Work summarized in this report was supported in part by Grant Nos. 88-38500-4070, 89-38500-4356, 90-38500-5211, 91-38500-5908 from the United States Department of Agriculture sponsored by the Cooperative State Research Service
NRAC
ANNUAL PROGRESS REPORT

For the Period
January 1, 1992 to December 31, 1992

Published May 10, 1993

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I. INTRODUCTION

The Northeastern Regional Aquaculture Center (NRAC) is one of five Regional Aquaculture Centers which were established and authorized by the U.S. Congress under Title XIV of the Agriculture and Food Act of 1980 and the Food Security Act of 1985. In addition to the Northeastern region, aquaculture centers have been established in the North Central, Southern and Western regions of the continental United States and one in Hawaii.

The Regional Aquaculture Centers are administered by the United States Department of Agriculture (USDA) which was designated Federal leadership in aquaculture by the National Aquaculture Act of 1980. USDA leadership is assigned to the Office of Aquaculture which is located in the Cooperative State Research Service of USDA. The Regional Centers are administrative agencies which encourage and fund cooperative and collaborative aquaculture research and extension educational programs that have regional and/or national application. The Centers are organized to take advantage of the best aquaculture science, education skills, facilities and extension services in the United States; all projects are driven by industry needs and are designed to directly impact commercial development of aquaculture in all states and U.S. territories.

II. ORGANIZATION AND ADMINISTRATION

NRAC encompasses 12 states (Connecticut, Delaware, Maine, Maryland, Massachusetts, New Hampshire, New Jersey, New York, Pennsylvania, Rhode Island, Vermont and West Virginia) and the District of Columbia. The Center is headquartered at the University of Massachusetts Dartmouth in North Dartmouth, MA where it is staffed by a full-time Executive Director, Secretary and Accounting Clerk and a part-time clerical assistant. The Center is responsible for coordinating proposals and managing grants, serving as fiscal agent for administering grant monies, arranging for external peer reviews of proposals, maintaining liaison with industry, research and extension participants, coordinating and facilitating interactions among parties in the region, maintaining liaison with other Regional Aquaculture Centers and USDA officials, and for other administrative matters as required in the daily management of the research grants.

A Board of Directors (BOD) representing the region's aquaculture industries, academic institutions, and government agencies establishes policy and provides overall direction for NRAC. The BOD elects from itself a 7-member Executive Committee which oversees matters of operating procedure for NRAC. A Memorandum of Understanding, enacted in 1988, describes and governs the relationships between member institutions in NRAC. NRAC's Manual for Cooperative Regional Aquacultural Research, Extension and Demonstration serves as a guide for the development, approval, conduct and review of regional projects sponsored by NRAC.

Research and extension priorities are established by a 24 member Technical/Industry Advisory Council (TIAC) in consultation with the aquaculture industry. The TIAC has a broad regional membership and represents scientists, extension agents and industry members with varied aquaculture expertise. The TIAC consists of a 12-member Technical Committee (TC) and a 12-member Industry Committee (IC). Nominations for the TIAC are solicited widely throughout the Northeast region.

During 1992 NRAC hired Dr. Victor J. Mancebo as Interim Executive Director to replace Dr. Hank Parker who was selected as the new Aquaculture Program Coordinator for USDA. Mr. David Morehouse served as Chairman of the NRAC Board of Directors; Dr. Reginal Harrell continued as Chairman of the Technical Committee and Mr. Kenneth Bergstrom replaced Mr. Dennis Walsh as Chairman of the Industry Committee.

Administrative highlights for 1992 included an Industry Summit and an internally generated and funded review of the overall functioning of the NRAC program. As a result of the summit and review, a recommendation to assign an Industry Advisor (IA) to each NRAC funded project to complement and work with the already assigned Technical Advisor (TA) was approved by the Executive Committee and Board of Directors of NRAC. The assignment of Industry Advisors helps keep projects oriented to industry needs. This has resulted in a change in the Organizational Structure of NRAC; the new Organizational Structure is depicted in Figure 1.
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D. Future Project Development.

On October 17-18, 1988 NRAC funded a conference with region wide attendance to establish Northeast Research and Extension Priorities. At that time seven broad areas of priority were established (see Table 2). NRAC continuously solicits inputs from industry, extension and research to update its priority areas. The Board of Directors at its meeting in Boston, MA on December 8, 1992 approved funding for Industry Summits to be held every second year to review the situation and outlook for northeastern aquaculture and establish any new research priorities. The next Industry Summit is tentatively scheduled for February 1994.

### TABLE 2

**SUMMARY OF NRAC PRIORITY AREAS**

I. User Conflicts and Social Resistance to Aquaculture.
   A. Develop educational and outreach programs.
   B. Disseminate information using extension techniques.
   C. Develop mechanisms for interstate information exchange.
   D. Establish system for providing more industry input to the research community.

II. Water Quality.
   A. Management of Nonpoint Source Pollution.
      1. Use/development/enforcement of planning and zoning ordinances.
      3. Development of watershed management plan.
      4. Coordination of watershed management activities.
      5. Public education and information program.
      7. Development of mitigating systems or programs (e.g. depuration, relay, microbial disinfection).
      8. Retrofitting of existing technology or developing new systems for stormwater pollution control devices.
   B. Water Quality Conservation, and Reuse.
      1. Design of aeration systems.
      2. Design of recirculation systems.
      3. Inventory regional water resources and conservation needs.
      5. Develop management procedures to prevent off-flavor.
      6. Improve algae and aquatic plant management techniques.

   A. Compilation, impact assessment and documentation of applicable laws and regulations.
   B. Workshops, site visits and seminars.
   C. Production of an "aquaculture regulation handbook".
   D. Develop models for improving legislation.
   E. Organization of a regional industry task force to pursue initiatives of responsible regulation.

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*Northeastern Regional Aquaculture Center*

A. Develop educational material and extension agents training sessions.
B. Conduct market research (including specialty, niche and ethnic markets).
C. Evaluate market structure to investigate cooperative marketing.
D. Analyze the economic risks and uncertainties associated with aquaculture enterprises.
   1. Develop models of aquaculture systems.

V. Health Management of Fish and Shellfish.

A. Develop a database of pertinent regulations and guidelines.
B. Conduct a symposium to facilitate regional cooperation in fish health management.
C. Develop sensitive, rapid, and economical testing procedures for pathogens included in "inspection list".
D. Develop a strategy for establishing a unified code for interstate transport of aquatic animals and products.

VI. Aquaculture Systems Technology.

A. Define "state-of-the-art" for aquaculture systems technology.
B. Develop new information for design of aquaculture systems and sub-systems.
   Topics include:
   1. Waste production of important species.
   2. Growth response as a function of environmental parameters.
   4. Development of sensors for environmental parameters.
   5. Off bottom culture methods.
   6. Closed cycle aquaculture systems.
   7. Improvement of pond, raceway and other systems.
   8. Improvement of harvesting, handling, and processing systems.
   9. Low cost oxygen maintenance.
  10. Product improvement.
  11. Improvement of depuration systems.
  12. Development of mathematical models of aquaculture systems.
C. Testing of designs (prototype or commercial scale).

VII. Increasing Aquaculture Production in the Northeast.

A. Genetics and reproductive physiology.
   1. Domestication of broodstock.
   2. Drain performance evaluation.
   3. Promoting, predicting and inducing ovulation of a species on demand.
   4. Ploidy and sex manipulation; genetic engineering.
B. Nutrition (develop nutritionally complete diets).
   1. Dietary requirements.
   3. Essential fatty requirements and color enhancement.
C. Disease.
   1. FDA approved efficacious drugs.
   2. Diagnostic service availability.
D. Growth.
E. Education (develop an education network).
IV. PROJECT PROGRESS REPORTS

NRAC88-1 "Genetic Improvement of the American Oyster (Crassostrea virginica) for Commercial Culture in the Northeast"

Total Funding $521,050

Period January 1, 1992 - December 31, 1992

Work Group Chair Susan E. Ford, Rutgers University

Participating Investigators/Cooperative Agencies

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<td>Susan Ford</td>
<td>Rutgers University</td>
</tr>
<tr>
<td>Harold Haskin</td>
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<tr>
<td>Robert Hawes</td>
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<td>Herbert Hidu</td>
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<tr>
<td>Dennis Walsh</td>
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<tr>
<td>Carey Mattheissen</td>
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<tr>
<td>Patrick Gaffney</td>
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<td>Karl Rask</td>
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Overview

Since 1984, the protozoan parasite, Haplosporidium nelsoni, the causative agent of the MSX disease of the American oyster, has been discovered in new areas of the northeastern United States. Steady declines in the size of oyster landings have accompanied the spread of this pathogen.

The oyster industry relies heavily upon hatchery-produced seed supplies as the base for replenishing local stocks. Thus, there is a clear need for a seed supply that is reliable both in quality and quantity.

The goal of this project is to produce genetically improved broodstock which meet the criteria of being disease resistant, having good growth rates, and producing oyster meat of high quality. The precise genetic mechanisms involved in resistance to MSX disease are also under investigation.

Objectives

- To develop an information base pertaining to the genetic basis for MSX-disease resistance.
- To disseminate experimental results to the aquaculture community.

Extension Component:

- Assemble and distribute technical and other relevant information on the genetic improvement of Crassostrea virginica, and field performance of oyster stocks to individuals or groups from industry, extension and the research community through extension publications and workshops.

- Organize a workshop session on genetic improvement of Crassostrea virginica at the Milford Shellfish Biology Seminar to present final reports and summarize overall results of the laboratory research and field evaluation carried out during the project’s three year term.

- Prepare and distribute fact sheets summarizing important information on oyster diseases, underlying principles of genetic improvement and disease resistance, and significant project accomplishments; and formulate practical recommendations for the production, deployment, and management of hatchery produced disease resistant stocks in the Northeastern Region.
Progress and Principal Accomplishments

The major objective of this final year of the project was to complete the evaluation of diploid and triploid resistant and susceptible crosses for disease resistance and growth in Delaware Bay. During the first year of the test (1991), the groups were tested for resistance to MSX disease. The general conclusion was that pure resistant stocks were most resistant, followed by the hybrids between resistant and susceptible stocks, and then by susceptible oysters. Within each of these groups, the diploids performed better than the triploids.

During 1992, the surviving oysters were challenged by Dermo disease in addition to MSX. There was relatively little difference in survival among diploid groups, although the homozygous resistant and susceptible oysters had somewhat lower mortality (16% - 18%) than did the hybrids (22% - 24%). On average, mortality of triploid groups was two to three times greater than that of their diploid counterparts.

Growth varied according to genetic make-up. Pure resistant triploid oysters were 21% larger, by weight at the end of the study, than were pure resistant diploids. The diploid hybrids, however, were 7 to 14% larger than the triploid hybrids and 22% larger than homozygous diploids.

A wrap-up session to present the results of the entire study to industry members and other researchers was conducted at the Milford Shellfish Symposium, Milford, CT in February, 1992. Four presentations were made by principal investigators and the session was concluded by a panel discussion, including investigators and industry members, on applicability of the results to oyster culture in the Northeastern and Middle-Atlantic regions of the United States.

Usefulness of Findings

The success of triploid oysters in commercial culture elicited a great deal of interest in their potential for enhancing disease resistance. The results of the triploid study clearly show that creating triploids, which entails expense and risks beyond that incurred in the production of diploid oyster larvae, provides no advantage in defense against either MSX or Dermo diseases. Calculations of total surviving biomass, which takes into account final size of survivors as well as survival rates, will provide more exact information on the relative value of the various genetic combinations.

