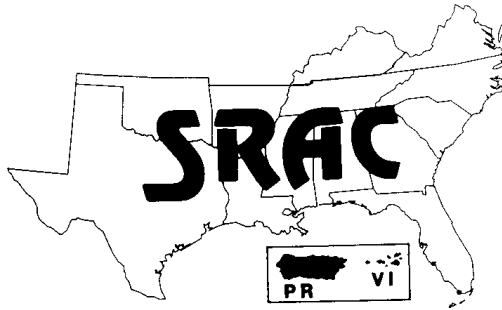


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SOUTHERN  
REGIONAL  
AQUACULTURE  
CENTER



## FOURTEENTH ANNUAL PROGRESS REPORT

For the Period Through August 31, 2001

December, 2001

Southern Regional Aquaculture Center  
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In cooperation with the U.S. Department of Agriculture,  
Cooperative State Research, Education & Extension Service

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# FOURTEENTH ANNUAL PROGRESS REPORT

SOUTHERN REGIONAL AQUACULTURE CENTER

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## **PREFACE**

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In 1980, Congress recognized the opportunity for making significant progress in aquacultural development, and included in Title XIV of the Agriculture and Food Act of 1981 and later, in the Food Security Act of 1985, the authority to establish aquacultural research, development, and demonstration centers in the United States (Subtitle L, Sec. 1475[d]) to enhance viable and profitable aquaculture production for the benefit of consumers, producers, service industries, and the American economy. It was envisioned that the centers would be used in a national program of cooperative research and extension activities in association with colleges and universities, state Departments of Agriculture, federal facilities, and nonprofit private institutions with demonstrated expertise in aquaculture research and development. Eventually, five such centers were established, one each in the northeastern, southern, north-central, western, and tropical Pacific regions of the country. The 1990 Farm Bill (Food, Agriculture Conservation, and Trade Act of 1990; P.L. 101-624) reauthorized funding for the Regional Aquaculture Center program.

Projects that are developed and funded by the Regional Centers are based on industry needs and are designed to directly impact commercial aquaculture development in all states and territories. The Centers are organized to take advantage of the best aquaculture science, education skills, and facilities in the United States. Center programs insure effective coordination and a region-wide, team approach to projects jointly conducted by research, extension, government, and industry personnel. Inter-agency collaboration and shared funding are strongly encouraged.

## **ACKNOWLEDGMENTS**

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The Southern Regional Aquaculture Center (SRAC) would like to acknowledge the contributions of the Project Leaders and Participating Scientists involved in the projects reported in this Fourteenth 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 our Board, IAC and TC, and those serving as Administrative Advisors.

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.

## INTRODUCTION

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Beginning with the first projects funded by SRAC, interest among aquaculture research and extension scientists in SRAC activities has been excellent. 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 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.

The Regional Aquaculture Center program is acknowledged to be an unusually efficient and productive use of Federal funds. The efficiency of the program lies in the unique approach to research, in which problems are identified at the local level and then solved using a coordinated, team approach. The team approach to problem-solving, which makes use of the best scientific talent in each region, results in a highly productive research and extension program. One measure of that productivity is the hundreds of high-quality, peer-reviewed scientific articles, graduate theses, and technical papers that have been generated since inception of the RAC program. As a further illustration of the productivity and impact of the Centers on American aquaculture, nearly 40% of the scientific articles contributed by U.S. scientists to one of the leading international aquaculture journals in a recent year were funded wholly or in part by the RACs. On a more local scale, some feeling for the productivity of the SRAC program can be gained by taking a glance at the number of extension publications produced through our Publications project or the number of peer-reviewed scientific publications generated as part of research projects (see, for example, the publications associated with the “Off-Flavor Management” and “Blue-Green Algae” projects).

A more important measure of success is the extent to which the results of projects have influenced or improved domestic aquaculture. As examples, results of recent or ongoing SRAC projects are being widely adopted by the industry to reduce feed costs, to improve the shelf-life of aquaculture products, and to reduce the incidence of environmentally-derived off-flavors in pond-raised fish.

Research to address the impact of aquaculture on the environment has proven to be particularly valuable. In August 2000, the United States Environmental Protection Agency announced its intention to develop effluent regulations for the aquaculture industry. Most of the technical information submitted to the USEPA to aid in development of sensible, yet effective, rules for warmwater pond aquaculture was derived from past SRAC projects. Further, the scientists and farmers who developed the most recent SRAC aquacultural effluents project in 1998 and 1999 showed remarkable foresight in anticipating the announcement of rulemaking by USEPA. Included in that project are research objectives that may provide key information leading to reasonable approaches to environmental management.

This Fourteenth Annual Progress Report of the Southern Regional Aquaculture Center covers the activities of the Administrative Center during the past year. Progress reports on the six multi-year research and extension projects supported by SRAC during this reporting period cover the life of the projects from their initiation date through August 31, 2001.

## **ORGANIZATIONAL STRUCTURE**

The Agriculture Acts of 1980 and 1985 authorized the 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.

The Board of Directors, the policy-making body for SRAC, utilizes recommendations from an Industry Advisory Council (IAC) and a Technical Committee (TC) to determine priorities for new and continuing aquaculture research and extension projects for the Southern Region. IAC membership represents different segments of the aquaculture industry throughout the region and provides valuable inputs for identifying priorities from an industry perspective. The TC is composed of research and extension scientists from essentially all states within the region and identifies priorities from a technical perspective. These groups provide valuable inputs into the SRAC program by identifying and developing priority research and extension needs in aquaculture. Using recommendations from these two groups, the SRAC Board of Directors selects priority categories for project development and funding.

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 of Directors, Industry Advisory Council, Technical Committee, Steering Committees and project Work Groups are provided by the Administrative Center. This includes monitoring the 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 Office personnel as needed.

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

## **BOARD OF DIRECTORS**

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

The structure of the Board is as follows:

- Three members of the 1862 Southern Extension Service Directors Association
- Three members of the 1862 Southern Experiment Station Directors Association
- One member of the 1890 Association of Research Administrators
- One member of the 1890 Association of Extension Administrators
- One AHS administrator from the host institution

Members of the Board are:

- Harold R. Benson, Kentucky State University
- W. S. Clarke, Virginia State University
- Ivory Lyles, Arkansas Cooperative Extension Service
- J. Charles Lee, Mississippi State University, Chairman
- David Morrison, Louisiana State University
- Daniel Smith, Clemson University Cooperative Extension Service
- Vance Watson, Mississippi State University
- Greg Weidemann, University of Arkansas

Ex-officio Board members are:

- Lester Myers, Chairman, Industry Advisory Council
- Michael Masser, Co-chairman, Technical Committee
- J. Larry Wilson, Co-chairman, Technical Committee
- Craig S. Tucker, Director, SRAC

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 allocation 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 Technical Committee and Industry Advisory Council and guidance from the National Aquaculture Development Plan; (4) review and approval of annual plans of work and accomplishment reports; and (5) final selection of proposals for funding by SRAC.



## **INDUSTRY ADVISORY COUNCIL**

The IAC, which meets at least annually, 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 as deemed appropriate by the Board of Directors.

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 chairman serves for two years and is elected by IAC members.

Members of the IAC are:

Steve Abernathy, LA  
J. Neal Anderson, AR  
James Bardsley, GA  
James P. Ekstrom, TX  
J. B. Hanks, LA  
R. C. Hunt, NC  
Austin Jones, MS  
Robert Mayo, NC  
John Morrison, AL  
Lester Myers, MS (Chairman)  
Bryan P. Plemmons, VA  
Marty Tanner, FL  
Jerry Williamson, AR  
Gary Youmans, SC

IAC members serve up to three-year appointments having staggered terms with options for re-appointment.

The IAC (1) recommends to the Board research and extension needs and priorities from an industry perspective; (2) reviews project proposals and accomplishment and termination reports; and (3) recommends to the Board, jointly with the Technical Committee, actions regarding new and continuing proposals, proposal modifications and terminations.

## **TECHNICAL COMMITTEE**

The TC is composed of representatives from participating research institutions and state extension services, other state or territorial public agencies as appropriate, and non-profit private institutions. Membership of the TC includes research and extension scientists representing essentially all states in the region. The TC meets as needed, but at least annually, and has a co-chairman for research and a co-chairman for extension. Co-chairmen serve for two years and are elected by TC members.

Members of the TC for research are:

David Brune, SC  
Gary Burtle, GA  
Frank Chapman, FL  
Jerry Crews, AL  
Harry Daniels, NC  
Allen Davis, AL  
Delbert Gatlin, TX  
Andrew Goodwin, AR  
John Hargreaves, MS  
Rebecca Lochmann, AR  
Ray McClain, LA  
Stephen Smith, VA  
Jim Tidwell, KY  
J. L. Wilson, TN (Co-chair)

Members of the TC for Extension are:

Jimmy Avery, MS  
David Cline, AL  
Dennis DeLong, NC  
Robert Durborow, KY  
Ruth Francis-Floyd, FL  
David Heikes, AR  
Tom Hill, TN  
Jeff Hinshaw, NC  
Andy Lazur, FL  
Greg Lutz, LA  
Michael Masser, TX (Co-chair)  
Brian Nerrie, VA  
Nathan Stone, AR  
Jack Whetstone, SC

Technical Committee members serve up to three-year appointments having staggered terms with options for reappointment.

The TC (1) recommends to the Board research and extension needs and priorities from a scientific perspective; (2) develops problem statements for research and extension areas under consideration; (3) plans, develops, and implements regional proposals; (4) reviews proposals and accomplishment and termination reports; and (5) recommends to the Board, jointly with the IAC, actions regarding new and continuing proposals, proposal modifications and terminations.

## **PROJECT CRITERIA**

Projects developed within SRAC should meet the following criteria:

- Involves participation by two or more states in the Southern Region.
- Requires more scientific manpower, equipment, and facilities than generally available at one location.
- Approach is adaptable and particularly suitable for inter-institutional cooperation, resulting in better use of limited resources and a saving of funds.
- Will complement and enhance ongoing extension and research activities by participants, as well as offer potential for expanding these programs.
- Is likely to attract additional support for the work which is not likely to occur through other programs and mechanisms.
- Is sufficiently specific to promise significant accomplishments in a reasonable period of time (usually up to 3 years).
- Can provide the solution to a problem of fundamental importance or fill an information gap.

## **PROJECT DEVELOPMENT PROCEDURES**

Research and extension priorities and statements of problems defining priority areas are jointly developed and recommended to the Board by the Industry Advisory Council and the Technical Committee. Using their recommendations as guidelines, the Board selects specific problem areas to be funded and appoints a Steering Committee (comprised of research, extension and industry representatives from the IAC, TC and other agencies) and an Administrative Advisor. The Steering Committee has full responsibility for developing a definitive research and extension Problem Statement, recommending levels of funding for each year of the proposed work, and preparation of the subsequent project proposal.

An Administrative Advisor is appointed by the Board for each active project area, and serves as the coordinator for activities related to the project, providing continuous linkage between the Work Group, Steering Committee and SRAC. Responsibilities of Administrative Advisors are outlined in the SRAC Operations Manual.

Following review of the Problem Statement by the IAC and TC, and review and approval by the Board, announcements to convene an *ad hoc* Work Group are made regionally to (1) institutions and individuals identified by the Steering Committee; (2) extension and research directors of 1862 and 1890 Land Grant Universities within the Southern Region; and (3) other institutions, agencies and organizations within the Southern Region having demonstrated capabilities in the area under consideration.

All *ad hoc* Work Group participants desiring to participate in a proposed research and extension activity must submit a "Commitment to Participate" form. Participants will also have an opportunity to make appropriate comments and suggestions relative to the development of the proposal and their interest and capability in participating. This information is used by the Steering Committee to draft a proposal, recommending the best qualified participants, as well as tentative funding allocations, to address objectives outlined in the Problem Statement.

Project proposals are reviewed by the Steering Committee, IAC, TC, all proposed participants and designated peer reviewers from within the region and from outside the region. The SRAC Director submits the project proposal and peer reviews to the Board of Directors for review and approval. Proposals not approved by the Board are returned for revision or eliminated from consideration.

Final selection of projects and levels of funding are determined by the Board. Most projects have an expected duration of three years. Following final approval by the Board of Directors and CSREES, work described in the research and extension project is implemented. Participating scientists, along with the Steering Committee, comprise the permanent Work Group for the research and extension effort and are responsible for implementation and conduct of the proposed work.

Separate allocations are made for research and extension to ensure strong programs in each of these areas. All funds allocated for extension activities are administered through the respective State Cooperative Extension Services.

## **ADMINISTRATIVE ACTIVITIES**

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The SRAC administrative staff consists of the Center Director and Administrative Assistant. A wide variety of support functions for the various SRAC components, including the Board, TC, IAC, Steering Committees and project Work Groups are provided:

- Center Director serves as an ex-officio member of the Board, TC, and IAC.
- Monitor research and extension activities sponsored by SRAC.
- Solicit and receive nominations for memberships on the TC and IAC.
- Coordinate submission of written testimony to the House Agriculture, Rural Development, and Related Agencies Subcommittee on Appropriations regarding RAC support.
- The Director of SRAC serves as a member of the National Coordinating Council for Aquaculture which consists of the Directors of the five Regional Centers and appropriate USDA/CSREES National Program staff.
- Prepare and submit the Grant Application entering into funding agreement with USDA/CSREES for each fiscal year, and Annual Plans of Work and Amendments to USDA/CSREES.
- Develop and execute appropriate Letters of Agreement with participating institutions in each funded proposal for the purpose of transferring funds and coordinating and implementing projects approved under each of the grants.
- Serve as fiscal agent to review and approve invoices and distribute funds to participating institutions as approved under the grants and as set forth in the Letters of Agreement.
- Prepare budgets for the Administrative Center, track administrative expenditures, and obtain USDA/CSREES approval for project and budget revisions.
- Prepare budget reports for the Board of Directors, tracking expenditures and status of funded projects and the Administrative Center.
- Assist Steering Committees and Work Groups with preparation and revision of proposals for technical and scientific merit, feasibility and applicability to priority problem areas.
- Solicit and coordinate national reviews of project proposals.
- Distribute extension fact sheets, research publications and videos to research and extension contacts throughout the Southern Region, other RACs, USDA personnel, and the Aquaculture Information Center.
- Produce and distribute the "SRAC Annual Progress Report," which includes editing and proofreading the project reports, designing and, using desktop publishing, producing camera-ready copy.
- Produce and maintain the web site for SRAC which provides downloadable copies of all SRAC fact sheets, the Operations Manual and Annual Reports, as well as lists of other research publications and extension contacts in the Southern Region.
- Prepare and distribute Work Group announcements and Requests for Proposals to research and extension directors and other interested parties throughout the Southern Region.
- Respond to numerous requests from aquaculture producers, the public, and research and extension personnel for copies of fact sheets, research publications and videos produced by SRAC and the other Centers, as well as requests for general aquaculture-related information.



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## **PROGRESS REPORTS**

The following cumulative reports detail the progress of research and extension work accomplished for the duration of the respective projects through August 31 of the current year. These reports are prepared by the Project Leaders in conjunction with the institutional Principal Investigators.

Publications, Videos and Computer Software .....	Page 15
Management of Environmentally-Derived Off-flavors in Warmwater Fish Ponds .....	Page 20
Verification of Recommended Management Practices for Major Aquatic Species .....	Page 34
Control of Blue-green Algae in Aquaculture Ponds .....	Page 40
Management of Aquacultural Effluents from Ponds .....	Page 65
Development of Improved Harvesting, Grading and Transport Technology for Finfish Aquaculture .....	Page 79

## **PUBLICATIONS, VIDEOS AND COMPUTER SOFTWARE**

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**Reporting Period**  
April 1, 1995 - August 31, 2001

<b>Funding Level</b>	Year 1 .....	\$50,000
	Year 2 .....	60,948
	Year 3 .....	45,900
	Year 4 .....	60,500
	Year 5 .....	67,000
	Year 6 .....	80,550
	Total .....	\$364,898

**Participants** Texas A&M University System serves as Lead Institution, with Dr. Michael Masser as Project Leader. Participants in this project include authors and co-authors from all states in the region as shown in the listing of publications at the end of this report.

**Administrative  
Advisor** Dr. Daniel Smith, Director  
South Carolina Cooperative Extension Service  
Clemson University  
Clemson, South Carolina

### **PROJECT OBJECTIVES**

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1. Review and revise, as necessary, all SRAC Extension printed and video publications.
2. Establish an ongoing project location to develop and distribute new SRAC educational publications and videos for Southern Region aquaculture industries. This project will be responsible for preparation, peer review, editing, reproduction, and distribution of all Extension and popular-type publications for all SRAC projects.
3. Place current, revised, and new publications in electronic format (e.g., Internet or compact disk) for more efficient use, duplication, and distribution.

### **ANTICIPATED BENEFITS**

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The most direct benefit from this project to the aquaculture industry is the widespread and ready availability of detailed information on produc-

tion and marketing of aquacultural products. SRAC fact sheets, videos, and other publications are distributed worldwide to a diverse clientele.

**Extension Specialists.** 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 benefit of utilizing a region-wide pool of expertise to develop materials for distribution through the nationwide network of Extension Specialists and County Agents. This process makes efficient use of personnel at the State level, and results in high-quality educational materials that are readily available to scientists, educators, producers, and the general public.

**Educators.** Several colleges and universities in the United States use SRAC technical fact sheets as reference material in aquaculture and fisheries courses. Educational institutions at the elementary and secondary level use SRAC extension materials in the classroom to make students aware of aquaculture production and associated trades as a possible vocation.

**Consumers.** Information is readily available for consumers who are seeking background information on aquaculture.

## *Results at a glance...*

*Titles of some recent SRAC publications:*

- ☆ *Cultivating the Eastern Oyster*
- ☆ *Water Gardens*
- ☆ *Processing Channel Catfish*
- ☆ *Trout Production: Handling Eggs and Fry*
- ☆ *Species Profiles of Mutton Snapper and Southern Flounder*
- ☆ *Common Farm-Raised Baitfish*

**Producers.** Information on the use of therapeutants, pesticides, methods of calculating treatment rates, and possible alternative crops and marketing strategies is in constant demand by aquaculturists. Videos that demonstrate such techniques are a ready source of “how-to” information.

**Potential investors.** Detailed information on production and marketing constraints and ways to alleviate or manage those constraints is particularly helpful to people making decisions about entering the aquaculture business. Economic information is used by lending agencies and potential investors, as well as established producers who use the information to help make day-to-day decisions on farm management.

**Internet access.** Availability of SRAC publications via the Internet and compact disk makes access faster and easier, facilitates searching for needed information, and reduces storage space requirements for printed documents.

## *Results at a glance...*

- ☆ *113 authors from across the United States have contributed to SRAC's publications projects.*

## **PROGRESS AND PRINCIPAL ACCOMPLISHMENTS**

During this project year, six new fact sheets were written and six were revised. All have been distributed throughout the Southern Region and

to interested Extension Specialists in other regions. A web site “AQUAPLANT” has been updated with information of aquaculture



producers. Approximately thirteen fact sheets are currently in some stage of writing or revision.

All SRAC publications are based on research conducted within the region or in surrounding areas. Research funding from universities

within the region, as well as funding from private sources, has been used to support the work on which the fact sheets are based. Copies of all fact sheets are available at <http://www.msstate.edu/dept/srac> on the Internet.

## WORK PLANNED

During the next project year, seven fact sheets will be revised and seven new fact sheets will be produced. Additionally, a CD will be developed with all fact sheets and a video produced on freshwater shrimp aquaculture. The new fact sheets will address 1) extensive culture of oysters, 2) broodfish management of catfish, 3) partitioned aquaculture system, 4) common fish parasites, 5) winter kill syndrome, 6) white

grub parasites, and 7) building a simple recirculating system.

Seven fact sheets will be revised on the following topics: 1) levee-type pond construction, 2) baitfish feeds, 3) baitfish feeding techniques, 4) crawfish harvesting, marketing, and economics, 5) pond aeration, 6) pond aeration equipment, and 7) ammonia in fish ponds.

## IMPACTS

This is a highly productive project with significant regional and national impact. Fact sheets and videos are requested and used by clientele in all 50 states on a regular basis. Within the Southern Region, more than 80 fact sheets and six videos are distributed on request daily. Fact sheets generated within the Southern Region are also widely distributed by RACs and extension personnel in other regions. An average of 5 to 20 SRAC fact sheets and 3 videos are distributed daily from each of the other four regions. This means that about 20,000 fact sheets and 3,200 videos per year are used by interested producers or consumers. In addition to direct requests for printed material, fact sheets and other informational materials are accessed daily from the SRAC web site by people searching for technical information. Since the fact sheets are also accessible through numerous other university research and extension web sites, the total usage and impact is undoubtedly several times greater.

### *Results at a glance...*

- ☆ *Twelve fact sheets were completed this year with 13 more in progress.*
- ☆ *Twenty-seven scientists from across the Southern Region contributed to publications produced by SRAC this year.*
- ☆ *SRAC has now published 163 fact sheets and 20 videos.*
- ☆ *Educators in schools and colleges use SRAC publications in classrooms throughout the U.S. and the world.*

Publications and videos produced by SRAC are increasingly used in educating high school and

college students about aquaculture. In recent years there has been a rapid expansion of aquaculture curricula in high schools. These programs heavily utilize our publications and videos for educational purposes but usage is impossible to measure because many people access the information from Internet sites. Aquaculture and fisheries courses taught at several colleges and universities also use SRAC technical fact sheets as part of the reference material used in the course.

### *Results at a glance...*

☆ All fact sheets completed by this project to date are available on the Internet at <http://www.msstate.edu/dept/srac>

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 indicates 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, although it has not been quantified. 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.

## PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

### Fact Sheets Completed (7/1/00 - 8/31/2001)

- Daniels, H. V. Species Profile: Southern Flounder. SRAC Fact Sheet 726.
- Hinshaw, Jeffrey M. and Skipper L. Thompson. Trout Production: Handling Eggs and Fry. SRAC Fact Sheet 220 (Revision).
- Lutz, Greg. Pet Turtle Production. SRAC Fact Sheet 439.
- Masser, Michael, Tim Murphy, and James Shelton. Aquatic Weed Management: Herbicides. SRAC Fact Sheet 361 (Revision).
- Silva, Juan L., Gale R. Ammerman, and Stuart Dean. Processing Channel Catfish. SRAC Fact Sheet 183 (Revision).
- Silva, Juan L. and Stuart Dean. Processed Catfish: Product Forms, Packaging, Yields and Product Mix. SRAC Fact Sheet 184 (Revision).
- Stone, Nathan and Hugh Thomforde. Common Farm-Raised Baitfish. SRAC Fact Sheet 120 (Revision).
- Stone, Nathan, Carole Engle, David Heikes, and Donald Freeman. Bighead Carp. SRAC Fact Sheet 438.
- Treece, Granvil. *Artemia* Production for Marine Larval Fish Culture. SRAC Fact Sheet 702.
- Treece, Granvil and D. Allen Davis. Culture of Small Zooplankters for the Feeding of Larval Fish. SRAC Fact Sheet 701.
- Wallace, Richard K. Cultivating the Eastern Oyster, *Crassostrea virginica* SRAC Fact Sheet 432 (Revision).
- Watanabe, Wade O. Species Profile: Mutton Snapper. SRAC Fact Sheet 725.

