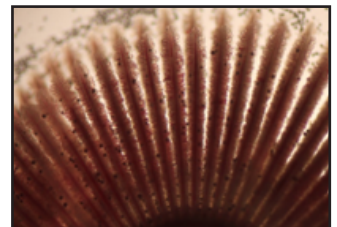
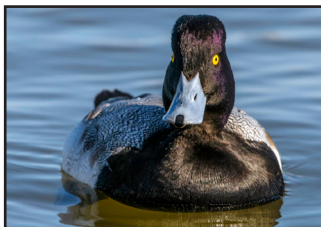
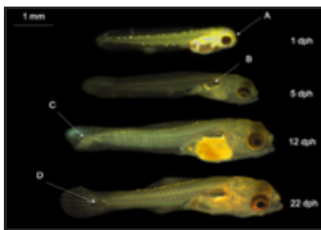


June 2019



**Supporting research and Extension projects based on industry needs and designed to directly impact commercial aquaculture development.**



For the period through August 31, 2018



United States Department of Agriculture  
National Institute of Food and Agriculture

# THIRTY-FIRST ANNUAL PROGRESS REPORT

USDA NIFA SOUTHERN REGIONAL AQUACULTURE CENTER

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## EXECUTIVE SUMMARY

This Thirty-First Annual Progress Report seeks to provide a summary of work completed and outreach activities of the Administrative Center during the past year. Full progress reports on the 11 multi-year research and Extension projects supported by SRAC during this reporting period are available at <http://www.srac.msstate.edu/annualprogressreports.html>. In the past year, SRAC funded projects totaling more than \$1.2 million. During the past year, these projects have resulted in 1 journal article, 3 abstracts, 1 book chapter, 4 Extension/Outreach publications, 20 oral presentations, 2 poster presentations, 5 digital products, and has supported 22 students.

The Center's Publications project is in its twenty-second year of funding. The Southern Regional Aquaculture Center started the Publications, Videos, and Computer Software Project in order to provide these materials in a timely and relevant manner. Since that time, more than 350 technical fact sheets (244 in the current catalog), 97 update revisions, 7 web presentations, 7 software programs or web tools, and 31 videos have been produced through the SRAC PVCS Project. Two new fact sheets and two fact sheet revisions were completed for this reporting period. The SRAC publications and AquaPlant websites were also updated with new materials. In the current reporting year, 59,101 unique users from 169 countries and territories used the SRAC Publications website, <https://srac.tamu.edu/>, to view or download SRAC publications 249,581 times. SRAC videos were viewed on the SRAC YouTube channel 41,455\* times during the current reporting period. The AquaPlant website, created with funding from the SRAC PVCS Project, had 233,279 unique users that viewed 592,196 webpages during the reporting period. These users were from 181 countries/territories. These analytics demonstrate that the SRAC Publications, Videos, and Computer Software project truly has worldwide reach and impact.

The "Improved Reproduction in Foodfish (Catfish and Largemouth Bass), Baitfish, and Ornamentals Using a New Spawning Aid (GnRH IIa)" project is evaluating the efficacy, reliability, safety, and mode of action of GnRH IIa in a range of species. GnRH IIa was deemed safe to induce spawning in channel catfish females. Forty-eight 5-year old blue males were implanted with either 0 µg, 100 µg or 500 µg of GnRH IIa. No significant differences were observed between the 3 groups regarding testis weight or motility. However, it is not possible to say at this point if the lack of effect is due to the implant type used, the protocol, or if GnRH IIa is not efficacious in maturing the testis. Spawning and milting trials for artificial propagation of Largemouth Bass were conducted for Year 2. Spawning trials for all four ornamental species were successfully completed. GnRH IIa was effective for spawning induction for both baitfish species (pinfish and pigfish) at all tested dosages. Data obtained in this project were submitted to the US Food and Drug Administration (FDA) in partial fulfillment of data requirements for an Investigational New Animal Drug (INAD) permit. The INAD was opened by FDA in February 18, 2018, and granted to AquaTactics LLC. Data collected during farm trials under the INAD will eventually be used to request full labelling of the drug for use in commercial hatcheries.

The "Predation Risk and Economic Impact of Lesser Scaup and Piscivorous Waterbirds on Commercial Baitfish and Catfish Production" project will allow researchers to estimate economic losses of fish caused by these birds and generate management recommendations for producers to ameliorate depredation of fish. During 2017-2018, 628 pond surveys were conducted on 423 individual baitfish ponds and 1,740 scaup were counted. Investigators collected 267 scaup and 29% contained fish parts. Investigators also completed 12 aerial surveys and counted 130,684 cormorants across 68 different night roosts. A total of 338 cormorants were harvested and catfish (*Ictalurus spp.*) represented 33% of the total prey biomass in their stomachs. This is the first study to develop on-farm costs of attempts by

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farmers to prevent losses due to bird depredation. Results demonstrate that the greatest costs are for the trucks and manpower used to chase birds. Fish farmers are spending more money and more time than had previously been thought in efforts to scare birds from their ponds.

The “Commercial Production of Selected Native Freshwater Ornamental Species” project aims to define effective culture protocols for nine species of freshwater fishes endemic to the U.S. Culture methods for the Gulf Coast Pygmy Sunfish and Metallic Shiner developed during this project are a solid foundation for the commercial production of select native North American ornamental fishes, which may be transferable to close congeners. Recommendations on Golden Topminnow broodfish size and expected egg output will help producers with production goals for this species as fecundity is low compared to many other ornamental species. Bluenose Shiner appear to require a nest mate associate such as a sunfish to initiate spawning as they potentially use the sunfishes nest as spawning substrate. Photo-thermal manipulation and introduction of artificial “chub mounds” has been unsuccessful for captive spawning of Mountain Red-belly Dace. Rainbow Darter at the Virginia Tech Hampton facility have also begun to reproduce and egg collection, disinfection, and rearing of fry will continue to aid in determination of fecundity, and establish disinfection and larviculture protocols.

The goal of the “Repeatability of Incidence and Time of Ovulation, Fecundity, and Fertility in Channel Catfish Females Induced to Ovulate for Production of Hybrid Catfish Fry” project is to determine a strategy to allow farmers to decide which females should be carried over to the next year and which should be culled at the end of the spawning season to increase hybrid catfish fry production efficiency. Based on ovulation rate only, surviving females are good for hybrid embryo production for at least 2 years. Letting a female “rest” for 2 years had no benefit. Degree of female readiness, fecundity and egg quality were not repeatable over a 2-year period. Hand-stripping increases the probability of death prior to the next spawning season. Brood stock handling procedures needs improvement if brood females are to be used multiple years. Management of brood stock densities is problematic because of potential high mortality that is not detected (fish often do not “float up” after death).

The “Techniques to Improve Production of Off-bottom Cultured Oysters” project seeks to evaluate the benefits associated with fine tuning methods to control biofouling when using the OysterGro™ system to grow high value single oysters. These include: reduced labor costs, improved product quality, improved yield, and shorter grow-out time. Within the project period, investigators deployed the experiment in each state and completed quarterly sampling. The industry partner in each state was responsible for adhering to the flipping routine. On the Gulf Coast, the experiment was completed because >70% of the oysters in the bi-weekly flipping treatment had reached harvest size and the gear had been exposed to the heavy fouling season. The project on Gulf Coast was terminated in June 2018 with harvesting of the product and final sampling. On the Atlantic Coast, by June 2018, 70% or more of the oysters in the bi-weekly flipping treatment had reached harvest size in North Carolina and South Carolina. In Georgia, however, the oysters were smaller. As of June 2018, the gear in each Atlantic state had not been exposed to what would be considered a heavy fouling season (i.e. summer). The decision was made to treat the June sampling trip as a “harvest” sampling for NC and SC. At that time the densities were reduced in each bag and the experimental treatments continued to be applied until the one-year post-deployment time.

The “Evaluation of Protein and Lipid Concentrations in Commercially Available Tilapia Feeds and Their Effect in Intensive Production Systems” project will evaluate typical commercial diet formulations with different levels of protein and lipid in commercial intensive tilapia recirculating aquaculture systems.

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The project start date was delayed due to some industry constraints. The project team needed to make some new arrangements to satisfy the industry participants on the project. Since one of the tilapia farms has recently installed a new production system, investigators continue to wait until the filtration systems achieve steady state (mature filters). To get things started, team members have held several conference calls, have met in person with all industry partners, and performed site visits at the tilapia RAS farms. They have also begun collecting baseline data to enter into the economic software and water quality model. After the site visits, economists realized that some adjustments needed to be made to the economic model. Those adjustments were completed.

The “Field-testing of a Rapid LAMP Assay to Detect the Marine Parasite *Amyloodinium ocellatum* in Commercial Aquaculture Facilities” project seeks to transition this technology to commercial application and provide enhanced opportunities for improving commercial outcomes. Unfortunately, this project was unable to definitively demonstrate its applicability in the field due to a couple of factors. Overall, the number of samples from the farms was lower than expected and produced only two microscopy-positive samples, which limits the statistical power required to robustly compare the sensitivities of LAMP and microscopy. Additionally, the LAMP assay experienced unresolved contamination issues that prevented its reliable use. As such, assessment of the limited number of positive or potentially positive samples was not done. Nevertheless, the investigators have generated cloned, positive controls that can be used to determine specific copy number of the target including future applications using qPCR. These cloned controls can be easily shared among diagnostic laboratories and could serve as reference material for future inspection protocols. Previous to this study, no information was available on freeze-tolerance of the pathogen. This work has demonstrated that frozen fish can vector viable, infective parasite life history stages that can establish infections in relatively biosecure facilities. The LAMP assay continues to have the potential for providing a substantial benefit to producers, but farm samples and lab samples destined for molecular analysis remain for future processing once resources to resolve the contamination issue are available.





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## INTRODUCTION

### **Mission**

The mission of the USDA NIFA Southern Regional Aquaculture Center (SRAC) is to support aquaculture research, development, demonstration, and education to enhance viable and profitable U.S. aquaculture production to benefit consumers, producers, service industries, and the American economy. Projects that are developed and funded are based on industry needs and are designed to directly impact commercial aquaculture development in the southern region and the nation.

### **Background**

The Agriculture Acts of 1980 and 1985 authorized establishment of aquaculture research, development, and demonstration centers in the United States. With appropriations provided by Congress for the 1987 and 1988 FYs, efforts were undertaken to develop the five Regional Aquaculture Centers now in existence. Organizational activities for SRAC began in 1987, with the first research and Extension projects initiated in 1988.

In 1980, Congress recognized the opportunity for making significant progress in domestic aquaculture development by passing the National Aquaculture Act (P.L. 96-362). The Act established USDA as the lead agency for aquaculture coordination and called for development of a National Aquaculture Plan. The next year, Congress amended the National Agricultural Research, Extension, and Teaching Policy Act of 1977 (P.L. 95-113) by granting, in Title XIV, Subtitle L, Sec. 1475(d) of the Agriculture and Food Act of 1981 (P.L. 97-98), authority to establish aquaculture research, development, and demonstration centers in the United States.

Congress envisioned the Centers as focal points in a national program of cooperative research, Extension, and development activities that would be developed in association with colleges and universities, state Departments of Agriculture, federal facilities, and non-profit private research institutions with demonstrated excellence in aquaculture research and Extension. Eventually, five such Centers were established: one in each of the northeastern, north central, southern, western, and tropical Pacific regions of the country.