Work Planned for Next Year

The project was terminated on December 31, 1992; however, final histological work-up and data analysis will be completed in early 1993.

Publications Issued or Manuscripts Approved During the Year

Issued:


Northeastern Regional Aquaculture Center


Approved

NRAC88-2  "Genetic Manipulation and Sex Control in Striped Bass (Morone saxatilis)"

Total Funding  $200,000

Period  January 1, 1992 - December 31, 1992

Work Group Chair  Reginal M. Harrell, University of Maryland

Participating Investigators/Cooperative Agencies

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<th>Investigator</th>
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<tr>
<td>Reginal M. Harrell</td>
<td>University of Maryland</td>
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<td>Howard J. Kerby</td>
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Overview

The striped bass, Morone saxatilis, is an excellent food and sport fish along the Atlantic coast. Declines in natural populations and recent state laws either banning or restricting commercial fishing have maintained high wholesale prices.

Striped bass have been successfully hybridized with both white bass and white perch. A great deal of commercial interest exists in the commercial culture of these hybrids due to their increased vigor over pure striped bass.

Limitations in culture technology are imposed by the limited availability of wild broodstock females. A further concern is that fertile hybrids may backcross and significantly affect the gene pool of the parental species.

This project investigates the role of polyploidy in enhancing marketability through improved growth rates and feed conversion. The desirability of triploids also rests in their functional sterility, which would allay the fears of natural resource managers.

Objectives

- To develop technologies for the commercial scale production of triploid striped bass larvae.
- To study the relative growth and survival of triploid larvae.
- To refine techniques for producing tetraploid larvae.
- To disseminate these technologies throughout the U.S. aquaculture industry.

Progress and Principal Accomplishments

This project was designed to determine if genetic manipulation of striped bass hybrids improved performance expression from an aquaculture perspective. The project had three objectives: 1) develop technology that will enable production of commercial scale numbers of triploid larvae and conduct relative growth and survival studies comparing them to normal diploids; 2) refine techniques for producing sterile striped bass (hybrids); 3) transfer technologies developed in the first two objectives to culturists to stimulate further development of a new major aquaculture industry in the United States.

We were successful in producing triploid hybrid striped bass using pressure induction at levels that would be commercially viable. Although polyploidy was achieved, inconsistencies in success was evident and believed to be due to variations in ambient incubation conditions of the gravid females prior to spawning. We successfully reared triploid progeny at two separate facilities, a freshwater site in West Virginia, and a saltwater site in Maryland. Both diploid and triploid hybrids reached commercial market size (> 500g) in the second year of grow-out. However, triploid performance was less than that of its diploid counterparts through the second year of growth with triploids being significantly smaller than the diploids at both culture sites.

Efforts to induce sex reversal (produce XX males) in pure striped bass through the use of hormones (17 a-methyl-testosterone and estradiol) was unsuccessful. Two years after treatment, gonadal development was minimal with only 7% of the fish showing sexual differentiation and only one of the control group was a female. Treatment group females were from the estradiol tests and the males were from both hormone treatments. The
remaining undifferentiated fish (93%) may have been males, but it was unknown whether these undifferentiated fish were expressing delayed sexual development or were sterilized by the use of the hormones.

Considerable effort was undertaken throughout the course of this project to transfer the technology as it became available. Three annual culture workshops were held in which participants experienced hands-on training in all aspects of striped bass culture, including polyploidy induction. Additionally, three extension videos were produced.

Usefulness of Findings

Although triploid induction methods resulted in fish that grew slower than its diploid counterpart, commercial market size was achieved within two years growth. Likewise, all triploid fish tested were functionally sterile while both male and female diploids were mature during their second year of growth. Comparative performance of triploid was still better than pure striped bass reared under similar conditions, and the fish appear to be sterile. This sterility greatly increases the value of hybrids for use in open water culture situations where culture would otherwise be prohibited due to potential outcrossing. We do not recommend disregarding the potential of triploid induction as a viable alternative based on this study’s growth and performance. The hybrid we used was the sunshine bass hybrid (white bass female x striped bass male) which, in affect, resulted in our working with a fish that had 2/3rds of its genetic make-up coming from white bass while the diploids only had 1/2 of its genome from the white bass. This may account for the differences seen. Future work should examine the palmetto bass (female striped bass).

Work Planned for Next Year

This project is terminated and the final report is in the process of being written.

Publications Issued or Manuscripts Approved During the Year

No publications were approved for this past year. However, two manuscripts are currently in preparation and will be submitted in 1993. Several scientific and extension presentations on results from this project were given during 1992.

Publications or videos associated with this project:

1992-Striped bass hatchery production. Extension video produced through grants obtained from the Northeastern Regional Aquaculture Center, the University of Maryland System Cooperative Extension Service Sea Grant Extension Program, and Maryland Sea Grant College.

1991-Fish farming in open ponds. Extension video produced through grants obtained from the Northeastern Regional Aquaculture Center, the University of Maryland System Cooperative Extension Service Sea Grant Extension Program, and Maryland Sea Grant College.

1990-Cage culture in farm ponds. Extension video produced through grants obtained from the Northeastern Regional Aquaculture Center, the University of Maryland System Cooperative Extension Service Sea Grant Extension Program, and Maryland Sea Grant College.

"Governmental Regulation of Growth and Development: Improving the Legal Framework for Aquaculture in the Northeastern United States"

Total Funding: $93,780

Period: January 1, 1992 - December 31, 1992

Work Group Chair: Alex W. Wypyszinski, Rutgers University

Participating Investigators/Cooperative Agencies:
- Alex W. Wypyszinski
- Norman K. Bender
- Timothy Eichenberg
- James M. Falk
- Bruce E. Lindsay
- Marilyn A. Altobello
- Rutgers University
- University of Connecticut
- University of Maine School of Law
- University of Delaware
- University of New Hampshire
- New Jersey
- Connecticut
- Maine
- Delaware
- New Hampshire
- Connecticut

Overview

Legal and regulatory obstacles are commonly cited as major impediments to aquaculture growth and development in the United States. Although the economic potential for aquaculture is promising, the permitting process is difficult, particularly when operations involve the use of public waters.

A complex legal framework confronts the potential aquaculturist. Policymakers who have the responsibility of striking a balance between protection of environmental and public interests should be made aware of the operational needs of the aquaculture industry.

This project is designed to assess existing regulations, analyze basic legal and policy issues and develop recommendations for changes in the legal framework. The ultimate goal is to improve public decisionmaking within a climate that is supportive of aquaculture ventures.

Objectives

- To develop recommendations for legal changes to enhance decision making and protect aquaculture ventures in the Northeast.

Progress and Principal Accomplishments

Phase 1 of the project continues; most of the information has been placed in computer files. Given the rapid changes taking place in the area of aquaculture law, regulation and policy, this phase should continue until the end of the project. In assembling this material the suggestion has been made that a procedure for periodic updates be designed for the future.

Phase 2 Analysis of basic legal and policy issues and preparation of background papers has been completed by the University of Maine School of Law participants. Among specific questions which were addressed in phase 2 were: the relationship of aquaculture to the public trust doctrine (state ownership of coastal waters and trusteeship of public rights of fishing, commerce and navigation), to riparian rights; to water quality and waste disposal; and to state responsibility for enforcement of wild stock finfish and shellfish regulations. Project participants defined the issues which were addressed in consultation with state officials, industry representatives, and government agency personnel.

Phase 3 Preparation of short case studies to document and assess the costs and impact of government regulation on the aquaculture industry in New Hampshire has been completed by University of New Hampshire participants. Their report has been circulated through the TLAC liaison, and will be included as part of the final report. Case study selection was based on criteria...
including the following: (a) species or type (marine or freshwater); (b) size of operation; (c) production methods employed; and (d) extent of vertical integration. Case studies were prepared through field research and interviews with aquaculturists, agency personnel, and local government officials, and through review of application and decision documents in the administrative record.

**Phase 4** This phase required development of recommendations for changes in the legal framework to improve public decisionmaking and to protect aquaculture ventures. These recommendations have been developed and are contained in the report filed by the University of Maine School of Law participants.

**Phase 5** Development of recommendations for changes in the legal framework to improve public decisionmaking and to protect aquaculture ventures, including model legislation and procedures.

**Usefulness of Findings**

The status of aquaculture legislation/regulations throughout the region has been in a state of flux since the inception of this project.

On June 6, 1992 Alex W. Wypyzinski made a presentation entitled "Permitting for Aquaculture: What You Don't Learn From Textbooks" at the Aquaculture in the Mid-Atlantic Symposium in Annapolis, MD.

The New Jersey Aquaculture Plan recommended as a result of work conducted by project participants will begin following the Governor's signature of an executive order drafted by the Rutgers Fisheries and Aquaculture Technology Center's Aquaculture Plan Subcommittee. Drafting of the Plan should take place in 1993.

**Work Planned for Next Year**

The final year of this three year project will be completed in 1993. **Phase 5** requires publications of a series of guides. Agreement on a final format should be reached in the next month. Outreach efforts will continue beyond project completion.

Issues of aquaculture policy will be discussed at a regional conference planned for 1993 by project participants. This conference will be part of the outreach component of this project.

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**Publications Issued or Manuscripts Approved During the Year**

"Northeast Regional Aquaculture Extension Program for a More Viable, Profitable Industry"

Total Funding $131,780

Period January 1, 1992 - December 31, 1992

Work Group Chair Joseph K. Buttner, State University of New York, at Brockport

Participating Investigators/Cooperative Agencies

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Overview

A primary goal of the Cooperative and Sea Grant Extension Programs is the timely and efficient transfer of information to prospective and practicing aquaculturists. The need for information is particularly acute in the Northeast where, despite an abundance of opportunities, definitive aquacultural data have not yet been standardized and made readily accessible. In addition, aquaculture information generated in different states and regions is often not transferred, resulting in lost opportunities and inefficient allocation of limited resources as states duplicate each others' efforts.

This program will compile existing northeast aquaculture information and develop ten publications which will address the highest priority concerns of the region's aquaculture community. The program also includes an industry and staff training component which will introduce extension personnel to aquaculture through workshop experiences.

Objectives

- To facilitate transfer of information to user groups in the northeast through the development and distribution of aquaculture extension publications.

- To provide hands-on training opportunities to Extension/Sea Grant staff and to industry representatives.

- To establish an aquaculture information network among Cooperative Extension and Sea Grant Marine Advisory personnel.

Progress and Principal Accomplishments

Expressed objectives of this project were to develop fact sheets for novice and established aquaculturists, to set up and hold workshops that address important needs of the aquaculture industry, and to facilitate networking between extension specialists and their clientele throughout the Northeast. The project in 1991/92 achieved all of its stated objectives: ten extension publications have been produced, two conferences that addressed Fish Health Regulations were held, support was provided for two important regional meetings (Milford Finfish and Shellfish Seminar, NRAC Industry Summit), and networking between extension people in different states and their clientele was enhanced (e.g., updated list of aquaculture specialists by state that includes name, telephone number, address, specialty, FAX number, and electronic mail box; list of aquaculture related publications available in each state).

Usefulness of Findings

Information and exchange mechanisms established by this proposal serve as a foundation for closer cooperation and continued growth in the future. Networking within extension and with clientele has been enhanced. A cadre of relatively few extension people has been expanded to include all northeastern states and the District of Columbia. Perhaps more importantly, industry people have become more familiar with the role and capabilities of extension. Increasingly, priorities and methodologies are defined through interactions between industry, research, and extension. The recently published NRAC fact sheets illustrate how the products of this project benefit aquaculture. Extension benefits because county agents can now respond directly to many initial inquiries about aquaculture; they have become part of the NRAC outreach effort. Industry benefits because inquiries by potential and novice culturists are handled by county agents thus affording extension specialists more time to address the needs of established producers.