### **Manuscripts in review**

Dunning, Rebecca and Harry Daniels. Hybrid Striped Bass: Economics.  
Gatlin, Delbert M., III. Improving Production Efficiency of Warmwater Aquaculture Species Through Nutrition. SRAC Final Project Report.  
Hargreaves, John and Craig Tucker. Operation and Maintenance of D.O. Meters.  
Santerre, Charles R. and George W. Lewis. Aquaculture Food Safety: Residues. SRAC Project Summary.  
Whetstone, Jack, Granvil Treece, and Craig Browdy. Marine Shrimp: Opportunities and Constraints in the Southeastern U.S.

### **Manuscripts in preparation**

Avery, Jimmy. Formulations and Application Techniques for Aquatic Herbicides.  
Brunson, Marty and Billy Griffin. Toxicity of Chemicals in Aquaculture.  
Engle, Carole and Nathan Stone. Economics of Small-scale Catfish Production.  
Hargreaves, John. Pond Mixing and Circulation.  
Hargreaves, John and Craig Tucker. Copper Use in Aquaculture.  
Masser, Michael P. Grass Carp for Aquatic Weed Control.  
Mims, Steve and Andy Lazur. Species Profile: Sturgeon.  
Whitis, Greg and Jeff Alred. Watershed Fish Production Ponds: Site Selection and Construction (Revision).



## MANAGEMENT OF ENVIRONMENTALLY-DERIVED OFF-FLAVORS IN WARMWATER FISH PONDS

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### Reporting Period

June 1, 1996 - August 31, 2001

<b>Funding Level</b>	Year 1 .....	\$250,827
	Year 2 .....	250,142
	Year 3 .....	229,266
	Year 4 .....	80,900
	Year 5 .....	56,100
	Total .....	\$867,235

<b>Participants</b>	University of Tennessee, Lead Institution .....	Thomas K. Hill
	University of Arkansas at Pine Bluff .....	Peter W. Perschbacher
	Auburn University .....	Claude E. Boyd
	Auburn University .....	R. Thomas Lovell
	Louisiana State University .....	Leslie C. Plhak
	Louisiana Tech University .....	H. Lynn Walker
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### PROJECT OBJECTIVES

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1. Evaluate the feasibility of decreasing the incidence of fish off-flavors by reducing the amount of phosphorus available to support phytoplankton growth.
  - a. Evaluate methods of reducing phosphorus input by diet modification by determining the minimum phosphorus requirement for food-sized channel catfish and quantifying the reduction in waste phosphorus generation by food-sized catfish fed "low-phosphorus" feeds relative to presently available feeds.

- b. Evaluate methods of removing phosphorus from pond waters by studying methods of enhancing rates of phosphorus removal from pond waters by pond bottom soils and determining the feasibility of precipitating phosphorus from pond waters as sparingly soluble aluminum or calcium salts.
2. Evaluate the feasibility of reducing the incidence of fish off-flavors by manipulating pond phytoplankton biomass and taxonomic composition using biological and chemical control measures.
  - a. Evaluate the effect of filter-feeding fishes on water quality and reduction or elimination of off-flavor in pond-raised channel catfish.
  - b. Develop microbial pathogens to control blue-green algal abundance.
  - c. Determine whether plant phenolics (tannins) can control growth of microorganisms that produce odorous compounds in warmwater fish ponds.
  - d. Evaluate the effect of routine, low-level treatments of ponds with copper sulfate on phytoplankton communities, off-flavor incidence, and water quality in channel catfish ponds.
3. Determine the feasibility of managing fish off-flavors by reducing rates of 2-methylisoborneol (MIB) uptake by fish and/or enhancing rates of MIB elimination from fish.
4. Develop statistical models describing the within-pond variation in the degree of off-flavor in fish populations under various conditions.
5. Develop analytical techniques for assessing flavor qualities in fish.
6. Develop publications to educate producers and processors on the ecology of environmentally-derived off-flavors, off-flavor management, and the results of this project.

## **ANTICIPATED BENEFITS**

The overall goal of this project is to reduce the incidence of unacceptable flavor quality in pond-cultured fish. If this goal is accomplished, the aquaculture industry will benefit from increased farm profits and market expansion resulting from improved consumer attitude toward aquaculture products.

Use of the revised phosphorus allowance in commercial catfish feeds should reduce the

phosphorus input to catfish ponds and thus reduce nutrients available to support algae growth. Similarly, use of alternative phosphorus supplements or phytase enzymes to increase utilization of phytate phosphorus in the feed may be beneficial in reducing phytoplankton growth and thus reduce occurrence of off-flavor. Regardless of the impact on algal communities, these studies will lead to more efficient use of phosphorus from feeds.

The use of chemical substances for reducing phosphorus levels in pond water could provide a simple procedure for channel catfish farmers and other aquaculturists to use in reducing the amount of phosphorus in waters of ponds to which large amounts of feed are applied. The benefits of the compounds (aluminum sulfate, calcium oxide, and calcium sulfate) chosen for use in this research are that they are common compounds, they are relatively inexpensive, they are environmentally safe and would not pose a food safety risk, and they would be easy to apply. If one or more of these compounds can reduce phytoplankton blooms, and especially blooms of blue-green algae, there does not appear to be any reason that farmers would not accept them readily.

The two biological control measures under investigation (use of filter-feeding animals and use of natural algal pathogens) are particularly attractive because they avoid the use of chemical control measures. In the case of control measures using filter-feeding fish or clams, economic returns from harvest of the animals stocked for algae control may be an added benefit. Other improvements in water quality may also occur. For example, a state fish hatchery is participating in the large-scale evaluation of planktivorous

fish with the hopes of addressing chronic problems with low dissolved oxygen levels.

Several chemical control measures are being investigated, including the use of copper sulfate and natural compounds such as plant phenolics. These studies should also lead to the development of one or more novel chemical treatments that can be used to control noxious phytoplankton blooms.

Additional studies focus on enhancing the elimination of MIB from channel catfish, so that off-flavor fish may be purged more effectively prior to processing. These approaches may be of significant economic value to the aquacultural industry. Also, improved methods of analysis for geosmin and MIB that are comparable or better than sensory methods with regards to sensitivity, and comparable or better than GC analysis in terms of objectivity will be developed. Immunoassay methods have these benefits and can also be formatted into rapid and simple test kits for industry. These methods will provide the industry with a better tool for quality control and fish grading as well as the research community with a better tool to study off-flavor development and control.

## **PROGRESS AND PRINCIPAL ACCOMPLISHMENTS**

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**Objective 1A.** *Evaluate methods of reducing phosphorus input by diet modification by determining the minimum phosphorus requirement for food-sized channel catfish and quantifying the reduction in waste phosphorus generation by food-sized catfish fed "low-phosphorus" feeds relative to presently available feeds.*

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The minimum dietary available phosphorus requirement for food-size channel catfish fed commercial type diets was determined in a 7-month pond study. The available phosphorus requirement based on subclinical measurements (bone breaking strength and alkaline

phosphatase activity) was found to be 0.3% of the diet, although the requirement for maximum growth was less than this. These data indicate that the available phosphorus requirement for commercial catfish feeds should be 0.3% of the diet, which is approximately 25% lower than the

present National Research Council recommendation.

The availability of different forms of phosphorus in practical feed ingredients was determined for channel catfish. A reference diet and test diets containing either menhaden fish meal, fish meal analog, meat and bone meal, soybean meal, cottonseed meal, corn, rice bran, wheat, and wheat middlings were fed to channel catfish after which fecal samples were collected and analyzed to determine the availability of different forms of phosphorus from the ingredients. The various feed ingredients varied considerably in terms of their phosphorus composition and availability to the fish. Wheat, sorghum, and cottonseed meal had the greatest phosphorus availability of the plant feedstuffs while fish meal analog had the greatest availability of the animal feedstuffs. Also in this study, the uptake and mineralization of different forms of phosphorus and nitrogen in feces from channel catfish fed the various ingredients were determined.

Digestibility trials were also conducted with channel catfish to evaluate several different feedstuffs which have been genetically modified to contain low concentrations of phytic acid. The phytic acid which is typically found in reasonably high concentrations in plant feedstuffs is indigestible and thus excreted by fish. The low-phytate varieties of barley, corn, and soybean meal which have been evaluated have elevated phosphorus availability to channel catfish. Thus, as these feedstuffs become more readily available, they may provide a dietary means of reducing phosphorus excretion by this species.

Another study was conducted to evaluate diet formulations and feeding strategies to marginally meet the phosphorus requirement of channel catfish while minimizing dietary phosphorus input. Fingerling channel catfish were fed one

## *Results at a glance...*

- ☆ *Low-phytate varieties of barley, corn and soybean meal elevate phosphorus availability to channel catfish so they may provide a dietary means of reducing phosphorus excretion as these feed stuffs become more readily available.*

of three practical diets with either no phosphorus supplementation (approximately 0.2% available phosphorus), minimal phosphorus supplementation from dicalcium phosphate (0.3% available phosphorus), or standard phosphorus supplementation from dicalcium phosphate (0.5% available phosphorus) for 8 weeks in aquaria. Two additional treatments included feeding the diet containing 0.2% available phosphorus with intermittent feeding (two days every other week or every fourth week) of the diet containing 0.5% available phosphorus. Samples of pectoral spines and whole-body tissues indicated adequate phosphorus status of channel catfish could be maintained with minimal phosphorus input by feeding the unsupplemented diet in conjunction with the phosphorus-supplemented diet for two days every fourth week.

A pond study was conducted to quantify the reduction in waste phosphorus generation by food-size channel catfish fed experimental diets formulated to contain 28% protein and 0.4% available phosphorus from either dicalcium phosphate (water-soluble) or defluorinated phosphate (water-insoluble) phosphate. No statistical differences were observed in weight gain, feed conversion, survival, bone ash, and bone phosphorus between fish fed the two diets. There were no significant differences in total phosphorus, soluble phosphorus, and chlorophyll *a*

concentrations in pond water between the two dietary treatments.

A pond study was conducted in Mississippi to quantify the reduction in waste phosphorus generation by food-sized channel catfish fed low phosphorus diets. A basal diet was formulated to contain 32% protein without supplemental phosphorus (0.2% available phosphorus). Supplemental phosphorus was added to the basal diet to provide available phosphorus of 0.3 and

0.4%, respectively, using dicalcium phosphate. There were no significant differences in total phosphorus, soluble phosphorus, and chlorophyll *a* concentrations in pond water between the two dietary treatments. However, in a similar study conducted in Alabama where phosphorus in catfish feed was increased from 0.6% (0.2% bioavailable) to 1.0% (0.6% bioavailable), there were significant increases in total phosphorus and phytoplankton production.

**Objective 1B.** *Evaluate methods of removing phosphorus from pond waters by studying methods of enhancing rates of phosphorus removal from pond waters by pond bottom soils and determining the feasibility of precipitating phosphorus from pond waters as sparingly soluble aluminum or calcium salts.*

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In 1996, laboratory and pond studies were conducted to determine the feasibility of precipitating phosphorus from waters as sparingly soluble aluminum or calcium salts through the application of aluminum sulfate (alum), calcium oxide (lime), or calcium sulfate (gypsum). The gypsum application had the greatest effect, and the treatment was repeated in 1997 at a higher application rate. At the higher rate (increasing total hardness to 200 ppm), gypsum significantly reduced total phosphorus and chlorophyll *a* levels. More frequent applications of alum to pond water (1998) showed distinct short-term effects but little long-term change in the pond water quality. Repeated applications of 7 pounds/acre of agricultural limestone at 2-day intervals also reduced soluble phosphorus concentrations but had little effect on phytoplankton. A 120-day pond trial was completed which evaluated the effects of periodic additions of alum on the availability of phosphorus in the pond environment. Addition of alum at 50 ppm every other week did not affect total production or feed conversion of channel catfish in 0.1-acre ponds. In addition, waterborne phosphorus concentration, primary

## *Results at a glance...*

- ☆ *Of several treatments tried, the treatment that resulted in less phosphorus availability and phytoplankton abundance combined pond bottom drying and tilling with gypsum application to the pond.*

productivity, algal species composition and sediment oxygen demand generally were not affected by alum addition; however, there was an obvious reduction in the amount of filamentous algae and other vegetation in ponds treated with alum.

In 1997 and 1998, drying and tilling empty pond bottoms before filling the ponds resulted in lower phosphorus levels in the water during the production season. Incorporation of alum, agricultural limestone, or sodium nitrate in the tilled soil did not reduce phosphorus levels or improve water quality above drying and tilling alone.



In 1999, the treatments were as follows: control, dry tilling of pond bottoms, and dry tilling of pond bottoms followed by periodic gypsum applications to maintain 200 ppm total hardness. The 1999 study showed that the dry-till with gypsum treatment was superior to dry-till alone in lessening phosphorus availability and phytoplankton abundance.

The experiment to evaluate the benefits of water circulation on pond water quality was

completed in October 2000. This study revealed no added benefit over dry-till with gypsum treatment of providing water movement with a 0.5-hp mechanical circulator. This experiment completed the trials to be conducted in this project.

During 2001, soil analyses and phytoplankton evaluations were completed on all samples from previous studies, and the findings of the research are being described in several manuscripts.

**Objective 2A.** *Evaluate the effect of filter-feeding fishes on water quality and reduction or elimination of off-flavor in pond-raised channel catfish.*

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Six species of filter-feeding macroorganisms were identified as candidates for biological control and tested in mesocosms in 1997 for their ability to filter off-flavor algae. Nile tilapia, blue tilapia, and silver carp significantly reduced numbers of *Oscillatoria perornata* and *Anabaena circinalis* (major producers of MIB and geosmin, respectively). Threadfin shad significantly reduced numbers of *A. circinalis*. Two species of local, unionid clams did not produce measurable effects. Based on these findings, a silver carp system was evaluated in 0.25-acre ponds in 1998. Abundance of *O. perornata* was reduced, but the alga was not eliminated. In addition, difficulty was encountered in adapting a silver carp system to channel catfish production ponds.

Testing of seven filter-feeding organisms and at several densities found a system based on Nile tilapia stocked at 2000 fish/acre in cages provided control of spiked *O. perornata* in 0.1-acre experimental ponds in one month. In two 2.5-acre ponds at a state fish hatchery, this system again eliminated the alga or reduced its abundance to below problem in one month. Genetically male tilapia grew from 3 inch stockers to 0.25 pounds in the 3 month evaluation, which is 10% faster growth than mixed-sex Nile tilapia. Additional water quality improvements (other than improving fish flavor) were also observed. For example, the incidence of low oxygen levels was reduced by 60-70% in ponds with the filter fish system compared to a control pond without the system.

**Objective 2B.** *Develop microbial pathogens to control blue-green algal abundance.*

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Fungal and bacterial pathogens of *Anabaena* and *Oscillatoria* were isolated from commercial catfish ponds. In laboratory studies these agents lysed cells of *Anabaena* and *Oscillatoria*, and selectively removed these species from mixed cultures

containing beneficial algae and blue-green algae.

A fungal pathogen was selected for evaluation in replicated tests that were conducted in 210-gallon tanks to study control of *Oscillatoria*

*perornata*. The tanks were filled with water from a commercial catfish pond, stocked with catfish fingerlings, and treated with preparations of a fungus. The fungus controlled *O. perornata*, but high oxygen demands were observed. Preparations of the fungus are being developed that will minimize the oxygen demand.

A bacterium that is pathogenic to species of *Anabaena* and *Oscillatoria* was isolated from pond water. When comparisons were made using a number of databases, no definitive match for the DNA sequence of the 16S rRNA gene for the bacterium could be established at the genus or species level. Therefore, the bacterium could represent a genus that has not been described. When the bacterium was evaluated as a biological control agent in laboratory studies, the average reductions in chlorophyll *a* were 94 to 98% for *Oscillatoria* spp. and 13 to 98% for *Anabaena* spp. No significant reductions in chlorophyll *a* were noted for *Chlorella vulgaris*, *Scenedesmus subspicatus*, *Selenastrum capricornutum*, *Microcystis aeruginosa*, or *Plectonema boryanum*. Shake flask cultures of the bacterium produced approximately 3 billion plaque-forming units/milliliter of culture broth. Therefore, 1 liter of culture broth, when uniformly distributed in one acre-foot of pond water, would result in an initial concentration of approximately 2,400 plaque-forming units/milliliter of pond water. In replicated tests conducted in 210-gallon tanks containing water from commercial catfish ponds, the bacterium selectively removed species of *Anabaena* and *Oscillatoria*. When the bacterium was inoculated to pond water, *Oscillatoria perornata* was reduced from an initial density of 2,700 filaments/milliliter to 0 filaments/

milliliter after 48 hours. The blue-green alga *Microcystis* became dominant as the species composition of the phytoplankton changed in the treated pond water. Results of laboratory and tank tests indicated that the bacterium did not adversely affect channel catfish fingerlings.

### *Results at a glance...*

☆ A bacterium isolated from pond water selectively attacks odor-producing blue-green algae while having no effect on beneficial algae or catfish. The bacterium shows promise as a biological control agent for the algae that cause off-flavors.

The bacterium was tested in ponds. Analyses of pond water following inoculation indicated that the bacterium was present in the water for up to 5 days after inoculation. While the results of preliminary pond experiments were encouraging, future tests in ponds need to be conducted under more uniform environmental conditions to facilitate interpretation of results.

Research is underway to optimize production of the bacterium and to produce preparations that have a shelf life of over 1 year. Three patents or patent applications related to this technology have been assigned to the Louisiana Tech Research Foundation. Negotiations are underway with two corporations that have expressed interest in commercial development of the bacterium.

**Objective 2C.** *Determine whether plant phenolics (tannins) can control growth of microorganisms that produce odorous compounds in warmwater fish ponds.*

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The bacterium *Streptomyces tendae* is known to synthesize geosmin, an earthy off-flavor con-

taminant of aquatic products. Experiments were conducted to determine the antimicrobial effects

of tannic acid and related compounds such as propyl gallate, methyl gallate, and gallic acid on the growth of *Streptomyces tendae*. Well-diffusion assays and biomass determinations were performed. The biomass determination method is more sensitive than the well-diffusion assay. The results of these experiments indicate that tannic acid is inhibitory to *S. tendae* at levels as low as 0.3 ppt. Propyl gallate is inhibitory at higher concentrations, but methyl gallate and gallic acid have no inhibitory effects at concentrations up to 1 ppt. Olfactory evidence suggests that tannic acid may inhibit geosmin synthesis.

It was also demonstrated that tannic acid and related compounds are inhibitory to the growth and pigment synthesis of off-flavor producing *Nostoc* sp. strain MAC. The minimum inhibitory concentrations of tannic acid, propyl gallate, and gallic acid in augmented pond water were 320, 240, and 500 micrograms/

disk, respectively. Tannic acid, propyl gallate, and gallic acid also exhibited inhibitory activity to *Cytophaga columnaris*, a ubiquitous, gliding fish pathogen, at 150, 300, and 300 ppm. Methyl gallate was effective at 500 ppm. The protein precipitation and polysaccharide binding capacities, lipophilicity and other physico-chemical properties of these compounds were measured in order to understand possible mechanisms for their antibacterial action. Tannic acid, a polymeric compound with multiple hydroxyl groups, had at least a nine times greater capacity for binding protein and glycogen than the other test compounds. These results suggest that the hydroxyl group availability of tannic acid is essential for antibacterial activity. Therefore, it is likely that these compounds may have some beneficial effect in controlling the microbial population in ponds and may have impact on the phytoplankton biomass.

**Objective 2D.** *Evaluate the effect of routine, low-level treatments of ponds with copper sulfate on phytoplankton communities, off-flavor incidence, and water quality in channel catfish ponds.*

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Eighteen, 0.4-ha earthen ponds in northwest Mississippi were used in a 3-year study to evaluate the effect of weekly copper sulfate applications on the incidence and economic impact of environment-derived off-flavors in channel catfish. Each spring when water temperatures increased above 70°F, nine of the ponds were treated weekly with 5 pounds/acre copper sulfate by placing the required amount of copper sulfate crystals in a burlap bag which was then placed in the current produced by a paddlewheel aerator. Copper treatments were discontinued each fall when water temperatures fell below 70°F. Overall prevalence of off-flavor was reduced by 80% for ponds treated with copper sulfate relative to control ponds, and episodes of off-flavor were of shorter duration in treated

ponds. Off-flavors never delayed fish harvest from treated ponds, whereas off-flavors delayed fish harvest on ten occasions in control ponds. Average annual fish harvest was 5,250 pounds/acre from ponds treated with copper sulfate and 4,760 pounds/acre from control ponds. The 9%

### *Results at a glance...*

☆ *Weekly, low-level treatments of catfish ponds with copper sulfate reduced the incidence of off-flavor by 80% and increased net revenues by over 40% compared to untreated ponds.*

reduction in fish harvest from control ponds was due to infectious disease outbreaks in one or two ponds each year where harvest was delayed due to off-flavor. Enterprise budgets showed that average net returns above variable costs were \$770/acre for control ponds and \$1,100/acre for ponds treated with copper sulfate. Variation in net returns was twice as great for control ponds as for treated ponds, indicating increased stability in production and economic returns when

off-flavors were managed using copper sulfate. High variation in annual economic performance on control ponds resulted from one or more ponds having high net returns while one or more ponds had extremely poor returns due to protracted episodes of off-flavor. Stability in production and costs is a means of reducing risk and is a positive factor farmers can use to better plan their cash flow needs throughout the production season and in the longer term.

**Objective 3.** *Determine the feasibility of managing fish off-flavors by reducing rates of 2-methylisoborneol (MIB) uptake by fish and/or enhancing rates of MIB elimination from fish.*

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Three compounds were initially identified as potential enhancers of MIB elimination based on their ability to increase the activity of cytochrome P450, the enzyme system thought to be involved in metabolizing the off-flavor compound, 2-methylisoborneol (MIB). One of the three compounds (3-methylcholanthrene) significantly increased the residence time of MIB in channel catfish. After initial success with clofibric acid, further investigation only showed a trend toward enhanced elimination. The last compound, ethanol, provided the best results observed in enhancing MIB elimination, but only following MIB exposure. Pretreatment with each chemical did not affect MIB uptake or elimination. Treatment with ethanol following MIB uptake nearly doubled the rate of MIB elimination. Although a direct correlation was observed between temperature and MIB elimination, temperature failed to have any synergistic effect on the enhanced elimination by any of the three compounds.