Although government agencies, particularly the United States Department of Agriculture, have provided significant support for aquaculture research and development, much of that funding is earmarked for specific use by specific institutions. The USDA NIFA Regional Aquaculture Center program is the only funding activity with the flexibility to stay abreast of industry development, identify problems on a region-wide scale, and implement cooperative, interstate projects to solve those problems.

Since its inception in 1987, SRAC has become the most important regional aquaculture activity in the southeastern United States. In its 31 years of operation, the Center has disbursed more than \$19.1 million to fund multi-state research and Extension projects. More than 200 scientists from 30 institutions in the southeast have participated in Center projects.

Productivity from SRAC research projects has been excellent since the Center's inception more than three decades ago. Information derived from SRAC-funded projects has been transferred to producers

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and other scientists in thousands of scientific papers and presentations. Currently funded projects continue this trend of high productivity.

Beginning with the first projects funded by SRAC, interest among aquaculture research and Extension scientists in Center activities has been excellent. In fact, funding and project coordination provided by SRAC has become so embedded in the fabric of southeastern aquaculture research and Extension that it is difficult to envision what these activities would be like without the program. We are pleased with the participation by our research and Extension scientists in the Southern Region in *ad hoc* Work Group meetings and Steering Committees, and their willingness to serve as Project Leaders and Principal Investigators for the projects. We believe this broad-based representation has resulted in strong, cooperative research that will be of long-lasting benefit to aquaculture producers and consumers, and to the growth of the aquaculture industry in the Southern United States.

### **Acknowledgments**

The Southern Regional Aquaculture Center acknowledges the contributions of the Project Leaders and Participating Scientists involved in the projects reported in this Thirty-first Annual Progress Report. Members of the SRAC Board of Directors, Industry Advisory Council, and Technical Committee have provided valuable inputs to the successful operation of SRAC during the past year. We particularly appreciate the assistance of the Chairs of these vital committees.

We also thank the scientists and aquaculturists from across the country who contributed their expertise and valuable time to review SRAC project proposals and publications. Without their help, it would be impossible to maintain the high quality of this program.

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## ORGANIZATIONAL STRUCTURE

Research and Extension problem areas for the southern region are identified each year by the Industry Advisory Council (IAC), which consists of fish farmers and allied industry representatives from across the region. The Technical Committee (TC), consisting of research and Extension scientists from states within the region, works with the IAC to prioritize problem areas. The two groups then work together to develop “Requests for Pre-proposals” describing objectives of work to solve problems with the highest priority. The best proposals submitted by individuals or teams are used to form a regional Work Group that plans and conducts the work. Regional aquaculture funds are allocated to participants in SRAC projects approved by the Board and NIFA. Reviews of project proposals, progress reports, and recommendations for continuation, revision, or termination of projects are made jointly by the TC and IAC and approved by the Board.

The thirteen states and two territories represented by SRAC are Alabama, Arkansas, Florida, Georgia, Kentucky, Louisiana, Mississippi, North Carolina, Oklahoma, Puerto Rico, South Carolina, Tennessee, Texas, U.S. Virgin Islands, and Virginia.

### **Administrative Center**

The Administrative Center is located at the Delta Research and Extension Center, Stoneville, Mississippi. Mississippi State University serves as the Host Institution. All necessary support services for the Board, IAC, TC, Steering Committees, and project Work Groups are provided by the Administrative Center. This includes monitoring status and progress of projects, preparing and executing Letters of Agreement, tracking administrative and project expenditures, reviewing progress reports, and assisting Project Leaders and participating institutional Grants Offices as needed.

Operation and funding are approved by the Board for inclusion in the Grant Application submitted annually to USDA NIFA. The Center staff also prepares and submits to USDA NIFA an Annual Plan of Work covering Center activities and projects to be funded. Following final approval, Letters of Agreement are prepared and executed with all participating institutions. The Center acts as fiscal agent to disburse and track all funds in accordance with the provisions of the grants.

### **Board of Directors**

The Board is the policy-making body for SRAC. Membership provides an appropriate balance among representatives from State Agricultural Experiment Stations, Cooperative Extension Services, 1890 Institutions, and the Administrative Heads Section of the Board on Agriculture Assembly of the Association of Public and Land Grant Universities.

The Board is responsible for 1) overall administration and management of the regional center program; 2) establishment of overall regional aquaculture research and Extension goals and allocations of fiscal resources to ensure that the center develops strong programs in both research and Extension; 3) establishment of priorities for regional aquaculture research and Extension education activities based on inputs from the TC and IAC; 4) review and approval of annual plans of work and accomplishment reports; and 5) final selection of proposals for funding by SRAC.

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Members of the Board for the reporting period were:

Gregory Bohach, Mississippi State University (Chair)  
Phil Elzer, Louisiana State University AgCenter  
Gina Eubanks, Louisiana State University AgCenter  
Steve Lommel, North Carolina State University  
Rick Cartwright, University of Arkansas Cooperative Extension Service  
Gary Lemme, Auburn Cooperative Extension Service, Auburn University  
Wes Burger, Mississippi State University

### **Industry Advisory Council**

The IAC is composed of representatives of state and regional aquaculture associations, federal, territorial and state agencies, aquaculture producers, aquaculture marketing and processing firms, financial institutions, and other interests or organizations. The IAC provides an open forum wherein maximum input from private and public sectors can be gained and incorporated into annual and ongoing plans for SRAC.

The IAC 1) identifies research and Extension needs; 2) works with the TC to prioritize research and Extension needs; 3) works with the TC to develop problem statements and recommend funding levels for projects addressing priority research and Extension needs; 4) reviews project proposals, progress reports, and termination reports; and 5) recommends to the Board, jointly with the TC, actions regarding new and continuing proposals, proposal modifications, and terminations.

Members of the IAC for the reporting period were:

Margie Saul, AR	Wec Terry, VA
Steve Sarten, KY	Kim Edge, GA
Ben Pentecost, MS	Douglas Kuenz, LA
Martha Campbell, FL	Shorty Jones, MS
Rob Ellis, NC	Chase Holub, TX
Marty Tanner, FL	Frank Roberts, SC
Travis Wilson, AL	David Heikes, AR
Townsend Kyser, AL	Jenny Davis Fagan, TN
Richard Eager, SC	

### **Technical Committee**

The TC consists of representatives from participating research institutions and state Extension services, other state or territorial public agencies as appropriate, and private institutions. Membership of the TC includes research and Extension scientists representing essentially all states in the region. The TC 1) works with the IAC to prioritize research and Extension needs; 2) works with the IAC to develop problem statements and recommend funding levels for projects addressing priority research and Extension needs; 3) reviews proposals, progress reports, and termination reports; and 4) recommends to the Board, jointly with the IAC, actions regarding new and continuing proposals, proposal modifications and terminations.

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Members of the TC for research for the reporting period were:

Brian Bosworth, USDA-ARS Warmwater Aquaculture Research Unit  
Ben Reading, North Carolina State University  
Ken Semmens, Kentucky State University  
Allen Davis, Auburn University  
Amit Sinha, University of Arkansas at Pine Bluff  
Amrit Bart, University of Georgia  
Delbert Gatlin, Texas A&M University  
Chris Green, Louisiana State University  
Cortney Ohs, University of Florida  
Chris Bentley, Virginia Tech University  
Mike Denson, South Carolina Department of Natural Resources  
Brian Alford, University of Tennessee

Members of the TC for Extension for the reporting period were:

Lance Beecher, Clemson University  
Mike Frinsko, North Carolina State University  
Gary Burtle, University of Georgia  
Luke Roy, Auburn University  
Todd Sink, Texas A&M University  
Greg Lutz, Louisiana State University  
Michael Schwarz, Virginia Tech University  
Craig Watson, University of Florida  
Forrest Wynne, Kentucky State University  
Anita Kelly, University of Arkansas at Pine Bluff  
Ganesh Kumar, Mississippi State University  
Marley Beem, Oklahoma State University  
Don Bailey, University of the Virgin Islands



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## PROGRESS REPORTS

### Publications, Videos, and Computer Software

**Reporting Period:** December 1, 2017 – August 31, 2018

**Length of Project:** March 1, 1995 – Ongoing

**Current Project Year:** 22

**Total Funds Committed:** \$28,354

**Principal Investigator:** Todd Sink, *Texas A&M University*

**Relevance:** When this project was initiated, fewer than half the states had educational materials covering the major aquacultural species in their state. The concept of using the SRAC program to produce timely, high-quality educational materials is based upon the benefits of centralizing the production process while using a region-wide pool of expertise to develop materials. Distribution is then decentralized through the nationwide network of Extension Specialists and County Agents including the National eXtension Initiative. This process assures an efficient publication process that makes use of the best available talent in specific subject areas.

**Response:** A committee of Extension Specialists and researchers solicit input on publication and digital product needs from their counterparts across the region. These suggestions are prioritized during an annual meeting of the committee based on need and available funding. The best talent from within and outside the region are then recruited to submit proposals to develop these products.

**Results:** The result is widespread availability of high-quality educational materials for scientists, educators, producers, students, and the general public which in turn leads to increased or improved efficiency aquaculture production, improved awareness of aquaculture products and the nutritional benefits of seafood, and increased aquaculture investment.

**Outreach Overview:** SRAC fact sheets and videos are distributed electronically, by direct request, and via Extension Specialists, County Extension Agents, and other RACs. These products are used regularly by clientele in all 50 states as well as internationally in 208 countries and territories. Fact sheets, videos, and web presentations are accessed daily from the SRAC Publications website and YouTube by people searching for technical information.



**Targeted Audiences:** The target audiences for this project are educators, consumers, producers, potential investors, students, and the general public.

**Outputs:** Two new fact sheets and two fact sheet revisions were completed for this reporting period. The SRAC publications and AquaPlant websites were also updated with new materials. All completed publications have been distributed electronically throughout the Southern Region and to interested Extension Specialists in other regions.



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**Outcomes/Impacts:** Publications and videos produced by SRAC are increasingly used in educating high school and college students about aquaculture. These programs heavily utilize SRAC publications and videos for educational purposes but usage is impossible to measure because access to the information is gained from many different Internet sites, through file sharing, and digital downloads of PDFs.

Another important impact is the education of local, state, and federal regulators about the aquaculture industry. This impact is difficult to measure but feedback from personnel in two states have indicated that the fact sheets are recommended reading for all new employees dealing with aquaculture, water quality, exotic species, and other permitting duties. This should be a positive influence toward making aquaculturists better understood and the development of more enlightened regulations.

The impact on consumers of aquaculture products is also likely significant. Consumers are primarily interested in a wholesome, safe, and inexpensive product, and it has been reported that the consumer-oriented fact sheets and videos developed within SRAC have generated more interest than the producer-directed materials. The fact sheets are in demand in both the English and Spanish versions and, as more information becomes available, Extension materials on food safety will be in increased demand by health conscious consumers.

The Southern Regional Aquaculture Center started the Publications, Videos, and Computer Software Project in order to provide these materials in a timely and relevant manner. Since that time, more than 350 technical fact sheets (244 in the current catalog), 97 update revisions, 7 web presentations, 7 software programs or web tools, and 31 videos have been produced through the SRAC PVCS Project. In the current reporting year alone, **59,101\*** unique users from **169\*** countries and territories used the SRAC Publications website, <https://srac.tamu.edu/>, to view or download SRAC publications **249,581\*** times. SRAC videos were viewed on the SRAC YouTube channel **41,455\*** times during the current reporting period. The AquaPant website, created with funding from the SRAC PVCS Project, had 233,279\* unique users that viewed 592,196\* webpages during the reporting period. These users were from 181\* countries/territories. These analytics demonstrate that the SRAC Publications, Videos, and Computer Software project truly has worldwide reach and impact.

