CENTER
FOR
TROPICAL
AND
SUBTROPICAL
AQUACULTURE

Center for Tropical and Subtropical Aquaculture

2000 Accomplishment Report

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In cooperation with

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EDUCATION · RESEARCH · EXTENSION

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INTRODUCTION

The mission of the Center for Tropical and Subtropical Aquaculture (CTSA) is to support aquaculture research, development, demonstration and extension education to enhance viable and profitable U.S. aquaculture.

Title XIV of the Agriculture and Food Act of 1980 and the Food Security Act of 1985 authorized establishment of five regional aquacultural research, development and demonstration centers in the United States (Subtitle L, Sec. 1475[d]) in association with colleges and universities, state departments of agriculture, federal facilities, and non-profit private research institutions.

CTSA is one of the five centers funded by the U.S. Department of Agriculture. Research projects span the American Insular Pacific, using its extensive resource base to meet the needs and concerns of the tropical aquaculture industry.

The five Regional Aquaculture Centers encourage cooperative and collaborative aquaculture research and extension education programs that have regional or national applications. Center programs complement and strengthen existing research and extension educational programs provided by the U.S. Department of Agriculture and by other public institutions. The Centers' objectives are to:

- promote aquaculture research, development and demonstration for the enhancement of viable and profitable commercial aquaculture production in the United States for the benefit of producers, consumers and the American economy;

- utilize the Regional Centers in a national program of cooperative and collaborative research, extension and development activities among public and private institutions having demonstrated capabilities in support of commercial aquaculture in the United States.

CTSA is jointly administered by the University of Hawaii and The Oceanic Institute. The Center offices and staff are located at The Oceanic Institute’s Makapu`u Point site on windward Oahu.

ORGANIZATIONAL STRUCTURE

CTSA funds aquaculture research, development and demonstration projects. Each year's program is the result of several groups working together for many months. A Board of Directors oversees the Center's programmatic functions, and an Executive Committee is responsible for the Center's administrative policy and functions.

In addition, CTSA has two working groups. The Industry Advisory Council (IAC) comprises members from financial institutions, aquacultural and agricultural enterprises, government agencies and other business concerns. The Technical Committee (TC) is made up of researchers, extension agents and fisheries officers.

The Board, the IAC and the TC draw their members from American Samoa, the Commonwealth of the Northern Mariana Islands, the Federated States of Micronesia, Guam, Hawaii, the Republic of Palau and the Republic of the Marshall Islands.

ADMINISTRATIVE CENTER

CTSA is co-administered by the University of Hawaii and The Oceanic Institute. CTSA's Administrative Center is located at The Oceanic Institute, on the island of Oahu in Hawaii. The Administrative Center staff provides all necessary support services for the Executive Committee, the Board of Directors, the Industry Advisory Council, the Technical Committee, various project review panels and delegations and project work groups. Dr. Cheng-Sheng Lee, Center Director, supervises operation of the Center.

EXECUTIVE COMMITTEE

The Executive Committee is the legal entity responsible for the Center's overall administrative policy formulation, budget and procedures. It also appoints the CTSA Director. The members of the Executive Committee are:

- Dr. Gary D. Pruder, The Oceanic Institute, {Executive Committee Chairman};
- Dr. Dean Smith, University of Hawaii, {Board of Directors Chairman}.

BOARD OF DIRECTORS

The Board of Directors is responsible for the development and implementation of the Center's program policy, including concurrence on total budget issues. The Board is also responsible for development of ancillary agreements with other agencies and institutions.

The members of the Board of Directors represent educational, state and non-profit private research institutions throughout the region. The Board of Directors:

- establishes initial guidelines for regional aquaculture research, development and demonstration activities;
- appoints and removes members of the Industry Advisory Council and the Technical Committee;
- approves the proposed strategy for project selection;
- approves the priority areas and goals for industry development identified by the Industry Advisory Council and Technical Committee;
- approves the Annual Plan of Work, including budget allocations;
- approves the Annual Accomplishment Report for consistency with the goals and objectives of CTSA and the authorizing legislation;
- develops ancillary agreements with other institutions.

The members of the Board of Directors are:

- Dr. Jeff Barcinas, College of Agriculture and Life Sciences, University of Guam;
- Mr. John Corbin, Hawaii State Aquaculture Development Program;
- Dr. E. Gordon Grau, University of Hawaii Sea Grant College Program;
- Dr. Michael Harrington, Hawaii Institute of Tropical Agriculture and Human Resources, University of Hawaii;
- Dr. Charles Helsley, Sea Grant College Program, University of Hawaii;
- Dr. Gary D. Pruder, The Oceanic Institute {Executive Committee Chairman};
- Dr. Singeru Singeo, Land Grant Program, College of Micronesia;
- Dr. Dean Smith, University of Hawaii, {Board Chairman}.

INDUSTRY ADVISORY COUNCIL

Members of the Industry Advisory Council include commercial aquaculture farmers, aquaculture suppliers and members of government bodies and financial institutions. Members are appointed by the Board of Directors for 3-year, renewable terms. As an advisory body, the Industry Advisory Council's capacity provides an open information exchange forum for those involved in the aquaculture business. With the approval of the Board of Directors, contributions of the IAC can be incorporated into annual and ongoing plans for CTSA. The Industry Advisory Council:

- recommends research and development needs and priorities from the perspective of the aquaculture industry;
- participates as needed in the review of proposals, project progress reports, program review delegations and other functions of the Center;
- recommends to the Board actions regarding new and continuing proposals, proposal modifications and terminations.

Members of the Industry Advisory Council are:

- Mr. David Barclay, Aquatic Culture and Design;
- Dr. Paul Bienfang, CEA Tech USA Inc.;
- Mr. Dennis Bishop, Kona Mariculture;
- Ms. Rebecca Bishop-Yuen;
- Ms. Mary Brooks, Pacific Aquaculture;
- Mr. Steve Chaikin, Molokai Sea Farms;
- Mr. Michael Crisostomo, Kurumaya Seahorse Restaurant;
- Mr. Richard Croft, Pohnpei Natural Products;
- Mr. Yimnang Golbuu, Palau Community College;
- Mr. John Gourley, Micronesia Clam Company;
- Ms. Linda Gusman, Island Aquaculture;
- Mr. Steve Katase, Royal Hawaiian Sea Farms;
- Mr. Robert Kern, Tropical Ponds of Hawaii;
- Mr. Jeff Koch, Mokuleia Aquafarm;
- Mr. Andrew Kuljis, Aquatic Farms;
- Dr. Todd Lorenz, Cyanotech;
- Mr. Richard Masse, Mangrove Tropicals;
- Mr. Ramsey Reimers, Robert Reimers Enterprises;
- Mr. Toshiuki Rudolph, Kukuoro Municipal Government;
- Mr. Neil Sims, Black Pearl, Inc.;
- Dr. Richard Spencer, Hawaiian Marine Enterprises {Industry Advisory Council Chairman and *ex officio* member of the BOD}
- Mr. Ron Weidenbach, Hawaii Fish Company;
- Dr. James Wyban, High Health Aquaculture;
- Dr. Leonard Young, Hawaii State Aquaculture Development Program.

TECHNICAL COMMITTEE

The Technical Committee's members represent participating research institutions and state extension services, other state or territorial public agencies as appropriate, and non-profit private research institutions. The Technical Committee provides research expertise to address priorities set by the Industry Advisory Council. The Board of Directors appoints members for 3-year, renewable terms.

The Technical Committee:

- prepares Problem Statements for priority areas identified by the Industry Advisory Council;
- participates as needed in project review panels, Program Review Delegations and other functions of the Center.

The members of the Technical Committee are:

- Dr. Harry Ako, University of Hawaii {Technical Committee Chairman and *ex officio* member of the BOD};
- Dr. Maqsudul Alam, University of Hawaii;
- Ms. Kristen Anderson, Hamilton Library, University of Hawaii;

- Dr. Brad Argue, The Oceanic Institute;
- Mr. Richard Bailey, Sea Grant Extension Service, University of Hawaii;
- Dr. James Brock, Hawaii State Aquaculture Development Program;
- Dr. John Brown, College of Agriculture and Life Sciences, University of Guam;
- Mr. David Crisostomo, University of Guam Cooperative Extension Service;
- Mr. Simon Ellis, Land Grant College Program, College of Micronesia;
- Dr. Robert D. Howerton, Sea Grant Extension Service, University of Hawaii;
- Mr. Tom Iwai, Anuenue Fisheries Research Center;
- Dr. Christopher Kelley, Hawaii Institute of Marine Biology;
- Dr. PingSun Leung, University of Hawaii;
- Dr. Shaun Moss, The Oceanic Institute;
- Dr. Anthony Ostrowski, The Oceanic Institute;
- Dr. James Szyper, Sea Grant Extension Service, University of Hawaii at Hilo;
- Dr. Albert Tacon, The Oceanic Institute;
- Mr. Howard Takata, Sea Grant Extension Service, University of Hawaii
- Dr. Clyde Tamaru, Sea Grant Extension Service, University of Hawaii.

EXECUTIVE SUMMARY

PROGRAM SCOPE

During 2000, the Center for Tropical and Subtropical Aquaculture completed work on projects funded under its Tenth Annual Plan of Work and continued work on projects funded under its Eleventh and Twelfth Annual Plans of Work. In addition, in June 2000, the Center initiated work on projects developed under its Thirteenth Annual Plan of Work and began developing its Fourteenth Annual Plan of Work.

Seven projects were funded under the Center's Thirteenth year program, which was approved by the Center's Board of Directors on December 2, 1999. Five were continuations of projects begun under the programs of previous years and two were continuations of projects funded in previous years with new components added.

Since the inception of the Center for Tropical and Subtropical Aquaculture in 1988, it has funded 131 research, demonstration, development and extension projects. Eleven projects were active during 2000. These projects fall into six categories:

- National Aquaculture Priorities;
- Information Dissemination;
- Extension Support to Further Industry Development;
- Marketing and Economics;
- Development of New Technologies;
- Demonstration and Adaptation of Known Technologies.

These projects address national aquaculture priorities:

* National Coordinator for New Animal Drug Applications

These projects address information dissemination:

* Library Aquaculture Workstation
* Publications

These projects address extension support to further industry development:

* Aquaculture Extension and Training Support for the U.S. Affiliated Pacific Islands
* Disease Management for Hawaiian Aquaculture

This project addresses marketing and economics:

* Development of Pacific Threadfin and Milkfish Growout Technology and Production of Live Feeds and Seedstock

These projects address development of new technologies:

* Development of Pacific Threadfin and Milkfish Growout Technology and Production of Live Feeds and Seedstock

* Marine Foodfish Seedstock Production
* Marine Ornamental Fish Culture Conservation
* Aquaculture of Marine Ornamentals

These projects address demonstration and adaptation of known technologies:

* Transitioning Hawaii's Freshwater Ornamental Industry
* Expansion & Diversification of Freshwater Tropical Fish Culture

A brief listing of the principal accomplishments of the active projects in these categories during 2000 is presented below. Details on each project's funding, participants, objectives, anticipated benefits, progress and future plans are presented in the Progress Reports section.

National Aquaculture Priorities

National Coordinator for Aquaculture New Animal Drug Applications

In the year 2000, the National Coordinator has established seven NADAs for human chronic gonadotropin, a supplemental NADA for formalin and an additional NADA for MS-222. Furthermore, data packages were submitted for AQUI-S™, chloramine-T, copper sulfate, formalin (extension), florfenicol, hydrogen peroxide, 17 alpha-methyltestosterone, oxytetracycline, potassium permanganate, and Pyceze™.

Information Dissemination

Library Aquaculture Workstation

During this time period, there were 7,007 logins to the ASFA database with a total of 16,383 queries. The website was accessed an average of 7,898 times per month during this reporting period with an average of 24 research or journal requests per month. PRAISE staff responded to requests by forwarding to their users 2,437 journal citations, delivering 692 documents totaling 774 pages and answering 99 miscellaneous inquiries. In addition, the Gray Literature database was expanded as was the website.

Publications

As planned, a new design was implemented for the quarterly newsletter and was published in November 1999, and April, July and October 2000, and disseminated thereafter. The Center assisted with the creation, production and distribution of four publications. The homepage was updated monthly and maintained as needed.

Extension Support to Further Industry Development

Aquaculture Extension and Training Support for the U.S. Affiliated Pacific Islands

The extension agent conducted seven workshops and several training courses for private and government agencies and provided other extension services to improve public outreach and training programs. Approximately 200 pamphlets and 10 videos were disseminated on a variety of aquaculture topics. Six new publications were printed.

Disease Management & Virology Service for Hawaiian Aquaculture

To assist aquaculture operations, 260 trips were made to the field. There were 658 case submissions of aquatic animals received for diagnostic laboratory analysis services. Ninety-five individuals or organizations requested assistance from the project. Additionally, the project was able to control a fish mortality in a shipping transfer, control an outbreak of *Cryptocaryon sp* and *Amyloodinium* disease. Additionally, several trials were performed with limited success from each one. Especially notable is the discovery that outbreaks of columnaris disease in juvenile catfish may respond best if the water temperature of the rearing unit is increased above 25 degrees and the fish receive treatment with hydrogen peroxide for 30 minutes at a dosage of 250 ppm.

Development of New Technologies

Development of Pacific Threadfin and Milkfish Growout Technology and Production of Live Feeds and Seedstock

Several growout trials were performed during the duration of this project, which allowed them to: refine threadfin growout techniques; develop alternative milkfish growout techniques; demonstrate the interaction between diet and pond environment on growout of milkfish; and determine that polyculture of shrimp and milkfish is beneficial to both because the overall biomass production from a pond is improved. These techniques were then transferred to the farmers in the form of site visits and/or telephone conversations or email. Three threadfin farmers have been established in Hawaii as a direct result of the fingerlings and technical assistance provided by this project in the past three years.

Marine Food Fish Seedstock Production

During the report period, the OI hatchery supplied 1.7 million threadfin and 1.34 million milkfish eggs to three growout and research operations on the islands of Hawaii and Oahu. Two threadfin production runs were conducted over the first 6 months of this project (April and July, 2000) following the quarterly schedule requested by farmers. A total 118,640 fish have been distributed to three farmers on the islands of Hawaii and Oahu. Efforts to implement a selected breeding program for Pacific threadfin have been initiated with the development of a domesticated broodstock group.

Aquaculture of Marine Ornamental Species

Under captive conditions at the Oceanic Institute, the project has been successful in obtaining flame angelfish spawns daily for almost one year with individual spawns from a single pair, reaching values as high as 12,000 eggs per spawn. Additionally, success has been documented in developing an external means of sexing individuals based on external characters for yellow tang. Broodstock populations for yellow tang and pygmy angelfish have been established and several species of juvenile wrasse were obtained and habituated to onshore tanks.

Marine Ornamental Fish Culture Conservation

The collection of eggs and larval and juvenile fish from Kaneohe Bay was attempted in two different ways and both proved to be unsuccessful. Several mitigating factors played into the failure of this

project including the location and duration of the trials. However, the project did succeed in collecting either the larvae or eggs of several different species of fish highly prized by aquarium hobbyists.

Demonstration and Adaptation of Known Technologies

Transitioning Hawaii's Freshwater Ornamental Industry

The project was able to demonstrate that sex ratio is clearly linked with the variety of swordtail being cultured. The freshwater rotifer variety, *Brachionus calciflorus*, has been made available to farmers as a live food for freshwater ornamental fish larvae in Hawaii. The procedure of artificially inseminating the swordtail as well as other live bearers was documented and project work group members also received training in the procedure. Two workshops were held. Project work group members produced a total of 11 newsletter articles, made presentations at two conferences, submitted two manuscripts for printing in conference proceedings and submitted one manuscript to a trade magazine during the reporting period.

Expansion and Diversification of Freshwater Tropical Fish Culture

The main priority of this project is to foster the development of ornamental fish culture in Hawaii. Success is difficult to measure, but is demonstrated by the ranking of freshwater ornamental culture as its own group since 1997, rather than grouped in "Other" as it had been previously. The project can also be attributed for the dramatic increase in farms from 22 in 1994 to 71 in 1998. On average, funded personnel responded to 600 requests for technical assistance annually during the course of the projects. A collaborative workshop series held previously which targeted rural communities was repeated.

A LOOK AHEAD AT YEAR 14

The development of the Year 14 program was initiated in February 2000 at the annual meeting of the Industry Advisory Council (IAC). The IAC reviewed the progress of funded projects and recommended Year 14 research priorities that would aid industry development. Members identified seven project areas, two of which were new and one was an expanded area with new components. The priority areas were:

1. Library Aquaculture Workstation – Year 14 *(continuing priority)*;
2. Research, Extension and Training in the U.S.-Affiliated Pacific Islands – Year 13 *(continuing priority with additional component);*
3. National Coordinator for Aquaculture New Animal Drug Applications – Year 6 *(continuing priority);*
4. Disease Management & Virology Service for Hawaiian Aquaculture – Year 9 *(continuing priority)*;
5. Marine Food Fish Seedstock Production – Year 2 *(continuing priority);*
6. Transitioning Hawaii's Freshwater Ornamental Aquaculture Industry – Year 2 *(continuing priority)*;
7. Aquaculture of Marine Ornamental Species – Year 2 *(continuing priority);*
8. Black Pearl Culture in the Pacific – Year 1 *(new priority);*
9. Capture and Growout of Juvenile Coral Reef Fish – Year 1 *(new priority);*
10. Publications *(continuing priority)*.

