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Annual Progress Report

North Central Regional Aquaculture Center

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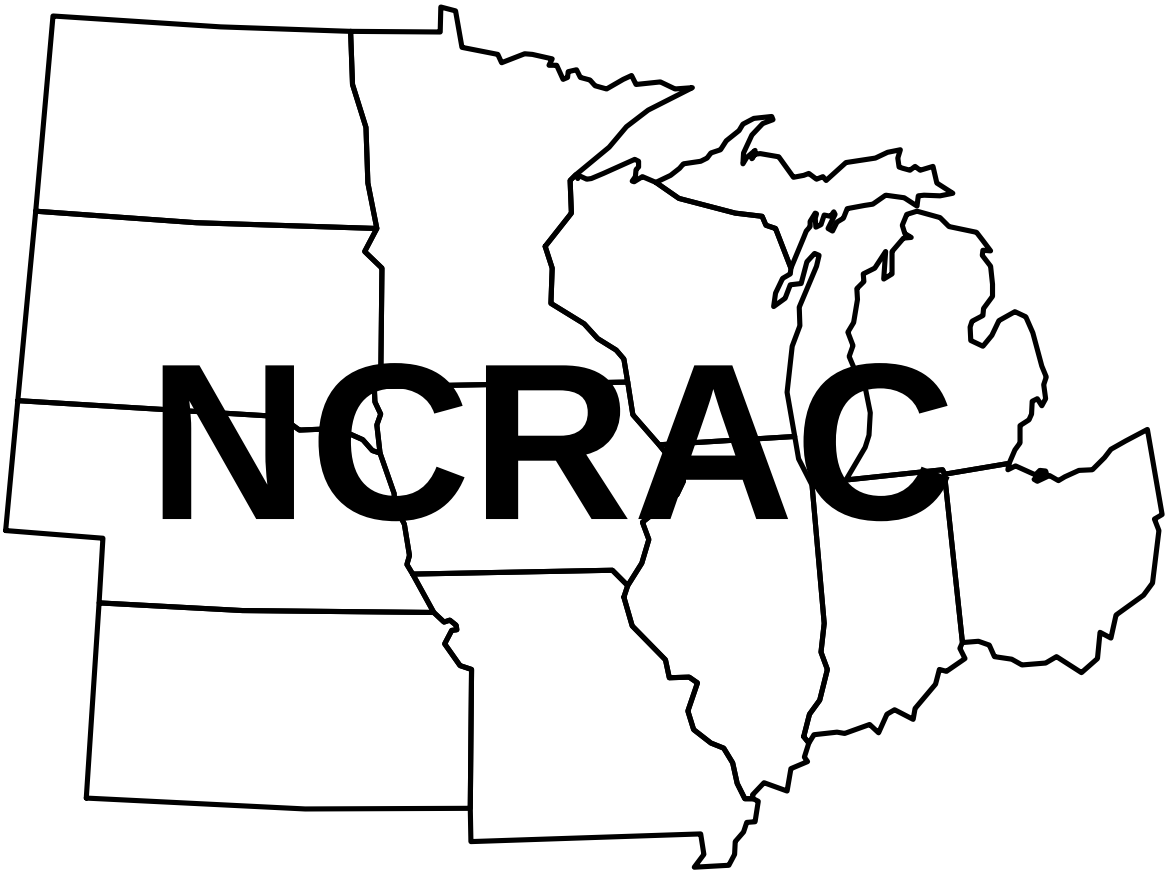
NORTH CENTRAL
REGIONAL AQUACULTURE CENTER



ANNUAL PROGRESS REPORT 1997-98

FEBRUARY 1999

ANNUAL PROGRESS REPORT

For the Period
September 1, 1997 to August 31, 1998

February 1999

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A table of commonly used abbreviations and acronyms can be found inside the back cover.

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INTRODUCTION

The U.S. aquaculture industry continues to be one of the fastest growing sectors within U.S. agriculture, although at a lesser rate than what occurred during the 1980s. Production in 1996 reached 694 million pounds and generated approximately $886 million for producers. The impact of U.S. aquaculture is substantial accounting for approximately 181,000 jobs and generating an estimated $5.6 billion annually. Yet, anticipated growth in the industry, both in magnitude and in species diversity, continues to fall short of expectations.

Much of what is known about aquaculture science is a result of institutional attention given to our traditional capture of wild fisheries with the goal of releasing cultured fishes into public waters for enhancement of declining public stocks. Despite extensive efforts to manage wild populations for a sustained yield, as a nation we consume substantially greater amounts than we produce. Much of the United States' demand for seafood has been met by imports. The U.S. imports over 40% of its fish and shellfish and, after Japan, is the world's second largest importer of seafood. Fisheries imports are the largest contributor to the U.S. trade deficit among agricultural products, and the second largest after petroleum, among all natural resources products. The value of imported fisheries products more than doubled during the 1980s and has continued to increase in the 1990s. In fact, the $14.5 billion value for 1997 was a record. In 1997, the trade deficit was $5.2 billion for all fisheries products, $5.0 billion of which was for edible fish and shellfish.

Landings for most commercial capture fisheries species and recreational fisheries of the United States have been relatively stable during the last decade, with many fish stocks being over exploited. In this situation, aquaculture provides an opportunity to reduce the trade deficit and meet the rising U.S. demand for fish products. A strong domestic aquaculture industry is needed to increase U.S. production of fish and shellfish. This can be achieved by a partnership among the Federal Government, State and local public institutions, and the private sector with expertise in aquaculture development.

Congress recognized the opportunity for making significant progress in aquaculture development in 1980 by passage of the National Aquaculture Act (P.L. 96-362). Congress amended the National Agricultural Research, Extension, and Teaching Policy Act of 1977 (P.L. 95-113) in Title XIV of the Agriculture and Food Act of 1981 (P.L. 97-98) by granting authority to establish aquaculture research, development, and demonstration centers in the United States in association with colleges and universities, State Departments of Agriculture, Federal facilities, and non-profit private research institutions. Five such centers have been established: one in each of the northeastern, north central, southern, western, and tropical/subtropical Pacific regions of the country. The 1996 Federal Agriculture Improvement and Reform Act (FAIR) (P.L. 104-127) otherwise know as the Farm Bill, has reauthorized the Regional Aquaculture Center program at $7.5 million per annum. As used here, a center refers to an administrative center. Centers do not provide monies for brick-and-mortar development. Centers encourage cooperative and collaborative aquaculture research and extension educational programs that have regional or national application. Center programs complement and strengthen other existing research and extension educational programs provided by the U.S. Department of Agriculture (USDA) and other public institutions. As a matter of

policy, centers implement their programs by using institutional mechanisms and linkages that are in place in the public and private sector.

The mission of the Regional Aquaculture Centers (RACs) is to support aquaculture research, development, demonstration, and extension education to enhance viable and profitable U.S. aquaculture production which will benefit consumers, producers, service industries, and the American economy.

The North Central Regional Aquaculture Center (NCRAC) was established in February 1988. It serves as a focal point to assess needs, establish priorities, and implement research and extension educational programs in the twelve state agricultural heartland of the United States which includes Illinois, Indiana, Iowa, Kansas, Michigan, Missouri, Minnesota, Nebraska, North Dakota, Ohio, South Dakota, and Wisconsin. NCRAC also provides coordination of interregional and national programs through the National Coordinating Council for Aquaculture (NCC). The council is composed of the RAC directors and USDA aquaculture personnel.

ORGANIZATIONAL STRUCTURE

Michigan State University (MSU) and Iowa State University (ISU) work together to develop and administer programs of NCRAC through a memorandum of understanding. MSU is the prime contractor for the Center and has administrative responsibilities for its operation. The Director of NCRAC is located at MSU. ISU shares in leadership of the Center through an office of the Associate Director who is responsible for all aspects of the Center's publications, technology transfer, and outreach activities.

At the present time the staff of NCRAC at MSU includes Ted R. Batterson, Director, and Liz Bartels, Executive Secretary. The Center Director has the following responsibilities:

- Serving as executive secretary to the Board of Directors, responsible for preparing agenda and minutes of Board meetings;
- Serving as an ex-officio (non-voting) member of the Technical Committee and Industry Advisory Council;
- Coordinating the development of research and extension plans, budgets, and proposals;
- Coordinating and facilitating interactions among the Administrative Center, Board of Directors, Industry Advisory Council, and Technical Committee;
- Monitoring research and extension activities;
- Arranging for review of proposals for technical and scientific merit, feasibility, and applicability to priority problems and preparing summary budgets and reports as required;
- Recruiting other Administrative Center staff as authorized by the Board of Directors;
- With assistance of the Economics and Marketing Work Group, Technical Committee, or others preparing a summary of regional aquaculture, including production statistics and sales, and identifying technical, financial, and institutional constraints to expanding production. The summary shall include sections addressing established industries, development industries, and opportunities for new product development, and recommended research needs;
- Maintaining liaison with other RACs; and
- Serving on the NCC.

At the present time the staff of NCRAC's Office for Publications and Extension Administration at ISU includes Joseph E. Morris, Associate Director, and Merry Rankin, Program Assistant. The Associate Director has the following responsibilities:

- Serving as head of Publications for NCRAC, including editor of the Center's newsletter;
- Serving as the NCRAC liaison with national aquaculture extension programs, including in particular, extension programs of the other four USDA RACs; and
- Serving as a member of NCRAC's Extension Executive Committee.

The Board of Directors (BOD) is the primary policy-making body of the NCRAC. The BOD has established an Industry Advisory Council (IAC) and Technical Committee (TC). Membership of the BOD consists of two persons from the IAC (the chair and an at-large member), a representative from the region's State Agricultural Experiment Stations and Cooperative Extension Services, a member from a non-land grant university and representatives from the two universities responsible for the center: Michigan State and Iowa State. The IAC is composed of representatives from each state's aquaculture association and six at-large members appointed by the BOD who represent various sectors of the aquaculture industry and the region as a whole. The TC is composed of a sub-committee for Extension (TC/E) and a sub-committee for Research (TC/R). Directors of the Cooperative Extension Service within the North Central Region appoint representatives to the TC/E. The TC/R has broad regional make-up and is composed of scientists from universities and state agencies with varied aquacultural expertise who are appointed by the BOD. Each sub-committee of the TC has a chairperson who serves as an ex-officio member of the BOD.

NCRAC functions in accordance with its *Operations Manual* which is periodically amended and updated with BOD approval. It is an evolving document that has changed as the Center's history lengthens. It is used for the development of the cooperative regional aquaculture and extension projects that NCRAC funds.

ADMINISTRATIVE OPERATIONS

Since inception of NCRAC February 1, 1988, the role of the Administrative Center has been to provide all necessary support services to the BOD, IAC, TC, and project work groups for the North Central Region as well as representing the region on the NCC. As the scope of the NCRAC programs expand, this has entailed a greater work load and continued need for effective communication among all components of the Center and the aquaculture community.

The Center functions in the following manner.

- After BOD approval of Administrative Center costs, the Center submits a grant to USDA/CSREES/Grants Management Branch for approval. To date the Center has received 11 grants from USDA for FY88 (Grant #88-38500-3885), FY89 (Grant #89-38500-4319), FY90 (Grant #90-38500-5008), FY91 (Grant #91-38500-5900), FY92 (Grant #92-38500-6916), FY93 (Grant #93-38500-8392), FY94 (Grant #94-38500-0048), FY95 (Grant #95-38500-1410), FY96 (Grant #96-38500-2631), FY97 (#97-38500-3957), and FY98 (#98-38500-5863) with monies totaling $7,949,181. Currently, five grants are active (FY94-98); the first six grants (FY88-93) have terminated.

- The Center annually coordinates a program planning meeting which sets priorities for the next funding cycle and calls for regional workshops to develop project outlines to address priority problem areas.
- Work Groups, which are formed at the workshops, submit project outlines to the Center. The projects are peer reviewed by experts from both within and outside the region.
- The BOD, using reviewers' responses, decides which projects are to be approved and funding levels. The Center conveys BOD decisions to all Project Work Groups. Those that are approved for funding are asked to submit revised project outlines incorporating BOD and reviewers' comments.
- The Center then submits the revised project outlines as a Plan of Work (POW) to USDA for approval.
- Once a POW is approved by USDA, the Center then prepares subcontracts for each participating institution. The Center receives all invoices for subcontractual agreements and prepares payment vouchers for reimbursement. Thus, the Center staff serve as fiscal agents for both receiving and disbursing funds in accordance with all terms and provisions of the grants.

Through August 31, 1997, the Center has funded or is funding 45 projects through 236 subcontracts from the first nine grants received. Funding for these Center supported projects is summarized in Table 1 below (pages 7-8).

During this reporting period, the Publications Office at ISU produced and distributed a number of publications including fact sheets, technical bulletins, videos, and the Center's newsletter. A complete list of all publications from this office is included in the Appendix under Extension.

Other areas of support by the Administrative Office during this reporting period included: monitoring research and extension activities and developing progress reports; developing liaisons with appropriate institutions, agencies and clientele groups; preparing, in coordination with the other RACs, both written and oral testimony for the U.S. House Appropriations sub-committee on Agriculture, Rural Development, Food and Drug Administration, and Related Agencies hearing in Washington, D.C.; participating in the NCC; numerous oral and written presentations to both professional and lay audiences; working with other fisheries and aquaculture programs throughout the North Central Region; and in conjunction with the Aquaculture Network Information Center (AquaNIC) maintaining a NCRAC web site (ag.ansc.purdue.edu/aquanic/ncrac).

PROJECT DEVELOPMENT

A joint Program Planning meeting of the BOD, IAC, and TC is held every year in the early winter. The IAC, with input from the TC, generates a list of priority areas for consideration by the BOD. Using their recommendation as guidelines, the BOD then selects priority areas for which project outlines will be developed. The BOD also specifies a maximum funding level for each priority area. Problem statements and objectives are then developed for each priority area by IAC and TC members at the Program Planning meeting. For projects with more than one objective, the IAC ranks the objectives by priority. The problem statement and objective(s) are then included in a workshop announcement that is broadly distributed throughout the North Central Region. The workshops are one-day events to establish a work group that will develop a project outline over the summer months.

Work group members will be those who have demonstrated that they have the expertise and facilities for undertaking the proposed work in regard to a particular objective or objectives. The proposed work cannot deviate from the objective or objectives included in the workshop announcement. The work group elects a chair and secretary. The chair is responsible for submitting the project outline to the NCRAC Director; the secretary is responsible for preparing minutes from the workshop that are distributed to all attendees. All project outlines are peer reviewed. The reviewers' comments are used by the BOD in making the final selection of projects and level of funding at the following year's annual Program Planning meeting. All work group members are apprised of the BOD decisions. Revisions of projects approved by the BOD are submitted by the work group chair to the NCRAC Director. The revised project outlines are then included in a POW that is submitted to USDA. Upon approval by USDA, the Center issues subcontracts to the funded work group members.

TIME FRAME

- Program Planning meeting: early winter.
- Workshops: late-spring, early summer.
- Project outlines developed over the summer by work group members who participated in the workshops. These project outlines are then submitted to the Center in the fall and peer reviewed.
- The Board of Directors at the following year's Program Planning meeting selects the projects to be funded.
- Project outline revised and submitted to the Center by May.
- Revised project outlines are then submitted in June as a POW (or an amendment to a POW) to USDA for approval. Once approved by USDA, subcontracts are let by the Center with a start date of September 1.

By following this procedure, it takes approximately 18 months from the time of identifying a priority area until inception of a project to address the issue in question.

WORKSHOPS

The purpose of the workshops is to bring together those who are best qualified to work on project objectives by virtue of a demonstrated record of expertise and access to facilities required in the project. These people form a work group for the purpose of writing a project outline to address the problem in question. The following criteria typically apply to those projects that are funded by NCRAC.

- Involves participation by two or more states in the North Central 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 to time (usually up to 2 years);
- can provide the solution to a problem of fundamental importance or fill an information gap;
- can be organized and conducted on a regional level, assuring coordinated and complementary contributions by all participants.

The NCRAC program pays no overhead to participating institutions nor tuition remission, has no brick-and-mortar money, and relies on in-place salaried personnel, equipment, and facilities to carry out the projects. Due to the collaborative and cooperative nature of these regional projects, no one individual or institution receives a significant portion of the total project funds.

PROJECT REPORTING

As indicated in Table 1, NCRAC has funded a number of projects for many of the project areas it has selected for research and extension activities. For example, there have been six separately funded projects in regard to Extension and Yellow Perch. Project outlines have been written for each separate project within an area, or the project area itself if only one project. These project outlines have been submitted in POWs or amendments to POWs for the grants as indicated in Table 1. Many times, the projects within a particular area are merely continuations of previously funded activities; while at other times they are addressing new objectives. Presented below are Progress or Termination Reports mostly for projects that were underway or completed during the period September 1, 1997 to August 31, 1998. Projects, or Project components, that terminated prior to September 1, 1997 have been reported on either in the 1989-1996 Compendium Report or the 1996-97 Annual Progress Report.

A cumulative list of all publications, manuscripts, papers presented, or other outputs for all funded NCRAC project areas is contained in the Appendix.

Table 1. North Central Regional Aquaculture Center funded projects.

Project Area	Project Number	Proposed Duration Period	Funding Level	Grant Number
Extension	1	5/1/89-4/30/91	$39,221	88-38500-3885
			$37,089	89-38500-4319
	2	3/17/90-8/31/91	$31,300	89-38500-4319
	3	9/1/91-8/31/93	$94,109	91-38500-5900
	4	9/1/93-8/31/95	$110,129	91-38500-5900
	5	9/1/95-8/31/97	$10,875	92-38500-6916
			$21,700	95-38500-1410
	6	9/1/97-8/31/99	$40,000	97-38500-3957
			$384,423	
Economics and Marketing	1	5/1/89-12/31/91	$127,338	88-38500-3885
			$34,350	89-38500-4319
	2	9/1/91-8/31/92	$53,300	91-38500-5900
	3	9/1/93-8/31/95	$40,000	93-38500-8392
			$254,988	
Yellow Perch	1	5/1/89-8/31/91	$76,957	88-38500-3885
			$85,723	89-38500-4319
	2	6/1/90-8/31/92	$92,108	90-38500-5008
	3	9/1/91-8/31/93	$99,997	91-38500-5900
	4	9/1/93-8/31/95	$150,000	93-38500-8392
	5	9/1/95-8/31/97	$200,000	95-38500-1410
	6	9/1/97-8/31/99	$200,000	97-38500-3957
			$904,785	
Hybrid Striped Bass	1	5/1/89-8/31/91	$68,296	88-38500-3885
			$68,114	89-38500-4319
	2	6/1/90-8/31/92	$101,000	90-38500-5008
	3	9/1/91-8/31/93	$96,550	91-38500-5900
	4	9/1/93-8/31/95	$168,000	93-38500-8392
	5	9/1/95-8/31/97	$150,000	95-38500-1410
			$651,960	
Walleye	1	5/1/89-8/31/91	$177,517	89-38500-4319
	2	6/1/90-8/31/92	$111,657	90-38500-5008
	3	9/1/91-8/31/92	$109,223	91-38500-5900
	4	9/1/92-8/31/93	$75,000	89-38500-4319
	5	9/1/93-8/31/95	$150,000	93-38500-8392
	6	9/1/95-8/31/97	$117,395	94-38500-0048
			$59,847	95-38500-1410
			$800,639	
Sunfish	1	6/1/90-8/31/92	$130,758	90-38500-5008
	2	9/1/92-8/31/94	$149,799	92-38500-6916
	3	9/1/94-8/31/96	$173,562	94-38500-0048
	4	9/1/96-9/31/98	$200,000	96-38500-2631
			$654,119	

Project Area	Project Number	Proposed Duration Period	Funding Level	Grant Number
Salmonids	1	6/1/90-8/31/92	$9,000	89-38500-4319
			$120,799	90-38500-5008
	2	9/1/92-8/31/94	$149,997	92-38500-6916
	3	9/1/94-8/31/96	$199,290	94-38500-0048
	4	9/1/97-8/31/99	$160,000	97-38500-3957
			$639,086	
NCR Aquaculture Conference	1	6/1/90-3/31/91	$7,000	90-38500-5008
National Aquaculture Extension Workshop/Conference	1	10/1/91-9/30/92	$3,005	89-38500-4319
	2	12/1/96-11/30/97	$3,700	95-38500-1410
			$6,7005	
Crayfish	1	9/1/92-8/31/94	$49,677	92-38500-6916
Baitfish	1	9/1/92-8/31/94	$61,973	92-38500-6916
Wastes/Effluents	1	9/1/92-8/31/94	$153,300	92-38500-6916
	2	9/1/96-8/31/98	$100,000	96-38500-2631
			$253,300	
National Aquaculture INAD/NADA Coordinator	1	9/1/93-8/31/94	$2,000	89-38500-4319
		5/15/95-5/14/96	$5,000	94-38500-0048
		5/15/96-5/14/97	$6,669	92-38500-6916
			$3,331	95-38500-1410
		5/15/97-5/14/98	$15,000	96-38500-2631
		5/15/98-5/14/99	$13,241	94-38500-0048
			$45,241	
Tilapia	1	9/1/96-8/31/98	$120,000	96-38500-2631
Aquaculture Drugs	1	7/1/96-6/30/97	$27,000	95-38500-1410
	2	12/1/96-11/30/97	$5,000	95-38500-1410
			$32,000	

PROJECT TERMINATION
OR
PROGRESS REPORTS

EXTENSION[1]

Project Component Termination Report for the Period
September 1, 1995 to August 31, 1998

NCRAC FUNDING LEVEL: $10,613 (September 1, 1995 to August 31, 1997)

PARTICIPANTS:

Ying Q. Ji	Minnesota Department of Agriculture	Minnesota
John M. Leininger	North American Fish Farmers Cooperative	North Dakota
LaDon Swann	Purdue University	Indiana/Illinois

REASON FOR TERMINATION
This component of the project was completed and funding expended.

PROJECT OBJECTIVE
Quarterly survey of wholesale fish buyers in selected U.S. and Canadian cities with emphasis on the North Central Region (NCR).

PRINCIPAL ACCOMPLISHMENTS
The NCR wholesale market report was only partially successful due to difficulty in obtaining the needed information from the wholesale fish buyers network as well as personnel changes during the project time period. As a result only five Canadian and Midwestern reports were generated. However, Swann made an agreement with the Maryland Department of Agriculture (MDA) to distribute a bi-weekly text report that they compiled of wholesale market prices for selected cities along the Atlantic seaboard to the Aquaculture Network Information Center's (AquaNIC) Web page. After Purdue University personnel received a faxed copy of the MDA report, it was re-entered and converted for the Web. Twenty-eight MDA reports were posted on AquaNIC.

IMPACTS
This work was the first attempt for a market report for the NCR. Experiences garnered from this work will be useful for planning similar market survey projects in the future.

RECOMMENDED FOLLOW-UP ACTIVITIES
The wholesale market survey was something of interest for many of the aquaculture producers in the NCR. However, for such a survey to be successful, someone needs to devote at least half of their time in undertaking such an activity and have a strong connection with the fish wholesaling industry. The National Marine Fisheries Service (NMFS) provides daily and weekly mark prices for Boston, Long Beach, New Orleans, New York, and Seattle. With adequate funding NMFS's service could be expanded to other regions.

PUBLICATIONS, MANUSCRIPTS, WORKSHOPS, AND CONFERENCES
See the appendix for a cumulative output for all NCRAC-funded Extension activities.

[1]NCRAC has funded six Extension projects. This termination report is for one of the objectives of the fifth Extension project. A progress report for all other projects and objectives is contained elsewhere in this Annual Progress Report.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1995-97	$10,613						$10,613
TOTAL	$10,613						$10,613

EXTENSION[2]

Progress Report for the Period
May 1, 1989 to August 31, 1998

NCRAC FUNDING LEVEL: $333,810 (May 1, 1989 to August 31, 1998)

PARTICIPANTS:

Fred P. Binkowski	University of Wisconsin-Milwaukee	Wisconsin
James M. Ebeling	Ohio State University	Ohio
Robert D. Espeseth	University of Illinois	Illinois
Donald L. Garling	Michigan State University	Michigan
Jeffrey L. Gunderson	University of Minnesota-Duluth	Minnesota
F. Robert Henderson	Kansas State University	Kansas
John Hochheimer	Ohio State University	Ohio
Anne R. Kapuscinski	University of Minnesota	Minnesota
Terrence B. Kayes	University of Nebraska-Lincoln	Nebraska
David L. Klinkebiel	North Dakota State University	North Dakota
Ronald E. Kinnunen	Michigan State University	Michigan
Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
David J. Landkamer	University of Minnesota	Minnesota
Charles Lee	Kansas State University	Kansas
Frank R. Lichtkoppler	Ohio State University	Ohio
Joseph E. Morris	Iowa State University	Iowa
Kenneth E. Neils	Kansas State University	Kansas
Robert A. Pierce II	University of Missouri	Missouri
Shawn H. Sanders	North Dakota State University	North Dakota
Brian R. Stange	North Dakota State University	North Dakota
Daniel A. Selock	Southern Illinois University-Carbondale	Illinois
John P. Slusher	University of Missouri	Missouri
Fred L. Snyder	Ohio State University	Ohio
LaDon Swann	Purdue University	Indiana/Illinois
Laura G.Tiu	Ohio State University	Ohio

PROJECT OBJECTIVES

(1) Strengthen linkages between North Central Regional Aquaculture Center (NCRAC) Research and Extension Work Groups.

(2) Enhance the NCRAC extension network for aquaculture information transfer.

(3) Provide in-service training for Cooperative Extension Service, Sea

[2]NCRAC has funded six Extension projects, the first three of which were chaired by Donald L. Garling. The fourth project was chaired by Fred P. Binkowski. The fifth and sixth projects are chaired by Joseph E. Morris. A Project Component Termination Report for one of the objectives of the fifth Extension project is contained elsewhere in this Annual Progress Report. The sixth project is a 2-year project that began September 1, 1997.

Grant Advisory Service, and other landowner assistance personnel.

(4) Develop and implement aquaculture educational programs for the North Central Region (NCR).

