



Bigelow Laboratory for Ocean Sciences

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

**REU Symposium Program & Abstracts
Thursday, August 13, 2015**



Program

8:45 Opening Remarks

- 9:00 Kronauer Anna B., Colby College, Waterville, ME USA
MACHINE LEARNING: FORECASTING THE ABUNDANCE OF GELATINOUS ZOOPLANKTON IN THE GULF OF MAINE
Mentors: Dr. Record NR
- 9:15 Keady Caitlin A., Bates College, Lewiston, ME, USA
INVESTIGATING CHLOROPHYLL-A ANOMALIES IN THE SOUTHERN OCEAN AND THE TASMAN SEA
Mentors: Drs. Lee Y, Record NR
- 9:30 Harrison Amelia O., University of Delaware, Newark, DE, USA
CHARACTERIZATION AND RATE ANALYSIS OF THE ENZYME BROMOPEROXIDASE IN DIVERSE MICROALGAE
Mentor: Dr. Archer SD
- 9:45 Katz Sam D., Hampshire College, Amherst, MA, USA
DETERMINING THE MECHANISM FOR BROMOFORM FORMATION BY DIATOMS
Mentor: Dr. Aepli C
- 10:00 Morefield Robert D., Southern Maine Community College, South Portland, ME, USA
HEMOCYTE RESPONSE TO SECONDARY INFECTION OF DERM DISEASED OYSTERS
Mentor: Drs. Poulton N, Fernández-Robledo JA
- 10:15 **Break (15 minutes)** *****
- 10:30 Khana Daven B., University of Georgia, Athens, GA, USA
IDENTIFYING GENES INVOLVED IN THE IRON METABOLISM PATHWAY THROUGH TRANSCRIPTOMIC ANALYSIS
Mentor: Dr. Emerson D
- 10:45 Navarro-Guitz Hector, Shepherd University, Shepherdstown, WV, USA
THE EFFECTS OF OCEAN ACIDIFICATION OF THE ABUNDANCE OF DMSP-DEGRADING BACTERIA
Mentor: Dr. Countway P
- 11:00 Spaulding-Astudillo Francisco E., University of Chicago, Chicago, IL, USA
HYDROCARBON DEGRADATION PATHWAYS USED BY COASTAL SEDIMENT MICROBIAL COMMUNITIES EXPOSED TO CRUDE OIL
Mentor: Dr. Orcutt BN
- 11:15 Hayden Lauren C., Southern Maine Community College, South Portland, ME, USA
OPTIMIZING THE CRYOPRESERVATION OF *PAVLOVA* *sp.*
Mentor: Dr. Lomas MW
- 11:30 **Break (15 minutes)** *****
- 11:45 Chmiel Rebecca J., Colby College, Waterville, ME, USA
GLOEOTRICHIA ECHINULATA IN MAINE LAKES: AN ANALYSIS OF GENETIC DIVERSITY AND DNA-BASED DETECTION
Mentor: Dr. Countway P

- 12:00 Stemple Brooke, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA
BIOAVAILABILITY OF IRON SUBSTRATES TO DIATOMS
Mentor: Dr. Twining BS
- 12:15 Cold Emma R., Utah Valley University, Orem, UT, USA
HETEROLOGOUS EXPRESSION OF GENE OF INTEREST USING THE MARINE PROTOZOAN
PERKINSUS MARINUS
Mentor: Dr. Fernández Robledo JA
- 12:30 Goode Andrew G., University of Maine, Orono, ME, USA
PHYSIOLOGICAL RESPONSES OF *OXYRRHIS MARINA* TO THE ALTERED LIPID COMPOSITION OF
VIRALLY INFECTED *EMILIANIA HUXLEYI* CELLS
Mentor: Drs. Martinez Martinez J, Fields DM
- 12:45 Break (1 hr 15 min) *******
- 14:00 Fulton Allyson M., Bowdoin College, Brunswick, Maine, USA
THE ART OF SCIENTIFIC STORYTELLING: THREE STEPS TO COMMUNICATING YOUR SCIENCE
EFFECTIVELY
Mentor: Trew Crist D.
- 14:15 Marra Madison T., Colby College, Waterville, ME, USA
EFFECTS OF EHV-86-INFECTED *EMILIANIA HUXLEYI* ON EGG PRODUCTION AND BEHAVIOR OF
ACARTIA TONSA
Mentor: Drs. Martinez Martinez J , Fields DM
- 14:30 Aleem Aaminah, SUNY Rockland, Suffern, NY, USA
VIRAL INFECTION OF *EMILIANIA HUXLEYI*: IMPLICATIONS ON OXYGEN PRODUCTION AND
CONSUMPTION IN A MULTI-TROPHIC SYSTEM
Mentor: Drs. Fields DM, Martinez Martinez J
- 14:45 Ets-Hokin Jeremiah M., Humboldt State University, Arcata, CA, USA
AFFECTS OF TIDALLY DRIVEN VARIATION ON THE RESPONSE OF CORALLINE ALGAE TO OCEAN
ACIDIFICATION
Mentor: Dr. Price NN
- 15:00: Break (15 minutes) *******
- 15:15 McVeigh Halley N., Warren Wilson College, Asheville, NC, USA
DEVELOPMENTAL EFFECTS OF OCEAN ACIDIFICATION CONDITIONS AND ELEVATED
TEMPERATURE ON *HOMARUS AMERICANUS* LARVAE
Mentor: Dr. Fields DM
- 15:30 Fachon Evangeline, Northeastern University, Boston, MA, USA
OCEAN ACIDIFICATION DIFFERENTIALY AFFECTS THE PHOTOSYNTHESIS OF KEY NEW
ENGLAND MACROPHYTES
Mentor: Dr. Price NN
- 15:45 Maine Julia E., Bowdoin College, Brunswick, ME, USA
EFFECT OF OCEAN ACIDIFICATION ON THE FOOD QUALITY OF THE COCCOLITHOPHORE
E. HUXLEYI
Mentors: Drs. White, MM, Balch, WM, Milke, LM

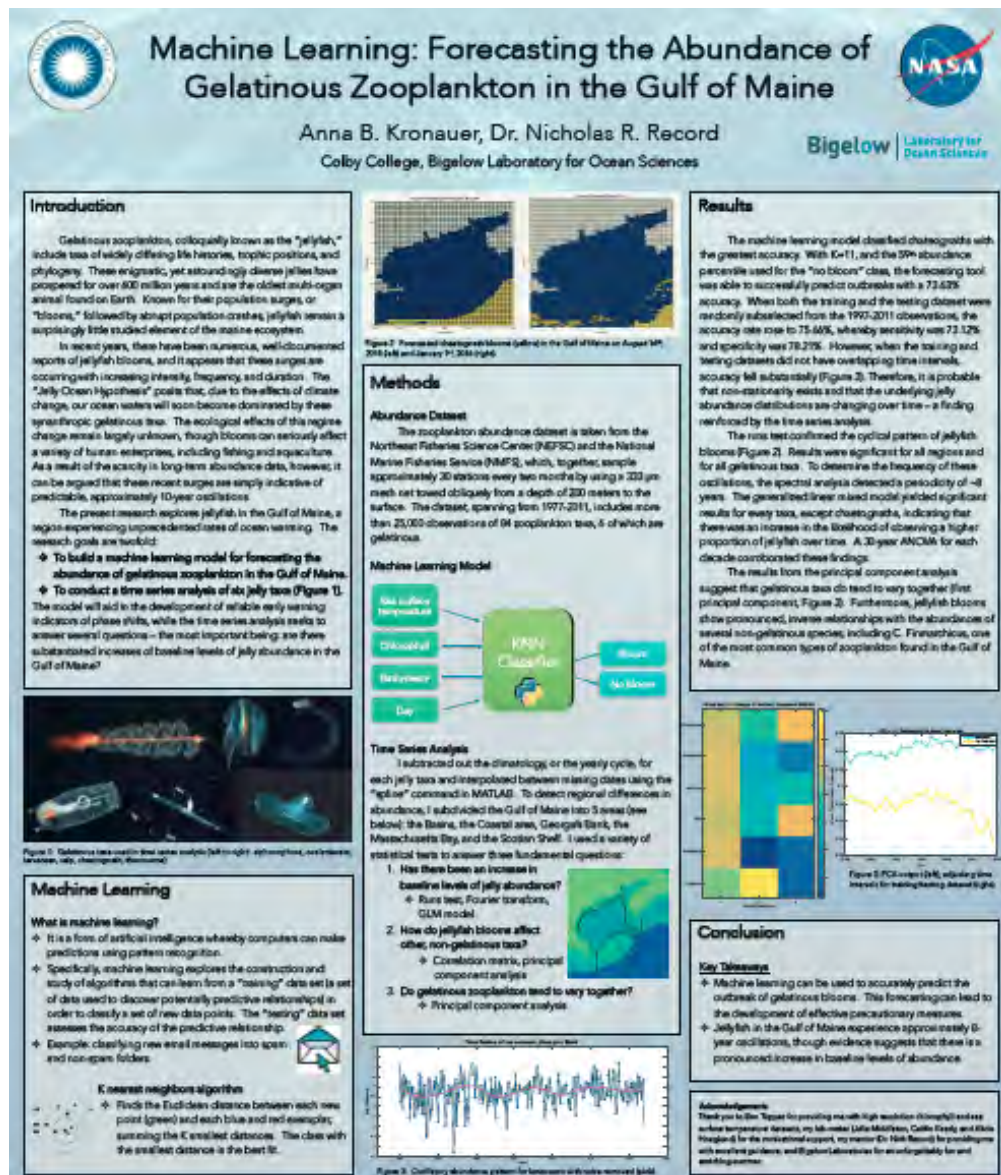
Abstracts

MACHINE LEARNING: FORECASTING THE ABUNDANCE OF GELATINOUS ZOOPLANKTON IN THE GULF OF MAINE

Kronauer AB & Record NR

Gelatinous zooplankton, colloquially known as the “jellyfish,” include taxa of widely differing life histories, trophic positions, and phylogeny. These enigmatic, yet astoundingly diverse jellies appear to be blooming with increasing intensity, frequency, and duration. The ecological effects of this phenomenon are presently unknown. As a result of the scarcity in long-term abundance data, however, it can be argued that these recent surges are simply indicative of predictable, approximately 8-year oscillations.

To achieve a greater understanding of jellyfish dynamics, the aim of the present research is two-fold: to build a machine learning model for forecasting gelatinous abundance and to conduct a time series analysis of jellies in the Gulf of Maine. The machine learning model uses the K-nearest neighbors algorithm to classify chaetognath blooms with the highest accuracy: 75.66%. Adjusting the model’s parameters revealed non-stationarity in the data – that the underlying gelatinous abundance distributions are indeed changing over time. This was reflected in the time series analysis, which detected an increase in the likelihood of observing a higher proportion of jellyfish over time.



INVESTIGATING CHLOROPHYLL-A ANOMALIES IN THE SOUTHERN OCEAN AND THE TASMAN SEA

Keady CA, Lee YJ, Record NR

The Southern Ocean is a globally important carbon sink, however its role has been weakening in recent years, possibly due to decreasing ocean primary productivity. Satellite-derived chlorophyll-*a* data are used as a proxy for phytoplankton biomass to investigate spatial and temporal patterns in high nutrient low chlorophyll (HNLC) waters proximal to Australia from 1998 to 2014. Variables such as sea ice extent, sea surface temperature, aerosol optical depth (AOD), and several global climate indices are examined using modeling techniques to find a possible relationship with chlorophyll-*a*. Both annual and interannual variability in this region could indicate changes in primary productivity and nutrient availability, which might suggest disturbances across multiple trophic levels. This study aims to explain chlorophyll-*a* anomalies, both in timing and abundance, and estimate future concentration changes in HNLC waters near Australia. Evidence is presented for a coupling between chlorophyll-*a* and both the Southern Annular Mode and AOD, which likely account for anomalies in the Austral spring and autumn, respectively.

Investigating chlorophyll-*a* anomalies in the Southern Ocean and the Tasman Sea

Keady CA^{1,2}, Lee YJ¹, Record NR¹

¹Bigelow Laboratory for Ocean Sciences, East Boothbay, ME USA; ²Bates College, Lewiston, ME, USA



Abstract

The Southern Ocean (SO) is a globally important carbon sink, however its role has been weakening in recent years, possibly due to decreasing ocean primary productivity (Johnston et al. 2010). Satellite derived chlorophyll-*a* data are used as a proxy for phytoplankton biomass to investigate spatial and temporal patterns in high nutrient low chlorophyll (HNLC) waters proximal to Australia from 1998 to 2014. Chlorophyll-*a* anomalies in this region could indicate changes in primary productivity and nutrient availability, which might suggest disturbances across multiple trophic levels. This study aims to explain chlorophyll-*a* anomalies and estimate future concentration changes in HNLC waters near Australia.

Sea Surface Temperature

In the Tasman Sea (TS) and the 55-60° S region of the Australian sector of the SO, sea surface temperature (SST) has a significant negative correlation with chlorophyll-*a* in the Austral spring, however particulate inorganic carbon (PIC), a proxy for coccolithophore biomass, shows no significant relationship with the SAM in that area. In other sectors of the SO, both chlorophyll-*a* and PIC are positively correlated with the SAM, suggesting that the SAM influences coccolithophore productivity in those regions.

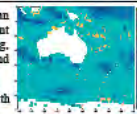


Figure 1 October-December chlorophyll-*a* correlated with August-October SST. *

Southern Annular Mode

The Southern Annular Mode (SAM) is characterized by latitudinal fluctuations in locations of the westerly wind band and high pressure regions, which suggest changes in mixed layer depth, upwelling, and nutrient availability. In the 55-65° S region of the Australian sector of the SO, the SAM is positively correlated with chlorophyll-*a* in the Austral spring, however particulate inorganic carbon (PIC), a proxy for coccolithophore biomass, shows no significant relationship with the SAM in that area. In other sectors of the SO, both chlorophyll-*a* and PIC are positively correlated with the SAM, suggesting that the SAM influences coccolithophore productivity in those regions.