In February 2000, the Technical Committee (TC), acting on the IAC's recommendations, drafted problem statements for new or expanded projects. Those formed the basis for the Preliminary Plan of Work, which was approved by the Board of Directors in March 2000. The Center staff then solicited proposals for projects, and ten proposals were submitted.

In June, the Center began its four-month review process. New proposals were first subjected to external peer review by at least three experts in the project topic area. The expert peer reviewers were identified with the assistance of the directors of the other Regional Aquaculture Centers and the U.S.D.A. program administrators. Proposals for both new and continuing projects then underwent review by panels comprising members of the Industry Advisory Council and the Technical Committee. The final version of the proposals are incorporated into the Fourteenth Annual Plan of Work, which will be presented to the Center's Board of Directors for approval in January 2001. Following Board approval, the plan will be submitted to the U.S. Department of Agriculture Cooperative State Research, Education and Extension Service for final approval.

PROGRESS REPORTS

An individual summary of the principal accomplishments of the active projects in these categories during 2000 is presented in the following pages. Details on each project's funding, participants, objectives, anticipated benefits, progress, impact, work planned and publications are presented. Information and results from previous years can be found in the correlating year's annual accomplishment report.

NATIONAL COORDINATOR FOR AQUACULTURE NEW ANIMAL DRUG APPLICATIONS – Year 5

Funding Level

Year 1	$5,000
Year 2	10,000
Year 3	10,000
Year 4	10,000
Year 5	10,000
Total	$45,000

Principal Investigator Rosalie Schnick, National Aquaculture NADA Coordinator
La Crosse, Wisconsin

PROJECT OBJECTIVES

The overall goal of this project is for the National Aquaculture Coordinator to coordinate activities for investigational new animal drug exemptions (INADs) and new animal drug applications (NADAs) to expedite approval from the Center for Veterinary Medicine (CVM) for the use of various drugs in aquaculture.

For this year, the objectives are to coordinate approval activities for these drugs:

Amoxicillin	AQUI-S™
Calcein	Chloramine-T
Crude carp pituitary	Copper sulfate
Earth Tec™	Erythromycin
17-beta estradiol	Florfenicol
Formalin (extension)	Fumagillin
Hydrogen peroxide	17á-methyltestosterone
MS-222	Ovaplant™
Ovaprim™	Oxytetracycline
Pet fish therapeuteants	Potassium permanganate
Pyceze™	Romet-30™
ReproBoost™	Sea lice control agents
Strontium chloride	Trichlorfon

Additionally, continuing objectives are to:

1. address antimicrobial resistance and aquaculture effluents issues and

2. develop minor species legislation and
3. promote international harmonization and cooperation on aquaculture drug approvals.

ANTICIPATED BENEFITS

New or amended approvals of aquaculture drugs will help reduce mortalities from disease and improve production efficiency; thus the aquaculturist can deliver more and healthier animals for consumption and recreational purposes and compete with foreign producers without drug regulations. The National NADA Coordinator will ensure that progress is made toward NADA approval and that resources (funds, people, and facilities) are used effectively.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Establishment of the National NADA Coordinator position in May 1995 has resulted in coordination, consolidation, and increased involvement in the INAD/NADA process on 18 of the 19 high priority aquaculture drugs and activities on 13 additional drugs of interest to aquaculture. Twenty-one INAD/NADA sponsors have initiated INADs or confirmed their commitment to gaining approvals of their products for the aquaculture industry.

Following are the accomplishments from this year:

Therapeutants

Chloramine-T – Major advances are being made to complete the data requirements for use to control mortalities associated with bacterial gill disease on freshwater-reared salmonids.

Copper sulfate – All submissions should soon be completed for control of *Ichthyopthirius* on catfish.

Formalin – In 1998, a supplemental NADA was obtained for formalin as a fungicide on all fish eggs and as an external parasiticide for all fish. All submissions should soon be completed for control of mortalities associated with fungal infections on *all* fish.

Florfenicol – The sponsor submitted a major package for approval for control of furunculosis in Atlantic salmon.

Hydrogen peroxide – Major advances are being made to complete the data requirements for use to control mortalities associated with saprolegniasis on finfish eggs and finfish and bacterial gill disease on freshwater-reared salmonids.

Oxytetracycline – Major advances are being made to complete the data requirements for use to control coldwater and columnaris diseases in salmonids.

RESULTS AT A GLANCE . . .

- 7 NADA for human chorionic gonadotropin
- Supplemental NADA for formalin
- Additional NADA for MS-222
- Data packages submitted for AQUI-S™, chloramine-T, copper sulfate, formalin (extension), florfenicol, hydrogen peroxide, 17 alpha-methyltestosterone, oxytetracycline, potassium permanganate, and Pyceze™.

Anesthetics

AQUI-S™ – The sponsor is ready to submit target animal safety and efficacy studies on salmonids completed in Canada to CVM. The sponsor submitted existing residue completion rates to CVM in December 1999 that gained a 21-day experimental withdrawal time. The IAFWA Drug Approval Working Group on AQUI-S™ decided on March 26th to continue to support current research on an active ingredient in AQUI-S™.

MS-222 – Two NADAs were approved for MS-222 as an anesthetic with a 21-day withdrawal time.

Spawning and Gender Manipulation Aids

Crude Carp Pituitary – The spawning and gender manipulation aid, Crude Carp Pituitary is approaching NADA approval. All submissions should be completed in 2000 for use on *all* fish.

Chorulon®--Human chorionic gonadotropin was approved on September 7, 1999 by CVM as a spawning aid by intramuscular injection for all fish and requires a prescription under the direction of a veterinarian. This approval is significant because it is the first original approval since 1986 when formalin was first approved for fish. Additionally, it was approved for *all* fish.

17 alpha-methyltestosterone – The environmental assessment has been reviewed, revised and resubmitted. All technical section submissions should be into CVM by the end of this year. The target animal safety study completed on percids by Southern Illinois University and was resent to Auburn for submission to CVM. Literature review on other species has been completed by Auburn University.

Crop grouping – Studies of crop grouping for the water-borne drug benzocaine have been completed in four species and the results have been analyzed. UMESC presented the results to CVM in the form of a seminar/meeting on August 30th. Acceptance of the crop-grouping concept by CVM will reduce residue chemistry data requirements and costs of approvals for all aquaculture drugs.

National Activities

JSA Aquaculture Effluents Task Force (AETF) met on June 7, 2000 to discuss the status of EPA's Effluent Guidelines Plan. A white paper on drugs and chemicals was submitted to EPA on August 24, 2000. A AETF Aquaculture Technical Information Exchange Forum was held September 20-21, 2000 where EPA distributed draft industry profiles and discussed issues related to approved analytical detection methods; industry profiles need revisions by October 27, 2000.

A bill entitled "Minor Animal Species Health and Welfare Act of 2000" was introduced into the House on June 27, 2000 (HR-4780) and into the Senate on October 5, 2000 (S-3169). The bill includes provisions for early life stages that should help expedite the approvals of aquaculture drugs that are of interest to public and private fish production. The draft Bill was reviewed by CVM in August 20, 2000 and changes need to be resolved with MUMS coalition.

Award – The National NADA Coordinator received the FDA Commissioner's Special Citation Award on June 9, 2000 for outstanding leadership, teamwork, and sustained efforts as the National NADA Coordinator.

IMPACT

The approval of the candidate drugs will aid the aquaculture industry to reduce mortalities associated with infectious and handling diseases and to increase their efficiency by

using spawning aids and gender manipulation aids. The domestic aquaculture industry will be better able to compete with foreign producers because there will be more legal drugs to use.

Efforts to develop the Minor Use/Minor species document into legislation will encourage more sponsors to support aquaculture drug approvals. Efforts to deal with the antimicrobial resistance issue and EPA's Effluents Standardization Plan will ensure that aquaculture can continue to legally use drugs and chemicals. Efforts on the international scale will result in more international harmonization related to aquaculture drug approvals.

WORK PLANNED

Coordinate approval activities for amoxicillin, AQUI-S™, 17 beta-estradiol, chloramines-T, common carp pituitary, copper sulfate, Earth Tec™l, erythromycin, florfenicol, formalin (extension), fumagillin, gonadotropin releasing hormone, hydrogen peroxide, 17 á-methyltestosterone, Ovaplant™, Ovaprim™, oxytetracycline, potassium permanganate, Pyceze™, and sea lice control agents. In addition there will be considerable effort on the MUMS legislation, antimicrobial resistance, Aquaculture Effluents Task Force, efficacy initiative, and international harmonization of aquaculture drug approvals and sensitivity tests.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Publications

Schnick, R.A. (In press). International harmonization of antibacterial sensitivity determination for aquaculture drugs. Aquaculture.

Griffin, B.R., R.A. Schnick, and W.H. Gingerich. 2000. Update on the Federal-State Aquaculture Drug Approval Partnership. Aquaculture Magazine 26 (3):56-68.

Schnick, R.A. 2000. Efficacy data needed for high priority aquaculture drugs. American Fisheries Society Fish Health Newsletter 28 (2):3.

Schnick, R.A. and P. Smith. 1999. International harmonisation of antibacterial agent approvals and susceptibility testing. EAFP Bulletin 19(6):293-294.

Schnick, R.A. 1999b. Use of chemicals in fish management and fish culture: past and future. Chapter 1, pages 1-14 in D.J. Smith, W.H. Gingerich, and M. G. Beconi-Barker, eds. Xenobiotics in fish. Kluwer Academic/Plenum Publishers, New York.

Schnick, R.A. 1998a. Approval of drugs and chemicals for use by the aquaculture industry. Veterinary and Human Toxicology 40 (Supplement): 9-17.

Schnick, R.A. 1997a. International regulatory aspects of chemical and drug residues. Pages 186-194 in R.E. Martin, R.L. Collette, and J. W. Slavin, editors. Fish inspection, quality control, and HACCP: a global focus. Technomic Publishing Company, Inc., Lancaster, Pennsylvania.

Schnick, R. A. 1996a. Chemicals and drugs. Pages 347-354 in R.C. Summerfelt, editor. Walleye Culture Manual. NCRAC Culture Series 101. North Central Regional Aquaculture Center Publications Office, Iowa State University, Ames, Iowa.

Schnick, R.A. 1996b. Cooperative fish therapeutic funding initiative: States in partnership with federal agencies to ensure the future of public fish culture. Transactions of the 61st North American Wildlife and Natural Resources Conference 61:6-10.

Schnick, R.A., D.J. Alderman, R. Armstrong, R. Le Gouvello, S. Ishihara, E.C. Lacierda, S. Percival, and M. Roth. 1997. Worldwide aquaculture drug and vaccine registration progress. Bulletin of the European Association of Fish Pathologists 17(6):251-260.

Schnick, R.A. and R.D. Armstrong. 1997. Aquaculture drug approval progress in the United States. Northern Aquaculture Supplement (Salmon Health Report): 22-28.

Schnick, R. A., W. H. Gingerich, and K. H. Koltes. 1996. Federal-State Aquaculture Drug Registration Partnership: A success story in the making. Fisheries 21(5): 4.

Papers Presented

Gingerich, W. H. and Schnick, R. A. 1997. Federal-state aquaculture drug approval partnership program. *In:* Book of Abstracts, World Aquaculture '97, Seattle, WA. 174.

Schnick, R. A. 1996. Aquaculture drug approval progress in the United States. Presented at Aquaculture Canada '96, 13th Annual Meeting of the Aquaculture Association of Canada, Ottawa, Ontario, June 2-5, 1996.

Schnick, R. A. 1996. Cooperative Fish Therapeutic Binding Initiative: States in Partnership with Federal Agencies to Ensure the Future of Public Fish Culture. Transactions of the 61st North American Wildlife and Natural Resources Conference. 61:6-10.

Schnick, R. A. 1997. Overview of partnerships for aquaculture drug approvals. Book of Abstracts, World Aquaculture '97, Seattle, WA. 415-416.

Schnick, R. A. 1997. Progress with registration of drugs and vaccines for aquaculture. Abstract for Workshop and round Table at the EAFP Eighth International Conference on Diseases of Fish and Shellfish, Edinburgh, Scotland, September 14-19, 1997.

Schnick, R. A. 1997. INAD and drug clearance update. Presented at the Midcontinent Warmwater Fish Culture Workshop. Springfield, MO. February 3-5, 1997.

Schnick, R. A. 1997. Current status and future needs for drugs in aquaculture: regional needs. Presented at the Workshop on International Harmonization for Drugs and Biologics, Seattle, WA. February 24, 1997.

Schnick, R. A. 1998. Upcoming successes for aquaculture drug approvals in the United States through unique partnerships. Presentation for a special session titled "The Aquaculture Drug Approval Process – The Good, The Bad and The Future is Now" to be held at Aquaculture '98, Las Vegas, Nevada, February 15 - 19, 1998.

Schnick, R.A. 2000. Overview of NADA Coordinator activities. USFWS - INAD Coordination Workshop, Bozeman, Montana, August 2-3, 2000.

Schnick, R.A. 2000. Aquaculture drug approval progress. Office of New Animal Drug Evaluations, Center for Veterinary Medicine, Rockville, Maryland, August 29, 2000.

Schnick, R.A. 2000. Aquaculture drug approval progress and need for efficacy studies. American Fisheries Society/Fish Health Section 2000 Annual Meeting, Pensacola Beach, Florida, September 6-8, 2000.

Schnick, R.A. 2000. Update on Federal-State Aquaculture Drug Approval Partnership. Annual Meeting of the International Association of Fish and Wildlife Agencies, Indianapolis, Indiana, September 15-16, 2000.

LIBRARY AQUACULTURE WORKSTATION – PACIFIC REGIONAL AQUACULTURE INFORMATION SERVICE FOR EDUCATION (PRAISE) – Year 12

Funding Level

Year 1 ……...$ 7,000
Year 2 ……..….6,700
Year 3 ……..6,000
Year 4 ……..7,000
Year 5 ……20,000
Year 6 ……14,100
Year 7 ……28,000
Year 8 ……49,000
Year 9 ……25,000
Year 10 ……..30,000
Year 11 ……..24,000
Year 12 ……..23,000
Year 13 ……..28,850
Total ……...$268,650

Current Participants

Lois Kiehl-Cain……University of Hawaii

Principal Investigator

Kristen Anderson, Information Specialist
Hamilton Library
University of Hawaii
Honolulu, Hawaii

PROJECT OBJECTIVES

The overall goal of this project is to make scientific information more accessible to the aquaculture community. Specific objectives related to that goal are:

1. Continue to provide established services.
2. Develop and maintain programs for user education (continuation).
3. Develop innovative webpages which exhibit Pacific aquaculture in an educational focus.
4. Technology transfer

ANTICIPATED BENEFITS

This project will increase the accessibility of scientific information throughout the Pacific region by providing:

- swift and accurate research literature to aquaculturists.
- increased exposure of the local aquaculture industry to the world via the internet.
- the development of a comprehensive Web resource for marketing and educational information as well as an enduser research portal specific for aquaculturists.
- a resource for the centralized accumulation and distribution of aquaculture information for the region.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

A breakthrough in the Pacific Islands' ability to access scientific information came in August 1993, when two remote workstations were established on Guam. Users at the site at the offices of the University of Guam's Cooperative Extension Service and at the Guam Department of Commerce gained access to PRAISE through a toll-free telephone line.

PRAISE entered a cooperative agreement with PEACESAT, a federally funded communications satellite, whereby residents at five Pacific Island sites can directly access the Aquatic Sciences and Fisheries Abstracts (ASFA) database through an Internet connection between the local PEACESAT station and the mainland vendor. In addition, PRAISE established a home page on the World Wide Web. Search requests can be sent to PRAISE personnel via the webpage.

The Pacific Islands Gray Literature project was established to address the inaccessibility of gray literature in the Pacific, where libraries and other organizations that collect and disseminate information are few.

Following are the accomplishments for this year:

Objective 1

Service has been greatly enhanced for PRAISE customers due to the advances in electronic technology. There are three major changes which are improving our service: 1) increased use and acceptance of the Ariel document delivery software which allows us to send articles as an email attachment, 2) the increasing number of people accessing the PRAISE website and utilizing the online request forms, and 3) the growing reliability of electronic access from Pacific Islands. Electronic access is still exceedingly expensive so time spent searching the Web is often extremely limited.

During this time period there were 7,007 logins to the ASFA database with a total of 16,383 queries. The Web site was accessed an average of 7,898 times per month during this reporting period with an average of 24 research or journal requests per month. PRAISE staff responded to requests by forwarding to users 2,437 journal citations, delivering 692 documents totaling 774 pages and answering 99 miscellaneous queries.

Objective 2

The Principle Investigator participated in the "Fish Health Seminar" in Hilo on the Big Island and discussed services offered by PRAISE as well as how individuals might do searching on their own and met with individuals to discuss searching and distributed brochures in the Kona area, specifically at businesses near the Energy Lab. Instruction

sessions were also given on Oahu, including ongoing participation with the MASSIP (Micronesia and American Samoa Student Internship Program). The Principal Investigator and the Project Assistant continue to develop instructional enhancements for the website. The Principal Investigator attended the annual conference of the International Association of Marine Science Libraries and Information Centers and focused on sessions dealing with instruction, document delivery, and digitizing projects.

