National Program 106
Aquaculture
Annual Report for Fiscal Year 2023

Introduction

This report communicates research and technology transfer highlights from ARS scientists and their support staffs working in the Aquaculture National Program (NP106) from October 1, 2022, through September 30, 2023. Additional information can be found in individual project annual reports located here.

The ***vision*** for ARS aquaculture research and technology transfer is *to enable science-based use of our natural resources to meet the seafood demands of a growing global population.*

The ***mission*** of NP106 is to conduct research and deliver technologies that improve domestic aquaculture production efficiency and product quality while minimizing impacts on natural resources.

NP106 aligns with the USDA Strategic Plan Goal 2, *Ensure America's Agricultural System is Equitable, Resilient, and Prosperous*; Goal 3, *Foster an Equitable and Competitive Marketplace for All Agricultural Producers*; and Goal 4, *Provide All Americans Safe, Nutritious Food*. Additionally, NP106 supports Objective 4.1, *Increase Food Security through Assistance and Access to Nutritious and Affordable Food*, and Objective 2.2, *Protect Agricultural Health by Minimizing Major Disease, Pests, and Wildlife Conflicts*.

NP106 also supports the USDA Science and Research Strategy Priority 1, *Accelerating Innovative Technologies and Practices and Priority 4: Cultivating Resilient Ecosystems*.

The aim of NP106 is to support a safe and affordable domestic supply of seafood products for 330 million U.S. consumers. Seafood products should be produced in a healthy, competitive, and sustainable aquaculture sector that is supported by almost 3,000 aquaculture farmers producing more than $1.5B farm gate value worth of goods annually. In 2019, the USDA National Agricultural Statistics Service (NASS) published the 2018 Census of Aquaculture updating these statistics for the first time since 2013. The report details many features of aquaculture in the United States, and shows that since 2005, the overall number of farms has dropped from about 3,093 in 2013 to 2,932 in 2018. However, farm gate sales in 2018 increased to more than $1.51B from 2013 level of $1.37B.

In fiscal year (FY) 2023 the ARS Office of National Programs (ONP) contributed to many federal aquaculture activities:

- Leadership in the National Science and Technology Council (NSTC) Subcommittee on Aquaculture was provided by an ARS Co-Chair, Executive Secretary and the Chair of the Science Planning Task Force on developing and reporting progress towards the National Strategic Plan for Aquaculture Research;
- Assistance provided to USDA APHIS towards the development of the National Aquaculture Health Plan and Standards.
- ONP staff co-organized the 2023 United States-Japan Natural Resources Aquaculture Workshop in Freeport, Maine.
- ONP staff led an interagency Federal Working Group that explores opportunities for reducing ocean acidification through the farming of seaweeds and seagrasses.

FY 2023 was the fourth year of externally-reviewed 5-year project plans (2020- 2024) that fall under the six components of the 2020 – 2024 Aquaculture National Program Action Plan:

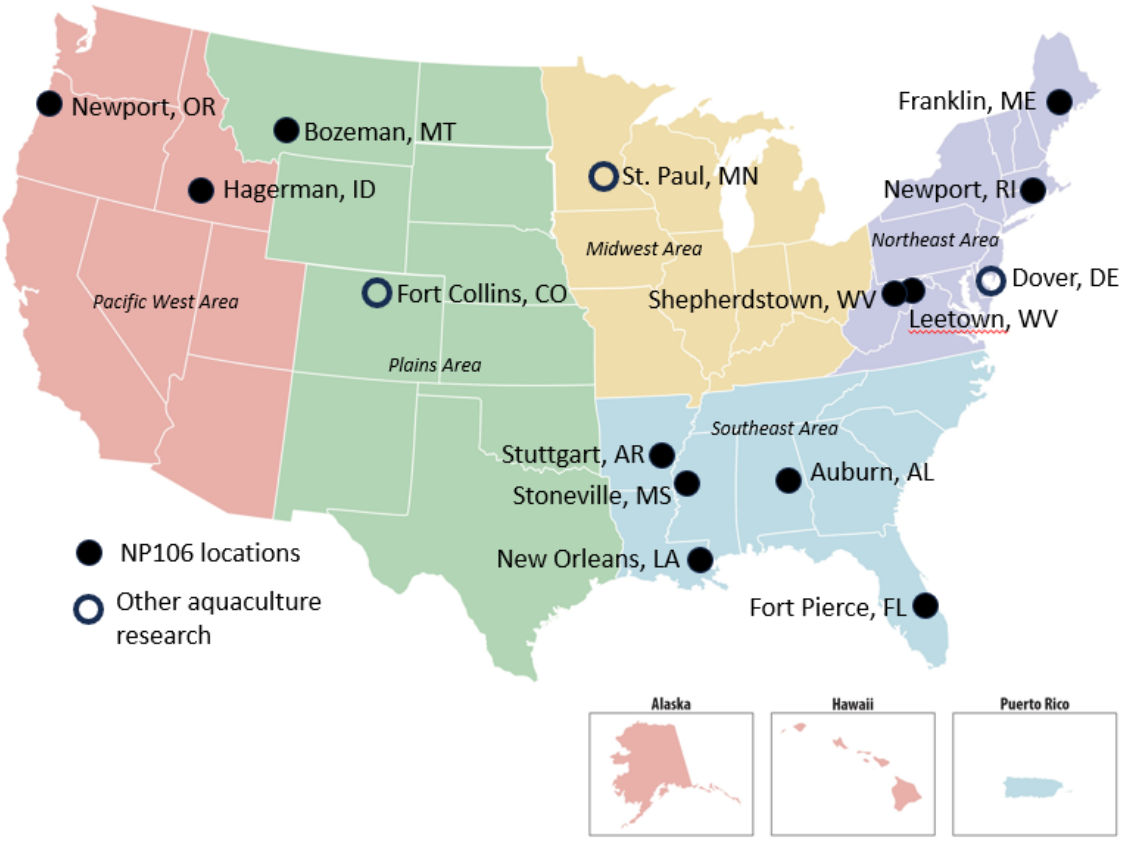
1. Improving the Efficiency and Sustainability of Catfish Aquaculture

2. Improving the Efficiency and Sustainability of Salmonid Aquaculture
3. Improving the Efficiency and Sustainability of Hybrid Striped Bass Aquaculture
4. Enhancing Shellfish Aquaculture
5. Developing Marine Finfish Seedstocks
6. Developing Sustainable Aquaponic Production Systems

Research themes include genetic improvement, reproduction and development, growth and nutrition, fish health, production systems, and product quality.

In 2023, the ARS Office of Scientific Quality and Review conducted a Retrospective Review of NP106 research from FYs 2018-2022. An external panel of U.S. aquaculture experts reviewed publications, technology transfer, research accomplishments, and other information and delivered an assessment that can be found here.

Following the Retrospective Review, ARS and NIFA program leaders hosted a series of eight stakeholder listening sessions to inform the development of the next Action Plan. Six sessions corresponded to the components of the current Action Plan and additional sessions were held to engage internal USDA stakeholders and Tribal aquaculture leaders. A new Action Plan was developed and can be found here. The updated plan provides linkages to higher level strategic plans and Administration priorities, describes programmatic updates for each component, adds a seventh component for tilapia research, and outlines stakeholder input activities and NP106 research capacity.



In 2023, NP106 conducted research at 10 main laboratories on 14 project plans including approximately 67 ARS scientists and University or private cooperators on 15 congressionally

mandated agreements. During FY 2023, ARS base funding for aquaculture research was approximately $55M, not including approximately $807K from incoming grants and agreements.

Although project plans guide most of the efforts of the laboratories, we remain flexible to respond to unanticipated challenges and opportunities. NP106 research covers the spectrum from fundamental to applied research and is focused on solving problems through long-term, high impact research.