*\*Web-based analytical tracking and reporting methods.*

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## Improved Reproduction in Foodfish (Catfish and Largemouth Bass), Baitfish, and Ornamentals Using a New Spawning Aid (GnRH IIa)

**Reporting Period:** September 1, 2017 - August 31, 2018

**Length of Project:** 2 years

**Current Project Year:** 2

**Total Funds Committed:** \$192,287

**Principal Investigators:** Sylvie Quiniou, Brian Bosworth, *USDA-ARS Warmwater Aquaculture Research Unit*; Chris Green, *Louisiana State University Agricultural Center*; Ken Semmens, Boris Gomelsky, Shawn Coyle, *Kentucky State University Aquaculture Research Center*; Matthew DiMaggio, Craig Watson, *University of Florida Tropical Aquaculture Laboratory*; Cortney Ohs, Jason Broach, *University of Florida*

**Relevance:** Spawning aids have been used by the aquaculture industry to improve fry production for many years as a large number of aquaculture species do not reproduce readily under captive conditions (GnRH IIa (D-Arg6-Pro9-NHet) has garnered recent interest as an alternative GnRH subtype which offers increased biological activity and reliability in channel catfish compared to mGnRH Ia and sGnRH IIIa. Researchers at four institutions will collaborate to evaluate the efficacy, reliability, safety, and mode of action of GnRH IIa in a range of species encompassing foodfish, baitfish, and ornamentals. Ultimately this investigation will help to assess the viability of GnRH IIa as spawning aid for a wide variety of fish species. In addition, the activities listed have been designed to support an INAD application for GnRH IIa.



**Response:** Objective 1: Efficacy of varying doses of GnRH IIa with Domperidone on successful ovulation, latency time, fecundity, fertilization, hatching success, and egg and larval morphometrics was evaluated in red-tailed black sharks, rainbow sharks, upside-down catfish, and featherfin squeaker catfish for Year 2. GnRH IIa was compared to Ovaprim, the current industry standard.

Objective 2: Administration of varying dosages of GnRH IIa as well as positive and negative controls for induced spawning in pinfish and monitoring of spawning performance.

Objective 3: The Safety of GNRH IIa to channel catfish was evaluated by injecting either 1x the recommended spawning induction dose of 100ug/kg versus 3x the dose or 5x the dose and controls (saline). Evaluation of testosterone and oestradiol release by late stage oocytes and pituitary tissue sections from sexually mature channel catfish was evaluated following incubation in *GnRH IIa*, *GnRH Ia*, and *GnRH IIIa* for Year 2. The effects of GnRH IIa implants on maturation of blue catfish males was evaluated. Spermiation and ovulation was evaluated in Largemouth Bass males and females following a single injection of GnRH IIa or GnRHIIIa + Dopamine versus HCG for Year 2.

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**Results:** Objective 1: Spawning trials for all four species were successfully completed during Year 2. Four treatments were evaluated, Ovaprim, 50, 100 and 200ug/kg GnRH IIa + 5mg/kg domperidone, as well as a negative control of propylene glycol. All species exhibited similar ovulation performance across hormone treatments except for *E. frenatum* injected with Ovaprim (17% ovulation). In general, fertilization and hatch success were comparable for both analogs among species. Embryo and larval morphometric measurements were also similar among treatments across all four species. Taken together, these results suggest cGnRH IIa to be a safe and reliable option for induction spawning of ornamental species.

Objective 2: Four dosages (25, 50, 100, 200 µg/kg) of GnRH IIa were evaluated and compared to the control Ovaprim and negative control saline treatments. GnRH IIa was effective for spawning induction for both pinfish and pigfish at all tested dosages. The response time for both pinfish and pigfish was typically 48-72 h after hormone administration. Pinfish experienced a delayed response to hormone administration; this was likely due to cooler water temperatures during the experiment. A spawn from a saline injected (negative control) pigfish was observed. Egg quantity appears higher but with greater variability for GnRH IIa treated pinfish and pigfish females compared to Ovaprim. Egg and larval morphometrics are being analyzed to determine any differences in treatment dosages and hormones.

Objective 3: We administered either 0x (saline control) 1x, 3x or 5x of the dose of GnRH IIa recommended for spawning induction (100 µg/kg) to groups of 10 females. After a week, the fish were euthanized and an autopsy conducted looking for macroscopic lesions on the body or organs. At the same time 10 different tissues were sampled (muscle, posterior and anterior kidney, heart, ovary, liver, spleen, posterior intestine, stomach and gill arch) fixed in 10% neutral buffered formalin and examined microscopically for any lesions or changes of tissue appearance. Both examinations (macroscopic and microscopic) were conducted by trained pathologists. Neither examination detected changes related to the GnRH IIa injections. Therefore, GnRH IIa is deemed safe to the animals to use to induce spawning in channel catfish females.

Estradiol was determined from ovarian fragments incubated for 8 and 16 hours within 10 and 100 µM concentrations of (GnRH IIa, mGnRH Ia, and sGnRH IIIa). Mean Estradiol concentrations increased from 8 to 16 hour incubations for all hormones for both incubation concentrations (10 and 100 µM). There were no significant differences in estradiol concentrations among hormones for either concentrations (10 and 100 µM) or incubation time points (8 and 16 hours). Based on these tissue incubations all three compounds (GnRH IIa, mGnRH Ia, and sGnRH IIIa) produce responses at the gonads. The *in vitro* incubations did not produce significant differences between the spawning hormones within ovarian tissues with the number of fish tested.

Forty-eight 5-year old blue males were implanted with either 0 µg, 100 µg or 500 µg of GnRHIIIa. Six weeks after implantation the fish were euthanized. The testis were dissected, weighted and the sperm extracted. Motility was evaluated for each sperm sample. No significant differences were observed between the 3 groups regarding testis weight or motility. However, it is not possible to say at this point if the lack of effect is due to the implant type used (Alzet, osmotic pump), the protocol (dose used and length of time chosen) or if GnRHIIIa is not efficacious in maturing the testis.

Spawning and milting trials for artificial propagation of Largemouth Bass were conducted for Year 2. Four treatments were evaluated, Saline as a negative control, Human Chorionic Gonadotropin (HCG) as a positive control, GnRH IIa, and GnRH IIa with a dopamine inhibitor. The respective ovulation rate for

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Saline, HCG, GnRH IIa and GnRH IIa with a dopamine inhibitor was 0, 75%, 90%, and 75%, with a mean response of 0, 25.7, 21.0, and 23.8 grams of eggs/kg brooder. Yield from incubated eggs improved from year 1, but remained low with 6.1%, 9.6% and 8.2% respective yields for HCG, GnRH IIa and GnRH IIa with a dopamine inhibitor. Regardless of spawning agent used, it was not possible to strip milt from males.

**Outreach Overview:** This project has not yet yielded results that could be delivered to the public as outreach. We are waiting on the final results before extending our recommendations.

**Targeted Audiences:** The target audiences are primarily stake-holders in fry production, either foodfish, baitfish or Aquarium fish as well as companies in animal health and drug regulatory agencies.

**Outputs:** Outputs are knowledge concerning 1) efficacy to induce ovulation, spermiation and fry production, 2) efficacious dose, and 3) some mechanisms of action of GnRH IIa.

**Outcomes/Impacts:** Data obtained in this project were submitted to the US Food and Drug Administration in partial fulfillment of data requirements for an Investigational New Animal Drug (INAD) permit. The INAD was opened by FDA in February 18, 2018, and granted to AquaTactics LLC. Data collected during farm trials under the INAD will eventually be used to request full labelling of the drug for use in commercial hatcheries.

**Partnerships Developed:** None.

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## Predation Risk and Economic Impact of Lesser Scaup and Piscivorous Waterbirds on Commercial Baitfish and Catfish Production

**Reporting Period:** July 1, 2017 - August 31, 2018

**Length of Project:** 2 years

**Current Project Year:** 2

**Total Funds Committed:** \$296,892

**Principal Investigators:** Luke Roy, *Auburn University*; Anita Kelly, *University of Arkansas at Pine Bluff*; Brian Davis, *Mississippi State University*; Brian Dorr, *USDA-WS NWRC*; Michael Schwarz, *Carole Engle, Virginia Polytechnic Institute & State University*

**Relevance:** Lesser scaup and piscivorous waterbirds, such as double-crested cormorants, consume fish raised via aquaculture and result in economic losses on commercial fish farms. In addition to the losses of fish, there is a financial cost associated with managing piscivorous waterbirds on commercial farms.

**Response:** This research will improve understanding of utilization of baitfish ponds by lesser scaup, species and sizes of fish consumed, and will ultimately generate an economic analysis of baitfish losses. This project will also generate contemporary information on cormorant roost locations, numbers of birds per roost, and roost distance from active and inactive catfish ponds in Mississippi as well as reveal how cormorants modify their use of roost sites as commercial aquaculture decreases. Ultimately, results from this study will allow researchers to estimate economic losses of fish caused by these birds, and generate management



recommendations for producers to ameliorate depredation of fish by waterbirds.

**Results:** During 2017-2018, 628 pond surveys were conducted on 423 individual ponds over 9 survey trips from mid-November through March. We counted 4,746 scaup during all surveys combined which was a substantial increase from the 1,740 scaup counted (173% increase) during all surveys in winter 2016-2017. We collected 267 scaup in winter 2017-2018. Of those, 29% ( $n = 77$  birds) contained sign of fish parts, and the majority of those ( $n = 71$ ) contained fish in the esophagus that we could use to estimate total fish consumption. Of the 267 scaup collected, 15% ( $n = 39$ ) contained <5mg of dried identifiable prey items and were not used in subsequent analysis. Much like the first winter collections, midge larvae was the most common prey item found in birds collected during the second winter and made up 40% of the diet by weight in winter 2017-2018. Unlike collections in the first winter, 2016-2017, fish comprised 18% of lesser scaup diet by weight in winter 2017-2018. Mean fish lengths consumed were 44.4, 39.5, and 48.3 for golden shiners, goldfish, and *Lepomis* spp. respectively and the maximum number of fish found in a single bird was 112 identifiable golden shiners. On average, baitfish farmers reported an average per-acre cost of \$250/acre (range of \$15/acre to \$553/acre). In addition to

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the costs of bird scaring and the direct fish losses, the problem of depredating birds has led to increased inefficiencies in the way farms are managed.

In our second year of study on catfish (2017-2018), we completed 12 aerial surveys and counted 130,684 cormorants across 68 different night roosts. Roosts ranged from 0.1 to 39 kilometers to the nearest aquaculture facility. A total of 338 cormorants were harvested from 22 different night roosts. Stomach contents contained 7,901 identifiable prey specimens, of which 3,333 were measurable. Catfish (*Ictalurus spp.*) represented 33% of the total prey biomass after length-weight formulas were applied to partially digested fish specimens. Other notable species found in the diet included shad (*Dorosoma spp.*), which comprised 57.6% of the total prey biomass, and sportfish, which comprised 9.2% of the diet.

This is the first study to develop on-farm costs of attempts by farmers to prevent losses due to bird depredation. Results demonstrate that the greatest costs are for the trucks and manpower used to chase birds. Fish farmers are spending more money and more time than had previously been thought in efforts to scare birds from their ponds.