(5) Develop aquaculture materials for the NCR including extension fact sheets, bulletins, manuals/guides, and instructional video tapes

ANTICIPATED BENEFITS

Members of the NCRAC Extension Work Group have promoted and advanced commercial aquaculture in a responsible fashion through an organized education/training outreach program. The primary benefits are:

- increased public awareness through publications, short courses, and conferences regarding the potential of aquaculture as a viable agricultural enterprise in the NCR;
- technology transfer to enhance current and future production methodologies for selected species, e.g., walleye, hybrid striped bass, yellow perch, salmonids, and sunfish, through hands-on workshops and field demonstration projects;
- improved lines of communication between interstate aquaculture extension specialists and associated industry contacts; and
- an enhanced legal and socioeconomic atmosphere for aquaculture in the NCR.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

Aquaculture Extension Work Group members have:

- Served as extension liaison, if not an active researcher, for every funded NCRAC project.
- Assisted in writing and developing the NCRAC Walleye Culture Manual that was edited by Bob Summerfelt of Iowa State University.
- Assisted with the planning, promotion and implementation of the hybrid striped bass, walleye and yellow perch workshops held throughout the region.
- Provided the NCRAC Economics and Marketing Work Group with information relevant to that group's efforts to develop cost of production budgets and expected revenues for the commercial production of food-sized hybrid striped bass, walleye, and yellow perch in the NCR.
- Participated as Steering Committee members for a regional public forum regarding revision of the National Aquaculture Development Plan and two National Aquaculture Extension Workshop/Conferences.
- Participated as Steering Committee members for the past three North Central Regional Aquaculture Conferences as well as the upcoming 4th Conference.
- Served as editors for regional aquaculture newsletters as well as in-state aquaculture associations.

OBJECTIVE 2

The demand for aquaculture extension education programs can not be met by the few specialists in the NCR (4.0 full time equivalents). Networking of specialists and Cooperative Extension Service (CES)-designated contacts has maximized efficiency of education programs and minimized duplication. The NCRAC Extension Project is designed to assess and meet the information needs of the various clientele groups through cooperative and coordinated regional educational programming. In fact, individual state extension contacts often respond to 10-15 calls per month from outside their respective state as well as

interacting with colleagues with mutual concerns related to developing aquaculture activities. Many of these requests have been met by providing fact sheets, technical bulletins, bibliographies and detailed responses to specialized questions.

Prior to mid-1994 little coordination of international aquaculture information sharing existed. National and international agencies producing information could only be obtained by contacting the respective sources of this information. Also, individual Sea Grant and CES personnel relied heavily on information produced by individual states or through regional cooperative projects. As Internet access extended beyond educational institutions and governmental agencies, a clear need developed to utilize the Internet to reach a much broader audience. In the age of an "information overload" the need for a centralized gateway to the ever-increasing number of aquaculture resources in electronic format was apparent.

Since the Aquaculture Network Information Center (AquaNIC) began more than 25,000 people from 49 countries have chosen to use AquaNIC as an alternative to or in conjunction with traditional means of obtaining information. Primary users by countries are: U.S. (40%), Canada (5%), Australia (3%), and the United Kingdom (2%). As a gateway to electronic resources in aquaculture, AquaNIC has increased the timeliness and variety of information available to outreach educators, governmental agencies, and individual users while more effectively utilizing existing personnel resources. AquaNIC can be accessed anytime and, therefore, does not face the challenges associated with office hours, time zones or weekends. Several groups have recognized the benefits AquaNIC provides to the world aquaculture industry and have established long-term partnerships with AquaNIC to assist them in distribution of their resources. Key groups using AquaNIC to house their Web sites include: World Aquaculture Society, NCRAC, Indiana Aquaculture Association, Illinois Aquaculture Industry Association, and the Indiana-based Aquatic Control, Inc.

AquaNIC began on a Gopher Server in July 1994 and moved to a World Wide Web server in January 1996. AquaNIC (http://ag.ansc.purdue.edu/aquanic) houses more than 1650 extension publications, governmental documents, image files, comprehensive e-mail lists, newsletters, calendars, job announcements, and résumés. In addition, AquaNIC has 190 pointers to other aquaculture and fisheries related Web sites. It is the gateway to the world's electronic resources in aquaculture including the Regional Aquaculture Centers. It also serves as the home of NCRAC's Web site http://ag.ansc.purdue.edu/aquanic/ncrac.

Swann has coordinated the distribution of NCRAC Annual Reports through AquaNIC. Currently, AquaNIC houses NCRAC Extension Fact Sheets and NCRAC Technical Reports and 1991-1997 Annual Progress Reports. Other services provided on the NCRAC Web Site include a directory of administrative staff and various NCRAC committee members, Extension contacts and the NCRAC Journal newsletter. Other activities related to the AquaNIC and NCRAC Web sites include the development of a World Wide Web 30-slide set for use in Extension and Sea Grant Educator training.

AquaNIC has been recognized by various groups including:

- Bronze award from the 1996 National Agriculture Communicators in Education in the category of publication for the AquaNIC mouse pad.

- Certificate of appreciation from the USDA was presented to Mark Einstein and LaDon Swann for leadership and service in creating and fostering the development of AquaNIC.
- Three star rating from Mckinley Group's online editorial team (1996).
- Best of the non-commercial sites on the Internet by Progressive Farmer On-Line (1996).
- Exceptional agriculture-related Web site by Ag View (1996).
- Editorial on the Success of the Aquaculture Network Information Center in *The Aquaculture News*. June 1996.

Aquaculture handbooks have been developed and distributed to each NCRAC-designated aquaculture extension contact and selected CES and Sea Grant field staff member.

As with any organization, there have been changes in NCRAC extension personnel since the inception of the project. Landkamer was the primary aquaculture extension contact for Minnesota. However, he left the university and Kapuscinski became the primary contact person; Gunderson has since assumed that responsibility. Two other individuals were replaced in 1994. In Kansas, Neils replaced Henderson and in Illinois, Kohler replaced Selock. There continues to be changes in NCRAC extension personnel since the inception of the project. Lee replaced Neils in Kansas in 1996. Hochheimer, who replaced Ebeling in Ohio, left Ohio State University; Tiu has been recently appointed as the aquaculture extension specialist for Ohio. Sanders has been recently appointed as the extension contact for North Dakota.

OBJECTIVE 3
In-service training for CES and Sea Grant personnel and other landowner assistance personnel have been held in most of the states in the region. Training has been in the areas of basic aquaculture and safe seafood handling including Hazard Analysis Critical Control Point (HACCP). Many of these individuals have, in turn, trained industry representatives in HACCP.

OBJECTIVE 4
A number of workshops, conferences, videos, field-site visits, hands-on training sessions, and other educational programs have been developed and implemented.

There have been workshops on general aquaculture, fish diseases, commercial recirculation systems, aquaculture business planning, crayfish culture, pond management, yellow perch and hybrid striped bass culture, rainbow trout production, in-service training for high school vocational-agricultural teachers and polyploid induction in sunfish held in the region. In several states, e.g., Iowa, Ohio and Wisconsin, potential fish farmers have been able to view aquaculture systems being operated by extension and research personnel.

Three North Central Regional Aquaculture Conferences have been held. The first in Kalamazoo, Michigan was held in March 1991. The second was held in February 1995 in Minneapolis, Minnesota and the third conference was held in Indianapolis, Indiana. These regional meetings were attended by hundreds of individuals including persons from Canada. The next conference is scheduled for February 1999 in Colombia, Missouri.

On April 10, 1993, over 700 viewers from 35 states and Canada watched the first national interactive teleconference on aquaculture, "Investing in Freshwater Aquaculture" that was broadcast from Purdue University. It was a televised

satellite broadcast for potential fish farmers. The program consisted of 10 five- to seven-minute video tape segments which addressed production aspects of channel catfish, crayfish, rainbow trout, hybrid striped bass, tilapia, yellow perch, baitfish, and sportfish. A set of course materials was available prior to the program. Three times during the program, a question and answer period was available to the audience through a toll free telephone number. Questions not answered during the program were answered by mail afterwards. The entire teleconference is available as a videotape from NCRAC's Publications Office as well as two other videotapes by the University of Nebraska-Lincoln that are reprises of the broadcast.

In support of extension activities being funded through research projects, i.e., hybrid striped bass and sunfish research projects, extension specialists have completed fact sheets/book chapters/videos. These extension materials arising from the combined efforts of both extension specialists and researchers will help to address many questions concerning aquaculture in the NCR.

In addition to the previously mentioned areas, several NCRAC extension contacts have been instrumental in fostering the continued growth of the aquaculture industry in the region. For example, Pierce has recently created the Cooperative Extension Aquaculture and Marketing Educational Program to facilitate the development and implementation of aquaculture educational programs in Missouri. Many of the NCRAC extension contacts have worked with industry and governmental representatives to produce state aquaculture plans and improved governmental regulations. Binkowski has worked with the Wisconsin Department of Agriculture, Trade and Consumer Protection in the production of: A Wisconsin Aquaculture Industry Profile Processor Survey 1998 and 1998 Wisconsin Aquaculture Directory.

All fish processors, including those who handle aquaculture products, are now required by law to process their fish following HACCP guidelines. Four 3-day HACCP training workshops were conducted by Kinnunen. These workshops served to train fish processors on the principles of HACCP and to give them knowledge on how to develop and implement a HACCP plan for their specific facility. Fish being processed at facilities running under HACCP now meet standards enforced by the FDA.

Kinnunen also worked with the Great Lakes Fish Health Committee on establishing a risk-based system to guide appropriate health decisions recognizing that zero risk is never attainable. If a risk system regarding specific fish diseases could be developed it may help relieve the burden of sacrificing 60 fish per lot for disease certification, which has been a hardship on fish farmers. This risk assessment work on fish diseases has been done based on epidemiological studies.

OBJECTIVE 5

Numerous fact sheets, technical bulletins, and videos have been written or produced by various participants of the Extension Work Group. These are listed in the Appendix.

Other extension-funded activities include: (1) a 4-H Guide for Aquaculture, (2) "Getting Started in Freshwater Aquaculture" CD-ROM and workbook, and (3) HACCP videos. The first two activities have been undertaken by Swann whereas Kinnunen has been working with Steven C. Ingham (University of Wisconsin-Madison) on the HACCP videos. NCRAC has only provided a small component of the funding for the first two activities. The 4-H guide will consist of

three youth guides and one leader's guide developed by a multi-disciplinary design team consisting of Sea Grant and CES experts. The guides will be understandable by leaders and youth regardless of their background in aquaculture. The "Getting Started in Freshwater Aquaculture" CD-ROM and workbook is computer-based instruction on the fundamentals of aquaculture. The workbook serves as a guide for use of the CD-ROM which contains technical information, business planning forms, and example examination questions for instructors teaching aquaculture courses.

Two HACCP videos will be developed. The first will cover the basics of sanitation in a fish processing plant and the development of a Sanitation Standard Operating Procedure. This video will be similar to one produced for the American Association of Meat Processors in 1996 which sold over 300 copies nationwide. The second video will describe the steps involved in smoking fish, with particular emphasis on the Critical Control Points in this process that must be monitored in a HACCP system.

WORK PLANNED

Efforts will continue in regard to strengthening linkages between research and extension work groups as well as enhancing the network for aquaculture information transfer. Participants will also continue to provide in-service training for CES, Sea Grant, and other land owner assistance personnel.

Educational programs and materials will be developed and implemented. This includes development of a sunfish culture guide, yellow perch culture guide and videos, hybrid striped bass culture guide, a publication on fee-fishing (sunfish), tilapia culture information packet, and a publication on yellow perch culture in flowing water systems. In addition, a draft of the 4-H Guide for Aquaculture will be completed and pilot-tested and the two HACCP videos completed.

Future HACCP workshops will be planned as needed in the NCR. Any additional workshops developed and hosted by state extension contacts will be advertised in surrounding states to take advantage of the NCRAC extension network and the individual expertise of Extension Work Group participants.

IMPACTS

- In-service training for CES and Sea Grant personnel has enabled those professionals to respond to initial, routine aquaculture questions from the general public.
- Development of aquaculture education programs for the NCR has provided "hands-on" opportunities for prospective and experienced producers. Approximately 5,000 individuals have attended workshops or conferences organized and delivered by the NCRAC Extension Work Group. Clientele attending regional workshops learned of aquaculture development strategies in other areas of the country and acquired information which was of direct use to their own enterprises. Education programs also created situations where problems encountered by producers were expressed to extension personnel who later relayed them to researchers at NCRAC work group meetings for possible solutions through the research effort.
- Fact sheets, technical bulletins, and videos have served to inform a variety of clients about numerous aquaculture practices for the NCR. For instance, "Making Plans for Commercial

Aquaculture in the North Central Region" is often used to provide clients with initial information about aquaculture, while species specific publications on walleye, trout, and catfish have been used in numerous regional meetings and have been requested by clients from throughout the United States. Publications on organizational structure for aquaculture businesses, transportation of fish in bags, and others are beneficial to both new and established aquaculturists. In a 1994 survey, NCRAC extension contacts estimated that NCRAC publications were used to address approximately 15,000 client questions annually.

- NCRAC extension outreach activities have helped to foster a better understanding and awareness for the future development of aquaculture in the region.
- AquaNIC has become an entry point for many people searching for aquaculture information on the Web. AquaNIC's home page now averages more than 3,000 visits per month by people from more than 50 countries. The Illinois-Indiana Sea Grant Program has also created Web pages for the Indiana Aquaculture Association, the Illinois Aquaculture Industry Association, NCRAC, and the World Aquaculture Society.
- The 4-H Guide for Aquaculture will offer a tremendous opportunity to teach math, biology, and chemistry using experiential learning. Aquaculture could also be easily adapted to teach life skills, e.g., communications and leadership to youth. Leadership and analytical skills can be strengthened through carefully choosing how content is selected and used. Incorporating aquaculture into 4-H Youth programs is not limited to rural farming communities; the curriculum could also be used in urban and inner city schools.
- Fish processors who have attended NCRAC-sponsored HACCP Training Workshops have learned the principles of HACCP with regards to its importance in insuring the production of a safe fishery product. They also learned how to work with their production employees on developing a HACCP Plan specific to their own processing facility. HACCP Plans have now been implemented by workshop attendees who are now keeping records of their daily processing and Sanitation Standard Operating Procedures. About 140 fish processors and/or aquaculturists attended one of the four HACCP Training Workshops.

PUBLICATIONS, MANUSCRIPTS, WORKSHOPS, AND CONFERENCES

See the appendix for a cumulative output for all NCRAC-funded Extension activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1989-91	$107,610	$237,107				$237,107	$344,717
1991-93	$94,109	$152,952				$152,952	$247,061
1993-95	$110,129	$198,099		$250,000	$55,000	$503,099	$613,228
1995-97	$21,962	$149,325	$5,000	$84,000		$238,325	$260,287
1997-99	$20,825	$52,900				$52,900	$73,725
TOTAL	$354,635	$790,383	$5,000	$334,000	$55,000	$1,184,383	$1,539,018

YELLOW PERCH[3]

Project Termination Report for the Period
September 1, 1993 to August 31, 1998

NCRAC FUNDING LEVEL: $350,000 (September 1, 1993 to August 31, 1998)

PARTICIPANTS:

Fred P. Binkowski	University of Wisconsin-Milwaukee	Wisconsin
Paul B. Brown	Purdue University	Illinois
Konrad Dabrowski	Ohio State University	Ohio
Donald L. Garling	Michigan State University	Michigan
Terrence B. Kayes	University of Nebraska-Lincoln	Nebraska
Jeffrey A. Malison	University of Wisconsin-Madison	Wisconsin
Extension Liaison:		
Donald L. Garling	Michigan State University	Michigan
Non-funded Collaborators:		
Harlan Bradt, etc.	Coolwater Farms, LLC, Cambridge	Wisconsin
William Hahle	Pleasant Valley Fish Farm, McCook	Nebraska
John Hyink/John Wolf	Alpine Farms/Glacier Springs Trout Hatchery	Wisconsin
Dave Smith	Freshwater Farms of Ohio, Inc., Urbana	Ohio
Michael Wyatt	Sandhills Aquafarm, Keystone	Nebraska
Nebraska Game & Parks Commission	Calamus State Fish Hatchery, Burwell	Nebraska
Forrest Williams	Bay Port Aquaculture, Inc., West Olive	Michigan

REASON FOR TERMINATION

The objectives for this project were completed and funding was expended.

PROJECT OBJECTIVES

(1) Continue to improve larval rearing techniques by developing and evaluating different starter diets in relation to size at transfer to formulated feeds under selected environmental conditions.

(2) Continue to improve pond fingerling production through examination of in-pond feeding techniques using physical/chemical attractants and improved harvesting strategies for different sizes of fingerlings from various types and sizes of ponds.

(3) Continue development of extension materials and workshops emphasizing practical techniques coinciding with production events to meet the needs of established and potential yellow perch culturists through on-site presentations at

[3]NCRAC has funded six Yellow Perch projects. Termination reports for the first three projects are contained in the 1989-1996 Compendium Report. This termination report is for the fourth and fifth Yellow Perch projects, both of which were chaired by Jeffrey A. Malison. The fifth project continued and expanded upon work undertaken in the fourth project. It was a 2-year study that began September 1, 1995.

two or more locations in different parts of the region.

PRINCIPAL ACCOMPLISHMENTS

As an integral component of this project, private producers cooperated by providing facilities, fish, feed, day-to-day husbandry, and routine data collection. At its inception, this project included the participation of eight different private fish farms in various parts of the North Central Region (NCR). Participating university researchers provided project oversight on experimental design, advice or direct assistance with the technical set-up of any specialized experimental systems, supervision and assistance on critical end-point data collection, and analyses of results.

In the project, significant progress was made at certain sites at testing selected research-based production technologies. Accordingly, from an extension perspective, the project successfully built and/or expanded working relationships between North Central Regional Aquaculture Center (NCRAC) researchers and certain regional fish farmers, testing various research-based technologies under practical production conditions, transferring knowledge from academia to the private sector, and identifying private producers who are both capable and willing to sustain a collaborative technology evaluation and demonstration effort. Several of the original private-sector collaborators have either met or have worked hard to meet their project commitments.

OBJECTIVE 1

Researchers at Michigan State University (MSU) directed their efforts towards: (1) identifying mouth gapes for first feeding yellow perch larvae, (2) correlating *Artemia* cyst diameter to nauplius hatching size, and (3) estimating heritabilities. Mouth gapes for larval yellow perch less than 10 mm (0.39 in) total length (TL) were described by linear regression models. For a first feeding 6.0-mm (0.24-in) TL larvae, the mouth gape width was approximately 322 μm (0.0127 in) and the mouth gape height was approximately 318 μm (0.0125 in).

Newly hatched *Artemia* nauplii from the Great Salt Lake strain (GSL) were as small or smaller than nauplii from the San Francisco Bay strain (SFB). The SFB strain has been marketed as producing the smallest nauplii. The demand and price for the SFB strain was higher, reaching as much as $220/kg ($100/lb), while the GSL strain was priced at less than $77/kg ($35/lb). Large statistical variations in cyst diameters were observed for the SFB and GSL *Artemia* strains. Nauplii hatched from cysts collected in the micro-sieves MS200 (mesh size in μm) and MS280 had statistically significant differences in length, width, and appendage length for both strains. These results indicate that there was a significant, positive correlation between *Artemia* cyst diameter and nauplii length, width, and appendage length.

Decapsulated *Artemia* cysts were used to culture first feeding yellow perch larvae with limited success. Hydrated, decapsulated cysts were also used with limited aeration, but dehydrated cysts would be better, because they tend to float on the surface and sink over a longer period of time. A flat bottom added to the yellow perch larval culture tanks helped to spread out the larvae when they exhibited resting behavior.

Initially, MSU researchers evaluated the effect of female spawner size on larval yellow perch characteristics. Positive linear relationships were significant between female spawner size (TL and weight) and egg ribbon weight and total fecundity. However, linear relationships were not significant

between female spawner size and number of eggs per gram of eggs, chorion shell diameter, and egg yolk diameter. The inability to establish a correlation between female spawner size and these egg characteristics may be due to large environmental variation. Examples of variables that can result in environmental variation are food availability, food quality, water temperature, and predation. Linear relationships could not be established between female spawner size and larval TL and size of the mouth gape, which could have been the result of large paternal influences or environmental variation. A genetics study was designed to identify the amount of influence from both maternal and paternal sources and from the environment.

The MSU genetics study was designed to partition out the influence of the maternal and paternal sources of variation on larval TL, mouth gape width, and height. Overall, the paternal contribution to the total variation was small (0–0.13). The overall residual term, including any environmental variance, was larger (0.05–0.21), which indicates the significant environmental influence. Heritabilities (h^2), based on spawner TL and age, were estimated for larval yellow perch TL ($h^2 = 0.14$), mouth gape width ($h^2 = 0.00$), and mouth gape height ($h^2 = 0.23$). All values of heritability were less than 0.50, which indicates that selection for improvement of these characteristics will be unlikely. Because this study included a fixed assignment of the parental stock, a true estimation of heritability could not be calculated. However, the estimates of heritability provide a valuable insight to the possibility of starting breeding programs, which could select for other desirable characteristics. The number of brood stock used and the number of larvae sampled were large enough to estimate the genetic variance components. However, by increasing the number of brood stock used, the amount of variation in the population would be better represented. The large dominance values (0.21–1.57) indicate that the variations in larval TL and mouth gape sizes occurred by chance.

Studies at Purdue University (Purdue) were designed to quantify the dietary requirements for sulfur amino acids (methionine plus cyst(e)ine) and the dietary choline requirements, providing the framework for the legal use of betaine as a flavor additive in diets for yellow perch. To date, the dietary requirements for lysine, arginine, total sulfur amino acids, the sparing effect of cyst(e)ine for methionine, and choline have been quantified in juvenile fish fed experimental diets.

The dietary requirements for lysine and arginine were 1.5% and 1.4% of the dry diet, respectively. The dietary total sulfur amino acid requirement for juvenile yellow perch is 1.0% of the diet and cyst(e)ine, a nonessential amino acid, can spare approximately 50% of the dietary requirement for the essential amino acid methionine. When total sulfur amino acid concentrations were held at the requirement, the dietary choline requirement was 750 mg/kg (ppm) of diet.

In 1996, Ohio State University (OSU) researchers spawned yellow perch out-of season during September–October by shifting the photothermal condition (light hours and temperature) by six months. The natural spawning of yellow perch occurs in April-May at 12–14°C (53.6–57.2°F) and a 12 h photoperiod. The brood stock was maintained at higher temperature and longer photoperiod during September–February (18°C [64.4°F] and 13 h). The photothermal conditions were decreased gradually until June. The chill period (10°C

[50°F] and 11 light h) was 60 days in duration (June–July) and was followed by gradually increased water temperature and longer day light (12°C [53.6°F] and 19 h). Following this period, 47% of the females were recorded as gravid and 24 were stripped or spawned naturally. The males spermiated during the entire shifted spawning period from August until September. The average relative weight of ovulated eggs as percentage of the female weight was 26.6 ± 10.7%. Embryo survival through the eyed-stage was 56 ± 24%. Larval skeleton abnormalities (45 ± 15%) and a low frequency of swim bladder inflation (44 ± 34%) were observed.

Hatching occurred seven days after spawning incubation at 14°C (57.2°F). Just before hatching, the eggs were transferred to 20-L (5.3-gal) aquariums with continuous water flow at 20°C (68°F). Three days after hatching, fresh-water rotifers *Brachionus calyciflorus* and microalgae *Dictyosphaerium chlorelloides* were added three times a day to aquariums at an average concentration of 10 rotifers/ml (296/oz). Eighty percent of the larvae were found to have 1–4 rotifers in the gut at first feeding. *Artemia* nauplii were added six days after hatching. The combination of rotifers, algae and *Artemia* was supplied until 14 days after hatching after which, only *Artemia* nauplii were offered to the larvae. Two different dry diets were tested for weaning 25 day old larvae, salmonid starter diet and experimental squid based diet. However, only 35 day old juveniles were found to accept dry diets and were not weaned completely from *Artemia* until an age of 45 days.

Nine diets were tested as weaning diets, including two commercial ("Zeigler" trout starter [Zeigler Bros., Gardiner, Pennsylvania] and "Biokyowa" [Biokyowa, Inc., Chesterfield, Missouri]), one semi-commercial (F.T. Barrows, Fish Technology Center, Bozeman, Montana), and six experimental diets. Live food (*Artemia* nauplii) was used as a control. In addition, the semi-commercial and one experimental diet ("walleye") were supplemented with 20% (initial fish biomass) *Artemia.* The "Zeigler" trout starter was coated with 5 or 10% (diet weight) krill hydrolysate as a feed attractant. One hundred fish (average wet weight 75.5 ± 5 mg; 0.0027 ± 0.00 oz) were placed in each of 44, 20-L (5.3-gal) aquariums. Fish were fed ad libitum, eight feedings per day. After 31 days, fish were sacrificed, counted, and sampled for length, wet weight, dry weight, and digestive tract enzyme activities. Percent survival to 31 days ranged from 35 ± 6.2% (French diet - based on freeze-dried liver and yeast extract with CMC was used as a binder) to over 70% on a walleye diet (based on krill meal and herring meal as protein sources, including 2% krill hydrolysate with gelatin used as a binder) or Barrows with 20% *Artemia* nauplii (manufactured by marumerization technique).

OBJECTIVE 2

Trials were completed by University of Wisconsin-Madison researchers at Coolwater Farms, LLC, to determine key parameters for producing yellow perch fingerlings habituated to formulated feed and reared in ponds for an entire growing season, and to compare the performance of two types of pond lighting and feeding systems. Their studies showed that rearing fingerlings in ponds for the entire first growing season can result in yields greater than 247,097 fish/ha (100,000 fish/acre), although variability in both pond productivity and fish size result in a wide range of production levels. Over two years of data collection, fingerling production in ponds harvested in the autumn ranged from 49,919–276,478 fish/ha (20,000– 112,000/acre, and averaged

about 148,250 fish/ha (60,000 fish/acre). Autumn-harvested fingerlings ranged in size from 6.4–20.3 cm (2.5–8.0 in) TL (2–100 g; 0.1–3.5 oz total weight). On a per acre basis, gross revenues (based on a fingerling price of $0.075/in) from the various production methods studied were as follows:

- traditional tandem pond/tank — $4,800-$8,000,
- improved tandem pond/tank — $6,000-$12,000,
- in-pond fingerling production with July harvest — $5,700-$9,500, and
- in-pond fingerling production with October harvest — $12,500.

University of Nebraska-Lincoln (UNL) researchers coordinated pond culture field trials in Nebraska. In 1994 and 1995, field trials were conducted at Pleasant Valley Fish Farm in two 0.08-ha × 1.5-m-deep (0.2-acre × 4.9-ft) rectangular ponds, which were drainable, aerated with low-pressure blowers connected to subsurface diffused-air distribution systems, and supplied with groundwater as needed for temperature moderation and to provide fresh water for flushing. Both ponds were filled and fertilized in early spring prior to stocking, then stocked with about 200,000 eyed-eggs (2,500,000/ha; 1,011,750/acre). Three major changes in procedures were made in Year 2: (1) the number of feeding stations in each pond was increased from five to seven, (2) the automatic feeders used were better maintained and more frequently filled with fresh feed, and (3) in one pond, a predetermined number of advanced fry were concentrated in a 1.8-m × 1.8-m × 0.9-m-(5.9-ft × 5.9-ft × 3.0-ft) deep net-pen around one of the feeders.