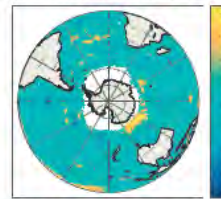


Figure 2 Correlation with September-November SAM and October-December chlorophyll-*a*. *

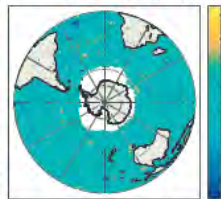


Figure 3 Correlation with September-November SAM and October-December PIC. *

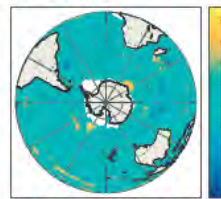


Figure 4 Correlation with November-January SAM and January-March chlorophyll-*a*. *

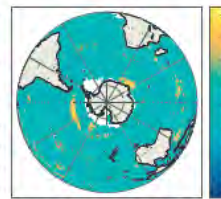


Figure 5 Correlation with November-January SAM and January-March PIC. *

Sea Ice Extent

Figure 6 Off the southeast coast of Australia, six month lagged chlorophyll-*a* concentration and April-June sea ice extent are positively correlated. Phytoplankton cells in ice are thought to stimulate productivity when the ice melts, however since ice does not reach these latitudes, this conclusion cannot be implied. *

Photosynthetically Active Radiation

Figure 7 Correlation between chlorophyll-*a* and photosynthetically active radiation (PAR) annual mean. PAR measures light available for photosynthesis, so PAR was expected to have a positive correlation with chlorophyll-*a*. Interestingly, they exhibit a negative correlation near the TS and show no significant relationship in the Australian sector of the SO. *

Wind and Currents

Figure 8 Water is driven south and east of Australia into the SO and the TS. Chlorophyll-*a* may be elevated due to currents that raise phytoplankton density in their terminal locations. The westerly wind belt pushes water and nutrients west, however its latitudinal location fluctuates with time.

Transportation of Iron Rich Dust

Aerosol Optical Depth (AOD) measures the amount of direct sunlight that is blocked from reaching earth's surface by atmospheric particles such as pollution or dust. In the Austral autumn, when light limitation and decreased stratification limit primary productivity, airborne iron-rich dust from Australia may boost ocean productivity and initiate a second bloom. It has been suggested that the main pathways for dust are off the south and southeast coasts of Australia (Bhattachan et al., 2014)

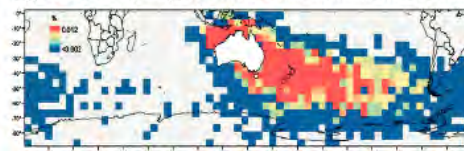


Figure 9 Distribution of the terminal location of 7-day forward dust trajectories from 1999-2009; from Bhattachan et al. (2014)

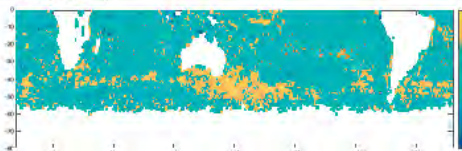


Figure 10 Correlation between AOD and chlorophyll-*a* in the Austral autumn (April-June). *

References

Bhattachan, A. and D'Odorico, P. (2014). Can land use intensification in the Mallee, Australia increase the supply of soluble iron to the Southern Ocean? *Sci. Rep.* 4, doi: 10.1038/srep06009
Johnston, B. M. and Gabric, A. J. (2011). Interannual variability in estimated biological productivity in the Australian sector of the Southern Ocean in 1997-2007. *Tellus B* 63: 266-286 doi: 10.1111/j.1600-0889.2011.00536.x
NOAA Climate Prediction Center. <http://www.cpc.ncep.noaa.gov>
NOAA Earth System Research Laboratory. <http://www.esrl.noaa.gov>
Satellite data merged by ACRI-ST <http://hermes.acri.fr/index.php?class=archive>

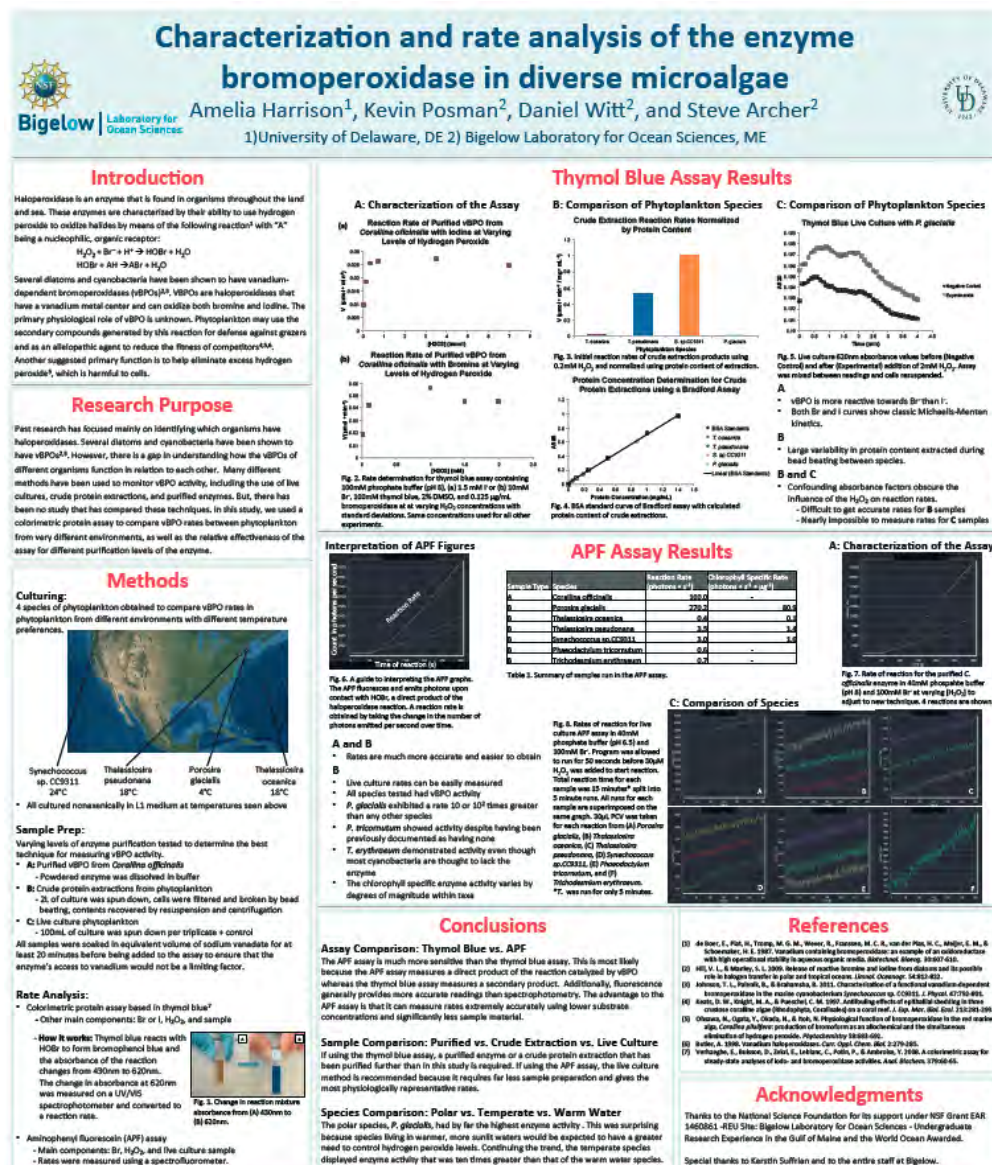
Acknowledgements

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*Note that only significant correlations ($p < 0.05$) are shown. Color bar indicates r -value.

Harrison AH, Posman KM, Witt DP, Archer SD

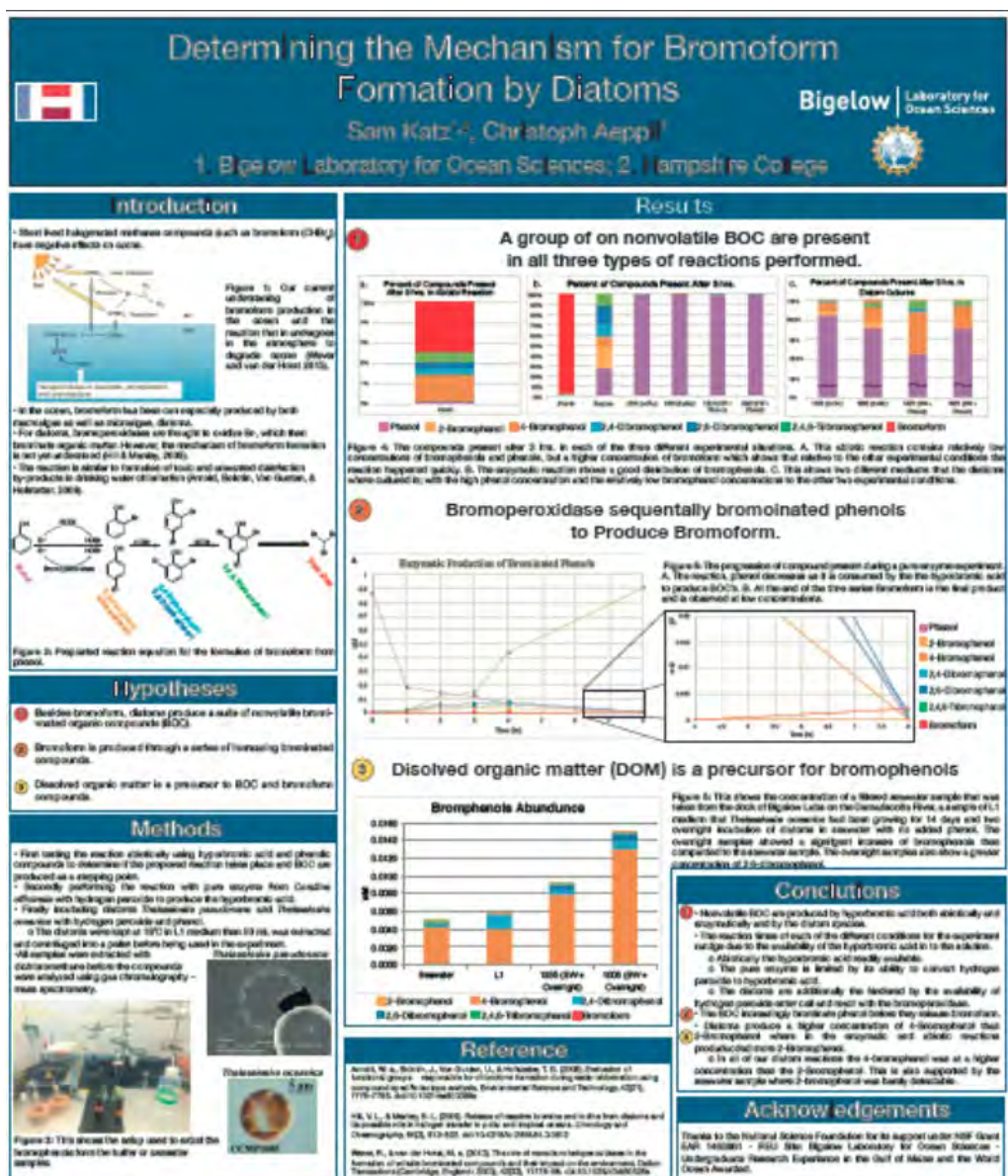
Vanadium-dependent bromoperoxidases (vBPOs) are a class of enzymes that use hydrogen peroxide (H_2O_2) to oxidize bromide and iodide. Several diatoms and cyanobacteria have been shown to have vBPOs, possibly to control H_2O_2 levels. While past research has focused on identifying organisms as having vBPOs, this study addresses the gaps in knowledge concerning differences in vBPOs function between organisms and compares techniques to measure activity rates. The effectiveness of a spectrophotometric thymol blue assay was compared to a spectrofluorometric aminophenyl fluorescein (APF) assay. The rates of several phytoplankton species from different environments were compared by normalizing to chlorophyll and protein content. Three different levels of sample purification were also tested: purified enzyme, crude protein extraction, and live culture. This is the first demonstration of the APF approach applied to microbial phytoplankton. It was the more sensitive and accurate assay and was ideal for live culture measurements. The thymol blue assay was only useful for more purified forms of the enzyme. All species tested showed vBPO activity. The polar species had the most activity while the warmest water species had the least.



DETERMINING THE MECHANISM FOR BROMOFORM FORMATION BY DIATOMS

Katz S & Aeppli C

Oceanic emissions of bromoform from macroalgae and diatoms have a critical impact on stratospheric ozone destruction. Here, we investigate the bromination mechanism of diatoms, which is not fully understood. Diatoms are hypothesized to contain bromoperoxidase, which use hydrogen peroxide to produce brominating species that ultimately lead to bromoform. We hypothesize that it reacts similarly to the abiotic formation of toxic and unwanted disinfection by-products in drinking water chlorination. The reaction was tested using three different conditions: abiotically, enzymatically with a pure bromoperoxidase, and biotically with two diatom cultures *Thalassiosira pseudonana* and *Thalassiosira oceanica*. All samples were incubated for 3 to 48 hours with phenol or seawater containing dissolved organic matter (DOM) and extracted with dichloromethane before being analyzed using gas chromatography-mass spectrometry. Besides bromoform, all three systems produced a suite of nonvolatile brominated organic compounds (BOC) that are consistent with sequential brominated of moieties of DOM. In the enzymatic reaction, the formation and degradation of certain BOC were observed before bromoform started to be released. During the diatom experiments with DOM, BOCs were significantly more abundant than the concentrations found in filtered seawater used for this experiments. Overall, this study shed light on the complex formation and degradation of BOC.



