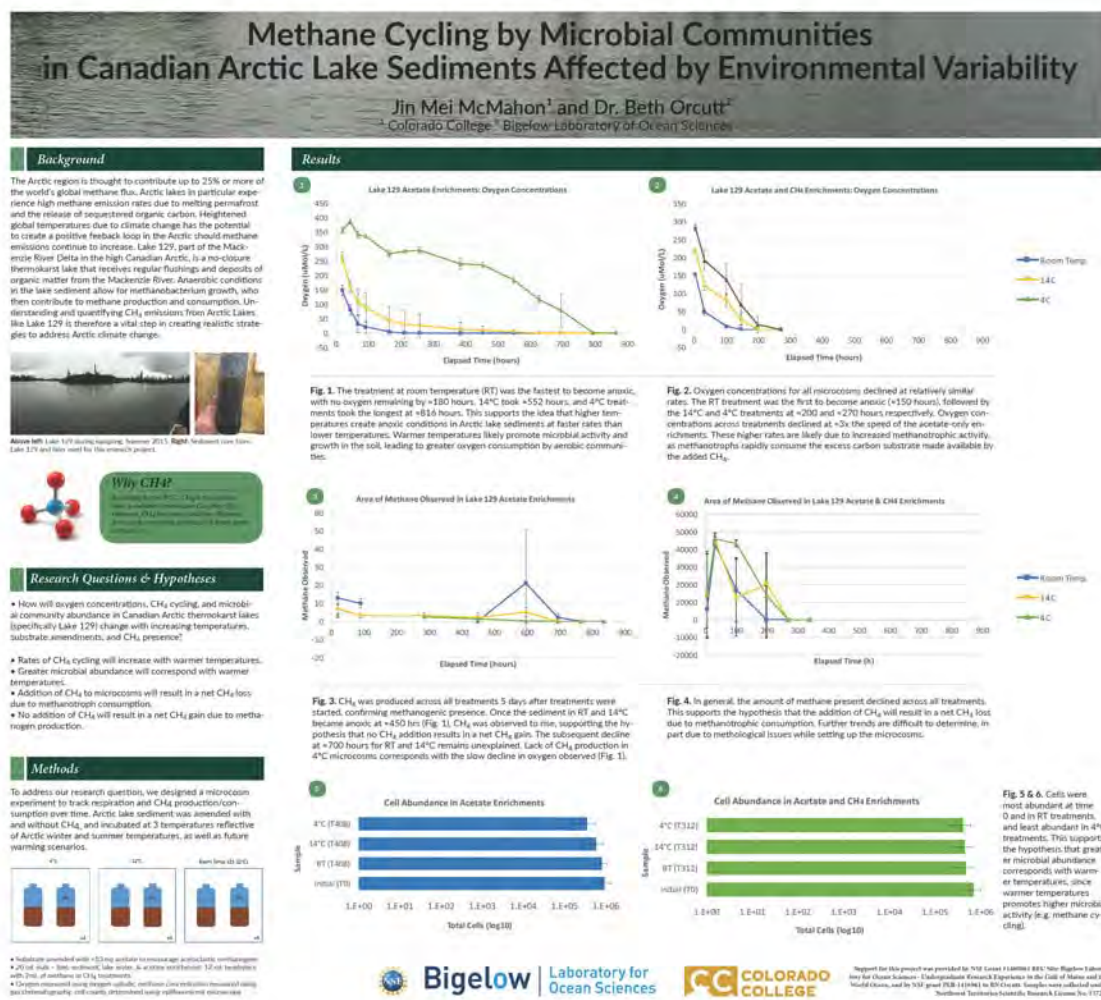
¹University of New Mexico, ²Bigelow Laboratory for Ocean Sciences

Predicted future climate change conditions are expected to drastically change ecosystems at all trophic levels. One understudied aspect of these changes is their impact on viruses which are known to play a critical role in shaping marine ecosystems. This current study yields new information about the interaction between viruses and climate change by determining how the abundance of representative dsDNA virus taxa change in response to increasing temperature and CO₂. Six mesocosms filled with seawater from the Damariscotta River were subjected to simultaneous temperature and CO₂ modulation, ranging from 500 -1200 ppm CO₂ and 16-20°C, for an 8 day period. Viral abundance was determined through qPCR using primers developed through study of the mesocosms' metagenome. A majority of virus taxa analyzed showed an initial drop in abundance following onset of treatment which the mesocosm with marine conditions most similar to current conditions did not experience. Viral production rates, in general, became positive after 6 days of treatment indicating accommodation to predicted climate change conditions. It is still unclear how these conditions affect viral dynamics.