Objective 3

This project is already underway. A page has been set-up with links to marketing information for aquaculture products.

Objective 4

PRAISE staff delivered 13 article reference submissions to Cambridge Scientific Abstracts for inclusion to the ASFA database. The focus was to ensure inclusion of all CTSA publications and area interest with proceedings of the PACON conferences. The PRAISE website and the Pacific Grey Literature Database are continually being updated and enhanced by the Principal Investigator and the Project Assistant. This ongoing work allows us to provide the best possible service.

RESULTS AT A GLANCE . . .

- Educated researchers, farmers, students and librarians regarding locating resources and acquisition of materials
- Updated and expanded the PRAISE website to include marketing information and education opportunities.
- Updated and expanded the Gray Literature Bibliography.
- Monetary value of services rendered: $38.759.20

IMPACT

The value of the PRAISE service is staggering. Based on rates one would pay to the information industry's major suppliers (Dialog Information Service, Inc. for access to ASFA, plus document delivery charges based on the average cost per article from UnCover, Inc.) the dollar value for our primary service may be presented as follows:

7,007 logins @ $1.20 ea. = $8,408.40
16,383 queries averaging
3 minutes each or
819 hours online @ $60/hr = $49,149.00
692 articles @ $15.50 ea. = $10,726.00

Total **$68,283.40**

The PRAISE website is a bonus. It allows users to make requests online, publicizes research being done in the Pacific via the Gray Literature Bibliography and gives local vendors a venue to advertise themselves to the world.

WORK PLANNED

- Continue to provide established services of database searching and document delivery.
- The Gray Literature Bibliography will be continually updated and improved.
- Methods to improve database access for Pacific Island residents will continue to be explored.
- Educational workshops for farmers, students, and researchers will be held on the island of Oahu and distribution of the information brochure will continue.
- The website will be continually updated. The current education webpage will be updated and expanded. A search-it-yourself page for aquaculturists is being developed. Pictures are being collected from local farms for a "virtual picture album" depicting Pacific aquaculture.
- A web tutorial is being planned and will be evaluated.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Publications

Brown, C. L. and D. E Coleman. 1991. Testing and Development of an Efficient, Remote CD-ROM System. *CD-ROM Librarian*. 13-18.

Coleman, D. E and R. L. Buettner. 1989. *A Union List of Aquaculture Journals in Hawaii*. Center for Tropical and Subtropical Aquaculture Publication #104. Waimanalo, Hawaii.

Coleman, D. E, D. Hanfman and S. J. Tibbet, eds. 1991. *Interactions of Aquaculture, Marine Coastal Ecosystems and Near Shore Waters: A Bibliography*. Bibliography and Literature of Agriculture #105. National Agricultural Library. Beltsville, Maryland.

Guenther, K. and D. E Coleman. 1994. *Pacific Islands Gray Literature Project: A Bibliography*. Center for Tropical and Subtropical Aquaculture Publication #115. Waimanalo, Hawaii.

Conference Proceedings

Coleman, D. E. 1990. Pacific Regional Aquaculture Information Service: Applied Technology and Development of a Long-Distance Information Service. *In:* International Association of Marine Science Libraries and Information Centers Conference Series: Proceedings of the Fifteenth Annual Conference.

Coleman, D. E 1991. Remote CD-ROM Searching and Satellite Communications: From Pie in the Sky to Application. *In:* International Association of Marine Science Libraries and Information Center Conference Series: Proceedings of the Sixteenth Annual Conference.

Coleman, D. E 1993. Gray Literature Project of the Pacific Regional Aquaculture Information Service. *In:* Proceedings of the Nineteenth Annual Conference of the International Association of Marine Science Libraries and Information Centers (IAMSLIC). Bethesda, MD.

PUBLICATIONS – Year 11

Funding Level	Year 1 ……..$10,000
	Year 2 ……10,000
	Year 3 ……12,000
	Year 4 ……15,000
	Year 5 ……38,000
	Year 6 ……18,000
	Year 7 ……18,000
	Year 8 ……18,000
	Year 9 ……18,000
	Year 10 ……..18,000
	Year 11 ……..33,600
	Total ……..$208,600
Current Participants	Kai Lee Awaya…….....…....CTSA
	Alcian Choy-Clegg...……..CTSA
	Jean McAuliffe.……..CTSA
Principal Investigator	Dr. Cheng-Sheng Lee
	Center for Tropical and Subtropical Aquaculture
	Waimanalo, Hawaii

PROJECT OBJECTIVES

1. Publish a quarterly newsletter to communicate information about the activities of the Center for Tropical and Subtropical Aquaculture and its funded projects and the latest information about aquaculture from the nation and the region.
2. Produce and publish informational reports of selected CTSA-funded projects, which will be distributed at no charge to commercial producers, aquaculture researchers and other interested parties throughout the Pacific region, with limited distribution in the continental United States.
3. Improve and maintain a graphically based website. The site will contain information on the Center, its activities, an annual request for pre-proposals, projects, and publications. It will also contain copies of the Center newsletter, and various publications suitable for download and printing at no cost to the user.
4. Duplicate and distribute CTSA's and other Regional Centers' publications and videos to information networks throughout the Pacific. This is coordinated with the Pacific Regional Aquaculture Information Service for Education (PRAISE), Hamilton Library, University of Hawaii and the University of Hawaii Sea Grant College Program.

5. Produce and publish the Year 2000 CTSA Accomplishments Report, which compiles information on the activities of all CTSA projects that were active during 2000 and distribute to USDA, the CTSA Board of Directors, the Industry Advisory Council and the Technical Committee.
6. Provide editing, layout and production assistance as needed on all publications produced by CTSA-funded projects to ensure that the standard of quality is kept at an exceptional level.

ANTICIPATED BENEFITS

The main benefit of this project is it enhances communication regarding aquaculture activity within the region by functioning as a nucleus for information exchange between the aquaculture industry and ongoing research programs. This in turn, will aid in the technological advancement of aquaculture.

By disseminating research results and other information related to commercial aquaculture production, the project also helps to overcome the limited information available in the region.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Since 1989, the Center has developed and published a newsletter four times a year, which is distributed to approximately 1,000 individuals, organizations and universities worldwide. The Center has also created *Project Updates*, technical bulletins that are distributed to the CTSA Board of Directors, Industry Advisory Council, Technical Committee, and to extension agents and other interested parties upon request. Additionally, the Publications project has produced two videos which provided the latest results from the Center-funded projects at the time.

By 1999, the staff at the Center had produced (entirely or cooperatively) and published 38 progress reports, manuals, or fact sheets; duplicated and distributed 16 videos produced by the other Regional Aquaculture Centers and distributed them to extension agents, libraries and aquaculturists throughout the region; assisted in development and maintained the CTSA webpage.

Following are the accomplishments for this year:

Objective 1

As planned, a new design was implemented for the quarterly newsletter and was published in November 1999, and April, July and October 2000, and disseminated thereafter.

Objective 2

Three publications were produced and published: *Effects of three maturation diets on spawning of the armored catfish (Corydoras aeneus)*, *What is the Best Feed for My Ornamental Fish?*, and *Hydrogen peroxide treatment for Amyloodinium sp. on mullet (Mugil cephalus) fry*.

Objective 3

The homepage was updated monthly and maintained as needed.

Objective 4

Two of CTSA's own publications along with the publications from the North Central Regional Aquaculture Center and the Southern Regional Aquaculture Center were distributed to the Board of Directors and other information networks.

Objective 5

The Year 2000 Accomplishment report was completed.

Objective 6

The Center assisted with the creation, production and distribution of four publications: the *Collecting Black-lip Pearl Oyster Spat* information sheet, the *Nursery and Growout Techniques for Giant Clams* (*Bivalvia: Tridacnidae)* manual, the *Production of Lemon Tetra Hyphessobrycon pulchripinnis* publication, and the *Producing pearls using the black-lip oyster* info sheet. Two other manuals staff members have edited should be published early next year.

IMPACT

This project has helped to disseminate aquaculture results and information throughout the region to enhance viable and profitable U.S. aquaculture production which will benefit consumers, producers, service industries and the American economy.

WORK PLANNED

The staff will continue to produce and publish the quarterly newsletter, maintain and upgrade the webpage, disseminate information, and provide assistance with publications as needed. Three manuals that staff members have provided editing and layout assistance for are due to be published in the next year: Black Pearl Farming, Black Pearl Oyster Grafting and the Commercial Production of Swordtails.

AQUACULTURE EXTENSION AND TRAINING SUPPORT IN THE U.S. AFFILIATED PACIFIC ISLANDS – Year 11

Funding Level

Year 1	$100,000
Year 2	85,870
Year 3	83,600
Year 4	70,000
Year 5	75,000
Year 6	98,000
Year 7	70,000
Year 8	75,000
Year 9	85,000
Year 10	85,000
Year 11	75,000
Total	$902,470

Current Participants

Eileen Ellis……..……..Marine and Environmental Research Institute of Pohnpei

Greg Muckenhaupt……Marine and Environmental Research Institute of Pohnpei

Principal Investigator

Simon Ellis
Land Grant Program
College of Micronesia
Kolonia, Pohnpei, Federated States of Micronesia

PROJECT OBJECTIVES

The goal of this project is to facilitate the development of a sustainable, economically viable aquaculture industry in the U.S. Affiliated Pacific Islands.

In order to attain this goal, the following objectives must be met:

1. Provide extension support and technical assistance to commercial producers, government aquaculture staff, and other interested parties to develop aquaculture within the U.S. Affiliated Pacific Islands. Priority will be given to private producers and government institutions targeting private sector development.
2. To strengthen institution and build capacity, the Regional Aquaculture Extension Agent will:

a) conduct on-site training and structured courses and workshops on the culture techniques for giant clams, sponges, black lipped pearl oysters, ornamentals and finfish.

b) hire and place an intern in the region during the summer months in an effort to improve regionally based aquaculture.

c) focus training efforts on local extension agents in government and private organizations.

d) continue to work with other members of PRAEP to establish long-term aquaculture development plans for island entities in the region.

3. To aid in the transfer of technology, the Regional Aquaculture Extension Agent will continue to produce a comprehensive extension series in multimedia format. A simplified manual will be completed on optimum farming practices of eight species of hard and soft coral.

4. The Regional Aquaculture Extension Agent will:

a) continue to collect and disseminate upon request aquaculture information in the form of papers, manuals and videos to interested parties in the private and public sector throughout the region.

b) expand and update the literature collection at the Regional Aquaculture Extension Agent's home office.

5. The Regional Aquaculture Extension Agent will be PI on a CTSA funded applied research project titled "Determination of optimum farming practices for 8 species of commercially valuable soft and hard coral".

ANTICIPATED BENEFITS

- Workshops, publications and information dissemination will lead to an increased perception of aquaculture and its potential benefit to the region, particularly in species identified as being regionally appropriate.
- Currently operating aquaculture facilities will benefit from the constant presence of an extension agent who can advise on problems, provide information on aquaculture topics and make site visits.
- Training and regular site visits will help to build capacity and skills among people interested or involved in aquaculture.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

This project began in 1989, when the Center for Tropical and Subtropical Aquaculture funded an aquaculture extension specialist for the region. The extension specialist provided technical advice and assistance to establish the FSM National Aquaculture Center in Kosrae and established a demonstration ocean growout farm on the reef outside the National

Aquaculture Center. A giant clam demonstration farm was established in Pohnpei, FSM. Three sponge demonstration farms were established in various states of the FSM. The agent provided information on aquaculture of various species in response to requests from parties throughout the region. Subscriptions to a relevant magazine were provided to operators of giant clam facilities. Slide presentations and accompanying written materials on general aquaculture were developed. Literature and advice on aquaculture of various species were provided to private and government agencies in the CTSA region. All locations were provided with information on the documentation required under the Convention on International Trade in Endangered Species (CITES) and by U.S. Fish and Wildlife Service to allow the export of giant clam products. Regional aquaculture businesses were assisted with developing markets for their products. Since 1997, the extension specialist has conducted 65 workshops with close to 500 participants.

Following are the accomplishments for this year:

Objective 1

The Nukuoro pearl project continues to be a priority. It is widely viewed as having huge potential but is struggling to attain profitability mainly due to funding shortages and the lack of a full-time manager. This year, the extension agent provided the contact details for a seeding technician for the farm and acted as a liaison between Nukuoro and the technician. He also wrote the basis of the work contract between the technician and Nukuoro to ensure they received the proper results. His proposed visit to the farm has been postponed due to the cholera outbreak on Pohnpei.

While there is still a desperate need to overcome the spat bottleneck in the RMI, this work has been stalled now for a number of reasons. During his recent trip to the RMI in August, the Land Grant office at CMI still has the intention of starting a small hatchery for training purposes. However, they are in the process of moving, and the move has been stalled by landowner disputes.

Black Pearls of Micronesia is a private company, which is developing proprietary technology, and is thus reluctant for him to visit any area of their facilities where he may learn any of their secret techniques. He respects their decision in this regard. He continues to meet with the local managers regularly to advise them if necessary and provide them any information they request.

The RRE pearl farm continues to expand slowly but is hampered by lack of spat.

Marine and Environmental Research Institute of Pohnpei (MERIP) continues to slowly build up their staff. They currently are predominantly reliant on volunteers who only remain for two years.

He has been communicating with the resident agent in Saipan regarding his intended visits later this year to continue extension and training in CNMI.

Aquaculture Cooperative of American Samoa did not receive the grant to start their backyard-farming project. Many of the prospective farmers continue to correspond with him although he has had few calls for technical assistance in recent months.

RESULTS AT A GLANCE . . .

- Printing of six new extension publications.
- Dissemination of over 200 pamphlets on aquaculture throughout the region
- Continued extension, training and capacity building in the region

Objective 2

The extension agent conducted seven workshops and several training courses for private and government agencies and provided

other extension services to improve public outreach and training programs in the past year.

In March, a high school senior intern from MERIP was hired for a 2-week work experience on a commercial clam farm in the RMI. Beginning in June, Amy Philip, a senior at UH Hilo, spent 10 weeks in his office. She received not only instruction on the office-based duties of an extension agent but also accompanied him on site visits and with his field-based research. With the expansion of aquaculture facilities in Pohnpei, it was possible for the intern to get a well-balanced exposure to all facets of aquaculture development.

Objective 3

Work on the simplified manual scheduled to be completed during the 2000/2001 project year, has not yet started because research is still being conducted. With his assistance the summer intern, however, did a basic economic analysis of coral farming which will be included in the finished manual. His office did produce an unplanned Pohnpei translation of the sponge manual. Created earlier in the year were the manual on the culture of soft corals, the manual on giant clam nursery and grow-out techniques, and the information sheets on pear oysters, collecting black-lip pearl oyster spat, lagoon farming of giant clams, and farming soft corals.

Objective 4

During the project year, approximately 200 pamphlets and 10 videos were disseminated on a variety of aquaculture topics. In addition, over 100 manuals were disseminated to a mailing list.

Objective 5

The species of coral to be used in the study were chosen and broodstock was collected. Methods for attachment were chosen and tested. Attachment rates were assessed and attached corals were measured 2 weeks after planting. Subsequent measurements have been taken at 4-week intervals. Broodstock regeneration is being monitored photographically and water quality is measured weekly. Measurements and monitoring will continue for 6 months.

The lights and equipment to observe the survival and interaction of cultured corals in a home aquarium setting have been ordered and received. Sources of shipping bags have been researched and bags have been ordered for the shipping and marketing trials. Notes and digital photographs are being compiled for the simple extension publication but manuscript preparation has not been started yet.

IMPACT

At the start of the project in 1989 aquaculture activity in the region was virtually non-existent. Today there are six giant clam hatcheries, four pearl oyster farms, 13 sponge farms, four coral producers and an estimated 15-20 tilapia farms operating in the region. Awareness of aquaculture and its income producing potential is growing rapidly. The constant presence of an extension agent combined with hands-on training of local individuals has led to substantial capacity building in the region. Native islanders are now operating most of the giant clam farms in the region. As the industry grows, this knowledge will spread to other species with the ultimate goal of a self-sustaining, economically viable aquaculture industry in the region.

WORK PLANNED

The extension agent will:

- continue to focus on MERIP until they have a fully trained Micronesian staff with the institutional knowledge to conduct aquaculture training themselves.
- travel to Saipan to provide training and extension support to NMC staff and will also work on stimulating interest in marine-based farming in the area.
- assist COM Land Grant with the installation of a small pearl hatchery at Nett Point in Pohnpei.
- hold a 2-day workshop on Rota and Saipan on culture techniques for commonly cultured marine species from the region. Attendees will be private farmers, government personnel and other interested parties.
- continue to provide information dissemination, outreach and capacity building through his participation in two meetings later this year where he will be presenting talks.

for the applied research project:

- attachment rates and broodstock regeneration measurements and monitoring will continue for 6 months.
- Aquaria will be setup and the trial will begin within the next 6 months for the survival and interaction of cultured corals in a home aquarium setting.
- prepare the manuscript for the simple extension publication.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Publications

Crawford, Christine. 1990. *Giant Clam Mariculture Information Sheet Number 1: Giant Clam Activities in the South Pacific*. College of Micronesia. Kosrae, Federated States of Micronesia.