**Outreach Overview:** Results from this project will be disseminated through a number of different outlets including state aquaculture association meetings, national aquaculture and wildlife society meetings, trade publications, extension publications, and peer-reviewed scientific journal articles. At this stage of the project stakeholders are being informed of preliminary data and progress made on the project mainly through presentations at state aquaculture association meetings. As more data is collected, other avenues of information dissemination will be utilized.

**Targeted Audiences:** Baitfish, sportfish, and catfish producers, the aquaculture scientific community, and state/federal agencies.



**Outputs:** To date there have been two extension articles published on this work. In addition, nine abstracts, seven oral presentations, and two poster presentations, have been delivered to aquaculture association and scientific meetings since the beginning of this project. Timely delivery of pertinent information gained through this study is being shared with stakeholders and interested parties as it becomes available. Lastly, aerial survey data of cormorant roost counts are provided to USDA APHIS Wildlife Services within 24 hours of collection to support their roost dispersal programs that provide a direct benefit to producers in reducing cormorant depredation.

**Outcomes/Impacts:** The final impact of this collective work cannot yet be ascertained as the study is not yet completed. However, the data related to scaup numbers and predation on baitfish and sportfish will be extremely valuable to commercial producers, as will the economic data revealing the true economic cost of running birds on commercial baitfish and sportfish farms. Likewise, the aerial surveys, diet study, and bioenergetics modeling being carried out with cormorants will also be of great value to the catfish industry. Updated economic costs tracking the cost of controlling piscivorous birds on catfish farms will assist commercial producers in developing management schemes to better control and increase farm efficiencies at managing the risk associated with cormorants.

**Partnerships Developed:** None to date.



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## Commercial Production of Selected Native Freshwater Ornamental Species

**Reporting Period:** September 1, 2017 – August 31, 2018

**Length of Project:** 2 years

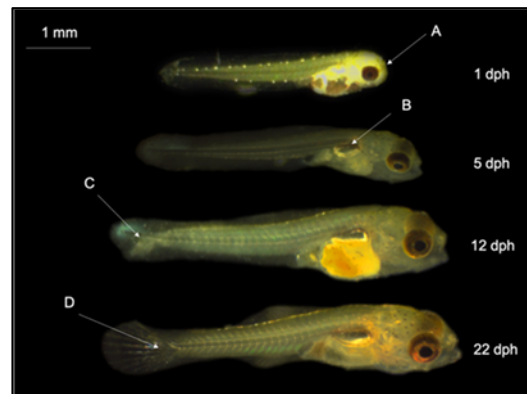
**Current Project Year:** 2

**Total Funds Committed:** \$149,426

**Principal Investigators:** Matthew DiMaggio, Joshua Patterson, Quenton Tuckett, *University of Florida*; Christopher Green, *Louisiana State University*; Donald Orth, Eric Hallerman, Michael Schwarz, *Virginia Tech University*

**Relevance:** Increasing interest in native North American fishes for ornamental use provides opportunities for novel aquaculture endeavors. Scant biological information on this group exists, let alone culture protocols for these species. Currently, native ornamental species sold in the aquarium trade are generally wild caught. The establishment of culture protocols is important for bringing new candidate species into commercial ornamental fish production. Once established, culture protocols can be transferred to industry partners, as well as applied and adapted to related species, including those of conservation concern. The objectives of the proposed research aim to define effective culture protocols for nine species of freshwater fishes endemic to the U.S. Species specific protocols will be immediately transferred to stakeholders within the southern region to aid in commercialization.

**Response:** Broodstock collection continued for the Black Banded Sunfish, Flagfin Shiner, and Sailfin Shiner as well as an additional shiner species, the Metallic shiner *Pteronotropis metallicus*. For the Gulf Coast Pygmy Sunfish and the Metallic Shiner, spawning substrate preference, embryo incubation techniques, egg disinfection protocols and larval feeds were all experimentally evaluated. In addition, development of Gulf Coast Pygmy Sunfish larvae was characterized, with milestones of interest being catalogued. Black Banded Sunfish were successfully spawned, albeit not consistently.



A study was conducted on Golden Topminnows to determine size related fecundity and egg production metrics to provide recommendations for female broodfish size (Small:  $5.7 \pm 0.19\text{g}$ ; Medium:  $8.53 \pm 0.16\text{g}$ ; Large:  $12.2 \pm 0.47\text{g}$ ). Golden Topminnow embryos and newly hatched larvae were measured for length at hatch and yolk volume morphometrics. Bluenose Shiner (10) were stocked in three 1,400-L outdoor mesocosms with 4-5 Longear Sunfish. Each tank was stocked with a sandy/pebble substrate and several species of submerged vegetation. In addition to natural prey items, fish were fed bloodworms twice a week.

Various environmental manipulations were assessed to elicit volitional spawning in captive populations of Rainbow Darter and Mountain Red-belly Dace. Broodstock populations of Rainbow Darter have been further expanded and new cohorts have been conditioned for spawning.

**Results:** Gulf Coast Pygmy Sunfish were characterized by very low fecundity, which may limit large-scale production of the species. A floating yarn substrate was preferred for spawning while the static



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incubation method yielded the best hatch ( $71.25 \pm 22.5\%$ ). Metallic Shiners, also preferred a yarn substrate for spawning and exhibited relatively high batch fecundity (12 eggs/female per spawn), continually spawning for more than four months. Safe egg disinfection protocols were elucidated for both species and elimination of live feeds from larviculture protocols did not negatively impact survival. Aquaculture production protocols established during this project are directly transferable to the commercial ornamental aquaculture industry and will circumvent costly research and development associated with new species production.

The number of Eggs female<sup>-1</sup> day<sup>-1</sup> (including all females each day) produced by Golden Topminnows were  $1.1 \pm 0.70$ ,  $2.0 \pm 0.69$ , and  $5.2 \pm 1.40$  for small, medium, and large female broodfish, respectively. This results in potential daily egg production from the two larger sized female groups at approximately 230 and 415 eggs per day for every 1000 g of female broodfish stocked. The use of outdoor tanks stocked with sunfish as a potential spawning cue for Bluenose Shiners resulted in Bluenose Shiner spawning within the early summer. Predation of larval and juvenile shiner by sunfish spawned at the same time was anticipated to limit the production of larvae from these initial outdoor trials.

Thermal manipulation of Rainbow Darter spawning tanks yielded volitional reproduction, with 722 eggs and 23 larvae collected over the experimental period. Egg disinfection was found to be unnecessary for this species and larvae readily consumed *Artemia* nauplii. Photo-thermal manipulation and introduction of artificial “chub mounds” has been unsuccessful for captive spawning of Mountain Red-belly Dace. Rainbow Darter at the Virginia Tech Hampton facility have also begun to reproduce and egg collection, disinfection, and rearing of fry will continue to aid in determination of fecundity, and establish disinfection and larviculture protocols.

**Outreach Overview:** Results of completed experiments have been disseminated through various presentations at professional meetings. Farm visits and tours of research facilities have also allowed for education of stakeholder groups. Interested commercial clientele are periodically informed of pertinent research develops.

**Targeted Audiences:** Commercial producers.

**Outputs:** Within the reporting period for this project, seven oral presentations and two posters have been delivered at conferences and professional meetings. No publications or manuscripts have resulted from this research to date.

**Outcomes/Impacts:** Culture methods for the Gulf Coast Pygmy Sunfish and Metallic Shiner developed during this project are a solid foundation for the commercial production of select native North American ornamental fishes, which may be transferable to close congeners. Recommendations on Golden Topminnow broodfish size and expected egg output will help producers with production goals for this species as fecundity is low compared to many other ornamental species. Bluenose Shiner appear to require a nest mate associate such as a sunfish to initiate spawning as they potentially use the sunfishes nest as spawning substrate.

**Partnerships Developed:** None to date.

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## Repeatability of Incidence and Time of Ovulation, Fecundity and Fertility in Channel Catfish Females Induced to Ovulate for Production of Hybrid Catfish Fry

**Reporting Period:** September 1, 2017 - August 31, 2018

**Length of Project:** 2 years

**Current Project Year:** 1

**Total Funds Committed:** \$126,619

**Principal Investigator:** Rex Dunham, *Auburn University*; Nagaraj Chatakondi, Brain Bosworth, *USDA-ARS Warmwater Aquaculture Research Unit*; Peter Allen, *Mississippi State University*

**Relevance:** The culture of hybrid catfish (channel catfish, *Ictalurus punctatus*, female X blue catfish, *I. furcatus*, male) is expanding, and this is a key component to the survival of the U.S. farm-raised catfish industry. It is not known if channel catfish females exhibiting good reproduction in one year continue to do so in subsequent years. If they do not have consistently good reproductive performance over time, a significant economic loss and inefficiency may occur. Our overall goal is to determine the repeatability of reproduction in channel catfish females to make hybrid catfish fry over two consecutive years.



**Response:** The repeatability of ovulation, fecundity, fertility and ultimately hybrid catfish fry production/kg in channel catfish females will be determined over a two-year period. A strategy will then be developed to allow farmers to decide which females should be carried over to the next year and which should be culled at the end of the spawning season to increase hybrid catfish fry production efficiency

**Results:** *USDA* - In 2017, a total of 280 catfish were selected, forty fish per week for 7 weeks during the spawning season. The percentage of fish gravid was quite low as 635 fish were evaluated to find the 280 spawnable fish, 37.9% gravid. Percent ovulation of channel catfish ranged from 65 to 95% with an average of 80% ovulation. Percent neurulation ranged from 8 to 32% with an average of 16%. This lower percent neurulation may be attributed to our hatchery water, which could not be stripped of gasses.

Percentage of gravid channel catfish females was much better in 2018, perhaps because of better weather patterns, as 59.8% of the fish evaluated were gravid, and of these, ovulation rate, 78.1%, was similar to 2017. Of the 68 females injected with LHRHa in 2018 that spawned in 2017, 75.0% ovulated, which was not different, 79.6%, for 133 virgin (injected for the first time) females.

Sexual development affected survival post-spawning season. Females that ovulated, gravid and non-gravid females had 34.9, 41.5 and 59.5% survival, respectively, in 2017, which was similar to results at Auburn University (see below). Gravid and non-gravid females had 37.3 and 63.7% post-spawning survival, respectively, in 2018.

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*Auburn University* - A total of 300 channel catfish females were injected over 6 spawning runs in 2017. The weather was unusually cold for the entire spawning season, and appeared to adversely affect the spawning preparation and spawning at Auburn University. Ovulation rates ranged from 38-69% with a mean of 59.6%. This had a major impact on fry output, but some runs produced as many as 2,000 fry/kg. In 2018, 76, 55 and 23 females that did not spawn in 2017, did spawn in 2017, were injected in 2016, but not 2017, respectively were injected with LHRHa to induce ovulation. There was no difference in ovulation rate for females that did not ovulate in 2017, 86.8% and those that did ovulate in 2017, 92.7%. Leaving females fallow for a year did not enhance ovulation rater, 78.3%.

Kansas random strain had higher ovulation percentage than Lake Marepas, 103KS, KxTH, Kmix select and Mix in 2017. However, genetic differences in ovulation rate were minor the following year.