Perhaps the most significant finding of the Year 1 field trail was that ponds stocked at high rearing densities produced at least three populations of perch of markedly different body sizes, a result that was almost certainly dependent on degree of acceptance of formulated feed. In the first pond, for example, about 25% of the perch harvested averaged about 34.5 g (1.22 oz), about 15% averaged about 8.2 g (0.29 oz), and the remaining 60% averaged about 1.4 g (0.05 oz) in body weight. Examination of gut contents revealed that at the time of harvest the large fish were consuming significant amounts of formulated feed, the medium-size fish were consuming "some" formulated feed, while the small fish were consuming almost no feed. By the date of harvest, the natural forage base in both ponds appeared to be depleted, which was not unexpected given the stocking rates. Conversely, post-harvest analysis in Year 2 revealed distinctly bimodal fish size distributions in both ponds. The weight distributions of the two distinct populations of perch in both ponds were found to be statistically normal. For the first pond, large fish comprised about 18% of those harvested and had a mean weight of about 30.5 g (1.08 oz), while the smaller fish had a mean weight of about 7.1 g (0.25 oz). For the second pond, the comparable figures were about 21%, 33.2 g (1.17 oz) and 7.4 g (0.26 oz), respectively. The perch confined to the net-pen supplied with formulated feed from one feeder had a near-normal (slightly positively skewed) size distribution, with a mean weight at harvest of only about 6.1 g (0.22 oz), suggesting that natural forage in ponds fed formulated feeds may still comprise a major source of food for much of the growing season. However, analysis of gut contents revealed that nearly all the fish in both ponds, as well as the net-pen, were consuming significant amounts of formulated feed by the final harvest date.

Comparison of the data generated by the Nebraska field trials revealed similarities between Year 1 and Year 2 in the estimated

numbers of perch produced and estimated survival rates in the two ponds used at Pleasant Valley Fish Farm. Specifically, the estimated numbers of perch produced and survival rates in Year 1 and Year 2 in the first pond were 12,833 and 11,145 and 13% and 11%, respectively. The same production estimates for the second pond were 8,011 and 9,831 fish, and 8.3% and 9.8% survival, respectively. However, the total biomass of perch harvested from the first pond was about 70.4 kg (880 kg/ha or 785 lb/acre) for Year 1 compared to 122 kg (1,525 kg/ha or 1,360 lb/acre) for Year 2, while the total biomass of perch harvested from the second pond was about 97.3 kg (1,216 kg/ha or 1,085 lb/acre) for Year 1 compared to about 126 kg (1,575 kg/ha or 1,406 lb/acre) for Year 2. Collectively, these findings demonstrated that providing formulated feed to perch fry and fingerlings in small (0.08-ha; 0.20-acre) rearing ponds by the procedures described can increase total production two- to four-fold, depending in large part on the number of feeders used per unit of pond surface area and the level of sustained attention given to maintaining feeders and keeping them supplied with fresh feed.

UNL investigators also evaluated several strategies of harvesting various sizes of young-of-the-year yellow perch (16-mm–35-mm [0.63-in–1.38-in] TL) using light attraction from various types and sizes of ponds. The studies evaluated four different lighting system designs to optimize light attraction and the capture of young perch in three different designs of passive capture gear. The four different lighting systems were tested in combination with all three designs of capture gear. Regardless of the equipment tested, UNL researchers found the “best” size at which to light harvest young perch to minimize physical injury while maximizing the number of fish captured seems to be between about 18-mm (0.71-in) and 25-mm (0.98-in) TL. However, a large number of variables influenced the number of fish captured and the catch per unit effort, combined with the widely varying harvest results observed under what appeared to be very similar conditions. Thus, a single night's effort under seemingly similar conditions with the same equipment might yield anywhere from 50,000–500,000 fish with no obvious explanation for the differences.

Many factors appeared to have major effects on harvest success, regardless of the combinations of harvesting equipment tested. Percentage success of total pond harvest seems to be inversely related to pond surface area, depth and the steepness of slope of pond banks. Thus with 0.2-ha (0.50-acre) ponds, harvest percentages as high as 50% can sometimes be achieved, while with 0.4-ha (1.0-acre) ponds harvest percentages higher than 50–60 are rare. Preliminary trials with larger ponds suggest that percentage harvest declines progressively with increasing pond surface area. Harvesting success can be impaired by poor weather and windy conditions, and can be particularly poor in ponds with steeply sloped banks where most of the littoral zone is deeper than 1.2-m (3.9-ft). High initial stocking rates of ponds with eyed-eggs or fry, if survival is good normally increased initial capture numbers and catch per unit effort. However, the percentage success of total pond harvest is often reduced by high initial stocking rates due in part to the great numbers of harvesting efforts required, each of which seems to have a negative influence on the strength of the phototactic response of perch of similar size to recurring exposures to artificial light. Two particularly important factors that appear to significantly reduce the utility of light harvesting young perch from ponds are: (1) the very short time period during which perch are in an appropriate size

range to harvest on a large scale, and (2) repeated or prolonged exposures to artificial light of perch of a size that are normally highly photopositive have a significant cumulative dampening effect on their overall phototactic response. The first of these factors is of major practical importance because of its limiting effects on the logistics of pond harvesting. Under good growing conditions, young perch may be in the desired range of 18-mm (0.71-in) to 25-mm (0.98-in) TL for only seven to ten days. Such a short time period makes the large-scale light harvesting of perch extremely sensitive to disruptions by poor weather or equipment failures, as well as the physical stamina of workers engaged in all-night harvesting efforts night after night. The obvious dampening effect of repeated or prolonged exposures to artificial light on the phototactic response patterns of young perch suggest that under practical conditions the practice of first habituating young fish to formulated feed in ponds using light as initial attractant to automatic feeders may be incompatible with any subsequent light harvesting strategies.

OBJECTIVE 3

During 1996, three yellow perch workshops were conducted. The University of Wisconsin Sea Grant Institute sponsored two workshops entitled "Intensive Aquaculture of Yellow Perch in Conjunction with Recirculating Aquaculture Systems," which included NCRAC Extension and Yellow Perch Work Group members. Alpine Farms (Sheboygan Falls, Wisconsin) personnel participated as aquaculture industry cooperators to provide their practical experience with, and knowledge of, yellow perch rearing in their recirculating aquaculture system technology.
UNL conducted a workshop in Nebraska.

In 1997, UW-Madison researchers sponsored an organizational meeting of producers of yellow perch who are using pond systems. The objectives of this meeting were to discuss common problems and opportunities facing these aquaculturists. The group was unanimous in their identification of fingerling size uniformity and pond production variability as being the most critical problem areas of production. The group also expressed interest in examining the potential of developing a cooperative mechanism to purchase commodities (e.g., fish food) and market products (e.g., fingerlings and processed fillets). A follow-up meeting of this group together with perch producers using other systems was held at the 1998 Wisconsin Aquaculture Conference (March 13-14, 1998, Eau Claire). These meetings have led to the formation of a yellow perch committee within the Wisconsin Aquaculture Industry Advisory Council. This committee will first meet in September 1998 and one of its first goals will be to develop a means of networking and communicating among all interested perch producers.

UNL delivered a total of eight extension programs that, in whole or in part, provided timely information on various aspects of yellow perch aquaculture. Some progress was made on the production of two videotapes on selected aspects of perch aquaculture: one on procedures for spawning perch, and the second on small-scale perch processing. A "rough-cut" edition of the perch spawning videotape has been reviewed by several aquaculture professionals, and has been shown at aquaculture conferences or workshops in several states, among them Indiana, Michigan, Minnesota, Nebraska, Ohio, Wisconsin, Maryland, and North Carolina. Both videotapes are expected to be completed by UNL by March 1, 1999.

IMPACTS

Quantifying critical nutritional requirements for targeted species reduces feed costs and allows variation in use of feed ingredients. The research completed at Purdue, MSU, and OSU, is defining a yellow perch diet and feeding strategies for use in the NCR.

Total sulfur amino acid concentrations are typically the first limiting amino acid in diets that contain high levels of plant protein feedstuffs. That is, if the requirements for methionine + cyst(e)ine are met, then other essential amino acid concentrations will be at or above the needs of perch. Once methionine is absorbed by perch, it is either used for synthesis of new protein, such as fillets, or catabolized (broken down) into cyst(e)ine, then choline. Given that methionine is limiting in most diets for perch, there will not be excess methionine for cysteine and choline synthesis. Thus, the values quantified at Purdue are vital pieces of information for dietary formulation and provide the basis of equally important work on flavor additives.

With the mouth gape of first feeding yellow perch fry identified, researchers and culturists can focus on providing suitable diets that are small enough for the larvae to consume.

The procedure of shifting the spawning season has to be accompanied with indoor larvae rearing. The larvae rearing protocol developed in this project is based on a combination of microalgae and rotifers as the larvae first feed. *Artemia* nauplii were offered from six days after initiation of feeding. Weaning period started at 35 days and the fingerlings were completely weaned from *Artemia* to dry diet at the age of 45 days. Co-feeding of dry diets and *Artemia* as well as coating starter diet with krill hydrolysate significantly increased growth of yellow perch juveniles.

Studies on pond fingerling production by UW-Madison researchers have shown that research-based production strategies can be used on a commercial scale to produce large numbers of perch fingerlings at a relatively low cost. Lights and automatic feeders can be used to improve the habituation of fingerlings to formulated feeds in tanks, and to feed-train perch directly in ponds. Improvements in feeder design has increased reliability and decreased capital and operational costs.

The Nebraska field trials conducted in ponds at Pleasant Valley Fish Farm in collaboration with the UNL clearly demonstrated that research-based production strategies can be used to culture both fingerling and food-size perch under commercial conditions in ponds, by stocking ponds at high densities and using intensive feeding methods. Field trial data collected at Pleasant Valley Fish Farm indicate that perch can be raised in ponds from eyed-eggs to fingerlings having mean weights of 11.0–12.8 g (0.39–0.45 oz) within one growing season at production levels as high as 1,216–1,525 kg/ha (1,085–1,360 lb/acre); and that age-1 fingerling perch can be raised in one growing season to food-size fish having weights averaging 135 g (4.76 oz), at production levels at least as high as 4,740 kg/ha (4,229 lb/acre). Rates of growth at Pleasant Valley Fish Farm aimed at the production of food-size perch were excellent, ranging from 0.55–0.82 g/day (0.02–0.03 oz/day). While many problems remain in perch culture, these field trials suggest that perch can be raised to food size in ponds within two years.

The extensive field trials conducted by UNL investigators have demonstrated both the utility and limitations of using light to harvest

young photopositive perch. Given the highly variable success rates of harvesting such perch with light and the nature and cost of the highly specialized equipment required to light harvest perch in large numbers, it is recommended that this harvesting practice by used only by experienced fish culturists for very targeted applications, such as the early harvest of very young perch for habituation to formulated feed. Comparatively small numbers of such feed-trained fish can potentially be used later in the growing season to facilitate the habituation of large numbers of perch fingerlings to formulated feed under intensive culture conditions.

Requests for information on yellow perch aquaculture continue to increase annually. Workshops done on yellow perch aquaculture in the NCR have enabled extension specialists and researchers to provide information on this species to established fish farmers, potential fish farmers, and the general public. The workshops have also provided a mechanism for yellow perch culturists to identify problem areas. For example, producers have identified the excessive variability in fingerling size and pond productivity as the critical problems currently faced by yellow perch fingerling producers. This provides valuable insight into future directions that are needed for yellow perch aquaculture research. Addressing these areas of concern expressed by current yellow perch producers will bridge the gap between research and solutions to real-world problems.

RECOMMENDED FOLLOW-UP ACTIVITIES

While the results of these studies have provided important information regarding larval and fingerling yellow perch production, they have also served to emphasize several areas in which improvements are greatly needed.

Results of these studies are being used to continue to improve larval culture. The first objective of the NCRAC Yellow Perch Project that began in September 1, 1997 was, "With the goal of larval intensive yellow perch feeding in tanks from the onset of first feeding, continue to develop methods to produce fingerlings." With the mouth gape identified, researchers can focus on providing suitable diets that are small enough for the larvae to consume. Work completed at OSU provides a strong basis for further study. Although separating *Artemia* cysts by size would enhance their use, the process used by MSU researchers would not be efficient for small-scale aquaculture operations. After the hydration and separation processes, the *Artemia* cysts would have to be dehydrated for storage. Better cyst processing techniques should be developed to separate cysts into smaller size categories after harvesting, but prior to dehydrating the cysts. The cysts could be graded, dehydrated, and sold according to diameter to improve utilization by small larvae.

The nutritional requirements data should be used in developing feeds specifically for larval yellow perch.

The results of heritability studies indicated that, through natural selection, larval TL and the size of the mouth gape for perch may have reached a plateau and cannot be increased through selective improvement. If artificial selection for these traits operates in the same direction as natural selection (i.e., larger mouths and longer lengths), then it may be difficult to improve on natural selection. However, other traits that are important for culture, such as larval survival should be investigated. Another genetics experiment should be conducted, but only after some of the environmental variables can be controlled (i.e., a captive brood stock). A selection program, which identifies perch

that grow well in intensive culture conditions, should be started to aid intensive larval culture techniques. This could be started by using perch raised entirely in intensive culture conditions as the brood stock for future cultures.

The high cost of fingerlings continues to be one of the greatest factors constraining the growth of yellow perch aquaculture. The extreme variability in the size of pond-reared fingerlings, coupled with relatively poor overall production rates (which are typically as much as an order of magnitude lower than theoretical production levels), continue to be critical problems facing yellow perch producers. Accordingly, efforts to develop improved methods of fingerling production need to be continued.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See the Appendix for a cumulative output for all NCRAC-funded Yellow Perch activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1993-94	$75,000	$87,240	$30,000	$10,000[a]		$127,240	$202,240
1994-95	$75,000	$81,587	$30,000	$81,000[abc]		$192,587	$267,587
1995-96	$107,086	$145,814	$20,000	$134,000[ac]		$299,814	$406,900
1996-97	$92,914	$106,095	$22,000	$86,911[a]		$215,006	$307,920
TOTAL	$350,000	$420,736	$102,000	$311,911		$834,647	$1,184,647

[a]Sea Grant/USDC/NOAA
[b]USDI, Bureau of Indian Affairs
[c]EPA

YELLOW PERCH[4]

Progress Report for the Period
September 1, 1997 to August 31, 1998

NCRAC FUNDING LEVEL: $95,300 (September 1, 1997 to August 31, 1998)

PARTICIPANTS:

Fred P. Binkowski	University of Wisconsin-Milwaukee	Wisconsin
Paul B. Brown	Purdue University	Illinois
Konrad Dabrowski	Ohio State University	Ohio
Donald L. Garling	Michigan State University	Michigan
Robert S. Hayward	University of Missouri-Columbia	Missouri
Jeffrey A. Malison	University of Wisconsin-Madison	Wisconsin
Extension Liaison:		
Donald L. Garling	Michigan State University	Michigan
Industry Advisory Council Liaison:		
Forrest Williams	Bay Port Aquaculture, Inc., West Olive	Michigan
Non-funded Collaborators:		
Marty Domer	Ohio Valley Fish Hatchery Inc., Mineral City	Ohio
Forrest Williams	Bay Port Aquaculture, Inc., West Olive	Michigan

PROJECT OBJECTIVES

(1) With the goal of larval intensive yellow perch feeding in tanks from the onset of first feeding, continue to develop methods to produce fingerlings.

(2) Increase growth rates of yellow perch greater than 150 mm (6 in) by evaluating diets, feeding strategies, environmental manipulation, and mono-sex/bi-sex comparisons.

(3) Develop out-of-season spawning methods for yellow perch.

ANTICIPATED BENEFITS

This project will address priority needs identified by the North Central Regional Aquaculture Center (NCRAC) Industry Advisory Council (IAC) for advancing yellow perch aquaculture in the North Central Region (NCR). The proposed research on Objective 1 will improve larval rearing techniques by developing and evaluating different starter diets and environmental conditions. The information generated by these studies will greatly assist perch producers in their efforts to reliably raise the large numbers of perch fingerlings needed by the industry. Research on Objective 2 will develop and evaluate methods for improving growth of perch as

[4]NCRAC has funded six Yellow Perch projects. Termination reports for the first three projects are contained in the 1989-1996 Compendium Report; a termination report for the fourth and fifth projects is contained elsewhere in this Annual Progress Report. This progress report is for the sixth Yellow Perch project, which is chaired by Jeffrey A. Malison. It is a 2-year study that began September 1, 1997.

they approach market size. The use of these methods by commercial perch producers will decrease the time needed to raise perch to market size and thereby increase the efficiency of production facilities and reduce production costs. One of the most promising strategies in this regard is the production of mono-sex female stocks of perch. A method for producing 100% female perch has been developed by researchers at the University of Wisconsin-Madison (UW-Madison) and is currently being used by several regional perch producers under an investigational new animal drug (INAD) exemption granted by the FDA. Current research under another NCRAC project entitled "Safety of 17α-Methyltestosterone for Induction of Sex Inversion in Walleye" is aimed at gaining a universal New Animal Drug Application approval for using this method in percids. The proposed research on Objective 3 will develop methods to induce out-of-season spawning in perch. The resultant availability of perch fry at different times during the year will increase the efficiency of existing pond and tank fry culture systems, by allowing multiple cropping of these systems. In turn, the availability of fingerlings at multiple times during the year would facilitate a fuller, more efficient use of grow-out facilities and equipment. The availability of fertilized eggs outside the normal spawning season would also greatly facilitate research on the culture of perch fry in tanks. Additional benefits of using the procedures developed in these studies include greater predictability of gamete production and reduced incidence of failed spawning, gamete resorption, and subsequent brood fish losses.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

Research at Purdue University (Purdue) was designed to evaluate the available larval diets fed to yellow perch and determine the appropriate size for transferring larvae to formulated diets. Protocols were developed and tested with larval perch and six larval diets were identified and acquired. Insufficient numbers of larvae were available during the year for complete evaluation of the existing diets. However, brood stock have been acquired and placed in environmental chambers at Purdue for testing later in 1998 and early in 1999.

Research at Michigan State University (MSU) was designed to build on studies that determined the mouth size of first feeding yellow perch fry. Protocols were developed to culture appropriately-sized live food and to monitor larval perch feeding behavior using videotaping. Larvae were obtained from Bay Port Aquaculture, Inc. and additional brood fish were collected from Lake Michigan and spawned using the dry method. The poor quality of eggs and larvae available during 1998 resulted in insufficient numbers of first feeding fry to complete the experiments.

Researchers at Ohio State University (OSU) have completed four feeding trials with yellow perch larvae and juveniles. To test the effect of krill hydrolysate as a feed attractant, two series of experiments were conducted. In the first, growth trials were conducted using commercial trout starter diet that was coated with liquid krill hydrolysate. Yellow perch juveniles of 40 ± 5 mg (0.0014 ± 0.0002 oz) average initial wet weight were used. The krill hydrolysate coated diet resulted in growth of yellow perch larvae that was 31% compared to the non-coated control diet (734 ± 33 mg; 0.0259 ± 0.0012 oz versus 559 ± 82 mg; 0.0197 ± 0.0029 oz final wet weight, respectively). Moreover, growth performances were not significantly different than for juveniles fed exclusively live *Artemia*. The food conversion ratio (FCR) was lower in fish fed the control diet,

although not significantly (2.95 ± 0.18 versus 3.69 ± 0.39, control and coated diets respectively). The second experiment determined the ingestion rates of yellow perch larvae fed krill hydrolysate coated starter diet. Four treatments were evaluated (six replicates each, approximately 25 larvae /replicate of yellow perch): (1) live food control (*Artemia* nauplii), (2) commercial base diet (Zeigler trout starter diet #1), (3) commercial base diet coated with 5% krill hydrolysate, and (4) commercial base diet coupled with the addition (1 mL (0.03 oz) every 30 min) of krill supernatant (centrifuged for 30 min at 10,000×). Coated starter diet with 5% krill hydrolysate significantly increased ingestion rates by three fold. The ingestion rates of the coated diet were not significantly different than of *Artemia* nauplii. The supplementation of krill hydrolysate supernatant to the experimental tank water significantly improved ingestion rates of uncoated diet by more than 200%. The ingestion rates were not significantly different from the krill coated diet and *Artemia* nauplii.

OSU researchers have begun experiments to test the influence of digestive hormone (Bombesin, Sigma Chemical Co.) or enzyme (Pancreatin, Sigma Chemical Co.) supplementation. Four diets were offered to yellow perch juveniles (587 ± 45 mg; 0.0207 ± 0.0016 oz average wet weight): (1) Zeigler #2 crumble (Zeigler Bros., Gardiner, Pennsylvania), (2) basic experimental diet formulated at OSU, (3) basic diet supplemented with 0.1% wet weight pancreatin, and (4) the basic experimental diet supplemented with 0.01% wet weight of Bombesin. Growth of fish fed supplemented diets were equal to the non-supplemented diet and the trout starter. No differences were found in length, weight or survival between different dietary treatments. Additionally, no significant differences in trypsin or chymotrypsin activities were found in intestinal tissue of fish fed all tested diets. Total activities of pepsin in tested fish were not significantly different among treatments, though those for the enzyme supplemented diet were slightly higher.

To determine the weaning period and the dry/live food amount, OSU researchers tested four treatments with seven-day-old yellow perch larvae: (1) *Artemia* nauplii only, (2) 50% *Artemia* (calculated as initial fish wet weight biomass) and basic experimental diet ad libitum, (3) 10% *Artemia* and basic diet ad libitum and, (4) experimental diet only. After 29 days, larvae fed a combination of 50% or 10% *Artemia* nauplii and dry diet were not significantly different from larvae fed solely on *Artemia* nauplii (246.3 ± 75.3 mg [0.0087 ± 0.0027 oz], 217 ± 23.6 mg [0.0077 ± 0.0008 oz], and 229.7 ± 76.3 mg [0.0081 ± 0.0027 oz], respectively). Final wet weight of larvae fed the dry diet only was significantly lower (160.7 ± 27.4 mg; 0.0057 ± 0.0010 oz) compared to the other treatments.

OBJECTIVE 2

UW-Madison researchers have completed an experiment that evaluated the effect of genistein, a weakly estrogenic compound found in soybean products, on the growth and reproductive development of yellow perch. The findings from this experiment suggest that low levels (0.75 mg/g of diet) of genistein may have a positive effect on growth in yellow perch, but no apparent estrogenic effects on reproductive function. These experiments also demonstrated that obtaining the desired effects on growth and reproductive development are highly dependent on dose. Studies comparing the growth of male and female yellow perch in ponds are currently underway in two replicate ponds, but the data have not yet been collected.

A scoping study to identify compensatory growth feeding schedules that maximize growth and growth efficiency in yellow perch (>150 mm; 6 in) is presently into day 90 and will continue for another 40 days at the University of Missouri (UM). A control group of individually-held fish (N = 10 for control and treatments) is receiving daily feedings to satiation. Treatment groups are subjected to repeating cycles of no-feed followed by refeeding to excess. No-feed periods are fixed at either 2, 7, 12, 17, or 22 days; these durations of the no-feed periods being the distinguishing characteristic of the five treatment groups. Refeeding periods following no-feed periods are continued until mean consumption rates within the treatment group no longer exceed that of the controls (i.e., hyperphagia has ended).

To date, treatment groups with longer no-feed periods (e.g., 12 days of no feeding) are showing evidence of outperforming those with shorter no-feed periods. This is different than what was observed earlier with hybrid sunfish where the 2-day no-feed period performed the best. While no treatment group has yet outgrown controls, some of the longer-cycle feeding regimes appear that they may do so by the end of the experimental period.

MSU has nearly completed the construction of a system for a metabolism study of 10 –17.5 cm (3.9–6.9 in) male, female, and mixed sex populations of yellow perch. Techniques to identify male and female perch using external characteristics have been improved. The basal metabolic energy, efficiency of diet utilization, and maximum theoretical response to a standard feed will be determined using a saturation kinetic model based on a multiple non-linear regression analysis. Growth studies will begin as soon as the temperature control system is installed.

OBJECTIVE 3

Research under this objective has begun at UW-Madison. Out-of-season spawning has been induced with variable degrees of success in several year classes of yellow perch females. Due to its immediate commercial applicability, emphasis has been placed on inducing spawning in July, which may allow for the double cropping of fingerling ponds. Work on out-of-season spawning at other times of the year (e.g., October and January) has been limited by the failure of water temperature control systems, but will continue once the systems are repaired.

The University Wisconsin-Milwaukee was originally approved to participate in this objective. Remodeling of their laboratory necessitated liquidating the brood stock that were to be used in this project and they have withdrawn from the Work Group.

WORK PLANNED

OBJECTIVE 1

Research at Purdue will evaluate all identifiable larval diets as first feed to larval perch and the appropriate size/age for acceptance of larval diets. Larvae will be acquired from formal collaborators, private sources and from brood stock now held at Purdue for completion of this project.

MSU will evaluate the feeding behavior of first-feeding larval perch fed various live and artificial feeds during the spring of 1999. Larvae will be obtained from Bay Port Aquaculture, Inc. and additional brood fish will be obtained through the Michigan Department of Natural Resources, Fisheries Division.

OSU researcher will analyze the final results of digestive enzymes for the enzymes and hormones supplementation experiment to

complete the evaluation of these additives on the yellow perch digestive system.

A study evaluating the extent to which 11–15 mm (0.43–0.59 in) total length perch can be habituated to formulated food will be conducted by UW-Madison researchers in late spring of 1999.

OBJECTIVE 2
A manuscript describing the effects of genistein on growth and reproductive development will be completed and submitted for publication by UW-Madison researchers. Additional pond replicate studies comparing the growth of male and female perch reared in ponds will be conducted.

UM researchers will continue research to identify compensatory growth feeding schedules that maximize growth and growth efficiency in yellow perch (>150 mm; 6 in).

Studies to determine the basic metabolic response of 10–17.5 cm (3.9–6.9 in) male, female, and mixed sex populations of yellow perch to a reference commercial feed using a saturation kinetic model based on a multiple non-linear regression analysis will begin at MSU at the end of 1998.

OBJECTIVE 3
UW-Madison and Purdue studies on out-of-season spawning induction will continue.

IMPACTS
OBJECTIVE 1
Research on Objective 1 will improve larval rearing techniques by developing and evaluating different starter diets and environmental conditions. The information generated by these studies will greatly assist perch producers in their efforts to reliably raise the large numbers of perch fingerlings needed by the industry.