Analysis of MIB elimination in the Uvalde strain of channel catfish indicated tremendous variation in the response of fish and their ability to eliminate MIB. Variation between

individuals is nearly 35%. The factors controlling the variation in response are unclear. No relationship has been observed between individual isoforms of cytochrome P450 and MIB elimination in this particular strain. Examination of MIB metabolism in Uvalde strain channel catfish with induced enzymes indicated that MIB is not metabolized.

To determine whether extrahepatic (tissues other than the liver) biotransformation of MIB may be occurring, the metabolism and disposition of radio-labeled MIB was examined in Uvalde channel catfish as well as another strain of channel catfish (USDA 103) and the channel catfish x blue catfish hybrid. No metabolites were observed in plasma from animals treated with an intra-arterial dose of radio-labeled MIB. Elimination of MIB from the two strains and hybrid was accurately predicted using a three compartment pharmacokinetic model. There was no significant difference in terminal half-lives between strains, but significant differences in other predicted pharmacokinetic parameters, such as total clearance, were observed with the hybrid strain, which had a 10-fold greater clearance.

**Objective 4.** *Develop statistical models describing the within-pond variation in the degree of off-flavor in fish populations under various conditions.*

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A study was conducted in summer and fall in ten commercial catfish farms to determine the proportion of off-flavor fish in ponds. Flavor was assessed by sensory evaluation. A larger occurrence of off-flavor was found in summer than in fall. Proportion of off-flavor fish varied from 0% to 54% depending on the pond and the acceptance criterion used. Between-pond

variance of the proportion of unacceptable fish was greater than within-pond variance. Therefore, Bayesian sampling should be used instead of conventional sampling based on the binomial function. Different sampling plans are proposed depending on levels chosen for producer and consumer risks. It was recommended to consider a sample size of at least 30 fish.

**Objective 5.** *Develop analytical techniques for assessing flavor qualities in fish.*

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Monoclonal antibodies have been produced that bind to 2-methylisoborneol (MIB). This led to the development of immunochemical methods (ELISA) to detect MIB down to levels of 0.01 ppb, low enough to be comparable to the human sensory threshold for MIB. Zeolite was tested as a material for MIB absorption and concentration. Using ELISA, zeolite was shown to absorb small molecules (glycoalkaloids) but not antibodies. Zeolite, however, was shown to be less efficient for MIB absorption than activated carbons, when compared using a purge and trap apparatus.

An eight-member sensory panel was trained

using the Sensory Spectrum Method. A preliminary study was conducted to evaluate the effectiveness of various processing procedures in reducing off-flavor in catfish. Fillets of each flavor rating were either dipped or vacuum tumbled in water, dairy whey or 3% lemon juice. The panel gave significantly higher scores for the geosmin note and lower scores for the chicken-like note for off-flavor level 5 compared to level 1, regardless of the treatment. Lemon juice significantly increased the geosmin note, whereas dairy whey reduced it. Vacuum tumbling with lemon juice reduced the green/corn note (considered a desirable note) compared to dipping in lemon juice.

**Objective 6.** *Develop publications to educate producers and processors on the ecology of environmentally-derived off-flavors, off-flavor management, and the results of this project.*

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See list of publications on pages 25-28.

## **WORK PLANNED**

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Work on all objectives is proceeding on schedule and no changes in the project have occurred this year.

## IMPACTS

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Information generated in two of the project objectives is already being used in the aquaculture industry. First, work to investigate phosphorus availability of various feedstuffs has been used by feed manufacturers to refine commercial diet formulations, with a cost savings to the farmer. Second, the efficacy of routine, low-level copper sulfate treatments for preventing algae-related off-flavors has been verified in large-scale field studies and the practice has been adopted by many commercial producers in Mississippi and Arkansas.

Although other results of this project are too preliminary to have had an impact on the aquaculture industry, several of the treatments and practices being investigated show promise. For example, phosphorus levels in ponds can be reduced by precipitating phosphorus as aluminum or calcium salts, or by treating the pond bottom to reduce phosphorus flux from soils to

water. These practices could be an important management procedure for improving quality of pond water and effluents and in combating off-flavor.

Another example of a potentially effective practice is the use of filter-feeding fishes, which has been shown to be effective in controlling odor-producing algae in small-scale systems and in pond trials. Use of fast-growing, genetically male tilapia may help avoid problems of uncontrolled reproduction encountered when mixed-sex tilapia are used in polyculture with catfish.

Perhaps the most intriguing result is the success achieved using bacterial pathogens of odor-producing blue-green algae. If these results can be transferred to pond-scale ecosystems, the work may lead to a novel, safe, and effective method of controlling flavor problems in fish.

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## VERIFICATION OF RECOMMENDED MANAGEMENT PRACTICES FOR MAJOR AQUATIC SPECIES

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### Reporting Period

January 1, 1997 - August 31, 2001

<b>Funding Level</b>	Year 1 .....	\$31,410
	Year 2 .....	66,114
	Year 3 .....	66,925
	Total .....	\$164,449

<b>Participants</b>	University of Arkansas at Pine Bluff (Lead Institution) ....	Carole Engle, David Heikes, Steve Killian, Pierre-Justin Kouka
	Auburn University .....	Jerry Crews, Greg Whitis, David Cline, Claude Reeves
	Clemson University .....	William "Rockie" English, Tom Schwedler, Johnny Jordan, Jack Whetstone
	Louisiana State University .....	Greg Lutz, Jimmy Avery (now at Mississippi State University)
	North Carolina State University .....	Harry Daniels, Steven Gabel, Michael Frinsko, Rebecca Dunning

<b>Administrative Advisor</b>	Dr. Jack Bagent, Director (Retired) Louisiana Cooperative Extension Service Louisiana State University Baton Rouge, Louisiana
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## PROJECT OBJECTIVES

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The overall goal of this project is to initiate verification programs in participating states. The emphasis is on developing the interdisciplinary process and internal committees within each state. While actual field results of verification trials of different management protocols will be valuable, this project is intended as a stimulus to develop and utilize verification trials as a new extension tool. The specific objectives of this project are:

1. To develop and implement recommended management practices for catfish and crawfish production systems in participating states;
2. To publish guidelines for infrastructure development, program implementation, and assessing results/benefits of aquaculture management verification. This publication will be a joint effort of participants; and

3. To publish recommended management plans and results of Objective 1.

## ANTICIPATED BENEFITS

The principal benefit of verification is to determine if the total set of research-based extension recommendations produces yields, feed conversions, and costs consistent with results from research trials. Researchers and extension personnel learn whether their recommendations are valid in commercial settings and whether or not

recommendations and research programs need to be adjusted based on what has been learned. Adoption of verification practices is expected to increase industry yields. The development of the verification management plan encourages open dialogue among researchers, producers, and extension specialists.

## PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

*Objective 1. To develop and implement verification programs of recommended management practices for catfish and crawfish production systems in participating states.*

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**Alabama.** The Extension Fisheries team established recommendations for the production systems (levee ponds, watershed ponds, and cages) in the verification project. There are four cooperators and four levee ponds in West Alabama (two with channel/blue hybrid catfish and two with channel catfish), five cages (two in East Central and three in Southeast Alabama), and two watershed ponds enrolled in the verification program. Production and water quality parameters have been monitored and the Fishy '98 computer program is being used to track feeding and other data on most of the production units since 1998. Four levee-style ponds have been enrolled in the verification program for 2-3 years. Stocking rates have varied from 4,071 to 5,469 fish/acre per year. Harvested weights range from 5,241 to 10,749 pounds/acre per year, including fish scrapped from ponds. Three of the four ponds are pending final harvest and scrapping. Overall, survival has ranged from 57 to 91%. Gross feed conversion ratios (pounds of feed/pounds of fish harvested) ranged from 1.87 to 2.8. Both of the watershed ponds

enrolled in the program have been harvested completely. Cages in East Central Alabama were harvested. The three cages in Southeast Alabama were successfully harvested in the first year of the project. However, in the second year, under-sized fish were delivered. This, in combination with an outbreak of columnaris disease,

### *Results at a glance...*

☆ *Analysis of the production and economic data indicate that the recommended management practices result in greater profit to the producers.*

resulted in significant mortality and probable escape of fish from the cages. The second-year cage trial was abandoned. The final, complete description of results of the Alabama verification program will be published in the upcoming year.

**Arkansas.** An inter-disciplinary verification committee, consisting of extension specialists, researchers, economists, and county extension agents developed specific management protocols for the verification of recommended foodfish and fingerling management practices. Record-keeping forms were developed and printed in field-booklet form on waterproof paper. A spreadsheet computer program and sampling methodology were developed to be used with the Fishy 3.2 record-keeping program. A literature search was conducted to ensure that the management protocols reflect a progressive, practical, and profitable management scenario.

Arkansas conducted verification trials on six foodfish (four in northern Poinsett and St. Francis counties and two in southern Chicot County) and two fingerling ponds across the Delta production area. Production inputs and yield data were collected on a weekly basis, summarized weekly and posted on the Arkansas CYVT web site ([www.uaex.edu/aquaculture/arcyvp.htm](http://www.uaex.edu/aquaculture/arcyvp.htm)). All ponds have been harvested completely and the complete, final report and summary of all data is in preparation.

**Louisiana.** The verification committee was formed, a literature review completed, and fisheries/aquaculture agents, specialists, and administration were trained in verification procedures. Management protocols have been developed for three crawfish production scenarios: 1) rice-crawfish rotation; 2) permanent crawfish pond; and 3) growing crawfish behind two successive rice crops. Five cooperators participated in the Year 2 Crawfish Yield Verification Project. Cooperators included three producers from Vermillion Parish, one producer from St. Martin Parish, and one producer from Acadia Parish. There were nine ponds enrolled in the project with six ponds evaluating the rice-crawfish rotation, one pond evaluating the permanent pond scenario, and two ponds of crawfish behind double-crop rice. The

production season began in October and terminated in June. Agents collected pre-production samples of water source, forage, and soil. Forage depletion was monitored monthly. Farmers

## *Results at a glance...*

☆ *Good feeding practices resulted in feed conversion values below 1.6:1.*

were given recommendations on trap density, bait usage, and harvest regimes. The final, comprehensive report on the Louisiana verification program will be published this coming year.

**North Carolina.** A five-member committee consisting of industry, university, and extension representatives established recommended catfish management protocols. These protocols have been implemented in the management of three channel catfish production ponds on three separate farms since fall 1997. Data collection continued through 2000 on different production variables (feeding, aeration, labor, etc.) on a weekly basis. Final harvests have been completed. The harvests were delayed due to extensive flooding caused by several hurricanes during the fall of 1999. A complete final report has been prepared summarizing the data collected for the cooperators' ponds over the entire three years of the project.

**South Carolina.** The verification committee was formed, the management protocol to be implemented developed, and a cooperator identified. Background information on financial and production performance was evaluated. A change in farm managers in the middle of the year caused some delays in the implementation of the rotational plan. The proposed phased rotation management plan was updated to work with the farm's current inventory levels, and

modified to work within the farm's restrictions of capital and equipment.

In August of 1998, a 12-acre stocker pond was stocked with approximately 240,000 fingerlings weighing 60 pounds per 1000 fish. They were fed until mid-October when approximately 60,000 fingerlings weighing about 200 pounds/1000 were moved to a vacated pond. The fish were fed until August 15, 1999, and a portion of the fish were harvested (12,000 pounds) and

sold to the processing plant. The fish were seined with a 1 3/8-inch sock to estimate true average size of the fish. The average size of the fish harvested was 1.38 pounds. There were personnel changes at the farm so that no accurate information was available on actual feed fed and no conversion rates could be calculated. The stockers were moved at approximately the correct time and size and were within 5% of the targeted market size at harvest that was projected by the model.

**Objective 2:** *To publish guidelines for infrastructure development, program implementation and assessing results/benefits of aquaculture management verification. This publication will be a joint effort of participants.*

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A joint project publication was written that presents guidelines for developing aquaculture verification programs. This bulletin presents a brief history of verification programs in extension. Procedures for infrastructure development are discussed that include information on forming the verification committee, efficient ways to develop summaries of the relevant research base, efficient ways to develop recommended management plans, data collection procedures, and data synthesis procedures. The bulletin then describes key components of program implementation. Frequency of farm visits, the role of county extension agents

and specialists, role of cooperators, production cycles, and required resource commitments are presented based on the experiences of specialists that conducted verification in this program.

The bulletin also discusses the types of results, benefits, and impacts that verification programs can have. Potential pitfalls and problems review the types of issues that can arise among cooperators, agents, specialists, and the production environment. The bulletin concludes with discussions of information dissemination and potential funding sources.

**Objective 3:** *To publish recommended management plans and results of Objective 1.*

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Management protocols have been developed for use in all participating states. The management protocols and final, comprehensive reports of

the results are the responsibility of each participating state. North Carolina has completed theirs.

## **WORK PLANNED**

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Commercial fish production requires complex management decisions on stocking, harvesting, and marketing. There are numerous factors

such as market requirements, drought, disease, and hurricanes, that are outside the control of the manager that affect the operator's ability to

manage the fish farm according to a pre-set schedule. These same factors and conditions have prevented final harvest of several of the ponds enrolled in the verification program. Much of the work planned for the coming year includes final harvest followed by final data summaries, and synthesis. When all data have been synthesized, complete reports with complete data will be published in each state.

In Arkansas, a comprehensive report is under preparation that summarizes all data collected over the project period for all ponds and includes an economic analysis of the results. Alabama still has ponds to harvest. Complete data analysis along with an economic analysis will be completed upon harvest of all ponds. Louisiana crawfish crops lag about 6 months behind fish crops. Upon final harvest, a report to summarize all findings will be completed and published.

## IMPACTS

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**Alabama.** There has been an increased awareness of actual inputs required to produce a crop of catfish, more attention paid to tracking all the real costs of catfish production, and increased attention to and analyses of water quality in pond production. There is now an awareness that close monitoring of water quality and equipment condition (aerators, tractors, feeders, etc.) can prevent problems and decrease fish mortality. There is increased attention paid to record-keeping and tracking costs of production. One producer who normally stocked ponds in the 12 to 15,000 fish/acre range has reduced his stocking rates because of the production data in his verification pond. Also, the verification program showed that the routine use of a 1.75-inch mesh seine results in an average size of fish sold of 2 pounds, not the 1.25 pounds previously assumed. Overall, the project demonstrated without a doubt that current extension guidelines will result in profitable production.

**Arkansas.** Of particular interest is the impact that this program has had on producers in the northern half of Arkansas. Prior to this program, county agents had very little exposure to catfish producers and many producers have turned to non-Extension sources of information for years. Since the initiation of this program, word has spread through fish farmer networks

### *Results at a glance...*

☆ *These results demonstrate the importance of careful feeding and the positive effect feed management has on profitability.*

that Extension has important information and the county agents have seen a tremendous increase in the number of aquaculture-related calls in their counties. The agent in Poinsett County has asked specifically that we continue catfish verification in his county indefinitely. Also, the number of producers from the northern counties submitting disease cases to Extension Fish Diagnostic Laboratories in Arkansas has increased, indicating an increased level of trust with Extension services.

**Louisiana.** The major impact of the project to this point has been the interest of the field agents in participating in a proactive program. Field agents have increased awareness of the importance of population structure at the end of the previous season, summer management of natural forage or rice, precipitation patterns while crawfish are aestivating in burrows,

pesticide use, and fall flooding protocols. Two of the cooperators reported that they realized the difference that higher trap densities had on overall catch rate. Some cooperators have shared previous years' records with agents in an attempt to further refine their production

practices.

**North Carolina.** The cooperating farmers have expressed their satisfaction with the results of this project and, where practical, have implemented these same practices on the rest of their ponds.

## PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

### Papers presented

- Daniels, H., S. Gabel, M. Frinsko, and R. Dunning. In review. Channel Catfish Production Manual: Recommended Management Practices for North Carolina Producers.
- Heikes, D. L. 1998. Catfish Yield Verification Update. Poster presentation. Catfish Farmers of Arkansas Annual Convention, Little Rock, Arkansas. 22-23 January.
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- Heikes, D. L. 1998. Catfish Fingerling Verification. Presentation. Catfish Farmers of Arkansas Annual Convention, Little Rock, Arkansas. 22-23 January.



## CONTROL OF BLUE-GREEN ALGAE IN AQUACULTURE PONDS

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### Reporting Period

January 1, 1999 - August 31, 2001

<b>Funding Level</b>	Year 1 .....	\$307,574
	Year 2 .....	280,639
	Year 3 .....	253,326
	Total .....	\$841,539
<b>Participants</b>	University of Tennessee (Lead Institution) .....	J. Larry Wilson
	University of Arkansas at Pine Bluff .....	Nathan M. Stone
	Auburn University .....	David Bayne, Thomas J. Popma, Claude Boyd
	Clemson University .....	David A. Brune, John A. Collier, T.E. Schwedler
	University of Georgia .....	Gary J. Burtle, George W. Lewis, Eloise L. Styer
	Louisiana State University .....	Robert R. Romaine
	Mississippi State University .....	John A. Hargreaves, Susan K. Kingsbury, Edwin H. Robinson
	University of Mississippi.....	Dale G. Nagle
	North Carolina State University.....	Harry V. Daniels
	North Carolina State University.....	Ronald G. Hodson
	USDA, ARS, NPURU.....	Kevin K. Shrader
USDA, ARS, SRRC .....	Paul V. Zimba, Casey C. Grimm	
<b>Administrative Advisor</b>	Dr. Greg Weidemann, Associate Director Arkansas Agricultural Experiment Station University of Arkansas Fayetteville, Arkansas	

## PROJECT OBJECTIVES

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1. Develop chemical control methodologies to prevent the establishment of noxious blue-green algal communities.
  - a. Evaluate novel selective blue-green algicides identified through laboratory screening.
  - b. Isolate, identify, and test allelopathic chemicals produced by competing blue-green algae and other micro-organisms found in local aquatic communities.



2. Evaluate nutrient manipulation to promote desirable phytoplankton community structure.
  - a. Increase nitrogen-to-phosphorus ratios in the water.
  - b. Reduce the availability of phosphorus from pond bottom muds.
  - c. Enhance the availability of inorganic carbon.
  - d. Manipulate trace metal availability.
  - e. Increase potassium levels in the water.
  - f. Increase salinity levels in the water.
3. Evaluate water circulation as a means of altering the environment to promote desirable phytoplankton community structure.
4. Evaluate the use of plankton-feeding fish to alter the environment to promote desirable phytoplankton community structure.
5. Evaluate the development of phytoplankton communities in the Partitioned Aquaculture System.

## **ANTICIPATED BENEFITS**

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The overall goal of this project is to identify methods of controlling or eliminating blue-green algae from aquaculture ponds. The ability to control algal communities in ponds could benefit farmers in several ways.

Excessive abundance of blue-green algae, especially when combined with their habit of growing in surface scums, can cause low dissolved oxygen concentrations and other water quality aberrations that affect fish growth and health. Therefore, the ability to control the composition of blooms could result in better fish growth and lower costs for aeration and other water quality management procedures.

The largest and fastest growing segment of aquaculture in the United States is farm-raised channel catfish. Catfish that are off-flavor are

unmarketable, and farmers are forced to hold those fish in inventory until composition of the pond microbial community changes and flavor improves. Holding market-sized fish in inventory imposes an economic burden on farmers, and off-flavor is estimated to cost the industry well over \$20 million a year.

Baitfish mortalities associated with blue-green algae are common in the early summer. Historical use of high rates of granular fertilizers may be a factor in these excessive algae blooms, especially in baitfish ponds that have been in production for years and have accumulated sediments. Documenting potential improvements in water quality as a result of pond renovation and sediment removal will provide farmers with information to make informed decisions when weighing benefits of pond renovation against

costs. Alternatively, sodium nitrate has been proposed as a pond bottom treatment to improve water quality and has been shown to effectively control phosphorus release from soil in laboratory studies. Field studies in commercial ponds will provide producers with information to better evaluate the potential merits of this compound.

Most of the treatments and management prac-

tices considered in this project have been promoted for controlling blue-green algae, but their effectiveness has not been documented. It is anticipated that this research will reveal which, if any, of these treatments are beneficial. Any practice demonstrated to be effective in controlling blue-green algae has considerable potential for improving aquaculture management and enhancing profits.

## PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

**Objective 1.** *Develop chemical control methodologies to prevent the establishment of noxious blue-green algal communities.*

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**University of Mississippi.** More than 4,000 plant and algae extracts have been prepared from several thousand collections of plants, cyanobacteria and algae. These collections now include more than 300 collections of tropical cyanobacteria and marine algae, and 450 collections of aquatic and wetland plants, cyanobacteria (collections and cultures), algae, and thousands of collections of higher plants. These extracts were evaluated for selective blue-green algicidal activity in a rapid bioassay using 96-well microtiter plates.

Initial extract evaluations were conducted using *Oscillatoria agardii* as the cyanobacterial test organism, and *Selenastrum capricornutum* as a chlorophyte control for nonspecific algicidal activity. These organisms were grown in continuous flow culture and provide uniform algal material for biological evaluation. Lipid extracts of 33 tropical marine cyanobacteria and algae were evaluated in these assays at an initial concentration 100 ppm. Two related species of marine green algae and one species of marine brown algae were found to contain substances that were selectively algicidal against *O. agardii*.

While these results are promising, the cyanobacterium *O. agardii* is not known to produce odorous compounds that cause off-flavor problems in aquaculture. We have since established cultures of the *O. perornata*, a filamentous blue-green alga that produces 2-methylisoborneol (MIB), the major tainting substance in catfish grown in northwest Mississippi. These cultures are suitable for high-throughput extract evaluation.

In the first phase of the project, 356 extracts from collections of aquatic and marine cyanobacteria, plants and algae have subsequently been evaluated in replicate bioassays using *O. perornata*. Forty-three extracts (12%) were found to be strongly active and cyanobacterial-selective at the concentration of 100 ppm, used for initial screening. Dose response data was obtained for these extracts at half-log concentrations. Twelve extracts were found to be effective at 30 ppm or lower. The two most potent extracts were confirmed to be two of the related marine green algae species first identified in the initial bioassays that used *O. agardii* rather than *O. perornata*.

Bioassay-guided fractionation of several extracts has resulted in the purification of unusual cyanobacterial-selective natural products that are effective against both *Oscillatoria* strains at concentrations in the parts-per-billion range. The chemical structures of several of these metabolites have been solved and are undergoing additional toxicological and antimicrobial evaluation.