HEMOCYTE RESPONSE TO SECONDARY INFECTION OF DERMO DISEASED OYSTERS

Morefield RD, Poulton N, Fernández-Robledo JA

Perkinsus marinus (named Dermo) is a highly infectious parasite of the eastern oyster (*Crassostrea virginica*). Dermo is passively ingested by the oyster where it infects the blood cells (hemocytes) that have defensively phagocytosed the pathogen. Dermo resists phagocytotic destruction chemically and modulates apoptosis of the hemocyte cell. Recent studies have shown the prevalence of Dermo diseased oysters has risen significantly in Maine USA. The same study shows that infected oysters are 1.7 times as likely to carry the human and animal pathogens *Toxoplasma gondii* and *Cryptosporidium parvum*. Dermo's ability to modulate hemocytic function coupled with the significant secondary infections of Dermo diseased oysters raises the need for insight into Dermo infected hemocytes and their phagocytic response to secondary infection. This study attempts to answer whether Dermo prevents infected hemocytes from phagocytosing other non-*Perkinsus* particles. Flow cytometry, a GFP expressing mutant *P. marinus* (*P. marinus* MOE (MOE):GFP) and PerCP fluorescent microbeads were utilized in conjunction with in-vitro incubation of hemocytes to facilitate measurable phagocytosis. This study developed procedural protocol and achieved preliminary data of this parasite/host interaction.

Hemocyte response to secondary infection of Dermo diseased oysters

Robert D. Morefield^{1,2}

Bigelow¹ Laboratory for Ocean Sciences

Mentors: Nicole Poulton¹ and José A. Fernández-Robledo¹
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² Southern Maine Community College, 2 Fort Road, South Portland, ME 04106



Abstract

Perkinsus marinus is a highly infectious parasite of the eastern oyster (*Crassostrea virginica*) and is the causative agent of "Dermo" disease. *Perkinsus marinus* is passed by the oyster where it infects the blood cells (hemocytes) that have defensively phagocytosed the pathogen. *Perkinsus marinus* resists phagocytotic destruction chemically and modulates apoptosis of the hemocyte cell. The prevalence of Dermo diseased oysters has risen significantly in Maine USA. The same study shows that infected oysters are 1.7 times as likely to carry the human and animal pathogens *Toxoplasma gondii* and *Cryptosporidium parvum*. Dermo's ability to modulate hemocytic function coupled with the significant secondary infections of Dermo diseased oysters raises the need for insight into Dermo infected hemocytes and their phagocytic response to secondary infection. This study attempts to answer whether Dermo prevents infected hemocytes from phagocytosing other non-*Perkinsus* particles. Flow cytometry, a GFP expressing mutant *P. marinus* (*P. marinus* MOE (MOE):GFP) and PerCP fluorescent microbeads were utilized in conjunction with in-vitro incubation of hemocytes to facilitate measurable phagocytosis. This study developed procedural protocol and achieved preliminary data of this parasite/host interaction.

Introduction

Perkinsus marinus is a marine protistan parasite that causes "Dermo" disease in marine mollusks resulting in heavy losses, especially in the eastern oyster (*Crassostrea virginica*) (Pillay, 1996). *Perkinsus marinus* is found in the water column where they passively gain entry into the oyster via their feeding. Once inside the oyster, *P. marinus* is engulfed by granulocyte cells (blood cells). Granulocyte attempt to destroy *Perkinsus* using reactive oxygen species (ROS) (Pillay, et al., 2003).

Hypothesis

Once *Perkinsus marinus* trophozoites have been phagocytosed by the oyster granulocyte, these granulocytes are prevented from engulfing other non-*Perkinsus* particles (secondary infection). Hence, immunocompromising the oyster and making it more susceptible to other water born pathogens.

Objectives

- Validate the use of *P. marinus* mutant expressing green fluorescent protein (PmMOE:GFP) for in vitro flow cytometric studies.
- Optimize the detection of *P. marinus*, fluorescent microbead and oyster hemocyte populations using flow cytometry for in vitro studies.
- Developing and optimizing the protocol for in vitro challenge. Defining the gating for flow cytometry evaluation of the hypothesis.

Materials

- Perkinsus oyster**: mutant expressing GFP (PmMOE:GFP; PmMOE) (Fernández-Robledo et al., 2006; Shubbar et al., 2013). Culture were maintained in Dulbecco modified Eagle's (DMEM) supplemented with 10% fetal bovine serum (FBS) in 25 cm² (54 ml) polystyrene coated flask with vent cap (Corning), Corning, NY) in a 36-10° C. Incubator (ThermoFisher Scientific, MA).
- Oysters**: Crassostrea virginica (1 year old) from Wiscasset, Maine Island Oyster, were kept in a continuous flow seawater tank.
- Fluorescent microbeads**: SPHERO[®] 6.5 µm 3.4 µm particles (Spherotech, LLC, USA).
- Cell Sorting**: Using a Becton Dickinson FACS Aria III flow cytometer.
- Oyster hemocyte isolation**: Oyster hemocytes were isolated using a Dremel rotary tool and used to inoculate for 24h hemocyte suspension using a 200 µm mesh.
- Flow cytometry**: Routine analysis was obtained using a Becton Dickinson FACS Aria III flow cytometer.

Methods

Cell Sorting: Using a Becton Dickinson FACS Aria III flow cytometer. PmMOE:GFP culture was sorted for selecting the trophozoites with the highest fluorescence.

Oyster hemocyte isolation: Oyster hemocytes were isolated using a Dremel rotary tool and used to inoculate for 24h hemocyte suspension using a 200 µm mesh.

Flow cytometry: Routine analysis was obtained using a Becton Dickinson FACS Aria III flow cytometer.

Results

Protocol development

(1) Vortex (2) Fixation (3) Anti-coagulant (4) NI microbead (5) Cell dissociation (6) Phagocytosis inhibitor (7) Flow cytometry

Protocol development using flow cytometry

The initial flow cytometry results showed that the oyster hemocytes were not able to phagocytose the *P. marinus* trophozoites. The use of a phagocytosis inhibitor (PmMOE:GFP) was used to inhibit the phagocytosis of *P. marinus* trophozoites by the oyster hemocytes.

Flow cytometry results

The flow cytometry results showed that the oyster hemocytes were not able to phagocytose the *P. marinus* trophozoites. The use of a phagocytosis inhibitor (PmMOE:GFP) was used to inhibit the phagocytosis of *P. marinus* trophozoites by the oyster hemocytes.

Results cont.

Flow cytometric evidence of phagocytosis via developed protocol

The flow cytometry results showed that the oyster hemocytes were not able to phagocytose the *P. marinus* trophozoites. The use of a phagocytosis inhibitor (PmMOE:GFP) was used to inhibit the phagocytosis of *P. marinus* trophozoites by the oyster hemocytes.

Summary

- PmMOE:GFP can be used for flow cytometric evaluation.
- Oyster hemocyte populations were successfully separated via FC.
- Protocols were developed to study the *P. marinus* and oyster granulocyte interaction via FC.
- Biological sampling method for in vitro challenges of oyster hemocyte was optimized.
- Protocols were developed for in vitro challenges of *P. marinus* and hemocyte cells.
- Flow cytometric gating parameters and procedure were defined for in vitro challenges of the hypothesis.

Future Directions

- Utilize cell sorting to validate regions identified by flow cytometry.
- Investigate methods for reduction of bead adhesion or use an alternate bead with dual laser flow cytometry.
- Increase the number of replicates using the optimized protocol.
- Develop an optimal protocol for use of oyster hemocytes in active for research concerning other parasite/host interactions.

References

Amorim, J. A., et al. (2013). The oyster granulocyte: a review of its biology and role in the oyster immune system. *Journal of Invertebrate Pathology*, 101, 1-10.

Amorim, J. A., et al. (2014). The oyster granulocyte: a review of its biology and role in the oyster immune system. *Journal of Invertebrate Pathology*, 101, 1-10.

Amorim, J. A., et al. (2015). The oyster granulocyte: a review of its biology and role in the oyster immune system. *Journal of Invertebrate Pathology*, 101, 1-10.

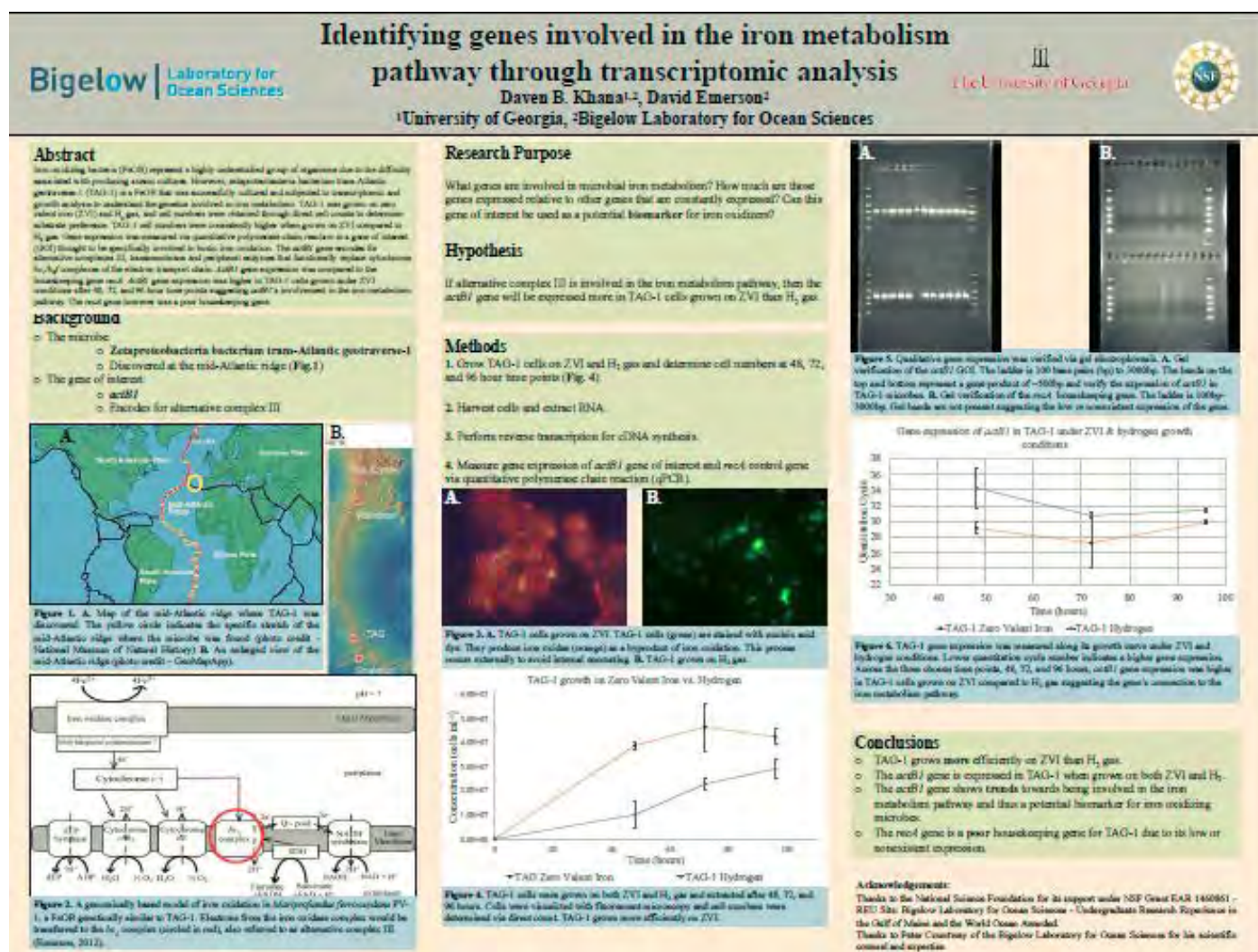
Acknowledgements

The authors thank the Bigelow Laboratory for Ocean Sciences for providing the facilities and equipment for this study.