Research at OSU has provided evidence that the use of krill hydrolysate as a feed attractant for coating commercial diets can increase utilization of dry diets. The coating of commercial starter diets increased ingestion rates and growth of yellow perch larvae as well as shortened the weaning period and the dependency on *Artemia* nauplii. Considering the current high price of *Artemia* cysts ($99–143/kg; $45–65/lb), these results can contribute to the decreasing costs of commercial intensive larvae rearing of yellow perch. The lack of effect of enzyme and hormone supplementation can be due to the advanced age of the tested juveniles. Farther studies need to be conducted on the effect of these in early age as well as with the improved microdiet's nutritional value.

OBJECTIVE 2
Research on Objective 2 will develop and evaluate methods for improving perch growth as they approach market size.

Previous work has led to the development of methods for producing monosex female stocks of perch, and this technology is currently being used by six regional perch producers under an INAD exemption granted by the FDA. The development of methods to promote perch growth with naturally occurring dietary supplements will further improve the profitability of the culture of food-size yellow perch.

OBJECTIVE 3
Research on Objective 3 will develop methods to induce out-of-season spawning in perch. The resultant availability of perch fry at different times during the year will increase the efficiency of existing pond and tank fry culture systems by allowing multiple cropping of these systems. In turn, the availability of fingerlings at multiple times during the year would facilitate a fuller, more

efficient use of grow-out facilities and equipment. The availability of fertilized eggs outside the normal spawning season would also greatly facilitate research on the culture of perch fry in tanks.

An additional site has been established for out-of-season spawning of yellow perch at Purdue without additional cost to NCRAC. Larval rearing techniques and feeding protocols have been established, including development of a new automatic feeder for fish larvae.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See the Appendix for a cumulative output for all NCRAC-funded Yellow Perch activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1997-98	$95,300	$98,565	$2,000			$100,565	$195,865
TOTAL	$95,300	$98,565	$2,000			$100,565	$195,865

HYBRID STRIPED BASS[5]

Project Component Termination Report for the Period
September 1, 1995 to December 31, 1997

NCRAC FUNDING LEVEL: $135,000 (September 1, 1995 to December 31, 1997)

PARTICIPANTS:

Michael L. Brown	South Dakota State University	South Dakota
Paul B. Brown	Purdue University	Indiana
Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
Jeffrey A. Malison	University of Wisconsin	Wisconsin
Robert J. Sheehan	Southern Illinois University-Carbondale	Illinois
Extension Liaison:		
Joseph E. Morris	Iowa State University	Iowa
Non-Funded Collaborators:		
Mike Freeze	Keo Fish Farm, Inc., Keo	Arkansas
Jerry Katt	Mid-Continental Fisheries	Illinois
Scott Lindell	AquaFuture, Turners Fall	Massachusetts
Robert Lyons	Lyons Enterprises, Morocco	Indiana
Gary Shirley	Shirley's Fish Farm, Lafayette	Indiana

REASON FOR TERMINATION

The objectives for this work on Hybrid Striped Bass were completed.

PROJECT OBJECTIVES

(1) Examine fry (phase I) to fingerling (phase II) production of three strains of white bass and three strains of hybrid striped bass (sunshine bass) in ponds with and without lights and vibrating feeders.

(2) Conduct field testing of fingerling (phase II) to advanced fingerling (phase III) production of three strains of hybrid striped bass (sunshine bass) in various culture systems.

PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

Adult white bass were acquired from three regions representing the extremes of the native range for white bass: Arkansas, South Dakota, and Lake Erie. Brood fish were held at Southern Illinois University-Carbondale (SIUC) and both white bass and hybrid striped (sunshine) bass fry were produced using these brood stock. At 4 days of age, the larvae were enumerated and subsequently stocked into fertilized 0.04 ha (0.10 acres) ponds 500,000/ha (202,350/acre). Training the fish to accept commercial diets began 21 days poststocking. At 36–41 days of age phase I fingerlings were harvested; survival rates

[5]NCRAC has funded five Hybrid Striped Bass projects. Termination reports for the first four projects are contained in the 1989-1996 Compendium Report. This project component termination report is for two objectives of the fifth project, which is chaired by Christopher C. Kohler. A progress report for the remainder of this project is contained elsewhere in this Annual Report. The fifth project continued and expanded upon the first four projects. It was a 2-year study that began September 1, 1995.

varied from pond to pond, but was generally poor (0–21% survival). Fish survival rates were markedly higher for hybrid striped bass ponds compared to white bass ponds, averaging 13% and 3%, respectively.

The phase I fingerlings were restocked for phase II production at SIUC. Due to a lack of fish, all three white bass stocks were eliminated from this segment of the experiment. Both Arkansas and South Dakota hybrid striped bass stocks were restocked in triplicate, while Lake Erie hybrid striped bass were only restocked in duplicate. The stocking rate used for phase II production was 25,000 fish/ha (10,118/acre). Fish were feed twice daily to satiation. At the end of the growing season, phase II fingerlings were harvested; survival rates ranged from 49–86%. Survival rates for both Arkansas and Lake Erie hybrid striped bass were about 72%, while the survival rate for South Dakota hybrid striped bass was only 57%. The average weight of individual fish also varied from stock to stock. The highest average weight was 90.2 g (3.2 oz) for Lake Erie hybrid striped bass, while South Dakota and Arkansas hybrid striped bass had average weights of 69.0 g (2.4 oz) and 58.4 g (2.1 oz), respectively.

In an attempt to feed-train hybrid striped bass fingerlings in ponds, 100,000 hybrid striped bass fry were stocked into a 0.4-ha (1.0-acre) pond at the University of Wisconsin-Madison's (UW-Madison's) facilities at the Lake Mills State Fish Hatchery, Lake Mills, Wisconsin in the spring/summer of 1997. The pond was equipped with a series of underwater lights and automatic feeders. The failure to observe any fish in the vicinity of the lights and feeders suggests that hybrid striped bass fingerlings are, or quickly become, photo-negative or photo-neutral; hence, the use of lights and automatic feeders to feed-train fingerlings in ponds does not appear to be an effective strategy for these fish.

Whole body proximate analysis was performed on phase II hybrid striped bass and white bass. Percent moisture for all strains of white bass (70.4%) was significantly higher than all hybrid striped bass strains (67.7%). Conversely, percent fat was significantly higher in hybrid striped bass (10.6%) than in white bass (9.0%). There was no difference in fat content, however, between Arkansas hybrids and South Dakota white bass. The only difference in percent protein was found between South Dakota white bass (15.5%) and South Dakota hybrid striped bass (14.2%). Percent protein for all other crosses were intermediate and not significantly different from either South Dakota taxon. White bass had significantly higher percent ash values than hybrid striped bass (4.1 and 3.6%, respectively). All hybrid strains and white bass strains were in separate groupings, but an intermediate grouping included Lake Erie white bass, South Dakota hybrids, and Arkansas hybrids. These results suggest that difference in body composition may exist between white bass and hybrid striped bass, but differences between strains are unlikely.

OBJECTIVE 2

Ponds

SIUC researchers investigated the use of ponds in producing phase III hybrid striped bass. Following phase II harvest, all three strains of hybrid striped bass were redistributed into newly filled ponds at 5,000 fish/ha (2,024 fish/acre). Fish were fed to satiation with a commercial trout feed (40% protein). Feeding rate was increased to twice per day during the warmer months, not exceeding the recommended 56 kg/ha (50 lb/acre). During the hot summer months of

the study, one replicate of each strain was lost from dissolved oxygen depletion.

Phase III fish were harvested in fall 1997 at the end of the second growing season. Percent survival averaged 90.4% in all ponds harvested. Lake Erie hybrids averaged 0.65 kg (1.43 lb), Arkansas hybrids averaged 0.64 kg (1.41 lb), and South Dakota hybrids averaged 0.57 kg (1.26 lb); mean weights were not significantly different. Production values averaged 2,853, 2,801, and 2,628 kg/ha (2,545, 2,499, and 2,345 lb/acre), respectively, for the above strains with no significant differences detected. Feed conversions were excellent (1.71, 1.49, and 1.67 for Lake Erie, Arkansas and South Dakota hybrids, respectively) and not significantly different. Fish grown in the small research ponds had production values similar to those obtained by the industry in large ponds when continual aeration is not provided.

Percent dress out for South Dakota hybrids (37.8%) and Lake Erie hybrids (37.3%) was significantly higher than that of Arkansas hybrids (34.6%). Sex ratios for all three strains were nearly 50:50. Gonadal Somatic Index of Lake Erie hybrids (1.4) was significantly higher than that of South Dakota (1.1) and Arkansas hybrids (1.0). No significant differences were detected between strains for Liver Somatic Index or Visceral Somatic Index.

Throughout all phases of pond production, Lake Erie hybrids performed as well or better than the other two strains of hybrid striped bass. The Lake Erie component of hybrid striped bass appears to be the strain of choice for North Central Regional aquaculture, but crosses with other strains of striped bass may yield different results. Perhaps the most promising results from phase III production is that commercially acceptable values for average weight, production, and feed conversion were achieved in an experimental setting in this region.

Tanks (Recirculation)

Two groups of hybrid striped bass fingerlings (Arkansas and South Dakota hybrids) were transported from SIUC to South Dakota State University (SDSU) to conduct strain comparison and density experiments. The culture system for both experiments consisted of 110-L (29.1-gal) glass aquaria connected as a closed freshwater recirculating system. Randomly selected fish from each strain group were stocked in individual aquaria to provide four replicates. The density experiment consisted of four replicates each of 5 (45/m^3; 1.3/ft^3), 15 (136/m^3; 3.9/ft^3) or 30 (273/m^3; 7.7/ft^3) South Dakota hybrids per 110-L (29.1-gal) aquaria. Performance characteristics (e.g., growth, conversion, condition, and survival) were monitored in both experiments. Strain comparisons were conducted at the low density.

Hand-feeding frequency was three times per day until the seventh week when belt feeders were incorporated, then feeding occurred continuously over the 12-h day period. Feed used was a commercial hybrid striped bass diet (38% protein, 5% lipid; Southern States, Farmville, North Carolina). Feed conversion was observed to decrease across all treatments when fish were switched to continuous belt feeding, as compared to three hand-feeding periods separated by 4-h intervals during the early portion of the study. Feed conversion differed little among treatments until week 11 of the study. During weeks 11 through 14 the high density treatment showed significantly poorer feed conversion than low and medium treatments. Strain comparisons did not reveal any conversion differences.

Growth pattern differences among treatments began to emerge during the second week of the study. Hybrid striped bass held at the medium density produced the largest proportional weight gain by the end of the study. Body condition at the conclusion of the experiment did not differ among treatments. No differences were detected between the two strains for growth and condition. Calculations from this study indicate that 200-mm (7.9-inch) hybrid striped bass can be safely reared at 8.8 kg/m^3 (0.6 lb/ft^3) up to 14.85 kg/m^3 (0.9 lb/ft^3) in recirculating systems.

Because uniform size is an important processing and marketing aspect, SDSU researchers examined the variability in individual weights over time to determine whether feeding hierarchies might have been established within tanks. There were distinct differences that corresponded with densities; however, the medium culture density provided fairly uniform fish size.

With the exception of ammonia, all monitored water chemistry was acceptable for the culture of hybrid striped bass. Ammonia concentrations in the high density treatment were sporadically well above baseline levels; those measurements were associated with feeding and observed to decrease shortly thereafter. Unionized ammonia concentrations in the high density treatment were not determined to be potentially toxic, rarely exceeding 0.011 mg/L (ppm) as NH_3-N.

The highest mortality (22%) occurred in the medium density treatment followed by the high (9%) and low (0%) density treatments. However, the mortality that occurred in the medium density treatment was a single event and a direct result of a plugged water jet.

Cages and Tanks

Studies at Purdue University were designed to examine maximum density of hybrid striped bass raised in cages and tanks. Two separate field studies were completed with two private producers in Indiana. In the first study, hybrid striped bass were grown through a production season and fed a standard diet. Initial densities ranged from 2.5–5.0 kg/m^3 (0.16–0.31 lb/ft^3) and final densities ranged from 6–31 kg/m^3 (0.17–1.95 lb/ft^3). There were no significant differences within or between sites. In the second field study, initial densities ranged from 4–18 kg/m^3 (0.25–1.13 lb/ft^3) and final densities ranged from 26–45 kg/m^3 (1.64–1.28 lb/ft^3). There were no significant difference within sites, but significant differences between sites were identified. Eviscerated dress out percentages were not significantly different in either study and ranged from 86–90%.

In the first tank culture study, densities ranged from 0.8–2.7 kg/m^3 (0.05–0.17 lb/ft^3) and final densities ranged from 8–28 kg/m^3 (0.23-1.6 lb/ft^3). Fish were fed a standard diet to satiation once each day and feed conversion ratios (FCR) ranged from 1.18–1.06. No significant differences were detected in weight gain, FCR, eviscerated dress out percentage, condition factor, or blood glucose or cortisol concentrations. In the second tank culture study, initial densities of 2.5–15 kg/m^3 (0.16–0.94 lb/ft^3) were used. At the end of the 86-day trial, weight gain and feed efficiency were not affected by the treatments in any of the stocking densities. There was no evidence of chronic stress in the fish. It appears that phase II hybrid striped bass can be stocked up to 15 kg/m^3 (0.94 lb/ ft^3) in recirculation systems; this finding is also reflective in SDSU studies.

IMPACTS

- The Lake Erie strain of white bass was identified as being the most suitable for the NCR of the strains evaluated.
- It has been demonstrated that hybrid striped bass grow as well or better in earthen ponds in the southern portion of the NCR as anywhere else in the United States.
- Many of the hybrid striped bass fingerling producers in the United States are adopting the white bass out-of-season spawning protocols developed through NCRAC funding.
- Stocking densities at levels best for fish production of hybrid striped bass cultured in ponds, tanks and cages have been identified

RECOMMENDED FOLLOW-UP ACTIVITIES

The combination of the findings of this study, identification of the best hybrid strain, along with the sperm storage protocols now being tested under industry settings, should allow for the culture of hybrid striped bass in the NCR. Continued demonstration of the technologies developed need to be undertaken with industry partners. In addition, economic analyses of hybrid striped culture at the densities identified in this study need to be undertaken.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See the Appendix for a cumulative output for all NCRAC-funded Hybrid Striped Bass activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1995-97	$135,000	$118,286	$55,019		$50,000	$223,305	$358,305
TOTAL	$135,000	$118,286	$55,019		$50,000	$223,305	$358,305

HYBRID STRIPED BASS[6]

Progress Report for the Period
September 1, 1995 to August 31, 1998

NCRAC FUNDING LEVEL: $15,000 (September 1, 1995 to August 31, 1998)

PARTICIPANTS:

Fred P. Binkowski	University of Wisconsin-Milwaukee	Wisconsin
Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
Joseph E. Morris	Iowa State University	Iowa
Extension Liaison:		
Joseph E. Morris	Iowa State University	Iowa

PROJECT OBJECTIVE

(1) Extension component:

(a) Coordinate selection of various culture systems and implement field testing (fingerling to advanced fingerling to food size).

(b) Write an initial culture manual using the information generated by all the hybrid striped bass research sponsored by the North Central Regional Aquaculture Center (NCRAC).

(c) Produce associated fact sheets, bulletins, and videos for hybrid striped bass research in the North Central Region (NCR).

ANTICIPATED BENEFITS

The overall goal for the five NCRAC-funded collaborative Hybrid Striped Bass projects is to enhance the culture potential of this fish in the NCR. The extension component of the fifth project will assure that the research information generated gets to the industry in a user-friendly form.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Kohler and Morris served as co-chairs for the NCRAC Hybrid Striped Bass Workshop that was held in November 1995 in Champaign, Illinois. The topics for the workshop included larval culture, cage culture, brood stock management, and an industry perspective. The 35 attendees were from Illinois, Iowa, Indiana, and Missouri. NCRAC-funded speakers included Chris Kohler, Sue Kohler, and Bob Sheehan of Southern Illinois University at Carbondale, George Brown and Joe Morris of Iowa State University, and LaDon Swann of Purdue University. Proceedings from this workshop are available from the NCRAC Publications Office at Iowa State University.

A hybrid striped bass fact sheet developed by Morris and Kohler has been reviewed and is

[6]NCRAC has funded five Hybrid Striped Bass projects. Termination reports for the first four projects are contained in the 1989-1996 Compendium Report; a project component termination report for two objectives of the fifth project is contained elsewhere in this Annual Progress Report. This progress report is for the remainder of the fifth project, which is chaired by Christopher C. Kohler. It was originally a 2-year study that began September 1, 1995.

scheduled for release in spring 1999. In addition, chapters and associated authors for a hybrid striped bass culture manual have been identified.

WORK PLANNED

The culture manual will be produced in two phases. In the first phase, the editors, Kohler and Morris, will review the current status of information using previously produced related materials, e.g., "Culture and Propagation of Striped Bass and its Hybrids" produced by the American Fisheries Society and "Farming a New Fish: Hybrid Striped Bass" available from North Carolina Sea Grant. The second phase will consist of producing materials that fill in the voids using information garnered from NCRAC research activities, e.g., nutritional and gamete storage and transportation. The culture manual will be completed in 1999 followed soon after by the development of associated videos.

IMPACTS

The proceedings from the 1995 NCRAC Hybrid Striped Bass Workshop has been used to address industry concerns and questions. The forthcoming culture manual should be useful in building upon previous related outreach materials in bringing forth new information to the public and in particular the aquaculture industry.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See the Appendix for a cumulative output for all NCRAC-funded Hybrid Striped Bass activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1995-98	$15,000	$16,968				$16,968	$31,968
TOTAL	$15,000	$16,968				$16,968	$31,968

SUNFISH[7]

Progress Report for the Period
September 1, 1994 to August 31, 1998

NCRAC FUNDING LEVEL: $373,562 (September 1, 1994 to August 31, 1998)

PARTICIPANTS:

Bruce A. Barton	University of South Dakota	South Dakota
Fred P. Binkowski	University of Wisconsin-Milwaukee	Wisconsin
Paul B. Brown	Purdue University	Indiana
Donald L. Garling	Michigan State University	Michigan
Robert S. Hayward	University of Missouri	Missouri
Terrence B. Kayes	University of Nebraska-Lincoln	Nebraska
Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
Jeffrey A. Malison	University of Wisconsin-Madison	Wisconsin
Joseph E. Morris	Iowa State University	Iowa
Douglas B. Noltie	University of Missouri	Missouri
Robert J. Sheehan	Southern Illinois University-Carbondale	Illinois
Robert C. Summerfelt	Iowa State University	Iowa
James R. Triplett	Pittsburg State University	Kansas
Industry Advisory Council Liaison:		
Charlie Stevens		Iowa
Extension Liaison:		
Joseph E. Morris	Iowa State University	Iowa
Non-Funded Collaborators:		
Denzil Hughes	Farmland Industries, Inc.	Kansas
Fountain Bluff Fish Farms		Illinois
Illinois Department of Conservation	Little Grassy State Fish Hatchery	Illinois
Jim Frey	Jim Frey Fish Hatchery, West Union	Iowa
Ron Johnson	Spruce Creek Fish Farm	Minnesota
Myron Kloubec	Kloubec Fish Farms, Amana	Iowa
Missouri Department of Conservation		Missouri
Tribal Council	Red Lake Band Chippewa	Wisconsin
National Biological Service	Midwest Science Center (formerly USFWS National Fisheries Contaminant Research Laboratory)	Missouri

[7]NCRAC has funded four Sunfish projects. Termination reports for the first project and all but the diet work of the second project are contained in the 1989-1996 Compendium Report. This progress report is for the third and fourth projects, which are chaired by Donald L. Garling and Robert J. Sheehan, respectively. These projects continued and built upon the first two projects. The third project was originally a 2-year study that began September 1, 1994; whereas the fourth project was originally a 2-year study that began September 1, 1996.

PROJECT OBJECTIVES

(1) Produce a production manual, accompanying videos, and other information as necessary to demonstrate the technology for culturing centrarchids.

(2) Determine the major nutritional requirements for centrarchids and to compare their growth and performance using available commercial feeds in laboratory and field settings.

(3) Determine the best feeding management strategies for culturing centrarchids in laboratory and field settings.

(4) Compare feeding trials for grow out of locally available 5.1–10.2 cm (2–4 in) black crappie (*Pomoxis nigromaculatus*) and male bluegill (*Lepomis macrochirus*) crossed with female green sunfish (*L. cyanellus*) hybrids in:

   (a) ponds at dissimilar latitudes in the region, and

   (b) recirculating systems using compensatory feeding strategies.

(5) Establish baseline physiological measures for small 2.5–7.6 cm (1–3 in) black crappie subjected to handling stressors and to test the effect of salt and temperature on stress reduction.

ANTICIPATED BENEFITS

A survey of the North Central Region (NCR), indicated that consumers and marketers wanted locally produced, farm raised, fresh fish products; marketers listed the bluegill as one of the top three species that they would like to purchase for their customers. A Sunfish Production Manual and associated video(s) for the NCR will provide research-based materials for information transfer to commercial producers and individuals interested in production of sunfish to meet these market demands.

Defining the critical nutritional requirements for targeted sunfish will enable development of diets that meet, but not exceed, their requirements; such findings will help to minimize feed costs, the largest annual variable cost in aquaculture. Protein requirements of sunfishes are poorly understood, which hinders their economic potential in food-fish culture.

Sunfish taxa that appear promising for food-fish production are the male bluegill × female green sunfish F_1 hybrid (BG × GS hybrid) and the black crappie. The BG × GS hybrid shows many of the characteristics sought in a commercial food fish, except that it appears to be difficult to economically grow to a size larger than 0.11 kg (0.25 lb). The black crappie appears to perform better than the white or hybrid crappie under culture conditions, and it may be more suitable than the BG × GS hybrid for production to a larger size. However, the black crappie has been difficult to habituate to commercial feeds, and it has shown poorer survival in tank and cage culture than is typical for the BG × GS hybrid. The fourth Sunfish project, which addresses Objective 4, will provide needed information on these two taxa.

There is a need for information that addresses the relative use of either fish for food-fish production in this region. Black crappie and BG × GS hybrid side-by-side pond production studies offer the best approach for providing this needed information. The fourth Sunfish project will provide information on whether stocking density and the inclusion of a period of

habituation to commercial feeds in the production cycle affects the performance of the two taxa. The Purdue University (Purdue) study will identify appropriate diets for BG × GS hybrids that can be purchased immediately. Further, growth rates, food conversion rates (FCRs), survival, and final weight at the end of the growing season at two different latitudes will be available from the Purdue and Southern Illinois University-Carbondale (SIUC) studies.

Recent evidence from the University of Missouri's (UM) compensatory growth (CG) studies (use of feeding and non-feeding strategies) with BG × GS hybrids indicates that a capacity exists for substantially reducing grow-out times for fish in aquaculture. Other possible benefits associated with CG include increased growth efficiency, influences on proximate composition of flesh, and delayed maturation.

Understanding the impact of CG on production in recirculating aquaculture systems is very important to commercial producers. Although reduced feed wastage is critical to optimize recirculating systems, fluctuations in ammonia levels from feeding and nonfeeding periods may result in performance problems by creating instabilities in the bacterial communities in biofilters. The application of process control technology in CG feeding trials at Pittsburg State University (PSU) will demonstrate the importance of continuous management on optimization of these production systems.

Clarifying the factors that influence the transition of pond-harvested black crappies to intensive tank culture will provide a foundation for industry efforts toward intensive food-fish production. Practical strategies of habituating large numbers of fingerlings to commercial diets and high rearing densities require that this transition be achieved in a cost-effect manner. University of Wisconsin-Milwaukee (UW-Milwaukee) researchers will explore the hurdles that need to be overcome. and determine the comparative costs of both methods.

Researchers at Iowa State University (ISU) have already demonstrated out-of-season spawning and production of fingerling sunfish raised in intensive culture. The present study at ISU compares performance of fingerling BG × GS hybrids produced by pond-spawned and tank-spawned parental stocks, and raised in a recirculating aquaculture system using compensatory and daily feeding regimens.

Initial experiments are required to establish the physiological response of black crappie to a standardized handling stressor as this has not been previously documented. Investigators at the University of South Dakota (USD) and the University of Wisconsin-Madison (UW-Madison) will conduct preliminary studies that will determine whether use of salt or temperature reduction may be useful for mitigating this stress response and reducing handling-related mortality.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

Michigan State University (MSU) and ISU personnel have completed drafts of a Sunfish Culture Guide; it is now being reviewed and should be completed during summer 1999. ISU personnel have produced a video addressing various production areas of BG × GS hybrids. The 8-minute video entitled "Sunfish (*Lepomis* spp.) Culture" is now available from the NCRAC Publications

Office at ISU. This video depicts methods for determining the sex of brood stock, species identification, and out-of-season spawning techniques.

OBJECTIVE 2

MSU

Researchers at MSU have empirically determined the optimal energy level for growth and protein retention in 125 mm (4.9 in) BG × GS hybrids utilizing a saturation kinetics model for curve fitting. Results demonstrate the semi-purified diet developed for these trials is well accepted by these fish; this results in a slightly lower but comparable growth to that obtained using a commercial control diet.

Purdue

Studies at Purdue were designed to quantify the dietary requirement for phosphorus (P) and the optimum lipid to carbohydrate ratio. Through four separate evaluations with four basal diet formulations, it appears the dietary requirement of BG × GS hybrids for P is $\leq$0.5% of the dry diet. In defining the optimum ratio of lipid to carbohydrate, researchers at Purdue compared BG × GS hybrids to pure bluegill. Pure bluegill grew significantly better than the BG × GS hybrid, a finding verified in a previous study. Based on weight gain, feed conversion, muscle lipid concentrations, and intraperitoneal (abdominal cavity lining) fat concentrations, both pure bluegill and BG × GS hybrids grow best when fed diets containing no less than 10% dietary lipid in the form of fish oil.

SIUC

Researchers at SIUC used practical diets containing crude protein levels of 32, 36, 40, and 44% and compared their ability to promote growth of BG × GS hybrids in recirculating aquaculture systems and ponds. Results indicated that under intensive culture conditions in recirculating aquaculture systems, where the formulated diet supplies virtually all the nutrition for the BG × GS hybrids, optimal crude protein levels are likely to be in excess of 40%. However, in ponds, where the diet is offered as a supplement to natural foods, a dietary crude protein level of 36% is adequate for promoting maximum mean harvest weight.

OBJECTIVE 3

ISU

One component of this objective was for ISU researchers to spawn sunfish (bluegill and BG × GS hybrids) out-of-season by manipulating temperature and photoperiod under laboratory settings. ISU researchers were able to spawn bluegills during a 6-month period (December 1994–May 1995); 40 spawns averaging 20,000 larvae each were obtained from 24 females. BG × GS hybrids were successfully produced fall 1997.