Work Planned for Next Year

The project has terminated, but momentum generated by the project will continue in at least three major avenues:
1. Under the leadership and coordination of John Ewart (DE) a follow-up extension project, "Development of a Northeastern Regional Aquaculture Extension Network," has been assembled and approved. Many participants in the original extension project are involved in the second project. Significantly, collaboration has grown substantially and the new project includes many new participants.

2. Concerns identified by the Workshop on Fish Health Regulations have been incorporated into the next Extension Project and a follow-up conference is scheduled. Karl Rask (MA) will continue to coordinate extension efforts in the area of fish health regulations.

3. Each NRAC supported research project now includes an extension person to facilitate communication with the Extension Work Group and timely transfer of information to industry. The approach initiated during the concluding Extension Project has become NRAC policy and will continue with future projects.

Publications Issued or Manuscripts Approved During the Year

"Initial Questions the County Agent Can Ask the Prospective Fish Culturist," NRAC Publication No.102, by G. Flimlin

"General Fish Health Management," NRAC Bulletin No.111, by P.R. Bowser and J.K. Buttners

"Aquaculture Systems for the Northeast," NRAC Fact Sheet No.120, by J. Buttners, G. Flimlin, and D. Webster

"Aquaculture Species for the Northeast," NRAC Fact Sheet No.130, by J. Buttners, G. Flimlin, and D. Webster

"Processing and Marketing Aquacultured Fish" NRAC Fact Sheet No.140, by J.M. Regenstein

"Business Planning for Aquaculture-Is It Feasible?" NRAC Fact Sheet 150, by D.B. Strombom and S.M. Tweed

"Growing Microalgae to Feed Bivalve Larvae," NRAC Fact Sheet No.160, by G. Baptist, D. Meritt, and D. Webster


"Predators of Hatchery-Reared Shellfish," NRAC Fact Sheet No.180, by G. Flimlin and B.F. Beal

"The Northern Quahog: The Biology of Mercenaria mercenaria," published cooperatively by the Northeastern Regional Aquaculture Center, Rhode Island Sea Grant, and Rhode Island Cooperative Extension, by M.A. Rice

Miscellaneous Presentations:

"Aquaculture Extension Specialists in the Northeast," September 1992, is a list of extension professionals by state that are actively involved in NRAC activities.

"Aquaculture Extension Publications," June 1991, is a list of state university, federal, state, and industry publications that may be useful to prospective and/or practicing aquaculturists.

"Industry Summit" provided approximately $10,000 support to facilitate participation by commercial producers.

Developed and held a Workshop on Fish Health and Regulations (5 August 1992) that included members of most state aquaculture associations in the Northeast, the National Aquaculture Association, and the United States Trout Farmers Association. A position statement was developed and approved.

Conference (5 July 1992) was held with federal officials and concerns about federal guidelines for fish health were discussed.
NRAC90-3

"Genetic Manipulation of Oysters Through Hybridization and Polyploidy"

Total Funding $134,759

Period January 1, 1992 - December 31, 1992

Work Group Chair Patrick M. Gaffney, University of Delaware

Participating Investigators/Cooperative Agencies

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<td>Patrick M. Gaffney</td>
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<td>Don Merritt</td>
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<td>Donald Webster</td>
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Overview

Hatchery production of bivalve seed stock has become an essential component of commercial aquaculture operations. Controlled hatchery production also offers the potential for the development of genetically superior lines. Shellfish industries in locations rendered unproductive or marginally productive because of disease or environmental degradation can possibly be revitalized by introduction of genetically improved lines.

In addition to developing commercial bivalve strains of demonstrated superiority through conventional selective breeding techniques, interspecific hybridization may be used to produce new, superior genetic types. In this project polyploids will be induced to create further modified oyster strains. Polyploidy is a condition created where individuals carry more than the normal number of chromosomes. Lines of these genetically altered species, tailored to particular environmental conditions, could be of tremendous benefit to the shellfish industry.

Objectives

- To develop methods for the production of interspecific polyploid hybrid oysters.
- To implement experimental methods in small-scale hatchery systems.
- To prepare educational materials and sponsor a workshop on oyster culture.

Progress and Principal Accomplishments

This project contained two components, research and extension. It began in summer 1990 and continued through 1992, with the majority of experimental crosses conducted in summer 1990 and summer 1991.

The research component was aimed at developing two tools, hybridization and polyploidy, for genetically manipulating oysters to produce improved lines for culture.

The extension segment was designed to bring general information about oyster culture to current and prospective cultured by means of an oyster culture workshop and extension fact sheets.

Hybridization:

Broodstock of five Crassostrea species - the Japanese oyster C. gigas, the Suminoe C. rivularis, the native oyster C. virginica, the Caribbean mangrove oyster C. rhizophorae and the Kumamoto oyster C. sikamea -- were maintained and conditioned for spawning in quarantine. We were not able to spawn the Kumamoto oysters successfully, and could only obtain limited numbers of C. rhizophorae, so we concentrated on crosses involving the other three species. We made large numbers of crosses, both as pair matings and mass spawns, to ensure that our results are broadly applicable.

In brief, our findings are straightforward. Most crosses yielded adequate to good fertilization and early larval development, but the larvae were inviable, dying at 10-12 days after fertilization.

C. virginica formed inviable hybrids with C. gigas and C. rivularis. Larvae from the cross with C. virginica and C. rhizophorae survived longer, but failed to settle; however, this work was limited in scope and does not necessarily mean that this cross cannot produce viable hybrids.

The Pacific oyster C. gigas formed inviable hybrids with C. virginica, but crosses with C.
**rivularis** yielded a good set of spat in 1991. However, subsequent analysis (summer 1992) by enzyme electrophoresis showed patterns resembling pure *C. rivularis* in these animals. We hope to use nuclear and mitochondrial DNA markers to determine whether they are simply contaminants (which seems unlikely, since they appear to be *C. rivularis*), or true hybrids with suppressed expression of *C. gigas* enzymes.

Hybridization with Pacific oyster species thus appears to have little potential role for the genetic manipulation of the eastern oyster. *C. rhizophorae* may prove useful in this regard, but further work is required.

Hybridization among the Pacific oyster species appears to be possible, and should be pursued further. If hybrids are sterile (by virtue of their hybrid status, or induced triploidy) as well as resistant to the diseases that plague the eastern oyster, they might eventually find a niche in Atlantic coast aquaculture.

**Triploids:**
In some cases, induction of triploidy (possession of three sets of chromosomes rather than the usual two) enables otherwise inviable hybrids to be "rescued". We crossed *C. gigas* females and *C. virginica* males and induced triploidy in the hybrid offspring. The resulting larvae showed a pattern of early mortality indistinguishable from that of diploid hybrids. However, this experiment should be repeated using various stocks of the two species before we can confidently conclude that triploidy cannot rescue hybrids.

**Dissemination of Results:**
The results of our research have been presented at annual meetings of the National Shellfisheries Association and the World Aquaculture Society. Details of the hybridization experiments, as well as a critical review of attempts at hybridization among species of the genus *Crassostrea*, will soon be published as three articles in the journal *Aquaculture*.

**Extension Component:**
A Fact Sheet on the remote setting of oyster larvae is nearing completion. This will include information on the use of this technique throughout the mid-Atlantic region, as well as results from another NRAC-sponsored study on larval development and mortality. A second fact sheet, on algal production for hatchery operators, is also to be issued soon.

A video on hatchery techniques and remote setting of oysters is also being prepared, with completion in 1994 anticipated.

Two remote setting demonstrations were held in Maryland in 1992. Personnel from this project also participated in education seminars at the Fish Farm Expo held in New Orleans in January 1992.

**Usefulness of Findings**
Although the attempts at hybridization have not proven successful the work undertaken in this project has pointed towards practical difficulties in producing hybrid oysters. Attempts at hybridization should continue with the eventual goal of producing genetically improved oysters which would benefit the Northeastern oyster industry by resulting in increased yearly production.

**Work Planned for Next Year**
Work on this project terminated in mid-1992. A final termination report is being prepared.

**Publications Issued or Manuscripts Approved During the Year**


Allen, S.K., Jr., P.M. Gaffney, J. Scarpa, and D. Bushek. Attempted hybridization of *Crassostrea virginica* (Gmelin) with *C. rivularis* (Gould) and *C. gigas* (Thunberg). Aquaculture (in press).

Allen, S.K., Jr., and P.M. Gaffney. Genetic confirmation of hybridization between *Crassostrea gigas* (Thunberg) and *Crassostrea rivularis* (Gould). Aquaculture (in press).

NRAC90-4, NRAC91-5, NRAC92-8

"Northeastern Regional Aquaculture Center Newsletter"

Total Funding $44,415

Period January 1, 1992 - December 31, 1992

Work Group Chair Joseph K. Buttner, State University of New York, at Brockport

Participating Investigators/Cooperative Agencies

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<td>Gregg Rivara</td>
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Overview

A regional newsletter is being published to address the need for timely and accurate dissemination of information about aquaculture in the Northeast and to describe the activities and projects of NRAC. This quarterly publication deals with recent developments in aquaculture research and commercial operations in the region. It also publicizes the contribution of NRAC to regional aquaculture activities.

Objectives

- To publicize the contributions of NRAC to regional aquaculture development.
- To disseminate results of aquaculture research and extension activities supported by NRAC.
- To provide a forum for the regional and national exchange of aquaculture information.
- To encourage support for aquaculture development.

Progress and Principal Accomplishments

The Newsletter originated in 1989 as a mechanism to inform the aquaculture community and others in the Northeast about NRAC activities. Since 1990, the Newsletter has been edited by SUNY Brockport and Cornell Cooperative Extension. Number of issues printed and mailed has gradually increased from slightly over 1,500 to nearly 3,200. The format and layout has been standardized with two-colors on the front and back. Articles highlight industry activities and NRAC projects.

The Newsletter represents perhaps the most effective, efficient, and economical mechanism to facilitate the timely transfer of information, to ensure that all parties are kept informed of important developments, and to promote the efforts of NRAC. Feedback from the readership has increased dramatically. More articles are being submitted and revision of the mailing list is a dynamic event. Additions, deletions, and changes are common. Between 20-50 new names are added between issues.

Publications Issued or Manuscripts Approved During the Year

Developed, printed and distributed three issues of "Northeastern Aquaculture," newsletter for the Northeastern Regional Aquaculture Center:

"Increasing Aquaculture Production in the Northeast through Nutrition"

Total Funding $293,270

Period January 1, 1992 - December 31, 1992

Work Group Chair Susan H. Goldhor, Center for Applied Regional Studies

Participating Investigators/Cooperative Agencies

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Overview

Meeting the nutritional requirements of fish species raised through aquaculture is a critical element of the industry. Commercial production of salmon, striped bass, and hybrid striped bass is poised for a major expansion, but high feed costs continue to be a significant constraint.

Fish processing plant wastes that are currently being exported or discarded represent a potential underutilized nutritional resource for fish culture. Poultry industry wastes are another possible energy source for aquaculture. Recycling of these wastes into the diet of farm and hatchery reared fish may be ideal methods for the economic utilization of these materials. This project will examine the feasibility of this approach.

This project will also assess the dietary needs of these species, particularly during larval stages. Currently, larval culture of estuarine and marine fish depends on live food. Artificial diets have the advantage of being more efficient in terms of expense, storage, and ease of feeding. Development of these larval diets requires analysis of more traditional dietary components and attention to the physical properties of particle feed which relate to digestibility and nutrient leaching.