Technology Transfer

2023 NP106 New Technology transfer metrics are summarized in **Table 1** below.

Mechanism	# New
Peer Reviewed Journal Articles	85
Book Chapters	3
Cooperative Research and Development Agreements	4
Material Transfer Research Agreements	13
Material Transfer Agreements	10
New Patent Invention Disclosures	3
New Patents (patented)	1

Outreach

NP 106 scientists were also active in serving on committees and as advisors/mentors for undergraduate and post-doctoral students and serving as adjunct/affiliate faculty members as outlined in **Table 2** below.

Advising, Mentorship and Outreach Activities	# of Participants
Advising and Mentorship	
Students and Post-Docs (ARS and Non-ARS)	9
Mentorships	8
Scientists Serving as Student Advisors	2
Adjunct or Other Appointments	7
Student Targeted Outreach	
Student related outreach activities - # of activities (Presentations to schools, science fair participation, student tours/visits to ARS locations)	4
Student related outreach activities - # of student participants (Presentations to schools, science fair participation, student tours/visits to ARS locations)	345
Other Outreach	
Other Outreach Activities - # of activities	11
Other Outreach Activities - # of student participants	87
Other Outreach Activities - # of non-student participants	144

International Collaborations

In 2023, NP106 scientists took part in research collaborations with scientists in the following countries:

DENMARK: ARS researchers in Stuttgart, Arkansas, continue collaborating with researchers at the Technical University of Denmark in Hirtshals on research designed to provide information on peracetic acid used in aquaculture. Collaboration have taken place by phone, email exchanges, and formal meetings at conferences for many years.

FRANCE: A University of Wisconsin-Milwaukee cooperator worked with the Pasteur Institute, Paris, France, in FY 2023. The objective of this collaboration was to analyze F. columnare mutants for virulence against larval zebrafish. This allowed us to expand the significance of cooperator-led research by comparing the effects of mutations on different zebrafish life stages (larval and adult).

There is no formal agreement for this work, and no USDA funds were used to support this collaboration.

GERMANY: An ARS researcher in Stuttgart, Arkansas, continued collaborating with researchers at the Leibniz-Institute of Freshwater Ecology and Inland Fisheries in Berlin on research designed to study the toxicity/effectiveness of peracetic acid to fish and the effectiveness of this compound to control pathogens on fish. Collaboration has taken place by email exchanges, video conferencing, formal meetings at conferences, and reciprocal visits to labs for many years.

MALAYSIA: ARS scientists in Hagerman, Idaho, and Bozeman, Montana, are collaborating with feed companies and the U.S. Grains Council based in Malaysia and Ohio Corn in the United States. The objective of this collaboration is to investigate and increase incorporation of corn distiller's dried grains with solubles (DDGS), a byproduct of ethanol production, in tilapia feeds to expand use in domestic and Asian tilapia aquaculture markets. This collaboration is informal, with no external funding source or formal agreement.

MOZAMBIQUE: Through Mozambique's Embassy in the United States, ARS NP106 was contacted to provide international consultation to researchers at The Center for Aquaculture Research (CEPAQ). CEPAQ is an institution created by the Ministry of Sea Inland Waters and Fisheries of the Government of Mozambique in 2017 as part of a national plan to establish and support the freshwater aquaculture industry. CEPAQ supports breeding of Mozambique tilapia and catfish. A one-day virtual meeting was held on October 20, 2022, during which results of the ARS NCCCWA rainbow trout breeding program for disease resistance were presented and discussed. This research had no external funding source and no formal agreement.

NORWAY: ARS researchers in Stuttgart, Arkansas, continue collaborating with researchers in the Fish Health Department of Nofima (Norwegian Institute of Food, Fisheries and Aquaculture Research) on research designed to show the importance of the potent disinfectant peracetic acid to the global aquaculture industry. Collaboration has taken place by email exchanges, video conferencing, and formal meetings at conferences during the last several years.

NORWAY: ARS researchers in Auburn, Alabama, are conducting collaborative research with Benchmark Genetics Norway AS under a formal agreement. The goal of the research is to determine the feasibility of selectively breeding Nile tilapia for resistance to Streptococcus species and other tilapia pathogens.

NORWAY: Beginning in 2015 and concluding in March 2023, The Conservation Fund Freshwater Institute (TCFFI) personnel have been collaborating in the CtrlAQUA project, an 8-year research initiative by Nofima and funded by the Norwegian Research Council and industry partners. The project aimed to make closed-containment aquaculture systems reliable and economically viable by identifying biological and technological solutions for producing Atlantic salmon. Our specific research contributions included investigating 1) hydrodynamic challenges and optimizing fish culture in huge 1,000+ m3 culture tanks to reduce fixed and variable costs for land-based salmon production; 2) how photoperiod regimes affect Atlantic salmon quality and robustness; 3) whether water ozonation will reduce sexual maturation in post-smolt Atlantic salmon; and 4) the best approaches to disinfecting recirculating aquaculture systems (RAS) and biofilters between production cohorts.

NORWAY: TCFFI personnel began collaborating in 2022 with The Arctic University of Norway (UiT) to contribute and develop RAS expertise and research towards "CandRAS", a 7-year project funded by the Norwegian Research Council. TCFFI personnel contributions involve collaboration on

the design, execution, evaluation, and reporting of experiments in RAS salmon production to address specific areas identified as challenges by RAS salmon producers. A significant component of this collaboration also involves providing expertise and advice on education directed towards employees in the industry, as well as individual courses designed for deeper understanding at the university level. The collaboration also involves short-term visits of personnel, and potential exchange of students, to support mutual U.S.-Norwegian interests in developing and expanding a successful Atlantic salmon RAS industry. Most recently, TCFFI hosted Nofima scientist Dr. Rene Alvestad for a week in March 2023.

NORWAY: EUROPEAN UNION. In 2021, the TCFFI Research Director joined the advisory panel of iFishIENCi (Intelligent Fish feeding through Integration of Enabling technologies and Circular principles), a European Union Horizon 2020 project involving many diverse scientists and industry partners in pursuit of improving aquaculture management and practices through innovation and advanced digital information technology. A major expected benefit of participating in this research initiative is fostering future collaboration to support TCFFI's recent development of a Precision Aquaculture research program. Activities in the reporting period include discussions with other panel members and contributing to policy recommendations for circular aquaculture.

NORWAY: Two one-day meetings were held with scientists from Institute of Marine Research (IMR) Bergen, Norway. ARS scientists from the National Center for Cool and Cold Water Aquaculture (NCCCWA) discussed the use of specific biomarkers for fish health monitoring. The scientists discussed a collaboration and extended work stay for developing non-lethal sampling methods for salmonid fish with the goal of early detection of viral diseases. The meetings were held on December 16, 2022, and May 15, 2023. This research had no external funding source and no formal agreement.

PERU: ARS scientists from the NCCCWA consulted with a scientist from the Instituto del Mar del Perú (IMARPE) Lima via email on *Weissella tructae* and *Yersinia ruckeri* disease outbreaks in Peru and the development of vaccines against these pathogens. This research had no external funding source and no formal agreement.

UNITED KINGDOM: ARS researchers in Bozeman, Montana, established a collaboration between ARS and Aquanzo in the United Kingdom to test novel single cell ingredients. The initial project involves testing artemia meal as a proof of concept for their patented methods of single cell organism production. Single cell organisms offer promise as alternative feed ingredients if cost-effective production methods that do not compromise nutritional content of the meals can be identified.

UNITED STATES: THE CENTER FOR RESPONSIBLE SEAFOOD. Beginning in 2023, TCFFI personnel are collaborating with The Center for Responsible Seafood and The Nature Conservancy to develop solutions to intensify sustainable shrimp production in tanks and small-scale ponds. The focus is on developing strategies to create tank and pond hydrodynamics that optimize the self-cleaning of waste solids and a completely mixed water volume. Technologies developed for optimal salmonid RAS hydrodynamics are being adapted and tested using computational fluid dynamics modeling. Initial results were presented at the Shrimp Summit in Vietnam in July 2023.

