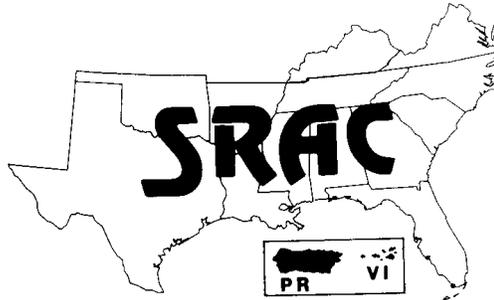

SOUTHERN
REGIONAL
AQUACULTURE
CENTER



TWELFTH ANNUAL PROGRESS REPORT

For the Period Through August 31, 1999

December, 1999

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

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TWELFTH ANNUAL PROGRESS REPORT

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PREFACE

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 1980 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.

ACKNOWLEDGEMENTS

SRAC would like to acknowledge the contributions of the Project Leaders and Participating Scientists involved in the projects reported in this Twelfth 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.

INTRODUCTION

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 United States.

By any measure, the Regional Aquaculture Center program is a productive and effective use of Federal funds. One yardstick of success is the amount of technical information generated through Center-sponsored programs. Some feeling for the productivity of the program can be gained by taking a quick glance at the number of extension publications produced through the SRAC publications project (pages 13-15) 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” project listed on pages 26-28).

A more fundamental measure of success is the extent to which the results of Center 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 the impact of aquaculture on the environment, to improve the shelf-life of aquaculture products, and to reduce the incidence of environment-derived off-flavors in pond-raised fish.

Research in aquatic animal nutrition has been particularly productive, and in one recently completed SRAC project, scientists identified the most cost-effective levels of vitamin and protein supplementation in feeds. Their work resulted in improved feed formulations and feeding practices that have saved the catfish, baitfish, and striped bass industries millions of dollars a year. For example, in the catfish industry alone, feed costs have been reduced \$2-4 a ton as a direct result of work on this project. Assuming overall feed use of 600,000 tons per year in the catfish industry, cost savings average \$1.8 million annually—over three times the amount spent on this project over its 3-year duration.

This Twelfth Annual Progress Report of the Southern Regional Aquaculture Center (SRAC) covers the activities of the Administrative Center during the period September 1, 1998 through August 31, 1999, and includes progress reports on the six multi-year research and extension projects supported by SRAC during this reporting period.

Publications, Videos and Computer Software
Management of Environmentally-Derived Off-Flavors in Warmwater Fish Ponds
Optimizing Nutrient Utilization and Waste Control Through Diet Composition and Feeding Strategies
Verification of Recommended Management Practices for Major Aquatic Species
Control of Blue-green Algae in Aquaculture Ponds
Management of Aquacultural Effluents from Ponds

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 FY's, 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
- David E. Foster, Arkansas Cooperative Extension Service
- Stephen Jones, Alabama Cooperative Extension Service
- Charles Lee, Mississippi State University
- David Morrison, Louisiana State University
- Daniel Smith, Clemson University Cooperative Extension Service
- David H. Teem, Auburn 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 allocations of fiscal resources to ensure that the center develops strong programs in both research and extension; (3) establishment of priorities for regional aquaculture research and extension education activities based on inputs from the 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 on-going 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
Randy Deshotel, LA
Austin Jones, MS
Robert Mayo, NC
John Morrison, AL
Lester Myers, MS (Chairman)
Marty Tanner, FL
R. R. Waldrop, TX
Jerry Williamson, AR (Vice-chairman)
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:

Gary Burtle, GA
Wallis Clark, FL
J. A. Collier, SC
Harry Daniels, NC
Carole Engle, AR
Delbert Gatlin, TX
John Grizzle, AL
John Hargreaves, MS
Ray McClain, LA
Scott Newton, VA
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
Charles "Bo" Collins, AR
Robert Durborow, KY
G. J. Flick, Jr., VA
David Heikes, AR
Tom Hill, TN
Jeff Hinshaw, NC
Andy Lazur, FL
G. W. Lewis, GA
Greg Lutz, LA
Mike Masser, TX (Co-chair)
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

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 distribute funds 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. Approximately 400 copies of this report are distributed by the Administrative Center each year.
- 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.

PROGRESS REPORTS

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

Publications, Videos and Computer Software Page 14

Management of Environmentally-Derived Off-flavors
in Warmwater Fish Ponds Page 20

Optimizing Nutrient Utilization and Reducing Waste
Through Diet Composition and Feeding Strategies Page 33

Verification of Recommended Management Practices for
Major Aquatic Species Page 51

Control of Blue-green Algae in Aquaculture Ponds Page 56

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PUBLICATIONS, VIDEOS AND COMPUTER SOFTWARE

Reporting Period

April 1, 1995 through August 31, 1999

| | | |
|----------------------|--------------|-----------|
| Funding Level | Year 1 | \$50,000 |
| | Year 2 | 61,000 |
| | Year 3 | 45,900 |
| | Year 4 | 60,500 |
| | Year 5 | 74,000 |
| | Total | \$291,400 |

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

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

The most direct benefit from this project to the aquaculture industry is the widespread and ready availability of detailed information on production and marketing of aquacultural products. SRAC fact sheets, videos and other publications are distributed worldwide to a variety of 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 courses in aquaculture and fisheries. Educational institutions at the elementary and high school level use SRAC extension materials to make students aware of aquaculture production and associated trades as a way of life for many people.

Consumers. Information is readily available for consumers who purchase aquacultural products as well as background information on aquaculture.

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 practicing aquaculturists. Videos that demonstrate 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 for clientele throughout the U.S. and the world.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

During this project year, 26 new fact sheets were written and seven were revised. The popular compact disk containing all the fact sheets was revised. All have been distributed throughout the Southern Region and to interested Extension Specialists in other regions. Approximately 21 fact sheets are currently in some stage of writing or revision.

All of these 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 research on which the fact sheets are based. Copies of all of the fact sheets are available on the Internet at <http://www.msstate.edu/dept/srac>.

WORK PLANNED

During the next project year, six new fact sheets will be prepared and two fact sheets are scheduled for revision. In addition there are two research summaries and two fact sheets scheduled to be written by research personnel from other projects.

A new series of SRAC fact sheets—"Species Profiles"—was initiated this year. The series was developed to provide technical information on the biology and culture of various finfish and shellfish that have shown promise as candidates

for aquaculture in the southeast region. Species Profiles will serve two purposes. First, they will provide an accessible summary of general information for students and individuals interested in the culture of the species covered in the series. Second, they will provide a source of technical information for the SRAC Technical Committee and Industry Advisory Council when they consider funding projects involving minor aquaculture species in the region. A total of 15 Species Profiles will be developed, with six to be prepared during the coming project year.

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 6 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, the SRAC website is accessed over 20 times a day, and most of those visits are probably from people searching for technical information in fact sheets posted at the site. 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.

Though there has been no attempt to quantify the impact financially, one Extension agent has estimated that using this information to help prospective producers make the correct decision about entering the aquaculture business results in savings of at least \$100,000 per contact. For producers already in the business, the savings from enhanced production efficiency are probably on the order of \$5,000 per producer using the service per year.

RESULTS AT A GLANCE...

- Twenty-six new fact sheets were written this year.
- Forty-six scientists from across the Southern Region contributed to publications produced by SRAC in this project year.
- SRAC has now published 157 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.

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 (4/1/98 - 5/31/99)

- Avery, Jimmy L., Robert P. Romaine, and W. Ray McClain. Crawfish Production: Production Economics, Pond Construction and Water Supply. SRAC Fact Sheet 240 (Revision).
- Avery, Jimmy L., Robert P. Romaine, and W. Ray McClain. Crawfish Production: Production Systems and Forages. SRAC Fact Sheet 241 (Revision).
- Barziza, Daniel E., Wendy M. Sealey, James T. Davis, and Delbert M. Gatlin III. Feeding Practices for Baitfish. SRAC Fact Sheet 123.
- Beem, Marley. Aquaculture: Realities and Potentials When Getting Started. SRAC Fact Sheet 441 (Revision).
- Brunson, Martin W., Nathan Stone, and John Hargreaves. Fertilization of Fish Ponds. SRAC Fact Sheet 471.
- Camus, A.C, R. M. Durborow, W. G. Hemstreet, R. L. Thune, and J. P. Hawke. Aeromonas Bacterial Infections - Motile Aeromonad Septicemia. SRAC Fact Sheet 478.
- Dunning, Rebecca D., Thomas M. Losordo, and Alex O. Hobbs. The Economics of Recirculating Aquaculture Tank Production Systems: A Spreadsheet to Guide Individual Analysis. SRAC Fact Sheet 456.
- Durborow, Robert M., Andrew J. Mitchell, and M. David Crosby. Ich (White Spot Disease). SRAC Fact Sheet 476.
- Durborow, Robert M., Ronald L. Thune, John P. Hawke, and A. C. Camus. Columnaris Disease: A Bacterial Infection Caused by *Flavobacterium columnare*. SRAC Fact Sheet 479.
- Hargreaves, John. Design and Construction of Degassing Units for Catfish Hatcheries. SRAC Fact Sheet 191.
- Hargreaves, John. Control of Clay Turbidity in Ponds. SRAC Fact Sheet 460.

- Hawke, J. P., R. M. Durborow, and R. L. Thune. ESC - Enteric Septicemia of Catfish. SRAC Fact Sheet 477.
- Hinshaw, Jeffrey M. Trout Production: Feeds and Feeding Methods. SRAC Fact Sheet 223 (Revision).
- Killian, H. Steven, David Heikes, Peter Van Wyk, Michael Masser, and Carole Engle. Inventory Assessment Methods for Aquaculture Ponds. SRAC Fact Sheet 395.
- Lazur, Andrew and Deborah Britt. Pond Recirculating Production Systems. SRAC Fact Sheet 455.
- Lazur, Andrew M. Small Scale, On-farm Fish Processing. SRAC Fact Sheet 442.
- Losordo, Thomas M., Michael P. Masser, and James Rakocy. Recirculating Aquaculture Tank Production Systems: An Overview of Critical Considerations. SRAC Fact Sheet 451 (Revision).
- Ludwig, Gerald M., Nathan M. Stone, and Charles "Bo" Collins. Fertilization of Fish Fry Ponds. SRAC Fact Sheet 469.
- Ludwig, Gerald M. Zooplankton Succession and Larval Fish Culture in Freshwater Ponds. SRAC Fact Sheet 700.
- Lutz, Greg and Jimmy Avery. Bullfrog Culture. SRAC Fact Sheet 436.
- Masser, Michael P. and Rex Dunham. Production of Hybrid Catfish. SRAC Fact Sheet 190.
- Masser, Michael P. and Tom Popma. Tilapia - Life History and Biology. SRAC Fact Sheet 283.
- Masser, Michael P. Water Gardens. SRAC Fact Sheet 435.
- Masser, Michael P., James Rakocy, and Thomas M. Losordo. Recirculating Aquaculture Tank Production Systems: Management of Recirculating Systems. SRAC Fact Sheet 452 (Revision).
- Mitchell, Andrew J., Robert M. Durborow, and M. David Crosby. Proliferative Gill Disease (Hamburger Gill Disease). SRAC Fact Sheet 475.
- Robinson, Edwin H., Meng H. Li, and Martin W. Brunson. Feeding Catfish in Commercial Ponds. SRAC Fact Sheet 181 (Revision).
- Sealey, Wendy M., Daniel E. Barziza, James T. Davis, and Delbert M. Gatlin, III. Dietary Protein and Lipid Requirements of Golden Shiners and Goldfish. SRAC Fact Sheet 124.
- Sealey, Wendy M., James T. Davis, and Delbert M. Gatlin, III. Refinement of Vitamin Supplementation in Diets for Pond-raised Channel Catfish. SRAC Fact Sheet 188.
- Sealey, Wendy M., James T. Davis, and Delbert M. Gatlin, III. Restricted Feeding Regimes Increase Production Efficiency in Channel Catfish. SRAC Fact Sheet 189.
- Sealey, Wendy M., Daniel E. Barziza, James T. Davis, and Delbert M. Gatlin III. Improving Feeds for Hybrid Striped Bass. SRAC Fact Sheet 304.
- Steeby, Jim A., Nathan Stone, H. Steven Killian, and Dennis K. Carman. Repairing Fish Pond Levees. SRAC Fact Sheet 104.
- Stone, Nathan. Renovating Leaky Ponds. SRAC Fact Sheet 105.
- Wellborn, Thomas L. and Martin W. Brunson. Construction of Levee-type Ponds for Fish Production. SRAC Fact Sheet 101 (Revision).

Manuscripts in review

- Hinshaw, Jeffrey M. Trout Production: Handling Eggs and Fry. SRAC Fact Sheet 220 (Revision).
- Hinshaw, Jeffrey M. Trout Farming: Carrying Capacity and Inventory Management. SRAC Fact Sheet 222 (Revision).

Losordo, Thomas M., Michael P. Masser, and James Rakocy. Recirculating Aquaculture Tank Production Systems: A Review of Component Options. SRAC Fact Sheet 453.
Treece, Granvil. Laboratory and Tank Production of Live Feed for Larval Fish.

