Bigelow | Laboratory for Ocean Sciences

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

REU Symposium Program & Abstracts
Thursday, August 1, 2019

Oral Program

12:30 Opening Comments

12:40 Carlin Schildge – Colby College, Waterville, ME

An Assessment of Green Sea Urchin Settlement in the Gulf of Maine.

Carlin Schildge[1]; Douglas B. Rasher[2]
Colby College[1], Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[2]

12:50 Emmaeve Jourdain – University of Maine, Orono, ME

Effects of warming seawater temperatures on economically valuable brown macroalgae phenology

Emmaeve Jourdain[1,2]., Nichole N. Price[2], Brittney Honisch[3]
[1]University of Maine Orono, [2]Bigelow Laboratory for Ocean Sciences, East Boothbay, ME,

1:00 Rémi Massé – University of Michigan, Ann Arbor, MI

The Iron Blanket: Microbial Iron Reduction in Alaskan Permafrost Sediments

Massé R [1,2], Michaud AB[1], Emerson D[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], University of Michigan, Ann Arbor, MI[2]

1:10 Matt Sabin – Colby College, Waterville ME

Evaluation of qPCR as a Method to Detect and Quantify Microcystin Concentrations in Maine Lakes

Matthew Sabin[1], Peter Countway[2]
Colby College[1], Bigelow Laboratory for Ocean Sciences[2]

1:20 Kayla Clark – University of Mississippi, University, MS

Horizontal Gene Transfer of Heme-copper Oxidases in Microbial Communities

Clark KN[1,2], Beam J[1], Bezuidt O[1] , Stepanauskas R[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME [1] , University of Mississippi [2]

1:30 Katherine Squires - Colby College, Waterville, ME

An analysis of metal oxide cycling in continental margin sediments: The relationship between Cr abundance and metal oxide cycling

Squires KR[1,2], Rauschenberg S[1], McManus J[1]
Bigelow Laboratory for Ocean Sciences[1], Colby College[2]

1:40 Katelyn Smith – Maine Maritime Academy, Castine, ME

Identifying Redox Potentials used by Microbes within Crustal Fluid of the Juan de Fuca Ridge Flank

Smith KA[1,2], Booker A[2], Orcutt BN[2]
Maine Maritime Academy[1], Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[2]

1:50 Grace Andrews – Colby College, Waterville, Maine

Effects of maternal size on larval fitness of the American lobster (*Homarus americanus*) under starvation conditions

Grace Andrews[1,2], Maura Niemisto[1], Alex Ascher[1,3], Donaven Baughman[1,4], Richard Wahle[3], David M. Fields[1]
[1]Bigelow Laboratory for Ocean Sciences, [2]Colby College, [3]University of Maine School of Marine Sciences Darling Marine Center, [4]Wichita State University

2:00 Donaven Baughman– Wichita State University, Wichita, KS

Maternal size effect on larval growth, development, and respiration rates in the American lobster (Homarus americanus)

Donaven Baughman[1,2], Grace Andrews[2,3], Maura Niemisto[2], Alex Ascher[2,4], Rick Wahle[4], David Fields[2]
1. Wichita State University, KS 2. Bigelow Laboratory for Ocean Sciences 3. Colby College, ME 4. University of Maine, School of Marine Sciences, Darling Marine Center

2:10 José J. Orozco Juarbe – Universidad de Puerto Rico – Mayagüez, Mayagüez, PR

Gut Content Analysis of the American Lobster (*Homarus americanus*) Larvae in the Gulf of Maine.

Orozco JJ[1], Countway P [2], Fields DM[2]
Universidad de Puerto Rico Rico – Mayagüez, Mayagüez, PR[1], Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[2].

2:20 Theresa Hong – Pennsylvania State University, University Park, PA

Effects of Microplastic Fibers on American Lobster Larvae (*Homarus americanus)*

Hong TJ[1,2], Woods MN[1], Fields DM[1], Matrai PA[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], Pennsylvania State University, University Park, PA[2]

2:30-2:40 ****** Break (10 min) **********

2:40 Catherine Wilhelm – The College of William and Mary, Williamsburg, VA

Potential Shell Hash Mitigation of Coastal Acidification for Juvenile Oysters Reared in Upwellers

Catherine Wilhelm [1,2,3], Nichole Price [2], Meredith White [3], Tessa Houston [3,4], Rich Smith [3], Brittney Honsich [2], Curtis Bohlen[5]
[1]The College of William and Mary, [2]Bigelow Laboratory for Ocean Sciences, [3]Mook Sea Farm, [4]Colby College, [5]Casco Bay Estuary Partnership

2:50 Adrienne N. Tracy – Colby College, Waterville, ME

Development and validation of gene delivery methods for *Crassostrea virginica*

Tracy, A. N. [1,2], Yadavalli, R.[1], Fernández Robledo, J. A.[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], Colby College, ME[2]

3:00 Hannah Waugh, Southern Maine Community College

Evaluating PmMOE Expression in *Perkinsus marinus* using Different Gene Delivery Methods

Waugh HA[1,2], Yadavalli R[1], Fernández Robledo JA[1]
Bigelow Laboratory for Ocean Sciences[1], East Boothbay, ME, Southern Maine Community College[2] South Portland, ME

3:10 Cristina Tusei – Humboldt State University, Arcata, CA

Photodegradation of Chlorinated Organic Compounds in Surface Waters

Tusei C[1,2], DiMento B[1], Aeppli C[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], Humboldt State University[2]

3:20 Anna Zeleny – College of Saint Benedict, Saint Joseph, MN

How Biodegradable is Weathered Oil?

Zeleny A[1,2],Beirne E[1], Keyes P[1], and Aeppli C[1]
[1]Bigelow Laboratory for Ocean Sciences, ME [2]College of Saint Benedict, MN

3:30 Lucy Quirk – University of South Carolina, Columbia, SC

Methane in Maine: A Localized Study of Tidal Influence on Air-Sea Methane Flux in the Gulf of Maine

Quirk, LE[1,2], Posman, KM[1], Archer, SD[1]
Bigelow Laboratory for Ocean Sciences[1], University of South Carolina[2]

3:40 Zhouxin (Trix) Li - Wheaton College, Norton, MA

Ocean Artificial Upwelling Effects on Dimethyl Sulfide (DMS) and Dimethylsulphoniopropionate (DMSP) Production

Zhouxin (Trix) Li[1,2], Kevin M. Posman[2], Stephen D. Archer[2]
[1]Wheaton College, Norton , MA,USA ; [2]Bigelow Laboratory for Ocean Sciences, East Boothbay, ME, USA

3:50-4:00 ****** Break (10 min) **********

4:00 Gabriella Kim - Colby College, Waterville, ME

An Investigation on the Decoupling of Dissolved, Particulate, and Cellular Metal Ratios in the Ocean

Kim GH[1,2], Tagliabue A[3] , Twining BS[1]
Bigelow Laboratory for Ocean Sciences[1], Colby College[2], University of Liverpool[3]

4:10 Mariah Ricci- The College of Saint Scholastica, Duluth, MN

Phytoplankton Distribution in the Mediterranean Sea Based on Ocean Color Data

Mariah Ricci [1,2], Nicolas Mayot [1] , Nick Record[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], The College of Saint Scholastica, Duluth, MN[2]

4:20 Gabrielle Martinez – Rollins College, Winter Park, FL

Gulf of Maine Temperature Salinity Curves From the Early 1900s (Henry Bigelow) Compared to the Present (GNATS)

Martinez, G.A.[1], Balch, W.M.[2]
Rollins College[1], Bigelow Laboratory for Ocean Sciences[2]

4:30 Moriah Kunes – University of Rochester, Rochester, NY

A Novel Approach to Quantify the Grazing Rates of Microzooplankton

Moriah Kunes[1,2], Laura Lubelczyk[1], Kate McPhee[1,3], Kevin Posman[1], Nicole Poulton[1], Stephen Archer[1]
[1]Bigelow Laboratory for Ocean Sciences, [2]University of Rochester, [3]Scripps College

4:40 Catherine Mahoney- Maine Maritime Academy, Castine, ME

Cellular Response of *Emiliania huxleyi* to Growth on Phosphonates

Mahoney, C.R.[1,2] Whitney, L.P.[1,2] Lomas, M.W.[2]
Maine Maritime Academy, Castine, ME [1], Bigelow Laboratory for the Ocean Sciences, East Boothbay, ME. [2]

Abstracts and Posters

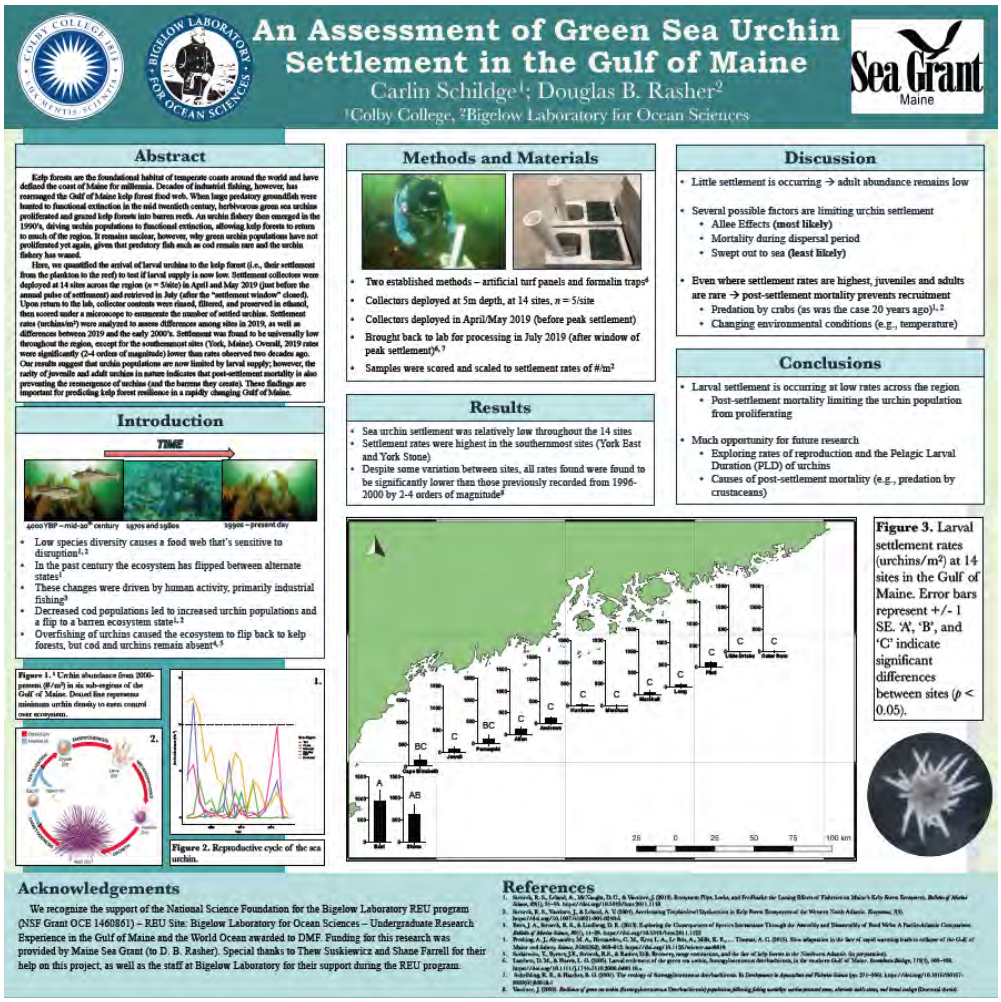
Carlin Schildge – Colby College, Waterville, ME

An Assessment of Green Sea Urchin Settlement in the Gulf of Maine.

Carlin Schildge[1]; Douglas B. Rasher[2]
Colby College[1], Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[2]

Kelp forests are the foundational habitat of temperate coasts around the world and have defined the coast of Maine for millennia. Decades of industrial fishing, however, has rearranged the Gulf of Maine kelp forest food web. When large predatory groundfish were hunted to functional extinction in the mid twentieth century, herbivorous green sea urchins proliferated and grazed kelp forests into barren reefs. An urchin fishery then emerged in the 1990's, driving urchin populations to functional extinction, allowing kelp forests to return to much of the region. It remains unclear, however, why green urchin populations have not proliferated yet again, given that predatory fish such as cod remain rare and the urchin fishery has waned.

Here, we quantified the arrival of larval urchins to the kelp forest (i.e., their settlement from the plankton to the reef) to test if larval supply is now low. Settlement collectors were deployed at 14 sites across the region (n = 5/site) in April and May 2019 (just before the annual pulse of settlement) and retrieved in July (after the "settlement window" closed). Upon return to the lab, collector contents were rinsed, filtered, and preserved in ethanol, then scored under a microscope to enumerate the number of settled urchins. Settlement rates (urchins/m^2) were analyzed to assess differences among sites in 2019, as well as differences between 2019 and the early 2000's. Settlement was found to be universally low throughout the region, except for the southernmost sites (York, Maine). Overall, 2019 rates were significantly (2-4 orders of magnitude) lower than rates observed two decades ago. Our results suggest that urchin populations are now limited by larval supply; however, the rarity of juvenile and adult urchins in nature indicates that post-settlement mortality is also preventing the reemergence of urchins (and the barrens they create). These findings are important for predicting kelp forest resilience in a rapidly changing Gulf of Maine.

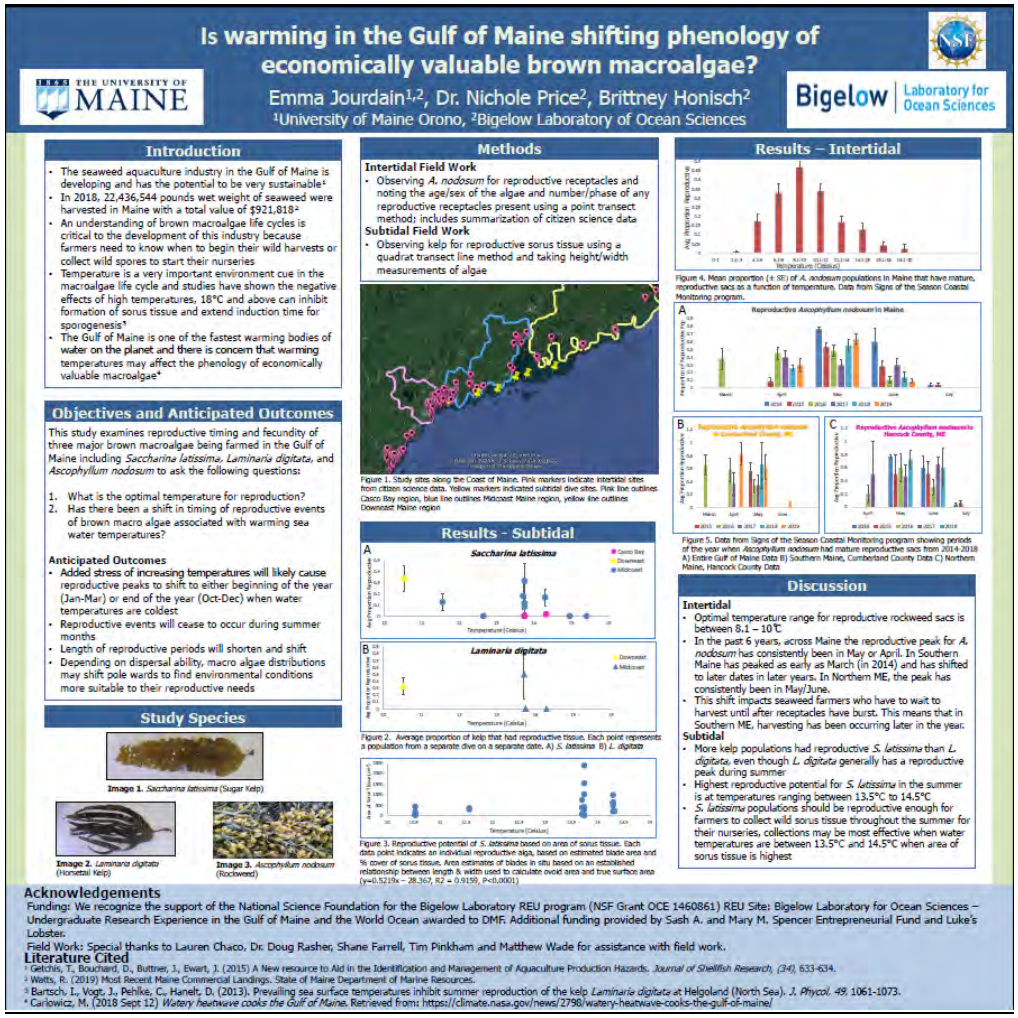


Emmaeve Jourdain – University of Maine, Orono, ME

Effects of warming seawater temperatures on economically valuable brown macroalgae phenology

Emmaeve Jourdain[1,2]., Nichole N. Price[2], Brittney Honisch[3]
[1]University of Maine Orono, [2]Bigelow Laboratory for Ocean Sciences, East Boothbay, ME,

An understanding of reproductive timing of brown macroalgae is crucial to the developing seaweed aquaculture industry in Maine so farmers can determine when wild harvest or nurseries can begin. However, there is concern because the Gulf of Maine is one of the fastest warming bodies of water in the world and temperature is an important environmental trigger for development of macroalgae reproductive tissues. In this study, we looked to see if there has been a shift in macroalgae algae phenology related to warming. Using the point-intercept method, we determined the proportion of the population of intertidal species, Ascophyllum nodosum, that had generated reproductive sacs. We also compiled data from the citizen science database, Signs of the Season Coastal Monitoring program that report the fecundity of A. nodosum since 2014. Also, we used a quadrat transect method on SCUBA to observe the proportion of the population that had developed sorus tissue for two subtidal kelp species, Saccharina latissima and Laminaria digitata. Our results show that the optimal temperature for reproductive A. nodosum is between 8.1 to 10℃, and in the past six years in Maine the month of reproductive peak for A. nodosum has been either April or May. There is evidence that reproductive peaks for A. nodosum are occurring later each year in Southern Maine. For the subtidal kelp, our results showed populations with the highest proportion of reproductive kelp were found at 13.75℃. We observed more populations with reproductive S. latissima than reproductive L. digitata, even though L. digitata generally reproduces more during summer while S. latissima reproduces more during winter. Knowing optimal temperatures associated with reproductive peaks for macroalgae is helpful for predicting how phenology may shift in the future. While no major shifts in phenology were observed for macroalgae, there is still concern for the seaweed aquaculture industry as the Gulf of Maine continues to warm.