Objectives

- To estimate the fatty acid requirements of striped bass.
- To develop nutritionally complete, cost-effective feeds using alternative protein and energy sources.
- To develop commercially feasible processes for producing aquaculture feeds.
- To share technology and management practices with the salmon and striped bass rearing industries.

Progress and Principal Accomplishments

This project was developed to explore promising ways to increase salmonid and striped bass production through the improvement of dietary formulations or lower feed costs. In effect, there are four sub-projects: The improvement of larval feeds for striped bass; the establishment of fatty acid requirements for striped bass and its hybrids; the use of liquid fish concentrates made from processing wastes in salmon feeds, and the use of modern extruder technology to produce slow sinking feeds for salmon.

Work on larval feeds was carried out cooperatively at the University of Rhode Island and at the Monell Chemical Senses Center/U.S. Fish and Wildlife
Service (Pennsylvania and Montana). Among the ways in which larval feeds might be improved are: better (more digestible or more balanced) protein sources, the development of ways to hold the feed together in the water while continuing to attract larvae and maximize digestibility, and the inclusion of minor ingredients to improve the diet's ingestion or digestion by larvae. Work carried out at URI during 1991 had shown that microbound diets performed significantly better than microencapsulated diets, so all of this year's diets at all institutions were microbound. Two hydrolysates were tested as major protein ingredients: dogfish and herring. The latter improved larval survival but not larval growth; the former had no effect. Diets composed of crystalline amino acids simulating the amino acid profiles of the eggs of striped bass or rainbow trout, striped bass larvae, or Artemia were poorly received and resulted in total mortality. A set of four practical diets, containing a variety of high protein, easily digestible ingredients were tested and were also used as the background for a set of varied inclusion rates of a commercial feed stimulant (FinnStim). Of the four practical diets tested, even the best produced survival and growth rates significantly lower than that produced by live Artemia; however, it does provide a reasonable base for future improved formulations. FinnStim offered no benefits at any levels of inclusion. Also tested for potential benefits was a protease. No benefits were seen; however, protease activity was lost so rapidly from the feed, that the data are inconclusive.

The University of Maryland's Horn Point Environmental Laboratory showed that both pure striped bass and palmetto bass (striped bass female x white bass male) benefitted significantly from HUFA-enriched diets. There was no indication that either fish had the ability to elongate shorter chain fatty acid into HUFA.

A collaborative effort by the Center for Applied Regional Studies and Universities of Maine and Massachusetts studied the inclusion of high levels of concentrated liquefied fish (enzyme hydrolysates) in Atlantic salmon diets. 65 day feeding trials tested (1) hydrolysed cod preserved by acidification with phosphoric acid or (2) by lactic acid fermentation using a commercially available, mixed culture with corn syrup as a carbohydrate source, as well as (3) hydrolysed dogfish preserved by acidification with phosphoric acid. In contrast to earlier trials testing hydrolysed cod fermented with molasses, hydrolysed cod fermented with corn syrup as a carbohydrate source proved statistically equivalent to the semi-moist control diet containing raw herring. Hydrolysed cod preserved with phosphoric acid resulted in a statistically significant reduction of final body weights and specific growth rates, while hydrolysed dogfish preserved in the same way proved equivalent to the control diet. At the end of these experiments, the fish were sacrificed and their flavor evaluated by a taste panel. The panel scored salmon fed on both cod-based diets below salmon fed either on the control or the dogfish-based diet. While differences between scores were small, they were statistically significant. Work on the economics of feeding concentrated liquefied fish proceeded along two lines. First, a survey of salmon producers was developed and pre-tested, to provide information on current salmon feed costs, markets and preferences. Second, with engineering input, a commercial scale concentrated liquid fish plant has been designed and costed out, and many of the logistical difficulties associated with the use of processing wastes have undergone preliminary analysis. Finally, a series of small tanks suitable for palatability studies was built and installed at the University of Maine.

Work at Cornell University proceeded in the library, where a literature search of various feed densities and the costs and benefits of achieving those densities was carried out, and in the food processing pilot plant where a new Wenger extruder was fitted for CO2 injection into the barrel.

The technology of liquid CO2 injection via extrusion, developed recently at Cornell, offers a novel way to control the density of feed pellets.

Usefulness of Findings

Larval diets: At least one reasonably successful practical-type diet has been demonstrated with larval striped bass, and should provide a base from which future experimental and commercial diets may be formulated. It is also of interest that herring hydrolysate provided a significant improvement in larval survival. Finally, a number of variables have been eliminated from future striped bass larval feed experiments.

Striped bass fatty acid requirements: The establishment of a clear HUFA requirement for striped bass and palmetto bass will help feed manufacturers to formulate high quality, practical feeds, and aid growers in choosing feeds for optimal growth.

Protein sources for salmonid diets: The findings in this area are unexpected and, although statistically significant, numerical differences in most experiments were small. For these reasons, they should be re-tested. If true, they are both interesting and useful. First, the raw material species utilized appears to affect hydrolysate
palatability for Atlantic salmon. Second, the choice of carbohydrate source in lactic acid fermentation appears to affect hydrolysate palatability for Atlantic salmon. Third, feeding cod hydrolysate to Atlantic salmon prior to slaughter appears to cause a small but statistically significant reduction in the salmon’s palatability to humans. Fourth, dogfish wastes, which are currently a serious disposal problem in the Northeast, appear to be a potentially attractive aquaculture feed ingredient.

Slow sinking feeds: The use of liquid CO2 injection during extrusion as a method for altering feed density is attractive, since it does not add any undesirable ingredients. If it succeeds, it will add a new technology to the armamentarium of the aquaculture feed industry.

Work Planned for Next Year

One larval feed trial will be carried out at URI, in which an attempt will be made to maintain higher levels of protease activity in feed.

The second feeding trial of dogfish hydrolysate, currently underway at the University of Maine, will be completed. Further feeding trials, utilizing condensed dogfish hydrolysate co-dried with soybean meal and/or poultry byproduct meal will be conducted. The new palatability testing facilities will be used to conduct further investigations of the effects of raw material species, methods of digestion, and methods of preservation on the palatability of hydrolysates. Utilizing the knowledge gained by these feeding trials to choose a final hydrolysate formulation, the costs and benefits of manufacturing hydrolysate, incorporating it into feeds, and feeding it to Atlantic salmon will be analyzed.

A slow sinking salmon feed will be developed using the new Cornell technology in which the density of the extruded pellets will be controlled by the injection of liquid CO2 into the extruder barrel. Post-extrusion fat absorption and sinking rate in salt water will be determined.

A series of workshops is planned in which the project’s findings will be extended to aquaculturists, feed manufacturers and, if warranted, to fish processors whose wastes might become a resource to the salmon farming industry.

Publications Issued or Manuscripts Approved During the Year

Publications in print or in press:


Approved manuscripts and theses:


NRAC90-6  "Commercial Field Trials of MSX-Resistant Strains of the American Oyster (Crassostrea virginica)"

Total Funding  $194,941

Period  January 1, 1992 - December 31, 1992

Work Group Chair  Susan E. Ford, Rutgers University

Participating Investigators/Cooperative Agencies

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Harold Haskin  Rutgers University  New Jersey
Gef Flimlin  Rutgers University  New Jersey
George Mathis  Mathis and Mathis  New Jersey
William Mook  Mook SeaFarms, Inc  Maine
Dennis Walsh  Aquacultural Research Corp.  Massachusetts
Karl Rask  University of Massachusetts Amherst  Massachusetts
Dick Nelson  Cotuit Oyster Company  Massachusetts
Richard Drew  Yarmouth Oyster Farms  Massachusetts
Michael Naughton  Stony Island Sea Farms  Massachusetts
Irving Puffer  Wellfleet Oyster and Clam Co.  Massachusetts
Sam Shriver  World's End Aquaculture Corp.  Maryland
Chip Petre  Intertidal Marine Aquaculture  Virginia
Gregg Rivara  Cornell University  New York
David Relyea  F. M. Flower & Sons Oyster Co.  New York
Donald Meritt  University of Maryland, Horn Point  Maryland
Mark Luckenbach  Virginia Inst. of Marine Science  Virginia

Overview

The protozoan parasite, (Haplosporidium nelsoni), which causes MSX-disease of oysters, has been a problem for the oyster industry for more than thirty years. Since 1984, MSX has spread from the mid-Atlantic states into areas of the northeastern United States where it had not previously been reported, and has become epizootic in regions where it had not caused problems before. Only New Hampshire and Maine are still MSX free.

Oyster growers in the region have identified MSX as a major hindrance to oyster culture, and many former growers have left the industry because of MSX. Others are operating with heavy losses. Industry growth is at a standstill because most oyster growers cannot afford to invest in, and then lose, another crop of susceptible stocks. A commercially proven MSX-resistant oyster strain could immediately revitalize the industry in the entire region.

For the past 2-3 decades, Rutgers University has been developing strains of American oysters which demonstrated resistance to MSX disease (see NRAC88-1). For this NRAC project a particularly promising strain (CXF 7th generation Delaware Bay line) will be provided to experienced commercial oyster growers. This will enable evaluation on commercially significant quantities of resistant oysters through the three growing seasons.

Objectives

- To provide MSX-resistant strains to oyster growers to compare their survival, growth, market value and MSX incidence with local controls under commercial conditions.
- To replicate these experiments in different parts of the region and under different growing conditions.
- To disseminate these results throughout the aquaculture industry.

Progress and Principal Accomplishments

Production in 1990 was beset with problems of poor growth and survival, especially of the Rutgers, MSX-resistant strain, due to factors not associated with MSX. In 1991 similar problems occurred in the hatchery and affected both resistant...
and control groups. Because these problems resembled the Juvenile Oyster Mortality syndrome, which has affected oyster growers in the Northeast for the past several years, we received approval to include histological investigations into possible causes for Juvenile Oyster Mortality in the current project (year one).

The work group decided to have two hatcheries produce seed in 1992, thus reducing the possibility of another seed shortage. In addition to the two hatcheries, 8 growers participated in the study during 1992. Sites in Maryland and Virginia were dropped because of excessive mortalities caused by Dermo disease; the New York site was also dropped because of problems with Juvenile Oyster Mortality at the participating company's growout site.

Seed oysters from MSX-resistant broodstock were produced by crossing individuals from a fast-growing Long Island strain and a Delaware Bay strain, both of which had been selectively bred for resistance for several generations. This cross has produced good results in earlier, small-scale experiments on Cape Cod and in Delaware Bay, as well as a more recent test at Rutgers. At both hatcheries the crosses, as well as spawns of control oysters from Wellfleet, MA, were made under the supervision of Rutgers geneticist Dr. Stan Allen.

The strategy of using two hatcheries proved to be wise as one hatchery lost most of its production whereas sufficient seed was produced by the other to supply each grower with 35,000 seed oysters from each of the resistant and the control stocks. To date, there have been no unusual mortalities in either batch at any location, although histological examination of samples collected in October 1992 indicate that MSX disease activity has not been high at any site. Growth performance has varied from site to site, although overall, the control oysters were slightly larger at the end of the growing season than were the resistant oysters.

Survivors of the 1990 spawnings (neither batch performed well) were sampled at one Massachusetts site in October 1992. Thirty-five percent of the control oysters had MSX infections compared to only 5% of the Rutgers strain, indicating that the poor performance of the latter was not related to a lack of resistance.

Usefulness of Findings

Although the seed produced and deployed in 1992 appears to be doing well so far, it is too early to judge the usefulness of the findings.