IDENTIFYING GENES INVOLVED IN THE IRON METABOLISM PATHWAY THROUGH TRANSCRIPTOMIC ANALYSIS

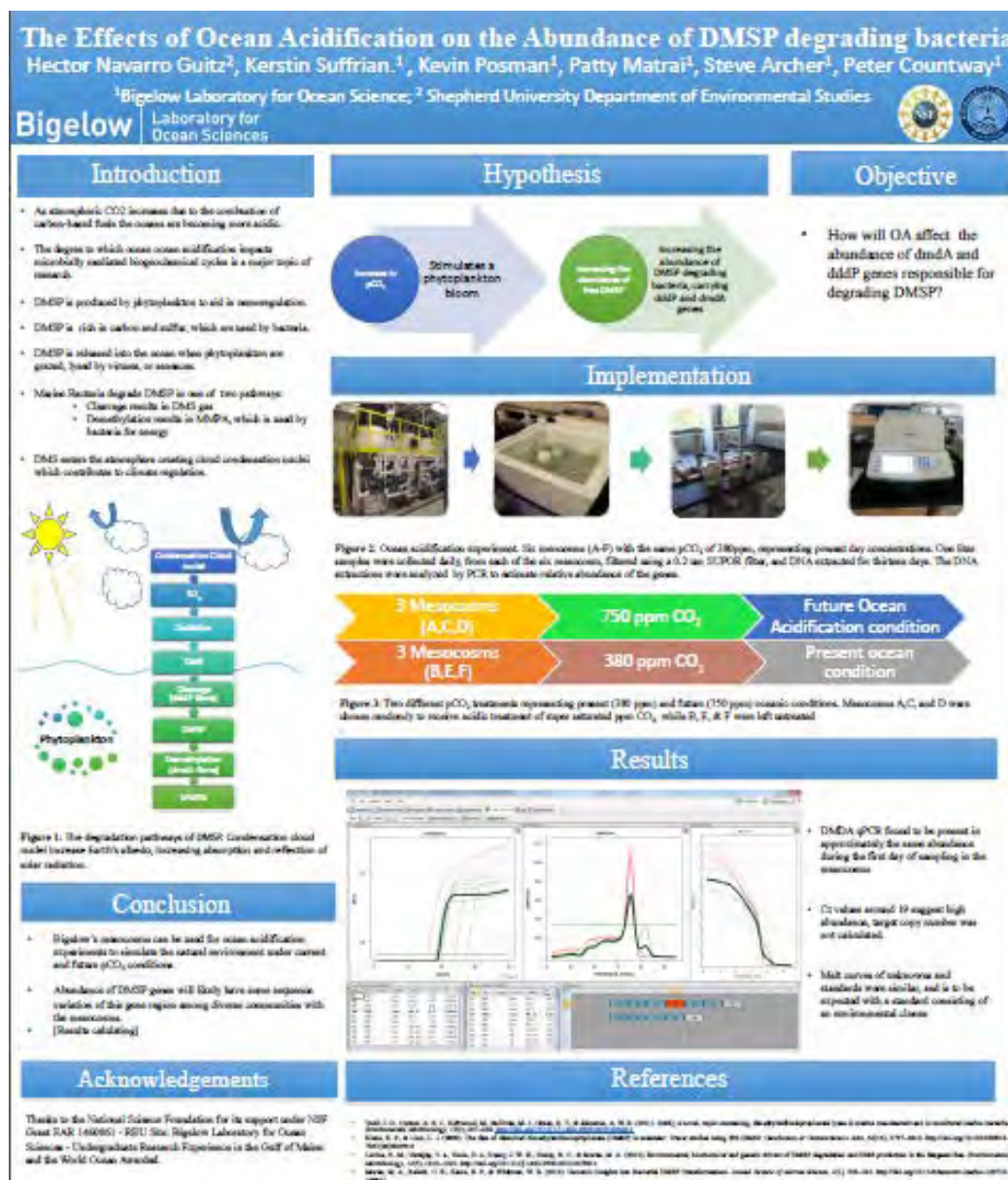
Khana D & Emerson D

Iron oxidizing bacteria (FeOB) represent a highly understudied group of organisms due to the difficulty associated with producing axenic cultures. However, zetaproteobacteria bacterium trans-Atlantic geotraverse-1 (TAG-1) is a FeOB that was successfully cultured and subjected to transcriptomic and growth analysis to understand the genetics involved in iron metabolism. TAG-1 was grown on zero valent iron (ZVI) and H_2 gas, and cell numbers were obtained through direct cell counts to determine substrate preference. TAG-1 cell numbers were consistently higher when grown on ZVI compared to H_2 gas. Gene expression was measured via quantitative polymerase chain reaction in a gene of interest (GOI) thought to be specifically involved in biotic iron oxidation. The *actB1* gene encodes for alternative complexes III, transmembrane and peripheral enzymes that functionally replace cytochrome *bc₁/b₆d* complexes of the electron transport chain. *ActB1* gene expression was compared to the housekeeping gene *recA*. *ActB1* gene expression was higher in TAG-1 cells grown under ZVI conditions after 48, 72, and 96 hour time points suggesting *actB1*'s involvement in the iron metabolism pathway. The *recA* gene however was a poor housekeeping gene.



Navarro Guitz H, Suffrian K, Posman K, Matrai P, Archer S, Countway P

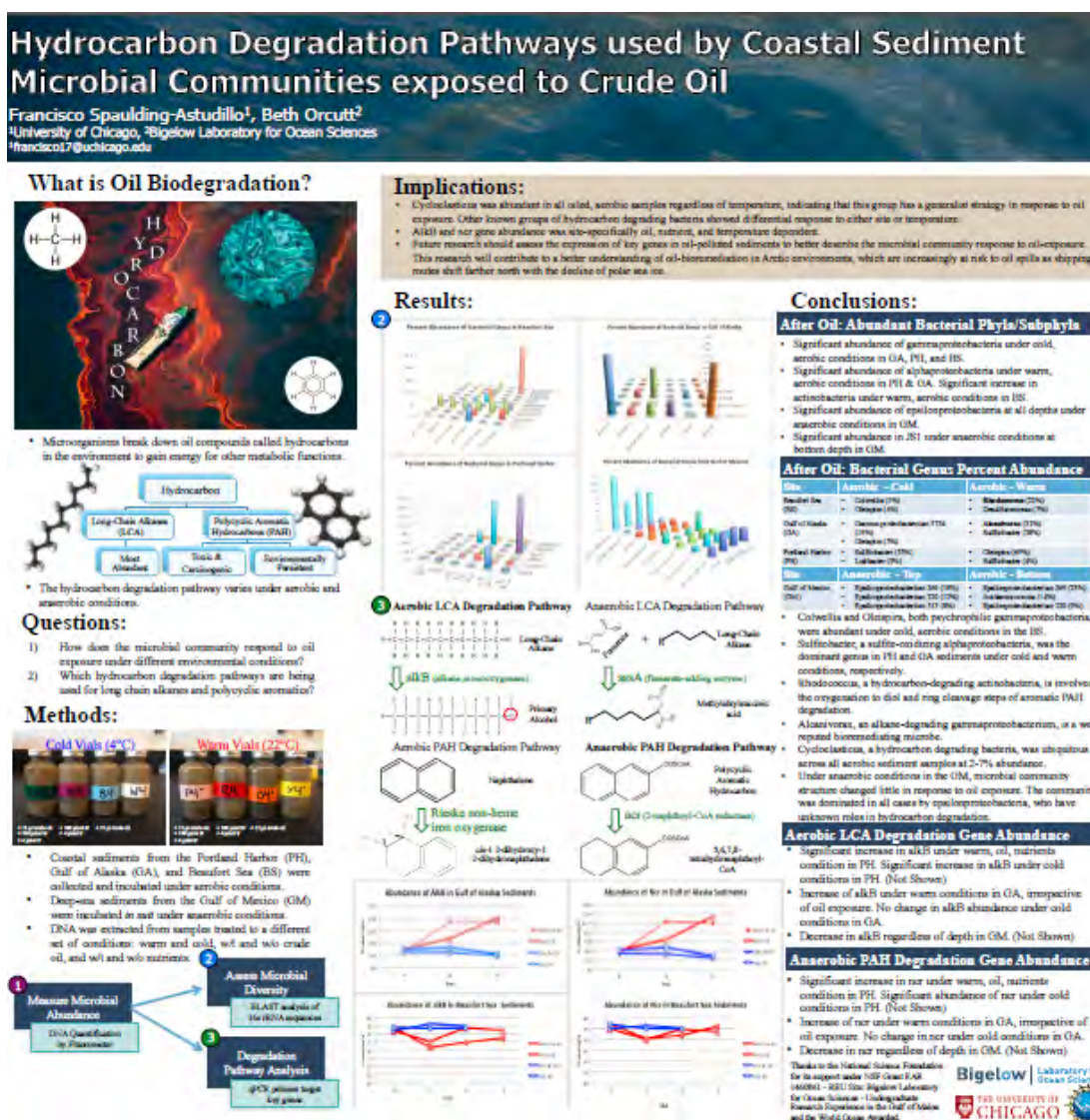
Dimethylsulfoniopropionate is a compound produced by phytoplankton and is an important of biogenic sulfur that compound that impacts microbial mediated biogeochemical cycles. DMSP is quickly metabolized by marine bacteria either through cleavage to DMS (dimethyl sulfide) or demethylation to MMAAP. The abundance of the genes encoding bacteria cleavage (dddP) and demethylation (dmdA) was measured in six mesocosms for twelve days. Measurements were taken daily, after a complete water column mixing to ensure that a representative sample was collected. Super concentrated CO₂ salt water was added to each mesocosm to raise the amount of CO₂ from present (380ppm) to future (750 ppm), to concentrations of the year 2100. Using qPCR, abundance of DMSP degrading bacteria was estimated from the abundance of the genes dddP and dmdA. Environmental standards (cloned PCR products), from West Boothbay, ME, were used to calibrate mesocosm samples in order to calculate gene copy number in experimental samples and to approximate the response of DMSP- degrading bacteria to ocean acidification.



HYDROCARBON DEGRADATION PATHWAYS USED BY COASTAL SEDIMENT MICROBIAL COMMUNITIES EXPOSED TO CRUDE OIL

Spaulding-Astudillo FE & Orcutt BN

The site-specific microbial community response to crude oil exposure in marine environments is not well described. Moreover, the abundance of genes implicated in long-chain alkane (LCA) and polycyclic aromatic hydrocarbon (PAH) degradation are not well understood. Coastal sediments from the Beaufort Sea, Gulf of Alaska, and Portland Harbor were treated with crude oil and incubated aerobically. Deep-sea sediments from the Gulf of Mexico were treated with the same crude oil and anaerobically incubated *in situ* for five months before recovery. *Cycloclasticus*, a known hydrocarbon-degrader, was abundant in all oiled, aerobic samples regardless of temperature, demonstrating a generalist oil-response strategy. Other hydrocarbon degrading bacteria showed differential response to either site or temperature. Primers for *alkB* and *ncr*, catabolic gene markers for aerobic LCA degradation and anaerobic PAH degradation, respectively, were found in literature and tested on DNA extracts in a QPCR-based assay. *AlkB* and *ncr* gene abundance was site and condition variable.



OPTIMIZING THE CRYOPRESERVATION OF PAVLOVA SP WITH SPECIAL ATTENTION TO LIPIDS

Hayden LC, Sexton J, Lomas MW

Cryopreservation is stable, efficient and commonly used to maintain microalgal collections. Unfortunately the process is often injurious to cells requiring the use of cryoprotective agents (CPA) such as dimethylsulphoxide (DMSO) which can also be injurious. *Pavlova sp.*, a high lipid golden algae, has been successfully cryopreserved without the use of a CPA at the National Center for Marine Algae and Microbiota at Bigelow Laboratory. *Pavlova sp.* is economically important as aquaculture feed and is a source of healthful polyunsaturated fatty acids. Hypothesizing that lipids provide protection against cryoinjury we grew six strains of *Pavlova sp.* at two light levels and cryopreserved each with and without CPA in lag, log and stationary growth stages. Population growth was followed by daily fluorometer readings and we used Bodipy fluoresce to quantify lipid content via flow cytometry as a proxy for lipid count. Most samples survived cryopreservation without CPA, but they are growing back more slowly than those with CPA, with success mostly strain dependent. Suggested future work includes scaling up culture size to provide higher cell densities at each growth stage and thorough lipid analysis.

Optimizing the cryopreservation of *Pavlova sp.*

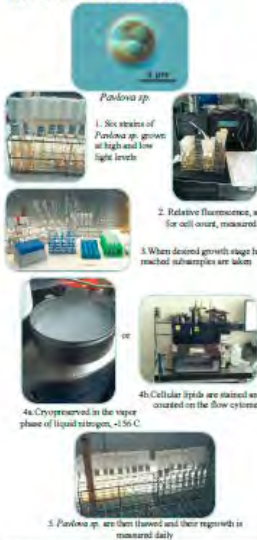
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Introduction

Cryopreservation, storage at ultra cold temperatures, is of great practical advantage in culture collections. The National Center for Marine Algae and Microbiota (NCMA) at Bigelow Laboratory maintains roughly half of their 1,000 strains in cryopreservation. It is archival, genetic changes and mutations which occur in continuous culture are greatly reduced, and requires the low labor. Unfortunately the process of freezing and thawing is often injurious to the cells. To reduce cell mortality cryoprotective agents (CPA) such as Dimethylsulfoxide (DMSO) are added. The trouble is that DMSO is toxic and can be deadly to the cells at room temperature. Some prior studies (Caron et al., 1995) have suggested that some algae might be able to be cryopreserved without the use of CPA. Preliminary experiments at the NCMA have shown that several strains of microalgae, including *Pavlova sp.* CCM393, have been successfully cryopreserved without the use of CPA. *Pavlova sp.* is a high lipid golden algae that is important in aquaculture. In our project we look at several different strains of *Pavlova sp.* cryopreserving them with and without CPA. We are interested in the cryoprotective potential of lipids within the cells and how they differ at various life stages and with different light treatments.

Materials and methods



Literature cited

Caron D and Lomas M (1995) Relationship between cold stress, cryoprotective concentrations and survival in the cryopreservation of marine microalgae. *Marine Biology* 128:225-234.
Dumas G, Villanar J, Dumas G and Dumas G (1995) Changes in the lipid composition and metabolism of the photosynthetic fatty acid content of some microalgae grown in mass culture. *Journal of Applied Phycology* 5: 31-40.
Gardner C, Michaels L, Azeiteiro M and Michaels P (2010) Change in Lipid Class and Fatty Acid Composition of Cultures of *Pavlova lutheri* in Response to Light Intensity. *J. Phycol.* 46: 873-881.
Halevy Z (2001) Protocols used in the cryopreservation of microorganisms. *Cryobiology* 44: 205-226.

Results

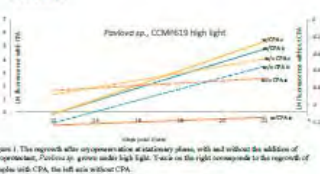


Figure 1. The regrowth after cryopreservation at stationary phase, with and without the addition of cryoprotective. *Pavlova sp.* grown under high light. Error in the right corresponds to the regrowth of samples with CPA, the left axis without CPA.



Figure 2. Photographs from flow cytometry showing on the right a population of unstained cells (yellow) and on the left a population of the same cells with the lipid stain (blue).

Table 1. Growth rates for *Pavlova sp.* strain 619 during stationary growth

Life stage	unstarved	lipid starved	delta fluorescence
lag	0.0	0.0	0.0
log	0.0	22.7	18.8
high	1.0	20	33
low	0.0	18.7	12.8
low 1	1.1	12.8	23.7
low 2	0.0	22	22.7

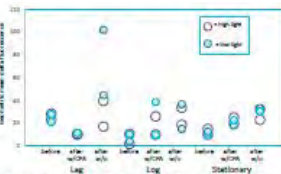


Figure 3. Data fluorescence as proxy for lipid content at three growth stages, before cryopreservation, after with and without the addition of CPA. Two replicates of each light treatment.

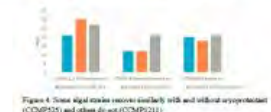


Figure 4. Some lipid strains recover similarly with and without cryoprotection (CCM393) and others do not (CCM392).