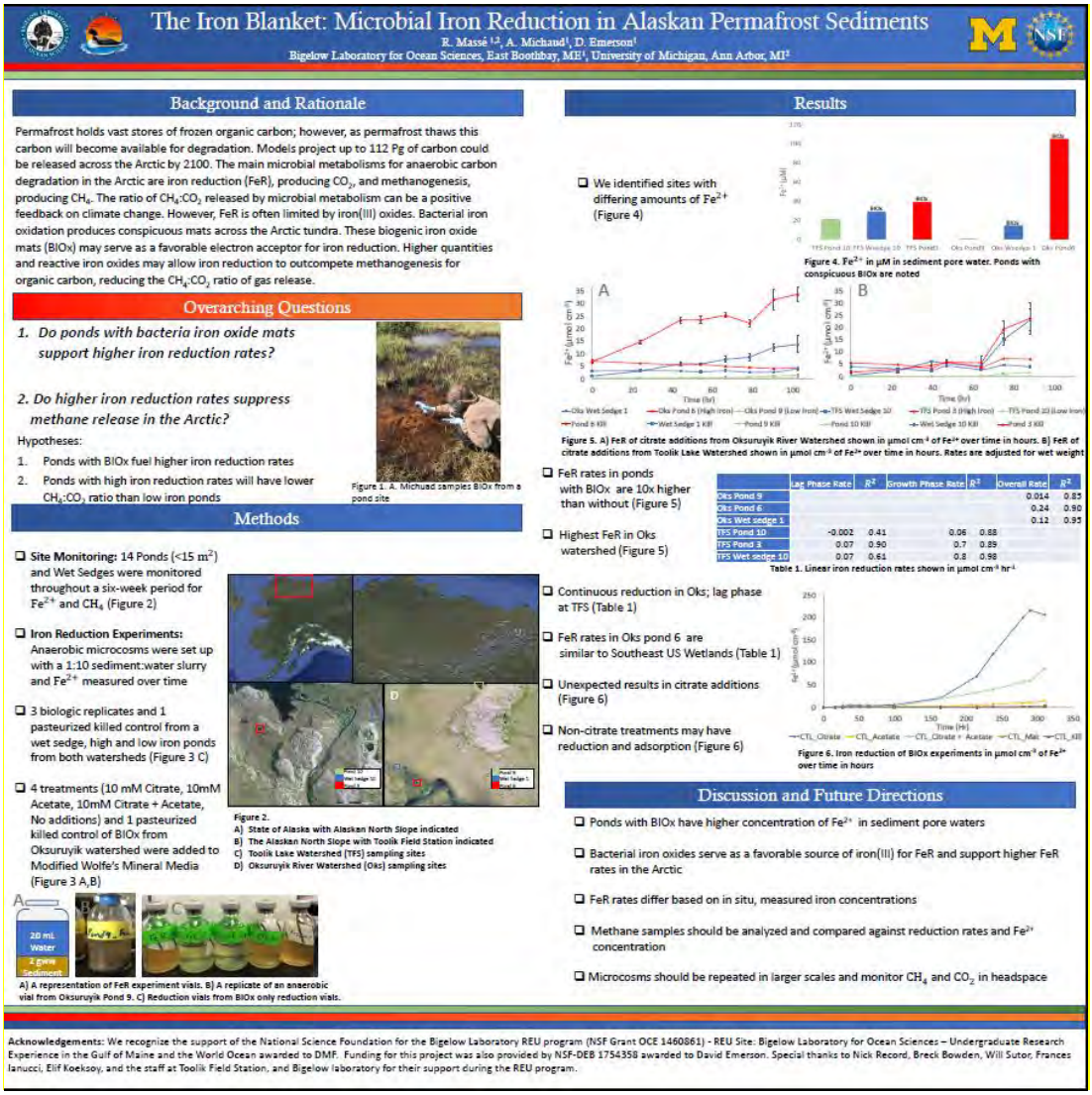


Rémi Massé – University of Michigan, Ann Arbor, MI

The Iron Blanket: Microbial Iron Reduction in Alaskan Permafrost Sediments

Massé R [1,2], Michaud AB[1], Emerson D[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], University of Michigan, Ann Arbor, MI[2]

Permafrost holds vast stores of organic carbon, much of which is currently unavailable to microbial processes; however, as permafrost thaws this carbon will become available for degradation. Some models project as much as 112Pg of carbon could be released across the Arctic by 2100, with several percent in the form of methane. How this permafrost carbon will be cycled and released by microbial metabolism, particularly through iron cycling and anaerobic production of methane, is important to understand and quantify given the impacts of positive feedback on climate change. The potential for microbial iron cycling through oxidation and reduction in arctic freshwater to suppress methane release is an important parameter for understanding the fate of arctic permafrost. Iron reduction potentials were quantified in pond and wet sedge environments using anoxic growth experiments with native water and sediment. Rates as high as $0.2383\ \mu molcm^{-3}hr^{-1}\ Fe^{2+}$ and $0.2536\ \mu molcm^{-3}hr^{-1}\ Fe^{2+}$ were measured at the Oksuruyik River and Toolik Lake watersheds, respectively. Iron reduction rates in the Arctic vary significantly with presence of bacterial iron oxides in sediment. This study finds thar bacterial iron oxide mats (BIOx) serve as a favorable electron donor for Iron Reduction and support higher rates of reduction.

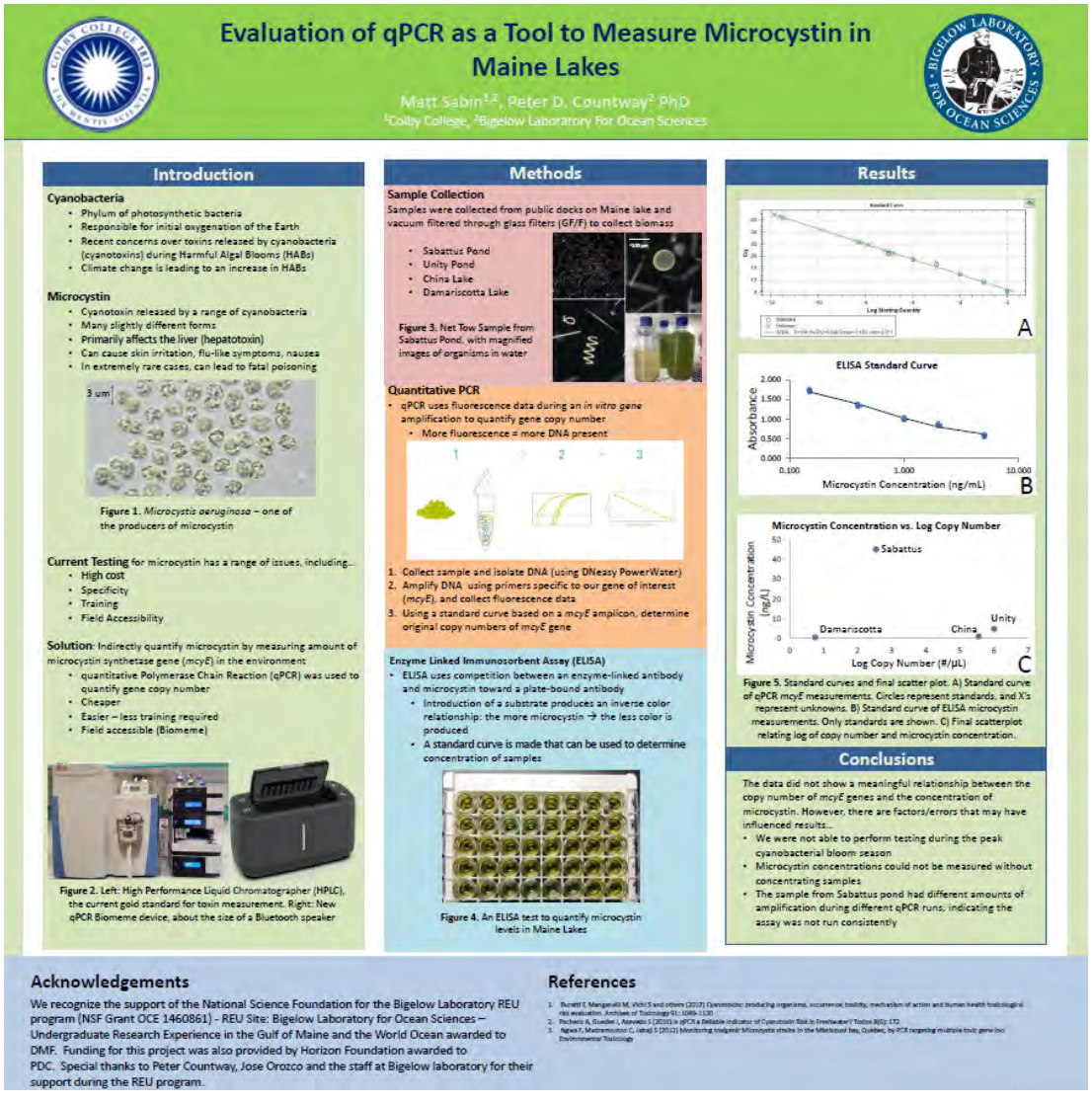


Matt Sabin – Colby College, Waterville ME

Evaluation of qPCR as a Method to Detect and Quantify Microcystin Concentrations in Maine Lakes

Matthew Sabin[1], Peter Countway[2]
Colby College[1], Bigelow Laboratory for Ocean Sciences[2]

Cyanobacteria play an important role in oxygenating the Earth. Recently, climate change and nutrient runoff from various human sources have led to an increase in harmful algal blooms (HABs). HABs are cyanobacterial in nature, and owe their to harmful nature to cyanotoxins. Cyanotoxins are toxins produced by cyanobacteria. One such toxin is microcystin. Microcystin is a hepatotoxin, meaning it harms the liver. Exposure to microcystin can lead to rashes and flulike symptoms. Fatal doses are extremely rare but have happened in Brazil. Current testing for microcystin is often less than ideal. It can be expensive, unable to detect multiple congeners of microcystin, and a number of other issues. A possible alternative method to measure microcystins would be using qPCR to measure the copy number of the microcystin synthetase gene. The *mcyE* gene was chosen as the target gene. Primers were taken from previous literature. Samples were collected from Sabattus Pond, Unity Pond, China Lake, and the Damariscotta Lake. An accepted microcystin concentration for each sample was found using ELISA testing. Although both qPCR and ELISA testing were performed successfully on all paired samples, there was no relationship found between the gene copy number of *mcyE* and the microcystin concentration.

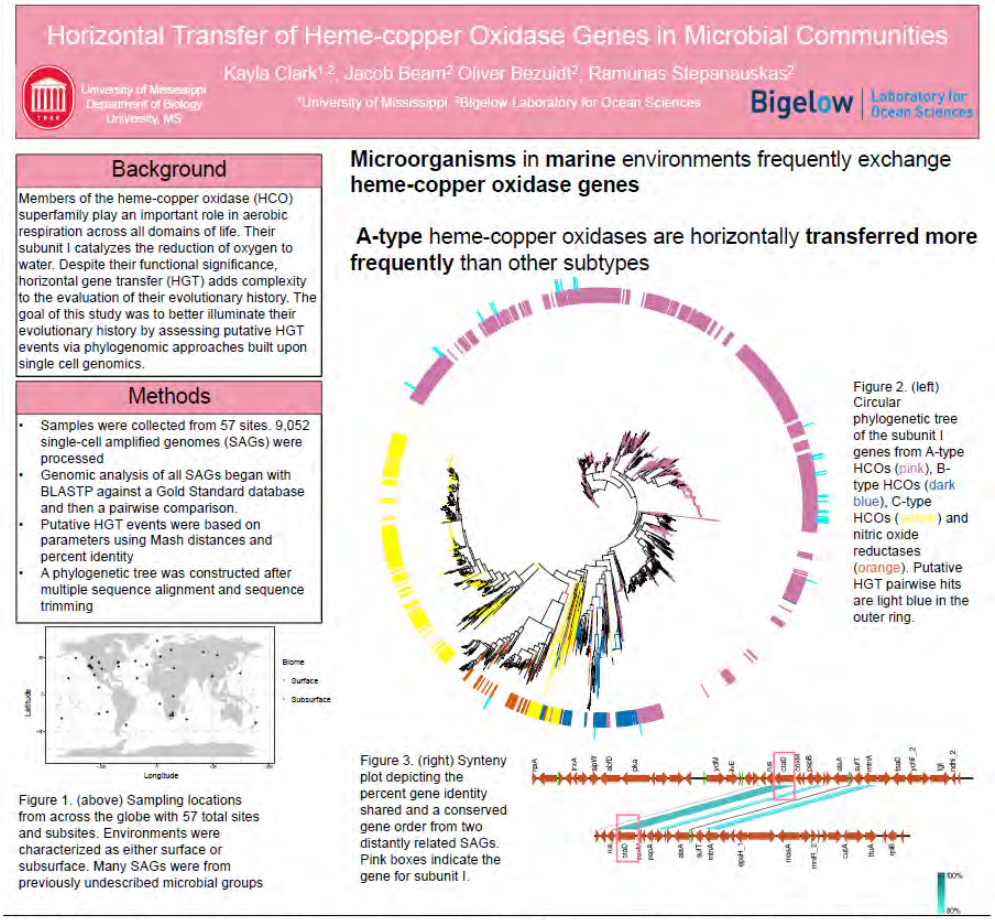


Kayla Clark – University of Mississippi, University, MS

Horizontal Gene Transfer of Heme-copper Oxidases in Microbial Communities

Clark KN[1,2], Beam J[1], Bezuidt O[1] , Stepanauskas R[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME [1] , University of Mississippi [2]

Members of the heme-copper oxidase (HCO) super-family make up the last enzyme complex in the aerobic respiratory chain, undoubtedly playing a large role in an important cellular process in all domains of life. Despite the functional significance of HCOs, horizontal gene transfer (HGT) within microbial dark matter adds complexity to the evaluation of their evolutionary history. Recent evidence suggests that microbial HCO genes were horizontally acquired on early Earth with only trace amounts of abiotic oxygen. Using a genomics approach, we search for putative horizontal gene transfer events to illuminate the evolutionary origin of HCO genes among disparate phyla of bacteria and archaea. A total of 9,052 single-cell amplified genomes (SAGs) from 57 sites were compared via BLASTP to a Gold Standard HCO database. Many of these genomes were from previously undescribed microbial groups - microbial dark matter. High sequence similarity of the subunit I genes across distantly related phylogenetic groups were suggestive of horizontal gene transfer (HGT). Sites from the subsurface with low oxygen content were considered to be analogous to an early Earth and thought to contain higher gene transfer frequencies. We show that putative HGT events are most common in A-type HCOs as well as in marine environments. Each of the four subtypes analyzed are part of a clear monophyletic group indicating they are evolutionarily distinct. SAGs selected for analysis of synteny showed that multiple subunits were most likely acquired together as an operon. These findings suggest that the horizontally acquired HCO genes have not yet ameliorated. The results build a more concise path towards understanding the evolution of heme-copper oxidases and aerobic life.