Work Planned for Next Year

Oysters will be monitored for survival, growth, and disease prevalence at all sites. We have also applied for a revision that would add histological examinations for NRAC-funded Juvenile Oyster Mortality studies as an objective of the project.

Publications Issued or Manuscripts Approved During the Year

None

24 Northeastern Regional Aquaculture Center
"Detection of Fish Pathogens for Fish Health Inspection by Non-Lethal Methods"

Total Funding $310,000

Period January 1, 1992 - December 31, 1992

Work Group Chair Pei W. Chang, University of Rhode Island

Participating Investigators/Cooperative Agencies

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Overview

Current practices associated with fish health inspections require that a sample of fish from a population be sacrificed to allow the collection of appropriate tissues for examination. This practice is costly when the population consists of a limited number of broodfish. Lethal sampling of the appropriate number of individuals can impact the producer both financially and logistically.

Objectives

- Develop a non-lethal tissue sampling method for obtaining kidney or other body tissues; and comparing the recovery of fish pathogens in fish tissues or body fluid collected by lethal versus non-lethal sampling methods.
- Develop and compare the rapidity, sensitivity, specificity and cost of IPNV detection in samples taken by lethal and non-lethal methods.
- Develop and test the efficacy of a time-resolving fluoroimmunoassay for the detection of *Renz bacteria* *salmonealorum* in tissues and body fluids collected by non-lethal sampling methods of fish.
- Develop and test the efficacy of monoclonal antibody-based indirect ELISA for the detection of *Aeromonas salmonicaida*.
- Develop a solid phase ELISA for the detection of IPNV-specific antibodies. Compare the sensitivity and specificity of the ELISA with serum neutralization test; correlate the antibody production and virus production of adult Atlantic salmon to IPNV following infection at 6° and 12°C; survey fish stocks in hatcheries and in the field for their IPNV antibody levels and correlate their immune status with virus isolation and current and past history of IPNV outbreaks among them and their progeny.
- Transfer the technology of non-lethal sampling and improved detection methods to workers doing fish inspections by means of workshops and scientific publications; and inform fish producers on the availability and benefits of fish health inspection procedures that include non-lethal sampling.

Progress and Principal Accomplishments

The work and accomplishments can be described in the following eight (8) categories:

I. Development of surgical procedures for collecting biopsies in fish: Non-lethal surgical procedures for collecting biopsies of liver and kidney tissues for use in fish health inspection have been demonstrated. Fish losses were minimal (4-8%) at 180 days post-surgery. The surgical procedure is recommended for use with valuable broodfish (Bowser).

II. Comparison of lethal and non-lethal sampling methods for the diagnosis of IPNV in trout: Studies were conducted at three trout hatcheries having asymptomatic IPNV carriers. Samples of kidney tissues (collected by lethal sampling methods) proved to be the most sensitive for virus detection; ovarian fluid pellets, body mucus and feces (non-lethal sampling methods) were useful but less sensitive. However, IPN virus was detected in the fish population of all three
hatcheries using non-lethal sampling. To achieve confidence of virus detection in a population of IPNV carriers, a greater number of fish would need to be sampled using the non-lethal sampling methods (McAllister).

III. Detection of *Ruminococcus salmonicida* (RS) from 300 feral and hatchery-raised Pacific salmon by direct fluorescent antibody test (DFAT) and monoclonal antibody-based ELISA was carried out. No RS was detected in the blood of feces of fish and rarely (1-1.6%) in the kidneys or sex products (ovarian fluid or milt). Kidneys (lethal sampling methods) and sex products (non-lethal sampling methods) are the tissues of choice for the detection of RS carriers (Reno).

The sensitivity of the fluorometric immunooassay in detecting RS in fish has been found to be comparable to DFAT. Efforts are being made to enhance its sensitivity (Reno).

IV. Monoclonal antibodies specific to *Aeromonas salmonicida* have been produced. A monoclonal antibody-based ELISA is now being developed (Bowser).

V. Development of a polymerase chain reaction (PCR) assay for IPNV as a non-lethal detection procedure. Primers identifying the polyprotein as well as the presumptive polymerase were developed. A reverse transcriptase PCR assay was developed to evaluate the ability of each primer to amplify strains of IPNV. Two primers were capable of identifying 10 serotypes of IPNV.

The sensitivity of the PCR assay is being determined with virus-infected cell cultures, fish body fluids and feces (Nicholson).

VI. Development of nucleic acid hybridization test for IPNV as a non-lethal detection procedure. A cDNA library having both RNA segments of IPNV (West Buxton) RNA has been constructed and compared to published Jasper and Sp strain sequences. A high degree of homology was shown. Probes were used to detect IPNV RNA in infected cell cultures using the dot blot hybridization assays, which showed that the probes hybridized well with RNA extracted from cells infected with American strains of IPNV, but failed to hybridize with some of the European strains. The IPNV probes did not detect viral RNA tissues of juvenile rainbow trout naturally infected with IPNV (Hetrick).

VII. Effect of body temperature on the immune response of Atlantic salmon. Atlantic salmon were raised at 3 temperatures (6, 10 and 15°C). At 17 months of age, all fish were infected with IPNV (West Buxton strain). Fish were then bled and sacrificed at various times post-infection; the immune status and virus carriage were determined. All fish showed similar immune responses which progressively increased during the 166 days of the study. IPNV was isolated from tissues of infected fish during the course of the study. The presence of humoral antibodies in blood serum can be used as an indicator of IPNV infection (Chang). Virus was detected by ELISA and isolation in tissue culture.

VIII. Extension/Technology Transfer. 1) A poster "Detection of Fish Pathogens" has been assembled that describes the project and its findings. The poster will be displayed at the Milford Aquaculture Seminar, Milford, CT on February 22, 1993. 2) Two technical publications were developed by Dr. Bowser, "A Manual for Non-lethal Surgical Procedures for Obtaining Tissue Samples For Use In Fish Inspections" and "Fish Health Inspections: What Are They?"

Usefulness of Findings

Non-lethal surgical procedures for collecting biopsies of kidney tissues for use in fish health inspection was successful. It is suitable as a non-lethal sampling technique for valuable broodfish (Bowser).

Ovarian fluid and skin mucus collected by non-lethal sampling methods can be used for the detection of IPNV in asymptomatic trout, provided that a large number of fish are sampled (McAllister). Sex products (ovarian fluid and milt) and kidneys are the tissues of choice for the detection of RS carriers (Reno).

The presence of humoral antibodies to IPNV in blood serum are an indication of previous infection in the fish and the possibility of virus carriage (Chang).

Sequencing of the genome of the West Buxton strain of IPNV allows a comparison between it and the Jasper and Sp strains. Further proof that the gene product of RNA segment B is the viral polymerase has been developed (Hetrick).

A non-serotype specific method of detection of any of at least 10 serotypes of IPNV has been developed. Further delineation of difference between North America and European strains by dot-blot assay has been developed (Nicholson).
Work Planned for Next Year

The biopsy procedure for collecting tissue samples will be used in a field trial (Bowser). The development and evaluation of a MCA-based indirect ELISA for the detection of RS will continue (Bowser). The extent to which the PCR based assay is suitable for field assay work will be determined (Nicholson). The sequencing work on the West Buxton strain of IPNV will be completed (Hetrick). Blood from feral and hatchery-raised fish will be collected and antibody titer to IPNV determined (Chang). The sensitivity of time-resolving fluoroimmunoassay for the detection of RS in fish tissue will be enhanced and compared with other immunoassays (ELISA and DFAT) (Reno). As part of the regular inspection programs at the National Fish Health Research Center, samples of fish are being collected to determine the applicability of non-lethal versus lethal sampling (McAllister). Extension and Technology transfer: to complete and display poster; to develop fact sheets to disseminate information (Buttner).

Publications Issued or Manuscripts Approved During the Year


Miscellaneous Presentations:


NRAC91-3  "Water Quality and Waste Management in Aquaculture Production"

Total Funding  $200,800

Period  January 1, 1992 - December 31, 1992

Work Group Chair  Fred Wheaton, University of Maryland, College Park

Participating Investigators/Cooperative Agencies

Kenneth Bergstrom  Red-Wing Meadow Farm  Massachusetts
John Ewart  University of Delaware  Delaware
Conrado Gempesaw  University of Delaware  Delaware
Thomas Handwerker  University of Maryland, Eastern Shore  Maryland
Reginal Harrell  University of Maryland, Horn Point  Maryland
Jerre Mohler  US Fish and Wildlife Service  Pennsylvania
Ernie Tresselt  Hunting Creek Fisheries, Inc., Maryland  Maryland
Fred Wheaton  University of Maryland, College Park  Washington, DC
Harriette Phelps  University of District of Columbia  Maine
Robert Blake  University of Maine  Maryland
Bradley Powers  Maryland Department of Agriculture  Hawaii
Gary Pruder  The Oceanic Institute  Pennsylvania
Steven Van Gorder  Fresh Culture Systems, Inc.

Overview

Maintenance of superior water quality is critical to commercial aquaculture production, and discharged wastes represent economic losses through inefficient use of resources. However, there is little data on discharges and environmental impacts of aquaculture wastes. This has resulted in a restrictive Federal and state regulatory environment related to aquaculture discharges.

Better understanding of the critical water quality aspects and dynamics of aquaculture production systems will aid in the development of rational regulations governing aquaculture wastes. This project will develop a database that describes critical water quality properties and waste discharges representative of several aquaculture production facilities in the Northeastern United States.

Effluent and water quality parameters that will receive particular attention are solids, BOD, phosphorus, and nitrogen. A 15-month program of sampling and analysis for these and other parameters will be carried out in close cooperation with industry partners.

The program proposed here parallels similar programs underway or in development at three other Regional Aquaculture Centers.

Objectives

- Develop a database describing water quality characteristics of representative aquaculture production facilities in the Northeastern U.S.
- Measure critical properties of culture systems dynamics and effluent waters in representative production systems.
- Develop criteria and methodology to test and evaluate production waste and system management technologies with a uniform format.
- Transfer information to industry and other production personnel.

Progress and Principal Accomplishments

The project was designed to sample the influent and effluent of two flow-through trout systems, one a raceway system and the other a tank culture system, and two pond culture systems. The pond culture systems consisted of a hybrid striped bass facility using brackish water and a commercial goldfish/koi farm. When the project was initiated, two of the farms were "out of compliance" for waste water discharge. Currently all of the farms are in compliance relative to waste water discharge. Achieving compliance by these farms was due to efforts of the farms, modifications made on the farms, and the assistance from investigators from this project.
Quarterly and 24 hour intensive sampling of the influent and effluent at the brackish hybrid striped bass facility has been completed on schedule. Due to unexpected delays only preliminary sampling has taken place on the other three farms.

In an attempt to correlate pond water quality with weather, a weather station has been installed at the goldfish/koi farm to monitor solar radiation, wind speed and direction, barometric pressure, relative humidity, water and air temperature, evaporation rate, and rainfall. In addition, sets of sensors were placed in four ponds to monitor PAR (photosynthetic active radiation), temperature, and oxygen concentrations. Four sensor sets were located in two locations (approximately in the center on the smaller pond dimension and 1/4 the distance along the longer dimension from each end of the pond) in each pond. One set of sensors was located about 6 inches above the bottom and another set was located approximately 6 inches below the water surface at each of the two locations. All the sensors were connected to a data acquisition system which collected a reading from each sensor once per hour.

Under the information transfer section of the project two efforts have been ongoing. The first is development of a computer literature search system by personnel at the University of Maryland, Eastern Shore at Princess Anne, Maryland. The system being used is a "micro" bridge and retrieval program that searches Current Contents. The system can store the material located in a "Procite" (Word Perfect compatible) format for transfer to the user. An aquaculture wastewater bibliography is being developed and will be available in the near future.