Manuscripts in preparation

Gatlin, Delbert M., III. Improving Production Efficiency of Warmwater Aquaculture Species Through Nutrition. SRAC Project Summary.
Hodson, Ron. Hybrid Striped Bass - Pond Production of Food Fish. SRAC Fact Sheet 303 (Revision).
Lutz, Greg and J. Avery. Production of Turtles.
Rakocy, James. Use of Aquaculture Effluents.
Rakocy, James. Recirculating Aquaculture Tank Production Systems: Integrating Fish and Plant Culture. SRAC Fact Sheet 454 (Revision).
Silva, Juan. Food Safety Techniques for Aquacultural Products.

CD ROM

Davis, James T. SRAC Publications CD (Revision).

Final Project Summary

Engle, Carole R. Analysis of Regional and National Markets for Aquacultural Products Produced for Food in the Southern Region. SRAC 601.
Tucker, Craig S. Characterization and Management of Effluents from Aquaculture Ponds in the Southeastern United States. SRAC 600.

RAC Results

Davis, James T. and Michael P. Masser. The Southern Regional Aquaculture Center's Publications, Videos and Computer Software Project.
Tucker, Craig S. Characterization and Management of Effluents from Aquaculture Ponds in the Southeastern United States.



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

Reporting Period

June 1, 1996 to August 31, 1999

| | | |
|----------------------|--------------|-----------|
| Funding Level | Year 1 | \$251,200 |
| | Year 2 | 250,900 |
| | Year 3 | 230,900 |
| | Year 4 | 80,900 |
| | Year 5 | 56,100 |
| | Total | \$870,000 |

| | | |
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PROJECT OBJECTIVES

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.

Improving the efficiency of phosphorus utilization in fish feeds should reduce the phosphorus input to catfish ponds. Since phosphorus is a key algal nutrient, reduced input may lead to decreased algal abundance, thereby reducing the 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 precipitating phosphorus from 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. Soil treatment methods that would enhance the ability of pond bottom soil to remove phosphorus from pond water would be equally valuable to aquaculturists.

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.

Several chemical control measures are being investigated, including the use of natural compounds such as plant phenolics. These studies could 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 and better than chromatographic 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 industry with a better tool for quality control and fish grading as well as provide the research community with a better tool to study off-flavor development and control.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

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.*

The minimum dietary available phosphorus requirement for food-size channel catfish fed commercial type diets was determined in a 7-month pond feeding experiment. 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 NRC (National Research Council) recommendation.

The availability of different forms of phosphorus in practical feed ingredients was determined for channel catfish by feeding diets containing feed ingredients that varied in phosphorus composition. Bioavailability varied from 31 to 89%. Of the plant feedstuffs tested, wheat, sorghum and cottonseed meal had greater phosphorus availability than soybean meal, corn, or rice bran. Of the animal feedstuffs, fish meal analog had greater availability than menhaden meal or meat and bone meal. Uptake and mineralization of different forms of phosphorus and nitrogen in feces from channel catfish fed the various ingredients was also determined.

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 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 (P) 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 P between fish fed the two diets. There were no significant differences in total P, soluble P, 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 P generation by food-sized channel catfish fed low P diets. A basal diet was formulated to contain 32% protein without supplemental P (0.2% available P). Supplemental P was added to the basal diet to provide available P of 0.3 and 0.4%, respectively using dicalcium phosphate. There were no significant differences in total P, soluble P, 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.



Catfish ponds in the Mississippi Delta.

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.*

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 8 kg/ha 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 mg/L every other week did not affect total production

or feed conversion of channel catfish in 0.04-ha ponds. In addition, waterborne phosphorus concentration, primary 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. The treatments used in the 1999 pond trial draw from the most promising treatments of the previous three years. Preliminary results show the greatest phosphorus reduction in the treatment that combines pond bottom drying and tilling with gypsum application to the pond water.

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

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* cf. *chalybea* 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 0.1-ha ponds in 1998. *O. chalybea* numbers were reduced, but not eliminated and difficulty was encountered in adapting a silver carp system to channel catfish production ponds.

Evaluations are continuing with threadfin shad and Nile tilapia in 0.05-ha ponds in polyculture with channel catfish. Stocking levels of Nile tilapia in cages are 1250 and 2500 Nile tilapia/ha, lower than have previously provided control of *O. chalybea*.

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

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 800-L (211-gallon) tanks to study control of *Oscillatoria chalybea*. 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 *Oscillatoria chalybea*, but high oxygen demands were observed. Preparations of the fungus are being developed that will minimize the oxygen demand.

A bacterium 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 3×10^9 plaque forming units per milliliter of culture broth. Therefore, 1-L of culture broth when uniformly distributed in 1,233.6 m³ (one

acre-foot) of pond water, would result in an initial concentration of approximately 2,400 PFU per milliliter of pond water. In replicated tests conducted in 800-L (211-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* cf. *chalybea*

was reduced from an initial density of 2,700 filaments per milliliter to 0 filaments per milliliter after 48 hours. *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 odorous blue-green algae while having no effect on beneficial algae. The bacterium has no adverse affect on catfish and shows great potential as a biological control agent for blue-green algae responsible for off-flavors.

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

Streptomyces tendae is known to synthesize geosmin, an earthy off-flavor contaminant 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 mg/mL. Propyl gallate is inhibitory at higher concentrations, but methyl gallate and gallic acid have no inhibitory effects at concentrations up to 1 mg/ml. 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 $\mu\text{g}/\text{disk}$, 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 $\mu\text{g}/\text{ml}$. Methyl gallate was effective at 500 $\mu\text{g}/\text{ml}$. 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.*

Eighteen, 0.4-ha (1-acre) earthen ponds in northwest Mississippi were used in a 3-year study to evaluate the effectiveness of weekly applications of copper sulfate pentahydrate at reducing the incidence of environment-derived off-flavors in pond-raised channel catfish. Each spring when water temperatures increased above 20°C (70°F), nine of the ponds were treated weekly with 2.3 kg/ha copper sulfate (5 pounds/acre) 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 20°C (70°F).

Over the 3-year duration of the study, incidence of off-flavor was reduced by 80% in copper-treated ponds relative to untreated ponds. Episodes of off-flavor in copper-treated ponds were also of shorter duration than in untreated ponds, and the presence of off-flavor never delayed harvest in copper-treated ponds. Off-

flavor episodes in untreated ponds were of highly variable duration, with off-flavors never detected in fish from one pond while fish in another pond were off-flavor for nearly a year. Off-flavors delayed fish harvest on ten occasions in untreated ponds.

Average annual fish harvest was 5,210 kg/ha (4,651 pounds/acre) in untreated ponds and 5,900 kg/ha (5,268 pounds/acre) in copper-treated ponds. The 12% reduction in fish harvest from untreated ponds was due primarily to infectious disease outbreaks in one or two ponds each year when harvest was delayed due to off-flavor. In other words, the inability to harvest fish in a timely fashion exposed fish to a greater risk of loss to diseases and other causes.

RESULTS AT A GLANCE...

A simple pond-treatment program using copper sulfate provided the following benefits:

- Over the 3-year duration of the study, incidence of off-flavor was reduced by 80%.
- Off-flavors never delayed harvest in copper-treated ponds.
- Enterprise budgets showed that managing off-flavor using copper sulfate increased annual net returns above variable costs by nearly 50%.

Enterprise budgets showed that average annual net returns above variable costs were \$1,405/ha (\$569/acre) for untreated ponds and \$2,095/ha (\$848/acre) for the copper-treated ponds. Variation in net returns was twice as great for untreated ponds as for treated ponds, indicating increased stability in production and returns when using copper sulfate. High variation in annual performance of control ponds resulted from one or more ponds having very good 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.*

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, was 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.*

Work on this objective will begin next year.

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

Polyclonal antibody (Pab) sera, provided by the USDA-Southern Regional Research Center for compounds similar in structure to geosmin and MIB (argosmin and camphor), were evaluated in ELISA. Both sera were found to be unacceptable, possibly due to the length of time the serum had previously been stored (7 years, frozen at -10 to -20°C) or an inherent problem with the Pab. Both sera showed very high background binding and very poor sensitivity to MIB and geosmin (between 10 and 100 µg/mL). In the last year, however, 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 an absorbant 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. The purge and trap distillation apparatus accompanied with GC/MS analysis for geosmin and MIB detection has been setup and now is being used. The conditions are those of Johnsen and Lloyd, 1992. We are currently optimizing the methods to enhance recoveries of geosmin and MIB. The sensitivity for the GC/MS is 0.025 ng MIB per injection.

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.*

See list of publications on pages 26-28.

WORK PLANNED

Work on all objectives is proceeding on schedule and no changes in the project have occurred this year. The work planned for the next two years to enhance removal of phosphorus from pond soils will not deviate from that outlined in the original proposal.

Another series of digestibility trials is currently underway with channel catfish to evaluate several different feedstuffs which have been genetically modified to contain low concentrations of phytic acid. The phytic acid found in reasonably high concentrations in plant feedstuffs is indigestible and thus excreted by fish. Low-phytate varieties of barley, corn and soybean meal are being evaluated and may provide a dietary means of reducing phosphorus excretion by channel catfish.

Commercial level scale-up of the most promising

system using filter feeders will be replicated in 2000 in a channel catfish polyculture. No significant changes in direction or emphasis are anticipated in studies of algal pathogens. Research will emphasize experiments to evaluate the bacterium to control species of *Oscillatoria* and *Anabaena* in tanks and small ponds. Efficacy of the bacterium as influenced by inoculum concentration will be determined.

Methods of MIB extraction (solvent and/or absorbant) methods and fish assay methods will be optimized using the MIB ELISA. Alternative methods for extraction and analysis will be investigated. The rapid clearance rate of MIB in hybrids suggest these animals may distribute MIB differently than other species. Further studies should be carried out to examine physiological differences between these and other catfish strains.

IMPACTS

Most of the work in this project has not been completed, so it is difficult to assess impacts. However, the information generated in work to investigate phosphorus availability of various feedstuffs is already being used to refine commercial diet formulations.

A major impact of this work has been the

development of a successful regimen for preventing algae-related off-flavors using copper sulfate. This treatment regimen has already been adopted by many commercial producers in Mississippi and Arkansas, and shows promise in reducing the economic burdens associated with the incidence of off-flavor.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

Publications in print

- Boyd, C.E. 1997. Practical aspects of chemistry in pond aquaculture. *The Progressive Fish-Culturist* 59:85-93.
- Boyd, C. E. 1998. Water quality for pond aquaculture. Alabama Agricultural Experiment Station, Auburn University, Alabama, Research and Development Series No. 43.
- Boyd, C. E. and A. Gross. 1999. Biochemical oxygen demand in channel catfish pond waters. *Journal of the World Aquaculture Society* 30:349-356.
- Boyd, C. E. and C. S. Tucker. 1998. *Pond Aquaculture Water Quality Management*. Kluwer Academic Publishers, Boston.
- Buyukates, Y. 1998. Determination of phosphorus composition and availability from various feedstuffs to channel catfish. Master's thesis, Texas A&M University.
- Chung, K. T. and C. I. Wei. 1997. Food tannins and human health: a double-edge sword? *Food Technology* 51:124.
- Chung, K. T. and T. Y. Wong, C. I. Wei, Y. W. Huang, and Y. Lin. 1998. Tannins and human health: a review. *Critical Review in Food Science and Nutrition* 38:421-464.
- Chung, K. T., Z. Lu, and M. W. Chou. 1998. Mechanism of inhibition of tannic acid and related compounds on the growth of some intestinal bacteria. *Food and Chemical Toxicology* 36:1053-1060.
- Eya, J. C. and R. T. Lovell. 1997. Available phosphorus requirements of food-size channel catfish fed practical diets in ponds. *Aquaculture* 154:283-291.
- Eya, J. C. and R. T. Lovell. 1997. Net absorption of dietary phosphorus from various inorganic sources and effect of fungal phytase on net absorption of phosphorus by channel catfish. *Journal of the World Aquaculture Society* 28:386-391.
- Eya, Jonathan C. 1998. Reducing phosphorus content of catfish feeds in ponds. Ph.D. dissertation, Auburn University, Alabama.
- Giri, B. J., III. 1998. The effect of regular addition of agricultural limestone on water quality in channel catfish production ponds. Master's thesis, Auburn University, Alabama.
- Gross, A. and C. E. Boyd. 1998. A digestion procedure for the simultaneous determination of total nitrogen and total phosphorus in pond water. *Journal of the World Aquaculture Society* 29:300-303.
- Gross, A., C. E. Boyd, and J. Seo. 1998. Evaluation of the ultraviolet spectrophotometric method for the measurement of total nitrogen in water. *Journal of the World Aquaculture Society* 30:388-393.
- Gross, A., C. E. Boyd, R. T. Lovell, and J. C. Eya. 1998. Phosphorus budgets for channel catfish ponds receiving diets with different phosphorus concentrations. *Journal of the World Aquaculture Society* 29:31-39.
- Massaut, L. 1998. Plankton trophic interactions in catfish and sportfish ponds in the presence of omnivorous, filter feeding fish. Ph.D. dissertation, Auburn University, Alabama.
- Perkins, E. J. and D. Schlenk. 1997. Comparisons of uptake and depuration of 2-methylisoborneol in male, female, juvenile and 3MC-induced channel catfish (*Ictalurus punctatus*). *Journal of the World Aquaculture Society* 28:158-164.