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The Arctic region is thought to contribute up to 25% of the world's global methane flux, with Arctic lakes in particular experiencing high emission rates due to melting permafrost. Lake 129, part of the Canadian Arctic's Mackenzie River Delta, is a no-closure thermokarst lake that receives regular organic matter deposits, allowing microbial communities to thrive. We set up acetate-amended Lake 129 sediment microcosms to investigate the effects of temperature variability and methane presence on microbial communities (specifically methanobacterium), methane cycling, and oxygen concentrations over time. Overall we saw that temperature had a significant influence on microbial activity, with warmer temperatures supporting larger communities of microbes and a more rapid oxygen depreciation. Addition of methane to microcosms had a noticeable impact on microcosm development, and both net gain and net loss of methane were observed, confirming active methanobacterium presence. While further research is necessary to fully understand the effects of temperature and microbial communities on methane cycling, this project constitutes a necessary step in determining what role methanobacterium play in influencing the Arctic methane flux.



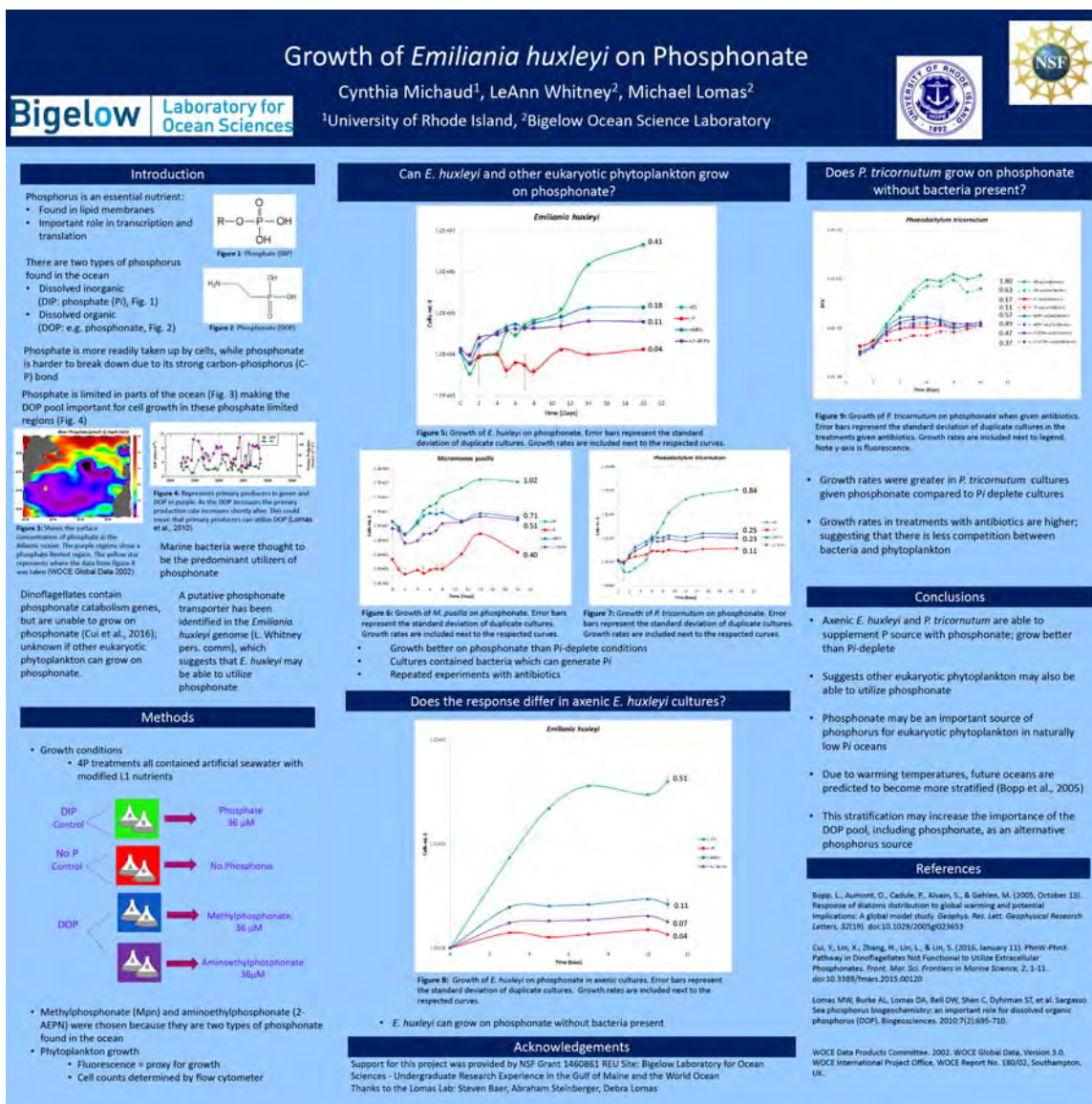
GROWTH OF *EMILIANA HUXLEYI* ON PHOSPHONATE