Personnel

New scientists in NP106 in 2023:

- **Dr. Luke Iwanowicz,** Research Molecular Biologist, joined the National Center for Cool and Cold Water Aquaculture Research, Leetown, West Virginia.

- **Dr. Nithin Muliya Sankappa,** Post-Doctoral Fellow, joined the Aquatic Animal Health Research Unit, Auburn, Alabama.
- **Dr. Linnea Andersen,** Post-Doctoral Fellow, joined the Aquatic Animal Health Research Unit, Auburn, Alabama.
- **Dr. Johanna Aldersey,** Post-Doctoral Fellow, joined the Aquatic Animal Health Research Unit, Auburn, Alabama.
- **Dr. Julio Garcia,** Research Biologist, joined the Aquatic Animal Health Research Unit, Auburn, Alabama.

Retirements in 2023:

- **Dr. Guangtu Gao,** Physical Scientist-Computational, National Center for Cool and Cold Water Aquaculture Research, Leetown, West Virginia.
- **Dr. Kevin Schrader**, Research Microbiologist, Warmwater Aquaculture Research Unit, Stoneville, Mississippi.

Prominent Awards *(not performance awards)*

The following scientists in NP 106 received prominent awards in 2023:

Dr. Benjamin LaFrentz, received the American Fisheries Society Fish Health Section Special Award for his contribution to the development of the AFS FHS Summer Seminar Series.

Research Results

The following section summarizes the specific research results addressing objectives in the current NP106 Action Plan.

Component 1: Improving the Efficiency and Sustainability of Catfish Aquaculture

Problem Statement 1A: Improve Catfish Aquaculture Production Efficiency

Snail trap prototype. Two species of aquatic snails serve as intermediate hosts for the trematode parasite *Bolbophorus damnificus,* which can infect pond-cultured catfish. Management strategies for the parasite are severely limited, and snail control is the best option for catfish farmers. However, copper sulfate pond treatments to control snails are largely arbitrary because they rely on the farmer's estimate of snail density in each pond. ARS researchers in Stoneville, Mississippi, and Mississippi State University collaborators developed a simple, low-cost snail trap prototype. The prototype used readily available materials, including a cricket cage, PVC pipe, and zip ties. The trap can be deployed into individual ponds to rapidly assess snail populations, enabling farmers to make more informed management decisions in their efforts to control *B. damnificus* host snails.

A vacuum degasser reduces gas supersaturation of well water. Most commercial catfish operations in the southeast United States use water that comes from deep aquifers, but this water is often supersaturated with nitrogen gas that is harmful to fish. ARS researchers in Stoneville, Mississippi, used water sources that mimicked commercial conditions to develop a vacuum degasser that eliminates nitrogen supersaturation from well water without changing other water quality parameters. This research will help researchers and producers develop best practices for improved water quality for fish production in commercial ponds.

Selecting for increased growth and carcass yield and disease resistance in the Delta Select line of channel catfish. ARS researchers in Stoneville, Mississippi, developed the Delta Select line channel catfish with superior growth and carcass yield, and industry responded favorably to the performance of this new line, which was released in 2020. However, more information was needed about how selecting for growth and carcass yield affected other important performance traits in the Delta Select line. ARS researchers in Stoneville, Mississippi, and Mississippi State University researchers compared how feed conversion efficiency and resistance to two important bacterial catfish pathogens (*Edwardsiella ictaluri* and *Edwardsiella piscicidia*) varied between Delta Select line catfish and randomly selected catfish. The Delta Select line catfish grew faster than the Delta Control line, but both lines demonstrated similar feed conversion efficiencies, indicating the faster growth of the Delta Selects was due to greater feed consumption. In experimental challenges, the Delta Control and Delta Select line survived *E. ictaluri* challenge similarly, but the Delta Select had higher survival rates after *E. piscicidia* challenge. The results suggest selection for increased growth and carcass yield in Delta Select line channel catfish has not had a negative impact on feed conversion efficiency or resistance to *E. ictaluri* and may have improved resistance to *E. piscicidia*. This information supports the continued use of the Delta Selects line in the U.S. catfish industry.

Reference genomes for channel and blue catfish. Channel and blue catfish are native to North America and are key to U.S. aquaculture. Efficiently implementing genomic selection in these species requires accurate, complete genome assemblies. ARS researchers in Stoneville, Mississippi, and collaborators utilized state-of-the-art molecular technologies and bioinformatics to produce highly complete DNA sequence assemblies that accurately represent the channel and blue catfish genomes. The new channel catfish genome assembly is a marked improvement over the previous reference assembly, while the blue catfish genome assembly is new. The researchers identified three chromosomes with large structural changes that likely contribute to lowered fertility observed in channel x blue hybrid F1 offspring. The research also revealed sets of genes that are unique within each species and genes that are unique to both species but different from other fish. The new genome assemblies serve as the reference genomes for these species in the GenBank database and will support genomic selection and selective breeding in both species that will improve U.S. catfish production.

Effect of rotenone on catfish ponds. Rotenone is a common chemical therapeutic used to rid ponds of undesirable and/or hold-over fish in catfish ponds before restocking, but previous research in natural water systems showed the recovery of phytoplankton and zooplankton communities could take months or even years following rotenone treatment. ARS researchers in Stoneville, Mississippi, and Mississippi State University collaborators evaluated how rotenone use impacted various aspects of catfish pond environments. Rotenone treatment did not affect water quality or macroinvertebrate communities, and phytoplankton and zooplankton populations recovered within 14 days after application, nearly the same timeframe as the typical wait time for normal management operations. These findings demonstrate that rotenone is an effective therapeutic with no significant long-term effects on key catfish pond ecology. The research was highlighted in the Fish Culture Section of the American Fisheries Society Winter/Spring 2023 newsletter.

Publications:

Engle, C., Hanson, T., Kumar, G. 2022. Economic history of U.S. catfish farming: Lessons for growth and development of aquaculture. Aquaculture Economics & Management. 26(1):1-35. https://doi.org/10.1080/13657305.2021.1896606.

Hedge, S., Kumar, G., Engle, C., Hanson, T., Roy, L., Van Senten, J., Johnson, J., Avery, J., Aarattuthodiyil, S., Dahi, S. 2022. Economic contribution of the U.S. catfish industry. Aquaculture Economics & Management. 26(4):384-413. https://doi.org/10.1080/13657305.2021.2008050.

Hedge, S., Kumar, G., Engle, C., Hanson, T., Roy, L., Van Senten, J., Johnson, J., Avery, J., Aarattuthodiyil, S., Dahi, S. 2022. Technological progress in the US catfish industry. Journal of the World Aquaculture Society. 53(2):67-383. https://doi.org/10.1111/jwas.12877.

Maina, A., Lochmann, R., Rawles, S.D., Rosentrater, K. 2023. Digestibility of conventional and novel dietary lipids in channel catfish *Ictalurus punctatus*. Animals. 13(1456):1-13. https://doi.org/10.3390/ani13091456.

Mischke, C.C., Richardson, B.M., Wise, D.J., Tiwari, A. 2023. Rotenone has little effect on water quality, phytoplankton, zooplankton or macroinvertebrates in aquaculture nursery ponds. North American Journal of Aquaculture. https://doi.org/10.1002/naaq.10276.

Mischke,, C.C., Richardson, B.M., Tiwari,, A., Griffin,, M.J., Wise,, D.J., Rehman,, J.U., Ashfaq,, M.K., Nanayakkara,, P., Khan,, I.A. 2022. Copper toxicity to the ghost rams-horn snail *Biomphalaria havanensis*. North American Journal of Aquaculture. https://doi.org/10.1002/naaq.10279.

Ott, B.D., Bosworth, B.G., Torrans, E.L., Waldbieser, G.C. 2023. Effect of brooder age and size on fry size variation in channel catfish. North American Journal of Aquaculture. https://doi.org/10.1002/naaq.10280.