As a second component of the information effort, four fact sheets are being developed. These four fact sheets will address the following topics: aquaculture production systems in the Northeast region, composition of aquaculture effluent from Northeast region commercial aquaculture facilities, an interim guide to policies and regulations for the disposal of effluent and other wastes from aquaculture facilities in the Northeastern states, and a resource guide to aquaculture management literature, technology, and research programs in the Northeastern region. Final reviewed copy for these fact sheets is due to the extension investigator in March of 1993.

Work Planned for Next Year

Due to changes in personnel a project revision is currently being developed for submittal to the NRAC. Additional monitoring will be done at the four sites selected in the original project for at least another year. The fact sheets and wastewater bibliography will be completed. An attempt will be made to correlate water quality in the goldfish/koi farm ponds with weather data, and the data collected from influent and effluent at the four sampling sites will be analyzed.

Publications Issued or Manuscripts Approved During the Year

None
NRAC92-1 "Alternative Marketing Options to Improve Profitability of the Northeast Aquaculture Industry"

Total Funding $200,000

Period May 1, 1992 - December 31, 1992

Work Group Chair Gregory Hanson, Pennsylvania State University

Participating Investigators/Cooperative Agencies

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Overview

The Northeast aquaculture industry is at a critical juncture in its development. Recent large reductions in the price of salmon, hybrid striped bass, hard clams, mussels, soft-shelled clams and other aquacultural species now threaten the survival and future development of the Northeast aquaculture industry. On the production side, active growers are finding that it is imperative to reduce current losses. Improved marketing practices and marketing management are the keys to restoring profitability at current production levels, or to expanding sales to achieve scale efficiencies.

To help ensure that the Northeast aquaculture industry is not seriously undermined by current price conditions, more resources must be directed to marketing issues. There is increasing recognition that factors such as a shorter growing season, higher labor costs and/or an inability to match foreign supports and subsidies will continue to place severe cost-competitive pressures on Northeast producers. Given the constraints of higher cost production, the region’s industry needs to increase efforts to capitalize on proximity to the nation’s largest population and income base along the Eastern Seaboard. Continued cost-cutting is imperative, but by itself will not ensure the long term growth and viability of the aquaculture industry. Perhaps the most pressing concern for Northeast aquaculture producers is lack of knowledge regarding the marketing basics of price relationships, consumer preferences, market niches and market institutions. Significant advances in knowledge of surrounding markets and market institutions are needed to expand sales and improve infrastructure.

Objectives

- To identify consumer and seafood buyer preferences on product attributes, value added products and seafood safety concerns affecting purchase decisions of Northeastern aquaculture products.
- To reduce institutional marketing constraints and to promote niche and value added marketing opportunities for Northeast aquacultural products.

Progress and Principal Accomplishments

The project was approved by NRAC in May. Much of the initial research effort was spent on preparation of survey documents and pretesting of the surveys. Baseline research was conducted with survey data for the Northeast made available by researchers in a recent marketing study funded by SRAC. Participants met twice, as a group, to coordinate project implementation. The first
meeting was in August in Baltimore, in conjunction with another professional meeting; the second meeting was in Boston in September, a location selected for its accessibility to the industry cooperators from New England.

During the project formulation period, it was pointed out to prospective project participants that the Southern Regional Aquaculture Center had recently completed three major inter-related survey studies on fish and seafood markets in all regions of the U.S. The surveys dealt with consumer usage patterns, food retailer sales and food service sales of fish and seafood. While the studies focused on catfish and crayfish, major parts of their surveys were general in nature, accounting for from one fourth to one half of the questions. Much of the SRAC data for the Northeast had not been analyzed nor published, and no in-depth analyses were done for our region. Thus it was suggested that marketing project participants gain access to the already-collected Northeast data, and where useful, to develop questions such that consumer trends between 1988, when the SRAC data were collected, and 1993, when the NRAC data would be collected, could be determined.

The SRAC data have proven useful to several subgroups of researchers in the marketing project. For example, key demographic factors influencing demand for aquaculture products in the Northeast have been identified. Strategies that target aquaculture products to identifiable subgroups, such as older female professional workers appear promising. The data have provided strong evidence that consumers view preparation time as a critical concern for fish products. The importance of fresh flavor and perceived nutritional value were also demonstrated by the SRAC data.

Following on the SRAC analysis, one subgroup is focusing on consumers' perceptions of product attributes and health safety risks associated with the consumption of northeastern aquaculture products including, mussels, oysters, clams, trout, salmon, hybrid striped bass and tilapia. Two mail surveys, one for finfish and one for shellfish, with questions specifically designed to address perceptions about product attributes and safety were developed last fall. Project members from the universities of Delaware, New Hampshire and Rhode Island met on three occasions to complete development of the above surveys. Pre-testing of the two surveys on individuals was completed in December, and recommendations for improvements to specific questions have been incorporated.

Another subproject focused upon the identification of marketing problems faced by seafood buyers and distributors. The associated team of researchers from the universities of Delaware, Maine and Rutgers met in September and developed a list of target species, a plan of action and a division of research responsibilities. The species identified are mussels, hard clams, salmon, trout, hybrid striped bass and tilapia. Scholarly literature was searched for relevant studies that could contribute to the development of survey instruments. A preliminary survey design was drafted, and revised. Results from the SRAC marketing data were also incorporated into the process of survey design.

In a related effort, a series of interviews with key personnel in the salmon industry, in both the Northeast and adjacent regions in Canada, were commenced by project cooperators from the University of Maine. The research focus is on institutional constraints to marketing, measurement of transactional costs and the development of strategies to overcome existing marketing constraints to facilitate orderly growth of the Northeast aquaculture industry.

One of the marketing subprojects focused on issues of value added fish products. A project participant at Rutgers University has begun efforts to compile ideas for a directory of enhanced or high-value aquaculture products appropriate for Northeast producers. Primary sources for product ideas have been in seafood trade shows. Recent issues of Seafood Business, Seafood Leader and Seafood International have been reviewed. Companies with value-added products derived from the target species which participated in Sea Fare Americas, in Miami, were interviewed in November.

A consumer intercept survey on value-added was developed by project cooperators from the New Jersey Department of Agriculture and New York Sea Grant. They developed both a Retail Survey and a Foodservice Survey to be conducted by consumer-intercept and foodservice operator, personal enumeration interviews. These surveys incorporate much more detailed price information than was available in the SRAC data. Foodservice operators are surveyed on the following product types: microwaveable prepared entrees, ovenable prepared entrees, prebreaded portions, portions in sauce, pasta sauces, shelf table salads, all natural salads, pates, mousses, terrines and shellfish in sauce. The origin of seafood products purchased is also well-developed in the survey document. A minimum of 1000 surveys are to be completed for each user group. This is being accomplished through surveying at retail and foodservice seminars and trade shows including the Boston Seafood Show, the New Jersey Restaurant Show and the Seafood Splash course conducted by the Food
Marketing Institute and the National Marine Fisheries Service.

The niche marketing subproject concentrated efforts on the joint analyses of the consumer, restaurant and grocery store SRAC survey data for the Northeast to determine niche marketing potential. New England, the Middle Atlantic and the North Central regions were selected for analysis. The latter represents an adjacent inland market readily accessible by Northeast producers. Profiles of heavy versus light users of fish and value-added products were prepared, and demographic factors including age, sex, income, occupation type and education were analyzed for their impact on consumption. 'Double-hurdle' analysis that first considers use/nonuse and then examines level of use was employed to identify combinations of consumer characteristics that offer increased market potential. For example, marketing efforts aimed at female/high income consumers appears to offer the prospect of expanded consumption. Research was begun on a logit regression model that will ultimately offer probabilities of consumer acceptance of aquaculture products based on particular combinations of demographic attributes.

Usefulness of Findings

The relatively short time the project has been in existence has not permitted that the analyses undertaken be completed. Virtually all of the personal enumeration and mail surveys will be conducted during 1993. The preliminary analyses of the three SRAC marketing surveys have obtained results for the Northeast that are promising. We anticipate that useful findings will be forthcoming, focused at all levels of the marketing chain, beginning with the consumer, including distributors, and ending with retailers, restaurants, supermarkets and other foodservice institutions.

Work Planned for Next Year

Initial analyses of the three SRAC data sets for the Northeast will be completed by April 30, the end of the first year of the project. The remainder of the first year will be dedicated to the completion of survey preparation and mailing. Analysis of survey results will commence during the summer and preparation of reports will begin during the fall of 1993. The surveys to be completed by topic and lead institution include: niche marketing (Penn State), value-added (two surveys, New Jersey Department of Agriculture), consumer preferences and safety (two surveys, Rhode Island, New Hampshire and Delaware), fish and seafood distributors (Delaware, Maine and Rutgers).
**NRAC92-2**  "Domestication of Striped Bass For Aquaculture"

**Total Funding**  $198,128

**Period**  May 8, 1992 - December 31, 1992

**Work Group Chair**  Bernard Petrosky, Delaware State College

**Participating Investigators/Cooperative Agencies**

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**Overview**

The culture of striped bass (*Morone saxatilis*) and its hybrids for commercial aquaculture,fee-fishing, and restoration operations is one of the more popular forms of aquaculture in the United States today, and it appears the trend will continue for many years to come. Although still in development, striped bass and hybrid striped bass aquaculture production is already expected to be greater than the wild harvest. In fact, production of striped bass hybrids as food fish has doubled in recent years and is expected to continue to nearly double for the next five years.

While the commercial culture of these fish is growing, it is not without certain problems. It is the hybrid crosses that are in demand for commercial aquaculture. Two primary technical problems impede the culture of this species: 1) dependency on wild striped bass brood stock for seed supply, and 2) the inability to spawn sexually mature adult fish on demand to provide fingerlings on a year-round, economically feasible basis.

This project will benefit the industry by aiding in the development of captive striped bass broodstocks and the results will provide information regarding handling, maintaining, and spawning fish in a commercial aquacultural milieu.

**Objectives**

- Develop effective techniques to rear captive striped bass and hybrid striped bass to reproductive maturity.
- Develop reliable techniques for successful year-round induced spawning of captive striped bass and hybrids. Study the effects of photoperiod and temperature on a) gametogenesis and controlling hormones, and b) spawning time and success. c) Identify and implement a potent gonadotropin releasing hormone analog-based (GnRHa) system to induce spawning in phase-shifted captive brood fish.
- Transfer information and technology to the striped bass cultivation industry.

**Progress and Principal Accomplishments**

This project was developed to address the critical problems of access to broodstock and limited spawning season faced by the striped bass/hybrid aquaculture industry.

**Objective 1)**
Timing of the release of funds for this project was such that most investigators in objective 1) missed the 1992 spawning season.
AquaFuture, Inc acquired and held both wild and domestic fish. Handling and transportation were accomplished without difficulty, but problems with disease and (for wild fish) converting to non-living feed resulted in virtually complete mortality.

AquaFarm Products, Inc. installed and tested prototype tank systems in which various fishes including striped bass and hybrids were successfully held.

Delmarva Aquatics (DA) spawned eight female Delaware striped bass. Six hundred thousand fry were sent to various pond rearing locations and tank culture to the fingerling stage was attempted. Pond culture was successful at several locations. Tank culture went well to about 25 days. Early in the season transient low temperatures appeared to permanently disrupt growth. Later fry did well while feeding on brine shrimp, but attempts to convert the fish to artificial feeds resulted in repeated major disease outbreaks and total fish loss.

