

Inventory of barriers in the northeast that limit the aquaculture industry

Funding level: \$181,083 Project start date: 1 October 2017 Project duration: 24 Months Participants: University of Maryland

Project Summary:

The workgroup team will contact appropriate federal, state, and if possible, local agencies to determine aquaculture regulations and permitting requirements. The team will also research other potential barriers to entry and identify all key personnel for assistance in starting an aquaculture operation.

Identification of the cause of hemic neoplasia in *Mercenaria mercenaria* and development of management methods

Funding level: \$175,336 Project start date: 1 October 2017 Project duration: 24 Months Participants: Roger Williams University

Project Summary:

Aquaculture of hard clams, Mercenaria mercenaria, is an economically important business venture in the U.S. The USDA's 2013 census of aquaculture in the U.S. reported hard clam commercial aquaculture production was worth over \$64 million. Diseases that threaten the viability of this industry are of significant concern to culturists, managers and consumers on the East Coast. In the last few years, Murphy and Smolowitz have identified a new disease in hard clams that appears similar to disseminated neoplasia, as described in Mya arenaria (the soft shell clam) and other bivalves. The disease is occurring in hard clams in Wellfleet, MA, and significantly affects the animals aquacultured in several disparate locations within the harbor. This new hard clam disease causes mortality during the spring in both sub-market and market sized animals. A recent experiment by Murphy and Smolowitz demonstrated that the disease was not hatchery-specific and occurred in animals from 3 different hatcheries when those animals were grown in Wellfleet Harbor. This disease in hard clams may be transmissible through the water column as is being identified in a similar disease in soft shell clams. In addition, there exists potential for transmission cross-species. This emergent disease situation presents several potentially dire consequences if not addressed since transfer to other locations in MA and on the east coast is probable.

There is a demonstrated need to develop "on the farm" strategies for this emergent disease risk to quahog production. Current research is in the early stages of understanding this disease and the ramifications to the entire industry could be significant. Right now it appears to be a localized problem, but without directed research to understand the epidemiology there are few to no recommendations for "on the farm" management of this disease.

Objective 1: We will conduct transmission experiments using infected animals and harvested neoplastic cells from heavily infected animals gathered in the spring from Wellfleet, MA. Naive animals will be injected with cells, exposed to cells in the water, exposed to infected individuals and injected with a cell free filtrate at different temperatures.

Objective 2: We will examine the extent of the disease in Wellfleet by planting seed from 3 different hatcheries at locations around Wellfleet and in year two by sampling water and sediment for neoplastic cells. Additional

bivalve species from the Wellfleet study area will be examined for neoplasia, including but not limited to oysters (*Crassostrea virginica*), surf clams (*Spisula solidissima*), and razor clams (*Ensis directus*).

Objective 3: We will determine the origin of the neoplastic cells through recovery and sequencing of the gene targets from hemolymph and tissues of infected and uninfected hard clams following method used by Metzger *et al.* (2016) to study the similar disease in soft shell clams. We will examine hemolymph from infected and uninfected clams for reverse transcriptase activity and determine whether candidate retroelements are present and abundant in the transcriptomes.

Objective 4: We will develop a quantitative PCR test method for the disease to be used in this research on hemolymph samples, water and sediment and for ultimate use for disease diagnosis in a diagnostic lab. We will use this test for detection of the disease in hard clams

Objective 5: The extension component of this study will result in at least one public meeting to explain the study and gather feedback from aquaculturists and regulators, will present results and provide fact sheets and will organize two regional workshops to share results of the study and possible management methods.

Development of a phage-based diagnostic test for the rapid detection of pathogenic *Vibrio species* in bivalves

Funding level: \$191,424 Project start date: 1 October 2016 Project duration: 24 Months Participants: Cornell University, University of New Hampshire

Project Summary:

The prevalence of gastrointestinal illnesses associated with the consumption of Atlantic coast shellfish contaminated with *Vibrio parahaemolyticus* and *V. vulnificus* has increased over the years, having a negative impact on the industry due to recalls and loss of consumer confidence in the product. We are proposing to develop a rapid and highly sensitive bacteriophage-based diagnostic test for early detection of shellfish contamination by virulent strains of these two bacterial pathogens. The first objective of this proposal is to isolate and characterize phages that infect *V. parahaemolyticus* and *V. vulnificus*. These phages will be isolated from water samples in which these bacteria thrive along the Atlantic northeast coast. Isolated phages will be characterized to differentiate those that infect isolates from clinical cases (virulent strains). A set of phages that infect specifically virulent isolates will be further characterized for efficacy of infection and multiplication. The best

performing phages will be selected and sequenced. The second objective of this proposal aims at developing the diagnostic test. Selected phages will be modified to express a marker that will be easily detectable in infected samples, bypassing the requirement for an enrichment phase. This detectable marker will be produced by phages multiplying in virulent isolates of *V. parahaemolyticus and V. vulnificus*. The ultimate goal of this research project is to develop a test that will be specific, sensitive, cost-effective, and rapid for the detection of shellfish contaminated with *V. parahaemolyticus and V. vulnificus*. This test will be beneficial to the industry and the consumers because it will provide a reliable and rapid tool to monitor contamination at any step of the process from pre-harvest to table.

A Novel Approach To Prevent Super Chill in Atlantic Salmon

Funding level: \$165,624 Project start date: 1 October 2016 Project duration: 24 Months Participants: University of Maine, National Cold Water Marine Aquaculture Center (ME), Cooke Aquaculture (ME), University of Rhode Island

Project Summary:

The project will research and demonstrate innovative production technologies focused on mitigation of super chill in Atlantic salmon aquaculture though new feed formulations. Project objectives are; to investigate the concentrations of simple sugars and sugar alcohols (SSASA) in plasma of Atlantic salmon to achieve a lower freezing point depression limit, to investigate the uptake of SSASA from the diet in both epithelia cells and the plasma of Atlantic salmon, to determine the efficacy of SSASA in reducing the impact of a super chill event under controlled conditions, to measure the physiological parameters of the fish subjected to super chill and compare to control, and to liaise with Atlantic salmon aquaculture industry, extension and technology transfer.

Impact Assessment of NRAC Funding on Aquaculture in the Northeast

Funding level: \$150,000 Project start date: 1 October 2015

Project duration: 24 Months

Participants: University of New Hampshire, Virgin Oyster Company, Salem State University, Marine Biological Laboratory, University of Maryland, NOAA, New Jersey Sea Grant, Profish, MAIC, Delaware State University, University of Maine, State of Pennsylvania, University of Rhode Island, Cornell University, West Virginia University

Project Summary:

The overall objective is to assess the scientific, socio-economic, and policy impacts of accomplishments made through NRAC's portfolio of recently (since 2005) funded aquaculture projects, including extension workgroup projects. Incorporated in this synthesis will be how these projects have or have not helped move the aquaculture industry close to solutions for the diversity of problems it faces within the region. From the resultant information, suggestions for achieving higher impacts will be identified that NRAC should consider in future funding initiatives.

Safe Feedstocks for Bivalve Aquaculture

Funding level: \$190,508 Project start date: 1 October 2015

Project duration: 24 Months

Participants: University of Rhode Island, Roger Williams University, Oyster Seed Holdings, Virginia Institute of Marine Sciences, Martha's Vineyard Shellfish Group, Fishers Island Oyster Group

Project Summary:

The overall goal of the project is to test the efficacy and safety of two bacteria strains to reduce or eliminate vibrios from phytoplankton feedstocks that are commonly used in bivalve larviculture and manage disease in hatcheries.

Specific aims:

1. Measure the reduction of vibrios and other potentially pathogenic bacteria in algal cultures following the addition of the two beneficial bacterial probiotic strains.

2. Measure the growth and survival of algae in response to probiotic treatment.

3. Measure the survival of bivalve larvae that are fed probiotic-treated algae prior to being challenged with bacterial pathogens.

4. Outreach and Extension: The results of this research will reach potential end users through direct collaboration, workshops, and meetings with commercial hatchery managers.

This work will be performed first at the laboratory scale and then tested at a commercial scale in bivalve hatcheries. Dependent upon successful outcomes of the proposed trials, the candidate strains will advance toward commercial development as new tools to prevent pathogen contamination of aquaculture systems. We anticipate that these strains will ultimately be useful to prevent economic losses due to infectious disease outbreaks in commercial hatcheries.