Richardson, B.M., Reifers, J.G., Walker, C.M., Byars, T.S., Mischke, C.C., Griffin, M.J., Wise, D.J. 2023. Evaluation of snail trap prototype for monitoring the intermediate gastropod hosts of Bolbophorus spp. in commercial catfish ponds in the southeastern United States. Journal of the World Aquaculture Society. 1-13. https://doi.org/10.1111/jwas.12962.

Sun, L., Engle, C., Kumar, G., Van Senten, J. 2022. Retail market trends for seafood in the United States. Journal of the World Aquaculture Society. 54(3):603-624. https://doi.org/10.1111/jwas.12919.

Sun, L., Engle, C., Kumar, G., Van Senten, J. 2022. Trends for U.S. catfish and swai products in retail markets. Aquaculture Economics & Management. https://doi.org/10.1080/13657305.2022.2147250.

Waldbieser, G.C., Liu, S., Yuan, Z., Older, C.E., Gao, D., Shi, C., Bosworth, B.G., Li, N., Boa, L., Kirby, M.A., Jin, Y., Wood, M.L., Scheffler, B.E., Simpson, S.A., Youngblood, R.C., Duke, M.V., Ballard, L.L., Phillipy, A., Koren, S., Liu, Z. 2023. Reference genomes of channel catfish and blue catfish reveal multiple pericentric chromosome inversions. BMC Biology. 21:67. https://doi.org/10.1186/s12915-023-01556-8.

Problem Statement 1B: Reduce the Impacts of Disease in Catfish Aquaculture

Proliferative gill disease risk assessment model. Proliferative gill disease (PGD) is caused by a ubiquitous myxozoan parasite and causes substantial losses in commercially raised catfish. Most losses occur when catfish fingerlings are moved to grow-out ponds for food fish production. ARS researchers in Stoneville, Mississippi, developed a PGD risk assessment model to determine the likelihood of fish losses in newly stocked production ponds or when fish are understocked for food fish production. The risk assessment model relies on comprehensive water analysis and uses eDNA methodologies and a quantitative polymerase chain reaction (PCR) assay specifically developed and validated through this project. By determining the parasite's infectious life stage levels in the pond water, they established correlations between parasite levels and mortality events observed in experimental pond trials, which

enabled them to accurately assess the probability of fish losses upon stocking. This program has been implemented as a demonstration project on farms where water samples are collected concurrently with sentinel fish exposures to identify ponds that can be safely stocked with minimal risk to fish health.

Publications:

Abdelrahman, H.A., Hemstreet, W.B., Roy, L.A., Hanson, T.R., Beck, B.H., Kelly, A.M. 2022. Epidemiology and economic impact of disease-related losses on commercial catfish farms: a seven-year case study from Alabama, USA. Aquaculture. 566:739206. https://doi.org/10.1016/j.aquaculture.2022.739206.

Abernathy, J.W., Zhang, D., Liles, M., Lange, M.D., Shoemaker, C.A., Beck, B.H. 2023. Whole genome sequencing and annotation of seven strains of *aeromonas veronii* isolated from channel catfish. Microbiology Resource Announcements. 12(2):e01231-22. https://doi.org/10.1128/mra.01231-22.

Churchman, E.M., Parello, G., Lange, M.D., Farmer, B.D., Lafrentz, B.R., Beck, B.H., Liles, M.R. 2022. Draft genome sequences of flavobacterium covae strains LSU-066-04 and LV-359-01. Microbiology Resource Announcements. 11(7):e00352-22. https://doi.org/10.1128/mra.00352-22.

Crider, J., Wilson, M., Felch, K.L., Dupre, R.A., Quiniou, S., Bengten, E. 2023. A subset of leukocyte immune-type receptors (LITR) regulates phagocytosis in channel catfish (*Ictalurus punctatus*) leukocytes. Molecular Immunology. 154:33-44. https://doi.org/10.1016/j.molimm.2022.12.009.

Gunn, M.A., Griffin, M.J., Ott, B.D., Rosser, G.T., Wise, D.J., Allen, P.J. 2022. Physiological response of channel (*Ictalurus punctatus*) and hybrid (*I. punctatus* x *I. furcatus*) catfish following *Bolbophorus damnificus* infection. Aquaculture. 563(2). https://doi.org/10.1016/j.aquaculture.2022.739016.

Lange, M.D., Churchman, E.M., Wise, A.L., Bruce, T.J. 2023. A recombinant 9E1 monoclonal antibody binds membrane and soluble channel catfish immunoglobulin M. Fish and Shellfish Immunology Reports. 4:100086. https://doi.org/10.1016/j.fsirep.2023.100086.

Nguyen, K.Q., Bruce, T.J., Oluwafunmilola, E.A., Liles, M.R., Beck, B.H., Davis, A.D. 2022. Growth performance, survival, blood chemistry, and immune gene expression of channel catfish (*Ictalurus punctatus*) fed probiotic-supplemented diets. Veterinary Sciences. 9(12):701. https://doi.org/10.3390/vetsci9120701.

Tuttle, J., Bruce, T., Abdelrahman, H., Roy, L., Butts, I., Beck, B.H., Kelly, A. 2023. Persistence of a wild-type virulent *aeromonas hydrophila* isolate in pond sediments from commercial catfish ponds: a laboratory study. Veterinary Sciences. 10(3):236. https://doi.org/10.3390/vetsci10030236.

Tuttle, J., Bruce, T., Butts, I., Roy, L., Abdelrahman, H., Beck, B.H., Kelly, A. 2023. Investigating the ability of *edwardsiella ictaluri* and *flavobacterium covae* to persist within commercial catfish pond sedi-ments under laboratory conditions. Pathogens. 12(7):871. https://doi.org/10.3390/pathogens12070871.

Wise, A.L., Lafrentz, B.R., Kelly, A.M., Liles, M.R., Griffin, M.J., Beck, B.H., Bruce, T. 2023. The infection dynamics of experimental *Edwardsiella ictaluri* and Flavobacterium *covae* coinfection in channel catfish (*Ictalurus punctatus*). Pathogens. 2023(12):462. https://doi.org/10.3390/. pathogens12030462.

Databases/Datasets Details:

Title: Potential roles of glutathione s-transferases in catfish skin and gills against bacterial infection.

Description: A set of glutathione S-transferases (GSTs) was PCR-amplified from cDNA of channel catfish (*Ictalurus punctatus*) skin and gill tissues. The genes were cloned and sequenced. All recombinant GSTs, expressed in pET system, were active in catalyzing conjugation of reduced glutathione to xenobiotic substrates. The potential role of these enzymes in anti-bacterial pathogens is under investigation. The following are GenBank accession numbers deposited in the National Center for Biotechnology Information (NCBI) database: OR137144-OR137150.

Problem Statement 1C: Improve Catfish Product Quality

The role of dietary geosmin in creating 'muddy' off-flavors in fish. The naturally occurring chemical geosmin is responsible for "muddy" or "earthy" off-flavors in fish and is a major quality issue for fish producers across the aquaculture industry. To date, nearly all off-flavor mitigation research has focused on geosmin accumulation in the water. ARS researchers in New Orleans, Louisiana, demonstrated that geosmin from feed can accumulate in fish fillets. By spiking feed with geosmin, researchers established a new method to impart the off-flavor compound into the edible portions of fish. Now ARS scientists can use dietary exposure to study off-flavor prevention and correction in catfish and other species, and study how geosmin affects sensory quality.

Publications:

Murillo, S. Ardoin, R., & Prinyawiwatkul, W. (2023). Consumers' acceptance, emotions, and responsiveness to informational cues for air-fried catfish (*Ictalurus punctatus*) skin chips. Foods. 12(7). Article 1536. https://doi.org/10.3390/foods12071536

Murillo, S., Ardoin, R., Prinyawiwatkul, W. 2023. Factors influencing consumers' willingness-to-try seafood byproducts. Foods. 12(6). Article 1313. https://doi.org/10.3390/foods12061313.

Schrader, K. 2023. Flavor wheel for sensory analysis of fish raised in recirculating aquaculture systems. North American Journal of Aquaculture. 85:87-91. https://doi.org/10.1002/naaq.10275.