ISU researchers also developed a procedure for tank-rearing larval bluegill and larval BG × GS hybrids. Results indicate that the protocol for tank-rearing larval bluegill and larval BG × GS hybrids should include using brine shrimp nauplii (*Artemia franciscana*) prior to using a commercial diet. It appeared that larval BG × GS hybrids could digest the commercial diet at the onset of exogenous feeding. However, without brine shrimp nauplii much lower survival rates resulted. Survival rates of about 25 and 37% can be expected for bluegill and BG × GS hybrids, respectively, by following this protocol.

UW-Milwaukee

The primary goal of UW-Milwaukee researchers was to utilize the early life stage feeding technology that they have developed for yellow perch and apply this approach to

centrarchids, specifically, black crappie. UM-Milwaukee researchers have been able to habituate young-of-the-year (YOY) pond-raised black crappie to commercial diets using either adult frozen brine shrimp or "green tank" water organisms, which included copepods, ostracods, and smaller cladocerans, and later brine shrimp nauplii and a beef liver mixture. The fish became habituated withing 14–26 days.

UW-Milwaukee researchers have continued to expand their efforts to habituate YOY black crappie to formulated diets. Past efforts to spawn adults in the laboratory or to collect wild adults have not been successful. The group of YOY black crappies acquired in October 1994 have been retained for use as captive brood stock; they have been maintained on a rearing regime that is intended to promote gonadal development.

UM

Researchers at UM were able to demonstrate under laboratory conditions a CG response (increased growth following a period of fasting) for BG × GS hybrids fed mealworms (*Tenebrio molitor*). Over a 105-day experiment, mean growth rates of BG × GS hybrids in the 2 and 14 day no feeding cycle groups were 2.1 and 1.5 times faster, respectively, than the controls that were fed ad libitum every day. These results represent the first demonstration that fish can be grown significantly larger than daily-fed controls over identical time periods by eliciting a CG response. Growth improvements from CG appeared to result from increases in both consumption rate and growth efficiency.

OBJECTIVE 4

SIUC

Growth of black crappie and BG × GS hybrids were compared using common pond aquaculture techniques at SIUC. A growth trial was initiated in April 1997 and completed in November of the same year. A second, on-going growth trial was initiated in April 1998. Similar methods were employed in both years, but fish were habituated to prepared diets prior to stocking in the 1998 study. Two densities were examined for both taxa: 8,000 and 14,000 fish/ha (3,239 and 5,668 fish/acre). Fish were fed 40% protein Silvercup™ trout feed throughout the study.

In 1997, mean survival was 75% for BG × GS hybrids and 49% for black crappie. Net production for BG × GS hybrids was 621.6 and 943.2 kg/ha (554.5 and 841.3 lb/acre) in the 8,000 and 14,000 fish/ha ponds (3,239 and 5,668 fish/acre), respectively. Black crappie net production was -111.0 and -193.9 kg/ha (-99.0 and -173.0 lb/acre), respectively, in ponds stocked at 8,000 and 14,000 fish/ha. The crappie did not appear to be using the prepared diet.

In spring 1998, BG × GS hybrids from the 1997 trial were stocked into production ponds to evaluate third-year growth. Mean size had increased from approximately 104 g (3.7 oz) in early November 1997 to 150 g (5.3 g) in May 1998, the time when they were harvested and redistributed into grow-out ponds.

In response to their poor performance, black crappie were habituated to commercial diets prior to stocking in 1998 (BG × GS hybrids were similarly treated) at SIUC. BG × GS hybrids accepted the commercial diet virtually immediately, whereas the crappie did not. Freeze-dried mysid shrimp and

Biokyowa™ 2000 mini-pellets were then given to the black crappie as starter diets. Freeze-dried mysids were eagerly taken the first time they were offered, and the Biokyowa™ diet was also readily accepted within a few days.

Purdue

Research at Purdue was designed to evaluate varying levels of dietary crude protein fed to grow out BG × GS hybrids in earthen culture ponds. BG × GS hybrids were acquired from the same commercial producer who supplied fish for the laboratory studies in previous years and were stocked in triplicate at the Purdue University Aquaculture Research Facility.

UM

UM researchers evaluated the potential to increase growth rates of BG × GS hybrids via CG using commercial diets, instead of a natural food (mealworms), as used in previous studies (see above under Objective 3). Laboratory results indicated that CG for those fish fed a commercial diet was not exhibited to the same degree as in the studies using a natural food. Numerous experiments were conducted to determine why CG responses, e.g., hyperphagia (increased appetite after no-feed periods) did not result in increased growth compared to the controls when the BG × GS hybrids were fed commercial diets. Although not directly evaluated, the findings from UM's laboratory experiments hinted that time-of-year may be a critical determinant in regard CG. It could be that better CG responses occur only during late winter to early spring when control fish growth rates have been low despite summer-like temperature and photoperiod.

ISU

During 1997 and 1998, researchers at ISU conducted two, 12-week feeding trials. These feeding trials compared growth and food conversion of fingerling BG × GS hybrids derived from tank- and pond-spawning grown in a recirculating aquaculture system using compensatory feeding strategies. Brood fish were spawned indoors, and their offspring were raised on brine shrimp nauplii and then on formulated feed to fingerling size (tank spawning). Other brood fish from the same parental stock were spawned in ponds and their progeny were raised to fingerling size in ponds on natural food (pond spawning). In the first study, each stock was raised in separate tanks, but in the second study, each stock was distinctively marked and raised together in the same tank. In both feeding trials, the restricted feeding group was fasted 2 days every week followed by 5 days of full feeding (hereafter referred to as 2–5 feeding regimen).

Food conversion in the 2–5 feeding regimen and daily feeding groups were similar. Most differences in growth between the two feeding groups were not statistically significant, but when statistical significance was found in comparing the two groups, fish in the daily feeding regimen had faster growth than fish in the 2–5 feeding regiment group.

Fingerlings of both tank- and pond-spawned fish were of similar size when the experiments started, but in both the 2–5 feeding regimen and the daily feeding treatment groups, growth was faster and feed conversion lower for pond-raised fingerlings than tank-spawned fingerlings. The most probable explanation for these results is the genetic differences among individual parental stocks, there was less

genetic diversity for the tank-reared fish because of the use of fewer brood stock.

UW-Milwaukee

UW-Milwaukee researchers continue to maintain a 1994 year class of black crappie captive brood stock. Using temperature and photoperiod as environmental conditioners for gonadal maturation they have been successful in producing a limited number of gametes. However, fertility was low, probably due to poor gamete quality. They speculate that the fish were too young to spawn successfully. They anticipate the fish will spawn in spring 1999.

As a back-up to their efforts to produce YOY from laboratory and wild spawns, UW-Milwaukee researchers obtained pond-spawned YOY black crappies from the Gavins Point National Fish Hatchery in Yankton, South Dakota. When offered brine shrimp nauplii on the day of arrival, approximately half the fish accepted the food. Trial feedings with commercial diets on the day of arrival were unsuccessful. These fish took longer to habituate to commercial diet than either the slightly larger YOY brought to the lab in October 1994, those habituated to a commercial diet within 14 days, or the larval crappies tested in July 1995 that habituated to commercial starter diet within 26 days. These results suggest that there is a strong preference for brine shrimp nauplii, and that habituation is not readily achieved by merely offering the commercial diet along with the transitional live food. Although there was limited interest in commercial diets as early as 6 days after the beginning of the trial, the fish consumed mainly brine shrimp nauplii. Full habituation to commercial diet appeared to closely follow the forced restriction of the live food. Survival during the trial was excellent; 99% over a rearing period of 103 days. UW-Milwaukee researchers intend to continue rearing this group of fish to demonstrate the growth that can be achieved under intensive flow-through culture with formulated diets.

PSU

Work at PSU has attempted to compare feeding trials for grow out of black crappie in recirculating systems under compensatory feeding strategies. Unfortunately most feeding trials to date have been unsuccessful, either because of water quality problems in the culture system or the lack of being able to train the fish to accept commercial feeds. However, one feeding trial using larger black crappie, 15.0–20.0 cm (5.9–7.9 in), obtained from Culver Fish Farm in McPherson, Kansas was completed during 1997-1998. Results indicated that there was no differences in food consumption between fish fed daily or those that were withheld feeds for either 2, 3, or 4 days when using Biodiet™ as the feed.

OBJECTIVE 5

USD and UW-Madison

Juvenile black crappie were reared extensively at the Gavins Point National Fish Hatchery in Yankton, South Dakota. In 1996 fish were collected by hatchery staff and transported directly to USD in late summer (pond temperature 21°C [69.8°F]).

Juvenile black crappies (mean weight, 2.6 g [0.4 oz]; mean total length, 59 mm [2.3 in]) were subjected to a 30-sec aerial emersion handling stressor and sampled by USD and UW-Madison researchers. Plasma samples were pooled for analysis because of the small volume; each sample thus represents a pool of plasma from 2–10 fish, depending on fish size. Samples were analyzed for levels of cortisol and chloride.

Preliminary results, subject to more complete statistical analysis, indicate that plasma cortisol was elevated following the disturbance from a mean pre-stress level of 46 ng/mL and reached a peak response of 231 ng/mL by 0.5 h. This response pattern is characteristic of those observed in other fishes generally and similar quantitatively to what has been reported in other non-salmonid species. Plasma cortisol was reduced by 3 h and returned to a level of 87 ng/mL by 24 h.

Plasma chloride levels declined from 73 meq/L in response to the stress by 0.5 h and reached their lowest value of 64 meq/L at 24 h after the disturbance. As with cortisol, this response is characteristic of those reported in other non-salmonid species, e.g., walleye and largemouth bass, and indicate a compromised osmoregulatory ability resulting from the stress. However, sample sizes for chloride determination were limited and the pre-stress value was also low compared with those found for other species.

WORK PLANNED

The Sunfish Culture Guide is scheduled for completion in 1999. Final analysis of the nutritional and proximate composition of fillets for SIUC data will be analyzed in 1999 and included in a termination report. Ponds used in the 1998 trial at SIUC will be harvested in November. Production, survival, mean length and weight, dress-out, feed conversion efficiencies, and liver and gonadal somatic indices will be determined and compared among treatments. Also, the hybrid sunfish in their third year of growth will be harvested and evaluated in November.

PSU researchers have been given a time extension to complete feeding trials of black crappie in their recirculating aquaculture system using compensatory feeding strategies. They will evaluate both a commercial feed as well as a natural food.

Additional stress response work on black crappie is planned by USD and UW-Madison for late October 1998 after the fish have acclimated to the laboratory environment following delivery from the hatchery. The objective of 1998 studies is to evaluate the use of altered water temperature and addition of salt to the recovery media as means of mitigating the physiological stress response associated with handling and transport and to enhance survivorship.

Fish acclimated to 25°C (77°F) will be stressed by subjecting them to a 30-sec handling stressor as in 1997. To simulate a transport scenario, handled fish will be transferred to one of the following conditions for 4 h: (1) 25°C (77°F) without salt (control), (2) 25°C (77°F) with 0.7% NaCl, (3) 20°C (68°F) without salt, or (4) 20°C (68°F) with 0.7% NaCl. After 4 h, fish will be returned to ambient conditions as they would if they were being stocked. Fish will be sampled before handling, at selected times after handling and transfer to experimental conditions, and after return to ambient conditions. Blood samples will be analyzed for cortisol and chloride levels; it is unlikely because of the small blood sample sizes provided by crappies of this size that additional physiological measurements will be possible.

IMPACTS

- Diets that have been specifically developed for targeted sunfish species should result in maximum performance at the lowest possible cost which in turn will help to maximize profit for the producer.

- It now appears that the intensive culture technology developed for yellow perch can be applied to black crappie.
- It is now possible to produce out-of-season bluegills and BG × GS hybrids in the laboratory by manipulation of temperature and photoperiod without the use of hormones.
- UM's findings suggest that the CG response differs according to the type of feeds used (natural versus commercial feeds).
- Two fish farmers in Iowa, one in Minnesota, one in Wisconsin, and another outside of the NCR are now using information on tank spawning of BG × GS hybrids developed by ISU.
- Pond studies at SIUC showed that hybrid sunfish grow rapidly during the second year of the production cycle, but not fast enough when beginning with small (6 g; 0.2 oz) fingerlings to produce highly desirable, market-size 227–340 g (0.50–0.75 lb) by the end of year 2. This suggests a 3-year production cycle may be necessary to achieve 227–340 g (0.50–0.75 lb) fish. Their studies also clearly showed that production methods suitable for BG × GS hybrids are not suitable for black crappie. The latter showed poor growth and survival, and they appeared to be subsisting on the natural food supply rather than the prepared diet. However, great success was obtained with crappie with two starter diets: freeze-dried mysids and Biokyowa™. The freeze-dried mysids were voraciously accepted by black crappie the first time it was offered — the intensity of the black crappie feeding response to the mysids was far greater than had been seen before in this species with non-living feeds. More work is needed to develop methods for weaning crappie from the starter to production diets.
- ISU's feeding trials of BG × GS hybrids in recirculating aquaculture systems indicate that family differences in performance characteristics are invariably observed. Thus, effort must be undertaken to avoid a culture program based on a limited gene pool.
- USD and UW-Madison results from 1997 indicate that black crappies are stressed from simple handling, the effect of which was still apparent after 24 h. Their results also indicate that routine handling, at least under these experimental conditions, was insufficient to cause significant mortality.

PUBLICATIONS, MANUSCRIPTS, AND PAPERS PRESENTED

See the Appendix for a cumulative output for all NCRAC-funded Sunfish activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1994-96	$173,562	$177,300	$12,012[a]			$189,312	$362,874
1996-98	$200,000	$146,368				$146,368	$346,368
TOTAL	$373,562	$323,668	$12,012			$335,680	$709,242

[a]Farmland Industries, Inc.

SALMONIDS[8]

Progress Report for the Period
September 1, 1997 to August 31, 1998

NCRAC FUNDING LEVEL: $80,403 (September 1, 1997 to August 31, 1998)

PARTICIPANTS:

Paul B. Brown	Purdue University	Indiana
Konrad Dabrowski	Ohio State University	Ohio
Donald L. Garling	Michigan State University	Michigan
Jeffrey A. Malison	University of Wisconsin-Madison	Wisconsin
Industry Advisory Council Liaison:		
David A. Smith	Freshwater Farms of Ohio, Inc., Urbana	Ohio
Extension Liaison:		
Ronald E. Kinnunen	Michigan State University	Michigan
Non-funded Collaborators:		
Myron Kebus	Wisconsin Aquatic Veterinary Service, Madison	Wisconsin
David Mueller	Rushing Waters Fisheries, Inc., Palmyra	Wisconsin
David A. Smith	Freshwater Farms of Ohio, Inc., Urbana	Ohio
Brad Strahm	Wenger, Inc., Sabetha	Kansas
Kathy Warner	National Center for Agricultural Utilization, ARS, USDA, Peoria	Illinois
Y. Victor Wu	National Center for Agricultural Utilization, ARS, USDA, Peoria	Illinois
M. Randall White	Purdue University	Indiana

PROJECT OBJECTIVES

(1) Develop and evaluate practical and economically viable diets that are fish-meal free or as fish-meal free as practical:
- using soy, or other oil-seed products that are regionally available, and
- using Shasta, Donaldson and Kamloop strains of rainbow trout and/or Arctic charr for the evaluation.

(2) Evaluate the effects of water temperature on the growth/stress response in salmonid strains or species (as listed in Objective 1) under outdoor commercial culture conditions in the upper and lower portions of the North Central Region.

(3) Investigate the effects of trace mineral supplementation on the growth and stress response of rainbow trout in high

[8]NCRAC has funded four Salmonids projects. Project component termination reports for objectives of the first two projects are contained in the 1989-1996 Compendium Report; termination reports for the remainder of the first two projects and all of the third are contained in the 1996-1997 Annual Progress Report. This progress report is for the fourth project, which is chaired by Paul B. Brown. The fourth project built upon the first three projects. It is a 2-year study that began September 1, 1997.

density culture, as evaluated by plasma cortisol levels and fin nipping behavior.

ANTICIPATED BENEFITS

OBJECTIVE 1

Work proposed and conducted under this objective is designed to develop diets for rainbow trout that are free of fish meal and rely on feed ingredients common in the North Central Region (NCR). This line of research will result in diets that are lower in cost, which will reduce overall production costs, and can be taken to local feed mills and thus reduce transportation costs.

North Central Regional Aquaculture Center (NCRAC)-funded research has shown that pretreatment of plant feedstuffs with the enzyme phytase can help improve utilization of phosphorus (P) and nitrogen (N) in limited fish meal and all plant diets for rainbow trout. In addition, insulin-like growth factor (IGF-1) levels can be used as a rapid indicator of nutritional status in rainbow trout.

OBJECTIVE 2

These studies will provide detailed information on the growth and stress responses of Kamloops and Donaldson strains of rainbow trout and Arctic charr reared under thermal conditions typically found in the NCR. Regional salmonid producers will be able to use this information to determine which of the three species/strains can be best utilized at their facility (i.e., under their specific thermal conditions) to maximize productivity and profitability.

OBJECTIVE 3

A survey will assess how widespread fin nipping behavior in rainbow trout is present in the NCR and determine if research needs to be done to address this problem if it exists.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

In the most recent study with rainbow trout fed fish meal-free diets, researchers at Purdue University (Purdue) evaluated the effects of supplemental and nutritionally complete vitamin and mineral premixes in addition to new sources of lipid, and compared growth and feed conversion of fish fed nine different formulations to those fed two different control diets containing fish meal as the primary source of crude protein. The basal experimental formulation was one identified in previous studies that contains 44% soybean meal and 31% corn gluten meal. In these studies, either the standard U.S. Fish and Wildlife Service vitamin or mineral premixes or Purdue nutritionally complete premixes were incorporated. Additionally, researchers incorporated flax, a new cold pressed soybean or fish oils, alone and in combination, into diets. Weight gain and feed conversion ratio were not significantly different in fish fed two of the experimental diets compared to fish fed the control diet. The best experimental diets contained the U.S. Fish and Wildlife Service mineral premix and the nutritionally complete vitamin premix from Purdue. Thus, vitamin supplementation seems to be an important factor in formulating diets that are free of fish meal.

Researchers at Ohio State University (OSU) compared the growth rates of rainbow trout fed six experimental diets in which fish meal protein was replaced with animal by-product protein mixture (APM) or with APM- and plant-protein mixtures (PPM). PPM was comprised of equal proportions of soybean and cottonseed meal. Three different regional sources of cottonseed meal (Chowchilla, California; Memphis, Tennessee; and Pine Bluff, Arkansas) were tested. These cottonseed meals differed in

free and total gossypol concentrations. Anticipated effects of diet formulations with cottonseed meal include gossypol toxicity, phytoestrogen effects, and decreased availability of essential amino acids. All diets contained 45–47% protein and 13.9–14.4% lipids. Initial weight of juvenile rainbow trout was 0.96 ± 0.03 g (0.034 ± 0.001 oz) and the experiment lasted 16 weeks. Water temperature increased gradually from 8–15°C (46.4–59.0°F). There were significant differences in growth between fish fed a control diet with fish meal protein only (1,370% body weight increase) and fish fed diets with 50 or 100% replacement with animal protein mixtures (1,129 and 1,235%, respectively). Fish fed two of the cottonseed meal containing diets (without fish meal) performed equally well as those fed the control diet (1,330–1,350%). Significant decreases in hematocrit values were observed in fish fed all experimental diets (39–40%) in comparison to control (44%). Free gossypol analysis in livers of juvenile rainbow trout revealed concentrations (1.22–1.78 ppm) which are much lower than described earlier in the literature in adult rainbow trout fed diets containing high levels of gossypol.

At Michigan State University (MSU) the IGF-1 isolation procedure has been refined for rainbow trout protocols. The first growth trial is near completion. Tanks were stocked with rainbow trout and experimental or reference diets were randomly assigned. Dietary treatments consisted of: (1) negative reference (sub-optimal protein), (2) positive reference (fish-meal based), (3) soybean meal substituted-untreated diet, and (4) soybean meal substituted-pretreated with phytase. Experimental diets were formulated with a 35% crude protein, protein to energy ratio of 100, and vitamin and mineral premixes added to meet the requirements of the fish. A two-week acclimation period was completed prior to the start of the study to allow fish to adjust to their new surroundings and feeding regime. Diets have been fed three times a day to three replicates of 16 fish per treatment. Feed rates were based on a percent body weight basis, determined during the two-week acclimation period by feeding the fish to satiation. Fish have been weighed every two weeks and feed rates have been adjusted according to weight gain. Preliminary results indicate that the phytase treated diet is out-performing the fish meal and the soybean meal untreated diet. This is the same trend that had been observed earlier by MSU researchers.

OBJECTIVE 2

The plan of work in the initial proposal was to conduct studies on Arctic charr and Donaldson strain rainbow trout at Rushing Waters Fisheries, Inc., Palmyra, Wisconsin. By the time of initiation of the project, however, Rushing Waters personnel determined that they would be unable to conduct the study. Accordingly, as per the backup plan detailed in the original proposal, the Arctic charr study was conducted at the University of Wisconsin-Madison (UW-Madison) campus.

To compare the stress responses of Arctic charr and Kamloops rainbow trout at different temperatures, approximately 120 Arctic charr (mean total length [TL] 160 mm [6.3 in], weight 31 g [1.1 oz]) and 100 Kamloops rainbow trout (mean TL 150 mm [5.9 in], weight 34 g [1.2 oz]) were obtained from Rushing Waters Fisheries, Inc., Palmyra, Wisconsin, and Trout Haven, Bryant, Wisconsin, respectively. The fish were held for three weeks in separate 750-L (198-gal) flow-through tanks at a water temperature of 12.5°C (54.5°F). The fish were then weighed, measured, and 25 fish were transferred into each of four 120-L (32-gal) flow-through tanks (two tanks of

rainbow trout, two of arctic charr). Over the next two weeks, the water temperature in one tank of each species was gradually raised to 15°C (59.0°F), and lowered to 10°C (50.0°F) in the other two tanks. After three weeks of acclimation, six fish from each tank were quickly removed, anesthetized, and bled via the caudal vasculature. The remaining fish were given an acute stress challenge test by holding them out of the water for one minute, then randomly placing them into separate tanks. Groups of six fish were then netted, anesthetized, and bled at 1, 3, and 24 h following the stressor. Assays for measuring serum cortisol, glucose, and chloride levels in Arctic charr are currently being developed, but the samples from the study described above have not yet been measured.

OBJECTIVE 3
No progress has been made during this reporting period.

WORK PLANNED
OBJECTIVE 1
In the next cycle of this project, research at Purdue will offer the new formulations mentioned above to various species of salmonid (Arctic charr) and strains of rainbow trout (Shasta, Kamloops, and perhaps Donaldson) and compare growth and feed conversion of fish fed the experimental diets to those fed commercial diets.

Mr. K.J. Lee will continue to work on related rainbow trout experiments to complete his Ph.D. dissertation at OSU in the first quarter of 2001. Fish body and fecal material will be analyzed for nitrogen, chromium, lipids, and ash, to complete nutritional evaluation of diets in relation to fish performance.

Tanks at MSU will be stocked with rainbow trout and experimental or reference diets will be randomly assigned. Dietary treatments will consist of: (1) negative reference (fasted), (2) positive reference (fish-meal based), (3) reference diet, (4) soybean meal substituted-untreated diet, (5) soybean meal substituted-pretreated with phytase, (6) fish meal limited-untreated diet, and (7) fish meal limited-pretreated with phytase.
Experimental diets will be formulated with a 35% crude protein, protein to energy ratio of 100, and vitamin and mineral premixes added to meet the requirements of the fish. A two-week acclimation period will be given to the animals prior to the start of the study to allow them to adjust to their new surroundings and feeding regime. Diets will be fed three times a day to three replicates of 16 fish per treatment for a period of at least six weeks. Feed rates will be calculated on a percent body weight basis, which will be determined during the two-week acclimation period by feeding the fish to satiation. Fish will be weighed every two weeks and feed rates will be adjusted according to weight gain. Waste products will be collected until the termination of the experiment and effluent P and N concentrations will be estimated by using a modified mass balance equation:

$$P_{ho} = P_{fed} - [P_f + (P_{tm} - P_{ti})]$$

where:
P_{ho} = P in effluent of hatchery origin
P_{fed} = P in feed
P_f = P in feces
P_{tm} = P in fish at end of growth period
P_{ti} = P in fish at beginning of the growth period.

OBJECTIVE 2
The blood samples will be analyzed and data summarized for the Arctic charr experiment. In the next year UW-Madison researchers

are planning to conduct an experiment comparing the stress responses of Donaldson and Kamloop strains of rainbow trout. Their present plans are to conduct this study at Freshwater Farms of Ohio, Inc., Urbana, Ohio, as described in the original proposal.

OBJECTIVE 3
MSU will conduct a survey to assess how widespread fin nipping behavior in rainbow trout is in the NCR. The survey will be mailed to a representative sample of trout producers in the NCR in the upcoming year.

IMPACTS
OBJECTIVE 1
It appears that research at Purdue has developed a dietary formulation that is free of fish meal and results in the same weight gain and feed conversion ratio as fish fed a standard trout diet. This will allow producers in the NCR the opportunity to take these formulations to local feed mills, use local ingredients, and significantly reduce feed costs.

Research at OSU has provided strong evidence that a diet with 25% cottonseed meal can be used to produce grow-out diets for rainbow trout without compromising growth rate. Taking into account current (August 3, 1998) prices for menhaden fish meal ($0.62/kg $560/ton) and cottonseed meal ($0.15/kg; $140/ton), this replacement should make a considerable difference in feed costs. It is recommended, however, that cottonseed meal should be used with caution for rainbow trout production because of the phytoestrogens and gossypol content. Further studies need to be conducted on possible impact of these substances on sex differentiation in salmonids. No more than 50% protein replacement with plant-protein and 50% with animal-protein analog mixtures is suggested in juvenile rainbow trout diets.

Improved utilization of dietary phosphorus and nitrogen will reduce the impact of aquaculture on the water quality of streams receiving discharge from fish farms. MSU research has demonstrated that phytase can be used to improve utilization of P and N and will determine if IGF-1 can be used as a rapid indicator of nutritional status in rainbow trout.

OBJECTIVE 2
The identification of trout strains or species which show improved performance under the thermal conditions found in the NCR will maximize productivity and profitability of aquaculture facilities in the region. In addition, the availability of rainbow trout strains or species with improved growth rate, feed conversion and disease resistance will greatly improve the production efficiency of private and public fish hatcheries throughout the region.