- Perkins, E. J. and D. Schlenk. 1998. Immunochemical characterization of hepatic cytochrome P450 isozymes in the channel catfish: assessment of sexual, developmental and treatment-related effects. *Comparative Biochemistry and Physiology* 121C:305-310.
- Robinson, E. H., L. S. Jackson, and M. H. Li. 1996. Supplemental phosphorus in practical channel catfish diets. *Journal of the World Aquaculture Society* 27:303-308.
- Tucker, C. S. 1998. Low dosages of copper may reduce off-flavor. *The Catfish Journal* XIII(3):5.
- Tucker, C. S. and M. van der Ploeg. 1999. Managing off-flavor problems in pond-raised catfish. Southern Regional Aquaculture Center Publication 192.
- Walker, H. L. and C. L. Patrick. 1998. Method of isolating and propagating microorganisms and viruses. U.S. Patent No. 5,739,019.
- Zhao, G., K. T. Chung, K. Milow, W. Wand and S. E. Stevens, Jr. 1997. Antibacterial properties of tannic acid and related compounds against the fish pathogen, *Cytophaga columnaris*. *Journal of Aquatic Animal Health* 9:309-313.

Manuscripts

- Buyukates, Y., S. D. Rawles and D. M. Gatlin III. Submitted. Phosphorus composition of various feedstuffs and apparent phosphorus availability to channel catfish (*Ictalurus punctatus*). *North American Journal of Aquaculture*.
- Boyd, C. E. In press. Microbiological and physiochemical characteristics of pond sediment and methods for improving oxygenation of the soil-water interface. In: *Proceedings of Biotechnology Conference, Phuket, Thailand*.
- Goktepe, I., C. Huang, and L. Plhak. In preparation. Measurement of 2-methylisoborneol in pondwater samples using ELISA.
- Gross, A., C. E. Boyd, and C. W. Wood. In press. Ammonia volatilization from channel catfish ponds. *Journal of Environmental Quality*.
- Huang, C. and L. Plhak. In preparation. Optimization of an ELISA for the analysis of 2-methylisoborneol.
- Kalinec, D., Y. Buyukates, S. D. Rawles, and D. M. Gatlin III. In preparation. Effects of alum addition on phosphorus dynamics in pond production of channel catfish. *Journal of the World Aquaculture Society*.
- Park, E.S. and L. Plhak. In preparation. Production of high affinity antibodies for the measurement of the flavor compound, 2-methylisoborneol.
- Park, E-S. and L. Plhak. In preparation. Production of high affinity antibodies for the measurement of the flavor compound, 2-methylisoborneol.
- Pavek, R. E. 1998. Effect of sodium nitrate enrichment on water quality variables, bottom sediments, and catfish production in earthen ponds. M.S. Thesis, Auburn University, Alabama.
- Perkins, E. J. and D. Schlenk. In press. Immunochemical characterization of hepatic cytochrome P450 isozymes in the channel catfish: assessment of sexual, developmental and treatment-related effects. *Comparative Biochemistry and Physiology*.
- Perschbacher, P.W. In preparation. Biological control of planktonic algae by filter-feeding fish for sustainable pond production. *Journal of Applied Aquaculture*.
- Perschbacher, P. W. and F. Fijan. In preparation. Channel catfish industry in the USA and the problem of off-flavor. *Ribarstvo (Croatia)*.

- Plhak, L. 1999. In press. Quality Control Methods to Improve Catfish Flavor. Louisiana Agriculture. Fall 1999 Issue.
- Schlenk, D., B. Hawkins, and E. J. Perkins. In press. Effect of ethanol, clofibrilic acid and temperature on the uptake and elimination of 2-methylisoborneol in channel catfish (*Ictalurus punctatus*). Fish Biochemistry and Physiology.
- Schlenk, D., B. Hawkins, E. J. Perkins, B. C. DeBusk, and D. Schlenk. In preparation. Comparison of 2-methylisoborneol elimination rates and cytochrome P450 expression in channel catfish: Lack of significant correlation and biotransformation. Drug Metabolism and Disposition.
- Schlenk, D., B. Hawkins, E. J. Perkins, B. C. DeBusk, and D. Schlenk. Submitted. Comparison of 2-methylisoborneol elimination rates and cytochrome P450 expression in channel catfish: Lack of significant correlation and biotransformation. Canadian Journal of Fisheries and Aquatic Sciences.
- Tucker, C. S. In press. Off-flavor problems in aquaculture. Reviews in Fisheries Science.
- Tucker, C. S. and T. Hanson. In press. Management of off-flavor with copper sulfate. The Catfish Journal.
- Walker, H. L. and L. R. Higginbotham. In preparation. Evaluation of a bacterial pathogen for control of cyanobacteria. Biological Control.
- Walker, H. L. and L. R. Higginbotham. Control of cyanobacteria with a bacterium. Patent application. (Submitted to U.S. Patent Office).
- Walker, H. L. and L. R. Higginbotham. Submitted. An aquatic bacterium that lyses cyanobacteria associated with off-flavor of channel catfish (*Ictalurus punctatus*). Biological Control.

Papers presented

- Boyd, C. E. 1998. Phosphorus chemistry in pond soils. Aquaculture '98, Las Vegas, Nevada, 15-19 February.
- Boyd, C. E. 1999. Water quality in channel catfish farming. Aquaculture '99, Sydney, Australia, 26 April - 2 May.
- Chung, K. T., Z. Lu and M. W. Chou. 1997. Effects of a tannins on growth of intestinal bacteria, 97th General Meeting of the American Society for Microbiology, A-111, Miami Beach, Florida, 4-8 May.
- Clarizia, L., K. T. Chung and S. E. Stevens, Jr. 1997. Effects of tannins on growth of *Streptomyces tendae*. 97th General Meeting of the American Society for Microbiology, Abstract 0-57, Miami Beach, Florida, 4-8 May.
- Park, E. S. and Plhak, L. 1998. Development of monoclonal antibody and enzyme immunoassay for 2-methylisoborneol, 4th. International Conference on Toxic Cyanobacteria, Beauford, North Carolina, 27 September - 1 October.
- Perschbacher, P. W. and J. L. White. 1999. Biological control of planktonic algae for sustainable pond production: Evaluation of filter-feeding macroorganisms. Aquaculture America 1999, Tampa, Florida, 27-30 January.
- Walker, H. L. 1997. Biological control of blue-green algae that cause off-flavor in channel catfish. Annual Meeting, Louisiana Catfish Farmers Association.
- Walker, H. L. 1998. Biological control of blue-green algae that cause off-flavor in channel catfish. Annual Meeting, Louisiana Catfish Farmers Association.



OPTIMIZING NUTRIENT UTILIZATION AND REDUCING WASTE THROUGH DIET COMPOSITION AND FEEDING STRATEGIES

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| | | |
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PROJECT OBJECTIVES

1. Determine the effects of diet composition on fish production, nutrient utilization, and excretion of organic and nitrogenous wastes.
 - a. Evaluate the effects of minimizing protein concentrations via amino acid supplementation of diets for channel catfish. The proposed research should be based on, and augment, available information concerning protein and amino acid nutrition of this species.

- b. Evaluate manipulations of dietary protein concentration and energy density as well as inclusion of specific diet additives to improve growth efficiency and nitrogen retention while limiting excretion of wastes by channel catfish and hybrid striped bass (sunshine bass).
 2. Assess the effects of various feeding strategies and techniques on fish production, nutrient utilization, and waste reduction.
 - a. Optimize feeding strategies in relation to water temperature for channel catfish production. Of particular interest is delineation of more precise feeding strategies when water temperatures are cool (spring, late fall) and extremely hot (late summer, early fall).
 - b. Evaluate alternative feeding strategies including manipulation of diet composition in relation to such variables as water temperature and fish size for channel catfish, baitfish, and hybrid striped bass (sunshine bass).
 - c. Develop and refine feeding strategies for crawfish that effectively enhance production by augmenting the forage-based system.
 3. Develop publications to effectively extend information derived from this project to feed manufacturers and fish producers.

ANTICIPATED BENEFITS

The overall goal of this project is to improve the efficiency of nutrient utilization in aquaculture feeds and forage, which has two important implications. First, efficient use of feeds and forage should make farming more profitable because feed costs represent a large fraction of the total cost of aquaculture production. Second, optimizing nutrient retention may improve culture system water quality and reduce the impact of aquaculture on the environment by decreasing waste production.

Objective 1. Work on this objective will provide information which will increase the efficiency of commercial diet assimilation by channel catfish and hybrid striped bass, with a concomitant reduction in waste generation. These improvements should increase the cost-effective-

ness of producing these fish in aquaculture and limit potential negative environmental impacts from waste production.

Protein is the most expensive component of channel catfish diets and a primary source of nitrogen in production ponds. Commercial catfish feeds contain proteins of animal and plant origin that enter the production pond in uneaten feed and fish feces. Nitrogen is released from feed and feces by bacterial decomposition which contributes to poor water quality. Thus, reducing the protein content of catfish diets could help to reduce feed cost and might improve water quality in catfish ponds.

Research with other aquatic species suggests that effective reduced-nitrogen diets can be

made by balancing dietary protein to match the amino acid composition of a species-specific “ideal protein.” An ideal-protein diet for channel catfish should possess an amino acid composition similar to that in the whole body of catfish, an indispensable amino acid content that meets all minimum dietary requirements, and a quantity of dispensable amino acids sufficient to meet the demands of protein synthesis during rapid growth. Such a diet might be utilized more efficiently than diets in which the amino acid composition is less strictly controlled.

Work under this objective includes evaluating the efficacy of several reduced-nitrogen, ideal-protein diets for channel catfish. It is hoped that these diets will increase dietary nitrogen utilization, improve fish growth, and reduce the amount of nitrogenous waste entering catfish ponds under typical production conditions.

The total cost of catfish diet ingredients can be reduced approximately 5% by using all plant protein ingredients and balancing acids in the diet with crystalline amino acids compared to a similar diet containing fish meal to balance amino acids. Nitrogen and phosphorus utilization may improve when channel catfish are fed diets with balanced amino acid content, leading to better overall dietary efficiency and improved profitability.

This objective also includes work on dietary

enhancement for the culture of reciprocal cross hybrid striped bass (sunshine bass). Increasing the lipid composition and supplementation of specific feed additives in hybrid striped bass diets may result in growth enhancement and increase nutrient utilization.

Objective 2. Precise feeding regimens for use when water temperatures are cool (spring, late fall) and extremely hot (late summer and early fall) may improve production efficiency and nutrient utilization in channel catfish farming. Improved feed management strategies that utilize daily feeding frequency and timing to optimize fish growth would significantly reduce feed costs by lowering labor requirements, reduce wear on machinery, and lead to greater production efficiency. Other feeding strategies, such as reducing daily feed allowance by feeding more concentrated feeds (high protein percentage), will allow the farmer to feed more conservatively and thereby waste less feed.

Formulating better baitfish and crawfish diets is expected to improve overall performance of golden shiners and crawfish in commercial production systems and improve profitability. Results from this objective will provide producers with viable, cost-effective feeding strategies that can enhance production by augmenting the forage-based food system. Identification of effective, low-cost feeds and practical feeding strategies will facilitate efficient supplemental feeding practices that minimize

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Objective 1a. *Evaluate the effects of minimizing protein concentrations via amino acid supplementation of diets for channel catfish. The proposed research should be based on, and augment, available information concerning protein and amino acid nutrition of this species.*

Texas A & M University. This project was initiated in March of 1997 with a 2-year pond feeding trial to evaluate the use of lysine supplementation to reduce total dietary protein and limit nitrogenous waste excretion in channel catfish production. Two experimental diets containing 25% crude protein and a standardized reference diet containing 30% crude protein from practical ingredients were fed to mixed sizes of channel catfish in earthen ponds. One of the experimental diets did not contain supplemental lysine, whereas the other was supplemented with 0.5% of lysine HCl to provide the same level of lysine as the reference diet. Fish in each pond were selectively harvested by grader seine in October 1997, May 1998 and October 1998 after which fingerling fish were added back to each pond. The final harvest took place in May 1999. Yield of marketable fish at each intermediate harvest was not affected by diet, and total yield data indicated that fish production was not negatively affected by reducing dietary protein from 30 to 25% of diet. Further, lysine supplementation of the diet with 25% protein did not confer any added benefits, and no effects on water quality could be attributed to the dietary manipulations. Body composition of fish fed the various diets is currently being determined to assess nitrogen retention.

Louisiana State University. The project was initiated in the spring of 1997. Objectives are to determine the effects of reduced-protein diets on

production yield, feed conversion efficiency, dressing percentage, and body composition of pond-raised channel catfish during a continuous, three-year production period, as well as effects of lowered-nitrogen diets on pond water quality. Fingerling fish (8-10 cm [3-4 inch] average length) were stocked in sixteen, 0.08-hectare (0.2-acre) ponds, at a density of 25,000 fish/hectare (10,000 fish/acre), in late spring. Fish are being fed one of four, isocaloric, extruded (floating) catfish feeds formulated to contain 26-30% crude protein. Each diet has been assigned to four, randomly selected ponds and fish are fed daily as much as they will consume in 30 minutes.