Component 2: Improving the Efficiency and Sustainability of Salmonid Aquaculture

Problem Statement 2A: Improve Salmonid Aquaculture Production Efficiency and Ensure Product Quality

Using artificial intelligence to detect fish mortalities. Early detection of elevated mortalities in aquaculture systems is crucial for timely management to prevent mortality escalation. Conventional mortality detection approaches rely on human observation and tracking that is sometimes augmented with underwater cameras. However, this approach can cause a delay in response times to prevent a significant mortality event, especially when personnel are often or always off-site. In addition, higher stocking densities and cloudy water can obscure visual observation of mortalities. Extramural ARS scientists in Shepherdstown, West Virginia, developed MortCam, an Artificial Intelligence- and Internet of Things (IoT)-enabled fish mortality detection and alert system that provides 24-hour surveillance for RAS conditions and reliably sends email and text alerts to fish production staff about mortality events. This technology will provide RAS farmers with a critical tool for reliable early-mortality detection and notification, which will support effective and timely treatments to prevent mortality escalation, improve fish welfare, and prevent economic losses.

Evaluating the effects of insect meal in fish feeds. The increased demand and reduced supply of fishmeal and fish oil have prompted the search for sustainable alternatives for aquaculture feeds. Insects are part of the natural diet of fish and have a small ecological footprint because they have a limited need for space. ARS researchers in Bozeman, Montana, collaborated with university scientists, trout producers, and insect growers to determine the nutritional value of insects for salmonids. Their study data indicates that insects are promising candidates for fish feeds and results provide feed companies with the information necessary to incorporate insect meal into commercial feed formulations. The global insect feed market is valued at $1.2M and is expected to reach $3M by 2030. A 2020 report indicated that using insect meal as feed resources in commercial salmonid feeds between 2016 and 2020 grew to low but reportable levels (0.4 percent of the total feed volume) for the first time. These levels are projected to increase, and aquaculture feed markets are projected to become the main market for insect meals by 2030.

Optimizing carbon dioxide removal in recirculating aquaculture systems. Fish release dissolved carbon dioxide (CO2), so CO2 levels must be managed in culture tanks to ensure levels are safe for fish. Recirculating aquaculture systems (RAS) commonly bubble air through the water to remove CO2 but management protocols for optimal performance have yet to be defined. Extramural ARS scientists in Shepherdstown, West Virginia, optimized protocols for removing CO2 in RAS using diffused air as the stripping gas in a relatively shallow water column and characterized how water flow, CO2 water levels, diffused airflow rates, and water depths affected CO2 removal efficiencies. These findings provide RAS farmers with management protocols for effectively removing dissolved CO2 using aeration basins in freshwater RAS, improving water quality, fish health, and production efficiency.

Water quality and waste production during Atlantic salmon depuration. Microbial biofilms can accumulate over surfaces within a RAS. The bacteria within these biofilms can produce and release off-flavor compounds that can be taken up by fish and impart objectionable flavors to fillets, thus requiring fish depuration. Depuration is done by holding fish in a separate, biofilm-free system before harvest to eliminate off-flavor compounds from flesh. Although feed is typically withheld while fish are in depuration systems, the fish still add waste (feces and urine) into the system. Best management practices for depurating fish are still being developed in the RAS industry, as water quality and waste production during the depuration process have not been investigated. Extramural ARS scientists in Shepherdstown, West Virginia, performed the first comprehensive analysis of water quality and waste production in an Atlantic salmon depuration system and determined that waste control in depuration systems fully stocked with Atlantic salmon is required for good product quality. A range of practical recommendations and procedural refinements were determined to optimize depuration system performance. These include 1) extending the depuration period, 2) identifying the optimal location for adding depuration system water to RAS to mitigate solids and ammonia contribution, 3) integrating an internal solids removal process within the depuration system design, and 4) managing dissolved oxygen in depuration systems. These novel management strategies are central to optimizing the removal of off-flavor compounds and improving product quality of fish produced in RAS, thus contributing to industry sustainability through better tasting fish.

Fish feed formulations for improving hatchery discharge water quality. Dietary formulation can greatly impact water quality in fish hatcheries. ARS scientists in Hagerman, Idaho, collaborated with the U.S. Fish and Wildlife Service and the University of Idaho and found that ammonia levels in water discharged from rainbow trout production facilities can be significantly reduced by lowering feed protein content from 41 percent to 35 percent. This reduction did not affect growth performance. Researchers also determined that phosphorus (P) bound as phytate-P in plant protein sources, normally

unavailable to rainbow trout, could be utilized by rainbow trout after treatment with phytase enzyme. This allowed rainbow trout to obtain physiological requirements for P, negated the need for additional dietary phosphate, and reduced P discharge in effluent. Phytase additions reduced water-soluble P waste loads by 43 percent from the fishmeal-based feeds and 56 percent from the plant-based feeds. Without phytase treatment of plant-based diets, trout growth performance declined and there was a marked increase in P output. Government agency water quality managers are continually reducing allowable limits of P release from production facilities into downstream rivers and streams, and these restrictions can reduce fish production and growth potential. This information is being used by feed manufacturers and producers to meet water quality requirements, which allows aquaculture producers to increase production while reducing phosphorus discharge from their facilities.

Hybridizing winter and summer spawning lines of trout yields fall spawning broodstock. Rainbow trout lines have been bred and selected to spawn at four different times of the year, necessitating the maintenance and selective breeding of each population to provide year-round egg production of genetically improved eggs. Rainbow trout females will not release their eggs in captivity, requiring hatchery staff to anesthetize and handle female fish once a week to squeeze the abdomen and determine whether eggs have ovulated and can be collected through manual expression. A spawning season with a 3-month window is manageable, but longer spawning seasons (more than 3 months) require more handling events, which can be stressful for fish and is labor intensive for hatchery staff. ARS researchers in Leetown, West Virginia, discovered summer-spawning females that are fertilized with frozen sperm from winter-spawning males produce eggs that yield females. These females, in turn, spawn within a manageable 3-month window in the fall, instead of over a 6 to 12-month window. This hybridization option eliminates the need to maintain and select using a fall-breeding population and suggests hatcheries with a single population can efficiently extend their egg production season with cryopreserved sperm obtained from males with an alternate spawn time.

High fillet yield trait persists across different commercial diets. There is considerable variation in how much fat is found in commercial rainbow trout diets. Some producers desire a lean diet (16-20 percent fat) while others feed a diet containing up to 35 percent fat. ARS scientists in Leetown, West Virginia, used selective breeding to produce a line of rainbow trout exhibiting high fillet yield (ARS-FY-H), but it is unknown if this trait persists across the range of fat levels commonly found in commercial feeds. The scientists compared how diets with three different fat levels affected growth performance, fillet yield, and fillet quality in the ARS-FY-H line and a low fillet yield line (ARS-FY-L). Both lines exhibited similar growth patterns, but the improved fillet yield trait persisted in the high yield line, regardless of dietary fat content, indicating that trout farmers can continue feeding their preferred dietary fat levels without losing the high fillet yield trait. Additionally, the high yield line exhibited a slightly firmer fillet, suggesting that selection for fillet yield does not compromise fillet quality.

Publications:

Ahmed, R.O., Ali, A., Al-Tobasei, R., Leeds, T.D., Kenney, B., Salem, M. 2022. Weighted single-step GWAS identifies genes influencing fillet color in rainbow trout. Genes. 13(8),1331. https://doi.org/10.3390/genes13081331.

Bare, W., Struhs, E., Mirkouei, A., Overturf, K.E., Small, B. 2023. Engineered biomaterials for reducing phosphorus and nitrogen levels from downstream water of aquaculture facilities. Nature Sustainability. 11(4). Article 1029. https://doi.org/10.3390/pr11041029.