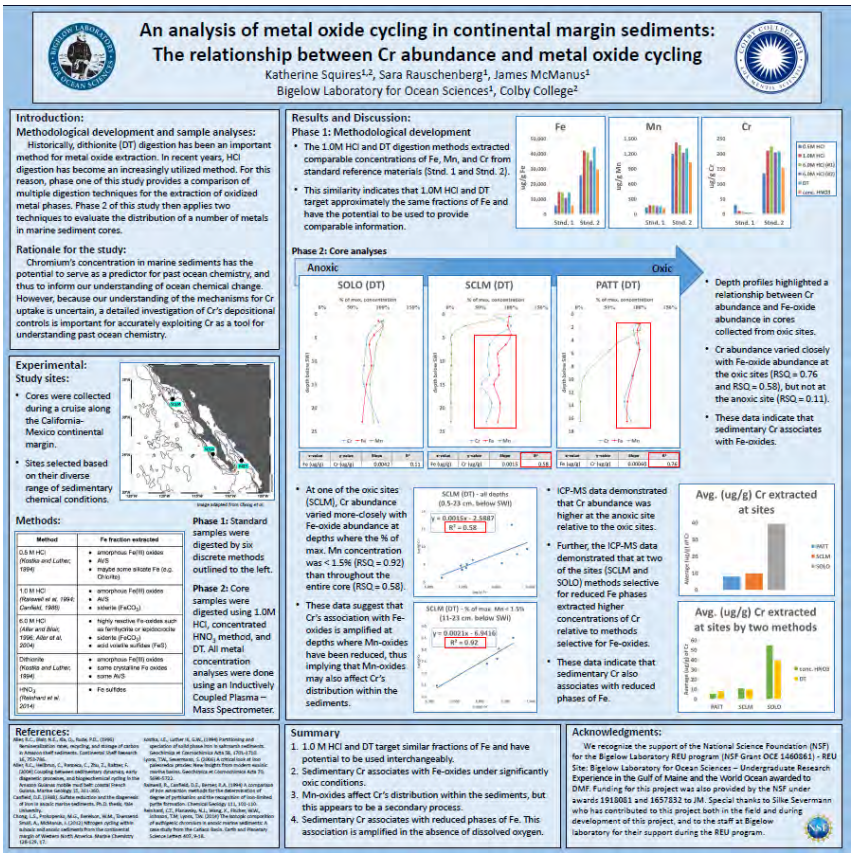
Acknowledgements: We recognize the support of the National Science Foundation for the Bigelow Laboratory REU program (NSF Grant OCE 1460861) - REU Site: Bigelow Laboratory for Ocean Sciences – Undergraduate Research Experience in the Gulf of Maine and the World Ocean awarded to DMF. Special thanks to the staff at Bigelow laboratory for their support during the REU program.

Katherine Squires - Colby College, Waterville, ME

An analysis of metal oxide cycling in continental margin sediments: The relationship between Cr abundance and metal oxide cycling

Squires KR[1,2], Rauschenberg S[1], McManus J[1]
Bigelow Laboratory for Ocean Sciences[1], Colby College[2]

Chromium's concentration in marine sediments has the potential to serve as a predictor for past ocean chemistry. However, because our understanding of the mechanisms for Cr uptake are uncertain, a detailed investigation of Cr's depositional controls is essential for accurately exploiting this potential tool. The central hypothesis of this research is that Cr will exhibit predictable behavior tied to metal oxide cycling. Historically, dithionite (DT) digestion has been an important method for metal oxide extraction. In recent years, HCl digestion has become an increasingly utilized method. For this reason, phase one of this project aimed to compare a number of HCl digestion methods to the DT digestion method. A number or common reference materials and laboratory standards were digested by six discrete methods: a 0.5 M HCl digestion, a 1.0 M HCl digestion, two different 6.0 M HCl digestions, a concentrated HNO_3 digestion, and a hot DT digestion. Fe, Mn, Cr, Cu, Mo, and Zn analyses were done using an Inductively Coupled Plasma – Mass Spectrometer (ICP-MS). The 1.0 M HCl and DT digestion methods extracted comparable concentrations of Fe, Mn, and Cr from standard reference materials. Phase two of this project applied the DT and nitric acid digestion methods to sediment cores that had been collected during a cruise along the California-Mexico continental margin. Core sites were selected based on their diverse range of sedimentary chemical conditions. Depth profiles highlighted a relationship between Cr abundance and Fe-oxide abundance in cores collected from oxic sites. Results confirmed that Cr abundance varied closely with Fe-oxide abundance at the oxic sites, but not at the anoxic site. This data indicates that sedimentary Cr associates with Fe-oxides under oxic conditions. Some data indicate that this association is amplified at depths where Mn-oxides have been reduced, thus implying that Mn-oxides may influence Cr's association with Fe-oxides; however, further work is necessary to support this conclusion. ICP-MS data demonstrated that Cr abundance was higher at the anoxic site relative to the oxic sites. Additional data demonstrated that methods selective for reduced Fe phases extracted higher concentrations of Cr relative to methods selective for Fe-oxides at two of the sites. These data indicate that in addition to its association with Fe-oxides, sedimentary Cr may be associated with reduced phases of Fe.

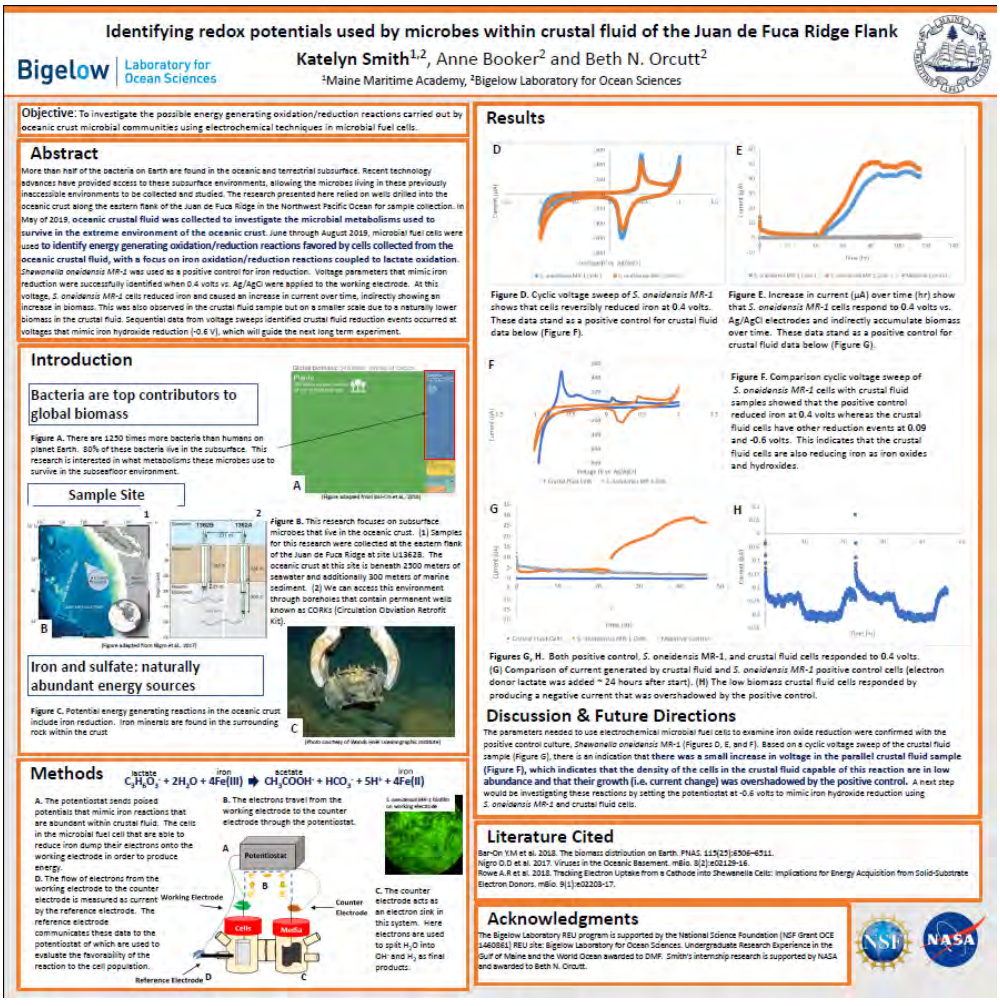


Katelyn Smith – Maine Maritime Academy, Castine, ME

Identifying Redox Potentials used by Microbes within Crustal Fluid of the Juan de Fuca Ridge Flank

Smith KA[1,2], Booker A[2], Orcutt BN[2]
Maine Maritime Academy[1], Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[2]

More than half of the bacteria on Earth are found in the oceanic and terrestrial subsurface. Recent technology advances have provided access to these subsurface environments, allowing the microbes living in these previously inaccessible environments to be collected and studied. The research presented here relied on wells drilled into the oceanic crust along the eastern flank of the Juan de Fuca Ridge in the Northwest Pacific Ocean for sample collection. In May of 2019, oceanic crustal fluid was collected to investigate the microbial metabolisms used to survive in the extreme environment of the oceanic crust. June through August 2019, microbial fuel cells were used to identify energy generating oxidation/reduction reactions favored by cells collected from the oceanic crustal fluid, with a focus on iron oxidation/reduction reactions coupled to lactate oxidation. *Shewanella oneidensis MR-1* was used as a positive control for iron reduction. Voltage parameters that mimic iron reduction were successfully identified when 0.4 volts vs. Ag/AgCl were applied to the working electrode. At this voltage, *S. oneidensis MR-1* cells reduced iron and caused an increase in current over time, indirectly showing an increase in biomass. This was also observed in the crustal fluid sample but on a smaller scale due to a naturally lower biomass in the crustal fluid. Sequential data from voltage sweeps identified crustal fluid reduction events occurred at voltages that mimic iron hydroxide reduction (-0.6 V), which will guide the next long term experiment.



Introduction

Bacteria are top contributors to global biomass

Figure A. There are 1250 times more bacteria than humans on planet Earth. 80% of these bacteria live in the subsurface. This research is interested in what metabolisms these microbes use to survive in the subseafloor environment.

Sample Site

Figure B. This research focuses on subsurface microbes that live in the oceanic crust. (1) Samples for this research were collected at the eastern flank of the Juan de Fuca Ridge at site U1362B. The oceanic crust at this site is beneath 2300 meters of seawater and additionally 300 meters of marine sediment. (2) We can access this environment through boreholes that contain permanent seals known as CORKs (Circulation Obviation Retrofit Kit).

Iron and sulfate: naturally abundant energy sources

Figure C. Potential energy generating reactions in the oceanic crust include iron reduction. Iron minerals are found in the surrounding rock within the crust

Methods

$$\underset{\text{lactate}}{C_3H_5O_3^-} + 2H_2O + 4\underset{\text{iron}}{Fe(III)} \rightarrow \underset{\text{acetate}}{CH_3COO^-} + HCO_3^- + 5H^+ + 4\underset{\text{iron}}{Fe(II)}$$

A. The potentiostat sends poised potentials that mimic iron reactions that are abundant within crustal fluid. The cells in the microbial fuel cell that are able to reduce iron dump their electrons onto the working electrode in order to produce energy.

B. The electrons travel from the working electrode to the counter electrode through the potentiostat.

C. The counter electrode acts as an electron sink in this system. Here electrons are used to split H_2O into OH^- and H_2 as final products.

D. The flow of electrons from the working electrode to the counter electrode is measured as current by the reference electrode. The reference electrode communicates these data to the potentiostat of which are used to evaluate the favorability of the reaction to the cell population.

Results

Figure D. Cyclic voltage sweep of *S. oneidensis MR-1* shows that cells reversibly reduced iron at 0.4 volts. These data stand as a positive control for crustal fluid data below (Figure F).

Figure E. Increase in current (μA) over time (hr) show that *S. oneidensis MR-1* cells respond to 0.4 volts vs. Ag/AgCl electrodes and indirectly accumulate biomass over time. These data stand as a positive control for crustal fluid data below (Figure G).

Figure F. Comparison cyclic voltage sweep of *S. oneidensis MR-1* cells with crustal fluid samples showed that the positive control reduced iron at 0.4 volts whereas the crustal fluid cells have other reduction events at 0.09 and -0.6 volts. This indicates that the crustal fluid cells are also reducing iron as iron oxides and hydroxides.

Figures G, H. Both positive control, *S. oneidensis* MR-1, and crustal fluid cells responded to 0.4 volts. (G) Comparison of current generated by crustal fluid and *S. oneidensis MR-1* positive control cells (electron donor lactate was added ~ 24 hours after start). (H) The low biomass crustal fluid cells responded by producing a negative current that was overshadowed by the positive control.

Discussion & Future Directions

The parameters needed to use electrochemical microbial fuel cells to examine iron oxide reduction were confirmed with the positive control culture, *Shewanella oneidensis* MR-1 (Figures D, E, and F). Based on a cyclic voltage sweep of the crustal fluid sample (Figure G), there is an indication that **there was a small increase in voltage in the parallel crustal fluid sample (Figure F), which indicates that the density of the cells in the crustal fluid capable of this reaction are in low abundance and that their growth (i.e. current change) was overshadowed by the positive control.** A next step would be investigating these reactions by setting the potentiostat at -0.6 volts to mimic iron hydroxide reduction using *S. oneidensis MR-1* and crustal fluid cells.

Literature Cited

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Nigro O.D et al. 2017. Viruses in the Oceanic Basement. mBio. 8(2):e02129-16.
Rowe A.R et al. 2018. Tracking Electron Uptake from a Cathode into Shewanella Cells: Implications for Energy Acquisition from Solid-Substrate Electron Donors. mBio. 9(1):e02203-17.