Table 2. Post-cryopreservation success of several *Pavlova sp.* strains

Strain	Life stage	CPA	Survival	Regrowth
CCM393	lag	low	100%	100%
CCM393	log	high	100%	100%
CCM393	log	low	100%	100%
CCM393	high	low	100%	100%
CCM393	low	low	100%	100%
CCM393	low 1	low	100%	100%
CCM393	low 2	low	100%	100%
CCM393	low 3	low	100%	100%
CCM393	low 4	low	100%	100%
CCM393	low 5	low	100%	100%
CCM393	low 6	low	100%	100%
CCM393	low 7	low	100%	100%
CCM393	low 8	low	100%	100%
CCM393	low 9	low	100%	100%
CCM393	low 10	low	100%	100%
CCM393	low 11	low	100%	100%
CCM393	low 12	low	100%	100%
CCM393	low 13	low	100%	100%
CCM393	low 14	low	100%	100%
CCM393	low 15	low	100%	100%
CCM393	low 16	low	100%	100%
CCM393	low 17	low	100%	100%
CCM393	low 18	low	100%	100%
CCM393	low 19	low	100%	100%
CCM393	low 20	low	100%	100%
CCM393	low 21	low	100%	100%
CCM393	low 22	low	100%	100%
CCM393	low 23	low	100%	100%
CCM393	low 24	low	100%	100%
CCM393	low 25	low	100%	100%
CCM393	low 26	low	100%	100%
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CCM393	low 93	low	100%	100%
CCM393	low 94	low	100%	100%
CCM393	low 95	low	100%	100%
CCM393	low 96	low	100%	100%
CCM393	low 97	low	100%	100%
CCM393	low 98	low	100%	100%
CCM393	low 99	low	100%	100%
CCM393	low 100	low	100%	100%

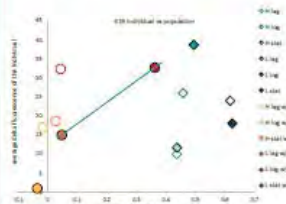


Figure 5. Comparing the lipid production per individual *Pavlova sp.* and growth rate of the whole population.

Conclusions

- Many strains of *Pavlova sp.* will survive cryopreservation without the addition of cryoprotective agents
- Regrowth after cryopreservation without CPA appears to be much slower than with CPA
- Lipid content, measured using Bodipy stain fluorescence as proxy, is different before and after cryopreservation regardless of CPA
- Some treatments without CPA show a linear correlation between individual lipid content and rate of population regrowth
- Survival is independent of light treatment
- Cell number going into cryopreservation is an important factor
- Cryopreservation is empirical

Future research

Continuing with the same *Pavlova sp.* strains from this experiment it could prove useful to grow much larger cultures, concentrating them at each growth stage and directly adding a known cell density going into cryopreservation. Also working with larger volumes we will be able to send out subsamples for thorough lipid analysis. Because different lipids are produced at different life stages it would be of great advantage and interest to see how the process of cryopreservation changes their development and how their identity influences successful cryopreservation.

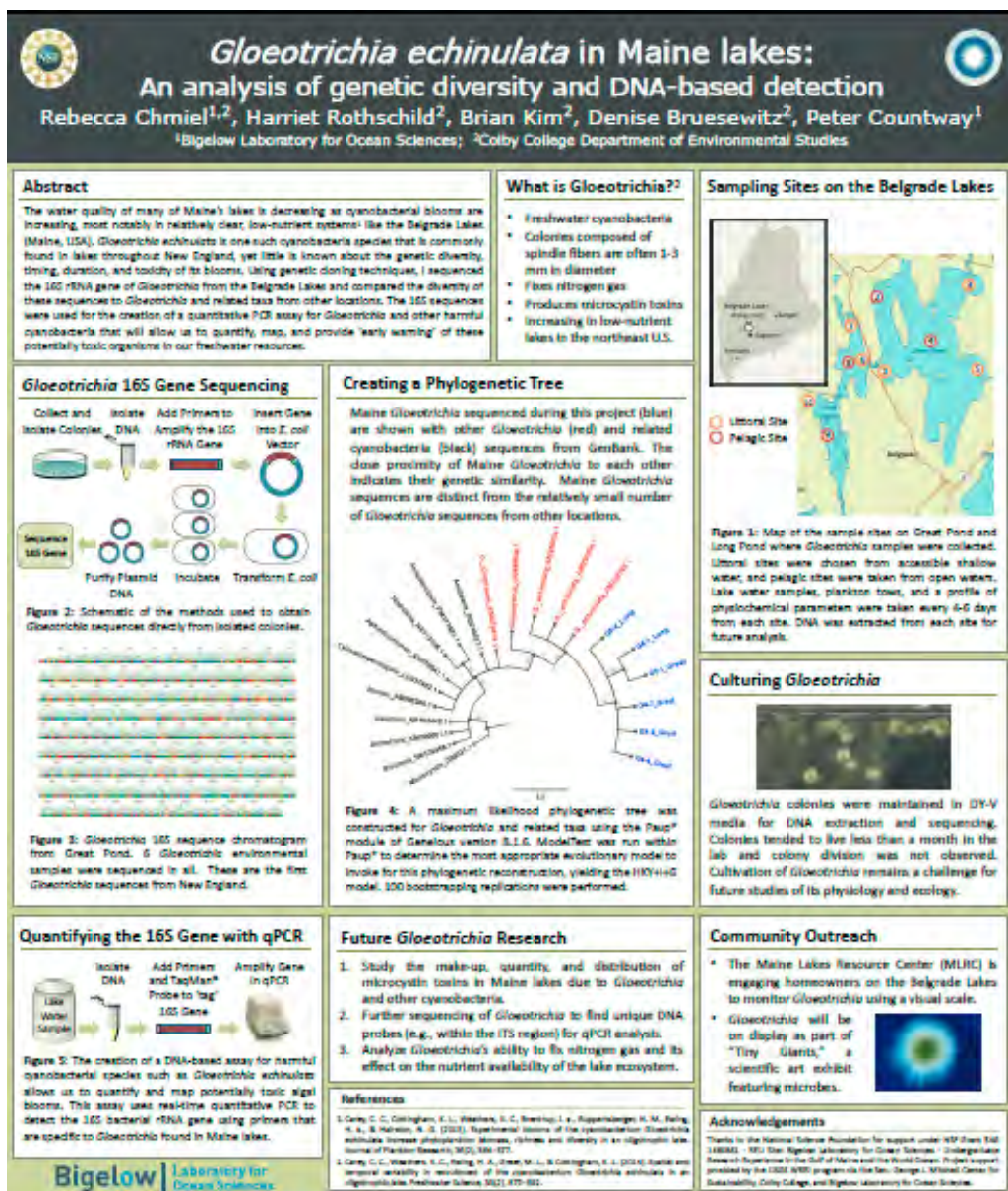
Acknowledgments

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GLOEOTRICHIA ECHINULATA IN MAINE LAKES: AN ANALYSIS OF GENETIC DIVERSITY AND DNA-BASED DETECTION

Chmiel R, Rothschild H, Kim B, Bruesewitz D, Countway P

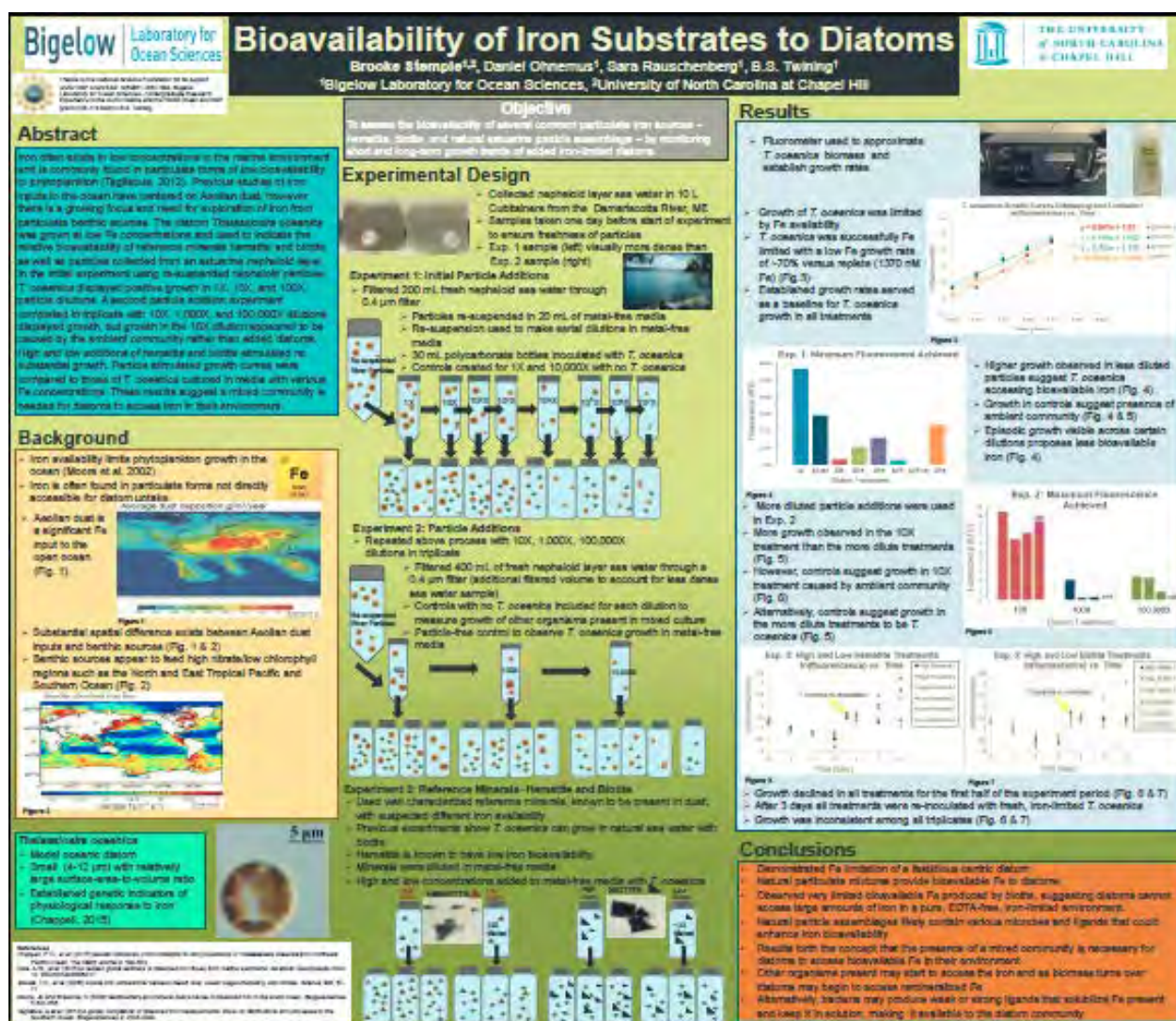
The state of Maine relies on the health and quality of its lakes, which make up a significant portion of the state's economy and public drinking water systems. However, the water quality of many of Maine's lakes is decreasing as cyanobacterial blooms are increasing, most notably in relatively clear, low-nutrient systems like the Belgrade Lakes (Maine, USA). *Gloeotrichia echinulata* is one such cyanobacteria species that is commonly found in lakes throughout New England, yet little is known about the genetic diversity, timing, duration, and toxicity of its blooms. Using genetic cloning techniques, I sequenced the 16S rRNA gene of *Gloeotrichia* from the Belgrade Lakes and compared the diversity of these sequences to *Gloeotrichia* and related taxa from other locations. The 16S sequences were used for the creation of a quantitative PCR assay for *Gloeotrichia* and other harmful cyanobacteria that will allow us to quantify, map, and provide 'early warning' of these potentially toxic organisms in our freshwater resources.



BIOAVAILABILITY OF IRON SUBSTRATES TO DIATOMS

Stemple B, Ohnemus D, Rauschenberg S, Twining BS

Iron often exists in low concentrations in the marine environment and is commonly found in particulate forms of low bioavailability to phytoplankton (Tagliabue, 2012). Previous studies of iron inputs to the ocean have centered on Aeolian dust, however there is a growing focus and need for exploration of iron from particulate benthic sources. The diatom *Thalassiosira oceanica* was grown at low Fe concentrations and used to indicate the relative bioavailability of reference minerals hematite and biotite, as well as particles collected from an estuarine nepheloid layer. In the initial experiment using re-suspended nepheloid particles, *T. oceanica* displayed positive growth in 1X, 10X, and 100X particle dilutions. A second particle addition experiment completed in triplicate with 10X, 1,000X, and 100,000X dilutions displayed growth, but growth appeared to be caused by a mixed community rather than added diatoms. High and low additions of hematite and biotite stimulated no substantial growth. Particle stimulated growth curves were compared to those of *T. oceanica* cultured in media with various Fe concentrations. These results suggest a mixed community is needed for diatoms to access iron in their environment.



HETEROLOGOUS EXPRESSION OF GENE OF INTEREST USING THE MARINE PROTOZOAN *PERKINSUS MARINUS* Cold E & Fernández Robledo JA

Perkinsus marinus is a marine protozoan parasite that causes “Dermo” disease in eastern oysters (*Crassostrea virginica*). *P. marinus* is closely related to *Plasmodium falciparum* which causes malaria. A recent study has showed that *P. marinus* causes no pathology damage but an immune response in humanized mouse providing the bases for a genetically modified *P. marinus* expressing *Plasmodium* genes to be used as a vaccination delivery system for malaria and other pathogenic diseases. A modified plasmid vector (pMOE-GFP) based on highly expressed gene tagged with green fluorescence protein and targeted to *P. marinus* cell wall was used to clone MSP8 and HAP2. MSP8 encodes for merozoite surface in *P. falciparum* and HAP2 is essential for fusion of male and female gametes; genetic disruption of the HAP2 locus revealed that parasite fertilization is prevented. Using electroporation, MSP8 and HAP2 plasmid were introduced into the *P. marinus* trophozoites. As controls pMOE-GFP was transfected into *P. mediterraneus*, *P. atlanticus* and *P. chesapeaki*. Transfection conditions included 5×10^7 *Perkinsus* trophozoites and 10 μ g of plasmid using Nucleofector® technology (D-023 program). The cells were recovered in 3 mL of *Perkinsus* culture media and transfected trophozoites were examined for green fluorescence. To facilitate subcloning of cells expressing GFP, we optimized a DME: HAM’s F12 -5% FBS -containing agar solid medium for plating *Perkinsus*. Examination of all transfected cells indicates expression of both MSP8 and HAP2. This is the first time that genes of a protozoan parasite have been expressed in a marine protozoan. At this point, transfectants are being monitoring to transient or stable and cloning using the plating methodology. The synthetic gene GP 1,2, a glycoprotein expressed in Ebola virus, is also being expressed into *Perkinsus* as a delivery system for a vaccine.