Betiku, O., Yeoman, C., Gaylord, T., Ishaq, S., Duff, G., Sealey, W.M. 2023. Evidence of a divided nutritive function in the rainbow trout (*Oncorhynchus mykiss*) midgut and hindgut microbiomes by whole shotgun metagenomic approach. Aquaculture Reports. 30. Article 101601. https://doi.org/10.1016/j.aqrep.2023.101601.

Choudhury, A., Lepine, C., Good, C. 2023. Methane and hydrogen sulfide production from the anaerobic digestion of fish sludge from recirculating aquaculture systems: Effect of varying initial solid concentrations. Fermentation. (9)2:94. https://doi.org/10.3390/fermentation9020094.

Cleveland, B.M., Radler, L.M., Leeds, T.D. 2023. Growth, fillet yield, and muscle quality traits are not affected by a genotype by environment interaction in rainbow trout consuming diets that differ in lipid content. Journal of the World Aquaculture Society. 1-15. https://doi.org/10.1111/jwas.12979.

Davidson, J., Raines, C., Crouse, C., Good, C., Keplinger, B. 2023. Evaluating Brook trout egg and alevin survival at different temperatures in simulated karst environments with marl sedimentation. Southeastern Association of Fish and Wildlife Agencies Conference. 10:27-35.

Davidson, J.W., Reman, N., Crouse, C., Vinci, B. 2022. Water quality, waste production, and off-flavor characterization in a depuration system stocked with market-size Atlantic salmon *Salmo salar*. Journal of the World Aquaculture Society. (54)1:96-112. https://doi.org/10.1111/jwas.12920.

Dupre, R. A., Ardoin, R., Trushenski, J., Jackson, C., Grimm, C., Smith, B. 2023. Dietary uptake of geosmin in rainbow trout (*Oncorhynchus mykiss*). Aquaculture. 571. Article 739458. https://doi.org/10.1016/j.aquaculture.2023.739458

Gao, G., Waldbieser, G.C., Ramey, Y.C., Zaho, D., Pietrak, M.R., Stannard, J.A., Buchman, J.T., Scheffler, B.E., Peterson, B.C., Palti, Y., Rexroad III, C.E., Long, R., Burr, G.S., Milligan, M.T. 2023. The generation of the first chromosome-level de-novo genome assembly and the development and validation of a 50K SNP array for the St John River aquaculture strain of North American Atlantic salmon. G3, Genes/Genomes/Genetics. jkad138. https://doi.org/10.1093/g3journal/jkad138.

Gulkirpik, E., Donnelly, A., Nowakunda, K., Liu, K., Andrade Laborde, J.E. 2023. Evaluation of a low-resource soy protein production method and its products. Frontiers in Nutrition. 10. Article 1067621. https://doi.org/10.3389/fnut.2023.1067621.

L. S. Garcia, A., Tsuruta, S., Gao, G., Palti, Y., Lourenco, T., Leeds, T.D. Genomic selection models substantially improve the accuracy of genetic merit predictions for fillet yield and body weight in rainbow trout using a multi-trait model and multi-generation progeny testing. Genetics Selection Evolution. 55:11 (2013). https://doi.org/10.1186/s12711-023-00782-6.

Lepine, C.A., Redman, N., Murray, M., Lazado, C., Johansen, L., Espmark, A., Davidson, J., Good, C. 2023. Assessing Peracetic Acid application methodolgy and impacts on fluidized sand biofilter performance. Aquaculture Research. 2023:6294325. https://doi.org/10.1155/2023/6294325.

Liu, K. 2022. A new method for determining protein solubility index (PSI) based on extraction with 5 mM alkali hydroxide and its correlation with trypsin inhibitor activity in soybean products. Journal of the American Oil Chemists' Society. 99(10):855–871. https://doi.org/10.1002/aocs.12643.

Liu, K. 2023. Chymotrypsin inhibitor assay: Expressing, calculating, and standardizing inhibitory activity in absolute amounts of chymotrypsin inhibited. Sustainable Food Proteins. 1(1):30-44. https://doi.org/10.1002/sfp2.1004.

Ranjan, R., Sharrer, K., Tsukuda, S., Good, C. 2023. Effects of image data quality on a convolutional neural network trained in-tank fish detection model for recirculating aquaculture systems. Computers and Electronics in Agriculture. 205:107644. https://doi.org/10.1016/j.compag.2023.107644.

Ranjan, R., Sharrer, K., Tsukuda, S., Good, C. 2023. MortCam: An Artificial Intelligence-aided fish mortality detection and alert system for recirculating aquaculture. Aquacultural Engineering. 102:102341. https://doi.org/10.1016/j.aquaeng.2023.102341.

Redmen, N., Straus, D.L., Annis, E.R., Murray, M., Good, C. 2022. Assessing the toxicity of peracetic acid to early Atlantic salmon *Salmo salar* life-stages. Aquaculture Research. (53)14:5097-5104. https://doi.org/10.1111/are.15997.

Weber, G.M., Martin, K.E., Palti, Y., Liu, S., Beach, J.N., Birkett, J.E. 2023. Effects of fertilizing eggs from a summer-spawning line with cryopreserved milt from a winter-spawning line on spawning date and egg production traits in rainbow trout. Aquaculture Reports. 29(101495). https://doi.org/10.1016/j.aqrep.2023.101495.

Problem Statement 2B: Reduce the Impacts of Disease in Salmonid Aquaculture

Improved salmon pathogen detection. Infectious salmon anemia virus (ISAV) is a serious viral pathogen of salmon and is internationally regulated based on virus phenotype. Standard phenotypic detection methods require using a molecular assay that takes days to generate results. ARS researchers in Orono, Maine, and University of Maine colleagues developed an improved molecular method that increased testing speeds five- to ten-fold while maintaining equal if not better accuracy for ISAV detection. This improved assay has been transferred and used by industry stakeholders in a high-pressure, high-volume situation to quickly screen 1,600 broodstock for the presence of the virus and prevented losses of hundreds of thousands of dollars.

Vaccine protection against different strains of Weissella tructae (formerly Weissella ceti). Weissellosis, caused by *W. tructae*, is an economically important emerging disease of farmed rainbow trout that can cause production losses as high as 40-80 percent. Until recently, all strains of *W. tructae* formed a genetically homogeneous group, suggesting a recent emergence of this pathogen. ARS scientists in Leetown, West Virginia, identified a *W. tructae* strain that is genetically distinct from previously characterized strains and likely represents an additional independent emergence of this pathogen. In addition, vaccination experiments established that strong cross-protection is conferred by vaccines prepared from either bacterial strain, indicating that only one strain is needed in the vaccine. This information has been vital to stakeholders for formulating a pre-exposure vaccine to protect against multiple distinct strains of this pathogen.

Water source influences the microbiome at a commercial trout aquaculture facility. Microbiomes can influence development, disease, and overall fish health, but more information is needed about their impact at commercial trout farms. ARS scientists in Leetown, West Virginia, and University of Connecticut partners analyzed 163 microbiome samples collected during 3 years from fish, water, and tank surfaces at a commercial trout production facility. Researchers found that the incoming water microbiome influences the type of microbes associated with fish and tank surfaces and that the fish pathogen *Flavobacterium columnare* was associated with source water and prevalent during disease outbreaks. This study identified a potential source and reservoir of an important pathogen and will lead to improved farm biosecurity and disease control.

Publications:

Conrad, R.A., Evenhuis, J., Lipscomb, R.S., Perez-Pascual, D., Stevick, R.J., Birkett, C., Ghigo, J., Mcbride, M.J. 2022. *Flavobacterium columnare* ferric iron uptake systems are required for virulence. Frontiers in Microbiology. Volume 12. https://doi.org/10.3389/fcimb.2022.1029833.

De Ruyter, T., Littman, E., Yazdi, Z., Adkison, M., Camus, A., Yun, S., Welch, T.J., Keleher, W.R., Soto, E. 2023. Comparative evaluation of booster vaccine efficacy by intracoelomic injection and immersion with a whole-cell killed vaccine against *Lactococcus petauri* infection in rainbow trout (*Oncorhynchus mykiss*). Pathogens. (12)5:632. https://doi.org/10.3390/pathogens12050632.