Diets being tested are 30%, 28% and 26% crude protein, plus a control diet. Reductions in dietary crude protein from 30 to 26% were achieved by decreasing the dispensable (dietary non-essential) amino acid content of the diets by 10 to 20% (28 and 26% CP diets, respectively), while maintaining concentrations of all indispensable (dietary essential) amino acids at minimum required levels. The ratio of each dispensable amino acid to lysine was held constant in all diets except the control. Diets were manufactured at a commercial feed mill.

Four top-harvests have been conducted to date, in the fall of 1997, the spring and fall of 1998, and the spring of 1999. After each harvest, fingerlings were restocked to maintain a density of 25,000 fish/hectare (10,000 fish/acre). The next top-harvest

RESULTS AT A GLANCE...

The protein level in catfish feed can be reduced to 25 or 26% without affecting fish production. Reduced protein levels in feeds will reduce feed costs and improve nutrient utilization.

will be conducted in October-November, 1999. At each harvest, 100 fish were taken from each pond for determination of body composition and dressing percentage. Water quality parameters and chlorophyll *a* concentrations

are monitored twice weekly to determine the effects of dietary treatments on pond water quality.

Results through spring 1999 harvest are shown in Tables A and B:

| Treatment | Total Diet Fed (kg) | Total Weight of Fish Harvested (kg) | Feed Conversion Ratio |
|-----------|---------------------|-------------------------------------|-----------------------|
| Control | 9,852 | 3,231 | 3.0 |
| 30% CP | 8,826 | 3,257 | 2.7 |
| 28% CP | 8,717 | 2,925 | 3.0 |
| 26% CP | 8,526 | 2,933 | 2.9 |

| Average Treatment | Visceral Fish Weight (g) | Dressed Fat (%) | Yield (%) |
|-------------------|--------------------------|-----------------|------------|
| Control | 515 (161) | 2.5 (0.1) | 58.6 (0.1) |
| 30% CP | 583 (98) | 2.8 (<0.1) | 59.3 (0.3) |
| 28% CP | 626 (29) | 2.5 (0.1) | 58.2 (0.3) |
| 26% CP | 470 (154) | 2.2 (0.1) | 58.5 (0.1) |

The University of Georgia. Channel catfish stocked in earthen ponds at the rate of 25,000 fingerlings per ha (10,000/acre) were fed soybean meal-corn-wheat middling diets that were formulated to contain 0.65% or 0.94% as methionine by the addition of DL-methionine or 0.94% as methionine by addition of menhaden fish meal. In the first year of a two-year trial, channel catfish yields were similar between all-plant ingredient diets and a diet that contained menhaden fish meal. The all-plant ingredient diet cost 5% less than the fish meal diet but had similar production. Water quality parameters,

including ammonia, nitrite, and total phosphorus, were similar in all treatments during the first year. After five partial harvests in the first year, a residual of smaller fish were carried over until the second year when stocking density was brought to 25,000 fish/ha (10,000/acre). The second year production cycle ended with similar gross catfish yield among treatments over the two year period, 11,728, 12,095, and 11,514 kg/ha, respectively (10,461, 10,789, 10,270 pounds/acre). The three diet formulas did not appear to affect proximate body composition. However, when feed intake was

considered, the net protein utilization was 8.7% higher when 0.94% methionine was added to soybean meal-corn-wheat middling diets versus the addition of 0.65% methionine. Although the implications for water quality improvement were

RESULTS AT A GLANCE...

Catfish feeds with all-plant protein can be used without affecting fish production. The cost of feed ingredients can be reduced by as much as 5% by using all-plant protein sources.

not shown in this short trial, the prospects for recovery of significantly more protein nitrogen by the channel catfish should encourage the practice of adequate methionine supplementation to all-plant-protein diets.

Objective 1b. *Evaluate manipulations of dietary protein concentration and energy density as well as inclusion of specific diet additives to improve growth efficiency and nitrogen retention while limiting excretion of wastes by channel catfish and hybrid striped bass (sunshine bass).*

Auburn University. Feeding 12.5% less of a 32% protein feed to catfish in production ponds resulted in the same yield of fish as feeding a 28% protein feed to satiation. Feed efficiency and economics were improved by feeding the 32% protein feed at the reduced rate; however, no differences were measured in nitrogen or phosphorus wastes, or phytoplankton production. Feeding 22.5% less of a 36% protein feed resulted in significantly less fish production.

Field research is currently underway to confirm the previous year conclusion that feeding 12% less of a 32% protein feed to catfish in production ponds resulted in the same fish yield as did feeding a 28% protein feed to satiation. A third treatment was incorporated to determine if fish fed the 28% protein feed to satiation have the same fish yield as fish fed the same quantity of 28% at higher density. All ponds were stocked with 15 g and 700 g fish at an approximate ratio of 8:1 to also determine if feeding regime affects size-related feed competition. Water quality parameters being monitored are water temperature, dissolved oxygen, ammonia, pH, nitrite, soluble phosphorus, chlorophyll, and COD concentrations.

Phosphorus budgets were prepared for channel catfish ponds that received one of five diets ranging from 0.60 to 1.03% phosphorus. Fish production did not differ ($P > 0.05$) among diets. There were few differences among treatments with respect to soluble reactive phosphorus, total phosphorus, and chlorophyll *a* concentrations or gross primary productivity. Phosphorus loss in effluents when ponds were drained for harvest did not differ among treatments ($P > 0.05$). Phosphorus removed from ponds in fish at harvest and the amounts of phosphorus adsorbed by bottom soils increased as dietary phosphorus concentration increased ($P < 0.05$). Low-phosphorus diets did not decrease phytoplankton productivity or improve effluent quality. Uptake of phosphorus by bottom soils is a major factor controlling phosphorus concentrations in pond water. Low-phosphorus diets can be beneficial in catfish pond management by reducing the phosphorus load to bottom soils and conserving their ability to adsorb phosphorus.

Three diets with different concentrations of crude protein were tested in channel catfish ponds. Fish were fed to satiation with 28% crude protein feed, and the other two diets (32% and

36% crude protein) were applied in amounts calculated to provide the same crude protein input as for the 28% crude protein diet. Using this practice, feed application was smaller as feed crude protein increased, and phosphorus and organic matter loads to the ponds decreased accordingly. The feeding practice and diets used in this study had no measurable effects on nitrogen concentrations in pond waters and effluents or on fish production. In spite of smaller phosphorus inputs with 32% and 36% protein feed, only a small fraction of the applied phosphorus remained in the water column, and the differences in phosphorus input in feeds among treatments did not affect pond water or effluent phosphorus concentrations. When fish ponds are drained for harvest, the quality of effluent did not change until about 75% of the water had been released. Water quality in effluents then deteriorated because the pond bottom was disturbed by outflowing water, fish activity, and harvest. By holding the last 25% of water in ponds for 12 to 24 h after fish removal, much of the suspended matter was removed by sedimentation. The water can then be released slowly to prevent resuspension of sediment, and a better quality effluent obtained.

A study of the biochemical oxygen demand (BOD) of waters from ten channel catfish ponds at Auburn, Alabama, revealed that the 5-d BOD (BOD_5) seldom exceeded 8 mg/L and that the ultimate BOD (BOD_u) was usually less than 30 mg/L. Water samples from catfish ponds usually needed to be diluted only 2 or 3 times to permit BOD_5 measurements, and nitrification occurred even during a 5-d incubation period. Catfish pond waters were not extremely high in ammonia nitrogen concentration, and ammonia nitrogen introduced in the ammonium chloride-enriched dilution water caused an appreciable increase in BOD of some samples. Plankton respiration is a major component of carbonaceous BOD (CBOD) in catfish pond waters. Thus, the BOD is not expressed as rapidly during 5-d incubations

as in typical wastewater. The ultimate BOD (BOD_u) would be a good measurement of oxygen demand for catfish pond effluents, but it is difficult to measure. Data from this study suggest that BOD_u can be estimated from BOD_5 , but the correlation is not strong ($R^2 = 0.62$). An alternative is to develop a short-term BOD measurement specifically for effluents from channel catfish and other aquaculture ponds. This study suggests that a 10-d BOD conducted without nitrification inhibition or addition of ammonia nitrogen in dilution water might be a better alternative to standard BOD_5 or BOD_u measurements normally used in wastewater evaluation.

Mississippi State University, Starkville. Two experiments were conducted in flow thru aquaria with sunshine bass at two water temperatures (26.7 and 32.3°C). Six semipurified diets were prepared which contained three protein levels (45, 40 and 35%) and two lipid levels (5 and 15%) to yield varying dietary energy/protein ratios of 6, 7, 8, 9, 10 and 11 kcal/g protein. Fingerling sunshine bass (about 3-4 g/fish) were randomly distributed at a rate of 25 fish /tank. Triplicate groups of fish were randomly assigned to each diet and fed to satiation for 8 weeks. Overall growth and nutrient utilization values were significantly higher for fish maintained at 26.7°C compared to fish kept at 32.3°C. Feed consumption decreased with increasing dietary E/P ratio. All the responses except hepatosomatic index (HSI) had the same pattern at both temperatures. Feed efficiency (FE), protein efficiency ratio (PER) and protein conversion efficiency (PCE) were highest at a dietary E/P ratio of 9 kcal/g protein. Whole body lipid deposition and intraperitoneal fat (IPF ratio) accumulation were increased with increasing dietary lipid levels. At these two temperatures HSI changed differently but HSI correlated with liver glycogen levels at both temperatures. At 32.2°C, liver glycogen deposition was positively

correlated with dietary carbohydrate levels. The lower energy conversion efficiency of fish held at 32.2°C indicates an increased energy requirement for maintenance and/or activity in these fish. We are unable to explain the reduced growth and nutrient utilization by the fish maintained at the elevated temperatures. Perhaps stress effects of high temperatures, associated with release of heat shock protein and/or metabolic changes mediated thru isoenzyme shifts may be responsible.

Several attempts were carried out to develop an assay to detect and characterize heat shock protein from different tissues of sunshine bass. Tissue proteins were extracted and isolated using SDS-PAGE and subjected to a western blot technique to identify HSP-70. Proteins isolated from tissues of sunshine bass did not cross react with mouse monoclonal antibody specific for the inducible form of HSP-70 (SPA 810), from Stress-Gen Biotech Corp., Canada. However, these proteins did cross react with monoclonal anti-heat shock protein from Sigma (Clone BRM-22), that recognizes both constitutive and induced forms of HSP-70. Further studies showed tissue extracts from sunshine bass cross reacted with HSP-70 rabbit polyclonal antibody (SPA-812) from Stress-Gen Biotech Corp. which is specific for the induced form. Many attempts were conducted to identify HSP-70 in red blood cells from sunshine bass but were not successful. Several other tissues (liver, gill, gut and brain) were studied and found to contain heat shock protein (HSP-70). However, we have been unable to detect significant differences in HSP-70 protein bands between heat stressed and control groups of fish. This could be due to our heat shock procedure (induction temperature, timing of the sampling) and also lack of sensitivity of our detection technique compared to a radio-labeled assay technique. Studies are underway to clarify the temperature which triggers HSP synthesis in sunshine bass and determine the best time for sampling.

An experiment was conducted to identify changes in certain enzyme or isoenzyme activities at elevated temperatures in sunshine bass. Liver esterase was found to be present in two isoenzyme forms at 25°C compared to only a single form at the higher (32°C) temperature.

An experiment performed to determine the maintenance protein and energy requirement was not successful due to a disease outbreak and associated mortality of fish. A preliminary study was conducted to determine metabolic reserve utilization during continuous starvation of sunshine bass at 25°C and 30°C. Lower energy retention was observed in fish kept at 30°C. This observation agrees with the findings of our initial growth study which showed an increased energy requirement for maintenance at the higher temperature.

Texas A & M University. Three studies have been conducted with hybrid striped bass to investigate a variety of dietary manipulations on growth and nutrient utilization. Two different feeding trials have been completed with hybrid striped bass in which the effects of dietary lipid level and carnitine supplementation were evaluated. Providing dietary lipid at 10% or 15% rather than 5% or 20% significantly enhanced weight gain of hybrid striped bass but dietary carnitine supplementation did not influence growth, nutrient utilization or body composition. In another study, dietary supplementation of a commercial proteolytic enzyme was evaluated with hybrid striped bass. This diet additive did not enhance fish growth in two separate feeding trials, nor did it increase nutrient digestibility or limit waste production. In a third study, supplementation of cholesterol and lecithin to the diet was found to have negligible effects on growth and body composition of hybrid striped bass.

