

**AQUAFISH CRSP
IMPLEMENTATION PLAN 2009–2011
ADDENDUM**

MARCH 2011



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Collaborative Research Support Program
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AQUAFISH CRSP IMPLEMENTATION PLAN 2009–2011, ADDENDUM

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Cover Photo

A Host Country researcher in Mexico investigating the feasibility of culturing the native Pacific Coast oyster *Crassostrea corteziensis* in coastal bays holds up a spat collector in use in a mangrove area of Boca de Camichin. Photo by HC Lead PI, Eladio Gaxiola.

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INTRODUCTION

This addendum to the AquaFish CRSP *Implementation Plan 2009-2011* includes seven add-on investigations undertaken by six US Lead institutions. These investigations focus on promoting the extension of CRSP technologies through outreach, commercialization, and partnerships; assessing impacts; and communicating the importance of CRSP research. Summaries of the add-on investigations are provided below.

AUBURN UNIVERSITY

A two-week training of six Master Trainers selected from Ghana, Tanzania, and Uganda (2 per country) will be held at Auburn University using its recently developed online Certification of Aquaculture Professionals (CAP) program. Upon completion, the Master Trainers will return to their countries to perform additional in-country trainings that they will organize and conduct. — 09TAP08AU

NORTH CAROLINA STATE UNIVERSITY

Host Country investigators will conduct surveys, workshops, and demonstrations to assess the impacts of (1) alternative feed reduction strategies in tilapia and milkfish farming communities in The Philippines and (2) polyculture training on fish and shrimp farming in Aceh, Indonesia. Workshops will be held on alternative feed practices and integrated milkfish polyculture. Six podcast modules designed to promote outreach and impacts of AquaFish CRSP research in Southeast Asia, Africa, and LAC also will be produced. — 09SFT06NC

PURDUE UNIVERSITY

In Ghana, AquaFish CRSP researchers will build capacity for an aquaculture training program at the Water Research Institute's Aquaculture Research Development Center. Farmer trainings will be held on tilapia and catfish propagation, hatchery management, and fingerling production. The workshops will also be open to researchers, extensionists, policy makers, and other interested stakeholders. Reference materials on fish life cycle and reproductive strategies will be developed. — 09QSD05PU

In Kenya, researchers will survey smallholder fish farmers who have participated in CRSP research and training programs dating back to 1996. They will collect comparative data on farmers' pre-CRSP production, management, and marketing practices versus their current practices. Assessment of these farm-level production data will help to identify factors influencing efficiencies of CRSP production technologies, which will be used to guide improved government policy making and extension services. — 09TAP07PU

UNIVERSITY OF ARIZONA

Mexican researchers will evaluate the commercial feasibility of bacteria-based degradation of methyltestosterone (MT), a hormone that hatcheries commonly use to masculinize tilapia fingerlings. Scale-up of the CRSP-developed MT-elimination technology will be tested in a large commercial hatchery. Farmer trainings will be held to introduce the use of bacterial bio-flocks for the dual purpose of MT-degradation and probiotic effects in aquaculture production systems. — 09MNE07UA

UNIVERSITY OF CONNECTICUT – AVERY POINT

This investigation will assess impacts of the integrated Mekong River Basin project that has been conducted in Cambodia and Vietnam. Participating student, researchers, and fisheries officers will initially be trained in impact assessment methodologies and data collection and analysis. They will then conduct impact assessments of the AquaFish CRSP work on snakehead aquaculture development and small-sized/low-value fisheries management. Reports will focus on technology adoption from the perspective of farm productivity, farmer welfare, and economic effects. — 09FSV03UC

UNIVERSITY OF MICHIGAN

In partnership with the Tongwei Co. Ltd in China, CRSP researchers will test the commercial applicability of a specially designed deep water aquaculture cage system that contains two novel features to reduce sediment discharge: (1) a collecting system to retain solid waste for safe removal and disposal and (2) a surrounding cage for secondary culture of fish that will feed on waste from the primary cage and small aquatic organisms that grow in the higher nutrient cage environment. Data collected on production statistics, nutrient dynamics, sedimentation rates, and water quality will be used to perform a cost-benefit analysis. — 09WIZ03UM

TOPIC AREA
SUSTAINABLE FEED TECHNOLOGY

IMPACT ASSESSMENT OF CRSP ACTIVITIES IN THE PHILIPPINES AND INDONESIA

Sustainable Feed Technology / Activity / 09SFT06NC

Collaborating Institutions & Lead Investigators

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Objectives

1. Provide training for and assess the impact of technologies and management practices that can improve incomes for small-scale aquaculture farmers in the Philippines and Indonesia.
2. Conduct extension activities and assess the impact of feed reduction strategies on tilapia culture in Central Luzon, Philippines
3. Enhance capacity building, provide training and assess impacts of alternative feeding practices, integrative culture systems, and value-added processing for milkfish production across a broader range of the Philippines.
4. Assess impacts of seaweed polyculture on fish and shrimp farming in Aceh, Indonesia
5. Produce podcast modules to promote outreach and impacts of AquaFish CRSP research in the Philippines, Indonesia and other host country sites in Southeast Asia, Latin American and Africa.