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Phosphorus (P), an essential nutrient for phytoplankton growth, is generally found in two forms in the oceans: dissolved inorganic (phosphate, P_i), the preferred form, and dissolved organic (e.g. phosphonate) which requires enzymatic processing. Phosphonate comprises up to 25% of the DOP pool in the ocean and can be an important source of P to prokaryotic phytoplankton in low P_i regions. The ability of eukaryotic phytoplankton to utilize phosphonate is underexplored. We investigated phosphonate utilization in axenic cultures of *Emiliana huxleyi* and other eukaryotic phytoplankton. The phytoplankton were able to grow on phosphonate at ecologically relevant growth rates. We also show differential expression of a putative phosphonate transporter in response to growth on different P sources. These results suggest eukaryotic phytoplankton may be able to supplement their P source with phosphonate; a key consideration given the prediction that future oceans may become more stratified and thus could increase the importance of DOP, including phosphonate, in supporting growth.

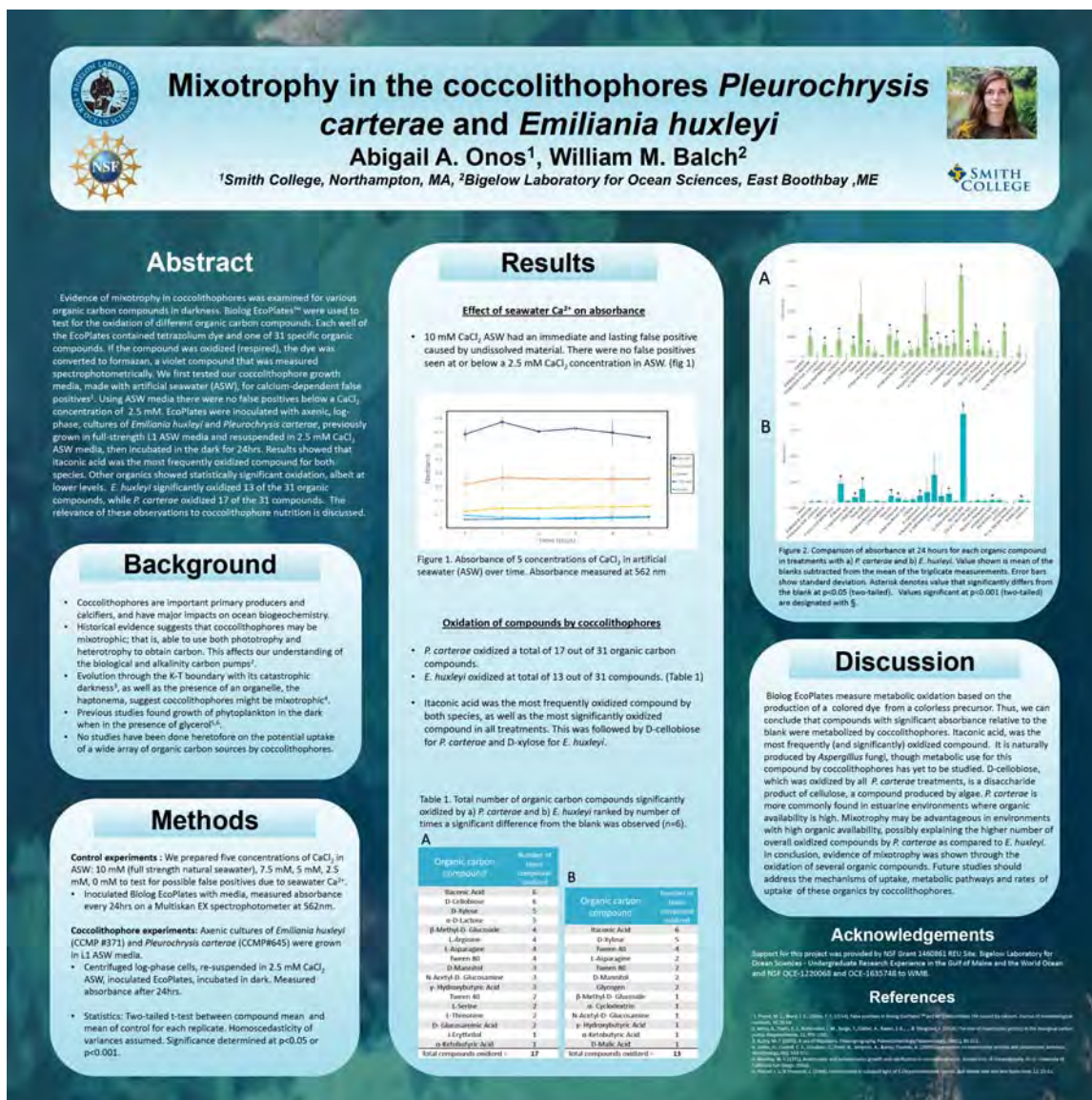


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Evidence of mixotrophy in coccolithophores was examined for various organic carbon compounds in darkness. Biolog EcoPlates™ were used to test for the oxidation of different organic carbon compounds. Each well of the EcoPlates contained tetrazolium dye and one of 31 organic compounds. If the compound was oxidized (respired), the dye was converted to formazan, a violet compound that was measured spectrophotometrically. We first tested our coccolithophore growth media, made with artificial seawater [ASW], for calcium-dependent false positives (reported previously). Using ASW media there were no false positives below a CaCl_2 concentration of 2.5mM. EcoPlates were inoculated with axenic, log-phase, cultures of *Emiliania huxleyi* and *Pleurochrysis carterae* previously grown in full-strength ASW media and resuspended in 2.5mM CaCl_2 ASW media, then incubated in the dark for 24hrs. Results showed that itaconic acid was the most frequently oxidized compound for both species. Other organics showed statistically significant oxidation, albeit at lower levels. *E. huxleyi* significantly oxidized 13 of the 31 organic compounds, while *P. carterae* oxidized 17 of the 31 compounds. The relevance of these observations to coccolithophore nutrition is discussed.