Objective 2a. *Optimize feeding strategies in relation to water temperature for channel catfish production. Of particular interest is delineation of more precise feeding strategies when water temperatures are cool (spring, late fall) and extremely hot (late summer, early fall).*

Mississippi State University, Stoneville. A pond study was conducted to evaluate effects of feeding strategies related to water temperature on optimizing nutrient utilization and reducing waste for food-size channel catfish. In March 1997, two sizes of channel catfish were stocked into 28, 0.4-ha (1.0-acre) earthen ponds at a rate of 24,700 fish/ha (10,000 fish/acre). After a one-month conditioning period, fish were fed to satiation with a 28% protein feed once every day, once every other day, or once every third day based on water temperature. Total nitrogen, total ammonia nitrogen, nitrite, nitrate, chloride, chlorophyll *a* and pH were measured monthly. All fish were harvested in December 1997 and samples taken for determination of dressout and fillet composition. Fish fed every day (treatment 1) throughout the growing season consumed the most feed and had the highest net production. Net production of fish that were not fed every day either in spring and fall (treatment 2) or during extremely hot summer (treatment 6 and 7) was not significantly different from that of fish fed every day except for treatment 3 in which fish were fed fewer days in the spring than fish in treatment 2. However, net production was significantly lower for fish that were not fed every day in spring and fall and during the extremely hot summer (treatments 4 and 5). Net production, feed consumption (feed input), feed conversion ratio, visceral fat, and total ammonia-nitrogen were positively correlated to number of days fish were fed. Net production, feed conversion ratio, visceral fat, and nitrite-nitrogen were positively correlated to feed consumption or feed input. No significant differences were observed in mortalities (based on daily recorded mortalities) and body proximate composition among treatments. Based

on these results it appears that catfish should be fed daily for maximum production. Impact of feeding strategies according to water temperature on water quality appears to be minimal although some significant differences were found among treatments. If catfish are fed to satiation daily from spring to fall and care is given to avoid waste of feed, they appear to reduce feed intake automatically during cool and extremely hot temperatures.

A second experiment was conducted to evaluate effects of diet composition (dietary protein and dietary protein-energy ratio) and feeding frequency (every day, every other day, or every third day) based on water temperature on optimizing nutrient utilization and reducing waste in channel catfish farming. In April 1998, two sizes of channel catfish were stocked into 28 0.4-ha (1.0-acre) earthen ponds at a rate of 24,700 fish/ha. After a one-month conditioning period, fish were fed to satiation with diets containing different protein levels and protein-energy ratios once every day, once every other day, or once every third day based on water temperature. All fish were harvested in November 1998. Data collection and analyses of water quality and fish samples were the same as described for 1997. Fish fed a 32% protein diet daily (treatment 1) had a lower net production than fish in treatment 4 (fish were fed a 28% protein diet every other day in the spring and fall at water temperatures below 26°C and fed a 26% protein diet during the rest of the growing season) and treatment 6 (fish were fed a 35% protein high energy diet every third day in the spring and fall at water temperatures below 26°C and fed a 28% protein diet during the rest of the growing season). This may have been

caused by a higher mortality observed for fish in treatment 1. Net production was positively correlated to feed consumption or feed input. Feed conversion ratio was positively correlated to number of days fish were fed and the feed input. Visceral fat and chlorophyll *a* concentrations were positively correlated to feed consumption. Net production, feed consumption, visceral fat, dressout, and water quality parameters were not correlated to number of days fish were fed, which appears to contradict results from the 1997 study. Reasons for the different responses between years may be related to the fact that several different diets were used and a shorter period for water temperatures below 26°C was observed in 1998 (37 days) as compared to that in 1997 (63 days). No significant differences were observed in proximate composition of fillet samples among different treatments. Data from the 1998 study confirm those from the 1997 study in which feeding strategies based on water temperatures do not markedly affect water quality. Until the data from the 1998 study are confirmed or refuted in future studies, we recommend daily feeding of either a 28% or a 32% protein diet during the growing season.

A third experiment was conducted to evaluate feeding strategies on optimizing nutrient utilization and reducing waste in channel catfish production. The feeding schedules follow:

- (1) daily;
- (2) 5 days on - 2 days off;
- (3) 4 days on - 3 days off;
- (4) 7 days on - 3 days off;
- (5) 6 days on - 1 day off;
- (6) 4 days on - 1 day off.

In March 1999, fingerling channel catfish were

stocked into 28, 0.4-ha (1.0-acre) earthen ponds at a rate of 24,700 fish/ha. After a one-month conditioning period, fish were fed to satiation with a 28% protein diet once daily according to the feeding schedules. All fish will be harvested in November or December 1999. Data collection and analyses of water quality and fish samples will be the same as described for 1997.

The University of Memphis. No effects on confinement stress on fish fed different feeding regimes in ponds were detected, however, an experimental design that will insure fish in all treatments were treated the same was difficult to develop. An experimental design has been developed to compare confinement stress responses between fish held on diurnally changing temperatures with fish held at constant temperature at the extremes of the diurnal oscillation. The diurnal change is about 10°C and the change occurs over 10 hours and then held for two more hours when the change to the other temperature is begun. The confinement stress test is conducted during the two hours of stable temperature at the end of each (cold and warm) cycle. Two experiments have been conducted using oscillations of 15.5 to 26.5°C and 24 to 35°C.

Texas A & M University. Two feeding trials have been conducted with channel catfish initially measuring 10-12 cm in length to investigate the effects of spring and fall photoperiod and temperature as well as dissolved oxygen on feed intake. Based on results of this study, a model was developed to describe the combined effects of temperature and dissolved oxygen in predicting feed intake of channel catfish to optimize feeding schedules and increase the efficiency of feeding.

Objective 2b. *Evaluate alternative feeding strategies including manipulation of diet composition in relation to such variables as water temperature and fish size for channel catfish, baitfish, and hybrid striped bass (sunshine bass).*

North Carolina State University. Pond trials have been conducted to evaluate the effects of feeding frequency on hybrid striped bass fingerling production. Feeding frequencies of two, three, and four times per day were tested. Reducing daily feeding frequency from three or four times per day to two times per day had no effect on total production or size distribution of fingerlings.

A second pond trial was conducted to evaluate the effect of time of day of feeding on the production of HSB foodfish. Preliminary results indicate that fish fed once per day either in the early morning or late evening had higher total production, average weight, and were more uniform in size distribution than fish that were fed either during mid-morning or mid-afternoon.

A final pond study was conducted to definitively establish the optimum feeding frequency for HSB fingerlings. Frequencies of one, two, and four times per day were evaluated to determine the effect of feeding frequency on overall production, feed conversion and size distribution. Total production, average weight and feed conversion efficiency were significantly improved when fish were fed twice per day versus once per day. Increasing feeding frequency to four times per day did not improve measured production variables.

RESULTS AT A GLANCE...

Reducing the frequency of feeding from three or four times a day to two times a day can save on labor costs and equipment wear when raising hybrid striped bass fingerlings. Feeding in early morning or late afternoon results in best feed conversion, uniformity of fingerling size, and total production.

A series of tests to determine the acute toxicity of ammonia and nitrite to different life stages was conducted. The 96-h LC50s were determined for eggs, larvae, 1-month old juveniles, 4-month old juveniles, and 18-month old adult fish. In general, the egg stage is the most tolerant of either ammonia or nitrite. The larval stage was the least tolerant to ammonia with tolerance increasing by the 4-month old juvenile stage. Larval HSB were highly tolerant to nitrite but this tolerance declined rapidly by the 1-month old juvenile stage.

The University of Arkansas at Pine Bluff. Diets containing different lipid sources are being evaluated in terms of their effect on standard performance measures (growth, survival, feed efficiency) and also indices of stress response (cortisol, glucose, chloride). A series of aquaria experiments using purified diets with different lipid sources has been conducted. Aquaria studies: Five purified diets per trial were formulated to be identical with the exception of the type of lipid(s) used. In trial 1 the lipid sources were: soybean oil (SBO), cod liver oil (CLO), equal amounts of SBO and CLO (SBO + CLO), canola oil (CAN) or olive oil (OO). In trial 2 the same types and amounts of lipid were used, but prior to addition of the oils the casein, gelatin, dextrin, Celufil and carboxymethylcellulose were extracted with boiling ethanol to remove residual lipid.

Fish in both trials were fed to satiation twice daily and weighed every three weeks. After six weeks, the feeding trials were terminated due to disease problems. A t-test conducted on six-week data from the two feeding trials revealed that there were significant differences in weight gain of fish fed the non-extracted versus extracted diets ($P = 0.002$). Weight gain was higher in fish fed the extracted diets. Analysis of variance revealed that there were no differences in weight gain of fish fed non-extracted diets with different lipid sources. However, among fish fed extracted diets weight gain was highest in fish fed the SBO+CLO and SBO diets versus those fed diet CLO, CAN or OO ($P = 0.05$). Survival did not differ among treatments.

Total lipid and fatty acid analysis of the diets was completed. Diets with n-6 to n-3 fatty acid ratios of 2.1 (CLO+SBO) to 7.0 (SBO) promoted fish growth best, while diets with ratios far below (CLO,0.3) or above (OO, 148; CAN, 198) this range resulted in reduced growth.

An additional feeding trial was conducted in aquaria using the same diet formulations for a longer period (eight months). The results were not consistent with the first two trials. Weight gain was highest in fish fed non-extracted diets with olive oil or cod liver oil alone and lowest in fish fed diets with soybean oil or canola oil alone. Survival was lowest in fish fed non-extracted diets with soybean oil or cod liver oil alone. There were no differences in weight gain of fish fed non-extracted or extracted diets regardless of lipid source. However, survival of fish fed ethanol-extracted diets was significantly higher than that of fish fed non-extracted diets, regardless of lipid source.

An outdoor feeding trial was performed (June-November 1998) to test the effects of practical diets with different lipid sources on performance of golden shiners in fertilized pools. Diets with soybean oil, cod liver oil or cottonseed oil alone, or a 50/50 mix of cod liver and soybean oils were tested. Weight gain of shiners fed diets with cod liver oil alone was significantly higher than that of fish fed diets with soybean oil or cottonseed oil at eight weeks, but the differences were not significant by 12 weeks. Twelve-week data also showed a negative correlation (-0.88) between

weight gain and survival, indicating that density-dependent growth may have masked diet effects even though all diets were offered in slight excess (4% body weight daily).

A feeding trial of golden shiners in ponds fed diets with 4 or 13% lipid as poultry fat or 13% lipid as menhaden fish oil is in progress. Subsamples have not revealed any differences in individual weights of fish fed different diets but the experiment will continue until the end of September. A companion trial comparing performance of juvenile goldfish fed diets with 4 or 13% poultry fat or menhaden oil is being conducted also in fertilized pools. The different lipid levels were incorporated into the experimental design in response to industry interest.

The University of Memphis. Golden shiners from the aquarium studies described above (UAPB) were subjected to a crowding stress. Samples were taken before, after two hours of low water crowding and two hours after the water levels were restored. The fish were very small which limited the blood sample volume. Some of the samples were combined and all samples could not be measured for cortisol and

RESULTS AT A GLANCE....

Feeds for baitfish production should be formulated with soybean oil, which was a much better source of lipid than canola, cod liver or olive oils.

electrolytes. However, no effect of diet on the stress response was detected.

An aquarium study was conducted on large golden shiners fed 8% lipid as soybean oil, cod liver oil, soybean oil plus cod liver oil, or cottonseed oil for about one month. Each diet was fed in duplicate. The system was supplied with flow through water and temperature ranged from 25 to 3° C. A confinement stress similar to that above was conducted after feeding the test diets for six weeks. Sufficient blood samples were taken and the analyses are in progress.

Louisiana State University, Baton Rouge and Rice Research Station. Several studies investigating inexpensive, locally available feedstuffs for crawfish have been completed by Louisiana Agricultural Experiment Station researchers. Feeding trials conducted in microcosms and outdoor fiberglass pools that simulated pond culture environments resulted in average crawfish growth responses 7 to 72 % and 30 to 173% greater when crawfish were fed supplements of rough rice seed (hull on) and whole raw soybeans, respectively, than when crawfish fed from the cultivated rice forage system alone. Average final weights for crawfish fed agricultural feedstuffs were 60 to 103% of

those fed formulated 25% crude protein crustacean feed and total yield averaged 86 to 103% of that achieved with the formulated feed. Data from field studies in earthen ponds were highly variable. It was found in one study that feeding (three days/week) while trap harvesting negatively impacted the catch, most likely by the presence of feed interfering with the effectiveness of the baited trap. In a second and third study, limiting feeding to one day per week following the last harvest day of the week failed to generate significant differences in yields, although supplements of soybean or soybean plus rice tended to provide the greatest quantity of large crawfish. In a fourth study, average total yield was significantly lower when crawfish received supplemental feeds (soybeans), although crawfish size-at-harvest was greater for that treatment. Supplemental feeds sometimes had a significant effect on sparing rice forage. It has become apparent that under field conditions when harvesting is accomplished by baited traps, supplemental feeding of crawfish to excess (even once per week) may not be conducive to increased yields. Furthermore, although economic data has not been fully summarized, it appears that the increased size of crawfish at harvest (i.e., increased revenues) may not always justify the cost of feeding, even with low-cost agricultural feedstuffs.

Objective 3. *Develop publications to effectively extend information derived from this project to feed manufacturers and fish producers.*

See list of publications on pages 44-46.