Females that did not ovulate in 2017 had a higher ( $P=0.0001$ ) survival rate (post-spawning 2017 until spawning season 2018) than females that ovulated in 2017 (30.7%). Strain affected survival and ranged from 0% for a commercial strain (limited sample) to 58% for 103KS.

For females that ovulated in both 2017 and 2018, repeatability of performance was essentially zero. The correlation between 2017 and 2018 for egg quality, relative fecundity, and female gravidness was near zero for each trait. Strain effects were minimal for these traits, however, there appeared to be a trend that the poorest ranking strains in 2017 became the best ranking strains in 2018, but this requires further analysis.

**Outreach Overview:** Results were presented at the annual December research meeting for farmers in Demopolis, Alabama. The results were also presented at the Catfish Session at Aquaculture America in New Orleans.

**Targeted Audiences:** The targeted audience includes catfish producers, processors, research scientists, and interested laypersons.

**Outputs:** Several graduate students have been trained. Within the reporting period for this project, two oral presentations have been delivered at conferences and professional meetings.

**Outcomes/Impacts:**

- Based on ovulation rate only, surviving females are good for hybrid embryo production for at least 2 years.
- Letting a female “rest” for 2 years had no benefit.
- Strain affected ovulation in bad spawning years and post-spawning survival.
- Ovulating one year does not affect the probability of ovulating a second year.
- Degree of female readiness, fecundity and egg quality were not repeatable over a 2-year period.
- Hand-stripping increases the probability of death prior to the next spawning season.
- Brood stock handling procedures at research institutions and farms needs improvement if brood females are to be used multiple years.
- Management of brood stock densities is problematic because of potential high mortality that is not detected (fish often do not “float up” after death).

**Partnerships Developed:** None to date.

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## Techniques to Improve Production of Off-bottom Cultured Oysters

**Reporting Period:** September 1, 2017 - August 31, 2018

**Length of Project:** 2 years

**Current Project Year:** 2

**Total Funds Committed:** \$196,780

**Principal Investigator:** Julie Davis, *South Carolina Sea Grant Consortium*; Thomas Bliss, Robert Bringolf, *University of Georgia*; Leslie Sturmer, *University of Florida*; John Supan, *Louisiana State University Agricultural Center*; William Walton, *Auburn University*; Charles Weirich, *North Carolina State University*

**Relevance:** The benefits associated with fine tuning methods to control biofouling when using the OysterGro™ system to grow high value single oysters include: reduced labor costs, improved product quality, improved yield, and shorter grow-out time. The methods used by the emerging oyster aquaculture industry in the Southern U.S. are effective, however, reducing or increasing the frequency of aerial drying and/or applying a fouling release coating could improve the profit margin of the business without impacting or improving product quality. These benefits will allow growers within the Southern U.S. to grow their businesses quicker and take advantage of strong and expanding markets for high value single oysters.



**Response:** The objectives of this project are to:

- 1) Determine the impacts of cage manipulation to decrease biofouling, and evaluate the effects on time to harvest, survival, and morphometric factors, such as meat weight and shell shape (height, length, depth).
- 2) Determine the impacts of antifouling agents to decrease biofouling, and evaluate the effects on time to harvest, survival, and morphometric factors, such as meat weight and shell shape (height, length, depth).
- 3) Determine the economic impact of each methodology on production costs.

**Results:** Within the project period our team deployed the experiment in each state and completed quarterly sampling. The industry partner in each state was responsible for adhering to the flipping routine. On the Gulf Coast the field component of the experiment was also completed during the reporting period because greater than 70% of the oysters in the bi-weekly flipping treatment had reached harvest size and the gear had been exposed to what is considered the heavy fouling season. The field component of the project on Gulf Coast was terminated in June 2018 with harvesting of the product and final sampling. On the Atlantic Coast, by June 2018, 70% or more of the oysters in the bi-weekly flipping treatment had reached harvest size in North Carolina and South Carolina within the reporting period. In Georgia, however, the oysters were smaller. As of June 2018, the gear in each

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Atlantic state had not been exposed to what would be considered a heavy fouling season (i.e. summer). The decision was made to treat the June sampling trip as a 'harvest' sampling for NC and SC. At that time the densities were reduced in each bag as outlined in our proposal and the experimental treatments continued to be applied until the one-year post-deployment time. Sample processing for the Gulf States commenced during the reporting period and carried on into 2019, as did sample processing for the Atlantic States. Each state's report follows and is very similar as the graduate students would travel to each state over the course of two weeks to completed each sampling. Julie Davis left the S.C. Sea Grant Consortium at the beginning of December, 2018. The project lead was changed to Consortium extension leader, Dr. Susan Lovelace on January, 11, 2019. Davis continues work coordinating this project under contract with the Consortium.

In September 2017, 12 cages were deployed at the following locations: Grand Isle, LA; Navy Cove Oyster Company, Fort Morgan, AL and Deer Island, MS; and Cedar Key, FL. At each location, a subsample of at least 100 oysters placed in the cages was saved for future analysis of start size shell metrics (shell height, length and width). Quarterly sampling took place in December 2017, March 2018, and June 2018. At each quarterly sampling, shell metrics of 10 oysters from each bag at each location were collected. During each quarterly sampling trip a photo of a standardized location on the bag was captured for future analysis of the degree of fouling based on flipping routine and/or coating treatment. In May 2018, based on March 2018 data and weekly observations by industry partners, the decision was made to terminate the field portion of the project in June 2018. A conference call amongst PIs determined that the cages had been exposed to a heavy fouling period and no further knowledge regarding the effectiveness of fouling control techniques would be gained from a density reduction and continuation to the one-year post-deployment date. The June 'harvest sampling' trip involved removing, without bias, 25 oysters from each bag for freezing and future measurement of shell metrics, condition index, and fouling. The remaining oysters were left in the care of the industry partner.

In September 2017, 12 cages were deployed at the UGA Demonstration Site, Skidaway Island, GA. Initial subsamples, quarterly sampling, and bag photos were the same as previously described. In May 2018, based on March 2018 data and weekly observations by industry partners, the decision was made to treat the June 2018 sampling as a 'harvest sampling' in order to coordinate with the fact that oysters in NC and SC had reached harvest size. A conference call amongst PIs determined that the cages had not exposed to a heavy fouling period and we would, therefore, continue, at reduced densities, to apply the experimental treatments through until the one-year post-deployment date (October 2018). The June 'harvest sampling' trip involved removing, without bias, 15 oysters from each bag for freezing and future measurement of shell metrics, condition index, and fouling. The remaining oysters were left in the care of the industry partner, with 50 oysters per bag remaining in each cage to be assessed at the one-year post-deployment date.

In September 2017, 12 cages were deployed at Lady's Island Oyster Inc., Beaufort, SC. Initial subsamples, quarterly sampling, and bag photos were the same as previously described. In May 2018, based on March 2018 data and weekly observations by industry partners, the decision was made to harvest the project in June 2018 because 70% or more of the oysters in the bi-weekly flipping treatment had reached 76mm. A conference call amongst PIs determined that the cages had not exposed to a heavy fouling period and we would, therefore, continue, at reduced densities, to apply the experimental treatments through until the one-year post-deployment date (October 2018). The June 'harvest sampling' trip involved removing, without bias, 15 oysters from each bag for freezing and future measurement of shell metrics, condition index, and fouling. The remaining oysters were left in the care

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of the industry partner, with 50 oysters per bag remaining in each cage to be assessed at the one-year post-deployment date.

In September 2017, 12 cages were deployed at Carolina Mariculture, Cedar Island, NC. Initial subsamples, quarterly sampling, and bag photos were the same as previously described. In May 2018, based on March 2018 data and weekly observations by industry partners, the decision was made to harvest the project in June 2018 because 70% or more of the oysters in the bi-weekly flipping treatment had reached 76mm. A conference call amongst PIs determined that the cages had not exposed to a heavy fouling period and we would, therefore, continue, at reduced densities, to apply the experimental treatments through until the one-year post-deployment date (October 2018). The June 'harvest sampling' trip involved removing, without bias, 15 oysters from each bag for freezing and future measurement of shell metrics, condition index, and fouling. The remaining oysters were left in the care of the industry partner, with 50 oysters per bag remaining in each cage to be assessed at the one-year post-deployment date.

**Outreach Overview:** None during the reporting period.

**Targeted Audiences:** Oyster producers in the Gulf and Atlantic States.

**Outputs:** None during reporting period.

**Outcomes/Impacts:** During the reporting period, there are no impacts as the project had yet to be deployed. In the South Atlantic, however, the project realized one significant accomplishment in that as a result of this project, the state of Georgia allowed import of oyster seed with no detectable level of disease into the state. This project also represents the first time floating oyster cages have been permitted for deployment in Georgia and Mississippi.

**Partnerships Developed:** None to date.





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## Evaluation of Protein and Lipid Concentrations in Commercially Available Tilapia Feeds and Their Effect in Intensive Production Systems

**Reporting Period:** September 1, 2017 - August 31, 2018

**Length of Project:** 2 years

**Current Project Year:** 1

**Total Funds Committed:** \$173,333

**Principal Investigator:** David Kuhn, Carole Engle, Jonathan Van Senten, Michael Schwarz, *Virginia Polytechnic Institute & State University*; Rob Ellis, *Astor Farms*; Clip Brock, *Brock Farms*; Delbert Gatlin, *Texas A&M University*.

**Relevance:** In the Southern region of the U.S. we have farmers that use recirculating aquaculture systems (RAS) for intense production of tilapia. Even though we have some understanding of general tilapia nutrition (e.g., in ponds with natural productivity), there is limited information regarding tilapia nutrition in production RAS (e.g., relatively sterile environment) under intense commercial grow out conditions. Furthermore, there is no consensus in the industry that exists whether farmers should use a low protein/lipid (e.g. 36/6) and or high protein/lipid (e.g. 40/10) feed. Both low and high protein/lipid commercial feeds are being used by various farmers. Understanding how these different diets impact fish production, water quality and waste management, and the overall economics will help farmers in the Southern region of the U.S. to be successful.



**Response:** Evaluate typical commercial diet formulations with different levels of protein and lipid in commercial intensive tilapia RAS:

- a) Assess dietary effects on fish production and water quality parameters (i.e., total suspended solids, biochemical oxygen demand, and production of organic matter).
- b) Test diets with fine-tuned protein and lipid levels to see if gross operating income can be increased.
- c) Conduct economic analyses to determine the most cost-effective formulations under RAS conditions.

The project start date has been delayed due to some industry side constraints. The project team needed to make some new arrangements to satisfy the industry participants on the project. The project is ready to begin on the feed manufacturer side. Since one of the tilapia farms has recently installed a new production system we continued to wait during this reporting period until the filtration systems achieve steady state (mature filters). Once the system achieves steady state we will be able to commence with Objective One. Due to logistics, these large feed deliveries will be coordinated for delivery at the same time for both tilapia farm participants. To get things started team members have held several conference calls, have met in person with all industry partners, and performed site visits at the tilapia RAS farms. We have also begun collecting baseline data to enter into our economic software and our water quality model. After the site visits we realized that some adjustments need to be made to the economic model. Those adjustments were completed.



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**Results:** None to report.