Acknowledgments

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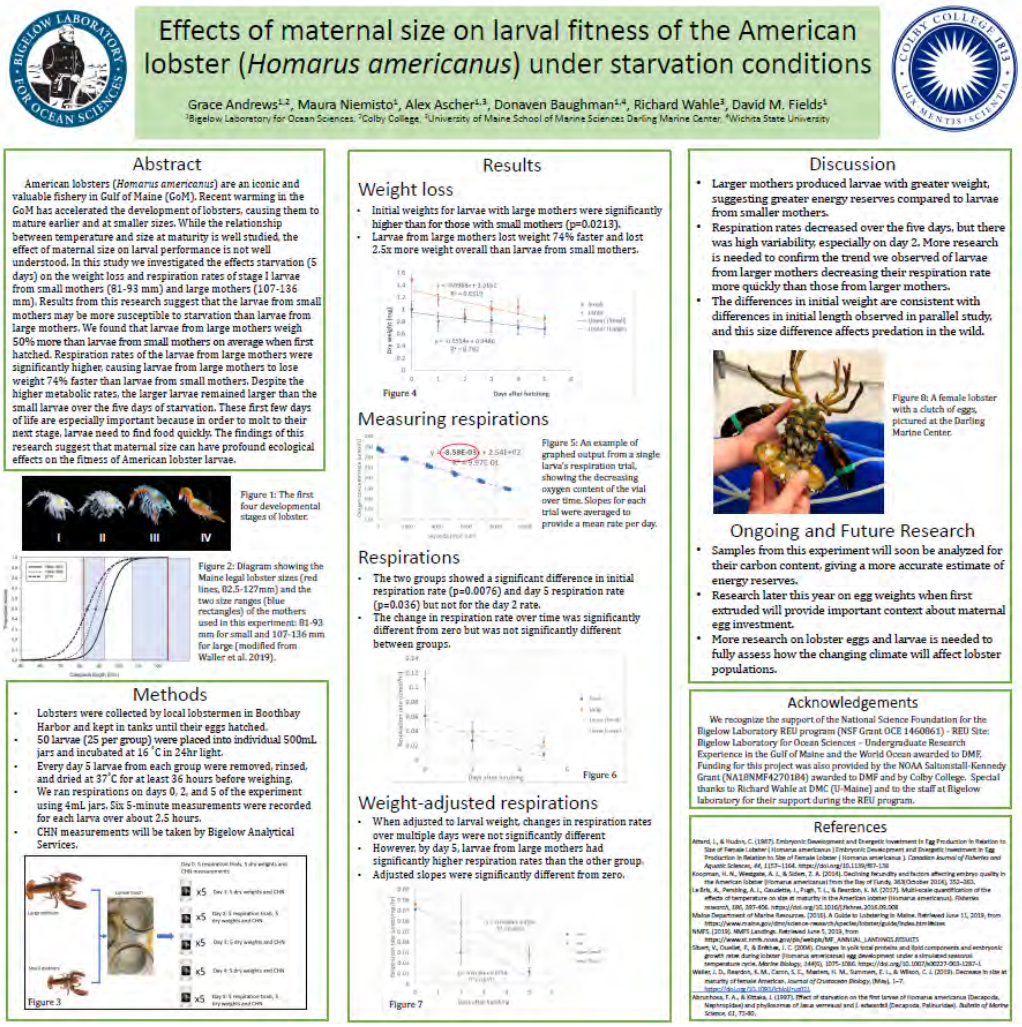
Grace Andrews – Colby College, Waterville, Maine

Effects of maternal size on larval fitness of the American lobster (*Homarus americanus*) under starvation conditions

Grace Andrews[1,2], Maura Niemisto[1], Alex Ascher[1,3], Donaven Baughman[1,4], Richard Wahle[3], David M. Fields[1]
[1]Bigelow Laboratory for Ocean Sciences, [2]Colby College, [3]University of Maine School of Marine Sciences Darling Marine Center, [4]Wichita State University

American lobsters (Homarus americanus) are an iconic and valuable fishery in Gulf of Maine (GoM). Recent warming in the GoM has accelerated the development of lobsters causing them to mature earlier and at smaller sizes. While the relationship between temperature and size at maturity is well studied, the effect of maternal size on larval performance is not well understood. In this study we investigated the effects starvation (5 days) on the weight loss and respiration rates of stage I larvae from small mothers (81-93 mm) and large mothers (107-136 mm). Results from this research suggest that the larvae from small mothers may be more susceptible to starvation than larvae from large mothers. We found that larvae from large mothers weigh 50% more than larvae from small mothers on average when first hatched. Respiration rates of the larvae from large mothers was significantly higher, causing larvae from large mothers to lose weight 74% faster than larvae from small mothers. Despite the higher metabolic rates, the larger larvae remained larger than the small larvae over the five days of starvation. These first few days of life are especially important because in order to molt to their next stage, larvae need to find food quickly. The findings of this research suggest that maternal size can have profound ecological effects on the fitness of American lobster larvae.

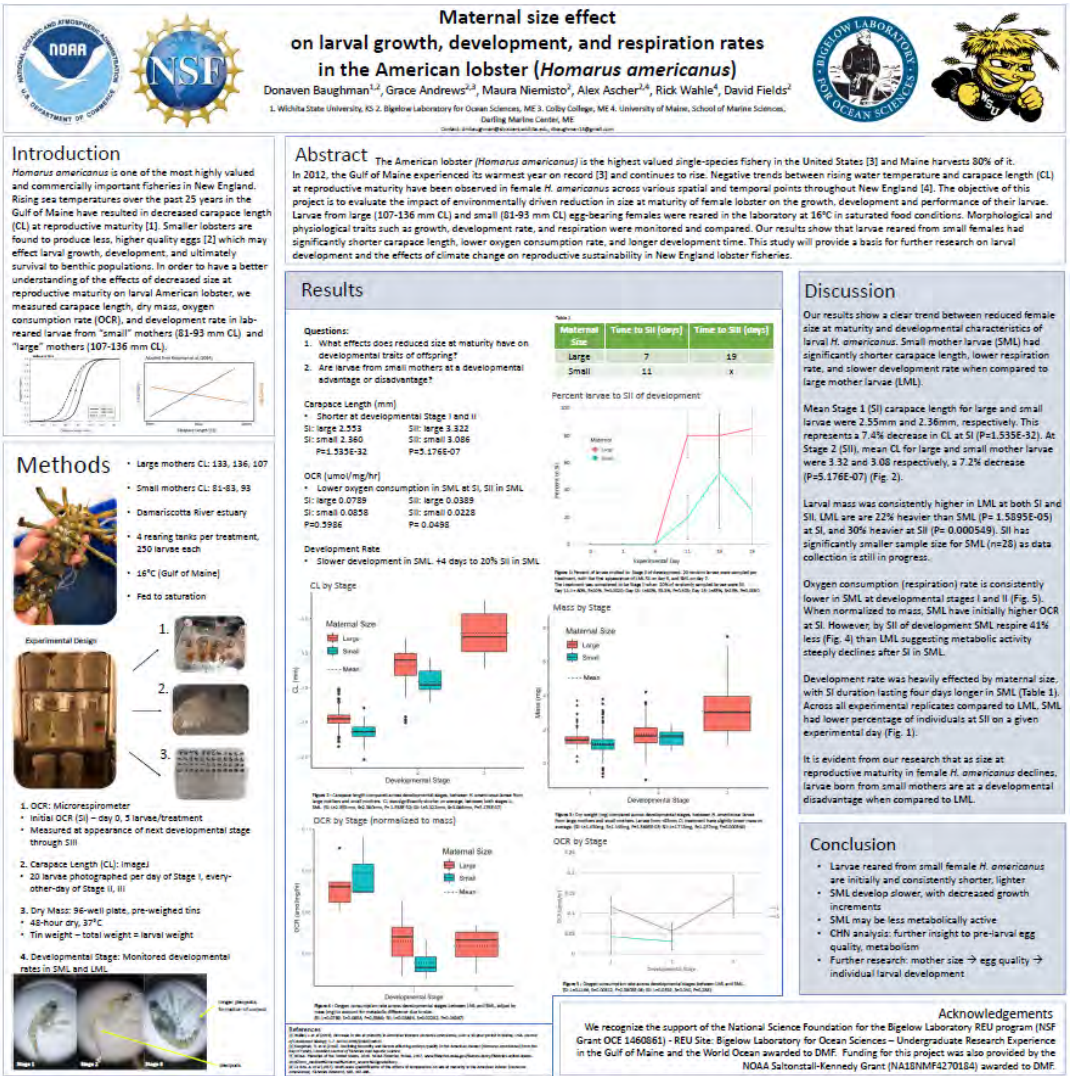


Donaven Baughman– Wichita State University, Wichita, KS

Maternal size effect on larval growth, development, and respiration rates in the American lobster (Homarus americanus)

Donaven Baughman[1,2], Grace Andrews[2,3], Maura Niemisto[2], Alex Ascher[2,4], Rick Wahle[4], David Fields[2]
1. Wichita State University, KS 2. Bigelow Laboratory for Ocean Sciences 3. Colby College, ME 4. University of Maine, School of Marine Sciences, Darling Marine Center

The American lobster *(Homarus americanus)* fishery is the most valuable single-species fishery in the United States, and Maine harvests 80% of it (NOAA 2017). In 2012, the Gulf of Maine experienced its warmest year on record and has broken that record 4 of the last 6 years since (NOAA, 2016). Negative trends between rising water temperature and carapace length (CL) at reproductive maturity have been observed in *H. americanus* across various spatial and temporal points throughout New England [Waller, LeBris]. The objective of this project is to evaluate the impact of environmentally driven reduction in size at maturity of female lobster on the growth, development and performance of their larvae. Larvae from large (107-136 mm CL) and small (81-93 mm CL) egg-bearing females were reared in the laboratory at 16°C in saturated food conditions. Morphological and physiological traits such as growth, development, and respiration were monitored and compared. Our results show that larvae reared from females <93mm CL had significantly shorter carapace length, lower oxygen consumption rate, and longer development time. This study will provide a basis for further research on larval development and the effects of climate change on reproductive sustainability in New England lobster fisheries.

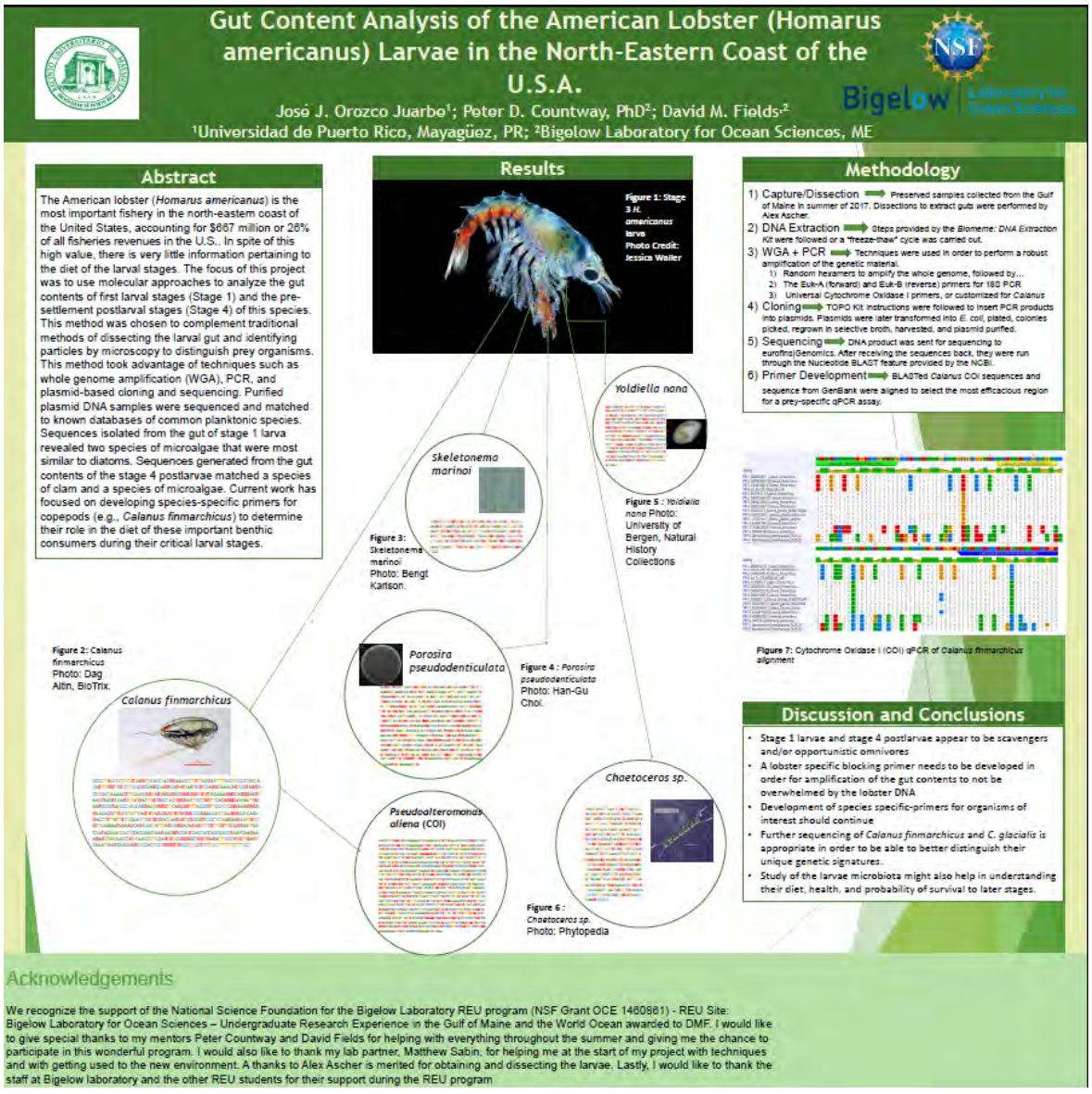


José J. Orozco Juarbe – Universidad de Puerto Rico – Mayagüez, Mayagüez, PR

Gut Content Analysis of the American Lobster (*Homarus americanus*) Larvae in the Gulf of Maine.

Orozco JJ[1], Countway P [2], Fields DM[2]
Universidad de Puerto Rico Rico – Mayagüez, Mayagüez, PR[1], Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[2].

The American lobster (*Homarus americanus*) is the most important fishery in the north-eastern coast of the United States, accounting for $667 million or 26% of all fisheries revenues in the U.S.. In spite of this high value, there is very little information pertaining to the diet of the larval stages. The focus of this project was to use molecular approaches to analyze the gut contents of first larval stages (Stage 1) and the pre-settlement postlarval stages (Stage 4) of this species. This method was chosen to complement traditional methods of dissecting the larval gut and identifying particles by microscopy to distinguish prey organisms. This method took advantage of techniques such as whole genome amplification (WGA), PCR, and plasmid-based cloning and sequencing. Purified plasmid DNA samples were sequenced and matched to known databases of common planktonic species. Sequences isolated from the gut of stage 1 larva revealed two species of microalgae that were most similar to diatoms. Sequences generated from the gut contents of the stage 4 postlarvae matched a species of clam and a species of microalgae. Current work has focused on developing species-specific primers for copepods (e.g., *Calanus finmarchicus*) to determine their role in the diet of these important benthic consumers during their critical larval stages.