The second phase of the program focused on the evaluation of plants found in tropical rainforests and temperate regions throughout the world for cyanobacterial-selective algicidal activity. Initial evaluations of plant extracts at 100 ppm picked up some activity associated with less selective antimicrobial substances such as tannins. In order to reduce the incidence of “false-positive” hits and to select for only those plants that contain potent and potentially more selective compounds, the primary phase of the high-throughput screening program evaluated plant extracts at 20 ppm. Extract plates were evaluated in duplicate. Plant extracts that showed cyanobacterial-selective activity at 20 ppm were reconfirmed by secondary evaluations at 20, 10 and 2.0 ppm. A bio-genetically diverse repository of plant extracts from over 170 plant families collected was examined. These plants were obtained from Peru, New Guinea, and the United States. This repository contained chemically distinct crude lipophilic extracts of separate plant parts (roots, leaves and stems, flowers, etc.) of each species collected. Over 2,300 crude extracts of more than 1,050 species of higher plant extracts were evaluated in this high-throughput screening system.

Over 70 plant species showed some level of selective activity against *O. perornata*. Since the major goal of this project was to identify cyanobacteria-specific agents, a process of “deselection” was undertaken to exclude compounds with broad activity against other microorganisms

(blue-green algae are gram-negative bacteria). Thus, the active extracts that showed broad antimicrobial activity against a panel of biomedically important microorganisms (bacteria and fungi) were used to deselect extracts from further study. The microorganisms used to dereplicate antibiotic-containing extracts were *Staphylococcus aureus*, methicillin-resistant *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Trichophyton mentarophytes*. In addition, all active extracts that showed mammalian toxicity in the monkey kidney Vero cell cytotoxicity assay were also deselected for further examination.

The most potent cyanobacteria-selective extracts (that were not significantly antibacterial and were non-toxic) were those obtained from the roots and stems of a Peruvian collection of *Dulacia candida* (family = Olacaceae). *Dulacia candida* is a widely distributed tropical Amazonian shrub. The crude *D. candida* extracts showed extremely potent anti-cyanobacterial activity, approaching the parts per billion range. The components of the extract were separated and the bioactivity was confined to several moderately non-polar chromatographic fractions. However, the substance (or substances) responsible for the potent anti-cyanobacterial have not yet been identified.

In addition, more than one hundred pure biologically active compounds from marine invertebrates and algae have been examined for anti-cyanobacterial activity. Some of these substances inhibited algae growth, but were either too cytotoxic or non-cyanobacteria selective.

**USDA-ARS.** Proprietary analogs of a promising natural product were tested in ponds for selective activity against blue-green algae. Efficacy testing yielded promising results, and one of the best proprietary compounds (as determined by laboratory screening studies)

significantly reduced the MIB-producing cyanobacterium *Oscillatoria perornata* and the cyanobacterium *Raphidiopsis brookii* at application levels of 125 ppb. Numbers of other cyanobacteria such as *Oscillatoria agardhii* and *Oscillatoria geminata*, green algae, and diatoms were not significantly affected by 125 ppb of this proprietary compound. Based upon the results, this proprietary compound is promising as a selective algicide to help prevent the growth of *O. perornata* and *R. brookii* in catfish aquaculture ponds, but it is not a broad-spectrum blue-green algicide. Additional efficacy testing is needed to confirm effective application rates.

Additional testing of several of the most prom-

ising proprietary compounds has determined that they are neither carcinogenic, antifungal, antibiotic, nor anti-protozoal. Analogs were also evaluated for acute toxicity and histopathology using channel catfish fingerlings. Acute toxicity was determined through measurement of mortality following 96 hours of aqueous exposure. The 96-hour LC50 for the analogs tested ranged from approximately 2-6 ppm. Histological analysis revealed abnormalities in the gills of treated fish. It was determined that the lethality of the analogs is related to histological changes in the gills of the catfish.

Additional screening of pure natural compounds and crude plant extracts has identified several other leads for blue-green algicides.

**Objective 2.** *Evaluate nutrient manipulation to promote desirable phytoplankton community structure.*

**Mississippi State University.** In experiments repeated over two years, eight 1,450-gallon enclosures (limnocorrals) were placed in a small research pond or a commercial earthen fish pond in which a dense phytoplankton community dominated by *Oscillatoria agardhii* was present. The N:P ratio of four enclosures was adjusted by addition of KNO<sub>3</sub> to provide a N:P ratio of about 30 and chelated iron was added to provide 1 ppm iron. Four enclosures did not receive nutrient additions. All enclosures were supplied with diffused aeration to produce gentle turbulence. Water samples collected every 2 to 3 days were analyzed for nutrients, solids, and indices related to phytoplankton biomass and community composition. After 2 weeks, the combination nutrient addition did not result in a shift of the phytoplankton community from dominance by *O. agardhii*. Nitrite concentrations increased and soluble phosphorus concentrations declined in nitrate-treated enclosures. Algal biomass in untreated enclosures

declined, suggesting that continued nutrient supply is necessary to sustain high algal biomass. Nutrient manipulation does not appear to hold promise as a technique to effect phytoplankton community structure in hypereutrophic aquaculture ponds.

### *Results at a glance...*

☆ *Studies in Mississippi and Alabama indicate that various manipulations of waterborne plant nutrients have little promise for controlling phytoplankton community composition in catfish ponds with high feeding rates.*

**Auburn University.** An initial laboratory study considered the effectiveness of the chelating agents ethylenediamine tetra-acetic acid (EDTA),

lignin sulfonate, and citric acid for maintaining iron in solution. EDTA was the most promising of the chelating agents, because iron remained at concentrations above 0.5 ppm for 30 days in soil-water systems treated with 1 ppm iron from iron-EDTA. Thus, it was decided to use EDTA chelated metals for pond research.

Pond studies in 1999 had three treatments: (1) chelated iron (0.5 ppm) plus chopped legume hay (40 kg/ha per week); (2) chelated iron, chopped legume hay, plus a trace element mix (1 kg/ha per week); (3) control. The hay applications resulted in low dissolved oxygen concentrations, and no benefits related to blue-green algae control were observed.

In 2000, two treatments (legume hay at 20 kg/ha per week and legume hay plus 0.5 ppm chelated iron and 1 kg/ha per week of trace element mix) are being compared to the control. No benefits of treatments on water quality or blue-green algae control were observed.

In 1999, potassium chloride treatments of 0, 30, 90, and 120 ppm did not result in significant differences in blue-green algal abundance. A sodium nitrate based fertilizer containing 8% N, 24% P<sub>2</sub>O<sub>5</sub>, and 15% K<sub>2</sub>O was as effective as a

standard, 10-34-0 liquid fertilizer in promoting sunfish production.

Studies conducted in bait minnow ponds failed to show a need for nitrogen fertilizer. Phosphate-only fertilization was as effective as nitrogen plus phosphorus fertilization. However, sodium nitrate was shown to be a more "environmentally-friendly" source of nitrogen than ammonium sulfate. This year, (2001), four different fertilizer treatments (20-0-0; 0-20-0; 20-20-0;10-20-0) are being conducted in earthen ponds stocked with bluegill. So far, no differences in algal composition or abundance have been noted among 0-20-0, 20-20-0, and 10-20-0 treatments. There is much less algal phytoplankton growth in the 20-0-0 treatment than in the other treatments.

The work on the pond soil cores from bait minnow ponds in Arkansas was completed this year. The major effect of pond aging on sediment quality was the accumulation of soft sediment of high phosphorus concentration over time. There was a drastic decline in sediment quality in 30- to 35-year-old ponds as compared to 7- to 25-year-old ponds. Laboratory experiments indicated that sodium nitrate treatment would not improve sediment quality in older ponds. The problem of impaired sediment quality in old ponds can probably be best resolved by removing soft sediment from ponds to provide firmer bottoms.

**University of Arkansas at Pine Bluff.** A laboratory study was conducted to evaluate the effects of incorporating sodium nitrate into pond bottom soils. Two common soils (Perry-Portland, known locally as gumbo, and Calloway-Calhoun-Loring, or crawfish) were collected from the bottoms of 20- to 45-year-old commercial baitfish ponds. A layer (4.5 L) of gumbo or crawfish soil was added to each of 24 microcosms (13-L buckets) and sodium nitrate was soil-incorporated at rates of 0, 25, 50

## *Results at a glance...*

- ★ *Phosphorus-only fertilization was as effective as nitrogen plus phosphorus fertilization in bait minnow ponds. Phosphorus-only fertilization is less expensive, it conserves nitrogen, and it lessens the possibility for nitrogen pollution of natural waters by pond effluents. Removal of soft sediment from old (25 years or more) bait minnow ponds should improve bottom soil quality.*

or 75 g N/m<sup>2</sup>. Buckets were filled with pond water, and sodium acetate was added throughout the study as a source of organic matter, to create anoxic conditions and simulate pond bottom waters.

Results showed that incorporating sodium nitrate suppressed phosphorus release in both soils for 11-19 days. However, elevated nitrite concentrations were also found during this period. Nitrite levels as high as 180-250 ppm were measured in the high rate treatment. The higher the rate of sodium nitrate, the longer was the duration of phosphorus suppression, but the additional time was not proportional to the dose.

A second laboratory study was conducted to evaluate the effects of adding different forms of sodium nitrate through the water column on pond bottom soil. A layer of gumbo soil collected from a 45-year-old commercial baitfish pond was added to each of 25 microcosms (13-L buckets), buckets were filled with pond water, and sodium acetate was added as a source of organic matter, to create anoxic conditions and simulate pond bottom waters. Sodium nitrate was added to the buckets at a rate of 50 g N/m<sup>2</sup> in the form of a powder, prill, or two types of coated prills (short-term and long-term). Coated prills were suggested as a mechanism to permit application of sodium nitrate through the water column in established ponds. The short-term coating was designed to release 20% the first day and to finish release by day 10 to 15, while the long-term coating was intended to slowly release sodium nitrate over a 2- to 3-month period. Results showed that compared to controls, adding sodium nitrate suppressed phosphorus release regardless of product form, with the long-term coated prill treatment lasting 10 days. Elevated nitrite levels were found in all buckets treated with sodium nitrate, however, nitrite concentrations were lowest in the long-term release prill treatment (highest concentration for this product form was 13 ppm NO<sub>2</sub>). Nitrite levels as

high as 180-250 ppm were measured in containers with the other three product forms.

Conditions in experimental microcosms differ significantly from those in ponds, and additional work in enclosures within ponds is currently in progress. Sodium nitrate in the form of time release (long-term) coated prills was applied to four of eight 4.7-m<sup>2</sup> limnocorrals installed within a goldfish brood pond. A blower system directs an air current over the surface of each corral to provide mixing without aeration. Water quality is being monitored to determine phosphorus and nitrite levels within the limnocorrals.

Pre-treatment water quality and plankton community data were collected from 12 commercial golden shiner ponds at monthly intervals. Study ponds ranged in size from 7 to 25 acres (3 to 10 ha) and represented two soil types and two ages (20-25 years and 40-45 years). Using a column sampler, water samples were collected from a single location in each pond. Results showed that for parameters linked to phytoplankton abundance (i.e., chlorophyll *a*, chemical oxygen demand, total phosphorus, and total nitrogen), seasonal water quality changes in golden shiner ponds were similar to those reported for channel catfish culture. Dissimilar results were found for dissolved inorganic nitrogen, which was highest in the fall rather than in the winter, as has been reported for catfish. This may reflect the relatively low feeding rates used for baitfish. In addition, relatively higher SRP concentrations were found in the summer months, perhaps a result of powdered feed used for young fish.

Pond renovation was evaluated as a technique to reduce problematic blue-green algae blooms and associated water quality problems. Monthly water quality and plankton community data were collected from 12 commercial goldfish ponds for a year. Six of the 12 ponds were renovated during the winter (1999-2000) and

returned to production in late spring. Water quality monitoring was then resumed in the renovated ponds and was continued for the growing season. Results indicated little difference in water quality in renovated ponds as compared to controls. Soluble reactive phosphorus was significantly higher in renovated ponds for the first two months after ponds were returned to production, reflecting pond management practices for rearing new crops of fish. Pre- and post-renovation soil testing showed no overall significant difference in sediment phosphorus concentrations. This may reflect the fact that sediment from inside ponds is used to rebuild ponds levees. However, pond renovation significantly reduced the phosphorus level in the upper 2 cm of sediment.

### *Results at a glance...*

☆ *Pond renovation had little effect on subsequent levels of soluble reactive phosphorus in baitfish pond waters or on average concentrations of phosphorus in bottom sediments. A possible explanation for these results is that pond bottom soils are used to rebuild levees during renovation and not removed from ponds. Bottom soils are mixed in the pond reconstruction process, and renovation decreased the average soil phosphorus concentration in the upper 2 cm of sediment.*

Sulfate-sulfur concentrations in pond bottom sediments averaged 98 ppm, and ranged from 3 to 417 ppm. Compared to typical levels in terrestrial soils in the study area, these concentrations are very high. Sulfate sulfur results from the decomposition of organic matter and typically is higher in clayey soils.

**Louisiana State University.** Concentrations of 2-methylisoborneol and geosin were determined from March, 1999, through February, 2000, in fifteen 0.2-acre channel catfish experimental catfish ponds located at the Aquaculture Research Station, Louisiana State University Agricultural Center, in Baton Rouge. Ponds were assigned randomly to three mineralization (salinity) levels: 0.0 parts per thousand (ppt) - control, 1.5 ppt, and 3.0 ppt. Nominal mineral concentrations were established and maintained with periodic additions of salt (NaCl). Water samples were collected twice a weekly and analyzed by solid phase micro-extraction gas chromatography mass spectrometry for MIB and geosmin concentrations. All ponds contained little or no MIB until late May. All ponds experienced off-flavor episodes at one time or another. Only trace levels of geosmin were observed throughout the year. Three major increases in the concentration levels of MIB were observed; one in early June, a second in early August, and a third in late September. Increases were not observed in all ponds. In several cases, concentrations of MIB increased rapidly from barely quantifiable levels (<0.1 ppb) to very high levels (>30 ppb) over a 3-4 day period. A slight reduction in the concentrations of MIB observed in the ponds with 3.0 ppt NaCl. Ponds with 1.5 ppt routinely contained higher levels of MIB than the control (0 ppt NaCl); but there is insufficient data determine the impact on the mitigation of off-flavor episodes by using low levels of NaCl.

A simplified method for analyzing for the muddy, musty off-flavor in catfish was developed that uses the liver of the fish rather than the muscle tissue. Slurried liver tissue permits partitioning of the analytes directly from the sample, thus eliminating the need for the microwave distillation step. Sample preparation takes less than five minutes per sample and GC/MS analysis takes 15 minutes from start to finish, for a total analysis time of 20 minutes. A comparison of the concentrations of MIB and geosmin from fish

fillet tissue and the liver was made by dosing fish (previously depurated for 96 hours) with MIB and geosmin at a final concentration of 3 ppb. Fish were sampled at 0, 2, 24, 48, and 72 hours after dosing to assess uptake of off-flavor compounds. The amount of 2-MIB and geosmin recovered directly from the liver was near 5%, while the recovery from muscle tissue employing microwave distillation approaches 60%. Fish were off-flavor after only one hour of exposure. The concentrations determined from the analysis of the livers are consistent with those determined from the fillets.

**North Carolina State University.** In 1999, a study was conducted at the Tidewater Research Station to determine the effect of applications of trace metals and organic matter on water quality and phytoplankton population dynamics in hybrid striped bass ponds. Twelve 0.25-acre hybrid striped bass (*Morone chrysops* ×

*M. saxatilis*) ponds were stocked (4,000/acre) in the spring and managed according to standard commercial hybrid striped bass culture procedures. Chelated iron, a mineral mix, and alfalfa pellets were periodically applied to six of the ponds during an entire growing season. Ponds were harvested in late fall. Water samples were taken weekly and analyzed for nutrients and phytoplankton composition, and soil samples taken at the beginning and end of the study were analyzed for total nitrogen, total carbon, organic matter, pH and metals. Blue-green algae began to appear in samples during the second week of September 1999 and dominated phytoplankton composition in all treatments during the month of October 1999. Application of iron, minerals, and organic matter did not result in any differences in phytoplankton species composition, fish production, soil quality or water quality when compared to control ponds.

**Objective 3.** *Evaluate water circulation as a means of altering the environment to promote desirable phytoplankton community structure.*

**Louisiana State University.** Twelve 0.1-acre ponds were stocked with multiple cohorts of channel catfish at a nominal stocking density of 10,000/acre. In eight ponds, fish were restricted to approximately a quarter of the pond area by a barrier placed across the pond width. In these ponds, a continuously-operating, horizontally-mounted pump mixed water between the area containing fish with the open area of the pond. In four of the mixed ponds, threadfin shad were stocked at 200/acre in the open area of the pond. Although the experiment is ongoing, there are no differences in water quality, phytoplankton community composition, or feeding rate among the three treatments.

Three water management practices were evaluated, each at two levels (presence or absence), alone, and in combination, to determine their

effects on blue-green algal community composition and water quality in experimental mesocosms managed to simulate commercial catfish production practices. In one treatment, aluminum sulfate (alum) was applied weekly at 3 ppm to reduce phosphorus (chemical control). In a second treatment, the water column was destratified by continuous vertical mixing in contrast to conventional surface aeration (physical control). In the third treatment, planktivorous gizzard and threadfin shad (*Dorosoma* spp.) were stocked at 25,000 juveniles/acre with channel catfish, to evaluate their ability to control blue-green algae (biological control).

The eight treatment combinations, arranged in a 2<sup>3</sup> factorial design, were randomly assigned to twenty-four 3,000-gallon fiberglass tanks with



soil bottoms (mesocosms) with three replicates per treatment combination. Catfish juveniles (mean = 52 g) were stocked in May at 10,000/acre, fed a 32% crude protein commercial feed daily at rates ranging from 40 to 150 pounds/acre, and harvested in November. Water samples were collected biweekly for nutrient and phytoplankton analysis.

Mean catfish survival was 88.5% and yield averaged 7,133 pounds/acre, with no observed differences related to water management practices. Shad biomass averaged 625 pounds/acre at harvest. The alum reduced soluble reactive phosphorus in October, but had no effect on phytoplankton density or community composition. Suspension of sediments in the water column from vertical mixing increased total nitrogen, total phosphorus, nitrate, and pH but had no discernible effect on the phytoplankton community. The presence of shad significantly reduced total algal biomass as evidenced by reductions in total nitrogen, total phosphorus, chemical oxygen demand, and chlorophyll *a*. Although the percentage of blue-green algae in the phytoplankton community was not significantly reduced compared to mesocosms without shad, odorous species of blue-green algae (*Oscillatoria perornata* and *Anabaena* spp.), known to cause off-flavor in catfish, were nearly eliminated by the presence of shad. Mesocosms with shad never had odorous species of blue-green algae that accounted for more than about 3% of the blue-green algal community, while mesocosms without shad were as high as 20%. Shad had no impact on catfish production.

**North Carolina State University.** Twelve ponds at the Tidewater Research Station were stocked in spring of 2000 with hybrid striped bass (*Morone chrysops* × *M. saxatilis*) at 4,000/acre to evaluate the effectiveness of water circulation for controlling blue-green algae abundance in hybrid striped bass ponds. Ponds were managed according to standard procedures and

harvested late fall. In six of the ponds, circulators were placed to produce whole-pond horizontal water circulation. Effectiveness of the water circulators in creating currents that circulated throughout the pond was evaluated with gypsum blocks. However, the water circulators used were very unreliable, and repeated mechanical problems throughout the duration of the study resulted in both decreased water circulation and the loss of several replicates in the treatment receiving circulation. Although water quality and phytoplankton population analysis revealed no differences between treatments, the frequent breakdowns of circulators resulted in inconsistent water circulation and prevented a meaningful evaluation of the circulation treatment. Consequently, a second evaluation of water circulation in hybrid striped bass ponds was planned for the 2001 production season.

In May 2001, hybrid striped bass (*Morone chrysops* × *M. saxatilis*) were stocked at 4,000/acre into twelve 0.25-acre ponds at the Tidewater Research Station. In six of the ponds, water is circulated during daylight hours (from 0900 to 1600) with a 0.5-hp pump (70 gallons/minute) placed on the pond bank. The pumps draw water from near the pond bottom and discharge at the surface approximately a third of the way down the length of the pond. Water samples are analyzed weekly or biweekly for concentrations of total and soluble reactive phosphorus, ammonia, nitrite, nitrate, total suspended solids, chemical oxygen demand, biochemical oxygen demand, pH, and chlorophyll *a*. In addition, phytoplankton abundance and composition were measured biweekly. Ponds will be harvested in November 2001. Preliminary analysis of the water quality data does not reveal any differences to date among treatments. However, phytoplankton analysis during the 2000 production season revealed that blue-green algae are usually not predominant in these ponds until late summer/early fall, and it is at this time

when any treatment effects would be most pronounced.

**Mississippi State University.** In three experiments, twelve 0.1-acre ponds were stocked with multiple cohorts of channel catfish at a nominal stocking density of 10,000/acre. In the first experiment, fish in eight ponds were restricted to approximately one-quarter of the pond area by a barrier placed across the pond width. In these ponds, a continuously-operating, horizontally-mounted ½-hp pump mixed water between the area containing fish with the open area of the pond. In four of the mixed ponds, threadfin shad were stocked at 200/acre in the

open area of the pond. In the second and third experiments, a baffle oriented along the long axis of the pond was placed in eight ponds. One ½-hp pump mixer was placed in each of four baffled ponds and two mixers were placed in each of the other four baffled ponds. Four ponds did not have a baffle and were not mixed. Results were similar in the three experiments: there were no differences in water quality, phytoplankton community composition, or feeding rate among the three treatments. Ponds with two mixers were more turbid than ponds in the other treatments. Turbidity in ponds with two mixers was dominated by suspended mineral matter.

**Objective 4.** *Evaluate the use of plankton-feeding fish to alter the environment to promote desirable phytoplankton community structure.*

**Auburn University.** Ten 0.1-acre earthen ponds were stocked with 9-g channel catfish at a density equivalent to 6,000/acre, with 0.3-pound grass carp at 20/acre. Five randomly selected ponds were stocked with 8-g threadfin shad at 800/acre. Each pond was fed once daily to apparent satiation with a commercial floating feed (32% crude protein). All ponds were harvested 8 November 1999 and fish were identified, sorted and weighed. One channel catfish was randomly selected from each pond for flavor analysis.

Total threadfin shad mortality occurred in one of the shad ponds (9 September 1999). One of the no-shad treatment ponds experienced a catfish kill during the final two weeks of September when they had attained an average weight of 0.7 pounds. Observed mortality during this period was 31% of the original stock and only 48% of the original stock was recovered at harvest. Following the shad mortality, that pond was eliminated for further consideration of water quality and phytoplankton analysis for the shad treatment.