PHYTOPLANKTON GROWTH IN THE PRESENCE OF MICROPLASTICS

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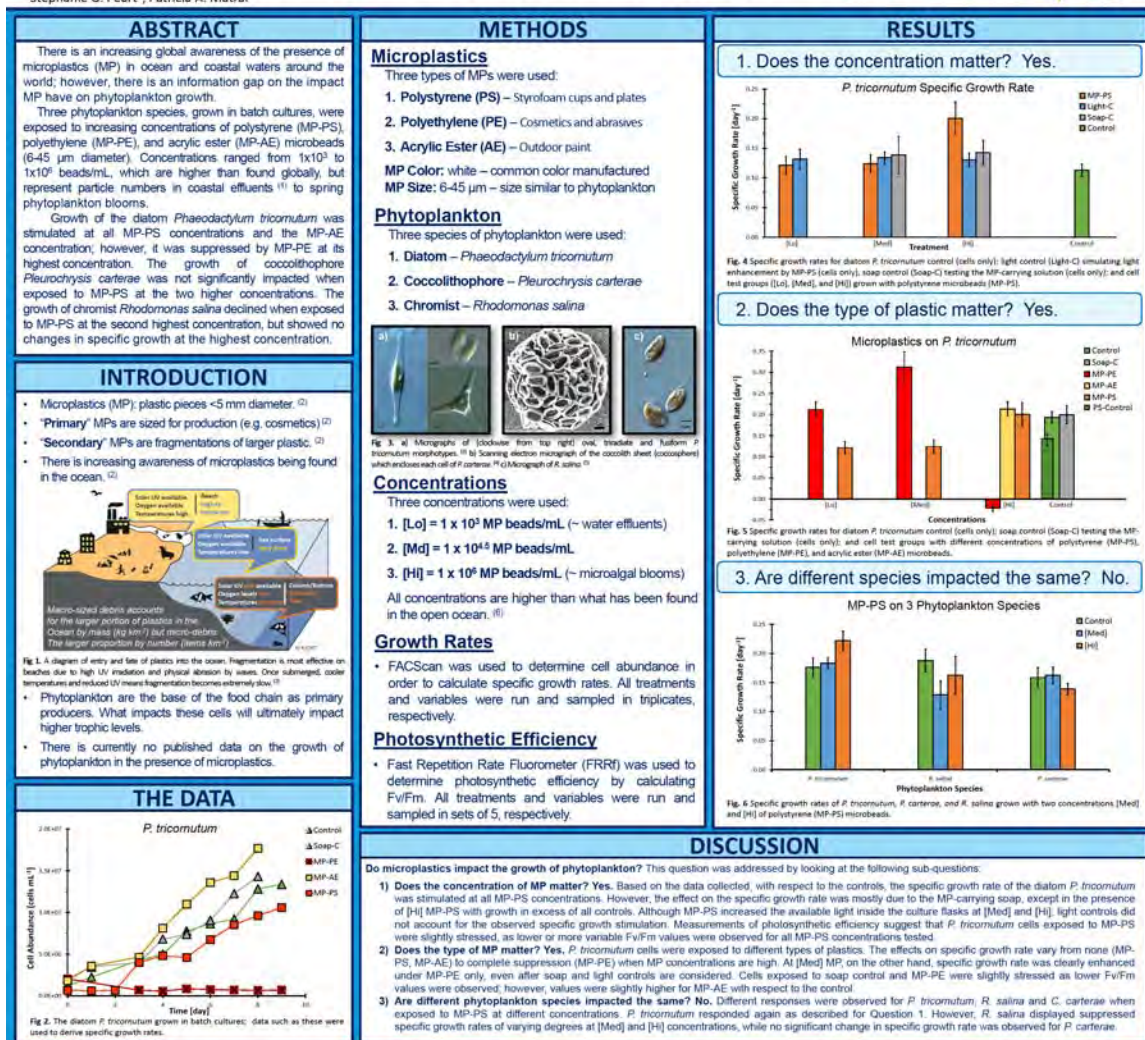
There is an increasing global awareness of the presence of microplastics (MP) in ocean and coastal waters around the world; however, there is an information gap on the impact MP have on phytoplankton growth. Three phytoplankton species, grown in batch cultures, were exposed to increasing concentrations of polystyrene (MP-PS), polyethylene (MP-PE), and acrylic ester (MP-AE) microbeads (6-45 μm diameter). Concentrations ranged from 1×10^3 to 1×10^6 beads/mL, which are higher than found globally, but represent particle numbers in coastal effluents¹ to spring phytoplankton blooms. Growth of the diatom *Phaeodactylum tricornutum* was stimulated at all MP-PS concentrations and at the MP-AE concentration; however, it was suppressed by MP-PE at its highest concentration. The growth of coccolithophore *Pleurochrysis carterae* was not significantly impacted when exposed to MP-PS at the two higher concentrations. The growth of chromist *Rhodomonas salina* declined when exposed to MP-PS at the second highest concentration, but showed no changes in specific growth at the highest concentration. These analyses provide insight on the variability of the impact on phytoplankton growth when exposed to different types and concentrations of MP.