Significance

We propose a series of activities to promote training and to assess the impact of technologies and management practices aimed at improving incomes for small-scale aquaculture farmers in the Philippines and Indonesia. This includes a focus on the promotion of accomplishments from previous Aquaculture CRSP activities as well as our current AquaFish CRSP projects.

AquaFish CRSP work in the Philippines and Indonesia is designed to develop and implement strategies to improve the cost effectiveness, sustainability and income opportunities of farming fish in these countries, in order to improve the subsequent livelihood of their people. Tilapia and milkfish are the two most prominent finfishes cultured in the Philippines and their culture is expanding rapidly both in inland and coastal regions. Milled feeds constitute the most costly component of fish farming, representing as much as 80% of total production costs for tilapia and 60-70% for milkfish. Therefore, procedures that reduce the amount of feed or its cost without negatively impacting harvest quality or yield can improve farmers' incomes.

Through delayed onset, alternate day, feeding at subsatiation levels, or combinations of the three, research shows that the amount of feed required to culture tilapia in ponds can be reduced by as much as 50% with little or no impact on the total yield of harvested fish (Brown et al. 2000; Bolivar et al. 2003; Bolivar et al. 2006). These practical strategies to improve production efficiency of tilapia have increased the incomes of some farmers and have the potential to benefit others similarly. Anecdotal information and informal surveys suggest that some farmers have adopted these new feeding strategies in the Philippines, including those who conducted the original research trials on their farms, but the extent to which this technology has been implemented is uncertain. This may arise in part from the lack of farmers' knowledge of alternate feeding strategies. The extent to which farmers do utilize the new methods has not been quantified. For these reasons, one of the aims of the proposed work is to conduct additional training on alternative feeding practices for farmers and to survey the commercial tilapia farming community to assess the extent to which new feeding practices are being adopted. These are critical elements to the promotion of new technologies developed by the AquaFish CRSP, and to assess the impacts of this technology.

Milkfish culture is the largest finfish aquaculture industry in the Philippines. As part of the Philippine government's food security and poverty alleviation programs, expansion of milkfish culture is highly prioritized, both to wean fishers off dwindling capture fisheries and to increase income of farmers and fishers alike, whose poverty levels are disproportionately high. Much of the growth in milkfish production is in cage culture in marine or brackishwater coastal areas. Cage culture of milkfish is done at higher densities and with significantly greater dependence on artificial feeds. This practice, however, has led to wastage of artificial feeds and to excessive nutrient loading in receiving waters, exacerbating pollution problems and contributing to periodic fish kills in areas of intensive milkfish culture. It also appears many farmers use substantially more feed than recommended. Our preliminary results suggest that a reduction of 2-3% below the recommended daily feed ration during the early fingerling stage can produce fish without substantially reduced yields. We will be evaluating whether reductions in ration levels throughout the entire production period can produce cost savings for farmers. We will also use seaweed and sea cucumber in an integrative milkfish culture system to reduce nutrient overload seen around milkfish culture clusters. Overall, these studies are focused on reducing feed inputs and environmental impact of milkfish culture while providing cost savings and additional income opportunities to farmers through seaweed and sea cucumber markets. With this supplemental application we would like to expand training on alternative feeding practices and on integrative culture systems for milkfish from one location to additional islands where milkfish culture is even more prominent, namely in the Samar / Visayas and Mindanao regions of the Philippines. Expansion of workshops to these regions would provide wider outreach and training to farmers and communities while enhancing capacity building through the participation of additional local and regional government agencies and other stakeholders in the Philippines. Also, we intend to survey farmers to gain a better understanding of the varied feeding practices utilized, which will enhance our capacity to assess impacts of CRSP-sponsored research.