The presence of shad had no effect on temperature, dissolved oxygen concentrations, pH, or

### *Results at a glance...*

☆ *In both Alabama and Georgia, stocking threadfin shad with channel catfish resulted in lower ammonia and nitrite levels late in the growing season. The improved environmental conditions apparently resulted in significantly better survival of catfish in the presence of shad in the Alabama study.*

total alkalinity of pond waters. Total organic carbon concentrations ranged from 6.3 ppm in April to 30.5 ppm in October in the shad treatment and from 4.7 ppm in April to 34.1 ppm in September in the no-shad treatment. Total organic carbon levels increased in both treatments during the growing season and were higher in

the shad treatment on 27 April and 25 May and higher in the no-shad treatment on 19 August.

Total ammonia-nitrogen (TAN) concentrations increased in both treatments throughout the growing season, but reached higher levels in the no-shad treatment. In the shad treatment, TAN concentrations ranged from 0.03 ppm in April to 1.92 ppm in October. In the no-shad treatment, TAN ranged from 0.03 ppm in June to 3.88 ppm in September. TAN concentrations in the no-shad treatment were significantly higher than concentrations measured in the shad treatment for the period 1 September through the end of the study. Nitrite-nitrogen concentrations in both treatments remained below 2.0 ppb until July and then began to increase, reaching a maximum of 37.2 ppb in the shad treatment in September and 94.5 ppb in the no-shad treatment in October. For the period 1 September through 25 October, nitrite-nitrogen levels were significantly higher in the no-shad ponds.

Phytoplankton abundance (as indicated by chlorophyll *a* levels) were low initially and increased progressively throughout the study to highs of 263 ppb chlorophyll *a* in the shad treatment in September and 285 ppb in the no-shad treatment in August. Chlorophyll *a* levels were significantly higher in the no-shad treatment on only one sampling date. There were no differences overall in phytoplankton abundance when data for chlorophyll *a* levels for all sampling dates in September and October were combined. Phytoplankton community characteristics and size distribution are currently being examined.

Survival of channel catfish in ponds with threadfin shad was 92%, which was higher than the 77% survival in ponds without shad. Catfish production was also higher in ponds with shad (4,651 pounds/acre) than in ponds without shad (3,980 pounds/acre). However, average weight at harvest was similar in both treatments (over-

all mean = 0.80 pounds). Feed conversion was marginally better in ponds with shad (1.30) than in ponds without shad (1.40).

Off-flavor analysis was performed on one catfish from each pond. The catfish were filleted with the skin-on, microwaved, and served to a panel of three taste testers. Slight off-flavor was detected by two of the three panelists in both treatments, but no significant differences were found.

### *Results at a glance...*

☆ *Polyculture of threadfin shad with channel catfish in relatively small experimental ponds resulted in improved water quality conditions and enhanced catfish survival. The stocking of threadfin shad in four commercial channel catfish ponds in West Alabama did not result in improved water quality when compared to four similar ponds with catfish but no threadfin shad.*

In 2000, eight commercial channel catfish ponds were selected for analysis on three catfish farms in West Alabama. Four ponds with catfish and established threadfin shad populations were selected along with four similar ponds stocked only with channel catfish. All ponds were managed with common commercial practices. Nightly aeration was used in all ponds to prevent fish loss caused by low dissolved oxygen concentrations. Copper sulfate was added periodically to six of the eight ponds for algal management. Partial harvests of catfish were carried out in ponds throughout the study. Water samples were collected twice a month, from May through October 2000.

Mean total organic carbon (TOC) increased

gradually through the entire growing season in both treatments. The TOC concentrations in the shad treatment (mean = 34.26 ppm; range = 22.1 ppm to 56.5 ppm) were significantly higher than TOC concentrations in the no-shad treatment (mean = 27.57 ppm; range = 20.8 ppm to 38.4 ppm).

Mean total ammonia-nitrogen concentrations also were higher in the shad treatment (mean = 1.6 ppm; range = 2.3 ppm in June to 0.3 ppm in September) than in the no-shad treatment (mean = 0.7 ppm; range = 0.3 ppm in October to 1.2 ppm in September). Nitrite nitrogen ( $\text{NO}_2\text{-N}$ ) concentrations did not differ significantly between treatments, remaining below 0.1 ppm throughout most of the growing season.

Mean chlorophyll *a* concentrations (corrected for phaeophytin) varied throughout the growing season in both treatments, but the mean concentration for the entire growing season was significantly higher in the shad treatment (mean = 299 ppm; range = 153 ppm to 407 ppm) than in the no-shad treatment (mean = 155 ppm; range = 66 ppm and 308 ppm). Phytoplankton community structure and size distribution are currently being examined.

This study did not reveal an improvement in water quality of commercial channel catfish ponds containing threadfin shad. In fact, concentrations of TOC, total ammonia, and chlorophyll *a* were significantly higher in the shad treatment than in the no-shad treatment. These ponds were stocked, harvested, fed, aerated, and chemically treated (with copper sulfate) independently by three different farm managers. As such, water quality differences measured between treatments were likely not solely caused by the presence or absence of threadfin shad.

**University of Georgia.** Threadfin shad or fathead minnows were stocked with catfish in 0.25-acre earthen ponds at Tifton, Georgia, and

compared to ponds with only channel catfish. At Cohutta, Georgia, two treatments were started comparing threadfin shad and channel catfish to channel catfish alone in 0.1-acre earthen ponds. Three replicate ponds were used for each treatment for a total of nine ponds at Tifton and six at Cohutta. Channel catfish were stocked as fingerlings in multiple sizes at 44,500/acre. Threadfin shad were stocked at about 2,500/acre and were 1.7 to 4 inches long. Fathead minnows were stocked at about 10 pounds/acre (8,900 to 10,000/acre) and were 1.4 to 2 inches long.

Threadfin shad stocking was difficult due to the fragility of this species during handling, hauling, and transfer into receiving waters. Five attempts were made to stock threadfin shad at both locations. The most successful method of threadfin shad stocking was to obtain 1.5 to 2 inch shad from local ponds in the months of January to April. Even under the best conditions, it is difficult to determine the survival of the threadfin shad after stocking. Stocking threadfin shad into holding ponds and seining after one or two weeks indicated that 30-90% of the threadfin shad could die a short time after stocking due to loss of scales during handling, temperature shock, alkalinity shock, salinity shock, or other stress due to handling or transfer. Sorting threadfin shad from gizzard shad, which often is found together with the threadfin in lakes, rivers, and aquaculture ponds, causes an increase in threadfin shad losses. Cast nets or seines can be utilized for capturing threadfin shad. However, each method of capture has disadvantages. Casting nets near paddlewheel aerators appears to be successful for capture of monospecific harvests of threadfin shad. Hauling aids should be utilized during transport and may include an anesthetic, sodium chloride, calcium chloride, antifoaming agents, or a buffer of pH 8.0 to 7.0. Tempering should be extended to two hours of gradual exchange of hauling water with receiving

water, even when similar water temperatures are measured in the two water sources.

Over 50 algal species have been identified from Georgia ponds during the growing season. Blue-green algal blooms are denser at the Tifton location than at the Cohutta location. Water temperatures are cooler at Cohutta and the water source is a spring from limestone caverns. The water source at Tifton is the Floridan aquifer. No differences among treatments were observed in 1999. All ponds had blue-green algae in abundant populations. Establishment of threadfin shad populations was variable and appeared to affect the observed phytoplankton population densities. Off-flavors were not detected in channel catfish harvested from the study in 1999 or 2000. Off-flavors were detected in fish from control ponds at Tifton in 2001.

In 1999, total ammonia concentrations were lower in the shad and minnow treatments than in the ponds with only channel catfish. Also, nitrite concentrations were lower in the minnow treatment than in the other two fish combinations. Soluble reactive phosphorus was similar in all ponds.

In 2000 at Tifton, blue-green algae became abundant in ponds with channel catfish only as early as April, in May with fathead minnows and catfish, and in June with threadfin shad and catfish. Blue-green populations reached 100 million cells/milliliter in ponds with channel catfish only, 80 million with fathead minnows and catfish, and 35 million with threadfin shad and catfish. At the Cohutta location, blue-green algae did not become abundant until August. It was apparent that blue-green algae were less abundant in the ponds with threadfin shad; however, all ponds had blue-green algae blooms by late summer.

In 2001 at Tifton, the presence of threadfin shad

reduced the number of blue-green algae colonies versus the control or fathead minnow treatment. Blue-green algal numbers were higher in fathead minnow ponds from March through August than in control ponds. At Cohutta, the blue-green algae, *Microcystis aeruginosa*, became abundant later than at the Tifton location. Few blue-green algal species were observed in ponds containing threadfin shad by August. The number of hours of aeration was reduced in ponds containing threadfin shad.

**Louisiana State University.** Eighteen 0.1-acre earthen ponds at the ARS-LAES, were stocked

### *Results at a glance...*

- ★ *Blue-green algae numbers were reduced by threadfin shad over a three-year period in intensively-managed catfish ponds.*

with channel catfish fingerlings in May 2000 at 10,000 fish/acre to study the efficacy of threadfin shad for control of blue-green algae. Juvenile and adult threadfin shad were captured from a local lake for stocking in the experimental ponds, but physiological stress associated with high water temperatures resulted in > 90% mortality in the shad. Surviving shad were held in ponds and were to be stocked into catfish ponds in November-early December 2000. However, the study was irreversibly compromised when a flock of 300 white pelicans entered the research facility in December and dramatically reduced catfish and shad populations, thereby requiring repetition of the experiment. The 18 ponds were re-stocked with catfish fingerlings in April 2001 at 10,000/acre, and 6 one-acre ponds were stocked with mixed size classes of channel catfish and channel x blue catfish hybrids at densities of ~ 7,000 fish/acre in summer. Threadfin shad

will be stocked half the replicate ponds (0.1 and 1-acre) in November/December 2001 when temperatures cool sufficiently to safely transport

and stock the fish. Heavy monofilament with reflectors will be placed over all experimental ponds to minimize depredation by avian predators.

**Objective 5.** *Evaluate the development of phytoplankton communities in the Partitioned Aquaculture System.*

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### **Clemson University**

#### Results from the 1999 Season

The 2-acre commercial scale PAS unit was brought into production in the spring of 1999 (Figure 1).

The unit was stocked with 33,000 catfish fingerlings in May. Stocked fingerlings averaged 15, 31, 48, 61 and 80 g. The catfish were stocked in 2 raceways consisting of 4 sections each. In addition, 1,320 pounds of tilapia (15,000 fish at

40 g each) were stocked into the 2-acre system. An additional 216 breeding tilapia (250-300 g) were stocked into the algal basin. The 2-acre unit successfully produced 14,500 pounds/acre of catfish at a carrying capacity of 17,000 pounds/acre. Feed applications reached 260 pounds/acre-day (Table 1). The 2-acre unit was dominated by green algal populations throughout the season and off-flavor in 1999 ranged from 1.0 to 1.5 (out of 5) at the time of harvest. This mild off-flavor was described as being "grassy".



Figure 1. Overview of 2-acre (above) and six-1/3 acre PAS units at Clemson University.

Year	Avg. max. catfish carrying capacity lb/acre	Avg. seasonal catfish yield lbs/acre	Avg. feed application lbs/acre	Avg. max. feed application lbs/acre	Tilapia co-production lbs/acre	Catfish feed conversion ratio
1995	3,258	3,078	20	60	0	1.34
1996	8,811	8,151	60	150	0/805 <sup>a</sup>	1.53
1997	14,054	12,965	94	200	1,826	1.41
1998	16,694	14,952	113	210	2,266	1.40
1999	10,479	11,474	100	260	3,843	1.60
2000	14,108	17,232	115	260	5,407	1.50

<sup>a/</sup> In 1996, three of the units contained tilapia (0 lbs/acre production) and three of the units contained tilapia (805 lbs/acre production).

Six 0.33-acre PAS units were stocked with both adult tilapia alone (breeding pairs) and with tilapia fingerlings and adults to see if successful algal species control could be sustained with reducing stocking requirement through the use of breeding pairs. By mid-season, four of the six 0.33-acre PAS units shifted from early blue-green dominance back to populations of more desirable green algae as the tilapia breeding pairs expanded in numbers and weight. In two of the 0.33-acre PAS units, late season algal populations shifted to predominantly blue-green populations suggesting that use of tilapia breeding pairs alone at these feed application rates is close to, or slightly beyond, the limit of blue-green population control.

Stocking density experiments were conducted showing that raceway catfish stocking could be increased from 4-5 pounds/cubic foot to 8-10 pounds/cubic foot with no adverse effect on

growth. These results demonstrated that overall system costs can be reduced by using a single high-density raceway and fewer raceway paddlewheels. A preliminary economic analysis projects that 40 acres of PAS units would produce catfish at a 5 to 15¢/pound lower cost than conventional pond culture (Table 2). However, this analysis is based on the assumption the net production would exceed 22,000 pounds/acre. Because of loss of winter fish carryover as a result of spring proliferative gill disease (PGD), this potential yield has yet to be realized.

Flow experiments were conducted in 1999 to determine the uniformity of the water velocity field that can be sustained with different combinations of paddles and paddle speeds. The results suggest that sufficient mixing and flow velocity in the algal channel can be maintained with 50% of algal channel width coverage by paddlewheels (Figure 2).

Table 2. Estimated PAS Annual Ownership Costs, Operating Costs, and Unit Costs Comparison Summary (1999).		
	160-Acre Convention Ponds	20, 2-Acre PAS Unit Farm
Annual ownership cost	\$ 95,301	\$114,181
Annual operating cost	418,325	418,617
Total Annual Cost	513,626	532,798
Total Annual Cost/Acre	3,669	13,319
Annual pounds harvested	700,000	909,840
Ownership cost/pound	0.136	0.125
Operating cost/pound	0.598	0.460
Total Cost/Pound	0.734	0.585
<b>TOTAL REVENUE</b>	\$525,000	\$681,630

#### Results from the 2000 Season

In May, the 2-acre PAS unit was stocked with 33,419 catfish fingerlings (2,050 pounds/acre) in size classes of 20, 65, and 88 g. In addition, 110 pounds/acre of tilapia breeding pairs were stocked at large in the algal basin (150 - 250 g males and 300 - 250 g females). The 1999 carryover fish (250 g each) were lost in spring 2000 in the 2-acre unit due to proliferative gill disease (PGD). Treatment of pond sediment with hydrated lime was observed to be effective in reducing the occurrence of PGD, although treatment was initiated too late to be effective.

The use of "breeding pairs" of tilapia alone in the 2-acre PAS produced marginal water quality results. By the third week of August, the phytoplankton community was completely dominated by blue-green algae. However, at the end of August, communities were undergoing a shift to green algal populations, an apparent result of increased filter-feeding activity of the expanding tilapia population.

In contrast, four 1/36-acre PAS units operating in August of 2,000 at 12,000 pounds/acre catfish carrying capacity, with 1,600 pounds/acre of native mussel in two units and 2,300 pounds/acre of silver

carp in two units, exhibited a dominance of green algal populations. However, the mussels were not as effective as silver carp or tilapia at controlling blue-green algal populations and, as a result, the visual color difference between the mussel and silver carp unit was obvious (Figure 3). In August 2000 the 0.33-acre units shifted to green algal populations using tilapia breeding pairs.

### *Results at a glance...*

☆ *The PAS technique offers the potential to provide a method to quadruple current fish production in a system which eliminates blue-green algal dominance and associated fish off-flavor problems, while recovering wasted nitrogen and phosphorus discharges, which currently pose an eutrophication threat to surface and groundwater supplies. Economic projections suggest that PAS catfish production costs are \$0.15 to \$0.17 per pound lower than conventional pond production costs.*



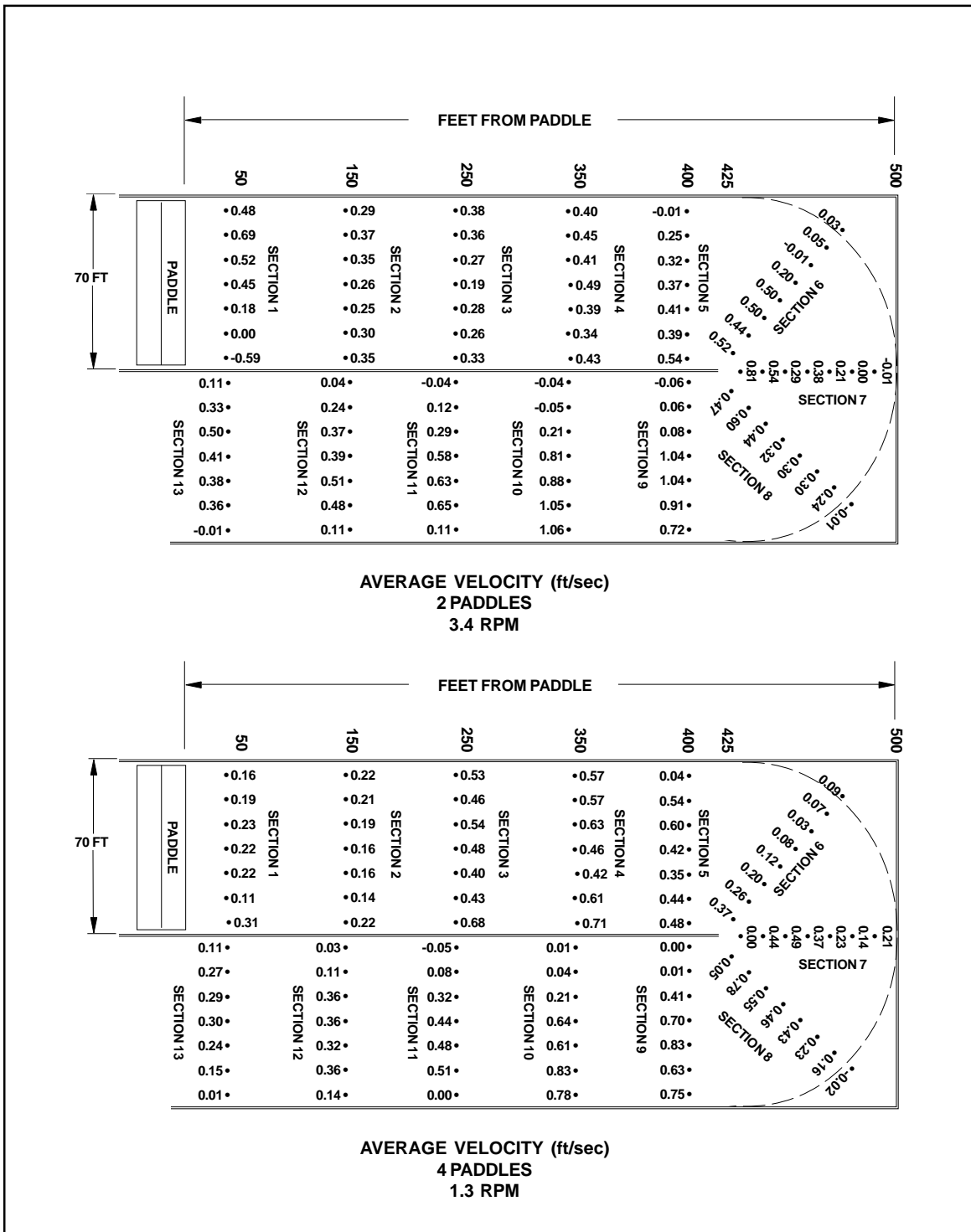


Figure 2. Velocity profiles in 2-acre PAS with 2 paddles and with 4 paddles.



Figure 3. Visual algal differences between mussel PAS unit (right) and silver carp PAS unit (left).

In Spring 2000, the net production for this season reached a 6-year average high in the six 0.33-acre units of 17,332 pounds/acre of catfish with 5,407 pounds/acre of tilapia. Average feeding rates reached 115 pounds/acre per day, with a maximum feed application of 260 pound/acre per day (Table 1). Because of excessive over-winter losses, the 2-acre prototype only produced 12,480 pounds/acre of catfish at the end of 2000.

#### Results from 2001 Season

In Spring 2001, the six 0.33-acre PAS units were stocked with an average of 2,931 carryover catfish (mean weight = 394 g), 3,484 large catfish fingerlings (mean weight = 71 g), and 2,092 small

catfish fingerlings (mean weight = 26 g). The six 0.33-acre units were also stocked with 25 male tilapia (160 g) and 50 female tilapia (76 g). The 2-acre prototype PAS unit was stocked with 10,500 carryover catfish 318-g, along with 8,734 (132 g), 9,872 (114 g), and 11,159 (21 g) catfish fingerlings. The 2.0-acre system was also stocked with 450 breeding pairs of tilapia. Prophylactic treatments for control of early spring PGD in the winter carryover fish were conducted in winter/spring of 2000. The incident of PGD was significantly reduced in the carryover fish in 2001. By the end of July of 2001, the 2-acre prototype system shifted to predominantly green algal population (Figure 4).

## **WORK PLANNED**

Work on all objectives is proceeding on schedule. No major changes in the work plan have occurred.

Refinements to the work plan or additional work is planned in some areas, as noted below.

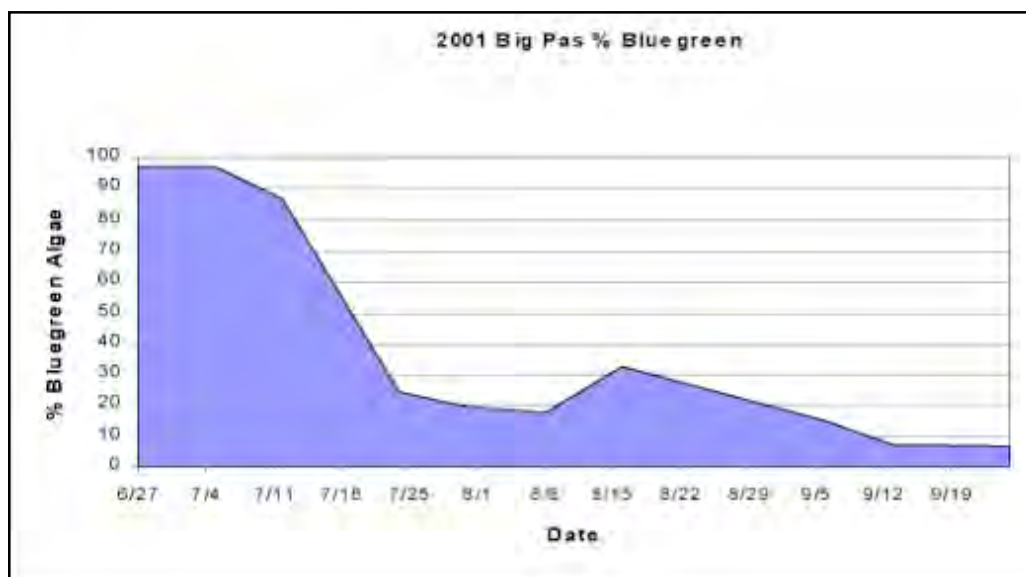


Figure 4. Blue-green algae occurrence in 2-acre PAS prototype in 2001.