OBJECTIVE 3
The incidence of fin nipping behavior in rainbow trout in the NCR will be determined and, if widespread, measures for its prevention will be assessed.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED
See Appendix for a cumulative output for all NCRAC-funded Salmonid activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1997-98	$80,403	$92,640				$92,640	$173,043
TOTAL	$80,403	$92,640				$92,640	$173,043

WASTES/EFFLUENTS[9]

Progress Report for the Period
September 1, 1996 to August 31, 1998

NCRAC FUNDING LEVEL: $100,000 (September 1, 1996 to August 31, 1998)

PARTICIPANTS:

Ira R. Adelman	University of Minnesota	Minnesota
Fred P. Binkowski	University of Wisconsin-Milwaukee	Wisconsin
Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
Industry Advisory Council Liaison:		
Harry Westers		Michigan
Extension Liaison:		
LaDon Swann	Purdue University	Illinois/Indiana
Non-Funded Collaborators:		
Antony Grabowski	Milwaukee County House of Correction Fish Hatchery	Wisconsin
John Hyink	Glacial Hills, Inc./Alpine Farms	Wisconsin
Jeffrey A. Malison	University of Wisconsin-Madison	Wisconsin
John Wolf	Glacial Hills, Inc./Alpine Farms	Minnesota

PROJECT OBJECTIVES

(1) Study and evaluate solid waste management by:

   (a) describing the relevant physical characteristics of fecal material from fish fed commonly used commercial feeds,

   (b) developing diets to maximize integrity of fecal pellets without loss of fish performance and compare the physical characteristics of these pellets to those in subobjective a, and

   (c) developing operational and engineering solutions to minimize destruction of larger particles and to remove all particulates.

(2) Develop a report that:

   (a) Describes the potential benefits of aquacultural by-products (effluents and solids) in the context of Integrated Resource Management and Sustainable Development,

   (b) Characterizes the differences between the aquacultural discharges and other agricultural and industrial discharges, and

[9]NCRAC has funded two Wastes/Effluents Projects. The termination report for the first project is contained in the 1989-1996 Compendium Report. This progress report is for the second project, which is chaired by Fred P. Binkowski. It was originally a 2-year study that began September 1, 1996.

(c) Identify case studies of previous controversies highlighting real versus perceived impacts of aquaculture.

ANTICIPATED BENEFITS

Characterization of the possible differences in fecal waste properties of important regional alternative species will assist in the engineering design and operation of rearing systems for waste removal.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1A

University of Wisconsin-Milwaukee (UW-Milwaukee)

Researchers investigated the physical properties of freshly deposited feces generated through intensive tank culture of yellow perch fed commercial feeds at various phases of the production cycle at commercial rearing densities and grow out temperature (18–23°C; 64.4–73.4°F). The phases examined to date include: (1) perch approaching market size (approximately 100–150 mm [3.9–5.9 in] total length) and fed Zeigler Bros. trout grower, (2) mature perch at marketable sizes (>150 mm; >5.9 in) fed Zeigler Bros. trout grower, (3) advanced fingerlings (50–100 mm; 2.0–3.9 in) fed Zeigler Bros. salmon starter, and (4) young fingerlings (25–75 mm; 1.0–3.0 in) habituated to feed exclusively on Biodiet #2 starter feed.

Initial attempts at individually isolating or anesthetizing fish to collect freshly egested fecal material were found to be unreliable in obtaining enough material to conduct investigations of specific gravity and fecal friability. Therefore, a low-head, side-stream siphoning device was constructed and installed on circular rearing tanks so that freshly settled fecal materials and uneaten food could be collected from the sump of the rearing tanks. The collecting basin (18.9-L [5.0-gal] pail) for the siphoned material was raised so that the water surface in the collection basin was just below that of the rearing tank. For 2.44-m (8.0-ft) diameter tanks, a smooth semi-flexible clear vinyl tube (approximately 28-mm [1.1-in] diameter) and a side-stream flow of around 4–7 L/min (1.1–1.8 gal/min) was used to siphon waste. In this way the water velocity within the siphon was low enough (adjustable in the range of <1–15 cm/sec; <0.4–5.9 in/sec) to collect the fecal material with minimal handling and disturbance. The clear tube allowed visual monitoring of the condition of the waste during collection. To insure freshly deposited material, the rearing tank was cleaned at the start of each collection period. By arranging for the inflow to the tank to rotate the water, settled material would rapidly settle and collect in the sump at the base of the central standpipe of the circular rearing tanks. By attaching the siphon tube to collars mounted on the outer diameter of the standpipe and holding the collecting end at a narrow fixed height above the bottom in the central collecting sump, settled solids could be continuously removed from the rearing tank. For perch larger than approximately 100 mm (3.9 in) in size this collection device could be left unattended even over night without fish entering the waste collection stream. Small fingerlings tended to enter the waste collector even though the opening was positioned very close to the tank bottom and feces from these fish were collected by hand-directing the low-head siphon hose.

In a tank of adult perch at a rearing density of 84 kg/m^3 (5.2 lb/ft^3) fed a ration of 2.4% (approximately 2 kg; 4.4 lb) this low-head siphon device would collect 4–5 kg (8.8–11.0 lb) of sludge that was 8–10% solid on a dry weight basis (320–500 g; 0.7–1.1 lb). This represents a recovery of 17–26% of the food solids entering the tank on a dry

weight basis. The literature reports that for salmonids, 25–30% of the dried weight of food is converted to feces on a dry weight basis. Therefore, between 69% to nearly 100% of the excreted solids produced in the rearing tank were removed by this simple device that concentrates the recovered waste in a side-stream of 4–5% of the tank's water inflow. In effect, with only a few pieces of inexpensive hardware, the rearing tank itself acts like a "swirl concentrator." With improvement this device could be incorporated as an inexpensive initial clarifier component, prior to further fine solids removal, in recirculating aquaculture systems (RAS). Fecal waste and uneaten food particles are removed directly from the rearing tank relatively intact before they are further broken up by more turbulent components of RAS.

Overall, freshly deposited feces (N = 887) ranged from 0.4–6.2 mm (0.02–0.24 in) in diameter and 0.6–23 mm (0.02–0.91 in) in length. Median fecal pellet diameters were 0.7 mm (0.03 in) (N = 344), 1.6 mm (0.06 in) (N = 240), 2.6 mm (0.10 in) (N = 182), and 3.2 mm (0.13 in) (N = 121) for perch in the size categories of 25–75 mm (1.0–3.0 in), 50–100 mm (2.0–3.9 in), 100–150 mm (3.9–5.9 in), and >150 mm (>5.9 in) total length, respectively. The corresponding median lengths of intact fecal particles for these size categories were 4.8 mm (0.19 in), 4.0 mm (0.16), 6.7 mm (0.26 in), and 5.4 mm (0.21 in), respectively. Fingerling perch on the Biodiet feeds tended to have longer feces in relation to their diameter and the feces tended to lack the multifolded rough character of the pellets of larger sized perch fed the Zeigler feeds.

Settling velocities of individual fecal and food particles were determined in a 180 cm (70.9 in) high settling column (10-cm [3.9-in] diameter). As anticipated by Stoke's law, settling velocities increased with increasing particle size and density. Settling velocities for feces increased gradually over a range of 0.4–5.0 cm/sec (0.16–1.97 in/sec) (N = 204) with increasing fish size. The settling velocities of the intact food granules and pellets were higher (5.0–16.0 cm/sec; 1.97–6.30 in/sec) than settling velocities of feces of similar diameter, except for the smaller granules of Biodiet #2 starter and feces of the fingerling perch with diameters <1 mm (<0.004 in) and settling velocities from 0.7–3.2 cm/sec (0.28–1.26 in/sec) and 0.4–1.8 cm/sec (0.16–0.71 in/sec), respectively. This difference mainly reflects the higher density of the pelletized food compared to the less dense fecal material after passage through the digestive tract.

Specific gravity measurements of freshly collected fecal solids were made before and after uniformly compressing the water from approximately 50 mL (1.7 oz) of collected sludge by centrifugation for 5 min at 2,500 rpm, pouring off the water, determining the resulting volume and weight of solid material and comparing it to the weight of an equal volume of deionized water and correcting for temperature. The overall mean specific gravity by this method was 1.055 (SD = 0.019; N = 36) after centrifugation and 1.029 (SD = 0.013; N = 24) without centrifugation. Differences in specific gravity of feces based on the type of food used were not detectable. It appeared that these mass techniques might create uniform conditions due to consistent packing of the material, while individual fecal particles can vary considerably in compactness and durability. Perhaps measuring settling velocities directly will be more expedient than using a representative value for specific gravity, that may be influenced by collecting and compaction techniques, to infer settling rates using Stoke’s law. It may be better to measure the settling velocity and calculate

specific gravity using Stoke's law and to use these values to make further estimates of settling velocities.

Initial attempts to examine the friability of feces of larger perch fed Zeigler grower diets using direct observation of small numbers of fecal particles in water in Erlenmeyer flasks were unsuccessful due to the heterogeneous nature of the fecal particles. Some of the larger fecal pellets appeared to consist of fine solid material approximately the size of the finely milled material in the formulated diet, encased in an outer more durable mucus-like shell that either was a solid smooth-surfaced stream of material or became folded on itself and compressed into a larger diameter rough surface pellet. In some fecal pellets the proportion and durability of the outer casing appeared to vary in thickness and a relatively small proportion of fine material was inside this casing. These fecal pellets were extremely compressed and durable and tended to have a grayish to white color compared to the less durable brownish type encasing large amounts of finely milled material. Some fecal pellets tapered along their length from wide easily friable character to the more durable grayish cast. This complicated the visual determination of when the pellet was completely broken down. For this reason it seemed better to use a larger (45–70 mL; 1.5–2.4 oz), more representative sample of fecal material to quantify fecal friability. Samples of intact settled fecal material collected by low-head siphoning and an approximate settled volume of 45–70 mL (1.5–2.4 oz) of material was scooped into a 150 mL (5.1 oz) graduated beaker, the contents of this beaker were poured into a 250 mL (8.5 oz) Erlenmeyer flask capped with parafilm and subjected to mechanical agitation of 0, 5, 15, 30, 60, 120, and 240 sec duration at 300 rpm on an orbital rotary shaker. The contents of the flasks were then poured into Imhoff cones and the settleable solids determined along with the volume of intact particles by visually determining the boundary between the fine broken settled solids (less than the diameter of intact fecal material) and the more rapidly settled mainly intact fecal material. The difference in settled volume of the intact feces versus the fines as a percent of the settled solids in the cone was used to express the degree of breakdown. Four or five repeated samples at each time duration were used to express the breakdown of fecal material over time.

Using this technique it was found that the feces of the larger perch fed Zeigler grower and salmon starter diets rapidly decreases from 60–80% intact material to around only 20% of the durable type of intact fecal material after only 5–60 sec of agitation at 300 rpm, while fecal material of fingerling perch fed Biodiet #2 starter also started out 70–80% intact and tended to remain around >60% intact even after 240 sec at 300 rpm. The fineness of the milling of the various components appeared to influence the durability of the fresh fecal material. The coarser "fines" in the grower diet fed to the larger sized perch appeared to give the fecal material a more friable consistency.

The smaller diameter and proportionately greater length of the intact fecal particle of fingerling perch fed semi-moist starter diet contributes to a slower rate of settling. This proportionately greater length suggests a greater resistance to mechanical agitation and further breakdown.

Southern Illinois University-Carbondale (SIUC)

To date most of what has been done on this component of the project involves building the fecal collectors and experimental system as well as developing a procedure that readily works with the materials at hand. The same systems with modest modifications

should be applicable to use with the other species (hybrid striped bass, yellow perch, rainbow trout, and possibly largemouth bass). Feeding trials for tilapia have commenced which will be followed by trials for hybrid striped bass, yellow perch, and rainbow trout.

The experimental recirculating system is equipped with a biofilter (1,000-L; 264.2-gal), a sump/particulate filter (110-L; 29.1-gal), and five aquaria (110-L; 29.1-gal) which serve as experimental units. Residence time for the recirculating system is approximately 48 h. Residence time for the individual aquaria is 15 min. Temperature is maintained against the ambient using immersible heaters and a chiller.

The experimental units are in the form of 110-L (29.1-gal) all-glass aquaria equipped with plexiglass fecal collectors. The tilapia must be at least 25 g (0.9 oz), preferably 50 g (1.8 oz), to confine the animals to the area over the fecal collectors without causing damage to fish when escaping around barriers. Stocking density needs to be at least 12 individuals per experimental unit to insure fecal samples of adequate mass/volume (approximately 3.0 g [0.1 oz]/sample [wet weight]) and to control aggression associated with lower stocking densities.

The fish, prior to use, are held in a holding system composed of a separate 1,000-L (264.2-gal) recirculating system and fed for maintenance using the 32% crude protein catfish diet. Environmental conditions are maintained to be the same as those in the experimental system.

The experiments are to be divided into three trials where replication is done through time using a Latin square design. A random sample of 60 fish from approximately 300 is pulled from the holding tank and divided into five groups of 12 each. Each group is placed into an experimental unit and given a 7-day acclimation period. During the acclimation period fish are fed a ration of the conditioner diet to be approximately 2.5% of their body weight divided into three feedings per day at 8:00, 12:00, and 16:00. During the experiment the fish are confined over the collector using a removable barrier except during feeding. Immediately prior to feeding, the fecal reservoirs are removed and cleaned. The barrier is then removed to allow the fish to approach the front of the tank for feeding. Food is applied at the fastest rate the fish will eat without allowing the pellets to hit the bottom. Upon completion of a feeding bout, the fish are gently driven back over the fecal collector, where the barrier is replaced. The tank is then siphoned of any waste feed or extraneous feces and the fecal reservoir is then replaced. At the end of the acclimation period the same protocol is followed with the feeding trial except the five treatments are randomly assigned to the five experimental units. Beginning on the second day of feeding, the feces within the fecal collector reservoir are isolated for further analysis prior to the 12:00 and 16:00 hour feedings.

Fecal sampling entails depositing the contents of the fecal collector onto a 100-μm mesh Nitex screen within a Buchner funnel. The sample is then subjected to aspiration for 30 sec. The sample is then carefully moved to a pre-weighed aluminum pan for weight determination. The sample is then subdivided to yield an approximately 1.5 g (0.05 oz) sub-sample to be placed into a 50 mL (1.7 oz) Erlenmeyer flask with 15 mL (0.5 oz) of experimental system water. The balance of the sample is stored for proximate analysis. Samples are then rated on a scale of 1–5 (5 indicates the fecal sample is nearly intact, 4

indicates approximately 75% intact, 3 indicates approximately 50%, 2 indicates approximately 25% intact, and 1 indicates form is lost). The samples are then exposed to shaking at a rate of 100 rpm with an orbit diameter of approximately 2.5 cm (1.0 in). Samples are rated again after five and 15 minutes. Treatments are compared while taking into account day within a trial (2–7). Rating data is then analyzed as ordinal data using non-parametric statistics. Proximate analysis data is to be analyzed using ANOVA and Duncan's multiple range test where significant differences are found.

OBJECTIVE 1B

UW-Milwaukee

In order to examine the influence of variations in composition of commercial diets, the effect of a "high-energy" versus a "grower" type commercial diet formulation on the physical properties of yellow perch wastes was compared.

Wastes from a 2.44-m (8.0-ft) diameter tank with approximately 3,000 perch that were 227 to 286 days post hatch were collected with the low-head siphon device used for Objective 1a. This group of fish was fed a typical “grower” diet (38.2 % protein and 8.2% lipid) for an approximate two-week period during which the size distribution and physical characteristics of freshly collected “intact” perch feces were evaluated with the same techniques used for Objective 1a. During a subsequent two-week period, a “high-energy” diet (42% protein and 15% lipid) was fed to this same group of fish and the fecal characteristics evaluated.

The size distributions of fecal diameters and fecal lengths, of feces from fish fed either the high-energy diet or the grower diet overlapped considerably. The settling velocities of individual feces produced by fish fed the high-energy diet were slightly but consistently lower than those produced by fish fed the “grower” diet. No difference was demonstrated in the specific gravity of the gathered and centrifuged fecal sludge samples, using the techniques employed in this investigation.

During agitation at 300 rpm feces produced from fish fed the “high-energy” diet generated a higher proportion of total suspended solids than did feces from the “grower” diet. Also, at least during the first 10 min of agitation, the “high-energy diet” feces appeared to remain slightly less intact as a percentage of the settleable material in the Imhoff Cones.

These differences in fecal characteristics suggest that feces produced by fish fed the “high-energy” diet are more likely to be resuspended by turbulence in the fish rearing tanks and break up to small-sized suspended particles slightly more readily than those produced from the “grower” diet.

Engineering strategies aimed at removal or recovery of biosolids from aquaculture rearing facilities must aim to separate solids before they are further broken up. Given the fragility of these particles, it seems that using the fish rearing tank itself as a settling unit is the most rapid means of accomplishing this with minimal mechanical disturbance.

The settling velocities of fecal material are important for rapid collection and removal.

Interestingly, by using highly digestible nutrient dense formulations to reduce waste output by the fish, fecal properties might also be altered in ways that make them more readily broken down and consequently more difficult to settle and remove. Strategies to reduce the output of waste by increasing the digestibility and incorporation of dietary nutrients into fish flesh may have trade-offs

in the characteristics of the fecal material produced. A combined strategy considering feed formulation and resulting fecal properties that influence engineered removal of biosolids, may result in more effective waste removal.

SIUC

The growth trial experiment has been completed with Nile tilapia originating from Purdue University. Five treatment diets were formulated to contain 32% crude protein using menhaden fish meal, gelatin, and casein as protein sources. Four dietary formulations utilized the fiber sources of alpha-cellulose or sugar beet pulp to yield dietary fiber levels of 8 and 18% for each fiber source. A control diet was formulated without the use of added fiber. A conditioner diet was also formulated to contain similar crude protein and energy levels as the treatment diets but contained 10% fiber as alpha cellulose. Carbohydrate and lipid levels were varied in response to fiber levels to conserve gross energy levels relative to dietary dry matter. Each treatment was replicated four times and assigned to experimental units in a randomized block design. The experimental units were five fish per 300-L (79.3-gal) circular tank. Tanks were part of a single recirculating system with temperature maintained between 28 and 30°C (82.4 and 86.0°F) and a photoperiod of 14-h light/10-h dark per 24 h cycle. Water quality in terms of total ammonia and nitrite concentrations was monitored weekly. Fish were acclimated to the experimental system for two weeks using the conditioner diet fed at a rate of 3% body weight/day divided over two feedings (dry feed/live fish weight). A reduced feeding rate was used due to apparently long handling times/low palatability of the diets. At the beginning of the feeding trial, initial weight of juvenile tilapia was 41.0 ± 3.0 g (1.45 ± 0.11 oz). Feeding protocol with treatment diets was the same as used for the acclimation period with corrections made for changes in fish weight every two weeks. Duration of feeding trial was 12 weeks. At the completion of the feeding trial, all fish were sacrificed, dissected for visceral weights and fillets, and are presently being analyzed for proximate composition of the fillets. Growth in terms of percent weight increase was not significantly different between treatments differing in fiber sources or levels.

The tilapia component has been completed. The hybrid striped bass, yellow perch, and rainbow trout components are requiring that the fecal collection system be modified to support higher flow rates and lower temperatures than used with the tilapia. The result is a need for a replacement of the biofilter and particulate units of similar volume to those used previously but more conducive to insulating from ambient air temperatures. This work has been completed and the system is being tested with two chillers to enhance reliability of temperature and water quality maintenance.

OBJECTIVES 1A AND 1C

University of Minnesota (UM)

A break in the production cycle at the UM facility occurred while adult tilapia, which became infected with *Aeromonas hydrophila*, were treated, held for a withdrawal period, and marketed. The stress from this disease limited the ability of the fish to tolerate decreased water quality and thus the degree to which the tanks could be loaded. During this time the fish were fed a maintenance ration. This lessened the nutrient input and resulted in better water quality and a lower suspended solids load than would normally be experienced in a production system. The disease problems necessitated a complete shutdown of the production system tanks. The shutdown

afforded an opportunity to rework system configurations and make repairs and improvements. In preliminary work with the tilapia production systems before the disease outbreak, total suspended solids levels in the three system types ranged from approximately 10–45 ppm. These values correspond well to average values reported in the literature.

OBJECTIVES 2A-C

Using the reference list from the first North Central Regional Aquaculture Center (NCRAC) Wastes/Effluents project as a base, a library search was conducted to identify and gather published literature (to date approximately 400 periodical articles, theses, and reference texts) relevant to the preparation of the report on the beneficial use of aquaculture waste by-products and alternative effluent treatment procedures applicable to the industry in the North Central Region.

In relation to subobjectives 2a and 2b, reference materials concerned with the fertilizing properties of aquaculture solids and sludges and the comparable properties and uses of agricultural manures and municipal sludges and their uses for land application and container media for vegetable and nursery stock have been identified and collected. Composting of these types of wastes to improve their utility has been another focus of the literature search. The coupling of aquaculture with hydroponics to reduce dissolved nutrient discharge and the use of composted products to suppress fungal disease are also a focus of the literature search. Information is also being gathered on the use of constructed wetlands and vegetative buffer strips as a possible means of reducing solids and nutrient discharge in aquaculture effluents. These references also include information on nutrient transport and availability on forested land, agricultural field crops, wild fields, cultivated grass and forage crops, and wetlands in relation to the general utilization of sewage sludge and manure that can provide a contrast and comparison to aquacultural effluents and solids.

WORK PLANNED

OBJECTIVES 1A-C

SIUC and UM

SIUC and UM work group participants have received no-cost extensions to continue their investigations beyond the period covered in this report. Their contributions will be included in a termination report for the project.

OBJECTIVES 2A-C

UW-Milwaukee

Researchers are in the process of completing this report and anticipate it will be finished by the end of October 1998.

IMPACTS

- Provide a broad base of information with regard to alternative species for rearing system design.
- System design of settling basins and clarifiers will be improved through the use of data gathered during this project.
- Literature review will allow knowledge-based decisions to be made regarding best management practices for the removal and treatment of aquaculture effluent.
- Fiber sources of alpha-cellulose and sugar beet pulp, when supplied at levels of 0, 8, or 18 % , do not appear to affect growth of Nile tilapia and may thus enable variations in dietary fiber levels to promote waste management without negatively impacting production.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See the Appendix for a cumulative output for all NCRAC-funded Wastes/Effluents activities.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1996-98	$100,000	$79,968				$79,968	$179,968
TOTAL	$100,000	$79,968				$79,968	$179,968

NATIONAL AQUACULTURE INAD/NADA COORDINATOR[10]

Progress Report for the Period
September 1, 1992 to August 31, 1998

NCRAC FUNDING LEVEL: $45,241 (September 1, 1993 to May 14, 1999)

PARTICIPANTS:

Robert K. Ringer	Michigan State University	Michigan
Rosalie A. Schnick	Michigan State University	Wisconsin

PROJECT OBJECTIVES

(1) Ensure effective communications among groups involved with Investigational New Animal Drug/New Animal Drug Applications (INADs/NADAs), including Canada.

(2) Serve as an information conduit between INAD/NADA applicants and the Food and Drug Administration's Center for Veterinary Medicine (CVM).

(3) Identify and encourage prospective INAD participants to become involved in specific investigational studies and NADA approval-related research.

(4) Seek the support and participation of pharmaceutical sponsors for INAD studies and NADAs and coordinate with INAD/NADA sponsors to achieve CVM approval more quickly.

(5) Guide prospective and current INAD holders on the format for INAD exemption requests and related submissions to CVM.

(6) Identify existing data and remaining data requirements for NADA approvals.

(7) Review, record, and provide information on the status of INADs and NADAs.

(8) Encourage and seek opportunities for consolidating the INAD/NADA applications.

(9) Coordinate educational efforts on aquaculture drugs as appropriate.

(10) Identify potential funding sources for INAD/NADA activities.

ANTICIPATED BENEFITS

Investigation and approval of safe therapeutic and production drugs for use by the aquaculture industry are some of the highest priorities currently facing the industry. At present, only a few approved compounds are available to the industry and further development of the aquaculture industry is severely constrained by a lack of approved drugs essential for treating more than 50 known aquaculture diseases. CVM

[10]Ted R. Batterson serves as the facilitator for this multi-year project interacting with a steering committee in overseeing the Coordinator's activities.

has afforded the aquaculture industry throughout the United States with a "window of opportunity" to seek approval of legal drugs to be used in their production practices. The need for additional drugs is great, but securing data necessary to satisfy the requirements of CVM for drug approval is time consuming, costly, and procedures are rigorous. The INAD/NADA process is the one method that allows the industry to provide CVM with data on efficacy and also aids producers in their production practices.

Coordination and educational efforts directed toward potential INAD/NADA applicants will save time and effort for both the industry and CVM. The National Coordinator for Aquaculture New Animal Drug Applications (National NADA Coordinator) serves as a conduit between an INAD/NADA applicant and CVM. The National NADA Coordinator helps to alleviate time demands on CVM staff, thus allowing more time to process a greater number of applications as well as increasing the breadth of research endeavors within the industry. The grouping of INAD applicants should help to alleviate redundancy, amalgamate efforts, and increase the amount of efficacy data, all of which should result in greater progress toward developing available, approved therapeutic and production drugs.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

In September 1992, Ringer, Professor Emeritus of Michigan State University, was hired on a part-time basis as National Coordinator for Aquaculture INAD Applications. He served in that capacity through August 31, 1994.

As National Coordinator for Aquaculture INADs, Ringer participated with CVM in educational workshops on INAD procedures and requirements. These workshops were conducted throughout the United States This included workshops held in conjunction with the U.S. Trout Farmers Association, Boston Seafood Show, and Aquaculture Expo V in New Orleans. The workshop at the Boston Seafood Show was videotaped and is now available on cassettes from the Northeastern Regional Aquaculture Center. In addition to the workshops, talks were presented on aquaculture drugs at the request of several organizations, including the World Aquaculture Society.

Ringer also helped in the preparation of a letter that CVM used in requesting disclosure information from those holding aquaculture INADs. By law, CVM cannot release any information about an INAD without such permission. A table containing information about these disclosures was made available to the general public. This included the names and addresses of the INAD holders as well as the drug and species of fish intended for use of the drug. It is intended that this table will be periodically updated after additional disclosure permissions have been obtained.

On May 15, 1995, Schnick, recently retired Registration Officer from the National Biological Service's Upper Mississippi Science Center (UMSC), was hired on a three-quarter time basis as National Coordinator for Aquaculture New Animal Drug Applications (National NADA Coordinator). On May 15, 1996, her position was increased to a full-time basis and the position has remained full time in Year 4 (May 15, 1998 to May 14, 1999).