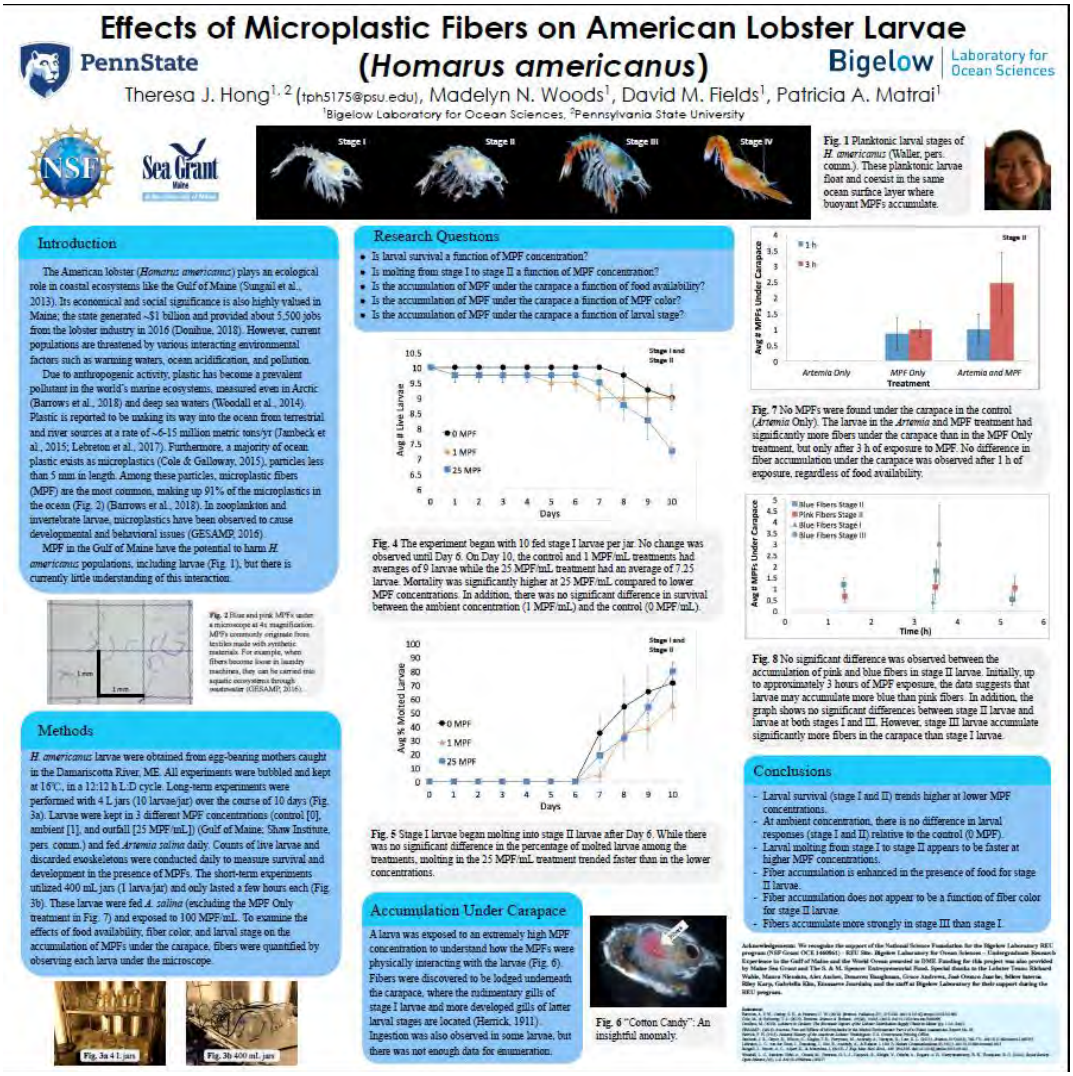


Theresa Hong – Pennsylvania State University, University Park, PA

Effects of Microplastic Fibers on American Lobster Larvae (*Homarus americanus)*

Hong TJ[1,2], Woods MN[1], Fields DM[1], Matrai PA[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], Pennsylvania State University, University Park, PA[2]

Due to anthropogenic activity, plastic has become a prevalent pollutant in the world's marine ecosystems, discovered even in Arctic and deep sea waters. Plastic is making its way into the ocean from terrestrial and river sources at a rate of ~6-15 MT yr^{-1}. The majority of ocean plastic exists as microplastics, particles less than 5 mm in length. Among these particles, microplastic fibers (MPF) are the most common, making up 91% of the microplastics in the ocean. In zooplankton and invertebrate larvae, microplastics have been observed to cause developmental and behavioral issues. MPFs pose a potential threat to larvae of the ecologically and economically significant American lobster (*Homarus americanus*), which is already threatened by climate change. Such planktonic larvae float and coexist in the same ocean surface layer where buoyant MPFs accumulate. Laboratory experiments were conducted to determine the effects of MPFs on the survival and development of *H. americanus* larvae under different MPF concentrations. Larval survival (stage I and stage II) was higher at lower MPF concentrations. Larval molting (stage I to II) appeared faster at higher MPF concentrations. Further experiments quantified the accumulation of fibers in the carapace as a function of food availability, fiber color, and larval stage. Fibers accumulated preferentially under the carapace in the presence of food for stage II larvae. MPF color had a slight effect on fiber accumulation under the carapace in stage II. MPF accumulated more in stage III than stage I. Food availability and larval stage contributed more strongly to carapace fiber accumulation than fiber color. It should be noted that for all treatments, there was no significant difference on the effect of ambient MPF concentrations ($\leq$ 1 MPF ml^{-1}) with respect to no fibers (control = 0 MPF ml^{-1}).



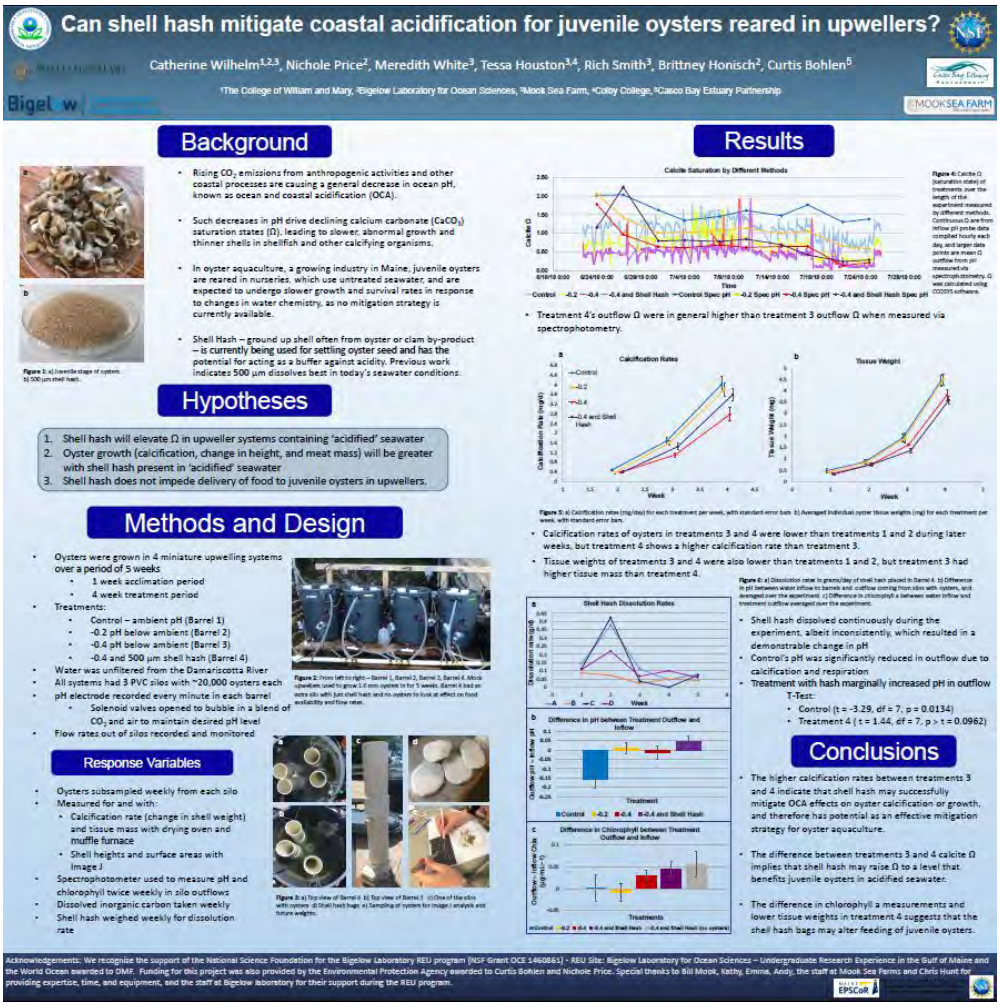
Catherine Wilhelm – The College of William and Mary, Williamsburg, VA

Potential Shell Hash Mitigation of Coastal Acidification for Juvenile Oysters Reared in Upwellers

Catherine Wilhelm [1,2,3], Nichole Price [2], Meredith White [3], Tessa Houston [3,4], Rich Smith [3], Brittney Honsich [2], Curtis Bohlen[5]

[1]The College of William and Mary, [2]Bigelow Laboratory for Ocean Sciences, [3]Mook Sea Farm, [4]Colby College, [5]Casco Bay Estuary Partnership

The phenomenon of decreasing pH in nearshore seawater due to increased CO_2 in the atmosphere and other land-sea interface processes, referred to as ocean and coastal acidification (OCA), is harmful to the development, growth, and survival of the larval phase of many calcified bivalve species. Within the aquaculture industry, oyster hatcheries have already begun implementing seawater buffer treatments to mitigate these OCA impacts. But, we know little about the vulnerability of older juvenile stages of oysters, which are grown-out in untreated upwelling systems, for which there is currently no mitigation strategy. Here we use a controlled experiment to test the mitigation potential of shell hash – finely ground up shell by-product (500 µm) – to determine whether hash can raise seawater Ω and oyster growth in miniature upweller mimics exposed to 'acidified' seawater. Juvenile oysters were subjected to naturally varying pH levels –an ambient pH control (T1), -0.2 pH below the ambient (T2), -0.4 pH (T3), and -0.4 + 500 µm hash (T4). We measured the calcification rates, tissue weight, and shell height of juvenile oysters weekly over 5 weeks. Calcification rates of oysters in T3 and T4 were lower than T1 and T2 during weeks 3 and 4, though T4 had greater average shell weight than T3 in week 4, indicating greater calcification. T4 also had significantly lower tissue weights than all other treatments, except T3 in the later weeks. Shell hash had an average dissolution of 0.12 g/day, and there was no difference in chlorophyll a abundance across the treatments. Our data indicates that the added 500 micron shell hash may increase calcification rates in response to 'acidified' seawater. This suggests that added shell hash may enough to mitigate future OCA effects on juvenile oysters in the aquaculture industry.

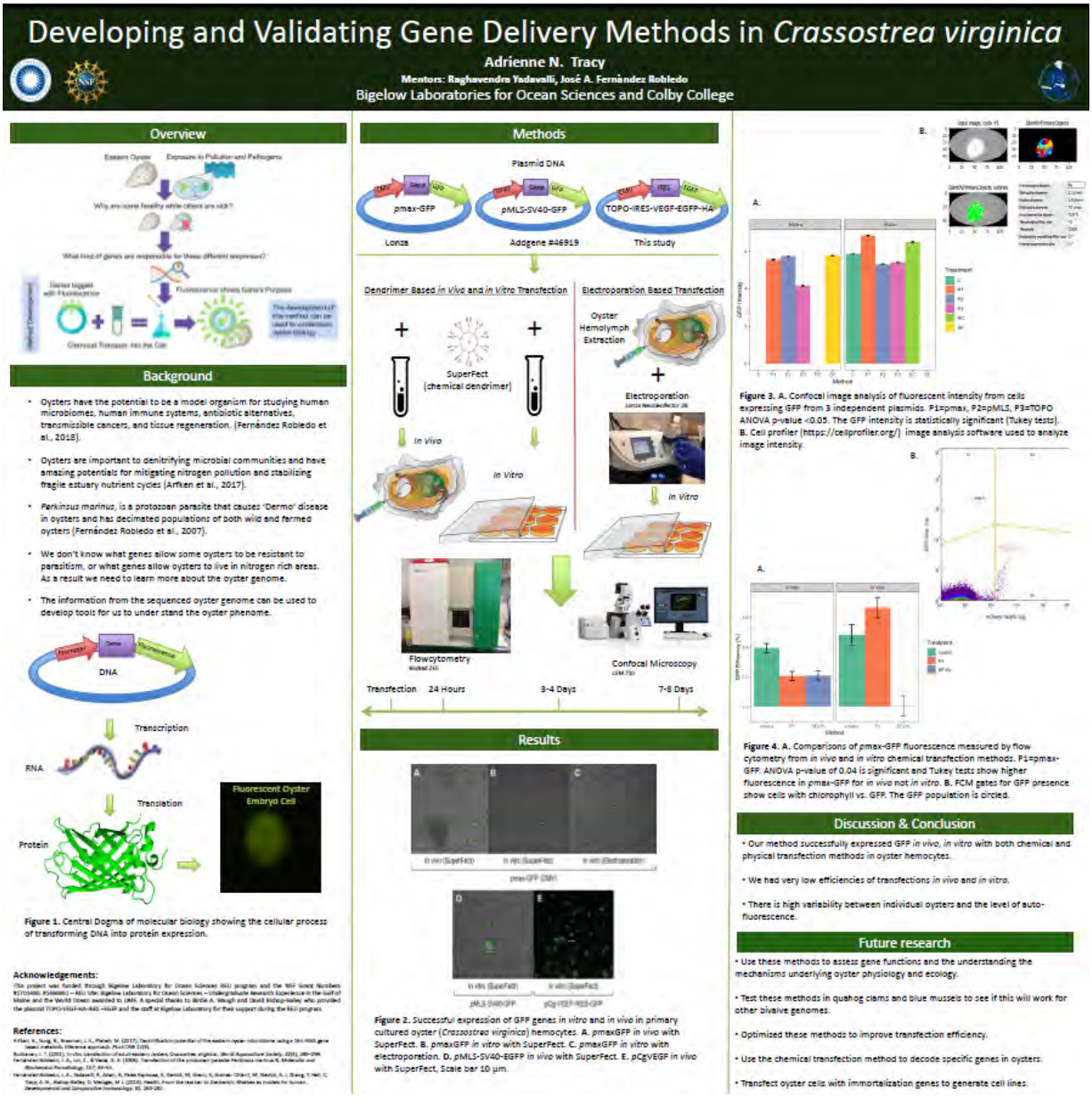


Adrienne N. Tracy – Colby College, Waterville, ME

Development and validation of gene delivery methods for *Crassostrea virginica*

Tracy, A. N. [1,2], Yadavalli, R.[1], Fernández Robledo, J. A.[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], Colby College, ME[2]

The Eastern oysters (*Crassostera virginica*), an important part of coastal economies, are being hurt by pollution, parasites, and diseases, but they have the potential to yield breakthroughs in environmental and medical research. With the oyster genome sequenced, scientists are focusing on deciphering the function of the predicted genes. This goal is now hampered by the limited number of molecular and cellular tools available. A more consistent gene delivery system needs to be developed to interrogate the genome in order to learn and assign gene functions to understand the phenome. In this study, we adapted and further developed methodologies for both chemical (dendrimers) and physical (electroporation) gene delivery into oyster hemoctyes *in vivo* and *in vitro* using plasmids under CMV and SV40 promoter control with GFP tagged genes. We expressed GFP in oyster hemocytes (blood cells) using all three methods and under both promoters. This methodology will allow for the assessment of gene functions and the understanding the mechanisms underlying oyster physiology and ecology and will give the opportunity to develop oyster cell lines.

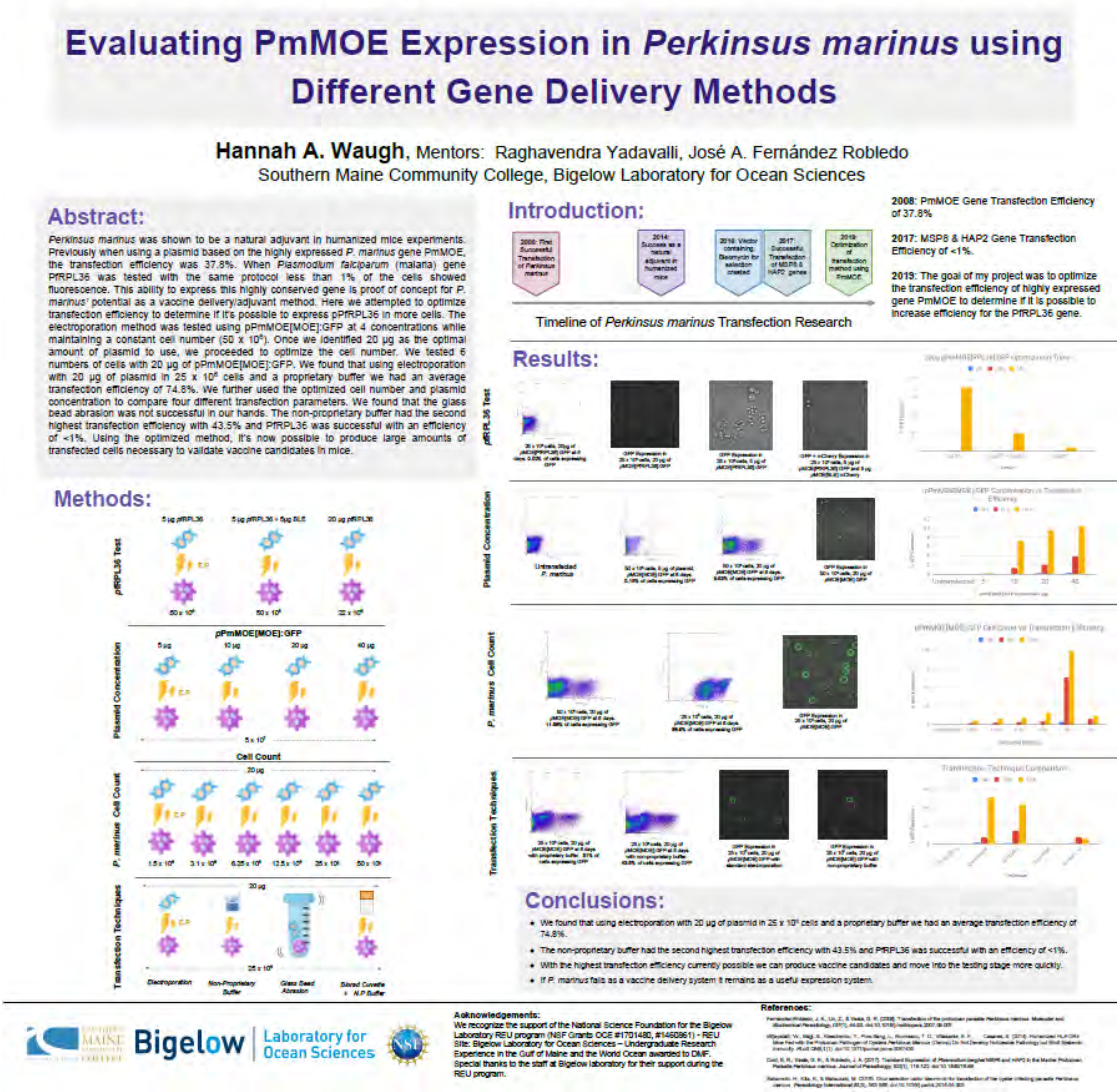


Hannah Waugh, Southern Maine Community College

Evaluating PmMOE Expression in *Perkinsus marinus* using Different Gene Delivery Methods