In Indonesia and the Philippines, interest has heightened in diversifying aquaculture crops, following the realization that intensive shrimp farming practices contributed to the deterioration of water quality in the mangrove coastal habitat. In our current AquaFish CRSP project we have conducted a series of workshops on incorporating seaweed in the polyculture of shrimp and fish as a more sustainable and environmentally benign, and profitable form of aquaculture. Although some farmers

have adopted these polyculture practices, the number of farmers, hectares, and communities impacted has not been assessed. We consequently propose to conduct household surveys in Aceh, Indonesia and to collect data (maps, surface hectares, number of farms, etc.) developed as communities were rebuilt following the 2004Tsunami. We will work with the Host Country Institute, Ujung Batee Aquaculture Center, the primary research and extension center in the Aceh province of Indonesia, to collate data and provide a full assessment of the impact of CRSP-funded training programs on alternative and sustainable management practices for the seafood farming communities of Aceh.

Podcasting is a wide-open, attractive format for digitizing procedures, technologies, pictures, data, and video clips set to music that can be used to distribute practically any digitized information that is desired. Unlike traditional brochures or fact sheets, it can be updated quickly and easily, and the updates are distributed automatically; unlike journals, texts, and fact sheets, they do not become obsolete. New advances can be incorporated into updates of old presentations for automatic distribution to holders of free subscriptions. Podcasts are an excellent means to convey information to the public, to include farmers, agribusinesses, extension personnel, congress, government personnel, and other stakeholders. We have proposed the production of a series of short podcasts to introduce aquaculture technology and other information focused on best management practices and feed reduction strategies for improving the efficiency of Nile tilapia culture, developed primarily in the Philippines. In this proposal we wish to produce an additional short series of podcasts to further convey ancillary CRSP activities in the Philippines and Indonesia. We propose to include impacts and accomplishments on sustainable polyculture technologies for culture of fish and shrimp, soft-shell crab farming, feed reduction strategies, integrative production systems, and value-added processing for milkfish and seaweed culture. An integral part of these podcasts will be the presentation of specific impacts that target indicators for the new Developmental Themes Advisory Panel reporting requirements of USAID.

We also propose to produce a series of podcasts on the accomplishments and impacts of research and training at other CRSP sites in Southeast Asia, Latin America, and Africa. Individual sites supported by the AquaFish CRSP address a range of research activities, building capability and sustainability into aquatic food production. Most but not all are elaborating technical approaches to sustainable Nile tilapia culture, such as the replacement of fish-meal based diets and other aspects of feeds technology, the phasing out of sex hormone treatments, initiation of polyculture methods, etc. Clean methods of aquaculture of other subjects are also under investigation, including new approaches to the production of African catfishes and oysters. At present, the sites of AquaFish CRSP research are introduced to the public primarily through a set of reports and material on websites. A Google search for "CRSP Aquaculture locations" reveals numerous websites including several that are based on previous CRSP activities; the net effect is a loosely integrated overview of AquaFish research sites. The relevance to current activities remains uncertain in a proportion of the URLs that turn up this way. We propose that a set of podcasts be assembled to present an easily accessible collection on the AquaFish CRSP website. Our goal in this series is to display the presentation of singular accomplishments at each of the sites, but more importantly to provide a means for visitors to look through a range of them in a short period of time, to gain an overview of the scope of CRSP activities and their impacts. The intended effect will be a sense that the total of these activities is greater than the sum of their individual pieces - that the AquaFish CRSP has had and continues to have a meaningful impact on the ability of developing countries to produce their own aquatic foods.

Collectively, the proposed supplemental activities will accomplish two primary goals: 1) the promotion of technologies to improve incomes and livelihood of farmers and fishers, and 2) the assessment of the impact of research conducted under the AquaFish CRSP and previous CRSP programs. Additionally, the work should develop the use of an attractive, effective and user-friendly tool (podcasts) to present and explain CRSP programs and accomplishments tailored for, and targeted at major stakeholders including the general public.

Quantifiable Anticipated Benefits

1. An estimate of the number of tilapia farmers (and hectareage) that use feed reduction strategies developed by CRSP research and the impact the management strategy has on improving household incomes will be quantified.
2. An extension paper on feed reduction strategies and cost savings will be produced for the tilapia farming community in the Philippines. This will complement podcast modules that will be developed for tilapia-related culture activities in the Philippines under current CRSP activities.
3. Increase the number of tilapia farmers who will adopt feed reduction strategies and improve household incomes. We anticipate through surveys and a workshop that 60+ farmers and stakeholders will receive direct training in improved feed management practices for tilapia culture.
4. Enhance capacity building and training on more environmentally sustainable milkfish farming to 6-8 communities and 80+ farmers, extension personnel, stakeholders, and government officials across a broader geographical range in the Philippines. This includes training on value-added processing of milkfish and integrated milkfish production systems aimed to enhance women's participation in aquaculture activities and income opportunities of households.
5. Identify and estimate the number of farmers and communities (and total hectareage) that have incorporated seaweed polyculture as a more sustainable technique for growing shrimp and fish in coastal regions of Indonesia. We anticipate that 40-50 farmers will also receive direct training on sustainable culture aquaculture through surveys.
6. Determine if household welfare and incomes have improved with the adoption of more sustainable coastal aquaculture techniques in Indonesia.
7. 2 - 4 podcast modules will be produced showing the impact of CRSP activities in the Philippines and Indonesia to include impacts of seaweed polyculture and sustainable production of shrimp/ fish in coastal aquaculture systems and of milkfish culture, integrated productions systems and value-added processing.
8. Six short podcast modules on major activities and impacts of CRSP-sponsored research at other major sites in Africa, Latin America and South/Southeast Asia will be produced and posted on the AquaFish CRSP website for dissemination to farmers, agribusinesses, extension personnel, congress, USAID and other stakeholders.
9. Essential data sets, additional training, and tools will be collected and developed to improve upon and enhance the reporting of specific indicators for the Developmental Themes Advisory Panel and the overall impact of USAID-CRSP