**Clemson University.** A detailed computer model capable of predicting PAS water quality has been developed. This model is being used to predict optimal PAS design and management. In 2001, experiments are currently underway to examine the costs and technical feasibility of supplementing the tilapia filter-feeding with physical/chemical techniques for enhanced algal removal. Prophylactic treatments for control of early spring PGD in the winter carryover fish will be continued. The size classes of fish used in the 2001 stocking will allow for the development of a model to predict end of season biomass as a function of stocking size distribution and weight.

**University of Georgia.** Phytoplankton identification will continue for the remainder of the growing season. Stomach contents will be identified for threadfin shad and fathead minnows. Fish harvests will be scheduled until November when ponds will be drained for a complete fish count. We anticipate a follow-up study to compare threadfin shad stocked together

with fathead minnows in catfish ponds with copper sulfate application for control of blue-green algae.

**University of Mississippi.** Laboratory culture, biological evaluation, and chemical studies will be continued. Extracts that have shown activity against blue-green algae in primary assays are currently being evaluated following serial dilution and further prioritized for chemical study. Bioassay-guided fractionation of the "active" extracts will be continued and the structures of new selective algicides will be determined by spectroscopic means. Purified active compounds will be evaluated for toxicity in cell-based toxicological screens. Several anti-algal compounds from marine organisms and plants are now under investigation in our laboratories. In addition to the work with the Peruvian plant *D. candida*, unusual terpenoid lipids (isolated from a marine brown algae) are currently under study. Once the chemical structures have been established a decision can be made regarding compound sourcing for further testing and development.

**Mississippi State University/USDA-ARS.** Additional efficacy testing of analogs of the natural product derivative will continue to determine the lowest effective concentration in catfish aquaculture ponds. The determination of the half-life of optimal natural product derivatives in catfish pond water will be performed to provide information related to environmental safety issues. Other analogs will be screened to determine their effectiveness as a broad-spectrum blue-green algicide. Pond testing (using entire catfish ponds) of the optimal selective com-

pounds (as determined by laboratory tests and efficacy tests using limnocorrals) is planned. Also, additional laboratory screening of other compounds will be performed.

**North Carolina State University.** Additional studies of pond water circulation using gypsum blocks are planned. A comprehensive analysis of the phytoplankton composition and nutrient concentrations of the ponds in the current study will be finished by the end of this year.

## IMPACTS

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Selected plant and algae extracts have shown strong anti-cyanobacterial activity. The findings indicate that natural products (small biologically active organic compounds) produced by organisms that live and compete in cyano-bacteria-rich environments are a valuable source of new cyanobacterium-selective algicides and may be of use in the control of blue-green algae in aquaculture ponds. The occurrence of both noxious and toxic blue-green algal blooms in fish aquaculture is a worldwide problem. The idea that chemicals in trees (and marine algae) may have the ability to control blue-green algal blooms is intriguing, especially in extremely poor regions (such as developing countries) where one could envision harvesting these natural materials for use in aquaculture ponds. A rather unexpected outcome of this research is the discovery of several new substances with herbicidal activity. As a result of natural products screening efforts, extracts from several plants and marine organisms were found to be selectively toxic only to the green alga control (*S. capricornutum*) and to specific types of higher plants. Some of the chemical constituents of these anti-algal/herbicidal extracts may hold promise as new natural product-based means of agricultural weed control.

Results of studies of baitfish ponds have increased our understanding of water quality and pond bottom sediments in this important sector of aquaculture in the southeast. Despite a history of long-term use of granular fertilizer in study ponds, sediment-bound phosphorus did not appear to exert a discernible effect on water quality under conditions of commercial production. Discovery of high soil sulfate sulfur levels in pond bottom samples contributed to affected baitfish farmers changing pond management practices to reduce chances of hydrogen sulfide problems.

Research at Auburn University showed that EDTA is an excellent chelating agent for iron (and presumably other metals) for use in pond aquaculture. Application of potassium chloride, legume hay, and iron and other trace elements to ponds do not appear to be useful for controlling blue-green algae in ponds. Sodium nitrate is a good nitrogen fertilizer for ponds; it has been shown to be environmentally superior to ammonium-based nitrogen fertilizers for use in aquaculture and sportfish ponds. Nitrate is not acid-forming, does not exert an oxygen demand, and its presence in effluents is less objectionable than ammonia. Pond fertilization work also revealed that

nitrogen fertilization was not needed in ponds at Auburn University, for fish production in ponds treated with nitrogen and phosphorus did not exceed that of ponds treated with phosphorus only. Sediment quality decreases as bait minnow ponds age and sodium nitrate treatment will not enhance soil quality in these ponds. Sediment removal seems to be the only feasible way of improving soil quality in old bait minnow ponds.

Research at Mississippi State University showed that nutrient manipulation techniques consisting of simultaneous adjustment of N:P ratios and addition of chelated iron does not affect phytoplankton community structure. Results from mixing studies suggest that some threshold level of turbulent mixing is necessary to overcome light limitation of phytoplankton production and shift phytoplankton community composition from dominance by cyanobacteria. Application of turbulent mixing should attempt to develop a uniform flow field to avoid areas of concentrated turbulence that can suspend pond soils.

Shad stocking is being considered by catfish farmers in Georgia who had off-flavor catfish and who could not utilize herbicides for control of blue-green algae. Shad stocking started in the winter of 2000 and spring of 2001 in Georgia. Shad stocking is limited by the availability of threadfin shad. Information from this study has helped with the fathead minnow/channel catfish stocking program for proliferative gill disease control. Behavior of fathead minnows in channel catfish ponds indicated a need to encourage spawning by adding spawning substrate or to restock the fathead minnows at regular intervals in order to maintain at least 3,600 minnows/ha.

The PAS technique offers the potential to provide a method to quadruple current fish production in a system which eliminates blue-green algal dominance and associated fish off-flavor problems, while recovering wasted nitrogen and phosphorus discharges, which currently pose an eutrophication threat to surface and groundwater supplies. Economic projections suggest that PAS catfish production costs are 15 to 17¢/lb lower than conventional pond production costs.

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## **MANAGEMENT OF AQUACULTURAL EFFLUENTS FROM PONDS**

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### **Reporting Period**

April 1, 1999 to August 31, 2001

<b>Funding Level</b>	Year 1 .....	\$227,603
	Year 2 .....	236,919
	Year 3 .....	150,740
	Total .....	\$615,262

<b>Participants</b>	Mississippi State University (Lead Institution) .....	John Hargreaves, Thomas Cathcart
	Auburn University .....	Claude Boyd
	University of Arkansas at Pine Bluff .....	Carole Engle, Nathan Stone
	Louisiana State University .....	Robert Romaine, Ray McClain
	North Carolina State University .....	Harry Daniels
	Virginia Polytechnic Institute and State University .....	Greg Boardman
	Waddell Mariculture Center .....	Craig Browdy

<b>Administrative Advisor</b>	Dr. Marty Fuller, Assistant Director Mississippi Agricultural and Forestry Experiment Station Mississippi State University Mississippi State, Mississippi 39762
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### **PROJECT OBJECTIVES**

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1. Develop additional information to characterize the components of aquaculture effluents that represent the greatest potential risk of deleterious environmental impact (e.g. suspended solids, total phosphorus).
2. Evaluate the impact of aquaculture pond effluent discharge on receiving stream water quality.
3. Evaluate a range of water management techniques appropriate for ponds as a means of reducing the quantity and improving the quality of discharged water.
4. Develop and evaluate models for predicting risks to the environment and the costs and benefits of implementing Best Management Practices (BMPs).
5. Based upon existing information, supplemented by project findings, develop a

comprehensive set of BMPs that can be implemented to reduce the environmental impacts of pond aquaculture in general. Develop supplemental BMPs particular to the various pond cultured species in the region that will complement the generic, pond-system BMPs. These BMPs would include best culture practices, waste handling and management, and water quality management and reuse.

6. Convene a series of workshops to educate and inform producers and regulators on the characteristics and management of aquaculture effluents from ponds, including BMPs based on the best available information and that minimize environmental impact and satisfy regulatory compliance requirements.

## ANTICIPATED BENEFITS

Results of this project will provide simple management alternatives to reduce the volume and improve the quality of effluents, possibilities for water reuse, and inexpensive treatment methods based on sedimentation. This project will provide beneficial effluent management practices to producers of channel catfish, striped bass, baitfish, crawfish, and marine shrimp. Development of

practical, environmentally sound management practices that minimize the effect of pond effluents on receiving streams will reduce the environmental impact and contribute to the sustainability of the regional aquaculture industry. Information generated by this project can be used by regulators and permit writers to provide effective and coherent regulation of aquaculture effluents.

## PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

**Objective 1.** *Develop additional information to characterize the components of aquaculture effluents that represent the greatest potential risk of deleterious environmental impact (e.g. suspended solids, total phosphorus).*

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**Auburn University.** Studies indicated that about 53% of total suspended solids, total phosphorus, total nitrogen, and biochemical oxygen demand were associated with particles less than 5 micrometers in diameter. Studies suggest that a water retention time in settling basins of 8 hours will improve effluent quality significantly and a settling time of 2 to 4 hours is sufficient to reduce total suspended solids in effluents to 75 to 90% of original concentrations. Solids removal is associated with declines in the mineral fraction with little change in organic solids concentrations in effluents. Application

of aluminum sulfate at 25 to 50 ppm did not improve the efficiency of solids removal in initial trials.

Estimates of runoff from watersheds suggest that settling basins to treat storm runoff from watershed-type catfish ponds require volumes of 30 to 40% of pond volume in order to provide a retention time of 8 hours. Thus, because of the large volume required, settling basins do not appear to be feasible for treating storm runoff. Settling basins for treating intentional discharge from partial drawdown or complete draining

would need to be only 10 to 20% of the volume of the largest pond on the farm because the quality of catfish pond effluents is relatively high except for the final 20 to 25% of water released when ponds are drained completely.

Most existing catfish farms in Alabama extend to property lines or streams, and there seldom is space for installing settling basins. However,

### *Results at a glance...*

☆ *Treatment of the final 20% of water discharged when ponds are drained can be accomplished with sedimentation basins designed with a hydraulic retention time of 8 hours. However, a settling time of 2 to 4 hours is sufficient to reduce total suspended solids in effluents to 75 to 90% of original concentrations.*

settling basins could be considered an essential component in the design of new farms. On farms without settling basins, it is possible to use the pond being harvested as its own settling basin. Water levels should be lowered to 20% to 25% of full volume, drains should be closed and fish harvested by seining. Once fish have been removed, the water should be allowed to stand until most of the suspended solids have settled. This will normally take only 2 or 3 days. The water should then be released slowly to prevent resuspension of solids. It is recommended that the valve only be opened to ¼ its maximum capacity during final draining, the valve should be closed at the beginning of rainfall and not reopened until water has cleared.

**Louisiana State University.** Water budgets were calculated over two production seasons in experimental crawfish ponds. The average

annual water requirement was 7.6 feet. More than 65% of the water outflow in both seasons was from evaporation, evapotranspiration by rice planted as crawfish forage, and seepage, with intentional and unintentional effluent release accounting for the remainder. Precipitation was the major water inflow in these shallow-water ecosystems, contributing nearly 60% of the total under conditions of normal average annual precipitation. Intentional water discharge during summer drawdown, an established management practice, was consistent over both seasons, averaging 0.81 feet. Unintentional discharge was highly correlated with the magnitude of precipitation events. Drought conditions in the first production season resulted in very low unintentional water release (3.2 inches). Unintentional water release in the second production season, representative of normal rainfall patterns in southern Louisiana, was 1.79 feet (24% of annual water budget).

Solids and nutrient chemistry in crawfish effluent samples from six ponds during the summer drawdown was determined at initiation of drawdown, 50% pond volume remaining, and < 10% of pond volume remaining. Additional water samples were collected as pond effluent passed through a highly vegetated and minimally vegetated filter strips.

**Mississippi State University.** In commercial channel catfish ponds dominated by a dense bloom of the blue-green alga, *Oscillatoria agardhii*, about 51% of total suspended solids had a diameter of less than 5 micrometers. Similar results were obtained from fractionation of solids in water collected from ponds dominated by inorganic turbidity (55% of the total suspended solids were less than 5 micrometers). Treatment of water collected from a pond dominated by a dense bloom of *O. agardhii* with alum between 0 and 50 ppm resulted in a solids reduction rate of 1.11 grams of solids per gram of

alum. Size fractionation of pond water following alum treatment indicated that the proportional removal of solids less than 5 micrometers was greatest, although the greatest absolute solids reduction rate occurred in whole (not fractionated) pond water. The proportion of solids in the smallest size fractions increased with alum dose, suggesting that larger solids were selectively settled by alum treatment.

**North Carolina State University.** Fractionation of solids in overflow water from hybrid striped bass ponds shows that more than 95% of total solids and between 53% to 65% of total suspended solids are less than 5 micrometers in diameter. More than 50% of biochemical oxygen demand in pond water is associated with particles less than 5 micrometers. The amount of settleable solids in hybrid striped bass pond water overflow is negligible, suggesting that sedimentation alone, without the use of chemical amendments to coagulate and precipitate solids, would not be an effective treatment of this fraction of pond water. Effluents from the initial and final stages of pond draining will be collected and characterized during the pond harvest scheduled for October, 2001.

**University of Arkansas at Pine Bluff.** A field study characterized suspended solids and 5-day biochemical oxygen demand (BOD<sub>5</sub>) in baitfish pond effluents by serial fractionation, and evaluated exiting drainage ditches as possible solids settling systems. Effluents from a total of ten ponds on five farms were sampled between 9 December 2000 and 3 June 2001. Total suspended solids (TSS) concentration at the point of discharge averaged 36.3 ppm during the draining of the first 10% of pond volume and increased by approximately 70% during the last 10% of the effluent volume. Volatile suspended solids (VSS) were more variable but averaged about 50% of TSS. BOD<sub>5</sub> averaged 9.0 ppm and did not increase significantly in the last 10% of effluent

volume. Screening through a 10-micrometer mesh removed more mineral than organic solids and reductions in TSS, although small, were statistically significant. Volatile suspended solids and BOD<sub>5</sub> generally did not change with reductions in TSS, except small but significant reductions in the first 10% of effluent after screening through a 41-micrometer mesh. Based on averages, 75-82% of suspended particles were less than 5 micrometers, with smaller particles being more organic. The last 10% of effluent was characterized by smaller particles than the first 10% of effluent volume. VSS in the last 10% of effluent had a stronger correlation ( $r = 0.91$ ) with BOD<sub>5</sub> than the first 10% ( $r = 0.71$ ). TSS had a weaker correlation with BOD<sub>5</sub> ( $r = 0.55$  in first 10% of effluent and 0.71 in last 10%).

**Waddell Mariculture Center.** In 2000, effects of sedimentation on shrimp pond harvest effluent were examined in a clay bottom, commercial pond stocked with approximately 20 *Litopenaeus vannamei* per square meter and managed with minimal water exchange. These results were compared to similar 1999 studies conducted on plastic lined ponds overlain with sand sediment, stocked with 100 *L. vannamei* per square meter, and managed with no water exchange. Particulate matter was substantially reduced in the 1999 ponds after 2 hours of sedimentation. In contrast, the 2000 pond had only modest reductions in particulate matter after 20 hours of sedimentation. Buoyant planktonic algae were a primary component of turbidity in 2000, but a relatively minor component in 1999. Reduced relevance of buoyant, free-floating algae is common in late season, intensive shrimp culture systems with no water exchange, in conjunction with increased floc comprised of non-living matter. Thus, the benefits from sedimentation of harvest effluent may increase with stocking density. Overall, these results further suggest that the benefits of sedimentation of harvest effluent may vary considerably depending

upon the physical and management characteristics of the pond. While factors such as sludge pile location in respect to outgoing water flow, pond bottom slope, aerator placement and operation during harvest, and shrimp behavior were noted variables in 1999, the 2000 study suggests that stocking density, water exchange, and associated water quality characteristics may also affect the magnitude of benefits from harvest effluent sedimentation.

Sedimentation was effective in reducing the pollution potential of total suspended solids in pond effluent. The flocculation process of the suspended particles was of variable importance in affecting the sedimentation rate. Moreover, the importance of the flocculation process and the effectiveness of sedimentation was greatest in the pond with the most turbid discharge.

The results of this study suggest that allowing sedimentation of the latter portion of shrimp

pond harvest effluent for only a few hours may significantly reduce total suspended solids, chemical oxygen demand, biochemical oxygen demand, and consequential effects on receiving waters. Thus, the simple and relatively inexpensive treatment of effluent by sedimentation may lead to a very marked reduction in the environmental effects of shrimp farming.

The results of this study suggest the possibility that resuspension of bottom mud may enhance the rate and extent of flocculation and increase the efficiency of sedimentation. An additional factor which may have affected flocculation was the stirring of the suspension, a process that enhances the probability of contact among the particles and increases the probability of efficient collision among the suspended particles. The effluents studied in this research were stirred to obtain a uniform suspension. The potential importance of this effect will also require further study and practical implications will be considered.

**Objective 2.** *Evaluate the impact of aquaculture pond effluent discharge on receiving stream water quality.*

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**Mississippi State University.** Seventeen stream sample locations were selected and geo-referenced within four sub-watersheds with variable development of the landscape to aquaculture ponds. Stream sampling occurred during base flow conditions and during the only significant runoff event that occurred during spring 2000. Despite elevated stream flow during this event, few ponds were observed to be discharging water. No changes in stream water quality could be attributed to aquaculture ponds. In some stream reaches, the magnitude of in-stream variation in water quality over short distances was large.

A study was conducted to evaluate the duration and settling characteristics of initial pond effluent from levee ponds. Settling characteristics of

effluent samples collected at the discharge point were determined. In addition, spatial and temporal variation of water quality in channels receiving the effluent was measured. When draining was initiated, shear forces generated by water moving into the drain pipe caused scouring in a zone around the entrance to the drain pipe. The initial flush of discharged water consisted of pond water and a slurry of sediment that had accumulated over the screen inside the pond. The initial discharge was very high in total suspended solids (43,860 ppm), volatile suspended solids (3,770 ppm), BOD<sub>5</sub> (118 ppm), and nutrients. Within 2.5 minutes of the initial discharge, total suspended solids decreased to 1,790 ppm, volatile suspended solids to 205 ppm, and BOD<sub>5</sub> to 29 ppm. By 30 minutes, the poorly

consolidated sediment from around the drain structure inside the pond had been discharged, and the effluent quality was identical to the bulk pond water for the remainder of the draining period (approximately 5 days). For the effluent, 15.2% of the total solids were discharged with the first 1.7% of the total volume discharged. The initial solids concentration was 900 times higher than the solids concentration in the bulk pond water. In contrast, the  $BOD_5$  of the initial flush of water discharged was only about 9 times higher than the  $BOD_5$  of the bulk pond water and concentrations decreased more rapidly than total suspended solids. Therefore, cumulative discharge of  $BOD_5$  as a function of water volume discharged did not have an initial spike of similar magnitude as for total suspended solids; rather, cumulative discharge of  $BOD_5$  was roughly proportional to the volume of water discharged. These results suggest that the ratio of mineral solids to organic matter of the initial flush of water was relatively higher than that of subsequently discharged water.

The median settling velocity decreased from 0.3 cm/second in the initial effluent to 0.06 cm/second after 30 minutes. Total suspended solids in a ditch 100 m from a catfish pond outfall increased to a maximum 30 minutes following the initiation of pond discharge, but then decreased to concentrations that were less than or equal to initial ditch concentrations. The duration of poor water quality of initial effluent from catfish ponds with internal drains is brief (less than 30 minutes) and discharged solids settle rapidly within receiving ditches.

**University of Arkansas at Pine Bluff.** A field study evaluated exiting drainage ditches for solids settling. Surveyed ditches ranged from 1,050 to 2,300 feet in length and from 12 and 39 feet in width. Study ditches included vegetated and non-vegetated types. Volatile suspended solids (VSS) decreased by 14.1% over the first 100 meters

(110 yards) of ditch. Total suspended solids (TSS) remained unchanged because of increases in fixed suspended solids (FSS). Current velocity in the ditch explained up to 65% of the variability in change in solids. There was a net increase in TSS when average velocity exceeded 2 feet/second. Theoretical settling rates of particles suggests that more VSS were removed in ditches than would have been possible based on particle distribution results obtained from fractionation. Fractionation may have affected the character of organic particles by breaking large particles into fine particles. The study suggests that ditches would be economical, effective solids settling systems if erosion in the ditch is prevented through vegetation, especially at the point of discharge, and ditches are engineered to minimize current velocity.

Additional work was conducted to characterize nutrient concentrations in baitfish pond effluents and receiving streams. Ten baitfish ponds were sampled and characterized from December 2000 through June 2001 in the central Arkansas Delta ecoregion. Effluent samples taken during the first and last 10% of pond drainage volume were analyzed for total nitrogen, total phosphorus, BOD, and TSS as well as general physical characteristics. Pond drainage ditch lengths were sampled to determine overall reduction of TSS, and ditch water quality was sampled and analyzed prior to stream discharge. Upstream samples were taken concurrently with pond and ditch samples and were analyzed for similar nutrient concentration and physical characteristics. There were no significant differences in effluent quality between the first 10% and the last 10% of effluent volume, except the last 10% had significantly higher TSS concentrations than the first 10%. There was no significant difference in nutrient concentrations in effluents sampled at the standpipe and effluents sampled at the end of drainage ditches. There was also no significant difference in standpipe effluent

concentrations versus those found upstream of the discharge point. In general, concentrations of measured parameters in commercial baitfish pond effluents were lower than or similar to those reported for commercial catfish ponds during the same seasons.

Data on nutrient concentration by sediment particle size resulting from serial fractionation of effluent samples are currently being analyzed, as

are changes in sediment and nutrients in receiving ditches during the first and last 10% of discharge. Nutrients and solids composition data were collected in an intermittent receiving stream at regular intervals for a 6-month period. Changes in chemical composition of stream water from the headwaters, past an urban area, downstream of a water treatment facility, and upstream and downstream of a large baitfish farm are being analyzed.

*Objective 3. Evaluate a range of water management techniques appropriate for ponds as means of reducing the quantity and improving the quality of discharged water.*

**Mississippi State University.** A study was designed to evaluate the effect of increasing the water storage capacity (depth) of one pond by 1 foot in an interconnected 2-pond or 4-pond module. Tested pond system configurations include 3 conventional production ponds linked to 1 production/storage pond; 1 conventional production pond linked to 1 production/storage pond; and 1 conventional (control) pond. In January 2000, ponds were filled with groundwater and data collection was initiated. During March 2000, each of the seven ponds were stocked with two sizes of commercial catfish. Market-size fish were harvested from all ponds (7) in fall 2000. Harvest rates were comparable to those of commercial production ponds. Ponds were restocked with additional catfish in spring 2001 and a second harvest will be conducted in fall 2001.