Phytoplankton Growth in the Presence of Microplastics

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Bigelow

Laboratory for
Ocean Sciences



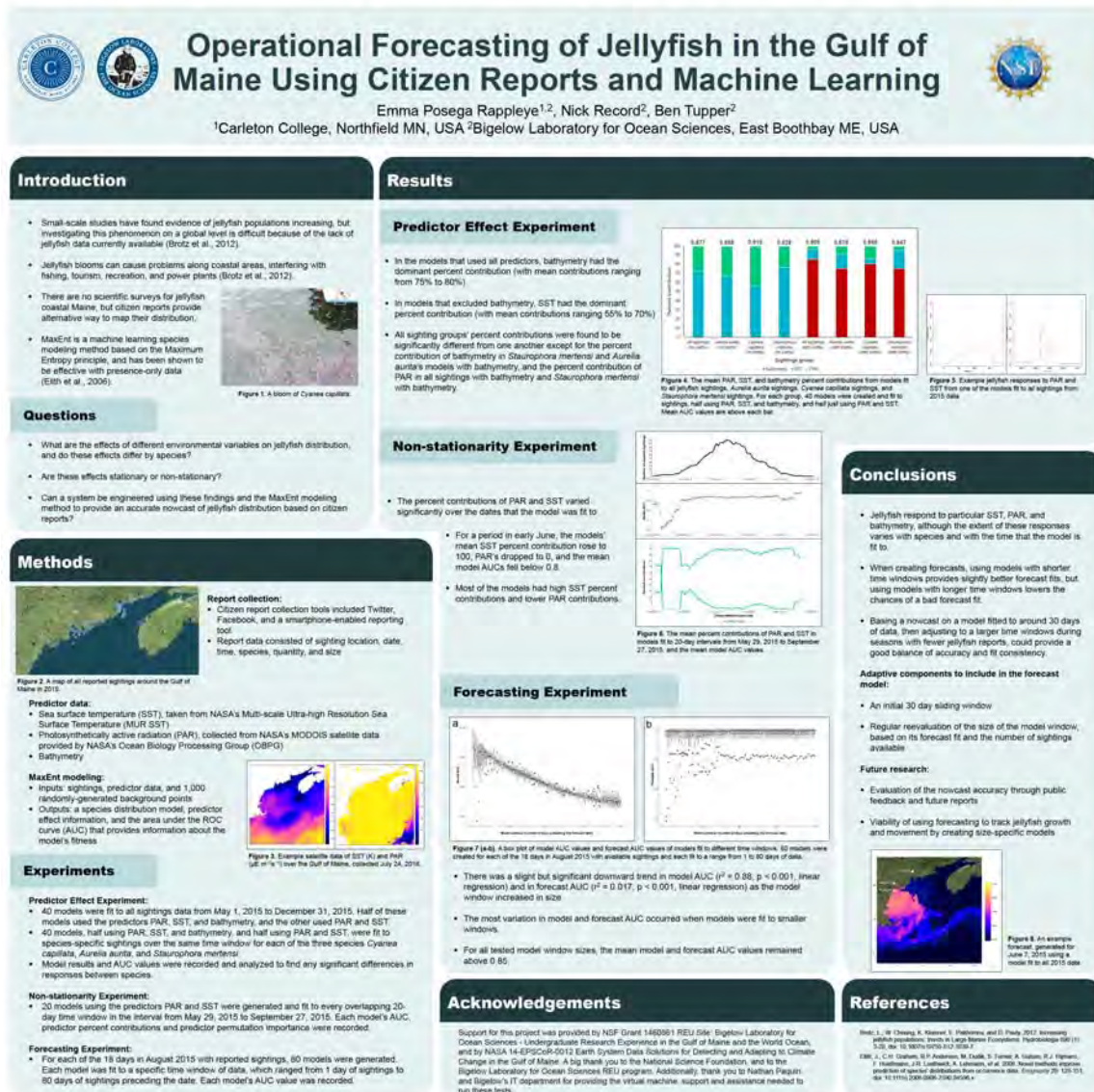
OPERATIONAL FORECASTING OF JELLYFISH IN THE GULF OF MAINE USING CITIZEN REPORTS

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As ocean conditions change, jellyfish blooms may be increasing; however, jellyfish data collection has historically been sparse and inconsistent, leading to difficulties investigating these global patterns. A better understanding of jellyfish distribution would aid in these investigations and would also help in addressing the problems that blooms can cause. While scientific surveys for jellyfish are not available in coastal Maine, citizen reports have emerged as an alternative way to map their distribution. In order to examine jellyfish responses to environmental predictors and explore the possibility of forecasting future blooms, these reports were used in conjunction with available satellite measurements to create distribution models and forecasts using MaxEnt. In these models, jellyfish appeared to be associated with increased sea surface temperature and photosynthetically active radiation, although the extent of their responses varied significantly by time window and species. Models tuned to smaller windows of time had good forecasting performances and the best fits, suggesting that models could be created using a small, recent window of citizen reports to provide an accurate jellyfish nowcast of the Gulf of Maine for public use.