**Outreach Overview:** None to report.

**Targeted Audiences:** Commercial tilapia producers in RAS.

**Outputs:** None to report.

**Outcomes/Impacts:** None to report.

**Partnerships Developed:** Cargill Feeds.

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## Field-testing of a Rapid LAMP Assay to Detect the Marine Parasite *Amyloodinium ocellatum* in Commercial Aquaculture Facilities

**Reporting Period:** September 1, 2017 - August 31, 2018

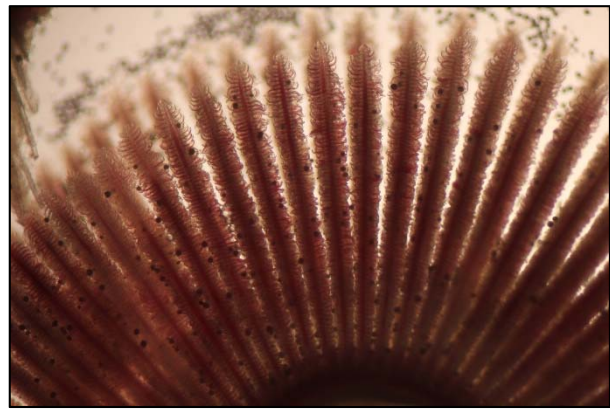
**Length of Project:** 1 year

**Current Project Year:** 1

**Total Funds Committed:** \$92,087

**Principal Investigator:** Reginald Blaylock, Jeffrey Lotz, *University of Southern Mississippi*; Covadonga Arias, Stephen Bullard, *Auburn University*; Roy Yanong, *University of Florida*.

**Relevance:** The dinoflagellate *Amyloodinium ocellatum* (AO) is a major constraint in warm water marine fish culture because it rapidly produces large numbers of infectious stages and is difficult to detect prior to the onset of morbidity. Current, farm-level diagnostics rely on microscopic identification of adult trophonts on the gills and skin. However, microscopic diagnosis is most likely to be accomplished when the trophonts are numerous or at the onset of morbidity, at which point options for treatment and control are limited. Field-testing of a LAMP assay that could detect single parasites in water or fish tissue prior to the onset of morbidity will transition this technology to commercial application and provide enhanced opportunities for improving commercial outcomes.



**Response:** Two molecular diagnostic methods for AO were set up at Auburn University: the PCR protocol described by Levy et al. (2007) and the LAMP assay described by Picon-Camacho et al. (2013). Both protocols target specific AO regions in the 18S rDNA. Positive control was generated from AO infected jack-knife (see below) using universal 18S primers. Regions flanking both LAMP and PCR targets were sequentially amplified and cloned. Several clones (positive controls) for each assay were sequenced to confirmed sequenced targets. The resulting 250 bp fragment was compared to others in GenBank for AO, revealing a small degree of polymorphism.

A total of 33 samples of water and fish (14 from ORA and 19 from ProAquatix) were received at Auburn University and processed. Gills from the fish were examined microscopically and water was filtered onto a membrane and stored at -80°C for future molecular analysis. Only two samples of fish tested positive for AO by microscopy. Positive results were confirmed by bioassay. Samples of the parasite were used for further investigation. Several jack-knife fish were infected with AO. All died within 72 hrs and were sampled for histopathology and microbiology. Tomonts were isolated from the aquarium sediment, skin, and gill of the dead jack-knife fish, placed in glass dishes to observe development and dinospore emergence, or used to infect 6 naive clown anemonefish. All clown anemonefish were readily susceptible to infection and mortality and were subsequently frozen to test freeze-tolerance of AO trophonts in gill. After 36 hrs of freezing, 3 clown anemonefish were thawed, and their gill arches were dissected into individual glass dishes. Tomonts from gill were observed for growth or division and

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exposed to naive clown anemonefish. The exposed fish became heavily infected with AO and died within 5 days.

A sample of AO from the University of Florida was used to establish a laboratory infection at USM/GCRL which is now maintained in a variety of wild and cultured fish including spotted seatrout and Atlantic croaker juveniles from the Thad Cochran Marine Aquaculture Center. A series of laboratory experiments to evaluate the diagnostic power of the LAMP assay in comparison to microscopy was commenced. Replicated groups of 20-L tanks were spiked with either 0, 1, 10, 100, 1000, 5000, or 10,000 dinospores. One-half the tanks in each group was stocked with four spotted seatrout. Ten 1-L aliquots of water taken from each of the fish-free tanks were filtered onto a membrane and frozen for later analysis. After 7 days, fish were killed and two gill arches were examined microscopically. Additionally, 10 samples of gill tissue were randomly taken from fish in each dosage and stored in RNA Later. Preliminary results suggest that the parasite is not detected microscopically at less than the 100 dinospore ( $5 \text{ L}^{-1}$ ) dosage, that the prevalence doesn't approach 100% until the 5,000 dosage ( $250 \text{ L}^{-1}$ ), and that mortality is not observed over 7 days at a dosage less than 10,000 ( $500 \text{ L}^{-1}$ ).

The two collaborating clown fish operations were visited to understand the commercial production process, AO impacts and cost of clownfish production. The cost of applying the LAMP assay was compared to the standard microscopy examination on the commercial clownfish operation. Because only two minor infections occurred during this study, the cost-benefit analysis was augmented by including a PCR detection method.

**Results:** Analytical sensitivity using purified plasmids containing PCR and LAMP target sequences was determined by serially diluting the target to extinction. Original analytical sensitivity for PCR was 9 pg/reaction, similar to what was described by Picon-Camacho et al. PCR primers AO18SR1 and AO18SF1 were redesigned and PCR conditions optimized in an attempt to increase analytical sensitivity. After those modifications, our PCR analytical sensitivity increased to 9 fg/reaction. This was in the same range as the analytical sensitivity reported for the LAMP assay previously. Unfortunately, we were unable to determine the analytic sensitivity for the LAMP assay in our laboratory. Negative controls were positive and despite many attempts to eradicate the contamination issue it was not possible to eliminate it.

Nevertheless, we have generated cloned, positive controls that can be used to determine specific copy number of the target including future applications using qPCR. This cloned controls can be easily shared among diagnostic laboratories and could serve as reference material for future inspection protocols.

We fulfilled Koch's postulates with frozen and thawed tomons demonstrating that freezing in domestic freezers does not kill the parasite. We further obtained a preliminary estimate of the threshold exposure density for microscopic detection. Dinospore densities less than  $5 \text{ L}^{-1}$  did not produce microscopically identifiable infections during the 7 day experiment. A dinospore density of  $500 \text{ L}^{-1}$  is required to produce mortality within 7 days. With respect to the LAMP assay, we obtained positive controls for the original PCR primers (targets LSU) and external primers (targets SSU) and demonstrated repeatedly in 10-fold dilutions that the analytical sensitivity for the original PCR primers using PCR was 4.2 pg/ul and that the analytical sensitivity level for the external primers using PCR was 9.9 fg/ul. We ran the LAMP assay 3 times on different days and demonstrated consistent results.

Cost comparisons between the LAMP assay, microscopy and PCR detection of AO detection was hindered by lack of AO outbreaks. However, equipment and annual maintenance costs to implement the

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three detection methods were calculated. Overall, the microscopy (~\$3,500 to 5,300) method was the least expensive, the LAMP assay (~\$5,300) cost about the same as microscopy, and both cost less than the PCR (~\$20,500 - \$60,000). On a per sample basis (including reagents, DNA extraction, and labor), the LAMP assay costs \$85 per sample; PCR costs \$105 per sample; microscopy, which requires no reagents, no DNA extraction, and minimal labor costs \$30 per sample. Benefits or sale of surviving clown fish were not determined for a specific production cycle but inferred from interviews with the two collaborating businesses which reported typical 50-80 percent survival with the microscopic AO detection methods. Early detection of AO by molecular methods (PCR and/or LAMP) might improve survival to 90%, which would increase the value per tank by \$2,000 to \$8,000. Therefore, IF the LAMP assay could be performed without contamination issues, it could effectively pay for itself by increasing the revenue per tank. The sophistication of each detection method varied greatly. PCR required the most skill followed by LAMP and then by microscopy.

**Outreach Overview:** No extension to date. Once applicability of the LAMP assay is determined, protocols for use will be written and made available to the wider scientific and user communities through refereed publications, articles in trade journals, fact sheets, and the American Fisheries Society Aquatic Animal Health Standards Committee (Blue Book).

**Targeted Audiences:** Producers of marine fish, particularly those producing marine ornamentals.

**Outputs:** None to date.

**Outcomes/Impacts:** Previous to this study, no information was available on freeze-tolerance of the pathogen. Our work has demonstrated that frozen fish can vector viable, infective parasite life history stages that can establish infections in relatively biosecure facilities. Marine aquaculture operations should adapt biosecurity SOPs to incorporate this knowledge. Our work also, for the first time, has quantified the dinospore density in the water required for subsequent microscopic detection of the parasite on fish. Because the parasite can escape microscopic detection at dinospore densities of 10 L-1 and infections in no more than half the fish are likely to be detected at dinospore densities up to 50 L-1, methods for better detecting these early infections would be of substantial benefit to the producer. Cloned controls are currently archived at Auburn University, but are available to aquatic animal health laboratories and researches interested in AO. Using these controls we could determine the exact copy number threshold for detection of the parasite using qPCR. Primers for PCR and LAMP were optimized and analytical sensitivities were assessed. We also demonstrated that the cost of the LAMP assay is not prohibitive because improved survival as a result of early detection potentially produces the revenue that covers the cost of the assay.

The LAMP assay continues to have the potential for providing a substantial benefit to producers, but this project was unable to definitively demonstrate its applicability in the field due to a couple of factors. Overall, the number of samples from the farms was lower than expected and produced only two microscopy-positive samples, which limits the statistical power required to robustly compare the sensitivities of LAMP and microscopy. Additionally, the LAMP assay experienced unresolved contamination issues that prevented its reliable use. As such, assessment of the limited number of positive or potentially positive samples was not done. Farm samples and lab samples destined for molecular analysis remain for future processing once resources to resolve the contamination issue are available.

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**Partnerships Developed:** Oceans, Reefs, and Aquariums (ORA), Ft. Pierce, FL. International industry partner. Clownfish farm that provided water and fish samples for analysis.

Proaquatix, Vero Beach, FL. International industry partner. Clownfish farm that provided water and fish samples for analysis.

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# Products Developed and Students Supported

## Journal Articles and Abstracts

Clements, S. A., B. Davis, B. S. Dorr, K. C. Hanson-Dorr, L. A. Roy, A. M. Kelly, C. R. Engle, S. C. Barras. 2018. Foraging Ecology and Depredation Impact of Scaup on Commercial Baitfish and Sportfish Farms in Eastern Arkansas. Mississippi Academy of Sciences, Summer Student Science Symposium 2018, July 26, 2018, Bost Conference Center, Mississippi State University, Starkville, Mississippi.

Clements, S. A., B. Davis, B. S. Dorr, K. C. Hanson-Dorr, L. A. Roy, A. M. Kelly, C. R. Engle. 2018. Foraging Ecology and the Resulting Economic Impact of Lesser and Greater Scaup on Commercial Baitfish and Sportfish Farms in Arkansas. Aquaculture Workshop and Arkansas Bait and Ornamental Fish Growers Association Annual Meeting. February 8th, 2018 Lonoke, Arkansas.