Based on 20 months of operation, effluent volume in the linked-pond systems was reduced by approximately 50 to 75%, and groundwater consumption was reduced by 40 to 50% compared to conventionally managed ponds. The first year of this study was drier than normal and the second year was wetter than normal. Therefore, these results should be representative

of those that may be achieved given the range of climatic conditions in the region.

Water quality (dissolved oxygen, temperature, total ammonia, nitrate, chlorophyll *a*, conductivity, alkalinity, and hardness) was not different

### *Results at a glance...*

☆ *Effluent volume can be reduced by increasing pond depth to increase rainwater storage capacity and linking the combined storage/production pond to one or three adjacent conventional ponds. After twenty months, effluent volume was reduced by approximately 50 to 75% and groundwater consumption was reduced by 40 to 50% compared to conventionally managed ponds.*

between ponds of the three configuration types. Disease (proliferative gill disease, enteric septicemia of catfish) epizootics have occurred in all ponds, but the spread of the disease could not be attributed to connections between ponds. The data indicate that the hydrological model used

as the basis for this study will require only minor modifications to be useful for analysis of various hydrological management strategies.

**North Carolina State University.** Data on water discharge quantity and quality is being collected from an 80-acre commercial hybrid striped bass farm. During selected pond drawdown events, water samples are collected from the farm drainage ditch and at 3 sites downstream and solids and nutrient concentrations are measured. Data on rainfall, runoff, and water use have been collected since fall of 2000, and a water budget for this farm is currently under development.

A study comparing water quality in annually drained hybrid striped bass ponds compared to ponds managed for zero discharge is in its third year. Twelve, 0.25-acre ponds are being managed according to common commercial practices for foodfish production. Water quality and fish production are not significantly different between the two treatments.

Two fixed-film filters (vertical brushes and a block honeycomb medium) are being evaluated for suspended solids removal from hybrid striped bass pond water. Vertical brush filters remove approximately 20% of solids on a single pass. Solids removal by the honeycomb filter is about 20-50% less than that of the vertical brushes. Solids removed by both filters and controls are approximately 15% organic material. During 2001 several trials are being conducted to evaluate the solids removal efficiency of the two filters over different hydraulic retention times.

**University of Arkansas at Pine Bluff.** As a water conservation technique in response to declining aquifer levels, re-use of pond water is growing popular in the Arkansas baitfish industry. Predation of fry by cyclopoid copepods present in re-used water is the greatest challenge to widespread adoption of this

practice. A study was conducted to evaluate treatments affecting zooplankton populations so that water can be re-used and the volume of effluent reduced. The aim of this study was to evaluate methods of restarting the zooplankton bloom in pond water held from previous production operations. The abundance and evolution of rotifer and copepod populations in ponds containing old water, old water treated with 0.25 ppm Dylox, and mechanically filtered old water were compared to ponds filled with ground water. Zooplankton were sampled and water quality was monitored daily for 6 weeks. Rotifer abundance increased in ponds in all treatments during the first 8 days. Average rotifer density over 8 days did not differ between treatments. However, average copepod abundance was affected by treatments. New water had significantly fewer copepods than Dylox-treated or old water, but did not have fewer copepods than mechanically filtered water. Mechanical filtration compared more favorably to ponds filled with ground water than to ponds treated with Dylox or not treated. Filtration minimized adult copepods, while maintaining sufficient rotifer density for baitfish culture. Mechanically filtered old water has good potential of providing sufficient food (rotifers and nauplii) for newly stocked fry while minimizing the risk of copepod predation on fry.

Cyclopoid copepod predation has been established as an important factor causing low and variable survival rates during sunshine bass fingerling production. A concentration of 500 copepods/L can result in 0% survival of 5-day-old fry stocked at 20 fry/L during a 24-h period. Concentrations of 50 copepods/L and below resulted in survival rates not significantly different from controls with no copepods. Farmers will encounter cyclopoid copepod concentrations between 50 and 500 copepods/L in pond water fertilized to enhance rotifer blooms or in water held from previous operations. This study investigated the effect of



cyclopoid copepod concentration on survival rates of sunshine bass, golden shiner, fathead minnow, and goldfish fry. Survival of golden shiner, fathead minnow, and goldfish was 95-100% at all copepod concentrations (0, 100, 200, 300, 400, and 500/L). Survival rates of sunshine bass (75.0-93.1%) were not significantly different among copepod concentrations ranging from 0 to 300/L. Sunshine bass fry survival at 400 copepods/L (62.5%) was significantly higher than at 500 copepods/L (39.4%), and significantly lower than at 0 and 100 copepods/L. On the

basis of these results, stocking sunshine bass fry into ponds with fewer than 300 cyclopoid copepods/L probably represents a low risk of predation. Concentrations of cyclopoids between 300 and 500/L represent a higher risk of predation. Concentrations of cyclopoids exceeding 500/L will probably result in extremely low survival and poor yields. Five-day-old golden shiner, fathead minnow, and goldfish stocked in old pond water with 500 cyclopoid copepods/L or less could have acceptable survival rates.

**Objective 4.** *Develop and evaluate models for predicting risks to the environment and the costs and benefits of implementing Best Management Practices (BMPs).*

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**University of Arkansas at Pine Bluff.** Partial enterprise budgets are being developed for the various effluent management strategies evaluated in this project. Budget analyses were completed for sedimentation basin management options for commercial catfish ponds. In all, 108 different scenarios were analyzed for sedimentation basins on catfish farms. Budgeting work on the water-storage/production pond strategies for catfish ponds was also completed. Seventy-two different scenarios were analyzed for production/storage ponds. Preliminary cost data have been collected on the fixed-film filter options under study in this project.

**Sedimentation Basins.** Costs associated with settling basins are dependent on the size and number of basins, and whether sufficient land is available for basin construction or if existing production ponds must be retrofitted and taken out of production. Sizing of settling basins is controlled by factors such as the type of effluent to be treated (draining or storm overflow), layout of ponds, size of the largest foodfish pond, the number of drainage canals, and the scope of regulations governing the release of aquacultural

effluents. The number of settling basins is affected by the hydraulic residence time (HRT) which is calculated from Stoke's Law. The HRT, in turn, is affected by the size of particles in suspension.

The number of settling basins is also affected by the number of drainage outlets on a farm. Some farms may drain in four to five different directions. Furthermore, farms that have ponds that are not contiguous would need a greater number of basins. Three farm size scenarios were considered in an analysis of settling basin costs: a 160-acre farm with approximately 140 acres of water, a 320-acre farm with 280 acres of water and a 640-acre farm with 560 acres of water. Average sizes of foodfish ponds in this analysis were assumed to be 10 and 15 acres, while fingerling ponds were 5 acres each.

Larger farm sizes will result in higher and more variable costs. Investment costs included excavation of settling basins and the installation of stationary re-lift pumps to drain effluents from excavated basins. Annual operating costs consisted of copper sulfate applications (to promote

sedimentation of phytoplankton cells), the annual cost of pumping, and levee mowing and maintenance, whereas annual fixed costs refer to depreciation of basins and pumps, interest on investment, and the opportunity costs associated with land taken out of production for the settling basin. Estimates of lost revenue due to production foregone from retrofitted production ponds were \$300, \$346, and \$480/acre for the 160-, 320-, and 640-acre farms, respectively. Larger farm sizes will result in higher and more variable costs. Investment costs included excavation of settling basins and the installation of stationary re-lift pumps to drain effluents from excavated basins.

Large investments are needed for the construction of settling basins. This investment cost depends heavily on the drainage layout of the farm and the scope of regulations governing the release of effluents. For instance, investment costs in a 160-acre catfish farm may range from \$28,648 to over \$375,000. For the 640-acre farm, investment costs under certain scenarios exceeded \$750,000.

Utilization of existing foodfish ponds to settling basins represents a more economical approach for the treatment of harvest/draining effluents than construction of settling basins. This is particularly true for those scenarios in which all effluent volume must be treated. This difference was a consequence of the extremely high

cost associated with excavating a sedimentation basin sufficiently deep to collect all farm effluents by gravity flow. Finally, compliance costs for the treatment of overflow effluents were moderate to high and strongly influenced by farm size.

**Production/Storage Ponds.** Two configurations (1:1 and 1:3), based on the number of production ponds served by each production/storage pond, were assumed. Increased depth of the combined production-and-storage pond increases the storage capacity of the system but incurs higher earthmoving costs. Three additional depths were considered: 12, 24, and 36 cm and seepage values of 0.0 and 1.0 mm/d were assumed. Cost estimates were developed for farms with average foodfish pond sizes of 10 and 15 acres. Fingerling ponds were not linked.

In total, 24 scenarios were defined for each farm size for a total of 72 scenarios. Depth of storage pond and pond configuration were the two most important factors affecting implementation costs of this technology. For instance, estimated total investment costs for a 160-acre farm with 10-acre foodfish ponds, 1:1 configuration, and 0-mm/d infiltration rate, ranged from \$76,123 to \$215,088 as the additional depth of storage ponds was increased from 12 to 36 cm. However, if the configuration is 3:1, investment costs decreased (ranging from \$44,782 to \$115,947).

**Objective 5.** *Based upon existing information, supplemented by project findings, develop a comprehensive set of BMPs that can be implemented to reduce the environmental impacts of pond aquaculture in general. Develop supplemental BMPs particular to the various pond cultured species in the region that will complement the generic, pond-system BMPs. These BMPs would include best culture practices, waste handling and management, and water quality management and reuse.*

**Auburn University.** An environmental audit form for assessing the status of environmental

management on catfish farms is under development. This instrument will be used to identify

potential problems that can possibly be solved with BMPs. The audit form will be used by project participants to conduct environmental audits of aquaculture production facilities. A document containing best management practices to reduce the volume and improve the quality of channel catfish farm effluents has been prepared. A cooperative effort has been established between Auburn University, Alabama Catfish Producers Association, Natural Resources Conservation Service (NRCS), and the Alabama Department of Environmental Management to develop a set of best management practices (BMPs) for Alabama catfish farming. These BMPs will be reviewed by all agencies involved and they will be presented to the farmers for comment. Moreover, farmer meetings will be held to assure that farmers are aware of the BMPs and have opportunity for input. Environmental groups also will be asked to comment on the BMPs. When formalized, the BMPs will be maintained by NRCS in the form of Guide Sheets and made available to farmers. Draft guide sheets have been developed for:

- Reducing Storm Runoff into Ponds

*Objective 6. Convene a series of workshops to educate and inform producers and regulators on the characteristics and management of aquaculture effluents from ponds, including BMPs, based on the best available information and that minimize environmental impact and satisfy regulatory compliance requirements.*

The first workshop convened on 6-7 November 2000 in Roanoke, VA. The objectives of the workshop were to (1) develop a prioritized list of practices that will minimize environmental impacts of aquaculture and be economically acceptable to producers; and (2) familiarize state regulators and consultants with the aquaculture effluents issue and provide the information necessary to develop effective and reasonable regulations.

- Managing Ponds to Reduce Effluent Volume
- Erosion Control on Watersheds and Embankments
- Pond Management to Minimize Erosion
- Control of Erosion by Effluents
- Settling Basins and Wetlands
- Feed Management
- Fertilization of Catfish Ponds
- Water Quality Protection to Improve Effluents
- Water Quality Enhancers
- Therapeutic Agents
- Fish Carcasses

**Louisiana State University.** The NRCS Code of Production was reviewed to assess their applicability to development of a set of BMP effluent and watershed management guidelines for the crawfish aquaculture industry. A draft set of BMPs completed and reviewed by various commodity, state, and federal agencies. The revised document — “Aquaculture Production: Best Management Practices” — was completed and is currently in press. The document includes individual reviews of BMPs for crawfish, catfish, recirculating systems, and cage culture systems.

In Arkansas, information on effluents and BMPs for pond aquaculture has been extended to producers. Two Extension newsletter articles were published. Results of the water re-use zooplankton study were presented to baitfish producers at the UAPB Aquaculture Field Day (attendance was approximately 300 people). A poster presentation on effluents and BMPs was exhibited at the Aquaculture Field Day and a presentation was made on the same topic at the

annual convention of catfish and baitfish producers (150 people). Extension faculty assisted the Arkansas Bait and Ornamental Fish Growers Association in adapting proposed BMPs for catfish production to baitfish, and in developing the association's BMP document.

Most scientists in this SRAC project are active participants in activities coordinated by the Joint Subcommittee on Aquaculture's Aquaculture Effluent Task Force, including the various subgroups representing the species and areas of specialization of project scientists.

## **WORK PLANNED**

The original work plan will be followed. The effort to formulate BMPs will continue.

## **IMPACTS**

The technical guidelines for several straightforward options for effluent treatment or volume reduction have been evaluated and are now available for consideration and implementation by producers of fish in ponds. Project personnel have been involved with the development and dissemination of information on best management practices to fish producers and to federal regulatory authorities considering regulation of aquaculture pond effluents. The comprehensive cost estimates and the best management practices developed in this study could potentially provide valuable information for both the Environmental Protection Agency (EPA) and the aquaculture industry as EPA proceeds with their rule-making effort to develop Effluent Limitation Guidelines.

Generalized results of the comprehensive cost estimates related to sedimentation basins have been discussed by members of the Economics Subgroup of the JSA Aquaculture Effluents Task Force with officials of the Environmental Protection Agency and the aquaculture industry. It is likely that the results of these studies will play a role in decisions made as EPA proceeds with their rule-making effort to develop Effluent Limitation Guidelines.

The Arkansas Bait and Ornamental Fish Growers Association used SRAC information in developing a set of Best Management Practices for bait and ornamental fish farms. Association members have committed to following these recommended practices in order to minimize any possible environmental impacts from their farms.

## **PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED**

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## Papers Presented

- Bodary, M., and N. Stone. 2001. Characterization of baitfish pond effluents and receiving stream water quality in central Arkansas. *American Fisheries Society Annual Meeting*, Phoenix, AZ, 12 - 16 August.

- Boyd, C. E. 2001. Aquaculture and water pollution. American Water Resources Association, Summer Specialty Conference, Snowbird, Utah.
- Boyd, C. E. 2001. Best waste management practices for the shrimp and catfish industries. Aquaculture Waste Management Symposium, Virginia Polytechnic Institute and State University, Roanoke, Virginia.
- Boyd, C. E. 2001. Role of BMPs in environmental management of aquaculture. Aquaculture 2001, World Aquaculture Society, Orlando, Florida.
- Cathcart, T. P., D. W. Rutherford, and J. A. Hargreaves. 2000. Field study for evaluating effluent and groundwater use reduction in catfish ponds. Water Resources Research Institute Conference Proceedings, Jackson, Mississippi, April.
- Engle, C.R. 2001. Efficient management of catfish farms and best management practices. Presentation and abstract. North American Association of Fisheries Economists, New Orleans, Louisiana.
- Frimpong E. A., and S. E. Lochmann. 2000. An evaluation of treatments affecting zooplankton populations for water re-use and effluent reduction. Aquaculture Field Day, University of Arkansas at Pine Bluff.
- Frimpong E. A., and S. E. Lochmann. 2001. Managing the impact of pond aquaculture on water resources of Arkansas, United States. Second Annual Students' Conference on Conservation Science, University of Cambridge, UK, 27- 30 March.
- Frimpong E. A., and S. E. Lochmann. 2001. An evaluation of treatments affecting zooplankton populations for water re-use and effluent reduction. UAPB Student-Faculty Research Forum, Pine Bluff, AR, 14 - 15 March.
- Frimpong E. A., and S. E. Lochmann. 2001. An evaluation of treatments affecting zooplankton populations for water re-use and effluent reduction. Joint annual meeting of the American Fisheries Society and the Wildlife Society, Arkansas Chapter, Heber Springs, AR, 6 - 8 February.
- Frimpong E. A., and S. E. Lochmann. 2001. An evaluation of treatments affecting zooplankton populations for water re-use and effluent reduction. Aquaculture 2001, World Aquaculture Society, Orlando, Florida.
- Hargreaves, J. A.. 2001. Research needs to evaluate best management practices for pond aquaculture. Aquaculture 2001, World Aquaculture Society, Orlando, Florida.
- Lutz, G. 2001. Best waste management practices for the alligator, crawfish, and turtle industries. Aquaculture Waste Management Symposium, Roanoke, VA, July 22-24, 2001.
- Stone, N. 2001. Best management practices for aquaculture. Arkansas Aquaculture 2001, Hot Springs, AR, 9 February.
- Stone, N., H. Thomforde and S. Lochmann. 2000. Best management practices and pond effluents. (Poster presentation). Aquaculture Field Day, University of Arkansas at Pine Bluff.
- Valderrama, D., and C.R. Engle. 2001. Estimating settling basin size for treating effluents from aquaculture. Presentation and abstract. North American Association of Fisheries Economists, New Orleans, Louisiana.
- Valderrama, D., and C.R. Engle. 2001. Estimating settling basin size for treating effluents from aquaculture. Presentation and abstract. Annual Meeting Arkansas Chapter AFS, Heber Springs, Arkansas.
- Valderrama, D., and C.R. Engle. 2001. Preliminary analysis of costs associated with settling basins and production/storage ponds to reduce effluents discharged from ponds. Presentation and abstract. UAPB Student/Faculty Research Forum 2001, UAPB, Pine Bluff, Arkansas.



## DEVELOPMENT OF IMPROVED HARVESTING, GRADING AND TRANSPORT TECHNOLOGY FOR FINFISH AQUACULTURE

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### Reporting Period

January 1, 2001 - August 31, 2001

<b>Funding Level</b>	Year 1 .....	\$287,053
	Year 2 (Projected) .....	\$272,391
	Year 3 (Projected) .....	\$190,556
	Total .....	\$750,000
<b>Participants</b>	Mississippi State University (Lead Institution) .....	Edwin H. Robinson, Jason E. Yarbrough, Terry R. Hanson
	University of Tennessee .....	Richard J. Strange
	North Carolina State University .....	Harry V. Daniels, Thomas Losordo
	University of Memphis .....	Bill A. Simco, Ken Davis (now at Harry Stuttgart National Aquaculture Research Center)
	University of Florida .....	Craig A. Watson, Roy P.E. Yanong
	University of Arkansas at Pine Bluff .....	David Heikes, Carole R. Engle, Hugh W. Thomforde
<b>Administrative Advisor</b>	Dr. David Morrison, Assistant Director Louisiana Agricultural Experiment Station Baton Rouge, LA	

### PROJECT OBJECTIVES

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1. Develop and evaluate new gear and methods or modify existing methods to improve harvest (seining and trapping) efficiency and fish grading selectivity and reduce stress during these activities.
2. Evaluate methods relative to loading and transport of fish to reduce fish mortalities and the negative effects of stress on product quality.
3. Conduct comparative analyses of new technology and current technology for harvesting, grading, and loading fish.

## ANTICIPATED BENEFITS

The primary benefit of this project will be to significantly improve profitability of the finfish aquaculture industry by improving harvesting

efficiency, grading selectivity, and methods for loading and hauling fish, and by reducing the stress associated with these practices.

## PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

**Objective 1.** *Develop and evaluate new gear and methods or modify existing methods to improve harvest (seining and trapping) efficiency and fish grading selectivity and reduce stress during these activities.*

### Channel Catfish

**Mississippi State University, NWAC.** Initial steps in determining mesh size to acquire the desired degree of grading of channel catfish took place in concrete raceways at the National Warmwater Aquaculture Center, Stoneville, Mississippi and Mississippi State University, Starkville, Mississippi. Three types of mesh [Conventional Twisted Polyethylene (CTPE),

fish with its size class. Each mesh sample was placed into a frame constructed to fit the raceway. The frame was then used to crowd fish to one end of the raceway. After fifteen minutes, a solid divider was placed into the raceway to stop the grading process. Fish remaining in the crowded area and those caught in the mesh sample were counted by size class. From these studies, a recommendation was made on the size mesh that was required to hold fish of a particular size using BPE (Table 1). The mesh size that was used to evaluate KPE was inadequate for grading food size fish. The decision to not conduct further evaluations using larger size KPE mesh was based on the high cost associated with manufacturing full sized grading socks.

### *Results at a glance...*

☆ *Studies in Mississippi indicate that braided polyethylene mesh is an excellent choice for constructing seines and socks for harvest of catfish. In addition, mesh sizes necessary to retain specific size fish were determined using the braided mesh.*

Knotless Polyethylene (KPE), and Braided Polyethylene (BPE)] of varying size (KPE: 38 and 39.5mm; CTPE: 1-5/8 inch, 1-3/4 inch, and 1-7/8 inch; BPE: 1-9/16 inch, 1-5/8 inch, 1-11/16 inch, 1-3/4 inch) were evaluated. Concrete raceways were stocked with channel catfish of five size classes. Fish of each size class were injected with dye in a specific location to identify that

Table 1. Recommended BPE Mesh size for Grading Food Sized Channel Catfish in Warm Temperatures.

Mesh Size (BK)	Desired Size Fish (lbs)
1-9/16"	¾ +
1-5/8"	1 +
1-11/16"	1 ¼ +
1-3/4"	1 ½ +



A prototype seine was used during the harvest of the USDA 103 strain of catfish at the National Warmwater Aquaculture Center (NWAC). Ten ponds (4 or 10 acre) were harvested with an average efficiency of catch of 93.5% in a single seine haul. Further work on seining efficiency is underway.

Full-sized commercial socks were built of BPE having a mesh size of 1-9/16 inch, 1-5/8 inch, and 1-11/16 inch. These socks were used to harvest commercial catfish ponds. Samples were taken immediately upon attachment of the sock, every two hours thereafter, and at load-out. The average population distribution for five size classes of fish is shown in Table 2. Based on this year's work, the recommendations in Table 1 appear to be valid for the BPE mesh. We will continue with data collection in the upcoming year to verify our recommendations.

In addition, commercial seining crews in Mississippi and North Carolina have purchased a prototype seine and socks constructed using BPE. We are working closely with these crews to instruct them in the use of the equipment and also to collect data on seining efficiency and grading.

BPE Mesh Size	Size Classes (lbs)				
	< 3/4	3/4-1	1-1 1/4	1 1/4-1 1/2	1 1/2 +
1-9/16"	2	15	25	15	43
1-5/8"	1	4	19	24	53
1-11/16"	0	0	3	6	91

**University of Arkansas at Pine Bluff.** Two projects are currently underway to develop

in-pond fish grading technology for both market-size channel catfish and channel catfish fingerlings. A horizontal floating platform grader with adjustable spacing has been designed and fabricated. An eductor-style fish pump mechanism has also been designed and fabricated.