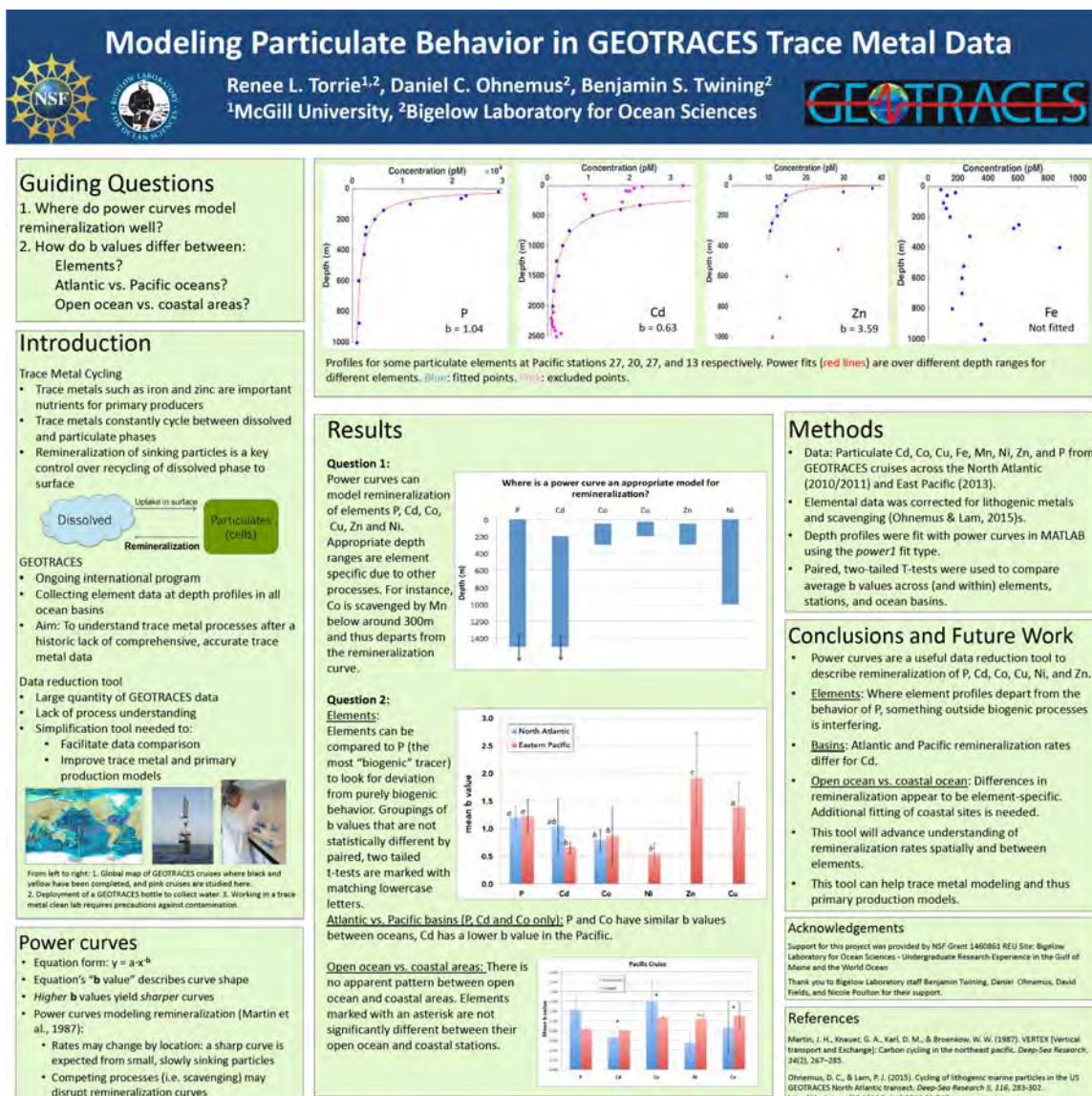


MODELING PARTICULATE BEHAVIOR IN GEOTRACES TRACE METAL DATA

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The collection of high-resolution, basin-scale trace metal data by the international GEOTRACES program has generated hundreds of depth profiles, allowing for unprecedented investigations of the vertical processes affecting trace metal cycling. A key vertical process is remineralization—the solubilization of biogenic particulate metals with depth. In this study, the remineralization of 8 elements was modeled with power curve fits to their depth profiles, using data from US GEOTRACES cruises across the North Atlantic and Eastern Tropical Pacific. After correction for lithogenic and scavenged phases, fits show that power curves can model the remineralization of P, Cd, Co, Cu, Zn, and Ni. Exponents (b values) in these power fits represent remineralization rates, with higher values indicating more rapid remineralization. These b values are compared across elements, basins, and regions. For instance, remineralization rates of Cd match those of P in the Atlantic [1.05 ± 0.48], but appear significantly slower in the Pacific [0.66 ± 0.11]. Power fits to depth profiles may prove useful in vertical modeling of trace metals and as a data reduction tool to study element-specific remineralization behaviors.





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