Crawford, Christine. 1991. *Giant Clam Mariculture Information Sheet Number 2: CITES Requirements for the Export of Giant Clams*. College of Micronesia. Kosrae, Federated States of Micronesia.

Crawford, Christine. 1991. *Giant Clam Mariculture Information Sheet Number 3: U.S. Food and Drug Authority Ruling on Giant Clams*. College of Micronesia. Kosrae, Federated States of Micronesia.

Crawford, Christine. 1992. *A Review of U.S. Food and Drug Administration Requirements to Market Giant Clam Meat in the United States of America*. CTSA Publication #109. Waimanalo, Hawaii.

Ellis, S. C. 1998. *Spawning and Early Larval Rearing of Giant Clams (Bivalvia: Tridacnidae)*. Center for Tropical and Subtropical Aquaculture Publication Number 130. Waimanalo, Hawaii.

Ellis, S. C., 1999. Aquafarmer Information Sheet: *Lagoon Farming of Giant Clams (Bivalvia: Tradacnidae)*. Center for Tropical and Subtropical Aquaculture Publication Number 139. Waimanalo, Hawaii.

Ellis, S. C., 1999. Aquafarmer Information Sheet: Farming Soft Corals for the Marine Aquarium Trade. Center for Tropical and Subtropical Aquaculture Publication Number 140. Waimanalo, Hawaii.

Ellis, S. C. 1999. Lagoon farming of giant clams (Bivalvia: Tridacnidae). Center for Tropical and Sub-tropical Aquaculture, Publication #139. Waimanalo, Hawaii, USA. 6 pp.

Ellis, S. C. 1999. Farming soft corals for the marine aquarium trade. Center for Tropical and Sub-tropical Aquaculture, Publication #140. Waimanalo, Hawaii, USA. 6 pp.

Ellis, S. C. 2000. Nursery and grow-out techniques for giant clams (Bivalvia: Tridacnidae). Center for Tropical and Subtropical Aquaculture, Publication Number 140. Waimanalo, Hawaii, USA. 103 pp.

Ellis, S. C. and L. Sharron. 1999. The culture of soft corals (Order: Alcyonacea) for the marine aquarium trade. Center for Tropical and Subtropical Aquaculture, Publication # 137. Waimanalo, Hawaii, USA. 73 pp.

Ellis, S. C. and L. Sharron. 1999. Manual: *The culture of soft corals (Order: Alcyonacea) for the Marine Aquarium Trade*. Center for Tropical and Subtropical Aquaculture Publication Number 137. Waimanalo, Hawaii.

Ellis, S. C. and M. Haws. 1999. Producing pearls using the Black-lip pearl oyster (*Pinctada margaritifera*). Center for Tropical and Sub-tropical Aquaculture, Publication #141. Waimanalo, Hawaii, USA. 8 pp.

Haws, M. and S. C. Ellis. 2000. Aquafarmer Information Sheet: Collecting Black-lip pearl oyster spat. Center for Tropical and Subtropical Aquaculture, Publication Number 144. Waimanalo, Hawaii, USA. 8 pp.

Haws, Maria. 1996. *Gems from the Sea: A Pearl Oyster Culture Manual*. Center for Tropical and Subtropical Aquaculture Publication Number 127. Waimanalo, Hawaii.

Heslinga, Gerald. 1996. *Clams to Cash: How to Make and Sell Giant Clam Shell Products*. Center for Tropical and Subtropical Aquaculture Publication Number 125. Waimanalo, Hawaii.

Lindsay, S. R 1991. Survival and Growth of Introduced Populations of the Giant Clam, *T. derasa*, on the Island of Yap Proper and the Outer Atoll of Woleai, FSM. Yap Marine Resources Division. Yap, FSM.

Conference Proceedings

Lindsay, S. R. 1995. Future trend of aquaculture – sponge culture. *In:* Friend, K. (ed.) Present and future of aquaculture research and development in the Pacific Island countries. Proceedings of the International Workshop, Ministry of Fisheries, Tonga. November 20 - 24, 1995. 423 pp.

Videos

Ellis, S. C. and G. Samson. 1998. *Spawning and Early Larval Rearing of Giant Clams*. Center for Tropical and Subtropical Aquaculture Video Number V003. Waimanalo, Hawaii.

Ellis, S. C. and G. Samson. (Producers) 1998. Video production: *Farming soft corals for the marine aquarium trade*. Center for Tropical and Subtropical Aquaculture video #137. Waimanalo, Hawaii, USA.

Heslinga, Gerald. 1996. *Clams to Cash: How to Make and Sell Giant Clam Shell Products*. Center for Tropical and Subtropical Aquaculture Video Number V002. Waimanalo, Hawaii

DISEASE MANAGEMENT & VIROLOGY SERVICES FOR HAWAIIAN AQUACULTURE – Year 7

Funding Level

Year 1	$41,638
Year 2	63,725
Year 3	45,956
Year 4	44,030
Year 5	66,451
Year 6	51,934
Year 7	81,991
Total	$395,725

Current Participants

Dr. Robert Bullis	The Oceanic Institute
Dr. Brad LeaMaster	University of Hawaii at Manoa
Dr. Yuanan Lu	University of Hawaii at Manoa
Mrs. Diana Montgomery-Brock	Aquaculture Development Program
Dr. Clyde Tamaru	Sea Grant Extension Program

Principal Investigator

Dr. James Brock
Hawaii Aquaculture Development Program
Hawaii State Department of Land and Natural Resources
Honolulu, Hawaii

PROJECT OBJECTIVES

1. To test tilapia from different locations for resistance to infection and disease from the piscirickettsia-like organism (PLO).
2. To determine the efficacy of hydrogen peroxide as a treatment for the control of columnaris disease.
3. To assist the high health shrimp producers in Hawaii acquire additional family lines of high health shrimp.
4. To establish a continuous cell line from each of the following fishes: the gray mullet (*Mugil cephalus)*, threadfin *(Polydactylus sexfilis)*, the green swordtail *(Xiphophorus helleri)* and the angelfish *(Pterophyllum scalare)*.
5. To provide culture diagnostic services for the isolation of fish viruses from 10 case submissions of marine or freshwater fishes.

6. To provide diagnostic support for the aquaculture sector in Hawaii and the Pacific Islands.

7. To communicate to the industry the findings from the studies conducted in this project.

ANTICIPATED BENEFITS

The correct identification of the direct and contributing cause(s) for diseases and impaired animal performance provides the basis for the implementation of effective corrective action. This project addresses field health management and disease issues hindering the aquaculture sector in Hawaii and the Pacific Islands. By identifying the direct and contributing cause(s) for diseases and impaired animal performance, an anticipated benefit is the improvement in production of Hawaii and Pacific Island aquaculture facilities or procurement of new information. This will eventually lead to improved health management strategies for improved aquaculture productivity in Hawaii and the Pacific Islands. Additionally, the services available through the project benefit aquaculture producers and researchers by helping people identify the causes for and reduce animal losses due to disease and unthrifty environmental conditions.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Since its inception in 1992, the project has substantial results from its activities, including:

- Identified the principal infectious diseases of Chinese catfish.
- Demonstrated that hydrogen peroxide is an effective therapeutant for the treatment of *Saprolegnia* sp. infections of eggs and *Trichodina* sp. and *Gyrodactylus* sp. infections of juveniles to adult Chinese catfish.
- Demonstrated that vaccination with an autogenous bacterin improves survival of Chinese catfish challenged with a fatal dose of *Aeromonas hydrophila*.
- Identified the principal biotic agents (various protozoan and metazoan ectoparasites) associated with mortalities of finfish cultured in a Hawaiian fishpond.
- Discovered in 1994 that an infection virus caused Taura syndrome, the most economically important disease of farmed *Penaeus vannamei* in the Western Hemisphere.
- Discovered that *Penaues stylirostris* is largely resistant to fatal disease from the Taura syndrome virus (TSV) and demonstrated the successful use of this species to mitigate the economic impact from Taura syndrome.
- Demonstrated that solar radiation heats the top centimeters of surface soil to > 60ºC, high enough to inactivate the known shrimp viruses.
- Demonstrated that the black crown night heron could be a transient transfer host for IHHNV.
- Identified the principal types of infectious agents found in imported groups of ornamental fish in Hawaii and implemented approaches to prevent the establishment of these agents on

commercial ornamental fish farms in the state.

- Conducted 16 half-day workshops on three islands on the diagnosis and control of diseases of ornamental fish.
- Discovered a new rickettsial agent was the cause for an emerging disease characterized by acute, high mortality in culture tilapia on Oahu, Hawaii.
- Demonstrated that feed medicated with oxytetracycline is effective to control mortality in tilapia exposed to the tilapia RLO.
- Facilitated the discovery by a commercial ogo farm of an effective means for the control of *Gracilaria* gall syndrome on that farm.
- Demonstrated the usefulness of ultrasound for sex identification of mature snakehead (*Channa striatus*).
- Demonstrated that hydrogen peroxide is an effective therapeutant for the control of *Amyloodinium* sp. infection on the Pacific threadfin, *Polydactylus sexfilis*.

Following are the accomplishments for this year:

Objective 1

The findings from the trial support the conclusion that prolonged exposure to low temperature induced a significantly higher mortality in juvenile tilapia subclinically infected with the PLO. These results support the conclusion that the sensitivity of testing tilapia for PLO would be enhanced if the fish were held for a period of time under chilled water conditions before being examined for PLO.

Objective 2

It was observed that the greatest effect on the Columnaris bacteria from hydrogen peroxide was with the highest dosage level (250 ppm). Therefore, the observations from the trial suggest that outbreaks of columnaris disease in juvenile catfish may respond best if the water temperature of the rearing unit is increased above 25 degrees and the fish receive treatment with hydrogen peroxide for 30 minutes at a dosage of 250 ppm.

At the hydrogen peroxide dosages tested, findings suggest that hydrogen peroxide is not an effective chemical for the treatment of epibiotic fouling in larval and early post larval *Penaeus (Litopenaeus) vannamei.*

Objective 3

Pathogen testing services were carried-out on shrimp sampled from eight facilities in Hawaii. Seventy-eight samples representing 606 juvenile to adult shrimp and 80 samples of post larval shrimp were assayed for IHHNV by the dot blot gene probe method. In addition, 238 shrimp tissue samples were submitted to the Aquaculture Pathology Group, the University of Arizona for PCR assay.

Objective 4

Cultivation of mullet (Mugil cephalus) cells in vitro – Two trials were performed. There was a loss of cells in the first passage, the cause of which is unclear. More culture trials will need to be run to test different growth media and fetal bovine serum for their support of the primary growth of mullet cells. Also, trials should be conducted to assess the effect of lower incubation temperature on the growth and survival of primary mullet cell cultures.

Cultivation of green swordtail (Xiphophorus helleri) and the angelfish (Pterophyllum scalare) cells in vitro – Among the six tissues selected for culture, snout had the fastest growth. However, cells also grew from the explants of the spleen, fins and kidney from both species. Cells did not grow from the liver explants. By the second day, contamination was apparent in the intestine cultures indicating an expected difficulty in the removal of endogenous contaminants (bacteria) from the intestinal tissues.

The combined use of M199 and L-15 resulted in the best growth of the freshwater fish cells. Cell proliferation appeared at day 2 for fin cultures of swordtail and formed a 25-50% cell monolayer by day 3 in this species. However, fin tissue cells began to die in both the primary culture and after the first passage. Only a small portion of cells remained attached at day 7 following the first passage. Nevertheless the fin-derived cells have continued to grow from swordtail and the cells are in their first passage.

Growing cells derived from snout for both species looked very good. These snout cells began to show up at day 3 and formed quite sizable cell colonies in about a week. Most cells are epithelial-like, while some fibroblastic-like cells are still present in the flasks. As of the date of this report, the snout-derived cells were growing well but have not been passed.

The spleen cells from swordtail and fin derived cells from angelfish displayed slow growth. No growing cells were seen from seeded tissue explants till day 6 and cell colonies remained vestigial and small. To date, these cells are still in the primary culture flasks.

The head soft tissue from angelfish had good cell growth. New cells were observed from the explants at day 3 and formed sizable cell colonies by day 8 of the incubation at 25°C. It appears that head soft tissue is a good tissue source for cell culture from the angelfish.

Objective 5

Three cases of diseased fish suspected or known to be infected by a pathogenic fish virus were evaluated by cell culture method in the period covered by this report.

Case A: Epidemic mortality of juvenile lionhead goldfish

The test was terminated after the third blind passage was completed. The negative results suggested that: a) The sample contained no infectious virus(es) or b) The sample contained infectious virus(es) that did not propagate in the test cell lines because the three cell lines were not susceptible to the virus.

As an additional approach, DNA was extracted from several of the pooled goldfish viscera samples. Amplification of the DNA using PCR primers derived from iridoviruses resulted in a specific gene fragment. Sequence analysis indicated that this amplified gene product showed a low sequence similarity to several reported iridoviruses. Characterization of this virus by obtaining more genome sequences is being attempted.

In addition, the establishment of a cell line from goldfish would be an aid in this process and work was initiated to accomplish this goal.

Case B: A survey of pathogenic viruses on juvenile mullet with dropsy (ascites)

Viral specific CPE was not observed during the initial infection in either of the two cell lines. The infected cells were subcultured three times and no CPE or cellular morphological abnormalities occurred during the three blind passages.

Although the histopathology examination performed on similar appearing mullet collected from the population did not indicate the presence of histopathology changes suggestive of a viral infection, the cell culture approach was undertaken. The negative cell culture findings support the histological conclusions that an infectious virus was not the cause for the ascites condition in the juvenile mullet. However, the application of cell lines derived from mullet that, after three blind passages did not display CPE, would have increased confidence in the conclusion that a virus infection was not the cause for the ascites syndrome in the juvenile mullet.

Case C: A survey of moribound rainbow trout juveniles for IPN and IHN viruses from an imported population with an unexplained mortality

Cytopathic effects (CPE) were not observed in the two cell lines inoculated with tissue

extracts from the fish. These results confirmed that IPN and IHN viruses were not present in the fish in the sample. The cause of the mortality in the juvenile trout was attributed to bacterial infection based on other laboratory findings and a positive response (cessation of mortality) after the fish were started on antibiotic medicated feed.

Objective 6

260 trips were made to the field to assist aquaculture operations. There were 658 case submissions of aquatic animals received for diagnostic laboratory analysis services. Ninety-five individuals or organizations requested assistance from the project.

5546 histology blocks and slides were prepared and examined from the 658 case submissions. 127 samples were submitted for culture, isolation and identification of bacteria. Tissue specimens from eight of the case submissions were evaluated by transmission electron microscopy for submicroscopic microorganisms. Viral pathogens were presumptively identified in the electron microscopy preparations in five of the eight submissions.

Surveillance for shrimp pathogens was carried out on shrimp tissue samples collected from seven companies or facilities in Hawaii. The following shrimp/prawn species were evaluated: *Penaeus vannamei, Penaeus stylirostris, Penaeus japonicus* and *Macrobrachium rosenbergii*. The tissue samples were evaluated for pathogens by PCR method and/or histopathology. For PCR assay the tissue specimens were submitted to the University of Arizona, Tucson, Arizona.

Hemolymph or pleopod tissues from 284 shrimp were assayed by PCR method for WSSV; hemolymph or pleopod tissues from 229 shrimp for IHHNV; lymphoid organ tissue from 80 shrimp for TSV and YHV; and hepatopancreas tissue from 20 shrimp for HPV. Also, 241 shrimp tissue samples were evaluated by histology methods for shrimp pathogens.

The viruses WSSV, TSV, YHV and HPV were not detected in any of the tissue samples from the four species assayed by PCR or histology methods from the seven facilities. From one location, five of the 49 samples (10%) of post larvae were positive for IHHNV and three (6%) of the parallel histology samples from that location had a few of the PIs that were positive histologically (CAIs) for low grade IHHNV infection, as well.

Objective 7

A 2-day Fish Health Management Workshop was held on June 24 and July 1, 2000 in Hilo. There were 18 participants. The workshop was well received. Two articles were submitted that reported results from work conducted in the Disease Management Project.

IMPACT

Staff from the companies that received the assistance made estimates of the economic impact based on the known or expected gain, or the reduction in the anticipated loss, once the problem was brought under control. In

ESTIMATED ECONOMIC IMPACT:

Gill fouling of tank-cultured shrimp – $500,000

Control of *Amyloodinium* disease -- $75,000

Control of an outbreak – $185,000
of *Cryptocaryon sp*

Control of a fish mortality – $4900
in a shipping transfer

Control of bacterial contamination – $828,000
of live brine shrimp fed to marine
larval shrimp

each case our recommendations for control were the result of our evaluation of the conducted in the field and our interpretation of the laboratory results. In the examples, the farm personnel implemented the strategies that were recommended. To a great extent the success of a given strategy reflects their efforts. The examples demonstrate the problem in the field, trials or studies

positive outcome that has occurred from the effective relationship that the Disease Management Project enjoys with the aquaculture community in Hawaii and the Pacific Islands.