### *Results at a glance...*

☆ *A horizontal floating platform grader with adjustable spacing can be effectively integrated into current harvest procedures to grade catfish in ponds.*

Preliminary field trials indicate that the prototype fish grading system can be effectively integrated into current harvesting procedures and has led to several design modifications and improvements. At present, the design modifications have been completed and field trials are being planned and scheduled.

### **Striped Bass**

**North Carolina State University.** An in-pond portable grader has been designed and constructed for use on hybrid striped bass farms. The grader was modified from the original design that was developed for channel catfish. Two on-farm demonstrations/trials have been conducted on commercial hybrid striped bass farms. Based on these trials, the transfer box has been modified to improve the passage of fish from the holding net onto the grader panels.

**University of Tennessee.** Progress toward evaluation of stress in striped bass during grading includes the establishment and characterization of the cortisol, glucose and chloride assays that will be employed and the training of the graduate student responsible for the work.

## Ornamental Fish

**University of Florida.** A survey has been developed and distributed to all the tropical fish farms in the state of Florida seeking information on existing technologies and practices, and suggestions for new technologies and/or

practices they would recommend investigating. A full-time biologist has been hired to assist in collection of the data, and evaluation of existing technologies and practices, including site visits and phone calls to collect survey results from those who do not return the completed survey.

**Objective 2.** *Evaluate methods relative to loading and transport of fish to reduce fish mortalities and the negative effects of stress on product quality.*

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## Baitfish

**University of Arkansas at Pine Bluff.** A graduate student has been hired to conduct work on the survival and profitability of varying loading rates and various commercial water quality stabilizers on closed system long-hauls. The student is currently being trained in the methods necessary to conduct the research.

facilities at The University of Memphis are under construction for use in this project. Systems are now available to evaluate stress responses of fish held in the laboratory under conditions that reflect some of the handling influences imposed under commercial pond situations. These systems will enable the manipulation of water conditions, such as temperature and hardness, to determine their influences on the strength and temporal patterns of physiological responses by fish subjected to handling stress.

**The University of Memphis.** New aquaculture

**Objective 3.** *Conduct comparative analyses of new technology and current technology for harvesting, grading, and loading fish.*

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## Channel Catfish

**University of Arkansas at Pine Bluff --** *Economic evaluation of grader technology.* Commercial farm data collection procedures have been developed based on discussions with commercial operations as to the most efficient ways to collect data on harvests of sub-marketable catfish from ponds. A computer template has been developed to generate enterprise budgets, and cost/kg attributable to the harvest of submarketable fish, for each farm scenario. These estimates, when complete and reviewed carefully, will provide a basis of production costs given current grading technology.

The analysis of the cost of constructing or purchasing the new grader has been initiated, but is not yet complete. Additional tests conducted this summer with the grader provided data to estimate the size distributions of fish and the mean weights harvested. Analysis is proceeding with this information to determine the cost/kg of fish loaded after grading with the new technology.

## Baitfish

**University of Arkansas at Pine Bluff --** *Economic evaluation of new transport technology.* Existing enterprise budgets for baitfish are over

five years old. These budgets have been updated to reflect current costs of production. Data acquisition forms have been developed to evaluate the current costs of transporting baitfish. These forms were developed with input from industry cooperators and are currently on

review by others. These will be used to develop a database of cost information related to fish transportation. This database will serve as the basis of comparison for comparing and evaluating the new technologies to be developed.

## WORK PLANNED

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### Year 2

**Objective 1.** *Develop and evaluate new gear and methods or modify existing methods to improve harvest (seining and trapping) efficiency and fish grading selectivity and reduce stress during these activities.*

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#### Channel Catfish

**Mississippi State University, NWAC.** Work will continue on evaluating prototype seines on commercial catfish farms as well as by controlled studies in NWAC ponds. Trials will be conducted under warm and cool water temperatures to account for seasonal variation in fish behavior. Grading socks will continue to be constructed from various materials and evaluated in research and commercial ponds.

**University of Arkansas at Pine Bluff.** Grader performance data will be collected beginning late summer/early fall 2001 and will continue through the following year to ensure performance data is collected throughout the entire temperature range.

**University of Memphis.** Stress responses by fish will be evaluated in association with research being conducted by Mississippi State University. MSU researchers will harvest fish from commercial ponds utilizing seines with different mesh sizes and materials. Blood samples will be taken at appropriate intervals during the harvesting and hauling of fish. The blood will subsequently be evaluated for appropriate physiological stress indicators. This information will be helpful to determine which methods/materials enable

handling of fish with the least adverse effects.

#### Striped Bass

**University of North Carolina.** Modifications to the transfer box will continue throughout the project period. Additional trials will be done to determine the optimum spacer setting on the grader panel for different sized fish. Trials will be conducted during all four seasons to determine the effect of water temperature on grading time, grading efficiency and associated mortality. Additional on-farm demonstrations are planned to introduce this technology to the industry and for training of harvest crews.

**University of Tennessee.** Work planned includes assaying striped bass blood after different harvesting and grading procedures in order to evaluate relative stress of the different methods.

#### Ornamental Fish

**University of Florida.** Working at the Tropical Aquaculture Laboratory and with a select group of farm collaborators, we will be collecting data on existing practices and technologies, emphasizing analysis of cost of materials, labor, and impact of practices on fish health and condition.

**Objective 2.** *Evaluate methods relative to loading and transport of fish to reduce fish mortalities and the negative effects of stress on product quality.*

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#### **Baitfish**

**University of Arkansas at Pine Bluff.** Work will begin on the evaluation of varying loading rates and using various commercial water quality stabilizers on closed system long-hauls on survival and profitability of baitfish.

**University of Memphis.** The effects of stress on loading rates and hauling of baitfish will be evaluated. Blood samples will be taken at appropriate intervals during hauling and subsequently analyzed for appropriate physiological stress indicators.

**Objective 3.** *Conduct comparative analyses of new technology and current technology for harvesting, grading, and loading fish.*

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#### **Channel Catfish**

**Mississippi State University, NWAC.** Benefits and costs for the harvest technologies developed for catfish will be estimated at the farm, processor, and consumer levels using various economic models. Economics of new technologies will be compared to current technologies.

**University of Arkansas at Pine Bluff --** *Economic evaluation of grader technology.* The costs and benefits of the new grading technology will be compared to the current technology in a partial budget framework to determine the resulting net benefits. The final phase of the project will be to incorporate the effects of the new grading technology into a

recently developed multi-year, dynamic whole-farm mathematical programming model for catfish production to examine farm-level effects of the new grading technology.

#### **Baitfish**

**University of Arkansas at Pine Bluff.** Economic evaluation of new transport technologies will be collected and summarized. Data on mortalities associated with the new technologies will also be collected. The additional costs and benefits associated with the new transport technologies will be compared to existing technologies within a partial budget framework. This analysis will then be extended to evaluate the effect on farm profits of adoption of the new technologies.

## **IMPACTS**

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#### **Channel Catfish**

Braided polyethylene (BPE) mesh is recommended for construction of catfish seines and socks based on performance characteristics determined in research and commercial catfish ponds. Mesh sizes that hold a particular size

class of catfish were determined. These data provide information that can be used to improve seining efficiency and grading selectivity.

The adjustable, in-pond fish grader allows for more control over the size of fish retained which should lead to more harvesting flexibility and

more marketing options for producers. The grader also reduces the time and labor required as compared to box graders that were used.

Improved grading technologies have the potential to improve farm profits theoretically by as much as 5-10%.

#### **Striped Bass**

In-pond graders have the potential to significantly reduce the labor and costs associated with

harvesting and minimize mortality caused by excessive handling.

#### **Ornamental Fish**

Initial surveys and analysis of existing practices will provide producers with accurate information of the economic cost and impact of their existing practices. This data can be used as a management tool and as a basis for development of new, improved management practices.

### **PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED**

None.

## SUPPORT OF CURRENT PROJECTS

Title	Yr	SRAC Funding	Other Support				Total Other Support	Total SRAC+ Other Support
			University	Industry	Other Federal	Other		
Publications, Videos and Computer Software	1	50,000	43,950	-0-	-0-	-0-	43,950	93,950
	2	60,948	30,737	-0-	-0-	-0-	30,737	91,685
	3	45,900	35,710	-0-	1,000	-0-	36,710	82,610
	4	60,500	41,000	-0-	-0-	-0-	41,000	101,500
	5	67,000	47,000	-0-	-0-	-0-	47,000	114,000
	6	80,550	52,975	-0-	-0-	-0-	52,975	133,525
Total		364,898	251,372	-0-	1,000	-0-	252,372	624,270
Management of Environmentally-Derived Off-flavors in Warmwater Fish Ponds	1	250,827	69,389	42,000	49,500	-0-	160,889	411,716
	2	250,142	69,389	53,000	28,380	20,000	170,769	420,911
	3	229,266	38,329	34,000	58,483	-0-	130,812	360,078
	4	80,900	25,829	26,000	36,000	-0-	87,829	168,729
	5	56,100	11,542	-0-	-0-	-0-	11,542	67,642
Total		867,235	214,478	155,000	172,363	20,000	561,841	1,429,076
Verification of Recommended Management Practices for Major Aquatic Species	1	31,410	60,286	1,000	-0-	-0-	61,286	92,696
	2	66,114	78,686	1,000	-0-	-0-	79,686	145,800
	3	66,925	78,986	6,000	-0-	-0-	84,986	151,911
Total		164,449	217,958	8,000	-0-	-0-	225,958	390,407
Control of Blue-green Algae in Aquaculture Ponds	1	307,574	171,746	27,000	172,500	-0-	371,246	678,820
	2	280,639	161,882	35,000	98,380	-0-	295,262	575,901
	3	253,326	149,662	16,000	120,983	-0-	286,645	539,971
Total		841,539	483,290	78,000	391,863	-0-	953,153	1,794,692
Management of Aquacultural Effluents from Ponds	1	227,603	105,319	-0-	-0-	-0-	105,319	332,922
	2	236,919	117,051	-0-	-0-	-0-	117,051	353,970
	3	150,740	109,516	-0-	-0-	-0-	109,516	260,256
Total		615,262	331,886	-0-	-0-	-0-	331,886	947,148
Development of Improved Harvesting, Grading and Transport Technology for Finfish Aquaculture	1	287,053	218,353	-0-	-0-	-0-	218,353	505,406
	2	272,391	227,188	-0-	-0-	-0-	227,188	499,579
	3	190,556	232,823	-0-	-0-	-0-	232,823	423,379
Total		750,000	678,364	-0-	-0-	-0-	678,364	1,378,364

## SRAC RESEARCH AND EXTENSION PROJECTS

Project	Duration	Funding	Grant No.
*Analysis of Regional and National Markets for Aquacultural Products Produced for Food in the Southern Region. Dr. J. G. Dillard, Mississippi State University, Principal Investigator	04/01/88-06/30/90 <b>Project Total</b>	<b>\$346,038</b>	87-CRSR-2-3218
*Preparation of Southern Regional Aquaculture Publications. Dr. J. T. Davis, Texas A&M University, Principal Investigator	01/01/88-12/31/90 <b>Project Total</b>	<b>\$150,000</b>	87-CRSR-2-3218
*Performance of Aeration Systems for Channel Catfish, Crawfish, and Rainbow Trout Production. Dr. C. E. Boyd, Auburn University, Principal Investigator	03/01/88-10/31/90 <b>Project Total</b>	<b>\$124,990</b>	87-CRSR-2-3218
*Develop a Statistical Data Collection System for Farm-Raised Catfish and Other Aquaculture Products in the Southern Region. Dr. J. E. Waldrop, Mississippi State University, Principal Investigator	06/01/89-11/30/90 <b>Project Total</b>	<b>\$13,771</b>	88-38500-4028
*Immunization of Channel Catfish. Dr. J. A. Plumb, Auburn University, Principal Investigator	Yr. 1-05/02/89-04/30/90 Yr. 2-05/01/90-04/30/91 <b>Project Total</b>	\$50,000 <u>49,789</u> <b>\$99,789</b>	88-38500-4028 89-38500-4516
*Enhancement of the Immune Response to <i>Edwardsiella ictaluri</i> in Channel Catfish. Dr. J. R. Tomasso, Clemson University, Principal Investigator	Yr. 1-05/02/89-04/30/90 Yr. 2-05/01/90-10/31/91 <b>Project Total</b>	\$46,559 <u>51,804</u> <b>\$98,363</b>	88-38500-4028 89-38500-4516
*Effect of Nutrition on Body Composition and Subsequent Storage Quality of Farm-Raised Channel Catfish. Dr. R. T. Lovell, Auburn University, Principal Investigator	Yr. 1-05/02/89-04/30/90 Yr. 2-05/01/90-04/30/91 Yr. 3-05/01/91-12/31/92 <b>Project Total</b>	\$274,651 274,720 <u>273,472</u> <b>\$822,843</b>	88-38500-4028 89-38500-4516 90-38500-5099
*Project Completed			

Project	Duration	Funding	Grant No.
*Harvesting, Loading and Grading Systems for Cultured Freshwater Finfishes and Crustaceans. Dr. R. P. Romaine, Louisiana State University, Principal Investigator	Yr. 1-05/02/89-04/30/90	\$124,201	88-38500-4028
	Yr. 2-05/01/90-04/30/91	124,976	89-38500-4516
	Yr. 3-05/01/91-04/30/93	<u>124,775</u>	90-38500-5099
	<b>Project Total</b>	<b>\$373,952</b>	
*Preparation of Extension Publications on Avian Predator Control in Aquaculture Facilities. Dr. James T. Davis, Texas A&M University, Principal Investigator	05/01/90-12/31/92		
	<b>Project Total</b>	<b>\$15,000</b>	89-38500-4516
*National Extension Aquaculture Workshop. Dr. Carole Engle, University of Arkansas at Pine Bluff, Principal Investigator	10/01/91-09/30/92		
	<b>Project Total</b>	<b>\$3,005</b>	89-38500-4516
*Educational Materials for Aquaculturists and Consumers. Dr. J. T. Davis, Texas A&M University, Principal Investigator	Yr. 1-05/01/91-04/30/92	\$3,971	87-CRSR-2-3218
		<u>35,671</u>	88-38500-4028
	Total Yr. 1	\$39,642	
	Yr. 2-06/01/92-05/31/93	\$59,000	91-38500-5909
	Yr. 3-06/01/93-12/31/94	<u>34,500</u>	92-38500-7110
<b>Project Total</b>	<b>\$133,142</b>		
*Characterization of Finfish and Shellfish Aquacultural Effluents. Dr. J. V. Shireman, University of Florida, Principal Investigator	Yr. 1-05/01/91-04/30/92	\$13,081	88-38500-4028
		82,747	89-38500-4516
		<u>49,172</u>	90-38500-5099
	Total Yr. 1	\$145,000	
	Yr. 2-06/01/92-05/31/93	\$168,105	91-38500-5909
Yr. 3-06/01/93-12/31/94	<u>\$128,936</u>	92-38500-7110	
<b>Project Total</b>	<b>\$442,041</b>		
*Food Safety and Sanitation for Aquacultural Products: Microbial. Dr. J. L. Wilson, University of Tennessee, Principal Investigator	Yr. 1-04/01/92-03/30/93	\$12,649	89-38500-4516
		<u>71,608</u>	90-38500-5099
	Total Yr. 1	\$84,257	
	Yr. 2-06/01/93-05/31/94	\$213,106	92-38500-7110
	Yr. 3-06/01/94-05/31/95	<u>\$237,975</u>	93-38500-8393
<b>Project Total</b>	<b>\$535,338</b>		
*Project Completed			



Project	Duration	Funding	Grant No.
*Aquaculture Food Safety: Residues. Dr. George Lewis, University of Georgia, Principal Investigator	Yr. 1-09/11/92-09/30/93	\$99,393	91-38500-5909
	Yr. 2-10/01/93-09/30/94	\$44,631	90-38500-5099
		<u>107,050</u>	91-38500-5909
	Total Yr. 2	\$151,681	
	Yr. 3-10/01/94-09/30/95	\$89,463	93-38500-8393
	Yr. 4-10/01/95-09/30/96	<u>\$11,392</u>	93-38500-8393
	<b>Project Total</b>	<b>\$351,929</b>	
*National Coordination for Aquaculture Investigational New Animal Drug (INAD) Applications. (In cooperation with other Regional Aquaculture Centers and USDA)	Yr. 1-09/01/93-08/31/94		
	<b>Project Total</b>	<b>\$2,000</b>	90-38500-5099
*Improving Production Efficiency of Warmwater Aquaculture Species Through Nutrition. Dr. Delbert Gatlin, Texas A&M University, Principal Investigator	Yr. 1-01/01/94-12/31/94	\$28,148	90-38500-5099
		123,705	91-38500-5909
		<u>128,444</u>	92-38500-7110
	Total Yr. 1	\$280,297	
	Yr. 2-01/01/95-12/31/95	\$38,059	92-38500-7110
		175,450	93-38500-8393
		<u>32,397</u>	94-38500-0045
	Total Yr. 2	\$245,906	
	Yr. 3-01/01/96-12/31/96	\$23,907	93-38500-8393
		<u>210,356</u>	94-38500-0045
<b>Project Total</b>	<b>\$760,466</b>		
*Delineation and Evaluation of Catfish and Baitfish Pond Culture Practices. Dr. Michael Masser, Auburn University, Principal Investigator	Yr. 1-04/01/94-03/31/95	\$75,530	92-38500-7110
		<u>43,259</u>	93-38500-8393
	Total Yr. 1	\$118,789	
	Yr. 2-04/01/95-03/31/96	\$113,406	94-38500-0045
	Yr. 3-04/01/96-03/31/97	\$28,517	93-38500-8393
		<u>72,281</u>	94-38500-0045
	<b>Project Total</b>	<b>\$332,993</b>	
Publications, Videos and Computer Software. Dr. Michael Masser, Texas A&M University, Principal Investigator (Continuing project)	Yr. 1-04/01/95-03/31/96	\$50,000	94-38500-0045
	Yr. 2-04/01/96-03/31/97	\$13,405	93-38500-8393
		<u>47,543</u>	94-38500-0045
	Total Yr. 2	\$60,948	
	Yr. 3-04/01/97-03/31/98	\$45,900	96-38500-2630
	Yr. 4-04/01/98-03/31/99	\$60,500	97-38500-4124
	Yr. 5-04/01/99-03/31/00	\$67,000	98-38500-5865
	Yr. 6-07/01/00-06/30/01	<u>\$80,550</u>	00-38500-8992
<b>Project Total</b>	<b>\$364,898</b>		
*Project Completed			

Project	Duration	Funding	Grant No.
Management of Environmentally-Derived Off-Flavors in Warmwater Fish Ponds. Dr. Tom Hill, University of Tennessee, Principal Investigator	Yr.1-06/01/96-05/31/97	\$29,349	93-38500-8393
		34,918	94-38500-0045
		<u>186,560</u>	95-38500-1411
	Total Yr. 1	\$250,827	
	Yr.2-06/01/97-05/31/98	\$68,718	94-38500-0045
		97,393	95-38500-1411
		<u>84,031</u>	96-38500-2630
	Total Yr. 2	\$250,142	
	Yr.3-06/1/98-05/31/99	\$154,621	96-38500-2630
		<u>74,645</u>	97-38500-4124
	Total Yr. 3	\$229,266	
Yr.4-06/01/99-05/31/00	\$80,900	98-38500-5865	
Yr.5-06/01/00-05/31/01	<u>\$56,100</u>	<u>99-38500-7375</u>	
<b>Project Total</b>	<b>\$867,235</b>		
*Optimizing Nutrient Utilization and Waste Control through Diet Composition and Feeding Strategies. Dr. Kenneth Davis, University of Memphis, Principal Investigator	Yr.1-12/01/96-11/30/97	\$241,476	95-38500-1411
	Yr.2-12/01/97-11/30/98	\$47,105	95-38500-1411
		<u>210,047</u>	96-38500-2630
	Total Yr. 2	\$257,152	
	Yr.3-12/1/98-11/30/99	\$34,365	96-38500-2630
		<u>199,811</u>	97-38500-4124
Total Yr. 3	<u>\$234,176</u>		
<b>Project Total</b>	<b>\$732,804</b>		
*National Aquaculture Extension Conference (In cooperation with other Regional Aquaculture Centers)	01/01/97-12/31/97	\$3,392	93-38500-8393
		<u>308</u>	95-38500-1411
	<b>Project Total</b>	<b>\$3,700</b>	
Verification of Recommended Management Practices for Major Aquatic Species. Dr. Carole Engle, University of Arkansas at Pine Bluff, Principal Investigator	Yr.1-01/01/97-12/31/97	\$31,410	95-38500-1411
	Yr.2-01/01/98-12/31/99	\$7,186	95-38500-1411
		<u>58,928</u>	96-38500-2630
	Total Yr. 2	\$66,114	
	Yr.3-01/01/99-12/31/00	<u>\$66,925</u>	
<b>Project Total</b>	<b>\$164,449</b>		
*Project Completed			

Project	Duration	Funding	Grant No.
Control of Blue-green Algae in Aquaculture Ponds. Dr. Larry Wilson, University of Tennessee, Principal Investigator	Yr. 1-01/01/99-12/31/99	\$25,147	96-38500-2630
		105,167	97-38500-4124
		<u>177,260</u>	98-38500-5865
	Total Yr. 1	\$307,574	
	Yr. 2-01/01/00-12/21/00	\$975	96-38500-2630
		17,394	97-38500-4124
		158,608	98-38500-5865
		<u>103,662</u>	99-38500-7375
	Total Yr. 2	\$280,639	
	Yr. 3-01/01/01-12/31/01	\$22,042	97-38500-4124
		7,202	98-38500-5865
		195,953	99-38500-7375
		<u>28,129</u>	00-38500-8992
Total Yr. 3	<u>\$253,326</u>		
<b>Project Total</b>	<b>\$841,539</b>		
Management of Aquacultural Effluents from Ponds. Dr. John Hargreaves, Mississippi State University, Principal Investigator	Yr. 1-04/01/99-03/31/00	\$100,000	97-38500-4124
		<u>127,603</u>	98-38500-5865
	Total Yr. 1	\$227,603	
	Yr. 2-04/01/00-03/31/01	\$236,919	99-38500-7375
	Yr. 3-04/01/01-02/31/02	<u>\$150,740</u>	
<b>Project Total</b>	<b>\$615,262</b>		
Development of Improved Harvesting, Grading and Transport Technology for Finfish Aquaculture. Dr. Ed Robinson, Mississippi State University, Principal Investigator	Yr. 1-01/01/01-12/31/01	\$287,053	00-38500-8992
	Yr. 2 - Projected	272,391	
	Yr. 3 - Projected	<u>190,556</u>	
	<b>Project Total</b>	<b>\$750,000</b>	