Heterologous Expression of Genes of Interest Using the Marine Protozoan *Perkinsus marinus*

Emma R. Cold ^{1,2}

Mentor: José A. Fernández Robledo ¹

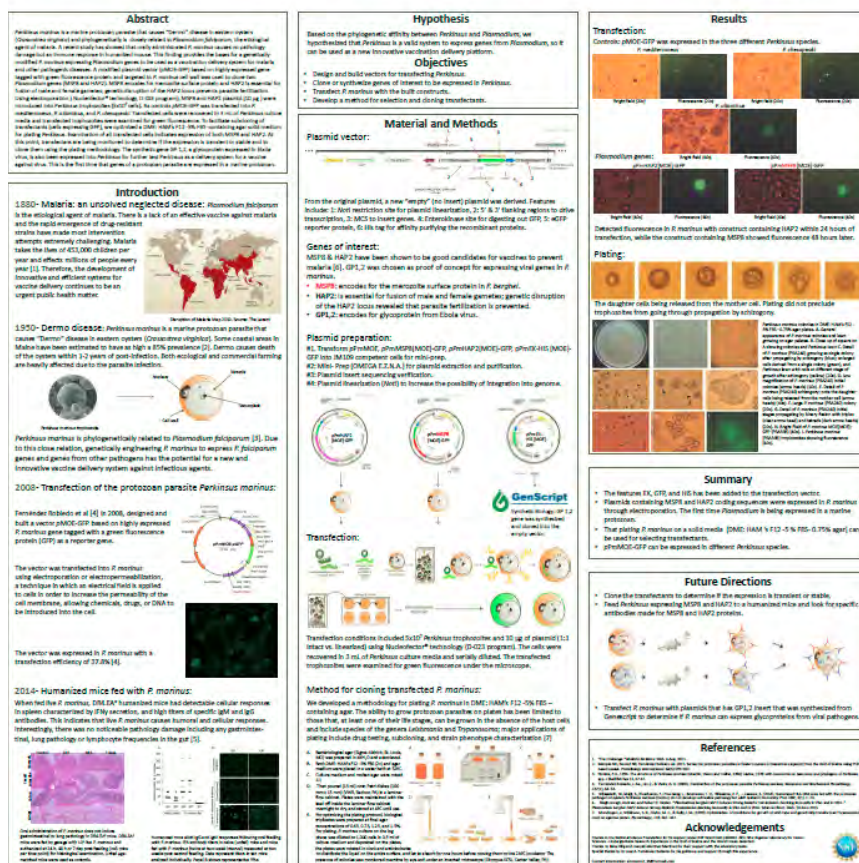
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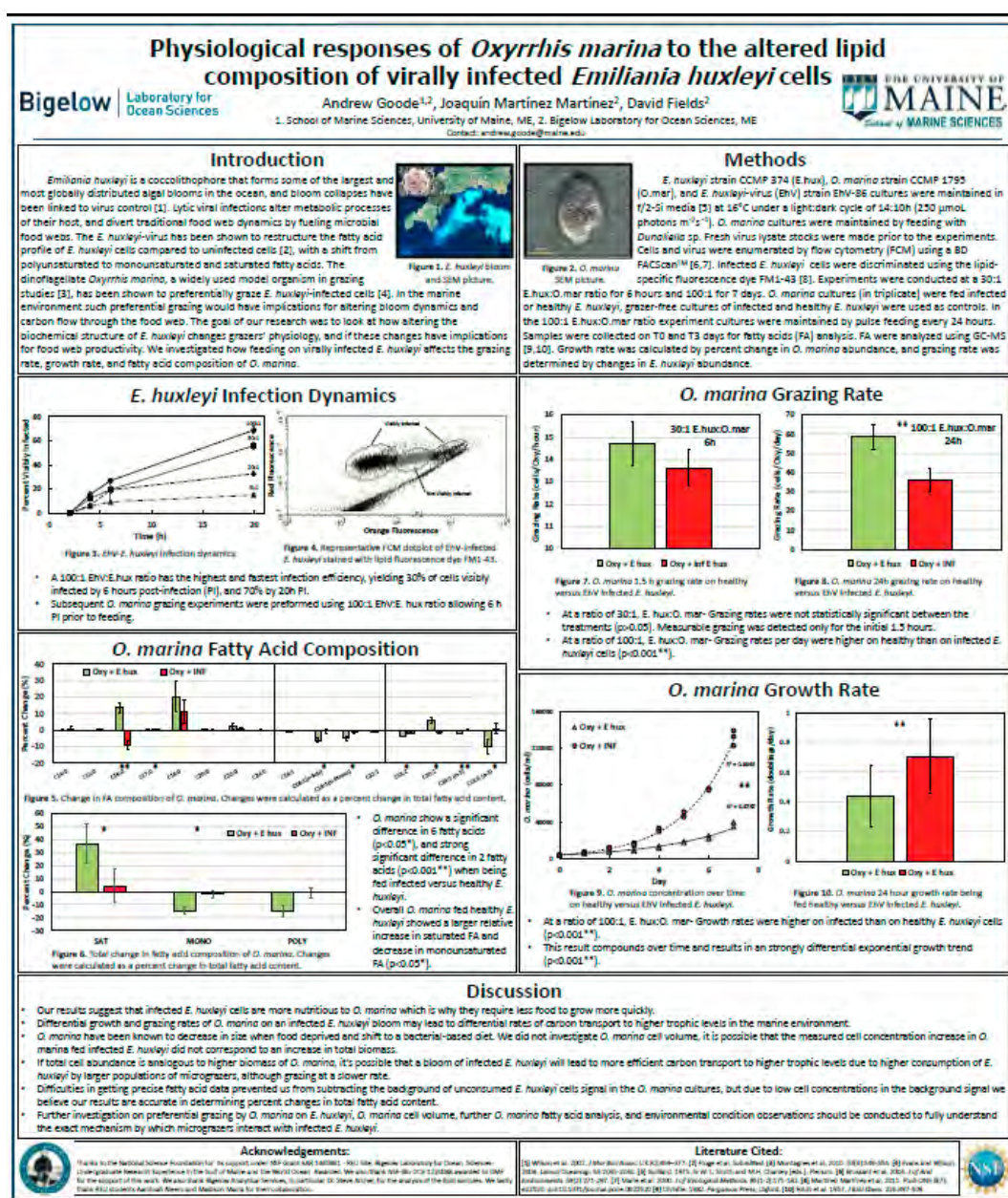
² Utah Valley University, Orem, UT 84058

UTAH VALLEY
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Goode AG, Martínez Martínez J, Fields DM






Emiliania huxleyi is a coccolithophore that form some of the largest phytoplankton blooms in the ocean. *E. huxleyi* abundance, distribution, and composition of essential fatty acids makes them a key component in various food webs. The *E. huxleyi*-virus has been shown to control the bloom duration and change the lipid composition of *E. huxleyi* cells. The alteration of essential fatty acids at the base of the food web may have downstream effects on other trophic interactions. *Oxyrrhis marina* has been studied extensively, and is used as a model organism for other micrograzers. Our experiment focuses on how virally infected *E. huxleyi* alters the physiological responses of *O. marina* and how these changes may have ecological implications. Long-term exposure of *O. marina* to high concentrations of *E. huxleyi* showed higher grazing rates on uninfected cells ($p < 0.05$), faster growth rates ($p < 0.05$), and a slower transition from monounsaturated to saturated fatty acids ($p < 0.05$) on infected cells. This suggests higher nutritional value of infected cells while also promoting larger carbon transport to higher trophic levels when blooms become infected.



THE ART OF SCIENTIFIC STORYTELLING: THREE STEPS TO COMMUNICATING YOUR SCIENCE EFFECTIVELY

Fulton AM & Crist DT

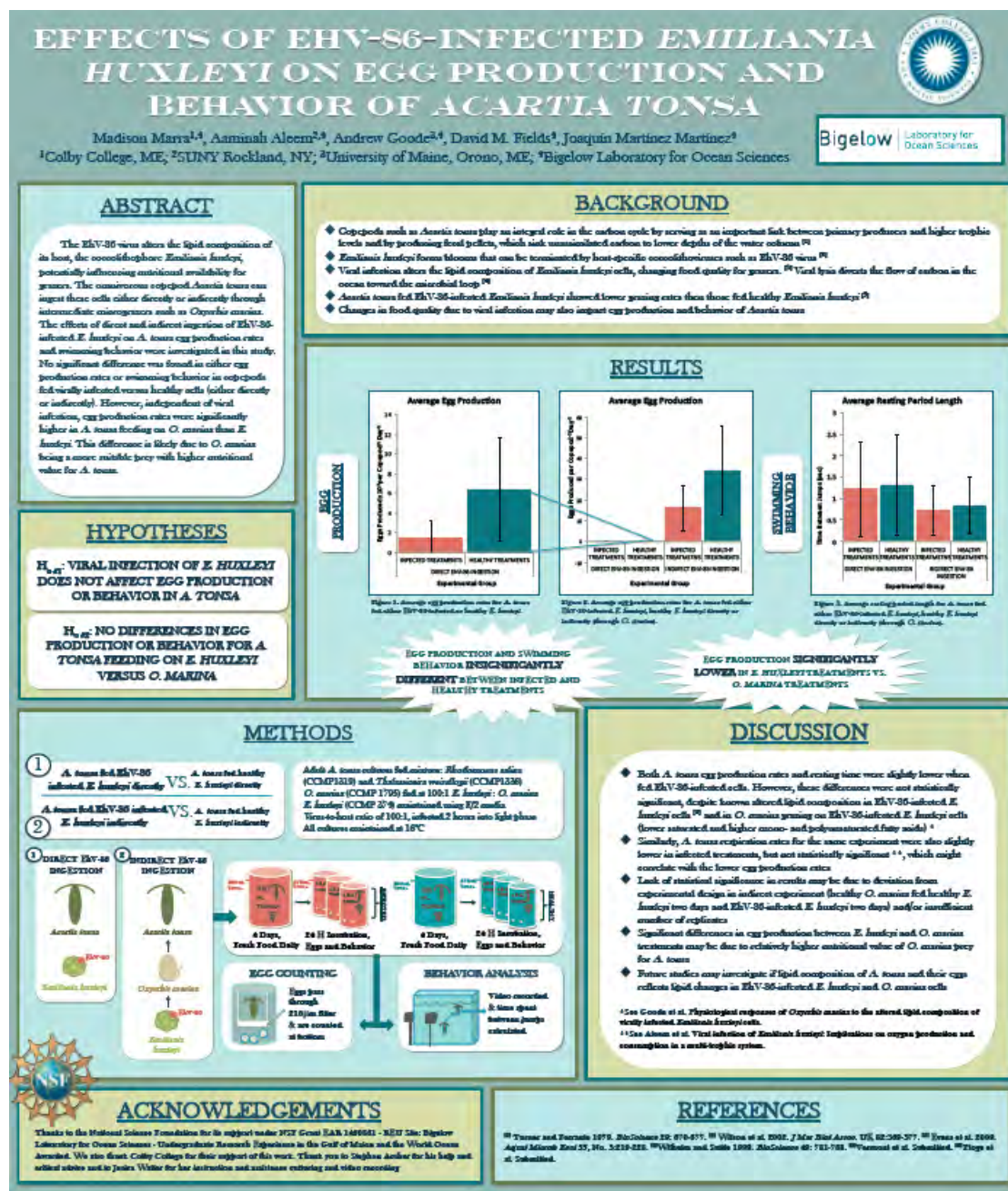
This final project consists of three steps that scientists can follow to successfully convey the importance and meaning of their research. Compiled from a series of practical tips gleaned from various literature, videos, tutorials, and articles on effective science communication, these three steps provide a roadmap to clearly, concisely, and convincingly explain research findings and their importance to a variety of audiences. The steps are: 1) to tailor how you explain your research to each audience, 2) to organize your research in a new way, and 3) to focus both on *what* you say and *how* you say it. Collectively, these steps provide the framework for scientists to successfully explain their work to a diverse range of audiences. In consideration of the endless number of ways that researchers are able to reach out to the public, the communication tips focused upon are versatile across multiple platforms.