WORK PLANNED

We plan to continue work to establish continuous cell lines from the snout and fin cell cultures started from the swordtail and the angelfish. Also, cell culture trials will be conducted with mullet and threadfin during the next reporting period.

Work on Objectives 5 and 6 will be continued.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Publications

Brock, J. A. 1995. Update on Recent Research Findings on Taura Syndrome of Farmed *Penaeus vannamei. In*: Proceedings of the III Simposio Centroamericano Sobre Camaron Cultivado. April 3-5, 1995. Tegucigalpa, Honduras.

Brock, J. A., D. V. Lightner, K. Hasson and R. Gose. 1996. An Update on Taura Syndrome of Farmed Shrimp in the Americas. Book of Abstracts, World Aquaculture Society '96. Bangkok, Thailand p. 50.

Brock, J. A., R. Gose, D. V. Lightner and K. Hasson. 1995. An Overview on Taura Syndrome, An Important Disease of Farmed *Penaeus vannamei.* Swimming Through Troubled Waters, Proceedings of the Special Session on Shrimp Farming. Aquaculture '95. Browdy, C. L. and J. S. Hopkins (eds.). pp. 84-94. World Aquaculture Society. Baton Rouge, Louisiana, U.S.A.

Brock, J., B. Poulos, R. Shimojo and R. Gose. 1999 Modification of the tissue preparation steps to reduce false positive reactions with the IHHNV dot blot gene probe. Proceedings Simposio V Centroamericano de Acuacultura, 18-20 August 1999, San Pedro Sula, Honduras. pp. 112-124.

Brock, J., B. Poulos, R. Shimojo and R. Gose. 1999. A report on the efforts to reduce false positive reactions with the IHHNV dot blot gene probe test. Proceedings of the V Congresso Ecuatoriano de Acuicultura, Enfocando Los Retos Del 2000, 28-30 October 1999, Guayaquil, Ecuador.

Klien, M. R. G. 1996. An Epizootiological Study of Chinese Catfish, *Clarias fuscus:* Mortalities on Two Farms in Hawaii. Master of Science Thesis in Animal Sciences. Submitted to the Graduate Division, University of Hawaii. 70 pp.

Montgomery, D., J. Brock, V. Sato, and B. LeaMaster. 1998. The Use of Hydrogen Peroxide to Treat Amyloodinium Ocellatum on Mobi (Polydactylus sexfilis). Vol. 20, No. 11, November 1998.

Montgomery, D., J. A. Brock and V.T. Sato.1998. Aquatips - Using hydrogen peroxide for Pacific Threadfin infected by Amyloodinium ocellatum. Center for Tropical and Subtropical Aquaculture - Regional Notes. 1998, 10:2, pp. 4-5.

Montgomery, D., J. Brock. 1999. Aquatips: Selecting products for water chemistry testing for your aquaculture operation. CTSA Regional Notes, Vol 10 (4), pp. 4-6.

Montgomery, D., J. Brock, C. S. Tamaru. 1999. Aquatips: Parasites in paradise: Ich. CTSA Regional Notes, Vol 11 (1), pp. 4-6.

Montgomery, D., R. Weidenbach, E. Weidenbach, B. LeaMaster, C. Tamaru, and C. Carlstrom. 1998. The Use of Ultrasound Technology to Determine Gender of Snakehead Fish (*Channa striatus*). Vol. 20, No. 9, September 1998.

Montgomery-Brock, D.R., J.Y. Sylvester, C.S. Tamaru, and J.A. Brock. 2000. Hydrogen peroxide treatment for *Amyloodinium* sp. on mullet (*Mugil cephalus*) fry. Aqua Tips, Regional Notes, Center for Tropical and Subtropical Aquaculture. Vol 11(4) pp. 4-6.

Montgomery-Brock, D.R., V. T. Sato, J.A.Brock, and C.S. Tamaru. 2000. The application of hydrogen peroxide as a treatment for the ectoparasite *Amyloodinium ocellatum* (Brown, 1931) on the Pacific threadfin, *Polydactylus sexfilis*. Journal of the World Aquaculture Society. Submitted.

Tamaru, C. S., F. Lum and J. Brock. 1998. Monitoring Water Temperature with Miniature Data Loggers. Vol. 20, No 7, July 1998.

Papers Presented

Brock, J. A. 1997. Taura syndrome: history, distribution and current status. Presented at the Special Session on Shrimp Virus Disease. World Aquaculture Society Meeting 1997. Seattle, Washington.

Brock, J.A. 1999. Approaches to some health issues for farmed marine ornamental fishes, Marine Ornamental Conference, Novermber 17-17, 1999, Kailua-Kona, Hawaii.

Brock, J.A. and R. Bullis. 2000. Disease prevention and control for gametes and embryos of fish and marine shrimp. Aquaculture Interchange Program Workshop, October 4-7, 1999, Honolulu, Hawaii.

Montgomery, D. R. and J. A. Brock. 1997. Preliminary trials on the application of hydrogen peroxide for treatment of the ectoparasites *Gyrodactylus* sp. And *Trichodina* sp. On the Chinese catfish (*Clarias fuscus*) and *Amlyoodinium* sp. on the Pacific threadfin (*Polydactylus sexfilis*). Presented at the World Aquaculture Society Meeting 1997. Seattle, Washington.

DEVELOPMENT OF PACIFIC THREADFIN AND MILKFISH GROWOUT TECHNOLOGY AND PRODUCTION OF LIVE FEEDS AND SEEDSTOCK – Termination Report

This project was terminated because all objectives were met.

Funding Level

Year 1	$112,000
Year 2	109,934
Year 3	110,000
Total	$331,934

Participants

Clyde Tamaru, Ph.D.	Sea Grant Extension Service
James Szyper, Ph.D.	Hawaii Institute of Marine Biology
Kevin Hopkins, Ph.D.	University of Hawaii at Hilo
PingSun Leung, Ph.D.	University of Hawaii at Manoa
Dean Toda	State of Hawaii

Principal Investigator
Anthony C. Ostrowski, Ph.D.
Finfish Program
The Oceanic Institute
Waimanalo, Hawaii

PROJECT OBJECTIVES

The ultimate goal is to support development of a sustainable aquaculture industry for Pacific threadfin and milkfish in Hawaii, which was accomplished by:

1. Providing Pacific threadfin and milkfish fingerlings to prime industry development.
2. Perform trials to determine optimum growout conditions and transfer that technology to farmers.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Objective 1

Over the past 3 years, this project has distributed 766,000 threadfin fingerlings and 487,500 milkfish fingerlings to close to 40 farms or research sites with the anticipated result being the development of a sustainable aquaculture industry for Pacific threadfin and milkfish in Hawaii. Before the inception of this project, threadfin production was non-existent in Hawaii. Three farms that received fingerlings are now successfully established and turning a profit from the threadfin they harvest. According to a survey conducted by the USDA, threadfin production has increased from 1,100 lbs in 1995 to 119,568 in 1999, an increase of more than 100 fold.

Besides providing the fingerlings, the project also offered technical support to the farmers in the form of on-site visits and phone or email communication. Because of the support, the survival rates of the fingerlings increased.

Additional market expansion has come from the promotion of threadfin to local and export markets by providing fish to media events and distribution outlets for evaluation. Because most consumers were unfamiliar with threadfin, the greatest obstacle was introducing it to the public. Participation in several media events proved to be successful and many high-end restaurants locally and on the mainland now have threadfin on their menu. Moreover, several surveys were conducted and response was always overwhelming that the taste of threadfin was exceptional.

Objective 2

Several growout trials were performed during the duration of this project, which allowed them to: refine threadfin growout techniques; develop alternative milkfish growout techniques; demonstrate the interaction between diet and pond environment on growout of milkfish; and determine that polyculture of shrimp and milkfish is beneficial to both because the overall biomass production from a pond is improved. These techniques were then transferred to the farmers in the form of site visits and/or telephone conversations or email.

Results from growout trials were able to prove that although individual shrimp and fish growth are adversely affected when pelleted feed is limiting, overall biomass production from a pond is improved when milkfish are raised with shrimp. Both milkfish and shrimp derive a large proportion of their nutritional needs from the pond environment, and there appears to be an additive, synergistic effect on exploiting the contribution from the pond when both species are raised together.

Feeding trials were also conducted and optimum feeds and formulations were determined for both milkfish and threadfin. This leads to a minimization of feed costs, which typically represent 50% of the production cost of any fish farming operation. It was determined as well that water use costs

RESULTS AT A GLANCE . . .

- Three threadfin farmers have been successfully established.
- Threadfin production in Hawaii has increased more than 100 fold since 1995.
- Upscale mainland restaurants have threadfin on their menus.
- Determined threadfin and milkfish feeding requirements for optimum growout.
- Distributed 766,000 threadfin fingerlings and 487,500 milkfish fingerlings.
- Farmers and distributors confirm increased sales of threadfin.
- There is a rising interest and market-demand for farm-raised fresh milkfish.

can be lowered with the identification of threadfin loading rate requirements.

Because costs of rearing 30-day-old fish from a hatchery are too high to yield acceptable profits, and there is no assurance that farmers will continue to be provided with seedstock, extensive or semi-extensive rearing methods needed to be developed for farmers to eliminate their reliance on an established hatchery. Green water trials were conducted enabling project members to find an alternative method for milkfish larval rearing, lowering the cost to raise a fry from $0.15/fry to $0.03/fry with no effect on on-farm survival. This was due to reduced labor and lower energy and feed costs by raising the fry in shrimp pond waters.

Results from an experiment designed to examine the effects of a high and low quality commercial feed in a pond water environment on growth and feed utilization, indicated that both feed type and the culture system had significant effects on biological performance of milkfish. There was an interaction between culture system and diet which indicated that the pond environment does contribute toward meeting the nutritional needs of milkfish, particularly when given low quality diets. It was concluded that while juvenile milkfish do obtain supplemental growth benefits from being raised in pond environments, farmers should employ use of high quality feeds to maximize growth.

Members of the project developed a spreadsheet for distribution to farmers. The spreadsheet was intended to be an easy to use enterprise budget for application on farm sites. The program was designed to take detailed production and financial inputs from the farmers for the first three years of operation and assumes that production will stabilize from the third year on for 20 years. The program has also been applied to evaluate the economics of threadfin production using the production parameters from the OI experimental production trials. It is now ready for distribution.

IMPACT

The most notable impact is the creation of an industry for threadfin. Previous to this project, the market for threadfin was non-existent. Through their distribution of fingerlings, three threadfin farmers have been successfully established in Hawaii. By providing fish to media events and distribution outlets for evaluation and exposure to local and export markets, the market was expanded and threadfin farmers and distributors have confirmed increased sales through several channels. Threadfin is now being served in upscale mainland restaurants throughout the country and is gaining national recognition.

The state's production of both threadfin and milkfish has dramatically increased since 1995. According to the USDA Hawaii Agricultural Statistics Service, threadfin production in Hawaii went from a reported 1,100 lbs in 1995 to 119,568 lbs in 1999, adding more than 80 fold to the value of the threadfin market in 1999. There is also a rising interest and market-demand for farm-raised milkfish in the state.

During the 3-year duration of the project $332,000 has been expended, this had led to over $1 million in revenue during its duration, a return of three times the dividend.

This project determined the socioeconomic and market characteristics of harvested threadfin through sales of fish to distributors and other outlets.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Papers

Kam, Lotus E., PingSun Leung, Anthony C. Ostrowski and Augustin Molnar. In press. Economics of a moi hatchery in Hawaii.

Martinez-Cordero, J. Francisco, PingSun Leung, Anthony C. Ostrowski and Michael D. Chambers. Profitability analysis of the commercial growout of Pacific threadfin *(Polydactylus sexfilis)* in Hawaii under different production systems. Accepted to Journals of Aquaculture in the Tropics.

Ostrowski, A. C. and A. Molnar. 1998 Pacific threadfin *Polydactylus sexfilis* (moi) hatchery manual. Center for Tropical and Subtropical Aquaculture. Publication Number 132. 96 pp.

MARINE FOOD FISH SEEDSTOCK PRODUCTION – Year 2

Funding Level
Year 1 ……..$110,000
Year 2 ……135,000
Total …….$245,000

Current Participants
Brad J. Argue, Ph.D. …….The Oceanic Institute
James A. Brock, Ph.D. ……..State of Hawaii
Christopher Kelley, Ph.D. …….University of Hawaii
Charles W. Laidley, Ph.D. …….The Oceanic Institute
PingSun Leung, Ph.D. ……...University of Hawaii
Robin J. Shields, Ph.D. ……..The Oceanic Institute

Principal Investigator
Anthony C. Ostrowski, Ph.D.
Finfish Program
The Oceanic Institute
Waimanalo, Hawaii

PROJECT OBJECTIVES

The ultimate goal of this project is to support the development of a self-sustaining marine food fish industry in Hawaii and transferring production capabilities to the commercial sector. Specific objectives to accomplish this goal are:

1. Maintain Pacific threadfin, milkfish and crimson snapper broodstock.
2. Produce 250,000 threadfin and 100,000 milkfish fingerlings for distribution to qualified farmers.
3. Refine disease-free certification program for finfish larvae and improve fry quality.
4. Begin to domesticate Pacific threadfin for aquaculture and produce a selected line for increased growth.
5. Determine cost structure and profitability of milkfish growout in Hawaii.
6. Continue phased fry payment schedule and expand program to other areas of the Pacific taking into account site-specific, economic, and genetic considerations.
7. Evaluate other bottomfish species as potential aquaculture candidates and develop broodstock capabilities as money and fish become available.

ANTICIPATED BENEFITS

- Phased fry payment program is anticipated to provide the incentive for farmers to begin to explore their own hatchery capabilities while not over burdening them with excessive costs for fingerlings or severing the supply of fingerlings completely.
- Initiation of a selective breeding program for threadfin will ensure that local farmers will be on the cutting edge of improved threadfin production to remain competitive.
- A budding marine foodfish industry for both threadfin and milkfish will be firmly established, which will represent half of all the finfish production in the State of Hawaii.
- The health monitoring program will assure farmers a quality product as well as help maintain a healthy industry.
- Development of selected lines of threadfin for improved performance will ensure continued maturation and growth of the industry.
- On-farm survival rates are expected to increase.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Researchers from the Oceanic Institute (OI) have provided both threadfin and milkfish fry to farmers for growout operations, as well as technical advice and site visits to assist farmers in hatchery and growout efforts. Year One of the project was successful in fulfilling overall objectives, and surpassing fry distribution targets. A total 273,449 threadfin and 144,071 milkfish fry were distributed to 8 threadfin and 27 milkfish farmers on the islands of Oahu, Kauai, Molokai, and Hawaii. In addition, over 1 million threadfin eggs were supplied to three farmers and two research institutions on Oahu and Hawaii to begin hatchery efforts. Threadfin and milkfish aquaculture in the state continues to grow. The impact of the project for Year One to the local farming community is estimated to be worth just under $750,000 in off-the-farm sales.

Following are the accomplishments for this year:

Objective 1

By the time this phase of the project was initiated, there were only four broodstock snapper remaining at OI. It was decided to cull these animals rather than risk any potential disease transference by combining them with established animals at HIMB. Broodstock snapper at HIMB continued to grow and develop well in the net-pens. Animals are currently being monitored and plans are to send eggs to the OI hatchery if and when they become available to attempt mass culture.

OI continues to maintain Pacific threadfin and milkfish broodstock for the project. Eggs are collected routinely and many have been distributed to local farmers to attempt mass culture. During the report period, the OI hatchery supplied 1.7 million threadfin and 1.34 million milkfish eggs to three growout and research operations on the islands of Hawaii and Oahu.

Objective 2

Two threadfin production runs were conducted over the first 6 months of this project (April and July, 2000) following the quarterly schedule requested by farmers. A total 118,640 fish have been distributed to three farmers on the islands of Hawaii and Oahu.

Only 3,600 milkfish fry have been distributed. A drop in algal production at the OI facility that resulted in low rotifer output has hampered production efforts. Efforts have been directed to correct the situation, and additional runs have been conducted as of this writing. Milkfish production runs are expected through October, and until broodstock stop spawning to reach the targeted production goal.

Objective 3

Threadfin and milkfish fingerlings (30 from each species) from each production run at OI were killed with an overdose of MS-222, weighed, necropsied, and tissues from major organ systems (gill, heart, liver, spleen, caudal kidney, digestive tract, and attached abdominal viscera) were collected and preserved. The specimens were blocked and processed by routine histopathology methods and slides were prepared. The tissue sections were stained with hematoxin and eosin, and inspections were conducted.

RESULTS AT A GLANCE . . .

- Necropsy and histopathology findings showed no gross change in the fry of threadfin or milkfish.
- Several farmers this year have begun hatchery efforts in earnest.
- More than 20 site-visits were made on farms on Oahu, as well as 10 site-visits to outer island farms.
- Efforts to implement a selected breeding program for Pacific threadfin have been initiated with the development of a domesticated broodstock group.

The necropsy and histopathology findings showed no gross change in the fry threadfin or milkfish, which disputes any presence of an infectious disease problem in the batches of fish.