Testing and Application of Novel Probiotic Bacteria for Use in Marine Aquaculture

Funding level: \$190,508 Project start date: 1 October 2015 Project duration: 24 Months Participants: University of Delaware, University of Maryland Baltimore County, University of MD Center for Environmental Science

Project Summary:

A 24 month study is planned in which we will build on the foundation laid by the 2012 NRAC Mini-Grant. Through that effort, we identified seven non-Vibrio bacteria from the intestines of Fundulus heteroclitus that had the ability to inhibit growth of not only the fish pathogens Vibrio harveyi (DNO1) and Vibrio damsela but also may inhibit growth of Vibrio sp. B183 (a shellfish pathogen) as determined by filter disk assays. Of these, we tested four probiotic bacteria (OY15, Iso5, Iso11 and Iso12) selected by their ability to inhibit pathogen growth and a glycerol-only control (the probiotic storage medium) in a short term growth trial with our model species, F. heteroclitus, to ascertain the potential effects of the novel probiotic bacterial strains on the larvae. In that initial trial, mummichogs exposed to the novel probiotics exhibited higher growth rates compared to the fish in the control treatment. The current proposal will continue to evaluate these potential probiotic strains on survival of growth of other commercially important species including striped bass, rainbow trout, Atlantic salmon and tilapia. In all cases, one day-old larvae will be stocked into triplicate experimental tanks at a density of 25 larvae per liter. Samples of larvae will be collected at the time of stocking and weekly to assess the growth effect of the respective probiotic strains. After six weeks, all surviving fish from each tank will be individually counted to determine survival. Due to space and labor considerations, individual target culture species will be tested in succession, as they become commercially/seasonally available.

Development and Evaluation of Novel, Non-toxic Solutions for Biofouling Control and Predator Exclusion in Shellfish Aquaculture

Funding level: \$193,582 Project start date: 1 October 2015 Project duration: 24 Months Participants: University of Connecticut, University of Maine, Martha's Vineyard Shellfish Group, Virgin Oyster Company, ePaint

Project Summary:

We propose to develop and refine an effective, environmentally-friendly, and affordable means to prevent biofouling on aquaculture gear and predation. We will 1) assess potential toxicity of new coatings (note: base materials being tested have been shown to be non-toxic and all are cleared with FDA and EPA regulations; it is still important to test new configurations to confirm non-toxic status); 2) assess adhesion and efficacy of newly developed formulations on test panels and gear on experimental facilities and farms to assess antifouling and antipredator properties, and 4) engage aquaculture producers in research and outreach, and disseminate the results to the industrial and scientific communities through presentations at workshops, conferences, outreach publications, web page, and peer-reviewed publications. We have assembled a strong team of scientists and aquaculturists from 4 states (ME, MA, NH, CT) as well as a cadre of industry supporters with decades of experience in shellfish biology, anti-fouling coatings, aquaculture, and outreach. All proposed experimental techniques have been used in our prior studies, and we have access to all necessary equipment and facilities. Successful development and demonstration of extended efficacy of biologically inert, inexpensive, and practical deterrents to biofouling will have a profound, positive impact on the global aquaculture industry, and our aim is to remove biofouling as one of the most costly problems in aquaculture.

Teleost spermatozoal transcriptomes: requisite foundation for functional genomics, sperm quality and male fertility

Funding level: \$20,000 Project start date: 1 Sept 2014 Project duration: 12 Months Participants: University of Maryland, Stuttgart National Aquaculture Research Center

Project Summary: The objective is to genetically sequence and evaluate spermatozoal transcriptome profiles from male striped bass with observed differences in fertilization rate.

White Worm, *Enchytraeus albidus*, production and marketing for live aquaculture feed Funding level: \$151,925 Project start date: 15 Aug 2014 Project duration: 24 Months Participants: University of New Hampshire, Washington DC

Project Summary:

White worms, *Enchytraeus albidus*, are a promising feed in the aquaculture industry for a variety of species (freshwater, marine, ornamentals, shrimps) across the U.S. Although they used to be mass produced in the former U.S.S.R. in the 1940s for sturgeon, white worm farming is a "lost art." At UNH, we have established a large white worm population using these dated Soviet techniques. There is enormous interest by the aquaculture industry to utilize white worms as a feed because more species are being cultivated, many of these still at the R&D stages, and many of whom are picky eaters during specific ontogenetic phases. The variety of available small live feeds for these picky eaters is limited. Also, it is not economically nor environmentally feasible to feed aquacultured species diets mostly composed of expensive, harvested wild fish meal. While we currently are working to provide worm samples to all interested parties, we cannot promote them as a readily available food source until we optimize worm production and are confident that we can ensure a large, steady, and safe supply. This research project would (1) develop "modern" white worm production protocols, which eventually could be adapted for commercial scale production, and (2) work with all interested aquaculture sectors to identify the white worm market(s) through a series of workshops resulting in identifying the target markets and providing worms nutritionally customized for those consumers (species). This research promotes sustainable, environmentally friendly tactics in its use of recycled, local, waste byproducts for worm feed, and a low carbon footprint. This research will yield economically viable techniques for those aquaculturists looking to diversify and a readily- available product for the aquaculture market.

Hatchery and Nursery Technologies for Improved Production of Blue Mussels

Funding level: \$198,077 Project start date: 15 Aug 2014 Project duration: 24 Months Participants: Marine Biological Laboratory, University of Maine, Massachusetts Sea Grant

Project Summary:

The overarching goals of the research we propose are to (i) develop hatchery and nursery technologies for cost effective year round seed production of blue mussels (*Mytilus edulis*), (ii) enhance the annual output per unit area of mussel farms and the market potential for domestically grown mussels and (iii) support the expansion and diversification of the northeastern shellfish culture industry which will create new opportunities for hatcheries, aquaculturists and fishermen. Our proposed research will facilitate the establishment of consistent, reliable sources of blue mussel seed from hatcheries that will promote steadier, higher volume and higher value harvests per acre. Ultimately, hatchery production of seed will result in "added value" by laying the foundation for genetic improvement of key traits. In addition to communicating and transferring the progress and results of our work directly to our hatchery and industry partners, we plan to have two major project deliverables. One will be to sponsor two Mussel Grower Workshops in Year 2 of the project to be held at the Darling Marine Center in Maine, and at the Cape Cod Cooperative Extension Office in Massachusetts. At these workshops, we will present not only the project results, but also

discuss blue mussel biology, current grow-out technologies, and current market opportunities and the implication of our results. Our goal is to increase participation in blue mussel culture region-wide. We will also communicate and transfer our project benefits through a *second major products*, including, but not limited to, a detailed hatchery manual for mussel seed, distributing mussel seed at the conclusion of the hatchery phases of our project, making the special declumping and grading equipment available to growers, presenting at regional scientific meetings, submitting articles in local and regional newsletters, and through our websites at our respective research institutions.

Measuring flow within shellfish growout systems: developing and evaluating a logging mini-flow meter

Funding level: \$8,634 Project start date: 1 July 2014 Project duration: 12 Months Participants: Roger Williams University

Project Summary:

The objective of this mini-grant application is to develop a small scale logging flow meter that will be of a size and a capacity capable of independent deployment in traditional shellfish aquaculture gear for an extended period of time. Once developed, we propose to test the application of the flow meter in multiple shellfish growout systems.

Workshop to Identify Constraints towards developing a Commercial eel aquaculture industry using Recirculating Aquaculture System (RAS) technology

Funding level: \$11,000 Project start date: 1 July 2014 Project duration: 12 Months Participants: University of New England, Cornell University, SUNY College at Oswego, Maine Sea Grant College Program and U Maine Cooperative Extension, University of Massachusetts Dartmouth, University of Maine

Project Summary:

We propose a two-day workshop at the University of New England in Biddeford ME where we will bring in experts associated with eel biology, marketing, and engineering to identify constraints and possible solutions towards creating economically viable culture systems using RAS technology.

There is Gold in Them Thar Mussels; Testing the Feasibility of Golden Mussel Culture for Branding and Market Expansion of Farmed New England Mussels

Funding level: \$10,000 Project start date: 1 July 2014 Project duration: 12 Months Participants: Marine Biological Laboratory, Martha's Vineyard Shellfish Group,

Project Summary:

The objective of this small study is to test the feasibility of utilizing the natural golden color morph of *Mytilus edulis* for aquaculture. Parameters of concern include larval competence (i.e. time to and completion of metamorphosis), growth rate from seed to market size, meat yield and shell strength, compared to those of the blue color morph. Shell strength will not be quantified in the study but should be explored in the future in order to fully evaluate the value of golden mussels.