NEW INAD/NADA SPONSORS

Schnick helped gain a new INAD/NADA sponsor for amoxicillin (INAD #9659) and met with Vetrepharm Limited (United Kingdom) in May 1996 in Fordingham,

United Kingdom, to discuss an action plan for the development of the INAD/NADA on their broad spectrum antibacterial product. Schnick also helped obtain and is working with INAD/NADA sponsors for hydrogen peroxide (external microbicide, INAD #9671), luteinizing-hormone releasing hormone (spawning aid, INAD #9709), common carp pituitary (spawning aid, INAD #9728), Aqui-S™ (anesthetic, INAD #9731), GB Research Inc., another sponsor for amoxicillin (oral antibacterial, INAD #9853), EarthTec™ (external microbicide, INAD #9996), copper sulfate (external microbicide, INAD #10-046), Ovaprim™ (spawning aid, INAD #10-040), fumagillin (myxozoan control, INAD #10-106), gonadotropin releasing hormone analog (spawning aid, INAD #10-087), azamethiphos (sea lice control, INAD #10-137), potassium permanganate (external microbicide, INAD #10-223), 17∝methyltestosterone (gender manipulation agent, INAD #10-296), and Pyceze™ (external microbicide, INAD #10-366). Another sponsor for MS-222 gained an approved NADA. Three sponsors renewed their commitment to their INAD/NADA process for formalin, chloramine-T, and oxytetracycline.

A major breakthrough has occurred in developing a new, oral antibacterial for aquaculture--Schering-Plough Animal Health has agreed to allow the development of florfenicol as a broad spectrum antibacterial for public and private aquaculture and as the model oral drug for crop grouping research. The stakeholders in the federal-state aquaculture drug approval partnership program (an International Association of Fish and Wildlife Agencies [IAFWA] project) voted overwhelmingly on a December 31, 1997 ballot to replace sarafloxacin with florfenicol as the oral antibacterial and model drug for the crop grouping research.

PROGRESS ON THERAPEUTIC DRUGS
Schnick and representatives of the Upper Mississippi Science Center (UMSC), La Crosse, Wisconsin held a special session at the Midcontinent Warmwater Fish Culture Workshop in February 1996 to consider label claims and identify potential pivotal study sites for chloramine-T under the federal-state drug approval partnership program. A meeting was held with CVM on October 30, 1996 to gain clarification on the design of the protocols for conducting pivotal efficacy studies on aquaculture drugs (especially chloramine-T) that are used in water treatments. That meeting was followed by a meeting on November 7-8, 1996 with INAD holders of chloramine-T to coordinate efforts on draft label claims, design protocols for pivotal clinical field trials, and identify pivotal study sites for chloramine-T. Several large, active compassionate INADs are held by public aquaculture agencies and organizations. Several of these INAD holders (e.g., U.S. Fish and Wildlife Service [USFWS]) are conducting pivotal efficacy studies for several potential label claims of chloramine-T. Akzo Nobel Chemicals, Inc. (Dobbs Ferry, New York) submitted a letter to their existing INAD (INAD #8086) file on July 21, 1997 committing to the development of a NADA on chloramine-T for aquaculture use. Akzo Nobel Chemicals, Inc. has also indicated that the company will fund genotoxicity studies required by CVM and is currently assessing mammalian and environmental safety requirements delineated by CVM on November 3, 1997.

A regulatory analytical method for para-toluenesulfonamide in fish tissue to support residue depletion studies was submitted on May 15, 1998 to CVM for review. Efficacy and residue chemistry technical sections were

submitted on July 30, 1998 by the IAFWA Project for review by CVM.

Based on residue and environmental data, CVM determined on July 11, 1996 that there are no human food or environmental safety concerns over the use of copper sulfate as a therapeutant, thus making approval relatively easy. Two meetings were held in July and August 1996 with a potential NADA sponsor and CVM to discuss the data requirements for approval and develop an action plan needed to obtain approval of copper sulfate as a therapeutant. Phelps Dodge Refining Corporation (El Paso, Texas) submitted an application for an INAD/NADA (INAD #10-046) on April 3, 1997, a request for two labels on the same package on October 24, 1997, and the product chemistry data on March 12, 1998. The product chemistry technical section was reviewed by CVM on April 15, 1998 and the sponsor responded to the items on July 31, 1998.

Efficacy and target animal safety data and the environmental assessment have been submitted to CVM by the Stuttgart National Aquaculture Research Center. All the data needed for an approval of copper sulfate have been submitted as of early 1998 and all the technical section packages are under review by CVM.

A supplemental NADA for formalin by Western Chemical, Inc. was approved on June 18, 1998 for control of certain fungi on the eggs of all finfish and certain external protozoa and monogenetic trematodes on all finfish. An additional data package was submitted to CVM on April 22, 1998 to expand the NADA for its use to prevent mortalities associated with external fungal infections on all cultured freshwater fish.

The National NADA Coordinator met with the potential sponsor of fumagillin, Sanofi Sante Nutrition Animale (Libourne Cedex, France) on April 19, 1996 in Paris, France to discuss cooperative efforts and the potential for development of a NADA in the United States. Sanofi committed to an INAD/NADA on fumagillin in June 1997. The NADA Coordinator is determining the potential of fumagillin to control or prevent hamburger gill disease in catfish and whirling and proliferative kidney diseases in salmonids. Meetings were held with the sponsor on September 16, 1997 in Edinburgh, Scotland and on February 14, 1998 in Las Vegas, Nevada to discuss the strategy and action plan for the development of a NADA on fumagillin. Meetings were also held with several potential researchers on November 12, 1997 in Stoneville, Mississippi and on December 11, 1997 in Bozeman, Montana to discuss a research action plan for generating efficacy data on fumagillin to control hamburger gill disease and whirling disease, respectively. Efficacy research is ongoing for both diseases.

A meeting was held April 12, 1997 with CVM to discuss the data requirements for hydrogen peroxide as an external microbicide and how to obtain the data so that an approval can be achieved for all the uses for which the drug seems to be efficacious. Efficacy (control or prevention of saprolegniasis on fish eggs) and target animal safety technical sections were submitted to CVM by the IAFWA Project on April 21 and 27, 1998. Data on the efficacy of hydrogen peroxide to control or prevent mortalities associated with saprolegniasis on fish was submitted by the IAFWA Project to CVM on September 18, 1998. A meeting was held with CVM on June 18, 1998 at CVM headquarters to discuss the remaining data requirements for hydrogen peroxide. Discussions centered on the mechanisms for

transferring the Canadian dossier to CVM and for setting up a Public Master File of data from UMSC. Enough data may exist for an early approval in the United States. CVM determined that the human food safety data requirements are met.

A meeting was held on April 11, 1997 with CVM to discuss remaining data requirements to obtain full approval for oxytetracycline. CVM indicated that additional data would be required to expand the NADA for oxytetracycline as an oral antibacterial at temperatures below 9°C (48.2°F) and at doses above the current label and that at least one pivotal efficacy study would be required for these uses to be added to the label.

A call-in of efficacy data was initiated on October 7, 1997 for any and all efficacy data on oxytetracycline that can support the extension and expansion of the NADA. Data were received November 18, 1997, organized in December 1997, evaluated in May 1998, and the summary report is in preparation for submission to CVM.

Abbott Laboratories (North Chicago, Illinois) was in the process of preparing the last portion of a technical section to complete the data requirements for NADA approval of sarafloxacin when concern for development of disease resistant pathogens in humans with the use of fluoroquinolones in animals was raised as an issue by the Centers for Disease Control and Prevention. The Catfish Farmers of America (CFA) sent a letter written by the National NADA Coordinator to the U.S. Food and Drug Administration commenting on the impending ruling regarding the prohibition of extra-label use of fluoroquinolones. The CFA was concerned that this regulation (1) establishes that these drugs, when used in the catfish industry, "are capable of increasing the level of drug resistant zoonotic pathogens (pathogens that are infective to humans) in treated animals at the time of slaughter" and (2) will negatively impact or stop the approval of a fluoroquinolone, sarafloxacin, and other fluoroquinolones for the catfish industry. The catfish industry and researchers have agreed to consider developing a risk assessment on the use of sarafloxacin in catfish to control enteric septicemia to alleviate concerns of disease resistant pathogens developing in humans from the use of this fluoroquinolone. It is doubtful that a new NADA will be allowed for aquaculture uses by CVM. Sarafloxacin was replaced by florfenicol in January 1998 by an unanimous vote of the IAFWA Project stakeholders.

The National NADA Coordinator met with the potential sponsor of Pyceze™, Grampian Pharmaceutical Ltd., in Edinburgh, Scotland on September 16, 1997 to discuss its plans for development of Pyceze™ in the United States. Meetings were held with Grampian Pharmaceutical Ltd. in Seattle, Washington and Stoneville, Mississippi in mid-April 1998 to determine the potential for use of Pyceze™ in the salmonid and catfish industries in the United States.

A meeting was held with CVM, Grampian Pharmaceutical Ltd., and the National NADA Coordinator on April 24, 1998 at CVM headquarters to discuss the requirements to complete a NADA application for Pyceze™ and the preliminary evaluation of the existing data.

Grampian Pharmaceutical Ltd. sent a letter of intent to commit to development of Pyceze™ as an external microbicide on freshwater fish along with a basic data package for approval as a fungicide on fish eggs to CVM on September 3, 1998.

A meeting was held with CVM and the new sponsor of potassium permanganate, Carus Chemical Corporation, on January 28, 1998 at CVM headquarters to discuss the requirements to complete a NADA application and the progress that has been made on the various technical sections. The residue chemistry technical section was submitted by the IAFWA Project to CVM for review.

The National NADA Coordinator worked with the National Aquaculture Association, private producers, state extension personnel, and Bayer Business Group Crop Protection to develop a plan to gain legal use of trichlorfon in non-food fish culture. Because this Bayer group did not think that it was economically feasible to develop trichlorfon as a drug, the registrant worked with the aquaculture industry on "Special Local Need (SLN)" registrations for use of trichlorfon to control predaceous insects, zooplankton, and adult lernea. Several states have either obtained or submitted requests for SLNs.

PROGRESS ON ANESTHETICS

Two meetings in June and August 1996 were held with representatives of Aqui-S™, an anesthetic approved for use on fish in New Zealand, to discuss the potential for development of Aqui-S™ in the United States. Aqui-S™ is approved in New Zealand with a zero withdrawal time and offers a potential alternative to benzocaine. UMSC decided to evaluate the comparative efficacy and regulatory requirements needed for approval on both benzocaine and Aqui-S™.

UMSC completed an efficacy and safety evaluation of Aqui-S™ in two size ranges of six representative freshwater fish species. The report was sent by the National NADA Coordinator on July 18, 1997 to all IAFWA Project stakeholders/cooperators for their decision on which anesthetic, benzocaine or Aqui-S™, should be the IAFWA Project drug. Twenty-four votes were cast for Aqui-S™ and no votes for benzocaine. Detailed assessments will be made of what data requirements will be addressed by the sponsor and what data requirements will be addressed by the IAFWA Project.

Western Chemical, Inc. obtained an approved NADA for MS-222 or tricaine methanesulfonate (Tricaine-S™) as an anesthetic on November 21, 1997.

PROGRESS ON HORMONES

A meeting was held at CVM headquarters on April 11, 1996 with Stoller, users of common carp pituitary (CCP), and researchers to determine a course of action for gaining approval of CCP. As a follow-up to that meeting, CVM coordinated a conference call on May 15, 1996 that covered: (1) the identification of researchers and the design of target animal safety studies; (2) the writing of the environmental assessment through the National Research Support Program Number 7 (NRSP-7), and (3) potential funding sources of the target animal safety studies. A literature review on efficacy and target animal safety of CCP was completed, presented on August 5, 1998 in Bozeman, Montana, and will be submitted to CVM for review in late 1998.

The National NADA Coordinator contacted all the holders of disclosed INADs on human chorionic gonadotropin (hCG) at the urging of CVM to send all the data to the sponsor, Intervet, Inc., that was incorporated in a February 1996 Intervet submission to CVM. CVM ruled on February 12, 1996 that enrollment in an INAD will not be required to use hCG as a spawning aid. CVM will defer regulatory enforcement if used by or on order of a veterinarian. Any hCG product may be prescribed, but CVM strongly

encourages the use of Intervet's product, Chorulon®. This policy was renewed in 1998 and will continue until hCG gains approval.

The National NADA Coordinator and Dr. David Erdahl (USFWS) met with Syndel International Inc. (Canada) in Seattle, Washington on February 23, 1997 to discuss the development of Ovaprim® in the United States. Another meeting was held with CVM on April 11, 1997 to discuss the strategy for development and the data requirements to gain an approval in food fish. Syndel International Inc. (Canada) recently obtained INADs for its gonadotropin releasing hormone analog product (#10-087) and Ovaprim® (#10-040). USFWS and other INAD holders are working with Syndel to develop the technical sections of the NADA package.

Schnick worked with CVM, Auburn University, Rangen, Inc. and tilapia producers to develop INAD #9647 on 17α-methyltestosterone (MT) for tilapia (obtained January 25, 1996) and then worked to obtain authorization from CVM and permission from Auburn University to allow the use of MT on percids under Auburn's INAD (obtained February 22, 1996). The North Central Regional Aquaculture Center (NCRAC) provided $27,000 to Southern Illinois University-Carbondale and the University of Wisconsin-Madison to conduct a target animal safety study on MT with walleye and provided $5,000 for Auburn University to conduct a literature review of the environmental data on MT and submitted an environmental assessment (EA) to CVM on November 7 and 26, 1998. CVM responded to the EA on June 9, 1998 and Auburn is preparing a response. The human food safety portion of the NADA submission on MT was recently submitted by Auburn University to CVM for review, and CVM has accepted MT as safe.

The Drug Enforcement Administration (DEA) has removed certain regulatory controls from the use of 17α-methyltestosterone feed for gender manipulation because the DEA perceives that there is no significant potential for abuse.

A meeting was held with CVM, Rangen, Inc., and the National NADA Coordinator on January 29, 1998 at CVM headquarters to discuss the requirements to complete a NADA application for 17α-methyltestosterone and the progress that has been made on the various technical sections. Rangen, Inc. submitted a letter of intent to CVM on February 27, 1998 to pursue the approval of 17α-methyltestosterone feed for gender manipulation under an INAD (#10-296).

PROGRESS ON THE IAFWA PROJECT

Several meetings were held at UMSC in May and June 1996 to review the whole IAFWA Project related to the following topics on each of the 10 study plans: (1) remaining data requirements; (2) tasks and jobs; (3) assignments for each job; (4) a time table for completing each assigned task; (5) budget projections by study plan and year; (6) budget shortfalls for the original IAFWA Project; and (7) assessment of the potential products at the end of the IAFWA Project. UMSC has reprogrammed its effort and direction under the IAFWA Project due to changes in requirements and circumstances for benzocaine, chloramine-T, hydrogen peroxide, oxytetracycline, and sarafloxacin. Efforts were made to save the entire IAFWA Project during government downsizing and budget reductions.

A Drug Approval Oversight Subcommittee was formed to aid the IAFWA Project to achieve its goal of obtaining drug approvals for United States public aquaculture. The first meeting was held May 5, 1997 in Hot Springs, Arkansas.

A meeting was held with representatives of the USFWS, Biological Resources Division (BRD) of the U.S. Geological Survey, CVM, American Fisheries Society, IAFWA, and the National NADA Coordinator on September 30, 1997 in Arlington, Virginia. The specific objectives of this meeting were to review and discuss: (1) the current status of public sector aquaculture drug and chemical approval activities; (2) the need for collecting pivotal field efficacy data at USFWS facilities; (3) the possibility of including non-USFWS entities on its INADs; (4) the steps needed to ensure continued support for public sector drug approval activities beyond June 1999; and (5) other areas of interest that will foster continued and future support for approval of aquaculture drugs and chemicals. A major topic of discussion centered on the lack of federal funding from BRD for the IAFWA Project after September 30, 1998. All groups agreed to search for mechanisms and sources of funds to continue the IAFWA Project for a total of at least eight years (to June 30, 2002). Subsequent meetings were held on March 20-22, 1998 and April 17-19, 1998 that produced the support needed for the three-year extension.

MEETINGS AND SPECIAL ACTIVITIES

As National NADA Coordinator, Schnick organized and coordinated a major INAD/NADA workshop in November 1995 under sponsorship of CVM that led to increased communications between INAD coordinators, better coordination of the data generation for each drug, and consolidation of several INADs.

CVM held a Joint Canadian-United States Workshop on Jurisdiction of Sea Lice Treatment and Control in September 1996 that will impact aquaculture drug approvals. One of the action items resulting from the workshop is the strategies and mechanics to institute forums for harmonization activities, i.e., the establishment of a joint Canada and United States Aquaculture Working Group. This means that data could be shared and certain requirements for all drugs could be harmonized so that there could be joint submissions leading to approvals being granted simultaneously in both countries.

The National NADA Coordinator met on October 30, 1996 in Rockville, Maryland with Dr. Meg Oeller, CVM Liaison to NRSP-7, and Dr. William Gingerich (UMSC) to discuss coordination of the mutual projects that NRSP-7 and the IAFWA Project have in common. Both projects are working on chloramine-T, copper sulfate, hydrogen peroxide oxytetracycline, potassium permanganate, and sarafloxacin. Schnick also discussed coordination of the other NRSP-7 projects on common carp pituitary, erythromycin, and amoxicillin.

CVM held a meeting on February 13, 1997 with several representatives from the aquaculture community to discuss the effects on aquaculture of two recent laws, the Animal Medicinal Drug Use Clarification Act and the Animal Drug Availability Act. CVM also released a document on April 30, 1997 that further summarizes the two laws and the associated regulations.

The National NADA Coordinator chaired a special session on partnerships for aquaculture drug approvals at World Aquaculture ‘97 held in Seattle, Washington on February 22, 1997.

NATIONAL AQUACULTURE INAD/NADA COORDINATOR

The National NADA Coordinator helped to coordinate the International Harmonization Workshop for Aquaculture Drugs/Biologics held in Seattle, Washington on February 24, 1997. The purpose of the workshop was to create an educational forum to exchange information and identify issues between public and private sectors and international organizations with the goal of initiating follow-up strategies to advance harmonization of drug maximum residue levels, aquaculture drug approval standards, and biological licensure. Several committees were set up to advance the harmonization of aquaculture drugs and biologics. The National NADA Coordinator is the chairman of the committee to identify approved drugs worldwide for aquaculture and which drugs are being pursued for approval.

To attract more pharmaceutical companies to aquaculture, the National NADA Coordinator is working on gaining information on the market for aquaculture drugs both in the United States and worldwide. She gave a seminar to the Pfizer Animal Health Group on May 5, 1997 to encourage the company's interest in developing its products for aquaculture.

The National NADA Coordinator presented a seminar on aquaculture and its drug needs to representatives of Schering-Plough Animal Health on August 26, 1997. She encouraged them to consider developing their oral antibacterial, florfenicol, for the United States market.

Starting February 1997, the National NADA Coordinator was elected to a two-year term on the Board of Directors of the U.S. Chapter of the World Aquaculture Society.

The National NADA Coordinator wrote a draft letter on July 30, 1997 about regulatory options that would encourage animal drug approvals for minor species and for minor use. The options included: (1) criteria for the determination of a minor species or a minor use, (2) creating additional statutory authority, (3) administrative and regulatory changes, (4) creating incentives, and (5) extending existing authority. On January 13, 1998, Schnick followed this letter with comments on a discussion draft "Proposals to increase the availability of approved animal drugs for minor species and minor use" (MUMS document).

On August 21, 1998, Schnick prepared a response to FDA objectives on the FDA Modernization Act of 1997, urging FDA to release the MUMS document to Congress as requested.

The National NADA Coordinator wrote a letter on May 29, 1997 in support of the efforts by the Office of New Animal Drug Evaluation (ONADE) to the Director of that office, Dr. Robert Livingston, because the progress that the aquaculture industry is making toward approvals has been helped by the ONADE.

The National NADA Coordinator wrote a letter on June 4, 1997 to Dr. Gary Edwards, Assistant Director-Fisheries, USFWS, in support of having the Bozeman National INAD Office expanded in its scope to include other entities under their INAD exemptions. Currently, USFWS is pursuing the mechanism that would allow other public agencies and private producers to be cooperators under USFWS INADs.

The National NADA Coordinator organized and chaired a follow up workshop and round table to the International Harmonization Workshop for Aquaculture Drugs and Biologics that was held in Edinburgh, Scotland on September 17, 1997 to identify approved drugs worldwide for aquaculture,

identify those drugs that are being pursued for approval, and determine where cooperative efforts can begin.

The Agriculture Research Service held a Program Planning Session at the Stuttgart National Aquaculture Research Center on February 2-3, 1998 to identify the principal target species and research priorities for a comprehensive research program for the next five years.

Schnick organized and chaired a producers session on compassionate INADs and was on the program committee for the International Harmonization Meeting for Aquaculture Drugs and Biologics at Aquaculture '98, Las Vegas, Nevada, February 15-19, 1998. Schnick also gave a presentation at a special session on aquaculture drug approvals at the same conference.

WORK PLANNED

The National NADA Coordinator developed an action plan that centers on coordinating all drugs of high priority for aquaculture toward NADAs through the INAD process. In particular, Schnick planned to: (1) develop a major initiative on amoxicillin to obtain approval for its use as a broad spectrum antibacterial in all fishes; (2) continue to seek the sponsorship of other oral antibacterials; (3) determine the potential of fumagillin to control or prevent whirling disease in salmonids and hamburger gill disease in catfish and pursue an INAD/NADA if feasible; (4) help determine the potential for approval of two anesthetics, benzocaine and Aqui-S™; (5) try to help the industry overcome negative attitudes about fluoroquinolones and in particular, sarafloxacin, so that sarafloxacin can be approved to control enteric septicemia in channel catfish; (6) identify potential funding sources for INAD/NADA activities; and (7) continue to coordinate efforts to obtain approvals for all 19 high priority aquaculture drugs and additional new drugs as they are identified.

Several meetings and workshops are planned that will benefit aquaculture drug approvals.

Schnick will give an overview presentation on international aspects of antibiotic sensitivity determination and the need for harmonization in aquaculture medicines at the workshop on minimum inhibitory concentration methodologies used in aquaculture, November 24-27, 1998, Weymouth, United Kingdom.

Schnick is chairing a producer INAD session at the Aquaculture America '99, January 27-30, 1999 in Tampa, Florida.

Schnick is organizing and chairing a session on worldwide cooperation toward aquaculture drug approvals at the World Aquaculture '99, April 26-May 2, 1999 in Sydney, Australia.

IMPACTS

Establishment of the National NADA Coordinator position in May 1995 has resulted in coordination, consolidation, and increased involvement in the INAD/NADA process on 18 of the 19 high priority aquaculture drugs and activities on 13 new drugs of interest to aquaculture. Twenty established or new INAD/NADA sponsors have initiated new INADs and progress has been made toward unified efforts on existing and new INADs/NADAs or have renewed their commitment to the INAD/NADA process on their drug products.

This enhanced coordination will help gain extensions and expansions of approved NADAs and gain approvals for new NADAs. In fact, a supplemental NADA has been

approved by CVM for formalin and a new NADA has been granted to Western Chemical Inc. for its MS-222 product.

The approval of the candidate drugs will aid the aquaculture industry to reduce mortalities associated with infectious and handling diseases and to increase their efficiency by using spawning aids and gender manipulation aids. The domestic aquaculture industry will be better able to compete with foreign producers because there will be more legal drugs to use.

PUBLICATIONS, MANUSCRIPTS, PAPERS PRESENTED, AND REPORTS

See Appendix.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1992-93				$17,000[a]		$17,000	$17,000
1993-94	$2,000			$12,180[b]	$4,000[c]	$16,180	$18,180
1995-96	$5,000		$22,750[d]	$63,519	$11,000[f]	$97,109	$102,109
1996-97	$10,000		$29,000[g]	$46,920[h]	$26,000[i]	$101,920	$111,920
1997-98	$15,000		$42,000[j]	$54,419[k]	$11,000[l]	$107,419	$122,419
1998-99	$13,241		$37,500[m]	$54,418[n]	$22,000[o]	$113,918	$127,159
TOTAL	$45,241		$131,250	$248,456	$74,000	$453,546	$498,787

[a]USDA funding through a Cooperative Agreement with NCRAC

[b]USDA funding through a Cooperative Agreement with NCRAC ($8,500) and FDA's Office of Seafood Safety ($3,680)

[c]Northeastern Regional Aquaculture Center ($2,000) and Southern Regional Aquaculture Center ($2,000)

[d]American Pet Products Manufacturers Association ($7,500), American Veterinary Medical Association ($10,000), Catfish Farmers of America ($2,000), Florida Tropical Fish Farm Association, Inc. ($500), Natchez Animal Supply ($1,000), National Aquaculture Council ($1,000), Striped Bass and Hybrid Producers Association ($500), and American Tilapia Association ($250)

[e]USDA funding through a Cooperative Agreement with NCRAC ($20,000), CVM ($20,359), and USDI/NBS International Association of Fish and Wildlife Agencies Project ($23,000)

[f] Center for Tropical and Subtropical Regional Aquaculture ($5,000), Fish Health Section of the American Fisheries Society ($1,000), and Northeastern Regional Aquaculture Center ($5,000)

[g]American Pet Products Manufacturers Association ($1,000), American Veterinary Medical Association ($10,000), Catfish Farmers of America ($10,000), Florida Tropical Fish Farms Association, Inc. ($1,500), Striped Bass & Hybrid Producers Association ($1,500), Simaron Fresh Water Fish, Inc. ($2,500), and Abbott Laboratories ($2,500)

[h]CVM ($18,400) and USDI/NBS International Association of Fish and Wildlife Agencies Project ($28,520)

[i]Center for Tropical and Subtropical Aquaculture ($10,000), Fish Health Section of the American Fisheries Society ($1,000), Northeastern Regional Aquaculture Center ($10,000), and Western Regional Aquaculture Center ($5,000)

[j]American Pet Products Manufacturers Association ($1,000), American Veterinary Medical Association ($10,000), AquaCenter, Inc. ($2,500), Aqui-S New Zealand Ltd. ($2,500), Catfish Farmers of America ($10,000), Earth Science Laboratories, Inc. ($2,500), Florida Tropical Fish Farms Association, Inc. ($1,500), Gurvey & Berry, Inc. ($5,000), National Aquaculture Association ($2,000), Simaron Fresh Water Fish, Inc. ($2,500), Striped Bass & Hybrid Producers Association ($1,500), and Western Chemical, Inc. ($1,000)

[k]CVM ($18,519) and USDI/BRD International Association of Fish and Wildlife Agencies Project ($35,900)

[l] Center for Tropical and Subtropical Regional Aquaculture ($10,000) and Fish Health Section of the American Fisheries Society ($1,000)

[m]American Veterinary Medical Association ($10,000), Aqui-S New Zealand Ltd. ($1,500), Carus Chemical Corporation ($1,000), Catfish Farmers of America ($10,000), Kent Seafarms Corporation ($1,000), National Aquaculture Association ($2,000), Phelps Dodge Refining Corporation ($5,000), Sanofi Sante Nutrition Animale ($2,500), Simaron Fresh Water Fish, Inc. ($2,500), and Striped Bass & Hybrid Producers Association ($2,000)

[n]CVM ($18,519) and USDI/BRD International Association of Fish and Wildlife Agencies Project ($35,899)

[o] Center for Tropical and Subtropical Regional Aquaculture ($10,000), Fish Health Section of the American Fisheries Society ($1,000), Fish Culture Section of the American Fisheries Society ($1,000), and Western Regional Aquaculture Center ($10,000)

TILAPIA[11]

Progress Report for the Period
September 1, 1996 to August 31, 1998

NCRAC FUNDING LEVEL: $120,000 (September 1, 1996 to August 31, 1998)

PARTICIPANTS:

Paul B. Brown	Purdue University	Indiana
Konrad Dabrowski	Ohio State University	Ohio
Paul A. Fuerst	Ohio State University	Ohio
Donald L. Garling	Michigan State University	Michigan
Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
Kerry W. Tudor	Illinois State University	Illinois
Industry Advisory Council Liaison:		
Curt Stutzman		Iowa
Extension Liaison:		
Donald L. Garling	Michigan State University	Michigan
Non-Funded Collaborator:		
Dr. Victor Wu	National Center for Agricultural Utilization, ARS, USDA, Peoria	Illinois

PROJECT OBJECTIVES

(1a) Develop and/or identify cost-effective feeds for tilapia culture in recirculating systems that minimize waste generation.