Objective 4

Efforts to implement a selected breeding program for Pacific threadfin have been initiated with the development of a domesticated broodstock group in which 10 females collected from the wild have been stocked with 10 F1 males. These animals reached reproductive condition in September with small spawns in September and October. It is anticipated that eggs derived from spawns in January will be used for selection purposes. Fifty randomly selected fish (controls) and the 50 largest fish (select line) will be tagged and then grown to sexual maturity to continue the selective breeding program.

Objective 5

Little progress has occurred with the determination of production cost structure and profitability of milkfish growout in Hawaii due to a delay in establishing a subcontract agreement with the University of Hawaii. The issue has been resolved and site visits to local milkfish farms are being arranged.

Objective 6

Seedstock provisions to farmers were logged for each shipment of the fry payment. To date, we have recovered only $1,484.80 of the $14,673.28 invoiced to farmers.

Site visits were conducted on the islands of Hawaii (Ben Krause) and Maui (Guy Ting) to assist in hatchery and growout set-up. Ben Krause of Pacific Harvest, Inc. is currently setting up a hatchery and conducting trial, larval rearing runs using eggs provided by OI. There were no requests for fry from Pacific island areas other than Hawaii.

Objective 7

There have been no spawns from any snapper species under any project.

IMPACT

Recent information provided by the State of Hawaii's Aquaculture Development program indicated a steady rise in sales of both threadfin and milkfish over the last several years. This is a direct result of the technology, fingerlings, and technical support supplied by this project. There has also been a steady rise in the demand for fingerlings to support expanded industry needs. This has prompted several private farmers to begin hatchery runs to not only supplement their own needs, but also to explore the potential to support industry needs. Pacific Harvest, Inc., a threadfin farm in Kona, has taken particular aim at establishing a threadfin hatchery. The company plans to be producing its own fry beginning September 2001. Project staff have assisted in the development of this hatchery.

The establishment of private hatcheries is consistent with the goal of supporting self-sufficiency of the industry.

Farmers are being better trained to raise threadfin larvae and on-farm survival rates are expected to increase. Completion of the hatchery economic model will help farmers estimate their required scale and costs of production, and provide them the basic equipment and supplies to develop their hatcheries. OI will attempt larval rearing while HIMB develops broodstock management techniques. The health assurance program will continue, but indications are that Hawaii-based farmers are receiving high-quality fry to provide them with the best chance of success in growout

WORK PLANNED

Over the next six months, substantial progress is targeted in the areas of milkfish economics and the threadfin selective breeding program. As indicated in this report, we have established an F1 generation *x* wild tank of broodstock that has initiated spawning over the last six months, and which will be used to establish our first group of fish for the selective breeding program.

The following is targeted:

- Spawning, larval rearing, and initiation of growout of the first F1 *x* wild cross of Pacific threadfin broodstock for the selective breeding program.
- Production and distribution of additional 130,000 threadfin and 96,000 milkfish fry.

- Continued assessment and documentation of fry health from the OI hatchery by the State of Hawaii's Aquatic veterinarian.
- Site visits to local milkfish farmers and generation of a synthetic enterprise budget for on-farm growout.
- Larval rearing of snapper fry if and when they become available.
- Continued distribution of threadfin and milkfish eggs to qualified farms, and technical assistance and site visits to local farmers.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Manuscripts

Kam, L.M., P.-S. Leung, A.C. Ostrowski, and A. Molnar. Economics of a moi hatchery in Hawaii. Draft. 31 pp.

Martinez-Cortero, F.J., P.-S. Leung, A.C. Ostrowski, and M.D. Chambers, Profitacility analysis of the commercial growout of moi (*Polydactylus sexfilis*) in Hawaii under different production systems. Draft. 23 pp.

MARINE ORNAMENTAL FISH CULTURE AND CONSERVATION – Termination Report

Based on the results presented, the IAC could not justify continuation of this project and chose instead to return the unspent money from this project to CTSA. Additionally, the Principal Investigator left his post to pursue his career outside of Hawaii.

Funding Level Year 1 ……...$49,200
Total.…….$49,200

Participants Dr. Jeri Fox…….……..Hawaii Institute of Marine Biology

Principal Investigator Dr. Christopher Brown
Hawaii Institute of Marine Biology
University of Hawaii
Honolulu, HI

PROJECT OBJECTIVES

1. Collect larvae from the wild, return them to the laboratory and rear them to market size.

2. Collect culture material from the Waikiki Aquarium.

3. Test the suitability of a green-water culture system primed with actively photosynthesizing monocellular algae and nutrient-enriched trocophores as a larviculture system.

4. Develop a prioritized list of marine ornamental species for aquaculture development.

5. Transfer the technology to industry.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Objective 1

The collection of eggs and larval and juvenile fish from Kaneohe Bay was attempted in two different ways. The first method was with nets made with small mesh sizes that were deployed from boats. The second method involved the use of land-based fish attracting devices referred to as FADs.

Two large nets were purchased, a seine net, custom made of knotless nylon (450' x 30' in size and 1/2 " mesh) and a neuston net, custom made with an aluminum frame (4' x 2' frame, 25' long with 1800 um mesh size). The nets were towed in different locations, at various times relative to the tides, and in a range of daylight as well as dark conditions. Tows were conducted in the area surrounding Coconut Island in Kaneohe Bay because if its proximity to populated reefs and to take advantage of the natural windbreak provided.

The size of the seine net proved it unsuitable because it takes four strong people to haul it out by hand. The hauling process is slow and allows a large number of larvae and juveniles to escape or be crushed in the folds of the net. In addition, the process requires two 17-foot Boston whalers, which are costly to rent. The wet net is extremely heavy and difficult to handle. Moving it onshore requires a forklift and a large space for hanging and drying the net. Tidal currents also affect the net. The neuston net was much more manageable and could be maneuvered by two to three people from one boat. The neuston net did prove to sample both larvae and eggs, but they were not those of the most sought after ornamental reef species. Also, Kaneohe Bay did not turn out to be the optimal place to employ this type of collection. The reefs in Kaneohe Bay even at high tide were too shallow to allow the net to be drawn directly over the reef without damaging the reef. The dynamics of the bay also seem to demonstrate that the recruitment of presettlement juveniles to the reef appears to be dependent on the incoming tide. Additionally, the dependence on one employee and volunteers as available, proved to impose limitations on the completion of this objective.

The land-based FADs consisted of an outdoor light mounted on a frame and positioned approximately 3 feet from shore. Three different areas were chosen. Many juvenile and larval pelagic schooling fish were collected in this manner as well as large quantities of invertebrates and copious numbers of copepods which were also used in later first feed experiments. None of the commercially important ornamental reef fish larvae were collected, although the ability of light attractants to accumulate fishes was viewed as an encouraging result. Other FAD designs were borrowed from previous research. One of the FADs consisted of rolled mesh and juvenile *Abedefduf abdominalis* quickly settled on it in large numbers. After a period of 3 weeks, these juveniles were collected from the FADs. Once the initial recruits were removed, the rate of recruitment dropped precipitously, to about one fish per week. Other fish were observed occasionally in the FADs, but the most consistent and easily caught were the sergeant majors. The duration of this experiment was three months. To get a larger and more varied recruitment, it may be necessary to encompass more seasons.

Some rearing trials were performed with *Abedufduf* from light-trapping and with frogfish that were collected. The former were not especially interesting because these fish had been reared before and the latter were unsuccessful.

Objective 2

With the cooperation of the Waikiki Aquarium, the collection was successful for either the larvae or eggs of several different species of fish highly prized by aquarium hobbyists.

Among these were yellow coral gobies, cardinalfish, sergeant majors, chromis, frogfish, as well as 12 spawns from the local Hawaiian seahorse, and 8 spawns from the masked angelfish. Egg collectors were specifically designed and constructed for individual exhibition tanks to maximize egg collection without interfering with the primary role of the aquarium, which is unobstructed viewing of the fish by the public. This was the product of a considerable effort, since it was an unacceptable risk to disrupt the public displays.

The most successful method used was observation and then siphoning.

Objective 3

Although green water was used in *Abedufduf* and frogrish culture, the plan of testing enriched trochophores remained out of reach for several reasons. Specifically, because a steady supply of larvae was never established from either wild collections or the aquarium.

Objective 4

A prioritized listing of possible candidates for culture based on feedback from all parts of the Hawaii Aquaculture community was created. The criteria for prioritization included likelihood of success, value, available technology, and availability of broodstock. Contributing factors included benthic eggs (more manageable than pelagic eggs) oral incubation, or the production of egg masses. The list is presented below in Table 1.

Objective 5

There was no attempt at the transfer of technology.

Table 1: Prioritized Species List

Species or Group	Florida Culture	Benthic Eggs	Hawaii Export	Expected Difficulty	Market
Yellow Tang	No	No	#1	High	Vast
Pygmy angelfishes	No	No	Yes	High	Solid
Sargassum fishes	No	*	Yes	Moderate/high	High Price
Hawkfishes (longnose, etc.)	No		Yes	High	Small but popular family, carnivorous
3-spot damselfish	No	Yes	Yes	Easy-moderate	Low price, high volume*
Misc. damselfishes	No	Yes	No	Moderate	Varies*
Neon goby	Yes	Yes	No	Moderate	Accepts tank-reared fish
Seahorses	? (Australia)	*	Few	Moderate	Solid*
True angelfishes	Few	No	Some	High	76 spp.
Butterflyfishes	No	No	Yes	High	Good, but requires some coral in diet
Surgeonfishes	No	No	Yes	High	Numerous spp.
Wrasses	No	No	Yes	High	Numerous spp.
Triggerfishes	No	Yes*	Yes	High	Good
Clownfishes Tomato Clarke's Pink Skunk Orange Maroon Wide-banded Cinnamon	Yes	Yes	No	Moderate	Established, competitive

Red saddleback					
Clownfishes Percula False Percula	Yes	Yes	No	Easy-moderate	High volume/low price
Bangai cardinalfish	Yes	*	No	Moderate	High Value
Neon dottyback	Yes	Yes	No	Moderate-high	Good
Striped dottyback	Yes	Yes	No	Moderate-high	Good

* indicates special considerations.

***Damselfishes** can be cultivated and constitute a significant segment of the marine ornamental market, but unrestricted imports of wild specimens undemine the profitability of captive rearing. It remains possible that restriction on such importations will be imposed, so the development of suitable technology for mass culture of the numerous species of damselfishes favored by the marine ornamental trade is strongly recommended. *The **seahorses** include a range of species (in this context, also pipefishes), at least one of which is endemic to Hawaii. Some are already being cultivated. Their nurturing of young contributes to the prospects of successful cultivation of a wider range of species, and continued technical development is considered advisable. A second asterisk also appears under "Market" for seahorses because additional markets exist besides the aquarium trade. Dried seahorses are a traditional dietary supplement in China – although supplying product to this market is almost certain to have unwanted image consequences for the business profiting from this sort of trade. *__Triggerfishes__ construct nests and consequently eggs can be collected efficiently; also they include numerous highly valued species, some of which are endemic to Hawaii. Cultivation appears to be a difficult task, however. *The **Bangai cardinalfish** is a mouthbreeding species, and can be bred in captivity with only moderate difficulty. The cohort size is small, however, so the cost of producing fry is relatively high.

IMPACT

The idea of collecting wild juveniles for growout has gained some acceptance.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Paper

Brown, C. L. and T. Ogawa. 1999. "History of Marine Ornamental Aquaculture in Hawaii", presented at the First International meeting on marine ornamental fish, Marine Ornamentals '99, Kona, Hawaii, November 1999.

TRANSITIONING HAWAII'S FRESHWATER ORNAMENTAL INDUSTRY – Year 1

Funding Level Year 1 ……..$100,000
Total ……....$100,000

Current Participants
Kathleen McGovern Hopkins..…..…..Sea Grant Extension Service
Matt Lyum…….. ……..Sea Grant Extension Service
Lena Asano……..……..College of Tropical Agriculture
Dr. Harry Ako……..……..College of Tropical Agriculture
Dr. Jim Szyper ……..Sea Grant Extension Service
Jennifer Olson……Windward Community College

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PROJECT OBJECTIVES

1. Conduct laboratory and field studies on sex determination in live bearers.
2. Demonstrate live feeds production systems to freshwater ornamental aquafarmers.
3. Investigate the role of Highly Unsaturated Fatty Acids (HUFAs) in the culture of freshwater ornamentals.
4. Provide technical assistance in the form of verbal consultations, written information, site visitations, conducting workshops and printing of manuals and production handouts.

ANTICIPATED BENEFITS

The morphology and coloration of the male swordtail is in large part its main attraction thus a preponderance of females (as the situation currently exists) ultimately result in lower profitability. Uncovering the parameters that influence the sex ratio of the swordtails will allow for establishing a best management practice that apparently is yet to be clearly defined for the swordtails.

It is anticipated that the results from the efforts being put on the identification of the sex determining characteristics in live bearers will have a profound effect on the overall productivity of Hawaii's freshwater ornamental industry as the largest component of the current inventory are the livebearers. Likewise, the hi-fin and lyretail traits are 2-3 times the value as that of their normal finned relatives and improving the production of these fin types will undoubtedly add to the overall efficiency and value of the crop as just the "culls" (normal fin) already fetch the same price of what is currently being produced.

The practice of rearing larvae presently involves use of commercially inert feeds which require a high amount of water exchange and are also relatively expensive. The dependence on the use of these feeds may be one reason that the production of egg layers has not caught on as expected. Ensuring that Hawaii's aquafarmers possess the culture technology for rotifers and other live feeds is essential for them to remain competitive as it will provide for a more cost effective operation as well as allowing for expansion and diversification of the industry by allowing for the mass culture of a broader range of fishes.

Bioremediation could increase potential farmer income at least 3-4 fold by allowing a farmer to conserve water. Likewise, recirculating systems allows a much broader range of potential opportunities for interested growers as the systems can be established with minimal amounts of water usage in a variety of locations.

By investigating the role of HUFAs in the culture of freshwater ornamentals, we can improve the quality of larvae and allow a more stable fry production in commercial hatcheries.

A relationship between the institutional partner, Sea Grant Extension Service and the private sector, guarantees that technologies are transferred in a timely manner and that modifications and other constraints can be identified.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Following are the accomplishments for this year:

Objective 1

A survey was taken of sex ratios of several varieties of swordtails and it demonstrated that sex ratio is clearly linked with the variety of swordtail being cultured.

Objective 2

Through the culmination of a 4-year effort, the freshwater rotifer variety, *Brachionus calciflorus*, has been made available to farmers as a live food for freshwater ornamental fish larvae in Hawaii. It is now listed in the Hawaii Revised Statutes as a conditionally approved organism and can be imported into Hawaii in the least restrictive category.

Culture trials using live algal preparations (Chlorella, "green water") technology as well as testing a variety of commercial dry mixes as potential feeds for the freshwater rotifers were completed during the reporting period. The Chlorella appears to be impractical to use unless there is enough of a demand to offset its cost and limited shelf life.

The procedure of artificially inseminating the swordtail as well as other live bearers was documented and project work group members also received training in the procedure. This method is useful for handling small numbers (5-10) of individuals but will require additional

investigation to make it more efficient for use to handle large (>1000) individuals at a time.

The spawning activities observed of the Corydoras given the various treatment feeds demonstrated consistent patterns. It was concluded that individuals fed the beef heart/seafood mix and beef heart only, produce twice the amount of eggs during the time they are fed that diet than those fed only the commercial salmon pellet.

RESULTS AT A GLANCE . . .

- Identified two principal factors (swordtail strain and stocking density) that appear to affect the resulting sex ratio of swordtails during 90-day growout trials.
- Freshwater rotifers are now available for use by Hawaii's aquafarmers.
- Completed two workshops held during the reporting period
- Printed 11 newsletter articles in Sea Grant, Honolulu Aquarium Society and Center For Tropical and Subtropical Aquaculture newsletters.
- Two conference presentations
- Two summary manuscripts submitted to conference proceedings.

Objective 3

A submerged biofilter, trickle filter and no filter controls (triplicated) were tested. The trickle filter containing 20 liters of biomedia was found to be the most effective filter capable of supporting 4.6 kg m^3 of fish at a daily feeding rate of 120 g per day.

Objective 4

Two workshops were held during the reporting period, "Fish Health Management in Hawaii Aquaculture" presented by Dr. James Brock of ADP at UH Hilo and "Producing and Selecting Fancy Livebearers" presented by Dr. Clyde Tamaru of Sea Grant Extension Service and Fred Morita, hobbyist.

Project work group members produced a total of 11 newsletter articles, made presentations at two conferences, submitted two manuscripts for printing in conference proceedings and submitted one manuscript to a trade magazine during the reporting period.

IMPACT

The most immediate impact is the acceptance of culturing Moina as an alternative live food to *Artemia*. The initial work was carried out during the previous project and part of the current project. Use of Moina represents a considerable savings in production costs in light of the escalating prices for Artemia cysts. The forecast for the future of Artemia is tenuous and it appears the trends of increasing prices and lower availability underscores the importance of this activity. It is anticipated that the use of freshwater rotifers by the private sector will also significantly impact production costs as well as allowing for a diversification of species being cultured in the state.