Dunham, R., Z. Taylor, D. Robinson, M. Coogan, J. North, J. Gurbatow, N. El Hussein, A. Salah, R. Odin, D. Olesen, N. Chatakondi, B. Bosworth and P. Allen. 2019. Repeatability of Incidence and Time of Ovulation, Fecundity and Fertility in Channel Catfish Females Induced to Ovulate for Production of Hybrid Catfish Fry. Aquaculture America 2019. New Orleans. <https://wasblobstorage.blob.core.windows.net/meeting-abstracts/AQ2019AbstractBook.pdf>

Sipos, M.J., T.N. Lipscomb, A.L. Wood, S.W. Ramee, C.A. Watson, and M.A. DiMaggio. (In Revision). Evaluation of cGnRH IIa for Induction Spawning of Two Ornamental Synodontis Species. (Submitted to Aquaculture).

## Book Chapters

Gomelsky, G., Semmens, K. J., Peatman, E., Coyle, S. D. and M.D. Matthews. Reproduction and Genetics. In book: Largemouth Bass Aquaculture, Tidwell, J.H., Coyle, S.D. and L.A. Bright eds., 5m Publishing. (in press)

## Extension/Outreach Publications

Hanson, L.A., W.G. Hemstreet and J.P. Hawke. 2019. SRAC Publication No. 0478, *Motile Aeromonas Septicemia (MAS) in Fish*. 5 pages

Mischke, C.C. 2019. SRAC Publication No. 0469/0471, *Fertilizing Fish Ponds*. 9 pages

Ray, A.J. 2019. SRAC Publication No. 2602, *Indoor Marine Shrimp Farming*. 7 pages

Schwarz, M.H., R. Blaylock, M.A. DiMaggio, E. Saillant and E. Henry. 2019. SRAC Publication No. 0703, *Introduction to Marine Copepod Culture for Live Feeds Production*. 5 pages

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## Oral Presentations

Blaylock, R.B., J.M. Lotz, S.A. Bullard, C.R. Arias, T. Hanson, and R.P.E. Yanong. Field testing a new diagnostic assay for *Amyloodinium ocellatum*. World Aquaculture Society, Cape Town, South Africa, 26–30 June 2017.

Christie, T. W., B. Davis, B. S. Dorr, K. C. Hanson-Dorr, L. A. Roy, A. M. Kelly, C. R. Engle. 2018. Predation Risk of Double-crested Cormorants (*Phalacrocorax auritus*) on Commercial Catfish Production in the Mississippi Delta. Poster presentation. Mississippi Academy of Science Summer Student Science Symposium. Starkville, Mississippi. July 26. (Placed second in the Graduate Student Division)

Clements, S. A., B. Davis, B. S. Dorr, K. C. Hanson-Dorr, L. A. Roy, A. M. Kelly, C. R. Engle, S. C. Barras. 2018. Foraging Ecology and Depredation Impact of Scaup on Commercial Baitfish and Sportfish Farms in Eastern Arkansas. Mississippi Academy of Sciences, Summer Student Science Symposium 2018, July 26, 2018, Bost Conference Center, Mississippi State University, Starkville, Mississippi.

DiMaggio, M., M. Sipos, T. Lipscomb, A. Wood, S. Ramee, E. Groover, and C. Watson. 2019. Evaluation of cGnRH IIa for induced spawning of four ornamental fish species. Aquaculture 2019. New Orleans, Louisiana.

DiMaggio, M., T. Lipscomb, Q. Tuckett, A. Wood, S. Ramee, J. Patterson, and C. Watson. 2019. Evaluation of Culture Protocols for two Florida Native Ornamental Species: *Elassoma Gilberti* and *Pteronotropis metallicus*. 39<sup>th</sup> Annual Meeting of the Florida Chapter of the American Fisheries Society. Haynes City, Florida.

Dunham, R. 2018. Catfish Genetics Update (sub-part: R. A. Dunham, N. Chatakondi, B. Bosworth and P. Allen). Repeatability of Incidence and Time of Ovulation, Fecundity and Fertility in Channel Catfish Females Induced to Ovulate for Production of Hybrid Catfish Fry). Catfish Update Meeting. Demopolis, Alabama.

Dunham, R.A., N. Chatakondi, B. Bosworth, I. Butts, N. El Hussein, A. Salah, Z. Taylor, M. Coogan, J. Gurbatow and P. Allen. 2019. Repeatability of Incidence and Time of Ovulation, Fecundity and Fertility in Channel Catfish Females Induced to Ovulate for Production of Hybrid Catfish Fry. Aquaculture America 2019. New Orleans.

Dutton, H.R., R. Yanong, W. Cai, C.R. Arias, and S.A. Bullard. *Amyloodinium ocellatum* infections in marine aquacultured fishes: A New Host Record and Anecdotal Observations that Indicate Freeze Tolerance. Southeastern Society of Parasitologists, Starkville, Mississippi, 19–21 April 2018.

Dutton, H.R., R. Yanong, W. Cai, C.R. Arias, and S.A. Bullard. *Amyloodinium ocellatum* infections in marine aquacultured fishes: A New Host Record, a Novel DNA Extraction Method, and Anecdotal Observations that Indicate Freeze Tolerance. 43<sup>rd</sup> annual Eastern Fish Health Workshop, Chattanooga, Tennessee. Abstract, National. 9–13 April 2018.

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Fetterman, J. A. and C. C. Green. 2019. Use of Historical Collections to Aid in Reproductive Assessments of the Bluenose Shiner. North American Native Fishes Association Annual Meeting. May 31, 2019, Jackson, MS.

Fetterman, J. A., Murr, C. E., and C. C. Green. 2018. Morphology, Gonadosomatic Index, and Gonad Histology Assessment of the Bluenose Shiner (*Pteronotropis welaka*). Louisiana Chapter of the American Fisheries Society, Annual meeting, May 24-25, 2018, Baton Rouge, LA.

Fetterman, J. A., Murr, C. E., and C. C. Green. 2019. Bluenose Shiner (*Pteronotropis welaka*) Spawning Trials and Suggested Culture Methods. Louisiana Chapter of the American Fisheries Society, Annual meeting, May 23-24, 2019, Thibodaux, LA.

Fetterman, J. A., Murr, C. E., and C. C. Green. 2019. Bluenose Shiner (*Pteronotropis welaka*) Spawning Trials and Suggested Culture Methods. Louisiana Chapter of the American Fisheries Society, Annual meeting, May 23-24, 2019, Thibodaux, LA.

Fetterman, J. A., Murr, C. E., and C. C. Green. 2019. Weight Dependent Intraspecific Variation in Female Golden Topminnow *Fundulus Chrysotus* Reproduction. World Aquaculture 2019, New Orleans.

Gonzales, R.D., C.R. Arias, R.B. Blaylock, and S.A. Bullard. Detecting *Amyloodinium ocellatum* in Cultured Fish: Efficiency Comparisons Between Direct Observation of Trophonts Using Light Microscopy and Detection of Waterborne DNA Using Loop-Mediated Isothermal Amplification (LAMP). Southeastern Society of Parasitologists, Athens, Georgia, 11–13 April 2019.

Lipscomb, T., Q. Tuckett, A. Wood, S. Ramee, J. Patterson, C. Watson, and M. DiMaggio. 2019. Development of Aquaculture Techniques for Two Florida Native Ornamental Fishes: *Elassoma gilberti* and *Pteronotropis metallicus*. Aquaculture 2019. New Orleans, Louisiana.

Semmens, Ken, Shawn Coyle, and Boris Gomelsky, 2019. Reproduction of Largemouth Bass. Abstract, Aquaculture America, 2019, New Orleans, March 7-11

Sharma, Amit, and Kenneth Semmens 2019. Efficacy of a Fertilization Solution Created with Testes Extraction and Maceration in Largemouth Bass, *Micropterus salmoides*, Abstract, Aquaculture America, 2019, New Orleans, March 7-11

Sharma, Amit, K Semmens and B. Gomelsky. 2019. Evaluation of Different Hormonal Spawning-inducing Agents in Largemouth Bass, *Micropterus salmoides*. Abstract, 2019 Association of Research Directors Research Symposium in Jacksonville, Florida (March 31st – April 3rd, 2019).

Sipos, M.J., T.N. Lipscomb, A.L. Wood, S.W. Ramee, E.M. Groover, C.A. Watson, and M.A. DiMaggio. 2018. Evaluation of cGnRH IIa for induced spawning of *Epalzeorhynchus bicolor* and *Synodontis nigriventris*. Aquaculture America 2018. Las Vegas, NV.



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## Poster Presentations

Murr, C. E., Fetterman, J. A., and C. C. Green. 2018. Application of Hormones as a Potential Spawning Aid for a Species of Conservation Concern. Louisiana State University Discover Day, April 10, 2018, Baton Rouge, LA.

Murr, C. E., Fetterman, J. A., and C. C. Green. 2018. Application of Hormones as a Potential Spawning Aid for a Species of Conservation Concern. Louisiana Chapter of the American Fisheries Society, Annual meeting, May 24-25, 2018, Baton Rouge, LA.

## Digital Products

SRAC Home Website: [www.srac.msstate.edu](http://www.srac.msstate.edu)

SRAC Publications Website: <https://srac.tamu.edu/>

SRAC Aquaponics Website: <https://srac-aquaponics.tamu.edu/>

SRAC YouTube Channel: <https://www.youtube.com/channel/UC1VFnlLef2WdHFEVF1O82jA>

AquaPlant Website: <http://aquaplant.tamu.edu/>

## Students Supported

**Jacob Fetterman**, Louisiana State University, Master of Science, Anticipated date of graduation - Summer 2019. Thesis title: *Reproductive Parameters and Methodologies for the Culture of Golden Topminnows (Fundulus chrysotus) and Bluenose Shiners (Pteronotropis welaka)*

**Courtney Murr**, Louisiana State University, Master of Science, Anticipated date of graduation – Fall 2020.

**Angel Cosillo**, Louisiana State University, Undergraduate Student, Anticipated date of graduation - Fall 2019.

**Taylor Lipscomb**, University of Florida, PhD Student, Anticipated date of graduation – Summer 2020, Dissertation title: *Evaluation of Digestive System Ontogeny in Selected Freshwater Ornamental Species to Guide Larval Nutrition Protocols.*

**Shane Ramee**, University of Florida, PhD Student, Anticipated date of graduation – Summer 2019, Dissertation title: *Potential Influences of Environmental Factors on Sex Differentiation in two Freshwater Ornamental Species, Rosy Barb and Dwarf Gourami.*

**Stephen Clements**, Mississippi State University, M.S. degree track, Degree has not been completed (anticipated completion date of May 2019), Thesis title: *Foraging Ecology and Depredation Impact of Scaup on Commercial Baitfish and Sportfish Farms in Eastern Arkansas*

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**Terrel Christie**, Mississippi State University, M.S. degree track, Degree has not been completed (anticipated completion date of May 2019), Thesis title: *Predation Risk of Double-crested Cormorants (Phalacrocorax auritus) on Commercial Catfish Production in the Mississippi Delta.*

**Michael Sipos**, University of Florida, Master's student, Graduated August 2018. Thesis title: *Evaluation of Induced Spawning and Embryo Disinfection Protocols for Four Ornamental Fish Species.*

**Jade Betancourt**, Louisiana State University, Undergraduate student, Graduated Spring 2018.

**Amit Sharma**, Kentucky State University, Master's student, degree in progress.

**Jacob Clark**, hourly undergraduate student at Kentucky State University.

**Haley R. Dutton**, Auburn University. Master of Science, Degree not completed during reporting period, thesis project indeterminate.