Good, C., Redman, N., Murray, M., Straus, D.L., Welch, T.J. 2022. Bactericidal activity of peracetic acid to selected fish pathogens in recirculation aquaculture system water. Aquaculture Research. (53)16:5731-5736. https://doi.org/10.1111/are.16031.

Graf, J., Testerman, T., Beka, L., Reichley, S.R., King, S., Welch, T.J., Wiens, G.D. 2022. A large-scale, multi-year microbial community survey of a freshwater trout aquaculture facility. FEMS Microbiology Ecology. 98:1-15. https://doi.org/10.1101/2022.05.03.490559.

Han, Y., Leaman, D.W., Shepherd, B.S. 2023. Ghrelin modulates differential expression of genes relevant to immune activities and antimicrobial peptides in primary head kidney cells of rainbow trout (*Oncorhynchus mykiss*). Animals. 13(10). https://doi.org/10.3390/ani13101683.

Riborg, A., Gulla, S., Colquhoun, D.J., Zeyl Fiskebeck, E., Ryder, D., Verner-Jeffreys, D., Welch, T.J. 2023. Pan-genome survey of the fish pathogen *Yersinia ruckeri* links accessory- and amplified genes to virulence. PLoS Pathogens. (18)5:e0285257. https://doi.org/10.1371/journal.pone.0285257.

Thunes, N.C., Mohammed, H.H., Evenhuis, J., Lipscomb, R.S., Perez-Pascual, D., Stevick, R.J., Birkett, C., Conrad, R.A., Ghigo, J., Mcbride, M.J. 2023. Secreted peptidases contribute to virulence of fish pathogen *Flavobacterium columnare*. Frontiers in Cellular and Infection Microbiology. 13. Article:1093393. https://doi.org/10.3389/fcimb.2023.1093393.

Vallejo, R.L., Evenhuis, J., Cheng, H., Fragomeni, B.O., Gao, G., Liu, S., Long, R., Shewbridge, K., Silva, R.O., Wiens, G.D., Leeds, T.D., Martin, K.E., Palti, Y. 2022. Genome-wide mapping of quantitative trait loci that can be used in marker-assisted selection for resistance to bacterial cold water disease in two commercial rainbow trout breeding populations. Aquaculture. 560(738574). https://doi.org/10.1016/j.aquaculture.2022.738574.

Component 3: Improving the Efficiency and Sustainability of Hybrid Striped Bass Aquaculture

Problem Statement 3A: Enhance Hybrid Striped Bass Aquaculture Production

Thermal protocol developed to create triploid hybrid striped bass. ARS researchers in Stuttgart, Arkansas, and Auburn, Alabama, collaborated with an industry partner on developing a protocol to produce triploid hybrid striped bass. Triploidy typically induces sterility, which producers regard as desirable because mature hybrid striped bass will otherwise divert energy from growth to fertility in the production cycle. In small-scale pilot trials, the researchers applied temperature shocks on freshly fertilized eggs and identified the best cold and warm combination to create triploid hybrid striped bass. They used these protocols in the full-scale commercial production of larvae grown in fish in ponds for 30 days. Results demonstrated that warm temperature shock was better than cold shock to induce a

greater percentage of triploid hybrid striped bass and these pond trials showed the potential for producing triploid hybrid striped bass in aquaculture.

Publications:

Rawles, S.D., Fuller, S.A., Green, B.W., Abernathy, J.W., Straus, D.L., Deshotel, M.B., McEntire, M.E., Huskey Jr, G., Rosentrater, K., Beck, B.H., Webster, C.D. 2022. Effects on growth, body composition, and survival of juvenile white bass (*Morone chrysops*) fed diets without marine fish meal and without supplemental amino acids. Aquaculture Reports. 26. Article 101307. https://doi.org/10.1016/j.aqrep.2022.101307.

Component 4: Enhancing Shellfish Aquaculture

Publications:

Aksoy, M., Eljack, R.M., Beck, B.H., Peatman, E. 2022. Nutritional evaluation of frass from black soldier fly larvae as potential feed ingredient for Pacific white shrimp, *Litopenaeus vannamei*. Aquaculture. 27:101353. https://doi.org/10.1016/j.aqrep.2022.101353.

Aksoy, M., Eljack, R.M., Peatman, E., Beck, B.H. 2022. Immunological and biochemical changes in Pacific white shrimp, *Litopenaeus vannamei*, challenged with Vibrio parahaemolyticus. Microbial Pathogenesis. 172:105787. https://doi.org/10.1016/j.micpath.2022.105787.

Delomas, T.A., Hollenbeck, C.M., Matt, J.L., Thompson, N. 2023. Evaluating cost-effective genotyping strategies for genomic selection in oysters. Aquaculture. 562:738844. https://doi.org/10.1016/j.aquaculture.2022.738844.

Dumbauld, B.R., Du, X., Hunsicker, M., Forster, Z. 2023. Multi-decade changes in the condition index of adult Pacific oysters (*Crassostrea gigas*) in response to climate in a US west coast estuary. Journal of Sea Research. 193. Article 102383. https://doi.org/10.1016/j.seares.2023.102383.

Hernandez, D.P., Abdelrahman, H.A., Galkanda-Arachchige, H.S., Kelly, A.M., Butts, I.A., Davis, D., Beck, B.H., Roy, L.A. 2022. Evaluation of aqueous magnesium concentration on performance of Pacific white shrimp (*Litopenaeus vannamei*) cultured in low salinity water of west Alabama, USA. Aquaculture. 565. Article 739133. https://doi.org/10.1016/j.aquaculture.2022.739133.

Lange, M.D., Abernathy, J.W., Rawles, A.A., Zhang, D., Shoemaker, C.A., Bader, T.J., Beck, B.H. 2023. Transcriptome analysis of Pacific white shrimp (*Liptopenaeus vannamei*) after exposure to recombinant *Vibrio parahaemolyticus* PirA and PirB proteins. Fish and Shellfish Immunology. 132. Article 108502. https://doi.org/10.1016/j.fsi.2022.108502.

Li, N., Pan, T., Griffith, A.W., Dellatore, M., Manahan, D. 2023. Integration of physiological and gene expression analyses to reveal biomarkers for protein dynamic mechanisms regulating higher growth and survival among larval oyster families (*Crassostrea gigas*). Aquaculture. https://doi.org/10.1016/j.aquaculture.2023.739918.

Strebel, L., Nguyen, K., Corby, T., Rhodes, M., Beck, B.H., Roy, L., Davis, A.D. 2023. On demand feeding and the response of Pacific white shrimp (*Litopenaeus vannamei*) to varying dietary protein levels in semi-intensive pond production. Aquaculture. https://doi.org/10.1016/j.aquaculture.2023.739698.

Component 5: Developing Marine Finfish Seedstocks

Problem Statement 5A: Develop Marine Finfish Seedstocks Optimized for Aquaculture Production Efficiency

Whole wheat grain is a good source of carbohydrates for farm-raised Florida pompano. ARS-funded researchers in Fort Pierce, Florida, tested several carbohydrate sources in Florida pompano feeds and found whole wheat grain was the best carbohydrate option for feed formulation. It resulted in better juvenile growth performance and better feed intake and feed efficiency than other carbohydrate sources. Using whole wheat grain to supplement diets ultimately reduces feed cost and waste ammonia is released to the environment without affecting the nutritional balance of feed, which will contribute to the economic sustainability of aquaculture operations.

Publications:

Bradshaw, D., Perricone, C.S., King, L., Allmon, E.B., Sepulveda Soledad, M., Willis, P.S., Riche, M., Kirchoff, N., Mejri, S. 2023. Commercial production of Florida pompano (*Trachinotus carolinus*) larvae at low salinity induces variable changes in whole-larvae microbial diversity, gene expression and gill histopathology. Frontiers in Marine Science. 10:1158446. https://doi.org/10.3389/fmars.2023.1158446.