(1b) Compare and evaluate economically important traits of current commercial tilapia strains in the North Central Region with other strains cultured in recirculating systems.

ANTICIPATED BENEFITS

Producers throughout the North Central Region (NCR) are raising tilapia. However, the combination of species and culture system are not operating at peak efficiency. Diets fed to tilapia are most often modified catfish diets. Those same diets are thought to cause increased muscle lipid concentrations in catfish. If the same problem exists in tilapia during the grow-out phase of production, then the same problems will occur as in catfish. Fish containing relatively high concentrations of lipid in the muscle are subject to more rapid uptake of off-flavor compounds from the water. Further, shelf life of the product can be impaired because of the higher degree of lipid oxidation that can occur. Higher lipid concentrations in fillets is often the result of imbalanced energy to protein ratio (E:P). Thus, the benefits of this line of research are continued improvement of diets fed to tilapia in recirculating systems, continued development of all-plant diets, enzymatic feedstuff enhancement, and use of animal

[11]NCRAC has funded only one Tilapia project to date, which is chaired by Donald L. Garling. This was originally a 2-year study that began September 1, 1996.

agriculture co-products that can be easily manufactured in this region, and continued improvement in product quality for the consumer.

The ability to evaluate genetic differences within and between strains, and to determine the degree of hybrid mixture within some strains will assist the design of future work to select strains which are better adapted to culture conditions which will be utilized in the northern United States, and to assist in the evaluation of genetic schemes such as the production of YY male lines, which can be used to improve aquacultural production. Gene markers for hypervariable neutral polymorphisms have been shown to be able to discriminate among populations and species with better resolution than morphometric traits. These gene markers also have the potential for application in aquaculture, including identification of individuals, families and species, and labeling of brood stocks. They can also be of importance in the identification of hybridization between stocks and species and in the monitoring of inbreeding rates in managed stocks for proper fisheries management.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Nile tilapia (*Oreochromis niloticus*) from the same genetic stock have been used for all nutritional experiments conducted at sites participating in research on Objective 1a. This stock has also been included in genetic research on Objective 1b.

This project has provided seed monies that have been supplemented by industry and the institutions involved. The ratio of other support to North Central Regional Aquaculture Center (NCRAC) funds was 2.2:1.

OBJECTIVE 1A

Research at Purdue University (Purdue) was designed to provide formulation guidelines for practical grow-out diets that are free of fish meal. In a previous phase of this project, 28% crude protein (CP) was identified as the minimum amount that resulted in maximum weight gain. In the second phase of the project, the optimum E:P was explored using the minimum CP concentration. The optimum energy and lipid concentrations of grow-out tilapia were similar to values developed for smaller fish using purified diets (3,000–3,200 kcal/kg [1,361–1,452 kcal/lb], or 4–6% dietary lipid). Dress out percentages, and nutritional composition were not significantly impacted at dietary lipid levels of 8% and lower.

Research at Illinois State University (ISU) was designed to compare 28% and 34% CP diets which were free of fish meal with a 36% CP commercial diet (Purina 5144) and a 36% CP experimental diet which contained 6% fish meal by weight under grow-out conditions. Grow-out conditions included separating small fish from large fish and removing small fish from tanks at four and eight weeks. Due to the fact that an extruder was not available at ISU at the time of diet preparation, diets were prepared by Triple F Products, Congerville, Illinois. Equipment available at Triple F required slight modifications to ingredients. In addition, dietary lipids used at Purdue were not available. Therefore, ISU all-plant diets cannot be compared directly to Purdue all-plant diets. Between weeks four and eight, the commercial diet generated greater rates of growth than the three experimental diets, while growth rates did not differ among the three experimental diets. Due to variability of growth conditions within commercial grow-out systems, additional studies of all-plant diets should be conducted.

In year one of the project, researchers at Ohio State University (OSU) replaced dietary fish meal protein with an animal by-product mixture. There were no significant differences in growth among fish fed on fish meal-free or animal by-product based diets. In year two, OSU researchers have completed a feeding experiment with five feed formulations where fish meal protein was gradually replaced (25, 50, 75 and 100%) with cottonseed meal protein, so that diet 5 was all-plant protein. Essential amino acids, lysine, and methionine were supplemented to diets 2 through 5 to account for an indispensable amino acid requirement. The feeding study was conducted during 16 weeks (May 21September 11, 1988) with fish of an initial weight of 10.2–13.4 g (0.36–0.47 oz). Anticipated effects of diet formulations with cottonseed meal include gossypol toxicity, phytoestrogen effects, and decreased availability of essential amino acids. There were significant differences in growth among fish fed diets with 75 and100% cottonseed meal and the rest of the dietary treatments. Growth depression amounted to 33.3 and 54.1%, respectively. Tilapia fed diets containing 75% or higher of cottonseed meal had significantly depressed hemoglobin and hematocrit values in comparison to fish fed a diet based on fish meal. For example, hematocrit values were depressed to 7–9% in comparison to 31–35% in fish fed diets with no or 25% cottonseed meal protein. This is the first observation of pathologies related to cottonseed meal inclusion in diets of Nile tilapia, which was considered to be one of the most resistant species to gossypol toxicity.

Researchers at Michigan State University (MSU) are evaluating the effect of phytic acid, contained in many oil seed meals, on protein digestion and availability and the use of the enzyme phytase to ameliorate these effects. In year one, they completed preliminary experiments that indicated feeding Nile tilapia to satiation three times per day improved growth and feed utilization compared to fish feed one, two, or five times per day. Two studies to determine the rate of feed and fecal movement through the intestine to determine an appropriate procedure for digestibility trials and phytate binding studies were also completed. In year two, MSU researchers fed juvenile tilapia a herring meal control diet or experimental diets which incorporated either untreated or phytase- treated soybean meal (SBM) substituted at 0, 25, 50, 75, or 100% of the total protein. The dry, untreated SBM diets contained 0, 0.20, 0.39, 0.58, and 0.77 % phytic acid, respectively. Phytic acid was below detectable limits in all the phytase-treated diets. During an eight week growth trial, fish were evaluated for weight gain, whole body CP, feed conversion ratio (FCR), protein efficiency ratio (PER), and apparent net protein utilization (ANPU). An inverse relationship was observed between percent substitution of SBM and growth, PER, and ANPU. Differences were not significant from the control for either treatment until SBM comprised 100% of the dietary protein. Significant differences detected between the treated and untreated groups were at 50% replacement for growth and FCR; 50% and 100% replacement for PER; and 50%, 75%, and 100% replacement for ANPU ($P < 0.05$). In all instances the fish fed the untreated diets performed better than the fish fed the phytase-treated diets.

Researchers at Southern Illinois University-Carbondale (SIUC) have been evaluating *Yucca shidigera* extract (Micro-Aide, Distributors Processing Inc., Porterville, California) as a feed additive to reduce fecal ammonia. They fed juvenile tilapia (22.8 ± 1.8 g; 0.80 ± 0.06 oz) diets containing the extract to determine its effects on growth.

The extract was added to a commercially available feed (Rangen Production™ 32% CP, floating 3/16" pellet) to yield treatments of 0, 0.5, 1.0, 1.5, and 2.0 g of extract per kilogram (0.008, 0.016, 0.024, and 0.032 oz per lb) of diet. Four replicates of each treatment were randomly assigned to separate circular tanks (300 L, 79.3 gal) stocked with five fish each. Culture conditions were maintained with a single recirculating system with water temperatures between 28–30°C (82.4–86.0°F) and a photoperiod of 14-h light/10-h dark cycle. Total ammonia and nitrite concentrations was monitored weekly. All fish were fed at a rate of 4% body weight divided over two feedings (dry feed per wet body weight) corrected for changes in fish weight every two weeks. After a two week acclimation period, experimental diets were fed for 12 weeks. At the completion of the feeding trial, all fish were sacrificed and are presently being analyzed for proximate composition of the whole body. Growth in terms of percent weight increase was significantly different only between treatments containing 0.0 and 1.5 g extract per kilogram of diet, with values of 361% and 258%, respectively. All other extract levels produced growth responses that were intermediate between those extremes and were not statistically different.

OBJECTIVE 1B

Previously, OSU researchers developed a series of short tandem repeat (STR; microsatellite) loci, which were isolated from the haplochromine cichlid species *Astatoreochromis alluaudi*. From these a subset of eight microsatellite markers was identified which have been used to characterize strains of tilapia and which are able to amplify genetic material from a series of seven tilapia species (*Oreochromis niloticus, O. variabilis, O. esculentus* and *O. leucostictus,* as well as hybrid strains of *Oreochromis,* and *Tilapia rendelli* and *T. zilli*, and *Seratherodon galileus*) to verify their utility and genetic variability. In addition to the microsatellite markers, OSU researchers have used a set of markers isolated by Thomas Kocher of the University of New Hampshire to determine genetic variation in populations of tilapia, especially *Oreochromis niloticus*. The populations being examined include an aquacultural stock being maintained at OSU's Piketon Aquaculture facilities to be used for growth and sex reversal studies, a stock of *O. niloticus* recently isolated from the wild, and a set of natural populations of *O. niloticus* from East Africa in the Lake Victoria basin and other lakes of Uganda. A set of studies have been completed on the use of randomly amplified polymorphic DNA (RAPD) applied to *O. niloticus* populations. These results are still being compared to the aquaculture strains. They show that the RAPD technology is a quick, relatively inexpensive approach to assessing genetic variation and interstrain divergence. OSU researchers have also become involved in a review of worldwide genetic resources of tilapia which will be published in an important compilation of information on the biology of tilapia as used in aquaculture. Finally, they have used their microsatellite markers to experimentally assess the impact of breeding structure on loss of genetic variability of small populations. Such populations are equivalent to many which would be found in aquacultural situations. Their results show that genetic variation is being lost at a much faster rate than usually assumed. This is likely to have important implications on efforts to maintain genetic variation in stock strains of tilapia. The comparison of genetic variation in natural and aquaculture strains will allow a better assessment of the variability between strains which OSU is studying in ongoing growth and sex-reversal

experiments being carried on in Dabrowski's laboratory.

Tilapia strain comparisons are continuing at SIUC. The Happy Knight™ red tilapia strain was replaced due to disease problems with the Arizona Red™ strain provided by International Strategies, Inc. of Arizona. Twenty-one fish from each strain were divided equally into three 400-L (106-gal) round black tanks in a semi-open recirculating system. All fish were acclimated to the system for two weeks and each tank had a mean fish weight of 50 ± 10 g (1.8 ± 0.4 oz). Fish are being fed Purina Aquamax 2000™ three times daily with a system water temperature of 30°C (86.0°F). This trial will run until the mean individual weight in each tank has reached 454 g (1 lb), growth of all the strains plateau, or 24 weeks has elapsed.

WORK PLANNED

The data generated by Purdue researchers will be provided to producers in the NCR and several new formulations will be evaluated on production scale in a new NCRAC-funded tilapia project that will begin September 1, 1998.

OSU researchers will complete their evaluation of cottonseed meal protein as a dietary ingredient. Samples of fish body and fecal material will be analyzed for nitrogen, chromium, lipids and ash, to complete nutritional evaluation of diets on fish performance.

MSU researchers have begun a growth trial incorporating graded levels of phytic acid into a herring meal based diet. The graded levels correspond to phytic acid levels found in diets incorporating 0, 25, 50, 75, 100, and 200% of the protein, on a dry matter basis, into a 32% CP diet. The diets will be evaluated as above. A study has been initiated to evaluate phytase treatment of SBM on CP, and individual amino acid digestibilities, on the above solvent extracted SBM diets. Samples are awaiting analytical and statistical analysis. A similar trial to evaluate CP and individual amino acids on the herring meal with graded phytic acid concentrations is in progress. Results from these trials will be completed by January 1999.

SIUC will complete a fecal collection experiment to determine the effect of feeding diets containing an extract of *Yucca shidigera* to Nile tilapia on fecal nitrogen content and leaching. Juveniles are being reared to a size of 25–50 g (0.9–1.8 oz). A Latin Square design will be used with three replicates over time per treatment. Treatment diets and feeding rates will be as described for the growth trial. Sixty fish will be randomly selected from a population of 300 and will be stocked at a rate of 12 fish per fecal collection tank.

OBJECTIVE 1B

The OSU lab will continue to participate with the tilapia genome effort coordinated by Thomas Kocher which has begun mapping the microsatellite markers which were identified by OSU over the last three years.

Researchers at SIUC will complete strain evaluations within the next nine months.

IMPACTS

Gross formulation guidelines for grow-out tilapia diets that are free of fish meal have been developed by Purdue and field tested a ISU. The basic formulation will be expanded to incorporate other ingredients that are readily available in the NCR. These can be taken to local feed mills, which should significantly reduce feed costs.

Research at OSU has provided strong evidence that a diet with 50% fish meal protein replaced with cottonseed meal can be used to produce marketable-size tilapia without compromising growth rate. Taking into account current (August 3, 1998) prices for menhaden fish meal ($0.62/kg; $560/ton) and cottonseed meal ($0.15 kg; $140/ton), this replacement should make considerable difference in feed costs. However, the decreased hematological parameters would most severely affect fish performance in conditions of low oxygen concentrations. Therefore, it is recommended that there should be no more than 50% protein replacement for tilapia production in recirculating systems. Further studies need to be conducted on possible impact of gossypol on marketability of fish fillet.

MSU research has lead to the development of more efficient feeding strategies and improved methods to determine digestibility for tilapia. Their research has indicated that soybean meal could be incorporated into juvenile tilapia diets at levels up to 75% of the protein in a 32% CP diet. Unlike mammals, poultry, and rainbow trout, their preliminary results suggests that treating SBM with phytase does not increase dietary nitrogen retention. This may reflect higher levels of proteolytic activity compared to carnivorous fish and terrestrial vertebrates.

Incorporation of *Yucca shidigera* extract into a practical diet for Nile tilapia does not appear to adversely affect growth response when incorporated at less than 2 g of extract per kilogram (0.032 oz per lb) of diet. Work in progress will determine the efficacy of the extract in enhancing protein utilization and/or manageability of nitrogenous wastes.

The development of genetic markers and assessment of genetic differences between strains will help aquaculturists better evaluate the importance of interstrain differences. In addition, development of new markers will have a significant contribution to the effort to develop a genome map for tilapia which can be used to direct future selective breeding for improved aquacultural production.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See Appendix.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1996-97	$86,500	$104,635	$56,566			$161,201	$247,701
1997-98	$33,500	$51,795	$46,000			$97,795	$131,295
TOTAL	$120,000	$156,430	$102,566			$258,996	$378,996

SAFETY OF 17α-METHYLTESTOSTERONE FOR INDUCTION OF SEX INVERSION IN WALLEYE[12]

Project Termination Report for the Period
July 1, 1996 to August 31, 1998

NCRAC FUNDING LEVEL: $27,000 (July 1, 1996 to December 31, 1997)

PARTICIPANTS:

Christopher C. Kohler	Southern Illinois University-Carbondale	Illinois
Jeffrey A. Malison	University of Wisconsin-Madison	Wisconsin
Industry Advisory Council Liaison:		
Rosalie A. Schnick		Wisconsin
Extension Liaison:		
Joseph E. Morris	Iowa State University	Iowa

REASON FOR TERMINATION

The objectives were completed and funding was terminated.

PROJECT OBJECTIVES

(1) To conduct a Target Animal Safety Study under Good Laboratory Practice (GLP) compliance for walleye fingerlings fed 17α-methyltestosterone for induction of sex inversion to produce genotypic females that produce viable sperm.

(2) To evaluate selected dosages of 17α-methyltestosterone for efficacy at inducing sex inversion in walleye.

PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

The safety of 17α-methyltestosterone (MT) fed to fingerling walleye (Percidae: *Stizostedion vitreum*) was assessed at Southern Illinois University (SIUC) in compliance with GLP regulations (21 CFR Part 58) for non-clinical laboratory studies. The investigation was based on the safety study protocol for Investigational New Animal Drug (INAD) #9647. Only minor deviations from the protocol occurred which were described in a 602-page report that was submitted to the INAD Sponsor. Two studies were conducted in a single room of the Vivarium at SIUC in 208.2-L (55-gal) glass aquaria . For the first study, groups of walleye fingerlings initially 45–55 mm (1.8–2.2 in) total length (TL) were fed diets containing MT at 0 (control), 15, 30, and 75 mg/kg (ppm) of feed whereas in the second study, groups of walleye fingerlings of initially the same size as the first study, were fed diets containing MT at 0 (control), 75, and 150 mg/kg of feed. In both studies, fish were fed their assigned diets for 60 days, followed by a 30-day withdrawal period in which all fish were fed the placebo, or control diet. Fish were observed twice daily for any overt clinical signs, and on day 91 were necropsied by a U.S. Food and Drug Administration's Center for Veterinary Medicine-certified veterinarian. No abnormal organs were observed, and no

[12]NCRAC has funded two Aquaculture Drugs projects. A termination report for the other project is contained in the 1996-1997 Annual Progress Report.

deaths or clinical abnormalities attributable to MT were noted. The results of these studies indicate that MT, used as described in the protocol, is safe for walleye fingerlings.

OBJECTIVE 2

At the University of Wisconsin-Madison, studies on the use of MT to induce sex reversal in female walleye have been completed. Groups of walleye fingerlings initially 35–45 mm (1.4–1.8 in) total length (TL) were fed diets containing MT at 0 (control), 3, and 15 mg/kg (ppm) of feed for 60 consecutive days. The fish were then grown to 200–250 mm (7.9–9.8 in) TL, sacrificed, and examined morphologically and histologically for reproductive status. One hundred percent of the fish fed MT at 3 and 15 mg/kg had identifiable testicular tissue. In the 3 mg/kg treatment group, 92% of the fish examined had apparently normal testes, and 8% had ovotestes. In the 15 mg/kg group, 95% of the fish examined had apparently normal testes, and 5% had ovotestes. Normal female development was observed only in the control group. Forty percent of the control fish were normal females and 60% were normal males. The presence of ovotestes coupled with the absence of normal female development in fish treated with MT indicates that MT at dosages as low as 3 mg/kg induces sex reversal in walleye.

IMPACTS

The successful and safe production of sex-reversed walleye brood stock provides information required for the use of MT in the production of monosex walleye, which may ultimately result in New Animal Drug Approval (NADA) for the use of MT in percids. The propagation of all-female populations of percids will result in improved production efficiency for the commercial aquaculture of these species, and a new management tool for resource enhancement programs.

RECOMMENDED FOLLOW-UP ACTIVITIES

The identification of a method to masculinize female walleye, and use of their gametes to produce broods of all-female offspring, offer great potential benefits for the commercial culture of walleye as a food fish as well as for resource enhancement. Work along several lines is still needed, however, to facilitate the commercial use of these findings. First, NADA approval of the method is needed. Second, a cooperative effort between the North Central Regional Aquaculture Center, commercial producers, and fisheries management agencies is needed to make the technology for monosex culture available to the public and private sectors. Third, research is needed to incorporate this monosex technology into the development of fast-growing strains of hybrid walleye (*Stizostedion vitreum* × *S. canadense*) that will further increase the productivity and profitability of commercial walleye culture.

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED

See the Appendix under Aquaculture Drugs.

SUPPORT

YEARS	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL SUPPORT
		UNIVER-SITY	INDUSTRY	OTHER FEDERAL	OTHER	TOTAL	
1996-97	$27,000	$21,375				$21,375	$48,375
TOTAL	$27,000	$21,375				$21,375	$48,375

APPENDIX

EXTENSION

NCRAC Extension Fact Sheet Series

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Kohler, S.T., and D.A. Selock. 1992. Choosing an organizational structure for your aquaculture business. NCRAC Fact Sheet Series #103, NCRAC Publications Office, Iowa State University, Ames.

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Meronek, T., F. Copes, and D. Coble. 1998. The bait industry in Illinois, Michigan, Minnesota, Ohio, South Dakota, and Wisconsin. NCRAC Technical Bulletin Series #105, NCRAC Publications Office, Iowa State University, Ames.

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Midwest Regional Cage Fish Culture Workshop, Jasper, Indiana, August 24-25, 1990. (LaDon Swann)

Aquaculture Leader Training for Great Lakes Sea Grant Extension Agents, Manitowoc, Wisconsin, October 23, 1990. (David J. Landkamer and LaDon Swann)

Regional Workshop of Commercial Fish Culture Using Water Reuse Systems, Normal, Illinois, November 2-3, 1990. (LaDon Swann)

First North Central Regional Aquaculture Conference, Kalamazoo, Michigan, March 18-21, 1991. (Donald L. Garling, Lead; David J. Landkamer, Joseph E. Morris and Ronald Kinnunen, Steering Committee)

Crayfish Symposium, Carbondale, Illinois, March 23-24, 1991. (Daniel A. Selock and Christopher C. Kohler)

Fish Transportation Workshops, Marion, Illinois, April 6, 1991 and West Lafayette, Indiana, April 20, 1991. (LaDon Swann and Daniel A. Selock)

Regional Workshop on Commercial Fish Culture Using Water Recirculating Systems, Normal, Illinois, November 15-16, 1991. (LaDon Swann)

National Aquaculture Extension Workshop, Ferndale, Arkansas, March 3-7, 1992. (Joseph E. Morris, Steering Committee)

Regional Workshop on Commercial Fish Culture Using Water Recirculating

Systems, Normal, Illinois, November 19-20, 1992. (LaDon Swann)

In-Service Training for CES and Sea Grant Personnel, Gretna, Nebraska, February 9, 1993. (Terrence B. Kayes and Joseph E. Morris)

Aquaculture Leader Training, Alexandria, Minnesota, March 6, 1993. (Jeffrey L. Gunderson and Joseph E. Morris)

Investing in Freshwater Aquaculture, Satellite Videoconference, Purdue University, April 10, 1993. (LaDon Swann)

National Extension Wildlife and Fisheries Workshop, Kansas City, Missouri, April 29-May 2, 1993. (Joseph E. Morris)

Commercial Aquaculture Recirculation Systems, Piketon, Ohio, July 10, 1993. (James E. Ebeling)

Yellow Perch and Hybrid Striped Bass Aquaculture Workshop, Piketon, Ohio, July 9, 1994. (James E. Ebeling and Christopher C. Kohler)

Workshop on Getting Started in Commercial Aquaculture Raising Crayfish and Yellow Perch, Jasper, Indiana, October 14-15, 1994. (LaDon Swann)

Aquaculture in the Age of the Information Highway. Special session, World Aquaculture Society, San Diego, California, February 7, 1995. (LaDon Swann)

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Walleye Culture Workshop, Minneapolis, Minnesota, February 17-18, 1995. (Jeffrey L. Gunderson)

Aquaculture in the Age of the Information Highway. Multimedia session, 18 month meeting of the Sea Grant Great Lakes Network, Niagra Falls, Ontario, May 6, 1995. (LaDon Swann)

AquaNIC. Annual Meeting of the Aquaculture Association of Canada, Nanaimo, British Columbia, June 5, 1995. (LaDon Swann)

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SOME COMMONLY USED ABBREVIATIONS AND ACRONYMS

BOD	Board of Directors
BRD	Biological Resources Division (of the U.S. Geological Survey)
°C	degrees Celsius
CES	Cooperative Extension Service
CG	compensatory growth
cm	centimeter(s)
CVM	Center for Veterinary Medicine
CP	crude protein
E:P	energy to protein ratio
°F	degrees Fahrenheit
FCR	food conversion ratio
FDA	Food and Drug Administration
ft, ft^2, ft^3	foot, square foot, cubic foot
g	gram(s)
gal	gallon(s)
h	hour(s)
ha	hectare(s)
HACCP	Hazard Analysis Critical Control Point
IAC	Industry Advisory Council
IAFWA	International Association of Fish and Wildlife Agencies
in	inch(es)
INAD	Investigational New Animal Drug
ISU	Iowa State University
kg	kilogram(s)
L	liter(s)
lb	pound(s)
m, m^2, m^3	meter(s), square meter(s), cubic meter(s)
mg/L	milligrams per liter = ppm (parts per million)
meq	milliequivalent(s)
mg	milligram(s)
min	minute(s)
mL	milliliter(s)
mm	millimeter(s)

MSU	Michigan State University
N	nitrogen
N	number
NADA	New Animal Drug Application
NCC	National Coordinating Council
NCR	North Central Region
NCRAC	North Central Regional Aquaculture Center
ng	nanogram
OSU	Ohio State University
oz	ounce(s)
P	phosphorus
P	probability
ppm	parts per million
PPM	plant-protein mixture
PSU	Pittsburg State University
Purdue	Purdue University
RAC	Regional Aquaculture Center
sec	second(s)
SDSU	South Dakota State University
SIUC	Southern Illinois University-Carbondale
TC	Technical Committee (TC/E = Technical Committee-Extension; TC/R = Technical Committee-Research)
TL	total length
UM	University of Minnesota University of Missouri
UNL	University of Nebraska-Lincoln
USD	University of South Dakota
USDA	U.S. Department of Agriculture
USFWS	U.S. Fish and Wildlife Service
UW-Madison	University of Wisconsin-Madison
UW-Milwaukee	University of Wisconsin-Milwaukee
YOY	young-of-the-year