WORK PLANNED

Channel Catfish

Louisiana State University, Baton Rouge.

To date, the project has progressed as described in the original proposal, with one unanticipated problem. On an evening in July, 1999, a lightning strike near our research station caused a power outage to the catfish ponds involved in this project. Power was not restored by the utility until the following morning, which resulted in a large loss of fish in our production ponds due to low oxygen levels. This effectively wiped out production for the third year of the study. The LSU Agricultural Center is pursuing reimbursement from the utility for funds expended in the third year of the project, so the experiment can be repeated in the next (March-October, 2000) production season under a 12-month, no-cost extension of the SRAC project. This constitutes the only significant change in the proposed procedures or activities associated with this project.

Texas A & M University. The pond feeding trial in which lysine supplementation is being evaluated will continue through September of 1999. Additional feeding trials are planned with channel catfish to further investigate the effects of temperature and dissolved oxygen as well as fish size on feed intake to augment the model being developed for improving feeding schedules for channel catfish.

Auburn University. Due to the retirement of Dr. Richard T. Lovell the nutrition component of the channel catfish effort will be completed by Dr. T. J. Popma. No changes in design or progress are expected by this change.

The University of Memphis. The experimental design problems with the catfish component have been overcome by a design which compares

the stress response among fish held on diurnally changing temperatures and fish held at constant temperature. Experiments which use spring, summer and fall diurnal high and low temperatures will be done. Fall and summer regimes have been completed and the samples are being analyzed.

Hybrid Striped Bass (Sunshine Bass)

Texas A & M University. The effects of dietary supplementation of exogenous digestive enzymes on nutrient utilization of hybrid striped bass are currently being investigated in a series of digestibility trials.

Mississippi State University, Starkville.

Further studies are planned to determine the induction temperature of HSP synthesis in sunshine bass and the duration of exposure required to elicit a significant response in target tissues. We also plan to investigate changes in various enzyme activities, such as malic dehydrogenase, due to acclimation of fish to different temperatures and investigate changes in the utilization of metabolic reserves at elevated temperature by analyzing whole-body composition and tissue composition of starved fish. This should give some insight how fish utilize body reserves and what metabolic shifts occur when fish are held at high temperatures.

Baitfish

The University of Arkansas at Pine Bluff. Proximate analysis of fish and feeds is underway as is analysis of water quality data.

The University of Memphis. Previous problems with small fish size have been overcome by using large golden shiners fed experimental diets in aquaria. We have been able to feed the

diets for about a month before administering the stress tests.

Crawfish

Louisiana State University. The field research under this project for this objective has been completed. For the remainder of the project period, efforts will be directed at further

summarizing, analyzing, and interpreting the data. The information will then be disseminated to fellow scientists, extension personnel, and practicing or prospective crawfish producers. The conclusions derived from this research will also be useful in determining the need and direction for further research regarding the practice of supplemental feeding of crawfish.

IMPACTS

Channel Catfish

- Reduction of the protein composition of fish feed to 25 or 26% without supplemental lysine should result in reduced feed costs and improve nutrient utilization. This reduction may be best achieved by decreasing the protein content by specifically decreasing the dispensable amino acid content.
- A cost savings of as much as 5% of feed production may be achieved by using an all-plant material protein source rather than animal origin protein. Vitamin and mineral supplementation may be required but sufficient methionine is apparently supplied by plant material protein.
- An alternate feeding strategy which has shown promise is to use lower feeding rates of a higher protein feed. This feeding practice is thought to result in less wasted food.
- Reducing the phosphorus content of catfish feed has little influence on water quality, but will conserve the

phosphorus adsorbing capacity of bottom soils. When ponds are drained, water should be released slowly to avoid disturbing the pond bottom and releasing sediment into the effluent. Feeding diets with different protein content had little impact on water quality, including ammonia nitrogen and BOD.

- Feeding strategies based on water temperatures do not markedly affect water quality or fish production. Daily feeding to satiation regardless of the protein content of the diet appears to be essential to achieve maximal production. The most important factor is carefully feeding to satiation every day. There is a possibility of predicting feed intake with a model using the interaction of temperature and dissolved oxygen. Such a model might increase the planning or efficiency of feeding schedules.

Hybrid Striped Bass

- Sunshine bass grow less and exhibit poorer nutrient utilization when reared at elevated water temperatures. Sunshine bass farmers should consider

this if they plan to culture this species in areas where the water temperature may exceed 32°C. The role of heat shock proteins in this response is still uncertain.

- Feeding 10 or 15% fat diets significantly enhanced weight gain over fish fed 5 or 20% fat. Supplementing the diet with carnitine, commercial proteolytic enzymes, cholesterol or lecithin had negligible effects on growth and body composition.
- Reducing feeding frequency from three or four times a day to two times a day can save on labor costs and equipment wear. Feeding in early morning or late afternoon appears to improve feed conversion and total production.

Golden Shiners

- Diets with n-6 to n-3 fatty acid ratios of 2.1 to 7.0 promoted growth better than those with low (0.3) or high (148 to 198) fatty acid ratios.

Soybean oil was a much better source of lipid than canola, cod liver or olive oils.

Crawfish

- The main impact has been to create awareness, among both scientists and producers, for both the potential benefits of supplementing crawfish in established forage systems of earthen ponds and the potential detriments of feeding under some conditions. Low cost agricultural feedstuffs such as whole raw soybeans and rice grains have been shown to be readily consumed by crawfish and contribute to increased growth. However, this project has demonstrated the practical problem whereby introduced feeds can interfere with the effectiveness of baited traps in harvesting crawfish. Furthermore, the results of this project indicate the need for, and possible direction of, further research to address the logistics of supplemental feeding in production systems that utilize baited-traps as the sole means of harvesting.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

Publications in print

- Boyd, C. E. and A. Gross. 1999. Biochemical oxygen demand in channel catfish *Ictalurus punctatus* pond waters. *Journal World Aquaculture Society* 30:349-356.
- Cho, S. H. 1998. Variable feed allowances with constant protein input in channel catfish ponds. Ph.D. Dissertation, Auburn University, Alabama.
- Gross, A., C. E. Boyd, R. T. Lovell, and J. C. Eya. 1998. Phosphorus budgets for channel catfish ponds receiving diets with different phosphorus concentrations. *Journal World Aquaculture Society* 29:31-39.
- Keembiyehetty, C. N. and R. P. Wilson. 1998. Effect of water temperature on growth and nutrient utilization of sunshine bass (*Morone chrysops* x *Morone saxatilis*) fed diets containing different energy/protein ratios. *Aquaculture* 166: 151-162.

- Lochmann, R. T. 1999. Diets of cultured fish. *Lab Animal*, 27(1): 36-39.
- McClain, W. R., R. P. Romaine, J. J. Sonnier, and K. R. Taylor. 1997. Effects of supplemental feeding on the harvests of procambarid crawfish in ponds. Annual Research Report, Rice Research Station, Louisiana Agricultural Experiment Station, LSU Agricultural Center 89:411-414.
- McClain, W. R., K. R. Taylor, and J. J. Sonnier. 1996. Effects of supplemental feeds on crawfish growth in microcosms. Annual Research Report, Rice Research Station., Louisiana. Agricultural Experiment Station, LSU Agricultural Center 88:548-551.

Manuscripts

- Buentello, J. A., W. H. Neill, and D. M. Gatlin, III. In press. Effects of water temperature and dissolved oxygen on daily feed consumption, feed utilization and growth of channel catfish (*Ictalurus punctatus*). *Aquaculture*.
- Gaylord, T. G. and D. M. Gatlin, III. Submitted. Effects of dietary carnitine and lipid on growth and body composition of hybrid striped bass (*Morone chrysops* x *M. saxatilis*). *Fish Physiology and Biochemistry*.
- Gaylord, T. G. and D. M. Gatlin, III. Submitted. Dietary lipid but not carnitine affects growth and body composition of hybrid striped bass (*Morone chrysops* x *M. saxatilis*). *Aquaculture*.
- Gross, A., C. E. Boyd, and R. T. Lovell. In press. Effects of crude protein concentrations in feed on quality of pond water and effluent in channel catfish culture. *The Israeli Journal of Aquaculture*.
- Harcke, J. E. and H. V. Daniels. In press. Acute toxicity of ammonia and nitrite to different life stages of reciprocal cross hybrid striped bass eggs and larvae. *Journal of the World Aquaculture Society*.
- Harke, J. E. 1998. Acute toxicity of ammonia and nitrite to different life stages of reciprocal cross hybrid striped bass (*Morone chrysops* x *M. saxatilis*). Master's thesis, North Carolina State University.
- McClain, W. R. and R. P. Romaine. In preparation. Relative contribution of different food supplements to growth of procambarid crawfish.
- McClain, W. R., R. P. Romaine, J. J. Sonnier, and K. R. Taylor. In press. Effects of supplemental feeding on the harvests of procambarid crawfish in ponds. Annual Research Report, Rice Research Station, Louisiana Agricultural Experiment Station, LSU Agricultural Center.
- Neill, W. H., E. L. Oberney, Jr., S. R. Craig, M. D. Matlock, D. M. Gatlin, III. In press. Estimating metabolism of fish (or shellfish) in aquacultural production systems. *Aquacultural Engineering*.
- Sealey, W. M., S. R. Craig, and D. M. Gatlin, III. In press. Dietary cholesterol and lecithin have negligible effects on growth and body composition of hybrid striped bass (*Morone chrysops* x *M. saxatilis*). *Aquaculture Nutrition*.
- Stone, N., E. Park, L. Dorman, and H. Thomforde. 1997. Baitfish culture in Arkansas: golden shiners, goldfish and fathead minnows. MP-386. Cooperative Extension program, University of Arkansas at Pine Bluff, Pine Bluff, Arkansas.
- Lochmann, R. T. and H. F. Phillips. Submitted. Nutritional aspects of health and related components of baitfish performance. Special Publication of the World Aquaculture Society.

Papers presented

- Boyd, C. E. 1998. Phosphorus chemistry in pond soils. Aquaculture '98, Las Vegas, Nevada.
- Boyd, C. E. 1998. Effluent management in channel catfish farming. Aquaculture '98, Las Vegas, Nevada.
- Boyd, C. E. 1999. Water quality in channel catfish farming, Aquaculture '99, Sydney, Australia.
- Buentello, J. A., D. M. Gatlin, III and W. H. Neill. 1999. Effects of water temperature, fish size and dissolved oxygen on daily feed consumption of channel catfish. World Aquaculture '99, Sydney, Australia.
- Gaylord, T. G. and D. M. Gatlin, III. 1998. Effects of dietary carnitine and lipid on hybrid striped bass. Twenty-sixth Fish Feed and Nutrition Workshop, Pine Bluff, AR.
- Gaylord, T. G. and D. M. Gatlin, III. 1999. The effects of dietary carnitine on growth and body composition of hybrid striped bass *Morone chrysops* x *M. saxatilis*. World Aquaculture '99, Sydney, Australia..
- Harcke, J. E. and H. V. Daniels. 1998. Acute toxicity of ammonia and nitrite to eggs, larvae, and juveniles of reciprocal cross hybrid striped bass. Aquaculture '98, Las Vegas, Nevada, February 15-19.
- Keembiyehetty, C. N., R. P. Wilson, C. Bradley. and D. S. Luthe. 1998. Investigations on summer temperature induced slower growth of hybrid striped bass fingerlings *Morone chrysops* x *M. saxatilis* Poster presented at 3rd annual Will Carpenter Adaptive Biotechnology Seminar series at Mississippi State University, 5-6 Nov. 1998.
- McClain, W.R. 1997. Relative contribution of different food supplements to growth of crawfish (*Procambarus clarkii*). 89th Annual Meeting, National Shellfisheries Association, Fort Walton Beach, Florida, April 1997.
- Romare, R. P. 1998. Evaluation of soybean grain and rice seed as supplemental feed for red swamp crawfish in pools. Aquaculture '98, Las Vegas, Nevada, February 15-19.
- Sealey, W. M. and D. M. Gatlin, III. 1998. Dietary cholesterol and lecithin have limited effects on hybrid striped bass. Twenty-sixth Fish Feed and Nutrition Workshop, Pine Bluff, AR.
- Sealey, W. M. and D. M. Gatlin, III. Evaluation of protein reduction and lysine supplementation of production diets for channel catfish, *Ictalurus punctatus*. Aquaculture America, 2000.



VERIFICATION OF RECOMMENDED MANAGEMENT PRACTICES FOR MAJOR AQUATIC SPECIES

Reporting Period

January 1, 1997 - August 31, 1999

| | | |
|----------------------|--------------|-----------|
| Funding Level | Year 1 | \$31,410 |
| | Year 2 | 77,525 |
| | Year 3 | 78,925 |
| | Total | \$187,860 |

| | | |
|---------------------|---|---|
| Participants | University of Arkansas at Pine Bluff (Lead Institution) | Carole Engle, David Heike, 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 | Jimmy Avery |
| | North Carolina State University .. | Harry Daniels, Steven Gabel, Michael Frinsko, Rebecca Dunning |

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| Administrative Advisor | Dr. Jack Bagent, Director Louisiana Cooperative Extension Service Louisiana State University Baton Rouge, Louisiana |
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PROJECT OBJECTIVES

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 an 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.