Waugh HA[1,2], Yadavalli R[1], Fernández Robledo JA[1]
Bigelow Laboratory for Ocean Sciences[1], East Boothbay, ME, Southern Maine Community College[2] South Portland, ME

Perkinsus marinus was shown to be a natural adjuvant in humanized mice experiments. Previously when using a plasmid based on the highly expressed P. marinus gene PmMOE, the transfection efficiency was 37.8%. When Plasmodium falciparum (malaria) gene PfRPL36 was tested with the same protocol less than 1% of the cells showed fluorescence. This ability to express this highly conserved gene is proof of concept for P. marinus' potential as a vaccine delivery/adjuvant method. Here we attempted to optimize transfection efficiency to determine if it's possible to express pPfRPL36 in more cells. The electroporation method was tested using pPmMOE[MOE]:GFP at 4 concentrations while maintaining a constant cell number (5 x 107). Once we identified 20 µg as the optimal amount of plasmid to use, we proceeded to optimize the cell number. We tested 6 numbers of cells with 20 µg of pPmMOE[MOE]:GFP. We found that using electroporation with 20 µg of plasmid in 2.5 x 107 cells and a proprietary buffer we had an average transfection efficiency of 74.8%. We further used the optimized cell number and plasmid concentration to compare four different transfection parameters. We found that the glass bead abrasion was not successful in our hands. The non-proprietary buffer had the second highest transfection efficiency with 43.5% and PfRPL36 was successful with an efficiency of <1%. Using the optimized method, it's now possible to produce large amounts of transfected cells necessary to validate vaccine candidates in mice.



Cristina Tusei – Humboldt State University, Arcata, CA

Photodegradation of Chlorinated Organic Compounds in Surface Waters

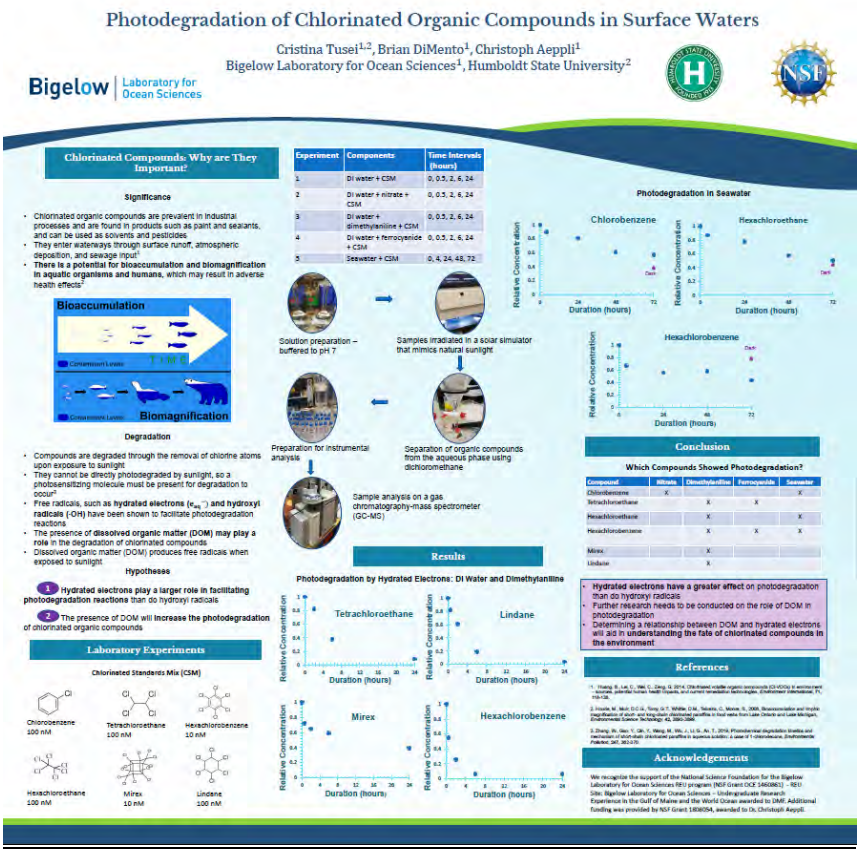
Tusei C[1,2], DiMento B[1], Aeppli C[1]

Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], Humboldt State University[2]

Chlorinated organic compounds are produced in large quantities and used in industrial products as pesticides and solvents. They easily leach into aquatic environments and pose a health risk to aquatic organisms and humans due to lengthy residence times and slow biodegradation. However, sunlight has the potential to photodegrade chlorinated organic compounds and abstract chlorine atoms. This is possible if photosensitizing molecules produce reactive species, such as hydrated electrons (e_{aq}^-) and hydroxyl radicals (·OH). Dissolved organic matter (DOM) present in natural waters forms eaq^- and ·OH when exposed to sunlight and may facilitate photodegradation. While much research has been conducted on ·OH, not much is known about the importance of e_{aq}^- in the degradation of chlorinated compounds in surface waters.

The research presented here explores the effects eaq^- and ·OH have on the photodegradation of a mixture of six chlorinated compounds, incubated in deionized and seawater matrices and irradiated by simulated sunlight. The results from these experiments suggest that hydrated electrons play a greater role in facilitating photodegradation of highly chlorinated compounds than do hydroxyl radicals. A clear depletion in four out of six chlorinated compounds was observed in irradiated samples. Although it cannot be said the results are statistically significant, a clear pattern is established. Initial experiments performed with natural seawater were not yet able to clearly identify the effects of e_{aq}^- and further experiments must be conducted to understand the role DOM plays in photodegradation.

These results may be used for future reference in studying the fate of chlorinated organic compounds. By understanding the characteristics of free radicals produced by DOM, it is possible to predict the degradation rate of harmful chlorinated compounds in natural waters.

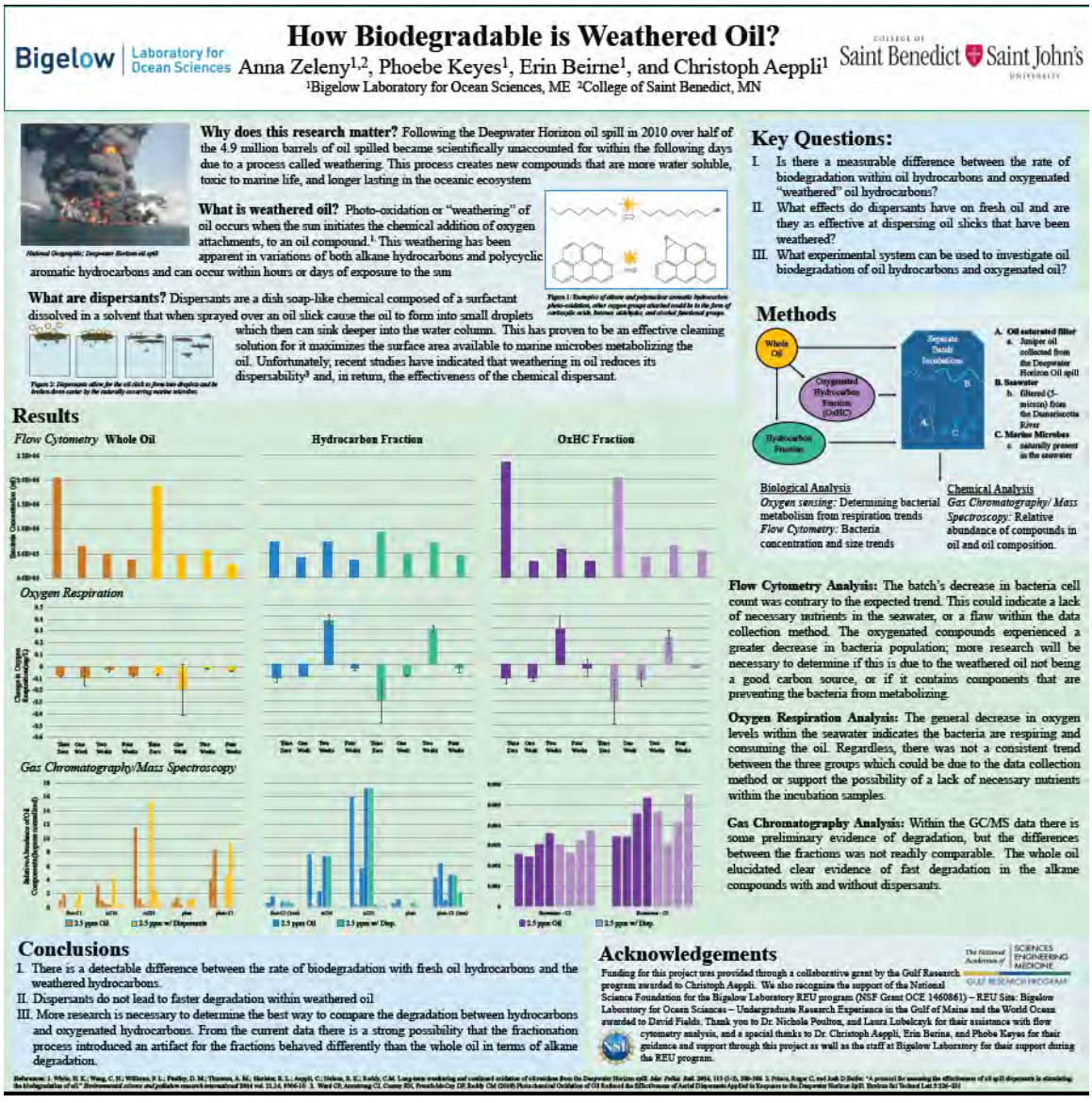


Anna Zeleny – College of Saint Benedict, Saint Joseph, MN

How Biodegradable is Weathered Oil?

Zeleny A[1,2],Beirne E[1], Keyes P[1], and Aeppli C[1]
[1]Bigelow Laboratory for Ocean Sciences, ME [2]College of Saint Benedict, MN

Following the Deepwater Horizon oil spill in 2010 research has indicated that over half of the 4.9 million barrels of oil released began to be photo-oxidized from the sun within the following days. Although research has concluded that naturally occurring marine microorganisms will degrade oil hydrocarbons, previous studies have neglected to quantify how biodegradable this photo-oxidized or "weathered" oil is. It is currently unclear if weathered oil is degraded in a similar time frame, and by similar microorganisms. In this study, the degradation of oil by marine microbes was compared between fresh and weathered oil through batch incubation experiments lasting three to four weeks. Furthermore, the influence of dispersants was tested. Dispersants are a common practice for responding to oil spills to facilitate oil biodegradation, but recent studies have indicated that oil photo-oxidation reduces oil dispersability. The cell density and oxygen respiration were measured during the experiment to determine bacterial growth and activity; gas chromatography was utilized to determine the degradation of oil hydrocarbons and oxygenated compounds within the oil. The results in this study supported that there is a difference in degradation rates of oil hydrocarbons and oxygenated compounds, and that dispersants do not lead to faster degradation within weathered oil.

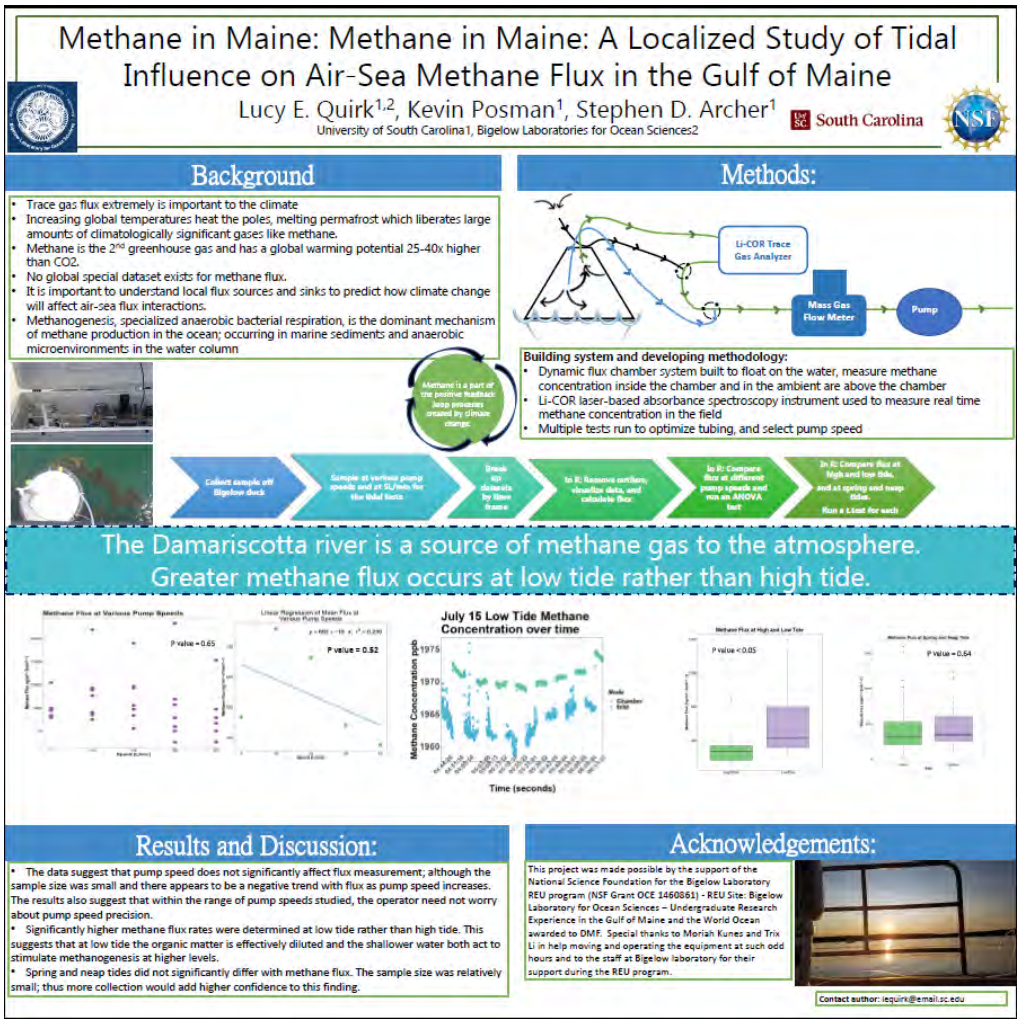


Lucy Quirk – University of South Carolina, Columbia, SC

Methane in Maine: A Localized Study of Tidal Influence on Air-Sea Methane Flux in the Gulf of Maine

Quirk, LE[1,2], Posman, KM[1], Archer, SD[1]
Bigelow Laboratory for Ocean Sciences[1], University of South Carolina[2]

Air-sea flux of methane influences global climate. This climatologically significant gas has a residence time in the atmosphere of ~10 years and over a 100-year period has a global warming potential 24-40x higher than CO_2. Methanogenesis, specialized anaerobic bacterial respiration, is the dominant mechanism of methane production in the ocean; occurring in marine sediments and anaerobic microenvironments in the water column. This study aimed to quantify methane air-sea flux in the temperate, coastal waters of the estuarine Damarisoctta River in the Gulf of Maine. A custom built, floating dynamic flux chamber linked to a laser-based absorbance spectroscopy methane-analyzer (Li-COR 7810) were used to quantify methane flux rates. A range of flow rates through the chamber were tested to optimize our approach. Methods for data handling and interpretation were developed in R. Flow rate had a non-linear effect on flux rate estimations; analyses were run at 5L/min but optimum flow rates may be as high as 15L/min. Data was collected at high and low tides to observe whether tidal cycles influence methane flux. Air concentrations of methane varied from 1938.79 to 2022.85 ppb during a tidal cycle. Calculated flux rates ranged from 77 to 1591 ng/m^{-2}/min, all from seawater to air. High and low tide flux varied significantly; for instance, the mean flux rate for high and low tide over one tidal cycle were 304.63 and 741.13ng/m^{-2}/min respectively. We hypothesized that higher flux at low tide compared to high tide resulted from more estuarine waters at low tide and increased exchange between methane and sediments sources, while incoming tides brought lower methane-containing seawater into the region. Tidal cycles must be considered when estimating the source-strength for methane in temperate estuarine systems. More improvements to the dynamic flux chamber will be made to ensure consistent airflow contact with seawater and establish optimum airflow rates for varied water types.