Delaware State College (DSC) stocked 300,000 fry from DA into small experimental ponds, yielding about 300 phase I/II fish. These fish (F1 ex-wild) are still being held in a pond. About 400 YOY fish produced by DA and trained to artificial feed were brought to DSC and are being held in an outdoor tank.

Objective 2)

a,b) University of Maryland, Center of Marine Biotechnology (UM COMB) has nearly completed construction of an aquaculture facility with state of the art photoperiod, temperature and salinity controls which will be used to provide the temperature and photoperiod regimes to phase shift the experimental fish. That portion of the work will be initiated within the next few months.

a) Radioimmunoassays for reproductive steroids were developed and validated. These assays will allow assessment of the reproductive response of fish to environmental manipulations. They have already been used to gather data on the “normal” cycle and its relation to gonadal development in striped bass exposed to natural environmental conditions.

b) Polymer-based, sustained-release GnRH delivery systems were developed and tested. Selected devices were used to induce spawning in female striped bass. GnRH formulations induced spawning in 90% of the treated fish versus none in the controls. Viable eggs were produced with good larval survival. These systems will be used with phase-shifted fish to induce spawning.

Objective 3)

Much of the activity in Objective 3) was delayed because of the slow start in objectives 1) and 2). University of Maryland, Cooperative Extension Service, Sea Grant Extension (UM CES) has proceeded with planning for the 1993 Striped Bass Short Course. The State University of New York, Brockport (SUNY) is prepared to move ahead with information dissemination as results come in.

Usefulness of Findings

This project is less than a year old and still in an early phase. Activities in objectives 1) have added to the experience of the investigators and resulted in a group of F1 ex-wild fish under cultivation as potential broodstock. In objective 2), results have provided an important tool for future studies (steroid assays) and an effective spawning induction system. In objective 3) the Striped Bass Short Course remains an important educational resource.

Work Planned for Next Year

The work group plans to continue with the project toward the outlined objectives.

Publications Issued or Manuscripts Approved During the Year


"A Comprehensive Investigation of Larval Development and Mortality in the Eastern Oyster, (Crassostrea virginica)"

Total Funding $198,698

Period April 6, 1992 - December 31, 1992

Work Group Chair Carter Newell, Great Eastern Mussel Farms Inc.

Participating Investigators/Cooperative Agencies

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Overview

Given the severe declines in oyster production in the Northeast, every effort should be made to stimulate production through the best methods available. For the Northeast to retain some of its share of the market, techniques such as remote setting should be explored. If these techniques can be adapted successfully to *Crassostrea virginica* in this region, then perhaps much of the once productive oyster grounds can be brought back into production.

Objectives

- To characterize the normal microscopic anatomy of *Crassostrea virginica* from fertilization through metamorphosis, to early juvenile.
- To document normal larval development and feeding activity through metamorphosis with non-invasive, time lapse video technology.
- To characterize physiological rate processes in normal and stressed organisms using both real-time and post-sampling techniques.
- To document changes in major catabolic energy substrates throughout development and under conditions of nutritional stress.

Progress and Principal Accomplishments

This project seeks to improve the technology of the remote setting of American oysters (*C. virginica*) in the northeast region by investigating the causes of mortality of post-set oysters. Comprehensive studies of larval anatomy, physiology and survival during different commercial-scale runs at oyster hatcheries in Maryland and Massachusetts were proposed to compare differences between cohorts with good and poor survival. This project is being carried out cooperatively with 5 sub-projects in Maryland, Massachusetts, Connecticut and New York.

The hatchery phase of the first season’s work on the larval mortality study was completed this summer. Hatcheries at the University of Maryland Horn Point Environmental Lab (HPEL) and at the Aquacultural Research Corporation (ARC) successfully reared and set different crosses involving oysters from Maryland and Massachusetts. There was a wide range of performance between the different crosses.

Data were collected on the performance of the crosses during the larval, settlement and early juvenile periods at both hatcheries. A series of larval transfers were conducted so that the effects of broodstock, spawning site, culture site and setting site could be assessed. Samples were obtained from all broods for analysis of biochemical and microbiological composition, as well as larval anatomy. Physiological data were also obtained at ARC, and during next season’s analysis it will be obtained at both hatcheries.

Ongoing biochemical analyses of the different cohorts suggests that differences in setting success...
and levels of biochemical stores (protein, carbohydrate and lipid) were closely associated with the sites at which the animals were raised, not where they were set. There appear to be several correlations between biochemical constituents and broodstock and rearing sites. Carbohydrate levels rose significantly from the D-hinge stage to about day 10 and leveled off, suggesting the importance of different types of algae during certain periods of larval culture. Metamorphic success of transferred larval groups was related to both stock origin and culture site.

Studies on larval physiology indicate greater capture of diatom Chaetoceros cells but greater ingestion rates of Isochrysis cells. Differences in ingestion rates were noted with larvae of different ages, indicating that larvae feed in a batch mode rather than continuously as previously thought. Data on juvenile heart activity and oxygen consumption are being processed. In addition, a time-lapse color video documentation of larval metamorphosis was initiated this year.

Of the 16 cohorts sampled during the first year, histologic evaluation is being done on only the 4 most important groups during the time of highest mortality. All the groups, however, were archived covering days 4 to 34 in larval culture.

Batches of oyster larvae from six reciprocal crosses were also cultured at the Milford laboratory with both flagellate and diatom diets. There were clear effects of shipping and/or different rearing conditions between the hatcheries and the Milford Lab. It was observed that pH was significantly positively correlated with survival in the different groups. Larvae fed on the flagellate diet showed more rapid growth and better survival than those fed on the diatom diet.

The extension portion of the project was successful in completing the filming for an instructional video on remote setting.

Usefulness of Findings

If the technology of remote setting can be successfully adapted to the northeast region, perhaps much of the once productive oyster grounds can be brought back into production. If the biological causes of larval and juvenile mortality can be understood, the industry may be closer to achieving historic production levels through aquaculture. If the role of nutritional stores in oyster larvae and their relation to metamorphic mortality can be clearly documented, the possibilities of improved survival by manipulating culture conditions may provide a breakthrough for the industry. Understanding biochemical and physiological processes in healthy organisms and determining the relative vulnerability to stress in these early stages is a prerequisite to determining the cause of mortalities during this particularly sensitive transition period. Studies of the histopathology will help identify the changes within organ systems and cells of the animals leading to the observed high mortality in some groups of remotely set oyster.

Work Planned for Next Year

The majority of the histopathologic work from year one will be accomplished during the early part of 1993. Collecting and archiving the cohorts in year 2 of the study will be accomplished; however, part or all of these will be examined at a later time as part of another grant. Data analysis of the respiration measurements and video tapes of larvae from different ages and crosses will occur early in 1993, and equipment has been obtained for parallel measurements at both ARC and HPEL. Statistical analyses of the biochemical data will continue, and further measurements, including a pilot project on immunocytochemistry for quantifying developmental rates in larval and juvenile oyster tissues, will be made on the crosses in 1993. The project will also include estimates of feeding and assimilation by the larvae using radiotracers both at ARC and HPEL. A meeting of the project PIs will take place at the Milford Shellfish Biology Meeting in February to coordinate the second year’s efforts and discuss the results from the first year’s results. During the second year, written results from the study will be disseminated to all hatcheries in the region with suggestions for improving survival of remotely set oysters. Two remote setting workshops will be held, in coordination with state aquaculture associations.

Publications Issued or Manuscripts Approved During the Year

None

Northeastern Regional Aquaculture Center
Total Funding $35,029

Period April 6, 1992 - December 31, 1992

Work Group Chair Cathy Wessells, University of Rhode Island

Participating Investigators/Cooperative Agencies

Cathy Wessells University of Rhode Island
James Anderson University of Rhode Island
Thomas Weaver University of Rhode Island
Nancy Balcom University of Connecticut
Linda Marek-Howe University of Vermont
Brian Beal University of Maine at Machias
Ralph Boragine American Seafood Institute

Rhode Island
Rhode Island
Rhode Island
Connecticut
Vermont
Maine
Rhode Island

Overview

It is generally accepted that aquaculture production and its contribution to the economy is growing. However, in the Northeast, only a few states have evaluated their aquaculture sectors and, regionally, only the pen-reared salmonid industry has been comprehensively surveyed. In order to assist investors, bankers and funding agencies in their distribution of capital and funding resources, it is important to identify areas of rapid growth and potential economic value of the various segments of the aquaculture sector. There is also a need to identify constraints to development and potential opportunities.

Objectives

- To compile preliminary estimates of private Aquaculture production and value for the Northeastern region.
- To identify future opportunities and current problems facing the industry.
- To assist identifying priority research directions based on industry need.

Progress and Principal Accomplishments

The project is being carried out by investigators at the University of Rhode Island, the University of Maine, the Sea Grant Marine Advisory Program at the University of Connecticut, the University of Vermont, and the American Seafood Institute.

A list of 1,174 potential aquaculture producers in the Northeast Region generated through various sources has been compiled. Various state and local government agencies were contacted in order to obtain lists of lease holders, fish culture permit holders, etc. This task was complicated by the heterogeneity of aquaculture regulatory agencies between states. However, most of the states that actually maintain lists of aquaculture producers were cooperative in supplying them. As a final source, various aquaculture industry directories were obtained and cross referenced with the existing list. Based upon work completed to this point, it is estimated that less than 50% of the list is comprised of active private aquaculture operations. Many of the individuals on the list are leaseholders with no current aquaculture production.

At this time, 137 in-depth interviews have been completed. Of this number, 75 have been completed in-person at the respective culture sites. The remaining 62 interviews have been conducted over the telephone. It is believed that most of the major producers have either already been contacted or will be personally contacted by the end of February. In addition, all potential producers on the list will be contacted through a brief mail survey and will be offered a free subscription to the American Seafood Institute Report in return for their cooperation. This mailing is presently being coordinated by the University of Rhode Island and the American Seafood Institute. The questionnaires will be mailed no later than February 15th.

As the lead institution in the project, the University of Rhode Island has compiled the list of potential producers and coordinated the efforts of the project participants. In addition, University of Rhode Island researchers have made trips to Maine, Massachusetts, New York, New Jersey, Pennsylvania, and Maryland in order to conduct personal interviews with major private aquaculture...
producers. University of Rhode Island researchers have conducted 117 interviews as of this date.

Linda Marek-Howe, Extension Assistant Professor at the school of Natural Resources, University of Vermont, is the principal investigator for the project in the State of Vermont. As of this date, Ms. Howe has completed eight interviews out of a list of ten potential private aquaculture producers in the state.

Nancy Balcom, with the Sea Grant Marine Advisory Program at the University of Connecticut, has assisted in interviewing producers within the State of Connecticut. Ms. Balcom has completed three interviews including the largest oyster producer in the region. Ms. Balcom is in the process of completing the remaining six shellfish interviews within the state.

Chris Van Orsdel, a Graduate Research Assistant under the advisement of Dr. James Wilson at the University of Maine, has participated along with a team of URI researchers in conducting interviews with major salmon producers in the state. Mr. Van Orsdel has also conducted an additional nine interviews with primary aquaculture producers.

The Aquaculture office of the Maryland Department of Agriculture has also been especially helpful in providing data from their own survey of aquaculture production in the state. These data, along with information gathered through personal interviews with key industry participants, will be useful in completing the Maryland portion of the Situation and Outlook report.

Usefulness of Findings

This project will yield a valuable report which can be used to identify the current status and trends in the industry, and in setting research priorities. This information will also be of value to industry participants, extension personnel, legislators, policy makers, researchers, and potential investors.