<div>  <div> <h2>The Art of Scientific Storytelling</h2> <h3>Three Steps to Communicating your Science Effectively</h3> <p>Allyson Fulton*, Darlene Trew Crist**</p> <p><small>*Bowdoin College, Brunswick, ME **Waggoner Laboratory for Ocean Sciences, East Brunswick, ME</small></p> </div> </div>			
<h4>ABSTRACT</h4> <p>This final project consists of three steps that scientists can follow to successfully convey the importance and meaning of their research. Compiled from a series of practical tips gleaned from various literature, videos, tutorials, and articles on effective science communication, these three steps provide a roadmap to clearly, concisely, and convincingly explain research findings and their importance to a variety of audiences. The steps are: 1) to tailor how you explain your research to each audience, 2) to organize your research in a new way, and 3) to focus both on <i>what</i> you say and <i>how</i> you say it. Collectively, these steps provide the framework for scientists to successfully explain their work to a diverse range of audiences. In consideration of the endless number of ways that researchers are able to reach out to the public, the communication tips focused upon are versatile across multiple platforms.</p> <h4>WHY DOES SCIENCE COMMUNICATION MATTER?</h4> <ol style="list-style-type: none"> 1. With federal funding comes the responsibility to communicate research findings to the broader public. <ul style="list-style-type: none"> — Federal funding is the primary source of basic research support, and 10% of funds are required to be allocated to sharing results with colleagues and the general public. 2. Effective communication gives the public accurate information. <ul style="list-style-type: none"> — Approximately three-quarters of American adults are currently qualified as scientifically illiterate. A lack of understanding the scientific process may lead to a distrust of research or purposeful misinterpretations of scientific results. This may have far-reaching consequences into highly-charged topics such as climate change. — Making science accessible helps to counter misinformation and misconceptions that citizens public, explore and debate. 3. Science communication enables a creative and expanded network. <ul style="list-style-type: none"> — Involving individuals outside of a particular niche field can foster innovative collaborations and promote creative problem-solving. — Learning the basics of communicating science clearly and concisely can serve as a template for navigating everything from grant writing, social media posts, to interviews. 	<h4>STEP 1: Tailor how you explain your research to each audience</h4> <p><i>One size does not fit all.</i></p> <p>➤ Know your audience. Identify who your audience is and what they already may know about your topic. Are they non-scientists, scientists from another field, policy makers, or journalists? You will be a more effective presenter if you understand your audience and present your work in a way that they can relate to and understand.</p> <p>➤ Put your research in context. Every audience member will come to the table with different interests and concerns, so tailor how you explain your work to meet those needs. Adjust what part of your research you focus on, and present your research in the context of a larger issue that your audience is already concerned about. This will provide context for your research and serve to enhance understanding and receptivity of your message.</p> <p>➤ Adjust the answers to these three questions to the needs and concerns of your intended audience:</p> <ol style="list-style-type: none"> 1. What have you found? 2. Why does it matter? 3. What difference does your research make? 	<h4>STEP 2: Organize your research in a new way</h4> <p><i>Arouse, then fulfill.</i></p> <p>➤ Your first point should be your main point. Begin your presentation with the main point you want to make or your "key message" and construct the rest of your presentation to support that key message. This is contrary to how scientists are taught to communicate with all supporting information before presenting a conclusion.</p>  <p><small>Scientists are taught to explain their research in the manner of a formal presentation: background, introduction, main point, conclusion, supporting details. To communicate with the general public, it is more effective to flip this structure and start with the conclusion that caught your eye.</small></p> <p>➤ Have three main take-away points. When explaining concepts to an audience with little or no background in your research, providing too many details can drown out the most important points. Prepare beforehand three main points that you want your audience to grasp by the end of your presentation.</p> <p>➤ Why expend the energy to change how you communicate? Not only will reorganizing your research enhance the audience's ability to understand your message, it helps your research stand out from everyone else's.</p> 	<h4>STEP 3: Focus on <i>what</i> you are saying and <i>how</i> you are saying it</h4> <p>Communication toolbox:</p> <p>➤ Visual literacy Visualize your information to keep an audience interested, to explain complicated concepts, and to enhance the message you would like to convey. Some examples include providing your audience simple and clear visual cues to follow the steps in your message, or drawing diagrams to help explain complicated concepts and patterns.</p> <p>➤ Analogies and metaphors Make use of analogies and metaphors to illustrate the concept you are trying to explain.</p>  <p><small>The volcano is an analogy for the process of a volcano erupting. It is used to explain the concept of a volcano erupting. The volcano is a metaphor for the process of a volcano erupting. The volcano is a metaphor for the process of a volcano erupting.</small></p> <p>➤ Jargon Avoid using jargon as much as possible. If some jargon is used, explain what you mean using vocabulary the audience is comfortable with. Be cognizant of words with "double meanings," or words that are used differently by the scientific community and the general public.</p> <p>Live/Dead: cold, nutrient rich water that comes to the ocean surface.</p> <p>Uncertainty: ignorance vs. statistical range of certainty.</p> <p>Positive feedback: good response/positive vs. self-reinforcing cycle.</p> <h4>SOURCES</h4> <p><small>See: Schultz, J. (2010). "The Art of Scientific Storytelling: A Guide for Scientists." <i>Science Communication</i>, 42(1), 1-10. https://doi.org/10.1177/1075547009358888</small></p>

EFFECTS OF EHv-86-INFECTED *EMILIANIA HUXLEYI* ON EGG PRODUCTION AND BEHAVIOR OF *ACARTIA TONSA*

Marra MT, Aleem A, Goode AG, Fields DM, Martínez Martínez J

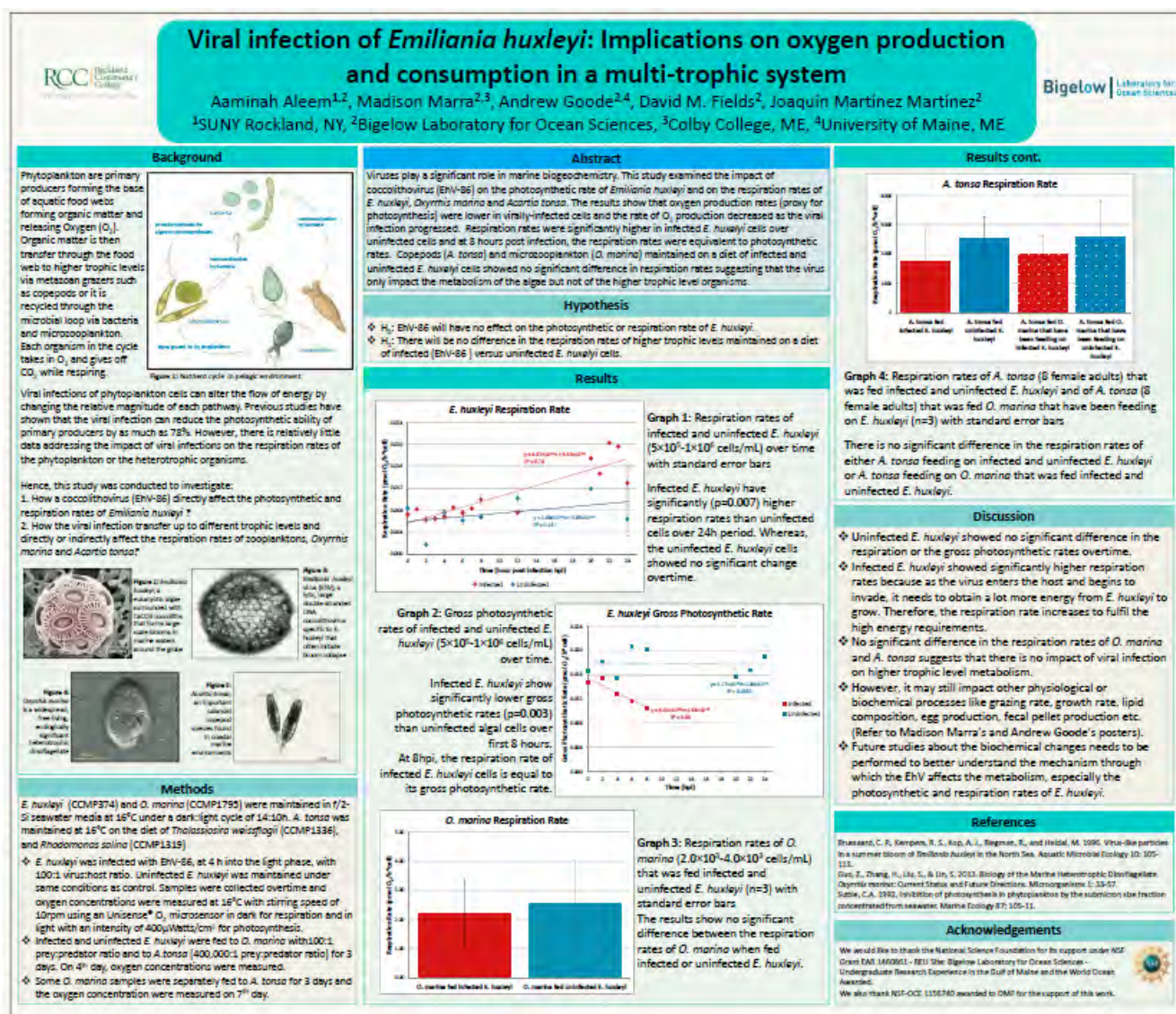
The EhV-86 virus alters the lipid composition of its host, the coccolithophore *Emiliania huxleyi*, potentially influencing nutritional availability for grazers. The omnivorous copepod *Acartia tonsa* can ingest these cells either directly or indirectly through intermediate micrograzers such as *Oxyrrhis marina*. The effects of direct and indirect ingestion of EhV-86-infected *E. huxleyi* on *A. tonsa* egg production rates and swimming behavior were investigated in this study. No significant difference was found in either egg production rates or swimming behavior in copepods fed virally infected versus healthy cells (either directly or indirectly). However, independent of viral infection, egg production rates were significantly higher in *A. tonsa* feeding on *O. marina* than *E. huxleyi*. The EhV-86 virus does not appear to affect these aspects of *A. tonsa* physiology. Differences found between *O. marina* and *E. huxleyi* treatments are likely explained by different grazing rates by *A. tonsa* due to nutritional differences between the two food sources.



VIRAL INFECTION OF EMILIANIA HUXLEYI: IMPLICATIONS ON OXYGEN PRODUCTION AND CONSUMPTION IN A MULTI-TROPHIC SYSTEM

Aleem A, Marra A, Goode A, Fields DM, Martínez Martínez J

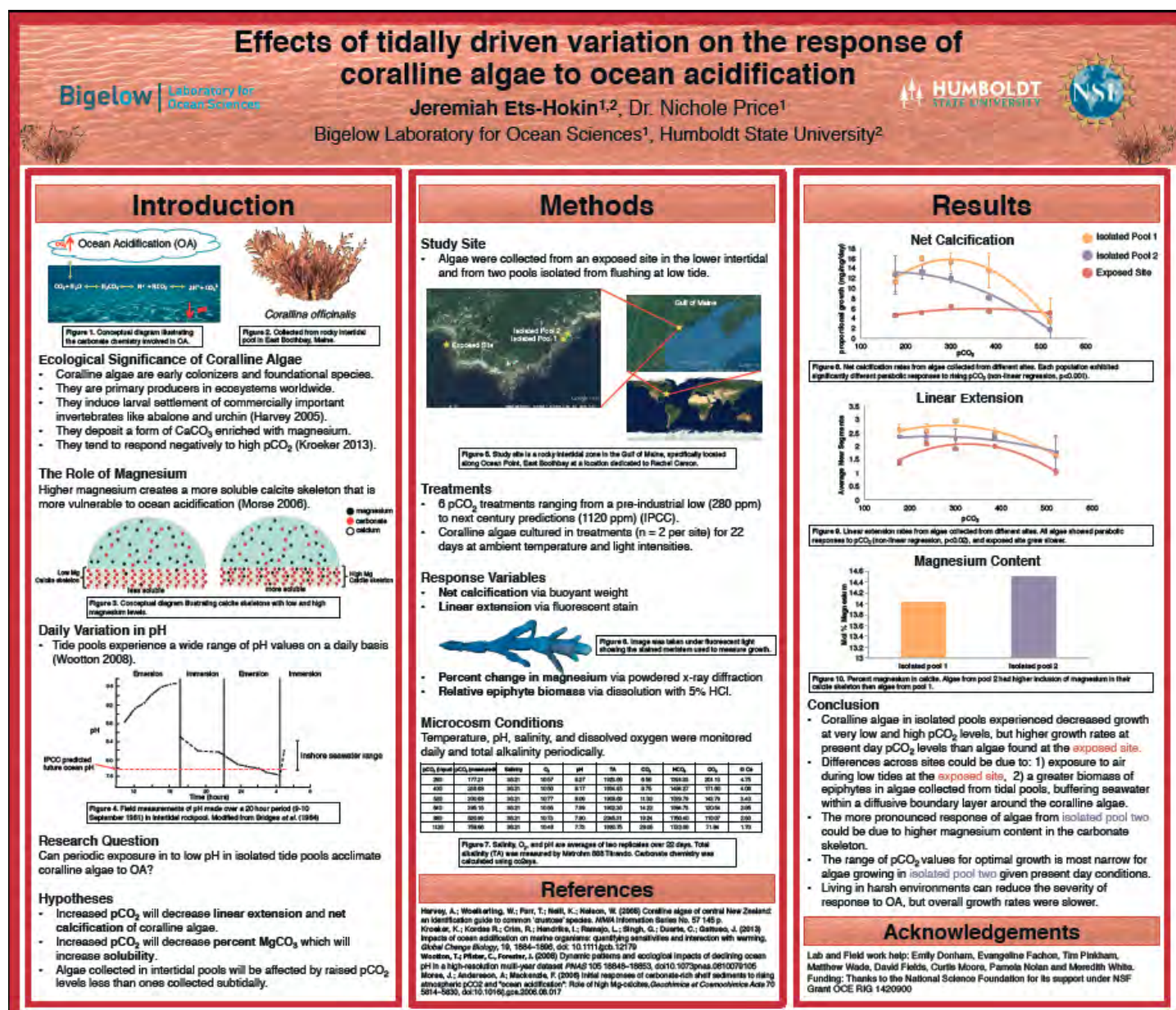
Viruses play a significant role in marine biogeochemistry, especially by altering the production and consumption of organic matter among microbial communities. This study was aimed to examine the impact of coccolithovirus (EhV-86) on the photosynthetic rate of *Emiliana huxleyi* as well as on the respiration rates of *Emiliana huxleyi*, *Oxyrrhis marina* and *Acartia tonsa*. Samples of infected (with EhV-86) and uninfected *E. huxleyi* were collected overtime. Later, both infected and uninfected *E. huxleyi* was fed to *O. marina* and *A. tonsa* as well as some of *O. marina* from each treatment was fed to a separate *A. tonsa* sample. Oxygen concentrations were measured at 16°C in the light for photosynthesis and in the dark for all respirations. For infected *E. huxleyi*, gross photosynthetic rate was significantly lower and the respiration rate was significantly higher overtime than the infected cells. However, there was no significant difference between the respiration rates of *O. marina* and *A. tonsa* among different treatments. This suggests that the virus only impact the metabolism of the algae but not of the higher trophic level organisms.