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), seven cages (four 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 Fishy '98 is being used to track feeding and other data on most of the production units since 1998. In West Alabama, four cooperators have harvested 110,266 pounds of hybrids and 398,070 pounds of channel catfish. These were all seined from ponds that were not drained and have been restocked in 1999. In Southeast Alabama, two cooperators harvested 140,004 pounds of hybrid catfish. Both ponds are watershed ponds and have been restocked. Three cages of channel catfish were harvested at the Wiregrass Substation, for a total weight of 4,604 pounds fish. These cages were restocked in spring 1999. In East Central Alabama, four cages have been harvested or partially harvested at this time, producing a total

of 4,032 pounds of fish and have been restocked.

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 is currently conducting 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 are being collected on a weekly basis, summarized weekly and posted on the Arkansas CYVT web site (www.uaex.edu/aquaculture/arcyvp.htm). In order to collect

two full production seasons of data on all sites, the trial will continue through the summer of 2000. In Desha County, two full fingerling production seasons have been completed and a third season currently is underway. Fry stocked in June, 1999, will be harvested as fingerlings through fall, 1999, and spring, 2000. This will complete the data collection phase of the fingerling verification trials.

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 1998 and terminated in June, 1999. Agents collected pre-production samples of water source, forage, and soil. Forage depletion was monitored monthly. Farmers were given recommendations on trap density, bait usage, and harvest regimes.

Problems developed concerning loss of labor that had been given primary responsibility of keeping records. At least two of the cooperators had harvesters quit during the height of the harvest season resulting in only partial records. Efforts are still underway to see if any of these records are salvageable or can be recreated. A better method of monitoring the collection of

field records needs to be developed.

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 is continuing on different production variables (feeding, aeration, labor, etc.) on a weekly basis. Five partial harvests have been completed.

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 lb. 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; and*

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

A joint project publication will also be published in the coming year that includes guidelines for infrastructure development, program implementation, and evaluation of aquaculture

management verification programs. Management protocols have been developed for use in all participating states and will be published in the near future.

WORK PLANNED

Alabama. Seven cages and six ponds will be harvested in 2000 and data recorded. The levee ponds in West Alabama will be drained and all fish harvested from them. The Southeast Alabama watershed ponds will be drained and all fish harvested, and data recorded. All cages under the CYVP program will be harvested. A summary of all information collected on the CYVP program will be written and submitted for the final report.

of program on producers will be analyzed during the summer of 2000.

Arkansas. All data (production inputs, water quality, stocking and harvesting) will be collected through the harvests scheduled for 2000. Foodfish and fingerling production ponds will be scrapped in early spring 2000. A comprehensive report will be prepared that summarizes all data collected over the project period for all ponds.

North Carolina. Complete harvest of the ponds is scheduled for late fall, 1999. An economic analysis of production costs will be done after the ponds are completely harvested and the final production data are available. Based on the data generated by this trial, a manual of recommended management practices for channel catfish farmers will be written during late fall, 1999.

Louisiana. The cooperators are committed to participating for two production cycles. The second production cycle will begin in October 1999. Yield, production economics, and impact

South Carolina. This was potentially an excellent project with great potential to help the catfish industry. Much work was put into establishing the system to collect data including a complete financial analysis of the current farm situation. However, in the middle of the project the owner fired the manager. A new manager was hired and he has now quit. The cooperator decided to sell out so he is no longer in business. Therefore, the participation by South Carolina in this project has been terminated.

IMPACTS

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.

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 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. 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. Although it is too early to quantify results of these trials, some of the cooperating producers are encouraged by their harvests and have begun to implement the recommended management protocols in the rest of their ponds.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

Papers presented

Heikes, D.L. 1998. Catfish Yield Verification Update. Poster presentation. Catfish Farmers of Arkansas Annual Convention, Little Rock, Arkansas. 22-23 January.

Heikes, D.L. and C. Davidson. 1998. Catfish Verification Web Site. Poster presentation. 1890 Extension System-Wide Conference, Atlanta, Georgia. 22-25 June.

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

Reporting Period

January 1, 1999- August 31, 1999

| | | |
|-----------------------------------|--|--|
| Funding Level | Year 1 | \$309,688 |
| | Year 2 (projected) | 281,986 |
| | Year 3 (projected) | 253,326 |
| | Total | \$845,000 |
| Participants | 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 |
| Administrative Advisor | Dr. Greg Weidemann, Associate Director Arkansas Agricultural Experiment Station University of Arkansas Fayetteville, Arkansas | |

PROJECT OBJECTIVES

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

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 practices 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*

University of Mississippi. Nearly 1,000 plant and algae extracts have been prepared from almost 500 collections of plants, cyanobacteria, and algae. These collections now include more than 200 samples of tropical cyanobacteria and marine algae, and 250 collections of aquatic and wetland plants, cyanobacteria (collections and cultures), and eukaryotic algae. These extracts are being evaluated in 96-well microtiter assays that select for blue-green algae selective algaecidal activity.

Initial extract evaluations were conducted using *Oscillatoria agardii*, as the cyanobacterial test organism, and *Selenastrum capricornutum*, as a chlorophyte control for non-specific algaecidal activity. These organisms are 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 algaecidal against *O. agardii*.

RESULTS AT A GLANCE...

Extracts from 91 plants and algae have been screened for algicidal activity against blue-green algae. Several of the extracts appear to be selectively toxic to odor-producing blue-green algae and will be further investigated as potential blue-green algicides.

While these results are promising, the cyanobacterium *O. agardii* is not known to produce odorous compounds that result in off-flavor problems in aquaculture ponds. We have since established cultures of the methylisoborneol-producing cyanobacterium *O. perornata* suitable for

high-throughput extract evaluation. Extracts from 91 collections of aquatic and marine cyanobacteria, plants and algae have subsequently been evaluated in replicate bioassays using *O. perornata*. Seventeen extracts were found to be strongly active and cyanobacterial-selective at the concentration of 100 ppm used for initial screening. Dose response data were obtained for these active extracts at half-log concentrations. Eight extracts were found to be effective at 30 ppm and two extracts remained both selective and effective at 10 ppm. 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 one of the green algal extracts has resulted in the partial purification of cyanobacterial-selective substances that are effective on against both *Oscillatoria* strains at concentrations in the ppb (parts per billion) range.

While these results are promising, the cyanobacterium *O. agardii* is not known to produce odorous compounds that result in off-flavor problems in aquaculture ponds. We have since established cultures of the methylisoborneol-producing cyanobacterium *O. perornata* suitable for high-throughput extract evaluation. Extracts from 91 collections of aquatic and marine cyanobacteria, plants and algae have subsequently been evaluated in replicate bioassays using *O. perornata*. Seventeen extracts were found to be strongly active and cyanobacterial-selective at the concentration of 100 ppm used for initial screening. Dose response data were obtained for these active extracts at half-log concentrations. Eight extracts were found to be effective at 30 ppm and two extracts remained both selective and effective at 10 ppm. 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 one of the green algal extracts has resulted in the partial purification of cyanobacterial-selective substances that are effective on against both *Oscillatoria* strains at concentrations in the ppb (parts per billion) range.

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

Mississippi State University. Eight 5.5-m³ (1450-gallon) enclosures were placed in an experimental earthen fish pond containing a dense phytoplankton community dominated by *Oscillatoria agardhii*. The N:P ratio of four enclosures was adjusted by addition of KNO₃ to provide a N:P of about 30 and chelated iron was added to provide 1 mg/L Fe. Four enclosures did not receive nutrient additions. All enclosures were supplied with diffused aeration to produce gentle mixing. Water samples collected every 2 to 3 days were analyzed for nutrients, solids, and indices related to phytoplankton biomass and community composition were measured by standard methods. 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.

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 mg/L for 30 days in soil-water systems treated with 1 mg/L iron from iron-EDTA. Thus, it was decided to use EDTA chelated metals for subsequent pond research in which (1) chelated iron plus chopped legume hay, and (2) chelated iron, chopped legume hay, plus a trace element mix, are being used as treatments for blue-green algae control. So far, results are not positive, and the use of legume hay has caused low dissolved oxygen concentrations in the ponds.

University of Arkansas, Pine Bluff. A study is currently in progress to evaluate the potential of two pond treatments (ammonium sulfate or sodium nitrate) on water quality and phytoplankton communities in 24 commercial golden shiner and goldfish ponds. Water quality and plankton community data are being collected monthly. As of mid-July, ponds in the sodium nitrate treatment appear to have better water quality than ponds in the other nitrogen treatment. Golden shiner ponds in the study are of two soil types and two age classes (20-25 years old and 40-45 years old). Cooperators at Auburn University have collected soil cores from representative ponds, and initial observations suggest a decline in soil quality as ponds age.

Louisiana State University. Water samples composited have been collected biweekly since March, 1999, from six locations in each of 18 experimental ponds at the Aquaculture Research Station stocked with catfish at commercial rates (25,000 fish per ha, or 10,000 fish per acre) and subject to low to moderate levels of mineralization (0, 1.5, or 3 ppt salinity). The water has been analyzed for the presence of 2-methylisoborneol (MIB) and geosmin using analytical techniques developed by USDA-SRRC scientists. Phytoplankton community composition and blue-green algal abundance are being enumerated in duplicate water samples by scientists at the Aquaculture Research Station-LAES.

Twenty-seven experimental 0.04-ha (0.1-acre) earthen ponds at the Aquaculture Research Station were renovated for use in experimental evaluations of the efficacy of threadfin shad and nutrient manipulation for control of blue-green algae in ponds stocked with channel catfish. Channel

catfish fingerlings and threadfin shad will be stocked in the experimental ponds in November after the danger of enteric septicemia catfish (ESC) disease has subsided. Shad will be stocked when water tem-

peratures cool in late November-early December to minimize physiological stress associated with capture from local lakes, transport, and stocking, in experimental ponds.

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

Louisiana State University. Twelve, 0.04-ha (0.1-acre) ponds were stocked with multiple cohorts of channel catfish at a nominal stocking density of 25,000/ha (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 500/ha (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 mg/L 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 62,500 juveniles per ha (25,000 per acre) with channel catfish, to evaluate their ability to control blue-green algae (biological control).

The eight treatment combinations, arranged in a 2³ factorial design, were randomly assigned to 24,

11.3-m³ (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 25,000 fish per ha (10,000/acre), fed a 32% crude protein commercial feed daily at rates ranging from 45 to 170 kg/ha (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 7990 kg/ha (7,133 pounds/acre), with no observed differences related to water management practices. Shad biomass averaged 700 kg/ha (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 evidence 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.

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

Auburn. In April 1999, ten 0.04-ha (0.1-acre) earthen ponds were stocked with 9-g channel catfish at a density equivalent to 15,000/ha (6,000/acre) and with 0.7-kg (0.3 pound) grass carp at 50/ha (20/acre). Five randomly selected ponds were also stocked with 8-g threadfin shad at 2,000/ha (800/acre). Each pond has subsequently been fed once daily to apparent satiation. Catfish survival has been apparently high and substantial threadfin shad recruitment has been observed. Samples have been collected every 2 weeks since 13 April 1999 for water quality and phytoplankton analyses. Data have been entered into computer spreadsheets and examined for accuracy.

University of Georgia. Threadfin shad or fathead minnows were stocked with catfish in 0.1-ha (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 0.04-ha (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 18,000/ha (44,500/acre). Threadfin shad were stocked at about 1,000/ha (2,500/acre) and were 4.5 to 10 cm (1.7 to 4 inches). Fathead minnows were stocked at about 10 kg/ha (3,600 to 4,000/ha, or 8,900 to 10,000/acre) and were 3.5 to 5 cm (1.4 to 2 inches).

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 4-5 cm (1.5 to 2 inch) shad from local ponds in the months of January to April. Even under the best conditions,

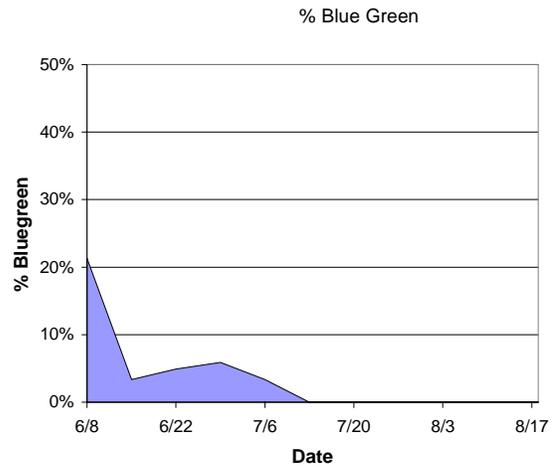
it is difficult to determine the survival of the threadfin shad after stocking. Stocking threadfin shad into holding ponds and seining after one or two weeks indicated that 30-90% of the threadfin shad could die a short time after stocking due to loss of scales during handling, temperature shock, alkalinity 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. An effort of 12 man-hours was required to capture 900 threadfin shad using cast nets compared to eight man-hours using a seine in the corners of large catfish ponds. Early morning seining is best during periods when the shad are swimming close to the shoreline.