Work Planned for Next Year

Project participants are continuing to identify and interview active private aquaculture producers in the Northeast region. At the same time, the American Seafood Institute in conjunction with the University of Rhode Island is preparing to mail brief questionnaires to all potential producers in the region. Data collection should be completed by the end of February. It is anticipated that the final document will be completed by the end of April.
"Unexplained Mortalities of Hatchery-Reared Oysters, \textit{(Crassostrea virginica)}, in the Northeast: A Followup Study"

**Total Funding**  
$20,000

**Period**  
March 23, 1992 - December 31, 1992

**Work Group Chair**  
Susan Ford, Rutgers University

**Participating Investigators/Cooperative Agencies**

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<td>Stan Czyzyk</td>
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<td>Frank Perkins</td>
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**Overview**

Heavy, unexplained mortalities of hatchery-reared oysters, \textit{Crassostrea virginica}, have been documented over the past two-three years by commercial oyster growers from several northeastern states. Losses have typically occurred during early field growout of seed held in suspended culture, and ranged from 50-92\% of the hatchery production. From north to south, oyster mortalities were observed in the Damariscotta River, Maine, at several sites in Massachusetts, Oyster Bay, New York, and in Fishers Island, New York. Mortalities associated with similar symptoms were also experienced by a Rutgers oyster strain of Long Island Sound origin, undergoing testing for MSX disease at Deal Island, Maryland in 1990.

**Objectives**

- To determine whether the mortalities are associated with a particular broodstock or hatchery.
- To determine whether the mortalities are associated with grow-out site.
- To determine whether the mortalities can be stimulated by experimental temperature elevation.
- To document the association of tissue and shell abnormalities with mortalities in the various experimental treatments.
- To continue efforts to determine whether the mortalities are associated with a pathogen.

**Progress and Principal Accomplishments**

The overall objective of this project was to continue the work begun with emergency funds from NRAC in 1991, to determine the cause of serious mortalities of hatchery-reared, juvenile eastern oysters, which have occurred in the Northeast over the past several years, and to suggest methods for alleviating the problem. The 1991 study produced evidence that a toxin, probably of bacterial or microalgal origin, may be associated with the mortalities. Nevertheless, concern remained that a pathogenic protozoan was causing the deaths and might be transmitted through the broodstock. The specific objectives in 1992 were to determine whether mortalities were, in fact, associated with a particular broodstock (either a genetic problem or a source of pathogen transmitted to offspring) or growout site, whether mortalities could be stimulated by experimental temperature elevation, whether an association existed between tissue and shell abnormalities and mortalities in the various experimental treatments, and whether a pathogen could be detected through light or electron microscopy.

High mortalities occurred for the third year at the experimental site on the north shore of Long Island, even though a different broodstock was used. Conversely, offspring of the original north shore broodstock experienced no mortalities when produced and grown on the south shore of Long Island. Oysters maintained inside the north shore hatchery at elevated temperature (25° C) in mixed well salt water and 25-μm filtered bay water supplemented with cultured algae, did not experience typical Juvenile Oyster Mortalities. Pathological results supported earlier findings: lesions occurred on the mantle just before and during the mortality episodes, but no evidence of a pathogenic protozoan was found.
Usefulness of Findings

Hatchery operators and growers can conduct business with the knowledge that there is no evidence to link a particular broodstock with the problem, and that intensive pathological studies have failed to detected a contagious protozoan. The results also support the conclusions of the 1991 study, that an agent in ambient waters, such as microalgae or bacteria, may be responsible for the losses, and point the direction for addition work.

Work Planned for Next Year

The project was for one year only; however, a new proposal has been submitted to concentrate on the potential role of bacteria or microalgae in the mortalities.

Publications Issued or Manuscripts Approved During the Year

None
"Fish Counting and Measurement in Situ: A Technology Assessment"

Total Funding $6,899
Period May 12, 1992 - December 31, 1992
Work Group Chair John E. Huguëtin, Massachusetts Maritime Academy

Participating Investigators/Cooperative Agencies
John E. Huguëtin Massachusetts Maritime Academy Massachusetts

Overview
The accurate counting and measurement of fish in situ, without having to individually handle and stress the fish, has become a critical need in fish farming. It is essential input information for financing, insurance, stock management and feeding activities. Since 40-60% of the total production costs often involve feeding, these data are essential for efficient and economical feed management. Inefficient feed management for large biomasses is not only a major economic loss but can also lead to serious environmental problems. While the economic importance of uncertainty about size and numbers has increased with the emergence of aquacultural industries, it is not a new problem and such data are also of value in wildlife management. There is a considerable history of attempted technical solutions, including acoustical, electrical and optical methods, involving a number of application areas. These application areas include stream monitoring, fish stock assessments, hatcheries, and culturing objectives. The relevant technical data are widely scattered.

Objectives
- To document past and ongoing research, and evaluate and recommend the most promising approaches for accurate counting and measurement of fish in situ.

Progress and Principal Accomplishments
The project has developed essentially as originally proposed and about as anticipated. The status and state of development is just about exactly where it should be at this time. The work plan as originally proposed had several different supportive aspects. These are summarized below:

Literature Search- A broad spectrum literature search has been carried out. A bibliography with about 50 citations and copies of all of them has been collected. Several of these are complete "unpublished" reports of considerable relevancy. This phase has essentially been completed.

Existing Equipment/Research- A large number of telephone calls and letters have been sent all over North America. About 30 letters have been sent to select individuals and groups in Europe and another 30 to the far east (mostly Japan). These have inquired about existing equipment and ongoing research projects. The responses from the U.S. and Canada, as well as Europe (mostly Norway), have been many and varied. This project has a good idea of the relevant current status and developments in these parts of the world. A list of 13 currently available commercial items has been assembled. Some of these have promise for use in New England but others are of marginal relevancy. The response from Japan has been disappointing and new leads into Japan would be appreciated. While there are a few loose ends in addition to Japan, this phase is largely completed.

Evaluation, Assessment & Recommendations- Information has been collected to date, evaluation is now underway.

Usefulness of Findings
A comprehensive evaluation of the widely scattered past and present efforts is needed for proper research planning. This includes documentation of past and ongoing research, evaluation of the most promising approaches and recommendations for future efforts. Without this perspective, research money's are likely to be wasted on redundant or unproductive approaches.

Work Planned for Next Year
The next phase is to organize and evaluate the collected information as well as pursuing any loose ends from the previous phases. A rough draft of this evaluation is expected to be completed around the end of Feb. 1993. The updated and final report is expected to be completed in early May with a
one or two page stand alone abstract. Depending on how the evaluation develops, a rough draft manuscript for publication based on the report may also be produced at the same time.

Publications Issued or Manuscripts Approved During the Year

None
NRAC92-7  "Creation of High Survival Resistant Lines of American Oyster Using MSX-Resistant Strains"

Total Funding  $8,625
Period  July 13, 1992 - December 31, 1992
Work Group Chair  Standish K. Allen, Rutgers University

Participating Investigators/Cooperative Agencies

| Standish K. Allen | Rutgers University | New Jersey |

Overview

Since 1958 Rutgers has been breeding American oysters for resistance to MSX-disease. Until about 10 years ago, this work has been relatively unnoticed. With the spread of MSX into the northeast region in the early 1980s, Rutgers Resistant (RR) oysters have received increasing attention for their considerable potential to mitigate MSX-disease losses in commercial aquaculture stocks.

Rutgers maintains a commitment to continuing these strains and to strategies for further improvements in the American oyster. This work is accomplished principally at the Cape Shore Hatchery located toward the mouth of Delaware Bay. Here MSX-disease pressure is probably the most consistent of any location in the US. Consistent disease pressure is essential to continued development and maintenance of resistant stocks.

The second major east coast disease, Dermo, has appeared recently in northeast sites as well. In 1990 it appeared in Delaware Bay, and in 1991--Massachusetts. RR oysters are not resistant to Dermo. Additionally, because genetic variability in RR stocks has been constrained through intense selection pressure and population bottlenecks, they may be more susceptible. Almost certainly, there is enough loss of genetic variability in RR oysters to question the wisdom of selecting for Dermo resistance from any one of the RR strains per se.

However, among all the resistant strains, much of the original genetic variation still exists. Reconstituting that variation is possible by mating the strains among themselves creating a superstock, a high survival line. Such a super-stock should be fully resistant to MSX, as all the individual strains had been, with the additional advantage of increased genetic variation. This variation would be advantageous for two major reasons. First, it would eliminate the apparent inbreeding problems seen recently in various commercial hatcheries during propagation of certain strains. Second, it would reconstitute additive genetic variance which could be exploited for selection to Dermo resistance. In fact this strategy is our best hope for creating stocks of oysters resistant to both MSX and Dermo.

Objectives

- To create two new strains of resistant oysters, consisting of:
  1. five founder populations derived from Delaware Bay MSX-resistant strains and
  2. five founder populations from Long Island MSX-resistant strains.

These strains will collectively be called High Survival Resistant Lines (HSRL).

Progress and Principal Accomplishments

From June 10 to July 21, 1992, we produced two geographic races of HSRL: a Delaware Bay (DB HSRL) and a northeast race (NE HSRL). Broodstock for DB HSRL comprised four strains of resistant oyster, now three years old, and wild stock from Delaware Bay. All these populations have been exposed to Dermo pressure for at least one generation. Constituent populations for NE HSRL included 2 succeeding generations of Long Island RR strains (BLA and CLA), and Flowers, Inc. and Ocean Pond, Inc. varieties of the BLA line. We did not introduce Long Island wild stock genes into NE HSRL.

Founder populations were produced by controlled matings among (but not within) each constituent population. Matings were made for both DB HSRL and NE HSRL using as many pairs as possible from each constituent population. Five founder sub-populations for each race (DB or NE) were produced, but we lost one of the NE sub-populations.
Through a series of mating among (but not within) strains, we produced a total of 2388 families comprising the five DB HSRL sub-populations and 3287 families comprising the five NE HSRL sub-populations. The current inventory of the strains going into the winter of 1993 is as follows: DB HSRL 10--6400; DB HSRL 20--2400; DB HSRL 30--2200; DB HSRL 40--19000; DB HSRL 50--46000; NE HSRL 10--25000; NE HSRL 20--33000; NE HSRL 30--3400; NE HSRL 40--19000.

Usefulness of Findings

The effort described above represents a new direction for the MSX-resistant stocks. It is expected that these new populations (already fully resistant to MSX-disease) will be available for industry use beginning in 1994. Resistance to Dermo will be gradual, and as resistance is acquired, the stocks will be more and more useful to areas affected by either or both diseases.

Work Planned for Next Year

Although our funding was for only one year (the year in which crosses were carried out), these 5 founder populations (F0), with restored genetic variability, will serve as the basis for future generations of HSRLs. The reason for subdividing the lines is so that each sub-population can be cross bred to other sub-populations (but not to itself) in future generations. Such a crossing scheme (rotational line crossing) prevents matings between closely related individuals. Also, because the high survival line comprises 5 sub-populations, high effective population sizes are maintained, minimizing genetic drift. Breeding adjacent sub-populations reciprocally (but not to themselves) will produce a new generation (F1) also consisting of 5 sub-populations in each geographical race.

Publications Issued or Manuscripts Approved During the Year

None
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The Office of Fiscal Affairs at the University of Massachusetts Dartmouth provides the necessary administrative support in the management of NRAC grants and subawards. Michelle Levesque, NRAC secretary, coordinates all daily activities of the Center and was central in the compilation and layout of this annual report. Betsy Tiedemann and Christine Phillips accurately typed the reports into the appropriate format. Jim Feeley of the University of Massachusetts Dartmouth Graphics/Photo department provided the illustration on the front cover.