Component 6: Developing Sustainable Aquaponic Production Systems

Problem Statement 6A: Optimize Aquatic Animal Species Production Systems for Aquaponics

Using black soldier fly larvae frass in an aquaponic system. Aquaponics integrates aquaculture with hydroponics by using fish waste as plant nutrients, but adding supplemental nutrients to enhance plant production is also common. Frass (insect manure and the substrate used to culture insects) can be used to enhance fish and plant production and, depending upon the culture substrate used to grow the insects, it may be possible to enhance both plant and fish growth in an aquaponic system using diet. ARS researchers in Auburn, Alabama, and Stuttgart, Arkansas, collaborated with University of Arkansas at Pine Bluff researchers to evaluate channel catfish juveniles raised in an aquaponic system that were fed diets with black soldier fly larvae (BSFL) frass. Each system had floating raft and media beds used to grow stevia and lavender. Catfish grew significantly better when fed a diet with BSFL frass and their intestinal histology exhibited reduced inflammation. Stevia and lavender had significantly more biomass when frass was added to the system, while plants grown in media beds were larger than plants grown in a floating raft subsystem. These results indicate that feeding catfish BSFL frass and culturing stevia and lavender in media beds can improve overall productivity.

Romano, N., Webster, C.D., Sinha, A., Beck, B.H., Yamamoto, F. 2023. Dietary inclusions of black soldier fly (*Hermetia illucens*) larvae frass enhanced production of channel catfish (*Ictalurus punctatus*) juveniles, stevia (*Stevia rebaudiana*, and lavender (*Lavaridula angustifolia*) in an aquaponic system. Aquaculture. 575:739742.

Publications:

Liu, A., Phillips, K., Jia, J., Deng, P., Zhang, D., Chang, S., Lu, S. 2023. Development of a QPCR detection approach for pathogenic *burkholderia cenocenpacia* from fresh vegetables. Food Microbiology. 115:104333. https://doi.org/10.1016/j.fm.2023.104333.

Thompson, K.R., Webster, C.D., Pomper, K.W., Krall, R.M. 2023. Use of aquaponics project-based environments to improve students' perception of science, technology, engineering, and mathematics (STEM) disciplines and career pathways. Interdisciplinary Journal of Environmental and Science Education. 19(2). Article e2309. https://doi.org/10.29333/ijese/13102.

Additional Research

Black soldier fly byproducts in tilapia diets harmful to liver. Black soldier fly farming offers a sustainable opportunity to provide ingredients for livestock feeds, including fish species. One of the byproducts for black soldier fly larvae (BSFL) farming is the exuviae (exoskeleton) that remains after larvae metamorphose to adults. Exuviae is rich in chitin, which might act as a prebiotic in some species of fish. ARS researchers in Stuttgart, Arkansas, and University of Arkansas at Pine Bluff collaborators evaluated how adding various amounts of ground exuviae to tilapia feed affected fish growth, biochemical composition, hepatic expression, and liver/intestinal histomorphology. Dietary BSFL did not affect growth, feeding efficiency, or fatty acid composition, but histological examinations found mild to severe liver changes ranging from localized inflammation to necrosis and hemorrhaging. Results appear to indicate that BSFL exuviae may be harmful to liver health in tilapia, which could limit its use in fish feed.

Selective breeding of Nile tilapia for resistance to francisellosis does not impact other important commercial traits. *Francisella orientalis* is an important tilapia pathogen that causes francisellosis and results in substantial economic losses for the global tilapia industry. ARS researchers in Auburn, Alabama, and collaborators from Bergen, Norway, and Miami, Florida, assessed options for selectively breeding tilapia with resistance to *F. orientalis* and how this might affect other traits of economic importance. Scientists found fish bred with increasing resistance to *F. orientalis* had high survival rates and did not find unfavorable genetic correlations between commercial traits such as resistance to other pathogens and growth. The results demonstrate that resistance to francisellosis in tilapia can be improved through selective breeding and this strategy will not impact other important commercial traits. This research has led to the development of a robust strain of tilapia that are resistant to disease and exhibit fast growth, both important traits for the tilapia industry.

Publications:

Johnston, A.E., Shavalier, M.A., Scribner, K., Soto, E., Griffin, M.J., Waldbieser, G.C., Richardson, B.M., Winters, A.D., Yun, S., Baker, E.A., Larson, D.L., Kiupel, M., Loch, T.A. 2022. First Isolation of a Herpesvirus (Family Alloherpesviridae) from Great Lakes Lake Sturgeon (*Acipenser fulvescens*). Animals. https://doi.org/10.3390/ani12233230.

Kaimal, S., Farmer, B.D., Renukdas, N., Abdelrahman, H.A., Kelly, A.M. 2022. Evaluating stress mediated microbial pathogenesis in golden shiners, *Notemigonus crysoleucas*. Frontiers in Physiology. 13. Article 886480. https://doi.org/10.3389/fphys.2022.886480.

Kelly, A.M., Renukdas, N., Barnett, L.M., Beck, B.H., Abdelrahman, H.A., Roy, L.A. 2023. The use of kaolin as a prophylactic treatment to prevent columnaris disease (*Flavobacterium covae*) in commercial baitfish and sportfish species. Veterinary Sciences. 10(7):441. https://doi.org/10.3390/vetsci10070441.

Malone, R.F., Alt, D., Pfeiffer, T.J. 2022. Food-based sizing criteria of bead filters with floating media in serial treatment with moving bed bioreactors. Journal of Aquaculture Engineering. 100:1-9. https://doi.org/10.1016/j.aquaeng.2022.102298.

Padeniya, U., Davis, D., Liles, M.R., Lafrentz, S.A., Lafrentz, B.R., Shoemaker, C.A., Beck, B.H., Wells, D.E., Bruce, T.J. 2023. Probiotics impact resistance to *Streptococcus iniae* in Nile tilapia (*Oreochromis niloticus*) reared in biofloc systems. Journal of Fish Diseases. 46:1137-1149. https://doi.org/10.1111/jfd.13833.

Pfeiffer, T.J., Ott, B.D. 2023. Biofiltration reactivation kinetics of a Cyclobio fluidized sand filter in a warmwater recirculating aquaculture system. North American Journal of Aquaculture. 85(2):166-173. https://doi.org/10.1002/naaq.10282.

Shoemaker, C.A., Lozano, C.A., Lafrentz, B.R., Mumma, W.P., Vela-Avitua, S., Ospina-Arango, J., Yazdi, M., Rye, M. 2022. Additive genetic variation in resistance of Nile tilapia (*Oreochromis niloticus*) to *Francisella orientalis* and its genetic (co)variation to both harvest weight and resistance to Streptococcus agalactiae Ib. Aquaculture. 561:738736. https://doi.org/10.1016/j.aquaculture.2022.738736.

Shoemaker, C.A., Lafrentz, B.R., Beck, B.H., Paulson, M.D., Garcia, J.C., Heckman, T.I., Soto, E. 2022. Coinfection by Group C Streptococcus dysgalactiae subsp. equisimilis and *Flavobacterium davisii* in Nile Tilapia, from the United States. European Association of Fish Pathologists. https://doi.org/10.48045/001c.38695.

Straus, D.L., Ledbetter, C.K., Farmer, B.D., Deshotel, M.B., Heikes, D.L. 2023. Toxicity of copper sulfate to largemouth bass fry in naturally soft water. North American Journal of Aquaculture. 85(2):174-177. https://doi.org/10.1002/naaq.10284.

Vela-Avitúa, S., Lafrentz, B.R., Lozano, C.A., Shoemaker, C.A., Ospina-Arango, J., Beck, B.H., Rye, M. 2023. Genome-wide association study for *Streptococcus iniae* in Nile Tilapia (*Oreochromis niloticus*) identifies a significant QTL for disease resistance. Frontiers in Genetics. 14:1078381. https://doi.org/10.3389/fgene.2023.1078381.