Hauling aids should be utilized during transport and may include an anesthetic, sodium chloride, calcium chloride, antifoaming agents, or a buffer of pH to 7.0. Tempering should be extended to two hours of gradual exchange of hauling water with receiving water.

Over 50 algal species have been identified from Georgia ponds during the first part of the 1999 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 has been observed to date due to the limited number of sampling intervals at this time. Off-flavors were not detected to date in channel catfish harvested from this study.

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

Clemson University. The 0.8-ha (2-acre) commercial scale PAS unit was brought on-line in the spring of 1999. The unit was stocked with 67,829 catfish fingerlings in May of 1999. Fingerling sizes were 15, 31, 48, 61 and 80 g. The catfish were stocked in two raceways consisting of four sections each. In addition, 15,000 tilapia weighing 40 g each were stocked into the system. An additional 216 breeding tilapia were stocked into the “algal” section of the system. As of August 1999, secchi disk transparencies are in the range of 15-18 cm and the system is dominated by green algae (Chlorophyta). The tilapia biomass in the system has reached approximately 850 kg/ha (2,100 pounds/acre) or 21% of catfish biomass. Similar results are being obtained in six 0.12-ha (0.3-acre) PAS units.



Impact of tilapia stocking (as breeding pairs and fingerlings) in 2-acre PAS unit; percent blue green algae vs date.



Two acre PAS unit showing the fish raceway and algal basin paddlewheels.

WORK PLANNED

Work on all objectives is on schedule and no major changes in the project have occurred or are anticipated.

IMPACTS

Work to date indicate that natural products (small biologically active organic compounds) produced by organisms that live and compete in cyanobacteria-rich environments are a valuable source of new cyanobacterium-selective algacides and that these algacides may be of use in the control of blue-green algae in aquaculture ponds.

The research has also revealed that EDTA is an excellent chelating agent for iron (and presumably other metals) for use in pond aquaculture.

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 waste nitrogen and phosphorus discharges, which currently pose a eutrophication threat to surface and groundwater supplies.

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.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

Manuscripts

Nagle, D.G. and V.J. Paul. In preparation. Production of secondary metabolites by filamentous tropical marine cyanobacteria: ecological functions of the compounds. *Journal of Phycology*.

Papers Presented

Nagle, D. G., D.E. Wedge, K.K. Schrader, I. Rajbhandari, and C.J.G. Duncan. 1999. Algae and aquatic plants as sources of new agrochemicals. The American Society of Pharmacognosy Interim Meeting, Tunica, Mississippi.

Brune, D. E., J. A. Collier, T. E. Schwedler, A. G. Eversole, and M. D. Hammig. 1999. Water Reuse Technology in Freshwater Ponds: Partitioned Aquaculture. World Aquaculture Society Meeting, Sydney, Australia.

Brune, D. E., J. A. Collier, T. E. Schwedler, and A. G. Eversole. 1999. Designed Ecosystems for Aquaculture: The Clemson Partitioned Aquaculture System. ASAE Annual Meeting, Toronto, Ontario, Canada.



MANAGEMENT OF AQUACULTURAL EFFLUENTS FROM PONDS

Reporting Period

April 1, 1999 to August 31, 1999

| | | |
|-----------------------------------|--|----------------------------------|
| Funding Level | Year 1 | \$236,833 |
| | Year 2 (projected) | 237,785 |
| | Year 3 (projected) | 141,411 |
| | Total | \$616,119 |
| Participants | 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 |
| Administrative Advisor | Dr. Marty Fuller, Assistant Director Mississippi Agricultural and Forestry Experiment Station Mississippi State University Mississippi State, Mississippi 39762 | |

PROJECT OBJECTIVES

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 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).*

Preliminary tests have been conducted that indicate that 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 mg/L did not improve the efficiency of solids removal in initial trials.

The annual crawfish aquaculture production

cycle in Louisiana begins with planting of forages in summer, flooding production ponds in fall, cultivation of the crawfish crop in fall, winter, and spring, and draining of ponds in late spring through early summer. Six experimental crawfish ponds at the Louisiana Agricultural Experiment Station's Aquaculture Research Station permanent crawfish ponds in Baton Rouge and 16 ponds at the Rice Research Station rice-crawfish culture systems in Crowley were planted with rice in mid summer 1999 as crawfish forage using standard commercial practices. Rice was

fertilized and herbicides applied to stimulate rice growth. The experimental crawfish ponds will be flooded in October 1999 for crawfish production and determination of annual water budgets and effluent quality at the two experimental sites.

In North Carolina, a study evaluating different fertilization regimes is currently being conducted in experimental hybrid striped bass ponds. Data on water discharge quantity and quality is being collected from a commercial hybrid striped bass farm.

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

In Arkansas, farmer-cooperators are being identified and receiving streams located. Historical streamflow daily values for hydrologic unit sampling stations near baitfish farms in Lonoke County, Arkansas, have been obtained from USGS records. Stream flow generally is highest in December through April, with periods of little to no flow in June through August.

Water use data from golden shiner, goldfish and fathead minnow farms has been collected to document pre-project practices.

In Mississippi, seventeen stream sample locations were selected and geo-referenced within four sub-watersheds with variable development of the landscape to aquaculture ponds.

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.*

In Mississippi, preliminary work was completed for a study evaluating the effect of increasing the water storage capacity (depth) of one pond in an interconnected 2-pond or 4-pond module. Earth

work required for increasing pond depth, modification of outflow pipes, and addition of pond linkage pipes and open channel flumes has been completed.

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

Work on this objective will begin in Year 3 of the project.

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.

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.

In Louisiana, a part-time research associate has been employed to begin conducting literature searches and reviews on best management practices (BMPs) that can be applied to management of effluents discharged from crawfish aquaculture 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.

Work on this objective will begin in Year 3 of the project.

WORK PLANNED

This progress report represents less than 5 months of project activities. Given the start date of the project, progress during this year was minimal.

However, project participants have initiated studies related to all project objectives described above and work on all objectives is on schedule.

IMPACTS

None to date.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

None to date.



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 | 61,000 | 30,737 | -0- | -0- | -0- | 30,737 | 91,737 |
| | 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 | 74,000 | 47,000 | -0- | -0- | -0- | 47,000 | 121,000 |
| Total | | 291,400 | 198,397 | -0- | 1,000 | -0- | 199,397 | 490,797 |
| Management of Environmentally-Derived Off-flavors in Warmwater Fish Ponds | 1 | 251,200 | 69,389 | 42,000 | 49,500 | -0- | 160,889 | 412,089 |
| | 2 | 250,900 | 69,389 | 53,000 | 28,380 | 20,000 | 170,769 | 421,669 |
| | 3 | 230,900 | 38,329 | 34,000 | 58,483 | -0- | 130,812 | 361,712 |
| Total | | 733,000 | 177,107 | 129,000 | 136,363 | 20,000 | 462,470 | 1,195,470 |
| Optimizing Nutrient Utilization and Reducing Wastes Through Diet Composition and Feeding | 1 | 246,715 | 261,465 | -0- | -0- | -0- | 261,465 | 508,180 |
| | 2 | 258,370 | 263,627 | -0- | -0- | -0- | 263,627 | 521,997 |
| | 3 | 234,915 | 258,545 | -0- | -0- | -0- | 258,545 | 493,460 |
| Total | | 740,000 | 783,637 | -0- | -0- | -0- | 783,637 | 1,523,637 |
| Verification of Recommended Management Practices for Major Aquatic Species | 1 | 31,410 | 60,286 | 1,000 ¹ | -0- | -0- | 61,286 | 92,696 |
| | 2 | 65,525 | 78,686 | 1,000 ¹ | 5,000 ² | -0- | 84,686 | 150,211 |
| | 3 | 66,925 | 78,986 | 6,000 ³ | 5,000 ² | -0- | 89,986 | 156,911 |
| Total | | 163,860 | 217,958 | 8,000 | 10,000 | -0- | 235,958 | 399,818 |
| Control of Blue-green Algae in Aquaculture Ponds | 1 | 309,688 | 77,403 | 27,000 | 172,500 | 70,000 | 346,903 | 656,591 |
| | 2 | 281,986 | | | | | | 281,986 |
| | 3 | 253,326 | | | | | | 250,503 |
| Total | | 845,000 | 77,403 | 27,000 | 172,500 | 70,000 | 346,903 | 1,191,903 |
| Management of Aquacultural Effluents from Ponds | 1 | 236,833 | 105,319 | | | | 105,319 | 342,152 |
| | 2 | 237,785 | 116,842 | | | | 116,842 | 354,627 |
| | 3 | 141,411 | 109,092 | | | | 109,092 | 250,503 |
| Total | | 616,119 | 331,253 | | | | 331,253 | 947,282 |

¹ Catfish Farmers of Arkansas² Submitted to Sea Grant³ \$1,000 from Catfish Farmers of Arkansas; \$5,000 proposed from Louisiana industry.

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 Project Total | \$346,038 | 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 Project Total | \$150,000 | 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 Project Total | \$124,990 | 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 Project Total | \$13,771 | 88-38500-4028 |
| *Immunization of Channel Catfish. Dr. J. A. Plumb, Auburn University, Principal Investigator | Yr. 1-05/02/89-04/30/90 | \$50,000 | 88-38500-4028 |
| | Yr. 2-05/01/90-04/30/91 | 49,789 | 89-38500-4516 |
| | Project Total | \$99,789 | |
| *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 | \$46,559 | 88-38500-4028 |
| | Yr. 2-05/01/90-10/31/91 | 51,804 | 89-38500-4516 |
| | Project Total | \$98,363 | |
| *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 | \$274,651 | 88-38500-4028 |
| | Yr. 2-05/01/90-04/30/91 | 274,720 | 89-38500-4516 |
| | Yr. 3-05/01/91-12/31/92 | 273,472 | 90-38500-5099 |
| | Project Total | \$822,843 | |
| *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 |
| | Project Total | \$373,952 | |
| *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 | | |
| | Project Total | \$15,000 | 89-38500-4516 |
| *National Extension Aquaculture Workshop. Dr. Carole Engle, University of Arkansas at Pine Bluff, Principal Investigator | 10/01/91-09/30/92 | | |
| | Project Total | \$3,005 | 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 |
| Project Total | \$133,142 | | |
| *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 |
| Project Total | \$442,041 | | |
| *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 |
| Project Total | \$535,338 | | |
| *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 | \$11,392 | 93-38500-8393 |
| | Project Total | \$351,929 | |
| *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 | | |
| | Project Total | \$2,000 | 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 |
| | Total Yr. 3 | \$234,263 | |
| | Project Total | \$760,466 | |
| *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 |
| | Total Yr. 3 | \$100,798 | |
| | Project Total | \$332,993 | |
| Publications, Videos and Computer Software. Dr. James T. Davis, 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 | \$74,000 | 98-38500-5865 |
| | Project Total | \$291,348 | |
| *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>76,279</u> | 97-38500-4124 |
| Total Yr. 3 | \$230,900 | | |
| Yr.4-06/01/99-05/31/00 | \$80,900 | | |
| Yr. 5 - Projected | <u>\$56,100</u> | | |
| Project Total | \$868,869 | | |
| 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 | \$241,501 | 95-38500-1411 |
| | Yr.2-12/01/97-11/30/98 | \$47,105 | 95-38500-1411 |
| | | <u>210,062</u> | 96-38500-2630 |
| | Total Yr. 2 | \$257,167 | |
| | Yr.3-12/1/98-11/30/99 | \$26,225 | 96-38500-2630 |
| | | <u>208,690</u> | 97-38500-4124 |
| Total Yr. 3 | \$234,915 | | |
| Project Total | \$735,000 | | |
| *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 |
| | Project Total | \$3,700 | |
| 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>70,339</u> | 96-38500-2630 |
| | Total Yr. 2 | \$77,525 | |
| | Yr.3-01/01/99-12/31/00 | <u>\$78,925</u> | |
| Project Total | \$187,860 | | |
| *Project Completed | | | |

| Project | Duration | Funding | Grant No. |
|--|-------------------------|------------------|---------------|
| Control of Blue-green Algae in Aquaculture Ponds. Dr. Larry Wilson, University of Tennessee, Principal Investigator | Yr. 1-01/01/99-01/01/00 | \$25,147 | 96-38500-2630 |
| | | 105,167 | 97-38500-4124 |
| | | <u>179,374</u> | 98-38500-5865 |
| | Total Yr. 1 | \$309,688 | |
| | Yr. 2 - Projected | \$975 | 96-38500-2630 |
| | | 17,394 | 97-38500-4124 |
| | | 159,955 | 98-38500-5865 |
| | | <u>103,662</u> | 99-38500-7375 |
| | Total Yr. 2 | \$281,986 | |
| | Yr. 3 - Projected | <u>\$253,326</u> | |
| Project Total | \$845,000 | | |
| 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>128,303</u> | 98-38500-5865 |
| | Total Yr. 1 | \$228,303 | |
| | Yr. 2 - Projected | \$237,785 | |
| | Yr. 3 - Projected | <u>\$150,740</u> | |
| Project Total | \$616,119 | | |