Zhouxin (Trix) Li - Wheaton College, Norton, MA

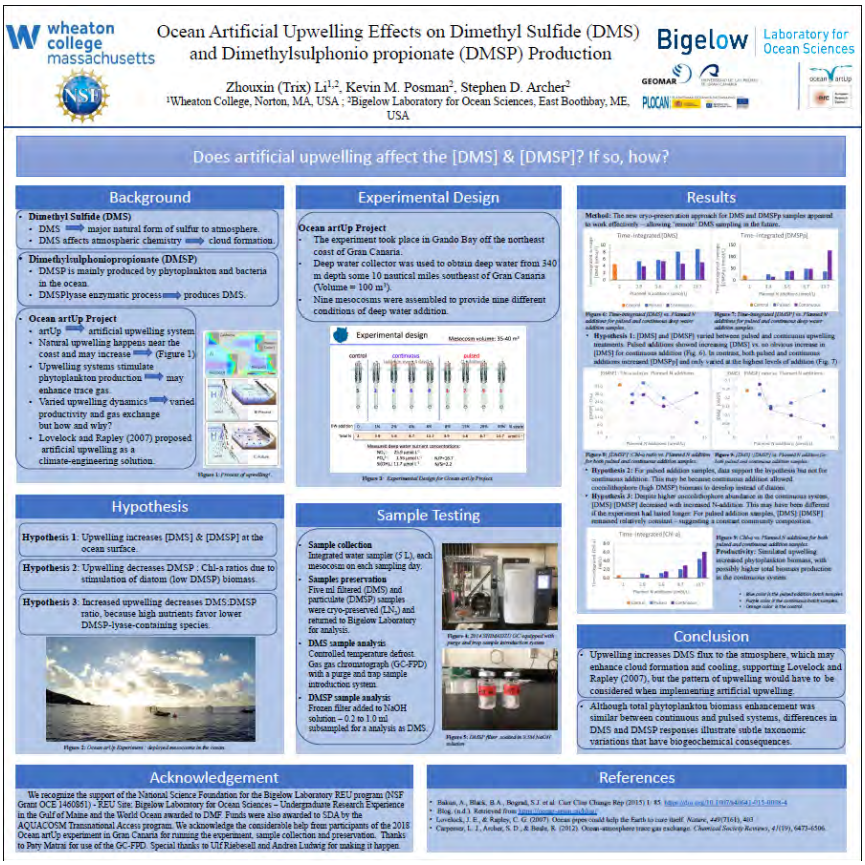
Ocean Artificial Upwelling Effects on Dimethyl Sulfide (DMS) and Dimethylsulphoniopropionate (DMSP) Production

Zhouxin (Trix) Li[1,2], Kevin M. Posman[2], Stephen D. Archer[2]
[1]Wheaton College, Norton , MA,USA ; [2]Bigelow Laboratory for Ocean Sciences, East Boothbay, ME, USA

Dimethyl sulfide (DMS) is the major form of sulfur release from the ocean to the atmosphere. Dimethylsulphoniopropionate (DMSP) produced by phytoplankton and bacteria, is the main precursor of DMS. DMS emissions affect cloud formation, lifetime, and albedo. The extent of changes in DMS emissions from the oceans and how this alters the Earth's radiative balance has implications for climate change.

Coastal upwelling brings nutrient-rich deep water to the surface ocean, stimulating growth of phytoplankton and potentially altering concentrations of DMSP and release of DMS to the atmosphere. The high primary productivity of upwelling systems also supports large pelagic fisheries. Lovelock and Rapley (2007)[1] proposed that artificial upwelling could be used as a climate-engineering solution. They hypothesized that increased upwelling would stimulate DMS emissions, increasing the formation of clouds, which reflect more sunlight and as a result, cool the Earth.

As part of the Ocean artUp Experiment carried out off Gran Canaria over 10-weeks in 2018, we examined the consequences of simulated upwelling on net DMSP and DMS production. Deep-water (340 m) additions to large-scale (~50 m^3) mesocosm bags were used to simulate continuous and pulsed upwelling patterns. A new cryo-preservation approach allowed DMS(P) concentrations to be determined by purge-and-trap GC-FPD analysis on samples collected ~6 months earlier. The results are interpreted in relation to changes in community composition stimulated by enhanced nutrient availability. Specifically, we examined temporal changes in the ratios of DMSP:Chl-a, DMS:Chl-a and DMSP:DMS between pulsed and continuous deep water addition scenarios. This information is used to assess Lovelock and Rapley's hypothesis and the consequences for DMS emissions if artificial upwelling was used to enhance fisheries productivity.

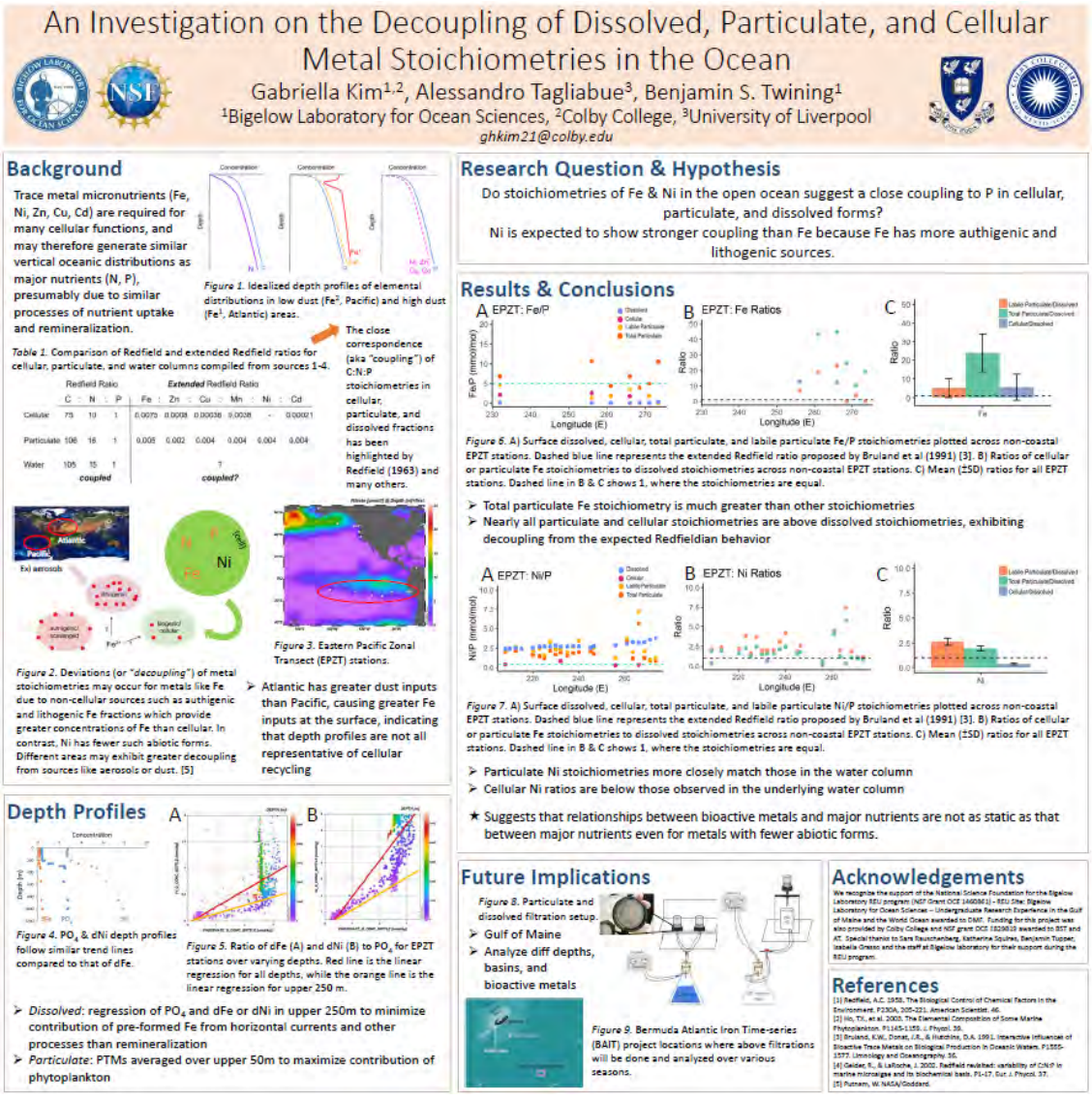


Gabriella Kim - Colby College, Waterville, ME

An Investigation on the Decoupling of Dissolved, Particulate, and Cellular Metal Ratios in the Ocean

Kim GH[1,2], Tagliabue A[3] , Twining BS[1]
Bigelow Laboratory for Ocean Sciences[1], Colby College[2], University of Liverpool[3]

Trace metal micronutrients such as Fe and Ni are required for many cellular functions and therefore often present similar vertical oceanic distributions as major nutrients such as N and P, presumably due to similar processes of nutrient uptake and remineralization. As a result, scientists have suggested the existence of an 'extended Redfield ratio', representing a constant correspondence, or "coupling", of the ratio of biomass and trace metals between cells and the water column. However, the ratios for metals such as Fe may deviate and "decouple" from the suggested constants due to non-cellular sources such as authigenic and lithogenic fractions. We investigated if the concept of an extended Redfield ratio is supported by data collected as part of the GEOTRACES project in the Pacific. We focused on metals with other abiotic forms (Fe), as well as those with fewer abiotic forms (Ni). To do so, we calculated ratios from dissolved phases and compared them to ratios in cellular and particulate material in the upper, non-coastal water column. Though both Fe & Ni have particulate ratios that are much greater than those in cells, Ni ratios more closely match the expected Redfieldian behavior. The cellular Fe ratio is significantly above that in the water column, while the cellular Ni ratio is below. Though further analysis will be done testing different constraints, these results suggest that relationships between bioactive metals and major nutrients are not as static as that between major nutrients.

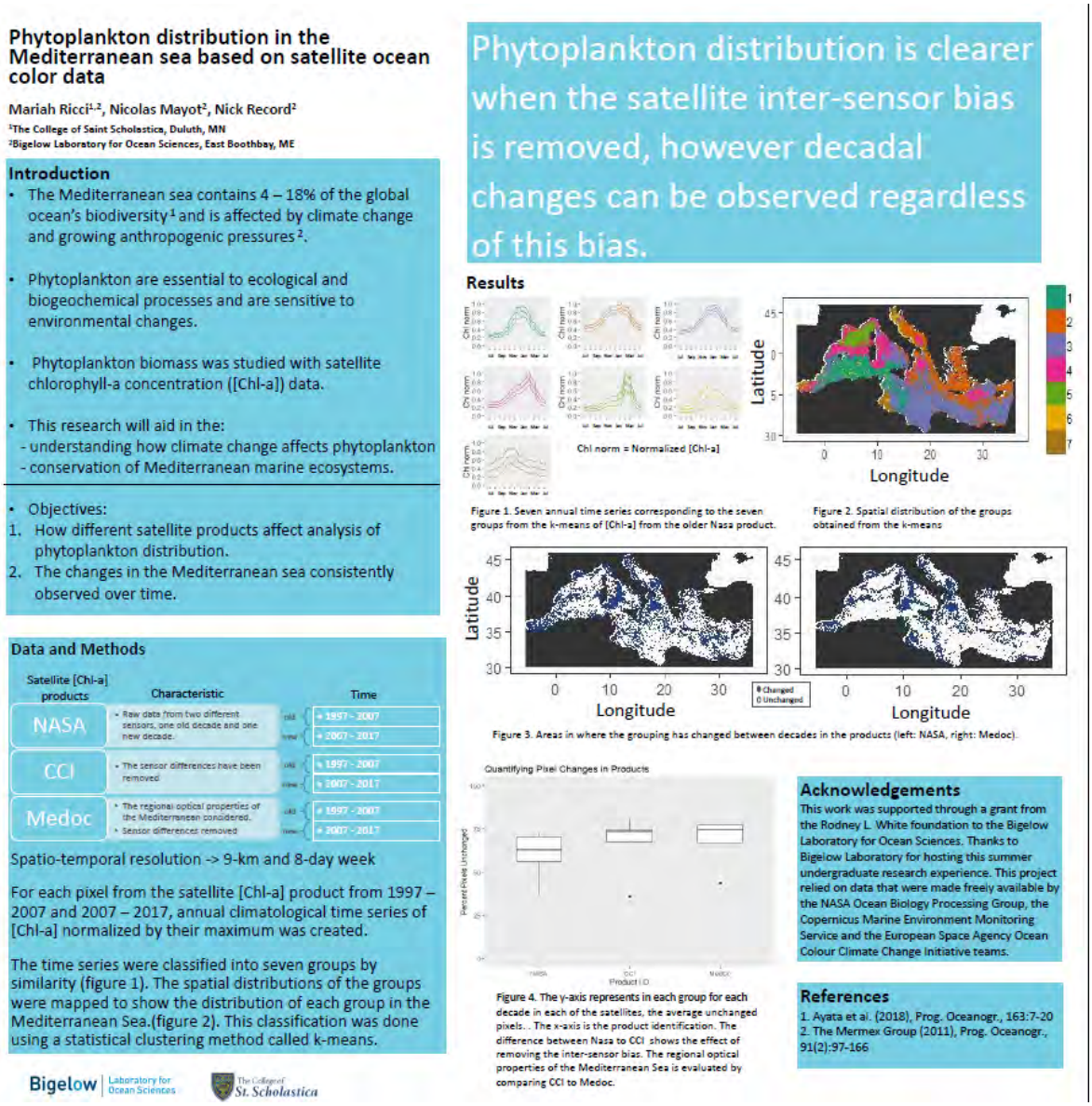


Mariah Ricci- The College of Saint Scholastica, Duluth, MN

Phytoplankton Distribution in the Mediterranean Sea Based on Ocean Color Data

Mariah Ricci [1,2], Nicolas Mayot [1] , Nick Record[1]
Bigelow Laboratory for Ocean Sciences, East Boothbay, ME[1], The College of Saint Scholastica, Duluth, MN[2]

Phytoplankton are essential to ecological and biogeochemical processes and are sensitive to environmental changes. Ongoing climate change and growing anthropogenic pressures are affecting the Mediterranean Sea. To study the effect on phytoplankton distribution in the Mediterranean basin, two decades (from 1997 to 2017) of satellite chlorophyll-*a* concentration data from different datasets (combine different satellite sensors and algorithms) were used. The spatial patterns in phytoplankton dynamics were obtained by using a statistical clustering method which classified the observed seasonal cycle of chlorophyll-a concentration. These patterns were more distinct from one another when using datasets in which the inter-sensor bias (from combining data from different satellite sensors) was reduced. For example, a quantitative analysis was done that revealed the method to classify data was more successful when the inter-sensor bias was removed Consistent changes over time in the phytoplankton distribution can be observed in the Adriatic and the North Ionian seas, as well as in the Tyrrhenian Sea, despite the satellite dataset used. This work emphasizes how climate changes affect the phytoplankton and could help for future management actions and studies of Mediterranean marine ecosystems.