The nutrition studies clearly show that a feed that has been demonstrated to be suitable for growing out fish does not necessarily mean it is suitable for maturation and spawning. Further investigations in this area will no doubt lead to an improved efficiency and cost savings in overall production of freshwater ornamentals.

WORK PLANNED

- Small-scale trials that combine stocking density and water temperature are close to being completed and the data will be summarized. Validation trials are to commence thereafter at Windward Community College facility, which is currently being renovated to accommodate the replicate tanks, needed for statistical validation.
- Modular phytoplankton and culture facilities are to be installed and field-tested for both rotifer and moina culture at the renovated WCC facility. Pilot-scale testing is to be done and results summarized. The facility is to be used to provide material to test the effects of enriching live feeds for growth and spawning of freshwater ornamental fish as well as serve as a hands-on demonstration facility for workshops covering this topic.
- Upon completion of the renovation, larval rearing trials and maturation experiments will be conducted to assess the effects of boosting HUFA content in live feeds.
- Technical assistance will be provided upon request.
- A workshop series that will cover fancy live bearers, moina culture and rotifer culture are being planned. Handouts and two manuals are being drafted and are to be finalized. A presentation will be made at the World Aquaculture Society Conference in May 2001 entitled "Spawning of the Armored Catfish *Corydoras Aeneus* fed three different maturation diets".

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Newsletter Articles

Ako, H., L. Asano and C.S. Tamaru. 2000. Factors affecting sex ratios in the swordtail, *Xiphophorus helleri*. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 10.

Ako, H. and C.S. Tamaru. 2000. Determining feed preference of koi feeds. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 5.

Ako, H. and C.S. Tamaru. 2000. What is the best feed for my ornamental fish? Aquatips, Regional Notes, Center for Tropical and Subtropical Aquaculture. Vol. 11, No. 2, Winter 2000.

Tamaru, C.S. and M. Lyum. 2000. Use of clove oil for anesthetizing freshwater ornamental fishes. I'a O Hawai'i, Vol. 2000, No. 5.

Tamaru, C.S., L. Pang and H. Ako. 2000. Effects of three maturation diets on spawning of the armored catfish (Corydoras aeneus). Aquatips, Regional Notes, Center for Tropical and Subtropical Aquaculture. Vol. 11, No. 3, Spring 2000.

Tamaru, C.S., G. Takeshita, F. Morita, and M. Yamamoto. 2000. Breeding fancy livebearers. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 4.

Tamaru, C.S. and V. Sato. 2000. Two new rotifers available for aquaculturists in Hawai'i. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 3.

Tamaru, C.S. and H. Ako. 2000. Essential fatty acids profiles of maturation feeds used in freshwater ornamental fish culture. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 2.

Tamaru, C.S., H. Ako and L. Asano. 2000. Culturing freshwater rotifer for use in the production of freshwater ornamental fishes - Part II. I'a O Hawai'i, Vol. 2000, No. 3.

Tamaru, C.S., H. Ako and L. Asano. 2000. Culturing freshwater rotifer for use in the production of freshwater ornamental fishes - Part I. I'a O Hawai'i, Vol. 2000, No. 2.

Tamaru, C.S. 2000. New Direction for moina research. I'a O Hawai'i, Vol. 2000, No. 1

Trade Magazine

Tamaru, C.S., H. Ako and L. Asano. 2000. Culturing Freshwater Rotifers For Use In The Production of Freshwater Ornamental Fishes. Tropical Fish Hobbyist. (Submitted).

Conference Proceedings

Ako, H., C. S. Tamaru, L. Asano, R. Yuen and M. Yamamoto. 2000. Achieving Natural Coloration in Fish Under Culture. Proceedings of the 28th U.S. Japan Aquaculture Panel Meeting. (In Press).

Tamaru, C.S. and Ako, H. 2000. Using Commercial Feeds for the Culture of Freshwater Ornamental Fishes in Hawai i. Proceedings of the 28th U.S. Japan Aquaculture Panel Meeting. (In Press).

Conference Presentations

Tamaru, C.S., L. Pang and H. Ako. 2000. Spawning of the armored catfish *Corydoras aeneus* fed three different maturation diets. World Aquaculture Society, World Aquaculture Society, May 2-6, 2000. Nice, France. Book of Abstracts, pg. 696.

Tamaru, C.S., H. Ako and L. Asano. 2000. Culturing freshwater rotifer for use in the production of freshwater ornamental fishes. Hawaii Aquaculture Association, 2nd Annual Conference. February 24, 2000, Windward Community College.

EXPANSION AND DIVERSIFICATION OF FRESHWATER TROPICAL FISH CULTURE – Termination Report

This project was terminated because all objectives were completed.

Funding Level
Year 1 ……$50,000
Year 2 ……100,000
Year 3 ……110,000
Total ……$260,000

Participants
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Lena Asano……Dept. of Environmental Biochemistry
Rich Bailey……Sea Grant Extension Service
Dr. James Brock……Aquaculture Development Program
Dr. Christopher Brown……..Hawaii Institute of Marine Biology
Calvin Chun……..Tropi Cal's of Hawaii
Brian Cole …….Sea Grant Extension Service
Michael Harrington ……...University of Hawaii
Ray Kosaka……Honolulu Aquarium Society
Paul Kotol ……..Sea Grant Extension Service
Meryl Miyashiro…….Moanalua High School
Resti Pagurigan……...Dept. of Environmental Biochemistry

Principal Investigator
Clyde Tamaru,
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PROJECT OBJECTIVES

Developing a freshwater ornamental fish industry in Hawaii is believed to be one means to diversify the agricultural output and stimulate the stagnant economy in Hawaii. However, it has long been recognized that considerable efforts are needed to develop the freshwater ornamental culture technologies along with the various other aquaculture technologies to an economically viable level within the State. The overall goal of the freshwater ornamental fish projects was to foster development of an ornamental fish culture industry in Hawaii. To accomplish this a variety of objectives were proposed and implemented during the reporting period:

1. Exposing aquafarmers to various culture scenarios

2. Introducing production/marketing infrastructure scenarios to tropical fish growers in Hawaii
3. Recirculating systems
4. Demonstrating out of season maturation and spawning of at least one freshwater ornamental fish species (i.e., rainbow or redtail sharks).
5. Completing a paper study of aquatic ornamental plants that hold potential for culture in Hawaii.
6. Conducting extension activities in the form of technical workshops, site visitation, verbal consultations and providing literature support of developments of additional farms geared towards the culture of freshwater tropical fish in Hawaii.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Objective 1

Activities under this portion of the projects ranged from operating an incubator hatchery for the production of first feeding larvae of selected egg layers to importation and amplification of five new species that represented different culture scenarios than the species brought in under the auspices of previous projects. Broodstock of tinfoil barbs, rainbow sharks, albino rainbow sharks, tiger barbs, blue and gold gouramis that were established at the Windward Community College facility were used to provide first feeding larvae. Mature individuals were either induced or conditioned to spawn to produce fry for distribution to prospective farmers. The number of fry distributed during the reporting period totaled 267,432 fry from the six species of freshwater ornamentals. From reviewing local wholesalers price lists of freshwater ornamentals being sold from Hawaii, the gouramis have become a sustainable crop along with the swordtails distributed under the auspices of the current and previously supported CTSA projects. Egg layers, particularly, the ones that require hormonal induction of spawning and larval culture have not become sustainable item(s) and will require additional technical assistance for it to be part of Hawaii's production portfolio.

RESULTS AT A GLANCE . . .

- The number of freshwater ornamental farms increased from 22 to 71 from 1994 to 1998.
- Freshwater ornamental fish production is now listed as its own category rather than in "Other".
- Sales increased $174,100 from 1995 to 1996.

From the information obtained it is clear that broodstock that require hormonal induction of spawning resulted in no production and marketing of product. The overall range in success (marketing and cultured product) was between 20-67% for the fishes for which information was obtained. In a survey sent to the recipients of fry or broodstock, information was requested as to whether additional freshwater ornamental species were requested. Of the 14 responses that were received, nine had purchased their own broodstock of other species or strains and were at various stages of production.

Based upon the responses and input from the project work group, the following species were decided upon as the five additional species to be imported under the auspices of the projects and amplified for distribution. The species selected were: Serpae Tetra, Lemon Tetra, Hi

Fin Red Wag Platy, Ram Cichlid, and Lyre tail swordtail. Broodstock of the serpae and lemon tetras were established at Windward Community College and two informational brochures were produced. Broodstock amplification of both the lyretail and hi-fin platy have been achieved and workshops are currently underway to introduce farmers to the characteristics for culturing these species.

Objective 2

A collaborative workshop series held previously which targeted rural communities was repeated during the reporting period. There were six individual workshops which covered the topics of: an introduction to freshwater ornamental culture; production concerns for freshwater ornamentals; marketing of freshwater ornamentals; business planning and financing ornamentals culture activities; niche marketing of other agricultural activities; and state and regional agricultural alliances of other agricultural activities.

Objective 3

Three recirculating tank systems equipped with three different biofilters (spray bar trickle filter, bead filter, and air lift system) were built during the reporting period and stocked with approximately 5000 tinfoil barb fry per tank. The results indicate that with regard to Total Ammonia Nitrogen (TAN) the trickle filter was comparable to the bead filter both of which were found to be superior to the airlift system. The highest rate of feeding reached 500 g/day which is considered to be indicative of intensive culture (5-10 kg of fish).

A concerted effort was made during the latter stages of the projects to develop adequate facilities to address the recirculating issues in an empirical fashion. Elimination of the rubber-lined ponds and installation of two gravel pads have been completed. The project site is now equipped with 10 x 600 gallon tanks, 10 x 500 gallon tanks and 24 x 40 gallon tanks. Triplicated treatments investigating different types of biofilters are currently in progress. At present this activity has been proposed as an objective in the second year of the Transitioning Freshwater Ornamental Fish Culture in Hawaii.

Objective 4

Gonadal maturation of three species (rainbow sharks, red tail black sharks and tinfoil barbs) of ornamental egg layers was monitored during the earlier project (Year 1). From the results obtained it is clear that the species investigated exhibit discrete spawning seasons. Broodstock rainbow sharks were held under extended day length (LD:14:10) and elevated water temperatures (20-24 ^{o}C) from November 1998. On April 26, 1999 females were found to possess oocytes in which germinal vesicle migration could be detected in the majority of oocytes examined. Females were successfully induced to spawn using a combination carp pituitary extract (CPE: 3mg per kg fish) and HCG (60 IU per kg fish). The first spawning trial resulted in approximately 45,000 hatched larvae (95% fertilization and hatching) demonstrating out of season maturation and successful production of fry out of season. During the second spawning trial (May 1999) eighty females were induced to spawn resulting in the production of approximately 125,000 fry. The results of the trial were summarized in a manuscript which has been submitted for publication.

Objective 5

Obtaining data on the market of freshwater aquatic plants has proven to be difficult and it will take a more serious input of resources to uncover important details of the market. During the reporting periods several attempts were made to obtain additional funds to support that effort but were largely unsuccessful. From the data obtained, it was concluded that the competition for breaking into the market with the plants will be very difficult. The initial findings are that the industry may have a chance at the transshipping level or niche level for specializing plants.

Objective 6

On average, funded personnel responded to 600 requests for technical assistance annually during the course of the projects. Approximately 50% of these were verbal consultations, 35% were providing written material and 15% were conducting site visits. A total of 35 workshops were held on three islands. Project work group members have completed four journal publications, eight technical handouts, 13 conference presentations and 34 newsletter articles.

IMPACT

The impact summarized describes the contributions made from the current and previously funded projects in a collective matter.

By reviewing the number of farms that report freshwater ornamentals as a commodity, a relative measure of success can be translated. From the start of the initial CTSA supported project in 1994, there has been an upward trend in the number of farms that produce freshwater ornamental fishes. The number was 22 in 1994 and has risen to 71 in 1998.

Data indicates that the freshwater ornamental growers in Hawaii have reached a sufficient critical mass to be considered by the Department of Agriculture's Aquaculture Development Program as a small industry in Hawaii's aquaculture scene. As of 1997, freshwater ornamental fish production is ranked as a discrete commodity group rather than categorized under "Other".

Since the inception of this project in 1996, there has been a dramatic increase in sales for ornamental fish and plants from $271,700 in 1995 to $445,800 in 1996 (statistic provided by USDA's Aquaculture Development Program).

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

Journal Articles

Ako, H. and C.S. Tamaru. 1999. Are feeds for foodfish practical for aquarium fish? International Aquafeeds, Issue 2, 1999: 30-36.

Brown, C. L., B. Cole, C. Farfan and C. Tamaru. 1999. Out-of-season spawning of the rainbow shark, *Epalzeorhynchus frenatus*. Progressive Fish Culturists (submitted).

Tamaru, C.S., H. Ako, and L. Asano. 2000. Culturing Freshwater Rotifers for use in the Production of Freshwater Ornamental Fishes. Tropical Fish Hobbyist (Submitted upon request from editor).

Tamaru, C.S., H. Ako, and R. Paguirigan. 1997. Essential fatty acid profiles of maturation feeds used in freshwater ornamental fish culture. Hydrobiologia 358:265-268.

Technical handouts

Bailey, R. and B. Cole. 1999. Spawning the tinfoil barb, *Barbodes schwanenfeldi* in Hawaii. Center for Tropical and Subtropical Aquaculture Publication Number 136.

Cole, B. and M. Haring. 1999. Spawning and production of the Serpae tetra, *Hyphessobrycon serpae*. Center for Tropical and Subtropical Aquaculture Publication Number 138.

Cole, B., P. Kotol, and M. Haring. 1999. Spawning and production of the lemon tetra *Hyphessobrycon pulchripinnis*. Center for Tropical and Subtropical Aquaculture Publication Number 142.

Cole, B., Tamaru, C.S., R. Bailey and C. Brown. 1998. A Manual for Commercial Production of the Gourami, *Trichogaster trichopterus*, A Temporary Paired Spawner. Center for Tropical and Subtropical Aquaculture, Publication Number 135. 24 pp.

Cole, B., Tamaru, C.S., R. Bailey, C. Brown and H. Ako. 1999. Shipping practices in the ornamental fish industry. Center for Tropical and Subtropical Aquaculture, Publication Number 131. 25 pp.

Tamaru, C.S., B. Cole, R. Bailey and C. Brown. 1997. Commercial production of the tiger barb, *Capoeta tetrazona*. Center for Tropical and Subtropical Aquaculture, Publication Number 128, 65 pp.

Tamaru, C.S., B. Cole, R. Bailey, and C. Brown. (In preparation). A manual for commercial production of the swordtail, *Xiphophorus helleri*. Center for Tropical and Subtropical Aquaculture. 55 pp.

Tamaru, C.S., H. Ako, R.S. Pagurigan, and L. Pang. 1998. Enrichment of artemia for use in freshwater ornamental fish production. Center for Tropical and Subtropical Aquaculture Number 133. 29 pp.

Conference Presentations

Ako, H., C.S. Tamaru, and L. Asano. 1999. Color, maturation and palatability of feeds. Conference Abstracts, AQUARAMA '99; World Conference on Ornamental Fish Aquaculture, June 3-6, 1999. Singapore.

Ako, H., C.S. Tamaru, L. Asano, and M. Yamamoto. 1999. Use of bioreactor produced cartenoids and foodfish feeds for growout of ornamental fish. Book of Abstracts, US Chapter of the World Aquaculture Society, Aquaculture America, January 27-30, 1999, Tampa, Florida. Pg. 1.

Ako, H., C.S. Tamaru, L. Asano, B. Yuen, and M. Yamamoto. 1999. Achieving natural coloration of aquaculture fish. US-Japan Natural Resources Aquaculture Panel 28th Joint Meeting, Spawning and Maturation of Aquaculture Species. November 10-12, 1999. Hawaii, USA.

Ako, H., C.S. Tamaru, L. Asano, B. Yuen, and M. Yamamoto. 1999. Carotenoids and ornamental fish. World Aquaculture Society, April 26- May 2, 1999. Sydney, Australia. Book of Abstracts, p. 10.

Ako, H., C.S. Tamaru, T. Nishimura and J. Rosowski. 1998. Feed palatability testing with ornamental fish. World Aquaculture Society, February 14-19, 1998. Las Vegas, Nevada USA, Book of Abstracts, p. 2.

Bailey, R., C.S. Tamaru, B. Cole and C. Brown. 1999. Live bearer production system developed for Hawaii. Conference Abstracts, AQUARAMA 99; World Conference on Ornamental Fish Aquaculture, June 3-6, 1999. Singapore.

Bailey, R., C.S. Tamaru, B. Cole, C. Brown. 1997. Performance of an intensive culture module for commercial production of the swordtail, *Xiphophorus helleri*, in Hawaii. Presented at the World Aquaculture Society, Feb. 18-24, 1997, Seattle, Washington.

Cole, B., C.S. Tamaru, R. Bailey, C. Brown. 1997. Effects of stocking density on growth and survival of the swordtail, *Xiphophurus helleri*. Presented at the World Aquaculture Society annual meeting, Feb. 18-24, 1997, Seattle, Washington.