AFFECTS OF TIDALLY DRIVEN VARIATION ON THE RESPONSE OF CORALLINE ALGAE TO OCEAN ACIDIFICATION

Ets-Hokin JM & Price NN

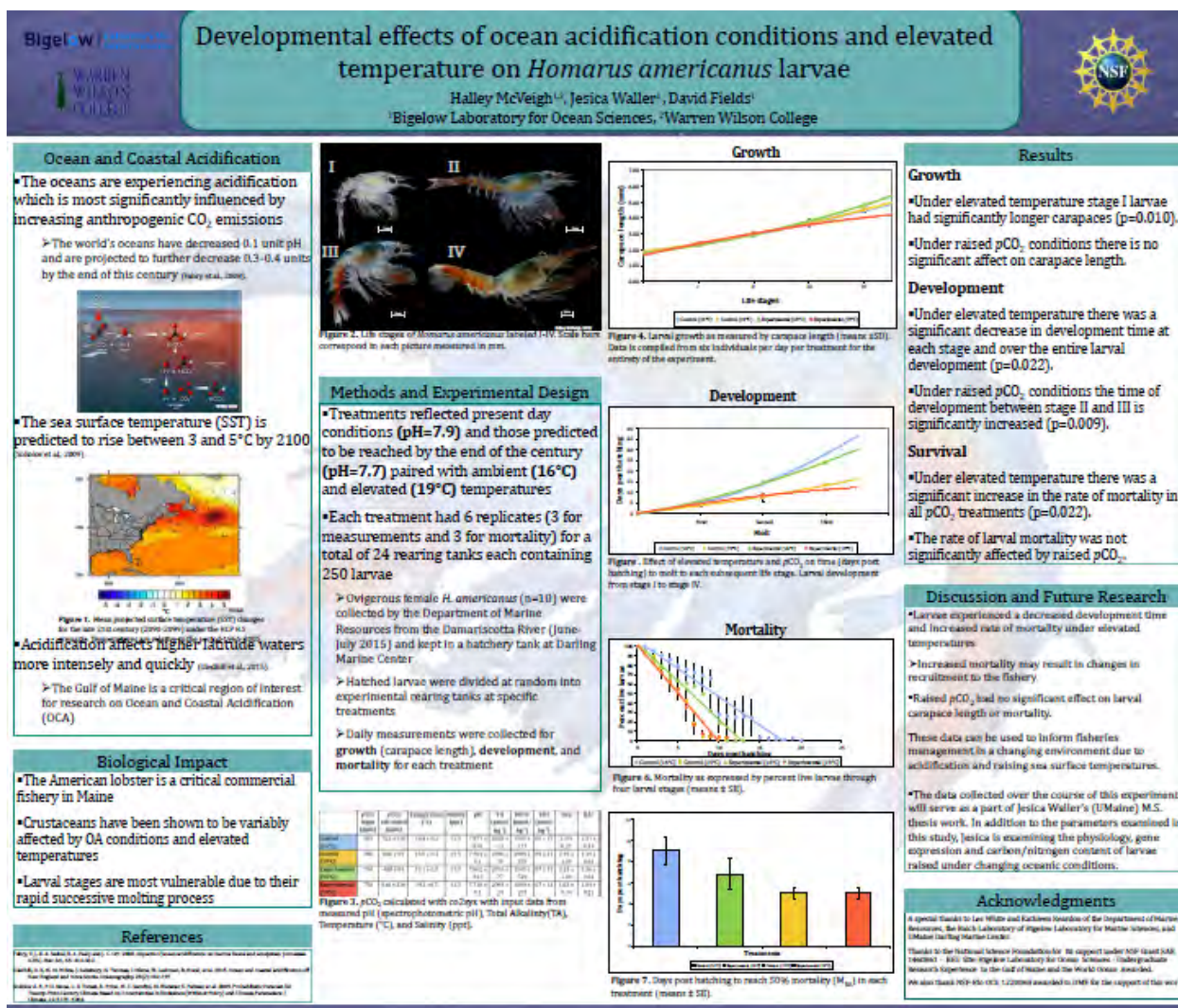
As atmospheric CO₂ levels rise, our oceans become more acidic and calcifying organisms like coralline algae are decreasing in their calcification potential. Coralline algae are early colonizers of destroyed ecosystems and help induce larval settlement of commercially important invertebrates. However, coralline algae are more susceptible to ocean acidification (OA) due to the higher magnesium content in their calcite skeleton, which makes them more soluble. Magnesium varies between individuals and therefore could be a mechanism of acclimation for algae living in harsh environments. To test this, we collected algal samples from tide pools that experience extreme tidally driven variation in pH and a site that experiences low daily variation. Samples were placed in microcosm treatments ranging from preindustrial low to a predicted next century high. Coralline algae collected in the isolated tide pools showed decreased growth in low and high pCO₂ levels compared to the exposed site that had no response to pCO₂ treatments but had lower growth overall. Overall living in harsh environments can reduce the severity of response to OA but growth rates are slower.



DEVELOPMENTAL EFFECTS OF OCEAN ACIDIFICATION CONDITIONS AND ELEVATED TEMPERATURE ON *HOMARUS AMERICANUS* LARVAE

McVeigh HN, Waller JD, Fields DM

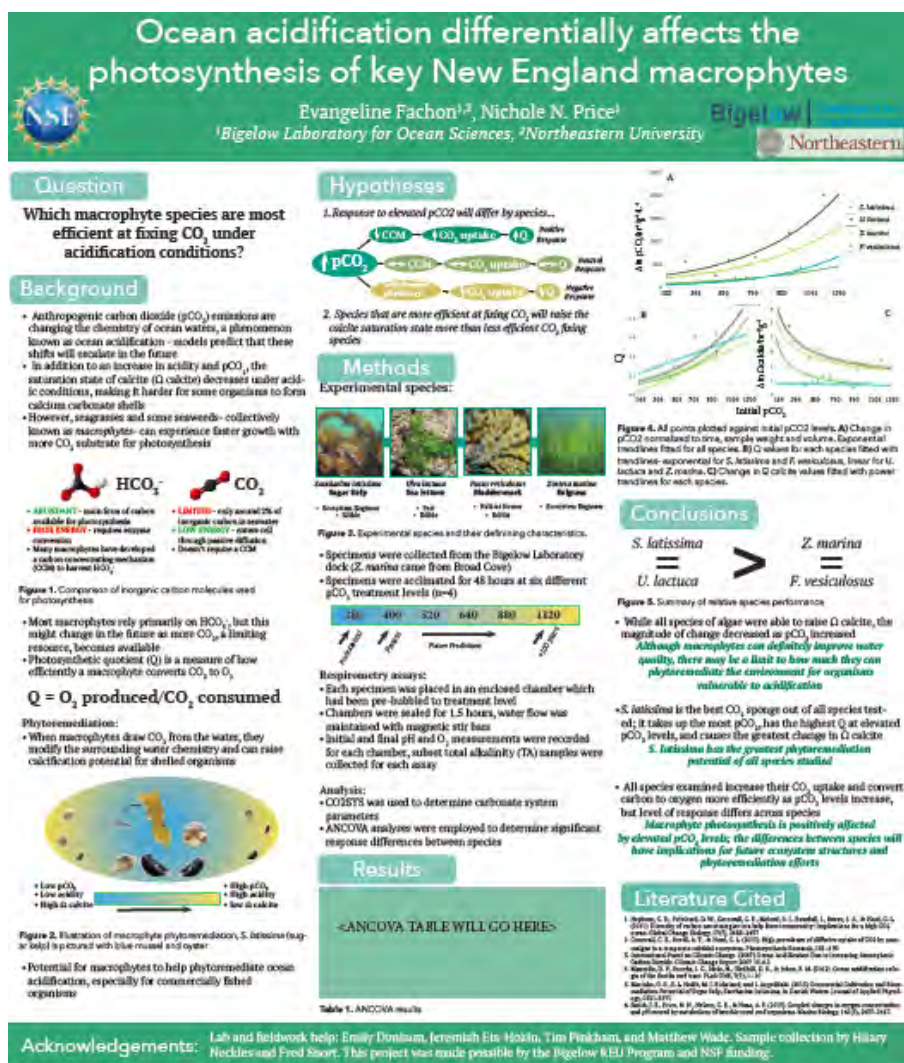
The Gulf of Maine is experiencing a rapid increase in temperature and a marked decrease in pH. This study aimed to quantify the impact of elevated temperature and decreased pH on the larval development of the iconic American lobster (*Homarus americanus*). Larvae were measured for growth (carapace length), development time, and survivorship over the larval duration. Elevated temperatures decreased development time across the larval stages of *H. americanus*. Consequently mortality increased at a significantly faster rate under elevated temperature. An increase in larval mortality may decrease recruitment to the fishery, thus impacting the most valuable single species in Maine. Experimental $p\text{CO}_2$ treatments yielded a significantly decreased development time between stages II and III, yet did not have a significant impact on carapace length or mortality. This study indicates that increasing temperature is a greater influence on larval development than elevated $p\text{CO}_2$. Determining how this species may respond to changing climactic conditions will better inform the sustainability efforts of such a critical marine fishery.



OCEAN ACIDIFICATION DIFFERENTIALLY AFFECTS THE PHOTOSYNTHESIS OF KEY NEW ENGLAND MACROPHYTES

Fachon E & Price NN

While the influence of anthropogenic CO₂ emissions on seawater chemistry is detrimental to the development of CaCO₃ reliant organisms, seagrasses and some seaweeds can experience enhanced growth under elevated pCO₂ conditions and even raise calcification potential (Ω calcite) for shell-producing organisms. Most marine macrophytes rely on the enzyme conversion of bicarbonate (HCO₃⁻) to supply the inorganic carbon needed for photosynthesis; the ability to down-regulate this energetic acquisition as more CO₂ becomes available will determine which species are successful under future conditions. Four commercially and ecologically relevant New England macrophyte species were exposed to past, present and future pCO₂ levels in respirometry assays; CO₂ consumption, photosynthetic quotient (Q), and change in Ω calcite were calculated for each sample. All species examined experienced increases in rate of CO₂ uptake and Q under elevated pCO₂ conditions, level of response differed across species. While all species of algae were able to raise Ω calcite, the magnitude of change decreased as pCO₂ increased. The varied responses observed across species have implications for future community structures as well as for phytoremediation efforts.

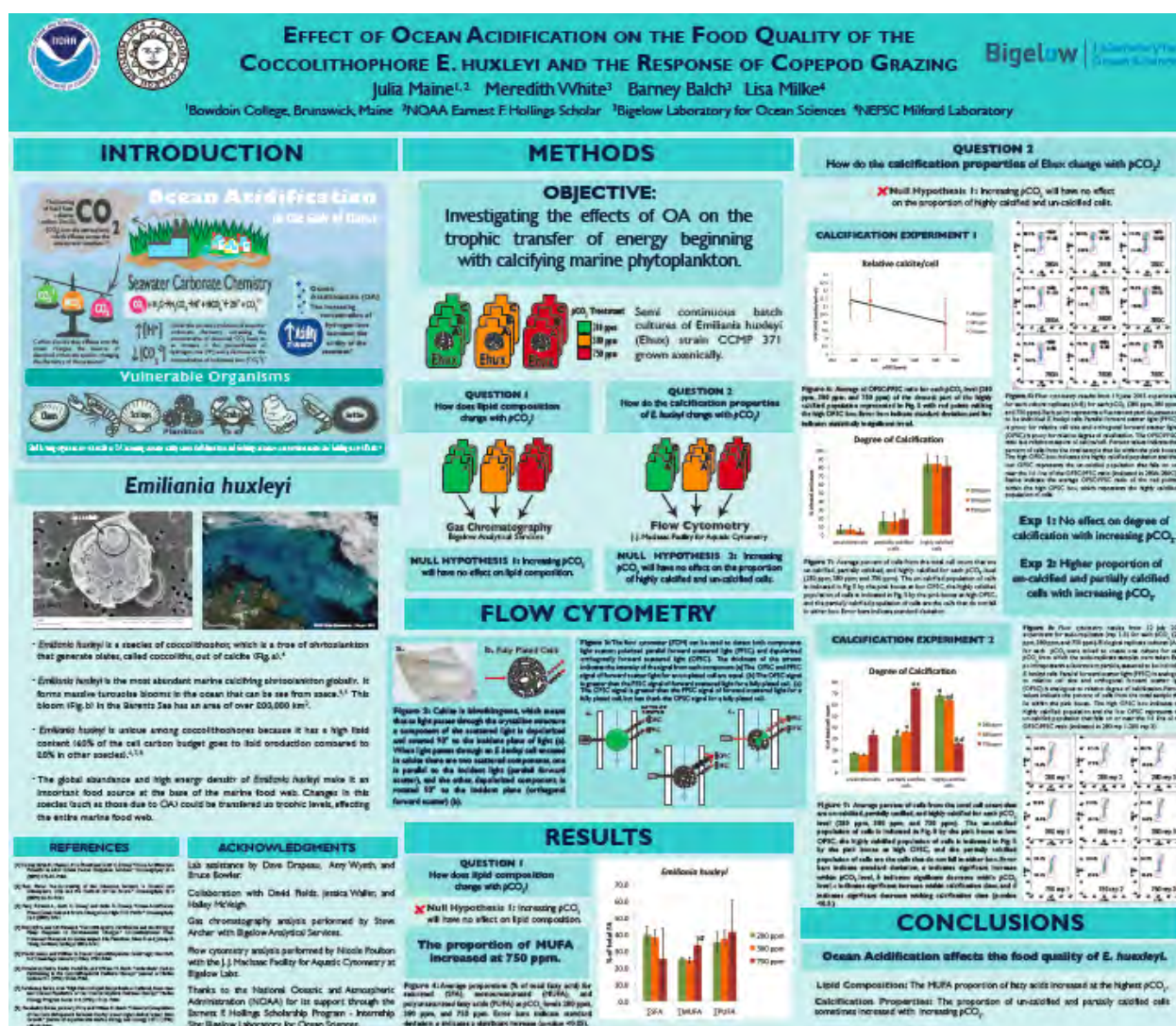


EFFECT OF OCEAN ACIDIFICATION ON THE FOOD QUALITY OF THE COCCOLITHOPHORE

E. HUXLEYI

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The burning of fossil fuels over the last 200 years has doubled atmospheric carbon dioxide (CO_2). CO_2 diffuses into seawater increasing acidity, a process called Ocean Acidification (OA). Calcifying marine phytoplankton, coccolithophores, are vulnerable to OA. *Emiliania huxleyi* is a lipid dense and globally abundant species of coccolithophore, therefore it is vital to higher trophic levels in the marine food web. The objective of this project was to determine how OA affects the food quality of *E. huxleyi*. Gas chromatography was used to determine how the proportions of saturated (SFA), monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA) in *E. huxleyi* varied with $p\text{CO}_2$. Flow cytometry was used to measure how the degree of calcification of cells changed with increasing $p\text{CO}_2$. The proportion of MUFA increased with $p\text{CO}_2$. The results of the first calcification experiment showed no effect on degree of calcification with increasing $p\text{CO}_2$, however the results of the second experiment showed that the proportion of un-calcified and partially calcified cells increased with increasing $p\text{CO}_2$. In conclusion, the food quality of *E. huxleyi* may be affected by OA.





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