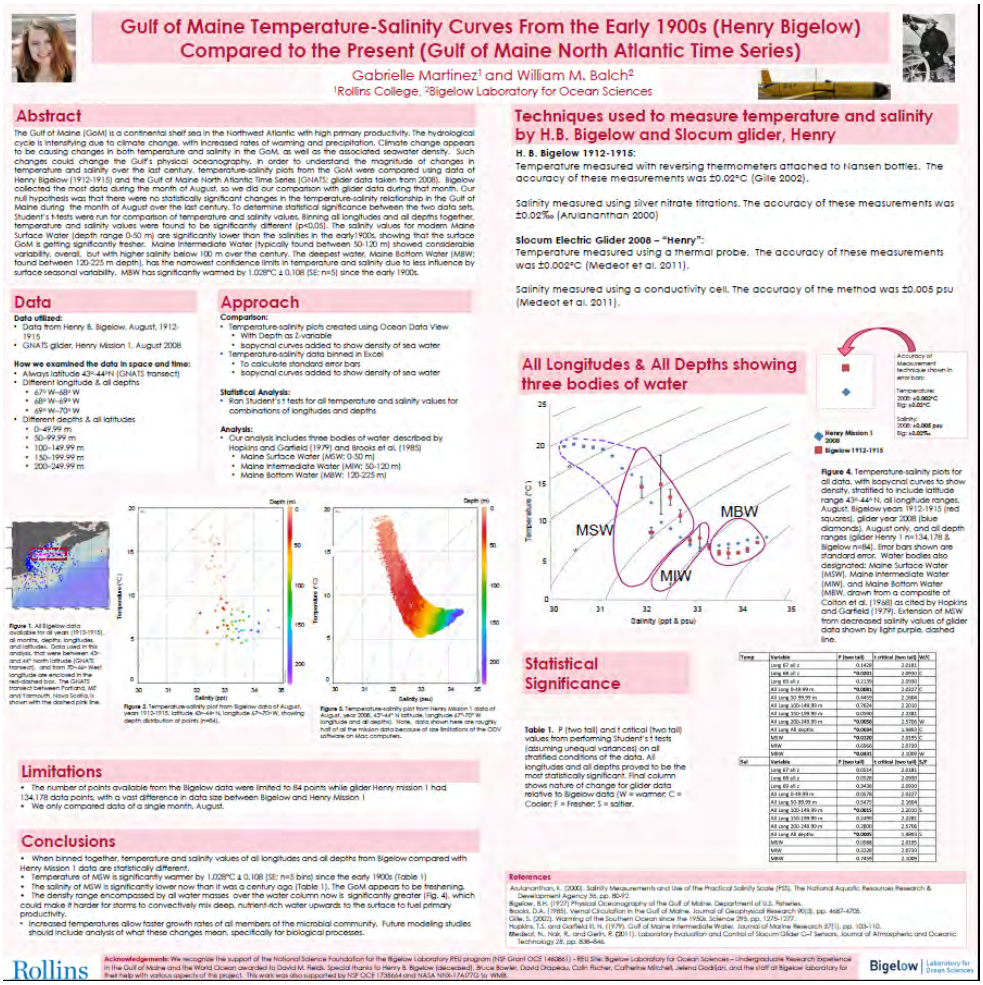


Gabrielle Martinez – Rollins College, Winter Park, FL

Gulf of Maine Temperature Salinity Curves From the Early 1900s (Henry Bigelow) Compared to the Present (GNATS)

Martinez, G.A.[1], Balch, W.M.[2]
Rollins College[1], Bigelow Laboratory for Ocean Sciences[2]

The Gulf of Maine (GOM) is a continental shelf sea in the Northwest Atlantic with high primary productivity. The hydrological cycle is intensifying due to climate change, with increased rates of warming and precipitation. Climate change appears to be causing changes in temperature (and possibly salinity) in the GoM, as well as the associated density. Therefore, this could change the Gulf's physical oceanography. In order to understand the magnitude of changes in temperature and salinity over the last century, temperature-salinity plots from the GoM were compared using data of Henry Bigelow (1912-15) and the Gulf of Maine North Atlantic Time Series (GNATS; glider data taken from 2008). Bigelow collected the most data during the month of August, so we did our comparison with glider data during that month. Our question was whether there were any statistically significant changes in the temperature-salinity relationship in the Gulf of Maine during the month of August over the last century. To determine statistical significance between the two data sets, t-tests were run for all temperature and salinity values. Binning all longitudes and all depths together, temperature and salinity values were found to be significantly different ($p<0.05$). The salinity values for modern Maine Surface Water (depth range 0-50m) are significantly lower than the salinities in the1900s, showing that the surface GoM is getting significantly fresher. Maine Intermediate Water (typically found between 50-120m) showed considerable variability, thus no significant changes could be discerned over the century. The deepest water, Maine Deep Water (MDW; found between 120-225m depth), has the narrowest confidence limits due to less influence by surface seasonal variability. MDW has significantly warmed by 1.028°C ± 0.108 (SE; n=5) since the early 1900s.



Acknowledgements: We recognize the support of the National Science Foundation for the Bigelow Laboratory REU program (NSF Grant OCE 1460861) - REU Site: Bigelow Laboratory for Ocean Sciences – Undergraduate Research Experience in the Gulf of Maine and the World Ocean awarded to David M. Fields. Special thanks to Henry B. Bigelow (deceased), Bruce Bowler, David Drapeau, Colin Fischer, Catherine Mitchell, Jeleny Godfrey, and the staff at Bigelow Laboratory for their help with various aspects of the project. This work was also supported by NSF OCE 1736664 and NASA NNX17AI77G to WMB.

Moriah Kunes – University of Rochester, Rochester, NY

A Novel Approach to Quantify the Grazing Rates of Microzooplankton

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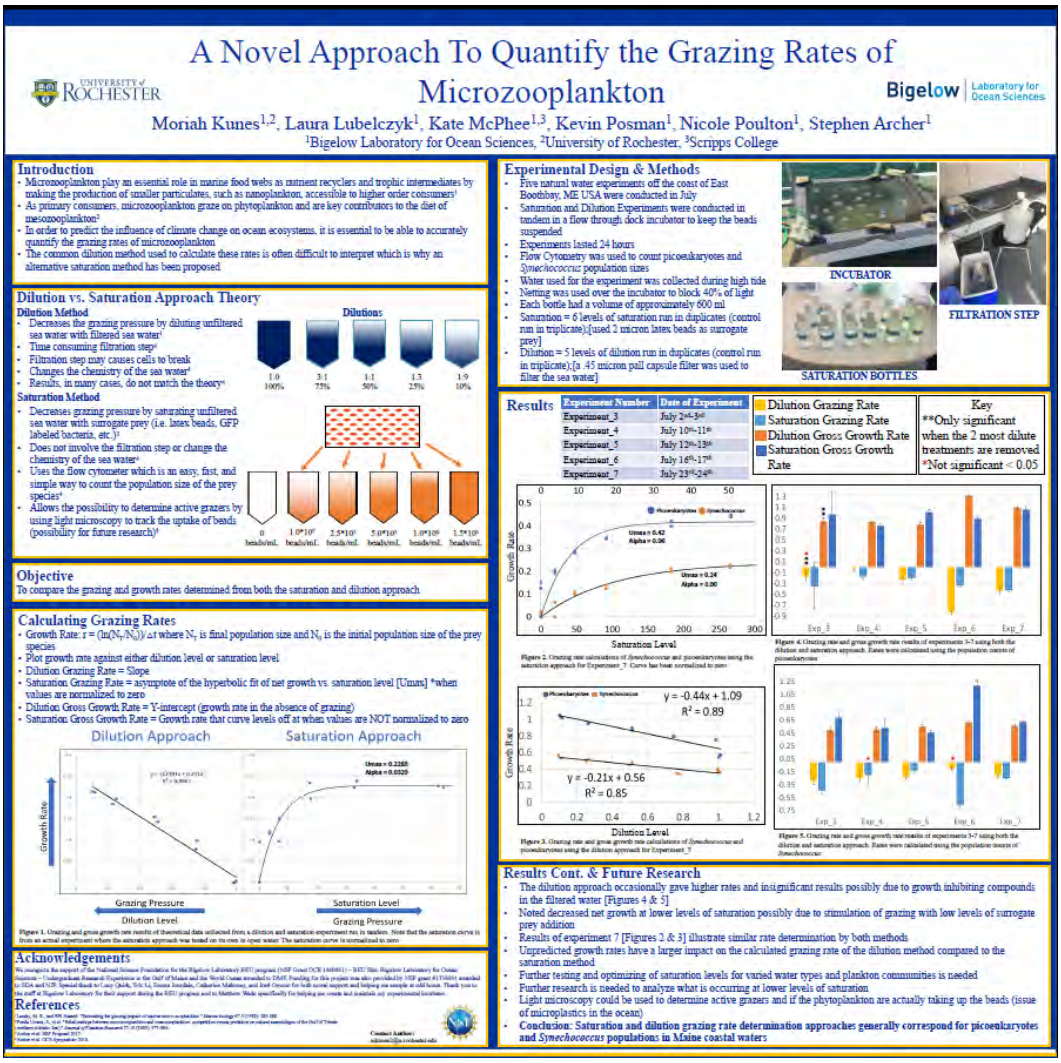
Microzooplankton are primary consumers in the ocean and play an essential role in marine food webs as nutrient recyclers and trophic intermediates. In order to predict the influence of climate change on ocean ecosystems, it is essential to be able to accurately quantify how microzooplankton affect primary production and hence, food webs. In this study we attempt to reform how their grazing rates can be determined. Currently, the most common method is the dilution approach which involves diluting seawater with filtered seawater sequentially reducing grazing pressure by decreasing predator-prey encounter rates. However, this approach changes the chemistry of the seawater sample and involves a time-consuming filtration step. This study tested a novel saturation technique that has the potential to eliminate the disadvantages of the traditional dilution method and increase sample throughput. In this case, grazing pressure is reduced through saturation of grazers with surrogate prey. Natural water experiments off the coast of East Boothbay, ME were used to compare the dilution and saturation methods. In July, five dilution and saturation experiments were conducted in tandem in a flow through dock incubator. Using flow cytometry, picoeukaryote population sizes were counted to calculate gross growth rates and grazing rates. Our results illustrate similar rate determination by both methods, plus some inconsistencies in both approaches, meaning that the results do not always fit expected mathematical interpretations. Specifically, we saw lower than expected growth rates at higher dilution levels possibly due to growth inhibiting compounds in the filtered water. We also noted decreased net growth at lower levels of saturation possibly due to stimulation of grazing with low levels of surrogate prey addition. The saturation approach generated encouraging results but requires further testing and optimizing for varied water types and plankton communities.



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CELLULAR RESPONSE OF *EMILIANIA HUXLEYI* TO GROWTH ON PHOSPHONATES

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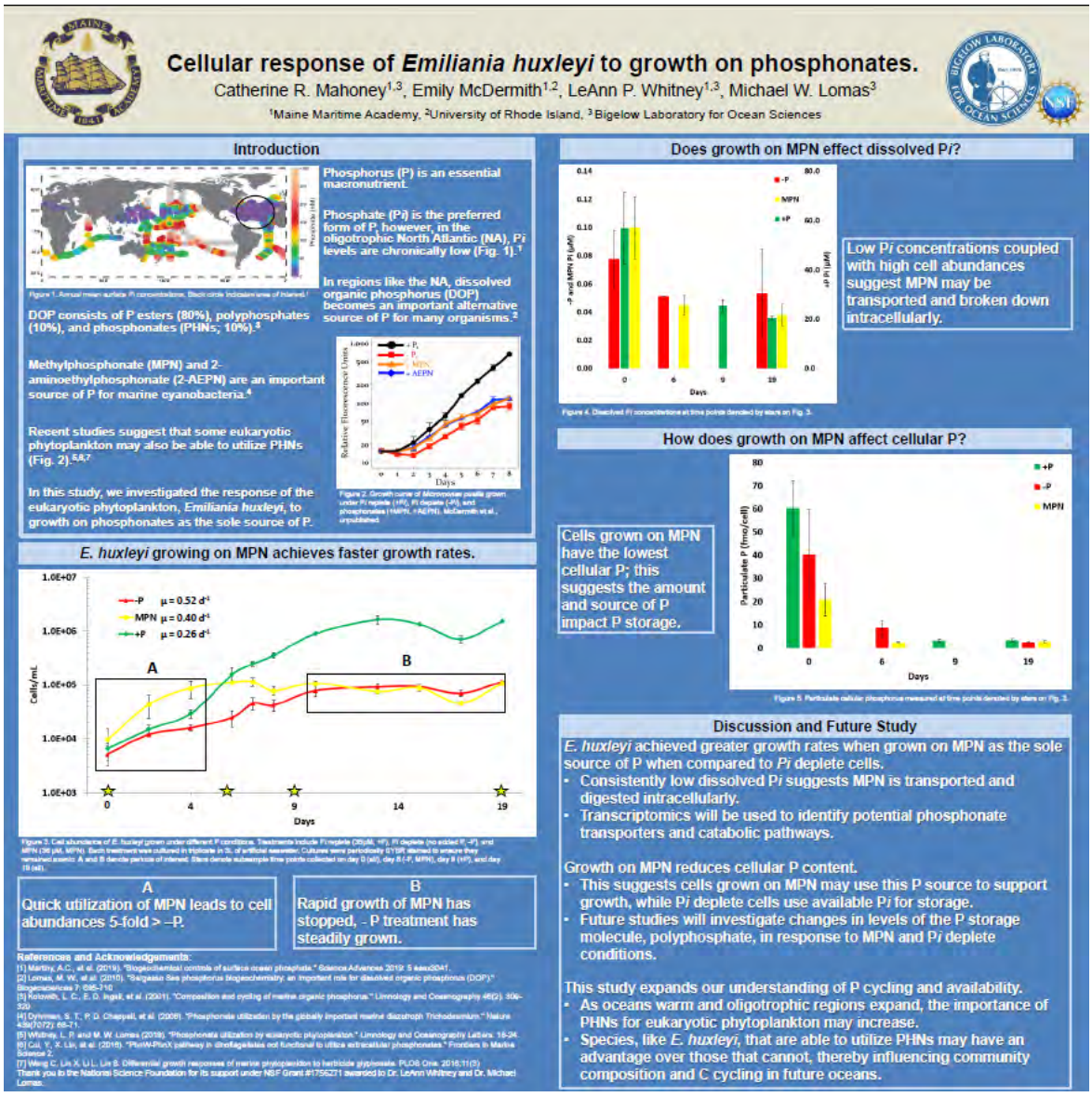
The importance of phosphorus (P) in cell structure and function make it an essential nutrient in the growth of phytoplankton. Large oceanic regions, such as the subtropical North Atlantic (NA), have growth-limiting levels of phosphate (P_i), the preferred form of P. In these regions, dissolved organic phosphorus (DOP) is an important alternative P source. The DOP pool largely consists of P-esters and phosphonates; both prokaryotic and eukaryotic organisms readily utilize P-esters, while phosphonates are thought to be an important P source only to prokaryotes. Recent studies have shown that some species of eukaryotic phytoplankton are able to utilize natural and/or chemically synthesized forms of phosphonates for growth, but the ability is not universal. In this study, we investigated the response of the eukaryotic phytoplankton, *Emiliania huxleyi*, to growth on phosphonates as the sole source of P compared to growth on replete and deplete levels of P_i . The effect of growth on methylphosphonate on cellular P and dissolved P concentrations were also measured. *E. huxleyi* supplemented with methylphosphonate achieved higher growth rates and final cell concentrations than cells grown under P deplete conditions, while cells grown under replete P_i conditions achieved the greatest growth rate and cell concentrations. This suggests that *E. huxleyi* is able to utilize methylphosphonate to support growth. As climate change continues to affect nutrient availability, alternate sources of P will become increasingly important. This research enhances our understanding of the physiological effect of DOP utilization on eukaryotic phytoplankton.



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