Cole, B., Tamaru, C.S., R. Bailey and C. Brown. 1999. Shipping practices in the ornamental fish industry. West Coast Live: Marketing and Shipping Live Aquatic Products '99. Nov. 14-17, 1999. Seattle, Washington, USA.

Tamaru, C.S. and H. Ako. 1999. Experiences on the use of commercial feeds on the culture and spawning of freshwater ornamental fishes in Hawaii. US-Japan Natural Resources Aquaculture Panel 28th Joint Meeting, Spawning and Maturation of Aquaculture Species. November 10-12, 1999. Hawaii. USA

Tamaru, C.S., H. Ako, L. Asano, and L. Pang. 1999. Growth and enrichment of *Moina* for use in freshwater ornamental fish culture. World Aquaculture Society, April 26- May 2, 1999. Sydney, Australia. Book of Abstracts, p. 751.

Tamaru, C.S., H. Ako, R. Paguirigan Jr., M. Miyashiro and C. Chun. 1998. Growth of angelfish, *Pterophyllum scalare*, and guppies, *Poecilia reticulata*, fed commercial feeds developed for foodfish. World Aquaculture Society, February 14-19, 1998. Las Vegas, Nevada USA. Book of Abstracts, 533.

Tamaru, C.S., L. Pang and H. Ako. 2000. Spawning of the armored catfish *Corydoras aeneus* fed three different maturation diets. World Aquaculture Society, World Aquaculture Society, May 2-6, 2000. Nice, France. Book of Abstracts, p. 696.

Newsletter Articles

Ako, H. and C.S. Tamaru. 1999. Are feeds for foodfish practical for aquarium fish? International Aquafeeds, Issue 2, 1999: 30-36.

Ako, H. and C.S. Tamaru. 1997. Palatability and growth supporting characteristics of Koi feeds. I'ao O Hawai'i, Vol. 1997, Issue 6.

Ako, H. and C.S. Tamaru. 1998. Astaxanthin, killing fish and the 13-inches per gallon rule. I'a O Hawai'i, Vol. 1998, Issue 9.

Ako, H. and C.S. Tamaru. 2000. Determining feed preference of koi feeds. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 5.

Ako, H. and C.S. Tamaru. 2000. What is the best feed for my ornamental fish? Aquatips, Regional Notes, Center for Tropical and Subtropical Aquaculture. Vol. 11, No. 2. Winter 2000.

Ako, H., D. Tagomori, K. Lum, C.S. Tamaru and R. Spencer. 1997. Palatability of different feeds using the swordtail, *Xiphophorus helleri*, I'ao O Hawai'i, Vol. 1997, Issue 10.

Ako, H., C.S. Tamaru, L. Asano, and D. Tagomori. 1998. Biofilters and water recirculation. I'a O Hawai'i, Vol. 1998, Issue 12.

Ako, H., L. Asano, K. Brittain, C. S. Tamaru. 1999. Enhancing color in ornamental fishes. University of Hawaii Sea Grant College Program, Makai, Vol. 21, No. 6.

Ako, H., L. Asano, M. Fukada, and C.S. Tamaru. 1999. Culture of rainbowfish *Psuedomugil furcatus*, and the use of a Hawaii-specific feed, Tropical Gold. I'a O Hawai'i, Vol. 1999, Issue 2.

Ako, H., T. Nishimura, and C.S. Tamaru. 1998. Palatability of flake feeds using the emperor tetra. I'a O Hawai'i, Vol. 1998, Issue 5.

Ako, H., T. Nishimura, and C.S. Tamaru. 1998. Testing palatability of flake feeds using the tinfoil barbs. I'a O Hawai'i Vol. 1998, Issue 8.

Ako, H., T. Nishimura, and C.S. Tamaru. 1998. Testing palatability of flake feeds using the guppy. I'a O Hawai'i Vol. 1998, Issue 6.

Tamaru, C.S. 2000. New Direction for moina research. I'a O Hawai'i, Vol. 2000, No. 1

Tamaru, C.S. and H. Ako. 1997. Essential fatty acid profiles of maturation feeds used in freshwater ornamental fish culture. I'a O Hawai'i, Vol. 1997, Issue 8 and 9.

Tamaru, C.S. and H. Ako. 1997. Growth of the angelfish, *Pterophyllum scalare*, using various commercial feeds. I'a O Hawai'i, Vol. 1997, Issue 4.

Tamaru, C.S. and H. Ako. 2000. Essential fatty acids profiles of maturation feeds used in freshwater ornamental fish culture. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 2.

Tamaru, C.S. and M. Lyum. 2000. Use of clove oil for anesthetizing freshwater ornamental fishes. I'a O Hawai'i, Vol. 2000, No. 5.

Tamaru, C.S. and V. Sato. 2000. Two new rotifers available for aquaculturists in Hawaii. University of Hawaii Sea Grant College Program, Makai, Vol. 22, No. 3.

Tamaru, C.S., F. Lum, and J. Brock. 1999. Monitoring water temperature in your aquariums. I'a O Hawai'i, Vol. 1999, No. 9.

Tamaru, C.S., G. Takeshita, F. Morita and M. Yamamoto. 2000. The breeding of fancy livebearers. Sea Grant College Program, Makai, Vol. 22, No. 4.

Tamaru, C.S., H. Ako and C. Carlstrom-Trick. 1998. We need to help each other keep up with the Hawaii Administrative Rules for the importation of biological commodities. I'a O Hawai'i , Vol. 1998, Issue 11.

Tamaru, C.S., H. Ako and L. Asano. 2000. Culturing freshwater rotifer for use in the production of freshwater ornamental fishes - Part II. I'a O Hawai'i, Vol. 2000, No. 3.

Tamaru, C.S., H. Ako and L. Asano. 2000. Culturing freshwater rotifer for use in the production of freshwater ornamental fishes - Part I. I'a O Hawai'i, Vol. 2000, No. 2.

Tamaru, C.S., H. Ako, R. Pagurigan, and C. Chun. 1997. Growth of guppies, *poecilia reticulata*, using various commercial feeds. I'a O Hawai'i, Vol. 1997, Issue 5.

Tamaru, C.S., H. Ako, R. Pagurigan, and C. Chun. 1998. Growth of guppies, *Poecilia reticulata*, using various commercial feeds. Makai, University of Hawaii Sea Grant College Program. Vol. 20, Issue 5.

Tamaru, C.S., H. Ako., R. Pagurigan, Jr. and M. Miyashiro. 1998. Comparison of various feeds on growth of the angelfish, *Pterophyllum scalare*. Makai, University of Hawaii Sea Grant College Program. Vol. 20, No. 1.

Tamaru, C.S., L. Pang and H. Ako. 2000. Effects of three maturation diets on spawning of the armored catfish (*Corydoras aeneus*). Aquatips, Regional Notes, Center for Tropical and Subtropical Aquaculture. Vol. 11, No. 3, Spring 2000.

Tamaru, C.S., L. Pang, and H. Ako. 1999. Spawning of the armored catfish, *Corydoras aeneus*. I'a O Hawai'i, Vol. 1999, No. 10.

Tamaru, C.S., L. Pang, H. Ako, and L. Asano. 1998. Growth and enrichment of *Moina* for use in freshwater ornamental fish culture. I'a O Hawai'i Vol. 1998, Issue 10.

Tamaru, C.S., L. Pang, V. Sato, H. Ako and L. Asano. 1999. Use of Moina as an alternative live food organism in the culture of freshwater ornamental fish larvae. University of Hawaii Sea Grant College Program, Makai, Vol. 21, No. 4.

Tamaru, C.S., V. Sato, L. Nakamitsu, H. Ako and L. Asano. 1999. Fatty acid profiles and use of Moina as an alternative to Artemia in the culture of freshwater ornamental fish larvae. I'a O Hawai'i, Vol. 1999. Issue 5.

Tamaru, C.S., V.T. Sato, and H. Ako. 1999. Use of *Chlorella vulgaris* for the culture of *Moina micrura*. University of Hawaii Sea Grant College Program, Makai, Vol. 21, No. 8.

Tamaru. C.S., V. Sato, and H. Ako. 1999. Update on the culture of Moina in Hawaii. I'a O Hawai'i, Vol. 1999, No. 6.

AQUACULTURE OF MARINE ORNAMENTAL SPECIES – Year 1

Funding Level Year 1 ……..$128,735
Total ……$128,735

Current Participants
Frank Alig……...……..Guam Aquaculture Development and Training Center
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PROJECT OBJECTIVES

The solicitation for this research effort called for a multi-institutional approach. Members of the Project Work Group were part of teams established to complement each other's efforts and thus have separate objectives to complete the overall goal to develop and transfer innovative culture technologies for high value ornamental fish and invertebrate species.

A. The Oceanic Institute

1. Establish broodstock populations of yellow tang (*Zebrasoma flavescens*) and flame angelfish (*Centropyge loriculus*), or other angelfish for project use.
2. Compare various live food organisms as first-feeds for larvae.
3. Conduct semi-intensive, "brown" water mesocosm trials and determine live food preferences of larvae.

B. The University of Hawaii and UH Sea Grant – Finfish

4. Compare rotifer and semi-intensive, "green" water mesocosm systems for culture of larvae.
5. Identify other fish species adaptable to "green" water systems.
6. Perform mass culture trials with identified species using, initially, a "green" water mesocosm, with transition onto nutritionally-enhanced rotifers.

C. The Guam Aquaculture Development and Training Center

7. Establish broodstock populations of Clown coris (*Coris gaimard*) or other wrasse.
8. Develop methods for wild zooplankton collection.
9. Conduct larval rearing trials if larvae and zooplankton are available.

D. The University of Hawaii and UH Sea Grant -- Invertebrates

10. Establish broodstock colonies of feather-duster worms (*Sabellastarte sanctijosephi*) and trocophore collection techniques at chosen farm facilities.
11. Develop methods to settle and culture metamorphosed worms.
12. Describe growth rates and stages of development.

ANTICIPATED BENEFITS

The project is designed to enhance the commercial production of marine ornamental fish and invertebrate species by developing new technologies for species that have a market, but are presently unexploited because of bottlenecks in technology.

The ultimate goal of this project is to assist in the development of a marine ornamental aquaculture industry in Hawaii and the Pacific. This represents a key economic opportunity for farmers in the state of Hawaii and Pacific Island affiliates such as Guam for several reasons. Firstly, there is a worldwide void in aquaculture production of marine ornamental species. Freshwater ornamental farmers in Hawaii must compete in saturated markets with well-established foreign and other domestic producers; however, it is estimated that less than 5% of all marine ornamental species traded on the open market are aquacultured, and that the actual numbers of cultured fish traded is miniscule compared to those traded by collectors. Secondly, it is well known that the health of coral reef ecosystems around the world is being severely degraded, and that wild collection practices are likely unsustainable unless alternatives are sought.

Moreover, the Hawaiian Islands are home to over 85% of the coral reefs in the United States, well-positioning the region to develop an aquaculture-based industry. Success of this project will not only provide new economic opportunity to farmers, but will also help ensure the long-term sustainability of the marine ornamental trade by providing alternatives to wild collection practices, and a means to practice resource conservation.

Successful completion of this project will provide hatchery techniques to culture several

species of marine ornamentals, thereby offering a more environmentally sustainable alternative to wild collection practices. Identification of the most appropriate food item would lead to development of methods to mass-produce them for larvae. Consistency in production would ensure a solid base for development of an industry and transfer of reliable technologies. Techniques to mature and spawn the species chosen could be transferred to other highly desired ornamental fish, allowing for the rapid development of new aquacultured species. Further, development of reliable methods to mass culture live feeds would benefit the entire spectrum of marine ornamental culture. As expressed in the June 1997 newsletter of the American Marine Life Dealers Association, several benefits, apart from cost savings, will accrue from the financial investment in research and development of captive propagation, including new economic development, job creation, and an increased emphasis on the importance of maintaining coastal resources. Additional economic benefits will flow throughout the industry, strengthening aquarium and pet retail stores and benefiting consumers with healthier fish.

PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Following are the accomplishments for this year:

Objective 1

Pygmy angelfish (Centropyge sp.)

Under captive conditions at OI, they have successfully obtained flame angelfish spawns daily for almost one year with individual spawns from a single pair, reaching values as high as 12,000 eggs per spawn. The species clearly acclimates well to the conditions of intensive culture. Further, tank cleaning and removal of fish for husbandry purposes are well tolerated, often resulting in slightly larger spawns. Care was taken to avoid buildup of aptysia in tank outflows, as it was shown they were veracious consumers of eggs.

Yellow tang (Zebrasoma flavescens)

A total 25 yellow tang broodstock collected off the Kona coast in 1999 have been successfully maintained in two broodstock tanks for approximately one year. They were successful in developing an external means of sexing individuals based on external characters. Despite being maintained in a healthy condition for over one year, we have yet to see any indication of spawning behavior or the appearance of spawned eggs in egg collection nets.

Objective 2

Experiments were carried out to quantify the hatch rate of flame angelfish, *Centropyge loriculus*, embryos spawned naturally at OI and to examine early larval development of this species. A new rearing system, suitable for replicated experiments with small quantities of fish eggs/larvae, was also commissioned and evaluated against existing rearing facilities. Preliminary feeding experiments were then undertaken to quantify the ingestion by flame angelfish larvae of mono-cultured prey items (SS-Type rotifers, *Brachionus rotundiformis*, nauplii of *Euterpina acutifrons* and

RESULTS AT A GLANCE . . .

- Established broodstock populations for yellow tang and pygmy angelfish.
- Several species of juvenile wrasse were obtained and habituated to onshore tanks.
- An external means of sexing individuals of yellow tang has been developed.
- Angelfish spawns have been maintained daily for almost year.

trochophore larvae of *Crassostrea gigas*).

An experiment was conducted to quantify hatch rate for flame angelfish embryos held in two types of incubation systems. Mean percentage hatch was higher in the 10L polyethylene tanks (80.0%±9.7) than in 1L glass beakers (69.2%±12.6), although these differences were not statistically significant.

A follow up experiment was conducted using the 10L rearing tanks to examine the effects of water treatment on survival of flame angelfish larvae during yolk resorption. In the absence of any significant treatment differences in survival, the multi-component water treatment process was adopted for subsequent rearing studies.

In an experiment to evaluate SS-type rotifers, *Brachionus rotundiformis*, and *E. acutifrons nauplii* as initial prey for flame angelfish larvae, embryos were stocked into each of twelve 10L tanks at a density of 28/L.

The dimensions of angelfish larvae receiving SS rotifers or *E. acutifrons* did not differ significantly from unfed larvae throughout the experiment, i.e. all larvae exhibited negative growth. A preliminary experiment to test the combined feeding approach was undertaken, using trochophore larvae of *Crassostrea gigas* (mean diameter 50µm). However, the experiment was aborted due to high mortality of the oyster trochophores.

Objective 3

Research is targeted during the next reporting period to evaluate semi-intensive larviculture methods for flame angelfish.

Objectives 4-6

No progress was made on the technical aspects of this segment over the last 6 months. Interviews for hiring a research associate position are currently ongoing. In addition, Dr Tamaru was approved in October for faculty affiliate status at the University of Hawaii to access facilities at the Hawaii Institute of Marine Biology (HIMB) which will be set up and used in lieu of the AFRC.

Objective 7

Several species of juvenile wrasse, including the clown coris, *C. gaimard*, were obtained by local collectors over the report period and habituated to onshore tanks. After several months, groups of Clown coris were combined and transferred to larger, 3,000L tanks located outdoors. Fish were combined based on relative age and behavior. Behavior of the fish is currently being observed for establishment of a social hierarchy. There have been no recorded spawns to date.

Objective 8

Efforts are underway to arrange collection trips. The project is presently exploring ways to ship samples from Guam to Oahu.

Objective 9

No progress was made due to lack of eggs.

Objectives 10-12

No progress was made. Mr. Richard Bailey resigned his position with USGES. A replacement is being sought.

IMPACT

Because this project has just begun, there is currently no immediate impact to report.

WORK PLANNED

Over the next 6 months, project participants plan to accomplish the following:

A. The Oceanic Institute

- Repeat trials on use of oyster trocophores as a first feeding item for flame angelfish.
- Conduct semi-intensive, "brown" water trials with flame angelfish.
- Continued collection of spawning data on *Centropgye loriculus*.
- Examination of the effect of tank size on natural spawning of *C. loriculus*.
- Continued habituation of yellow tang broodstock to captivity.

B. The University of Hawaii and UH Sea Grant Extension Service – Finfish

- Hire research associate to assist in feeding trials.
- Establish testing facilities at HIMB.
- Initiate "green" water feeding trials of fish.

C. The Guam Aquaculture Development and Training Center

- Recruit biologist aide.
- Inquire about the possibility of shipping formalin (fixed plankton samples).
- Start communication with C. Hunter for potential identification of collected plankton.
- Purchase supplies, assemble plankton collection cage and start data collection.
- Initiate the first of the plankton tows from boat and data collection.
- Draft memo to reprogram boat rental budget to plankton analysis budget/contract.

D. The University of Hawaii and UH Sea Grant – Invertebrates

- The Project PI will seek to find a replacement for Mr. Bailey to carry out this task or recommend a reevaluation of the task.

PUBLICATIONS, MANUSCRIPTS OR PAPERS PRESENTED

No publications have been produced to date.