**Robert D. Gonzales**, University of Southern Mississippi. Master of Science in progress. Thesis Title: *Assessing the Sensitivity of a LAMP Assay Versus Microscopy in Diagnosing Amyloodinium ocellatum.*

**Shannon Kirk**, University of Georgia, Master's student, Anticipated date of graduation – Summer 2019, Thesis Title: *Efficacy of Biofouling Mitigation Methods for Floating Cage Production of Southeastern Triploid Eastern Oysters.*

**Ellis Chapman**, Louisiana State University, Master's student, Anticipated date of graduation – Summer 2019, Thesis Title: *Comparing Off-Bottom Oyster Aquaculture Techniques in the Northern Gulf of Mexico on Biofouling.*

**Dalton Robinson**, Auburn University, Master of Science

**Nathan Backenstose**, Auburn University, Master of Science

**Zachary Taylor**, Auburn University, Master of Science

**Michael Coogan**, Auburn University, Doctor of Philosophy

**James North**, Auburn University, Doctor of Philosophy

**Jeremy Gurbatow**, Auburn University, Master of Science

**Nour El-Husseini**, Auburn University, Doctor of Philosophy



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## Appendix 1. List of Completed SRAC Projects to Date

### **Evaluation of Probiotic and Prebiotic Supplements with Catfish, Golden Shiners, Hybrid Striped Bass and Tilapia under Conditions of Commercial Production**

Duration: 2015-2017 Funding Level: \$274,308

Participants: TAMU, AU, USDA ARS WARU, UAPB, ESA

### **Improvement of Blue Catfish Germplasm for Hybrid Catfish Production**

Duration: 2014-2017 Funding Level: \$44,343

Participants: USDA ARS WARU, LSU

### **Integrated Approaches to Reducing Individual Variability and Providing Year Round Harvest of Channel-Blue Hybrid Catfish**

Duration: 2015-2017 Funding Level: \$275,232

Participants: AU, USDA ARS WARU

### **Performance Evaluation of Intensive, Pond-Based Culture Systems for Catfish Production**

Duration: 2012-2016 Funding level: \$292,891

Participants: USDA ARS WARU, AU, MSU, UAPB

### **Split-Pond Aquaculture Systems: Design Refinements for Catfish Production and Evaluation for Culturing Other Species**

Duration: 2014-2017 Funding level: \$452,824

Participants: USDA ARS WARU, MSU, AU, USDA ARS NPURU, UAPB

### **Studies to Improve the Control of Virulent *Aeromonas hydrophila* and Evaluate the Impact of Environmental Factors on its Abundance in Catfish Aquaculture Ponds**

Duration: 2014-2016 Funding level: \$354,287

Participants: AU, MSU, USDA NWRC

### **Using National Retail Databases to Determine Market Trends for Southern Aquaculture Products**

Duration: 2009-2015 Funding level: \$397,845

Participants: UAPB, TTU, AU, UF

### **Improving Catfish Broodstock Management by Manipulating Diet, Stocking Densities, and Sex Ratios**

Duration: 2011-2015 Funding level: \$382,463

Participants: UAPB, TAMU, USDA ARS WARU

### **Identification and Removal of Adhesive Proteins from Goldfish and Baitfish Eggs and Egg Masses**

Duration: 2014-2015 Funding level: \$32,432

Participants: LSU, UAPB, UF

### **Implementation of Collective Action Alternatives Identified for the U.S. Catfish Industry**

Duration: 2014-2015 Funding level: \$121,120

Participants: UAPB, AU, UCD, UMo

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**Effects of Mosquito Abatement Pesticides on Various Life Stages of Commercially Important Shellfish Aquaculture Species in the South**

Duration: 2011-2012 Funding level: \$39,973

Participants: Coll. of Charleston, Sanibel-Captiva Conservation Foundation Marine Laboratory

**Development of Baitfish, Goldfish and Ornamental Fish Hatchery Methods**

Duration: 2011-2012 Funding level: \$59,957

Participants: UAPB, LSU, UF

**Reproduction and Larval Rearing of Freshwater Ornamental and Marine Bait Fish**

Duration 2011-2014 Funding level: \$499,400

Participants: UF, LSU, MSU

**Potential Marketing Structures for the Catfish Industry**

Duration: 2011-2013 Funding level: \$244,591

Participants: UAPB, AU, KSU, UCDavis, UMo

**Evaluation of Impacts of Potential “Cap and Trade” Carbon Emission Policies on Catfish, Baitfish, and Crawfish Farming**

Duration: 2011-2013 Funding level: \$119,952

Participants: AU, UAPB, LSU

**Development and Evaluation of Cool-Water Crawfish Baits**

Duration: 2011-2014 Funding level: \$124,326

Participants: LSU, TAMU, AU

**Identifying Determinants for Development of Live-Market Grading Standards for Crawfish**

Duration: 2011-2012 Funding level: \$49,952

Participants: LSU, UAPB

**Improving Reproductive Efficiency of Cultured Finfish**

Duration: 2009-2011 Funding level: \$493,973

Participants: USDA/ARS/CGRU, TAMU-CC, TAMU, AU, UF, UT, UAPB, USDA ARS NRAC

**Economic Forecasting and Policy Analysis Models for Catfish and Trout**

Duration: 2007-2009 Funding level: \$148,335

Participants: UAPB, LSU, MSU, NCSU, UF, AU

**Improving Reproductive Efficiency to Produce Channel x Blue Hybrid Catfish Fry**

Duration: 2004-2008 Funding level: \$460,000

Participants: AU, LSU, MSU, UMem, USDA/ARS CGRU

**Development and Evaluation of Pond Inventory Methods**

Duration: 2007-2009 Funding level: \$294,976

Participants: UAPB, LSU, MSU, UF, UMiss

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**Feed Formulation and Feeding Strategies for Bait and Ornamental Fish**

Duration: 2005-2008 Funding level: \$335,063

Participants: UAPB, TAMU, UF, UG

**Innovative Technologies for Commercial-Scale Aquaculture**

Duration: 2004-2008 Funding level: \$935,726

Participants: AU, CU, LSU, MSU, UAPB, USDA ARS CGRU, USDA ARS NARC

**Identification, Characterization, and Evaluation of Mechanisms for Control of Bolbophorus Trematodes and Columnaris-Like Bacteria Causing Disease in Warm Water Fish**

Duration: 2003-2006 Funding level: \$598,947

Participants: USDA APHIS WS, USDA-ARS SNARC, AU, CU, LSU, MSU, NCSU, UAPB, UT

**National Aquaculture Extension Conference**

Duration: 2002 Funding level: \$4,500

Participants: University of Arizona

**Development of Improved Harvesting, Grading and Transport Technology for Finfish Aquaculture**

Duration: 2001-2003 Funding level: \$750,000

Participants: UMem, MSU, NCSU, UAPB, UF, UT

**Control of Blue-green Algae in Aquaculture Ponds**

Duration: 1999-2001 Funding level: \$836,247

Participants: AU, CU, LSU, MSU, NCSU, UAPB, UG, UMiss, UT

**Management of Aquacultural Effluents from Ponds**

Duration: 1999-2002 Funding level: \$555,353

Participants: AU, LSU, MSU, NCSU, UAPB, Waddell MC

**National Aquaculture Extension Conference**

Duration: 1997 Funding level: \$3,700

Participants: Univ. of Maryland

**Verification of Recommended Management Practices for Major Aquatic Species**

Duration: 1997-2000 Funding level: \$160,305

Participants: AU, LSU, NCSU, UAPB

**Optimizing Nutrient Utilization through Diet Composition and Feeding Strategies**

Duration: 1996-1999 Funding level: \$732,804

Participants: AU, LSU, UMem, MSU, NCSU, LSU, TAMU, UAPB, UG

**Management of Environmentally-Derived Off-Flavors in Warmwater Fish Ponds**

Duration: 1996-1999 Funding level: \$866,281

Participants: AU, LSU, LaTech, UMem, MSU, TAMU, UAPB, UMiss, UT

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**Publications, Videos and Computer Software (Years 1-12)**

Duration: 1995-2008    Funding level: \$826,000

Participants: TAMU

**Improving Production Efficiency of Warmwater Aquaculture Species through Nutrition**

Duration: 1994-1996    Funding level: \$760,466

Participants: AU, ECU, KSU, LSU, UMem, MSU, TAMU, UAPB, UG

**Delineation and Evaluation of Catfish and Baitfish Pond Culture Practices**

Duration: 1994-1997    Funding level: \$332,993

Participants: AU, LSU, MSU, TAMU, UAPB, UG

**Aquaculture Food Safety: Residues**

Duration: 1992-1995    Funding level: \$351,929

Participants: AU, LSU, MSU, TAMU, TennTech, UF, UG

**National Coordination for Aquaculture Investigational New Animal Drug (INAD) Applications**

Duration: 1992    Funding level: \$2,000

Participants: North Central Regional Aquaculture Center

**National Extension Aquaculture Workshop**

Duration: 1991    Funding level: \$3,005

Participants: UAPB, ACES, TAMU

**Educational Materials for Aquaculturists and Consumers**

Duration: 1991-1992    Funding level: \$133,142

Participants: AU, KSU, LSU, MSU, NCSU, OSU, TAMU, UF, UG, UVI

**Characterization of Finfish and Shellfish Aquacultural Effluents**

Duration: 1991-1994    Funding level: \$442,041

Participants: AU, CU, LSU, MSU, NCSU, TAMU, UAPB, UF, UG, VSU, Waddell MC

**Food Safety and Sanitation for Aquacultural Products: Microbial**

Duration: 1991-1995    Funding level: \$535,338

Participants: UT, AU, LSU, UF, UG

**Preparation of Extension Publications on Avian Predator Control in Aquaculture Facilities**

Duration: 1990-1992    Funding level: \$15,000

Participants: TAMU, MSU, UG, USDA APHIS ADC (MS, AR, LA, and S&T Field Station)

**Effect of Nutrition on Body Composition and Subsequent Storage Quality of Farm-Raised Catfish**

Duration: 1990-1992    Funding level: \$822,843

Participants: AU, KSU, LSU, MSU, TAMU, UG

**Harvesting, Loading, and Grading Systems for Cultured Freshwater Finfishes and Crustaceans**

Duration: 1990-1993    Funding level: \$373,952

Participants: LSU, AU, CU, UMem, MSU, UG, USL

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**Immunization of Channel Catfish**

Duration: 1990-1991 Funding level: \$99,789

Participants: AU, LSU, UG

**Enhancement of the Immune Response to *Edwardsiella ictaluri* in Channel Catfish**

Duration: 1990-1991 Funding level: \$98,363

Participants: CU, TAMU, UG

**Develop a Statistical Data Collection System for Farm-raised Catfish and Other Aquaculture Products in the Southern Region**

Duration: 1989-1990 Funding level: \$13,771

Participants: MSU, LSU, AU, UA, TAMU, UG, LU, CU, UF, UT, VTU, USDA NASS

**Performance of Aeration Systems for Channel Catfish, Crawfish, and Rainbow Trout Production**

Duration: 1988-1990 Funding level: \$124,990

Participants: AU, LSU, MSU, NCSU, TAMU

**Analysis of Regional and National Markets for Aquacultural Products Produced for Food in the Southern Region**

Duration: 1988-1990 Funding level: \$346,038

Participants: AU, CU, LSU, MSU, TAMU

**Preparation of Southern Regional Aquaculture Publications**

Duration: 1988-1990 Funding level: \$150,000

Participants: AU, UA, UF, UG, KSU, LSU, MSU, NCSU, UPR, USC, TAMU, UVI