sponsored research activities. Likewise, key constraints to the adoption or expansion of best management practices should be identified that could improve USAID targeted activities.

10. Graduate and undergraduate students in both host countries and the U.S. will be trained in aquaculture extension including survey development and analyses and podcast production.

Research Design & Activity Plan

Location

Surveys, workshops and demonstrations will occur in communities of Central Luzon, Leyte, and Mindanao, Philippines and Aceh, Indonesia. Podcast will be produced at NCSU and NOAA from content (data, graphs, pictures) provided by host country and U.S. investigators in Africa, Southeast Asia, and Latin America.

Methods

1. Conduct Extension Activities and Assess the Impact of Feed Reduction Strategies on Tilapia Culture in Central Luzon, Philippines

An extension fact sheet will be produced showing the alternate feed reduction strategies and their cost savings. The extension paper will be shared with the farmers interviewed during the assessment portion of the study and will be available to anyone at the Fisheries and Aquaculture Center (FAC) at CLSU, the primary tilapia research and extension center in Luzon, Philippines. This extension paper will complement the development of a series of short Tilapia Podcasts that is currently planned where alternate feed reduction strategies along with other information on tilapia culture will be produced in electronic format with graphics/pictures/photos for access by the tilapia farming and research/extension community.

We will conduct an assessment study through survey and interviews of households within the primary tilapia farming communities of Central Luzon, in Regions II (Cagayan Valley which comprises two landlocked provinces, Quirino and Nueva Vizcaya) and III (Central Luzon which comprises six provinces: Bataan, Bulacan, Nueva Ecija, Pampanga, Tarlac and Zambales) (BFAR, 2007). Respondents for this survey work will be obtained from the list of registered tilapia farmers from local government units in these regions and will be randomly chosen. The survey questionnaire will be developed and analyzed using the guidelines of Fink (2003) and methods of Edwards (2006) previously used in obtaining general socioeconomic data from rural tilapia farmers in this region. The survey instrument will be developed and reviewed among project personnel (U. Hatch, R. Bolivar, R. Borski) and the Philippines Bureau of Fisheries and Aquatic Resources (BFAR) staff to determine both the extent to which feed reduction strategies are used by farmers and if so, how they are modified to meet specific circumstances; whether household incomes/welfare have improved or not, and the risks and constraints to adopting alternative feeding strategies. Personal visits to farms will document the farm operation/management method, and each farmer-respondent will complete the survey questionnaire, with assistance as needed by project staff at CLSU through the personal interview. Survey analyses will quantify the adoption rate, number and percent of communities, amount of water hectare, etc. impacted by alternate feeding strategies developed by CRSP and provide recommendations for how the government might facilitate the adoption of cost containment feeding strategies.

Additionally, we will conduct a training session through a farmer's day workshop at CLSU to show and train farmers (30+), feed manufacturer's, other stakeholders and government officials on the alternate or "moderate" feeding strategies as well as on

more sustainable and least cost formulation of tilapia diets. The results of the survey and any recommendations to improve cost containment feeding strategies and least-cost formulations for tilapia farming will be presented.

2. Enhance Capacity Building, Training and Impacts of Alternative Feeding Practices and

Integrative Production Systems for Milkfish Culture in the Philippines.

We propose to expand our workshop training on integrative production systems, feed management practices, and value-added processing for milkfish from Guimaras Island to other islands where milkfish culture is prominent; Leyte-Samar, Visayas and Davao, Mindanao. 30-40 farmers, government officials, local extension personnel, and other stakeholders will participate in a 2-day workshop in each of these regions directed by SEAFDEC staff, Evelyn de Jesus-Ayson, Maria Luhan, Marie Nievaes and Gwen Anuevo. Fisher folks and other stakeholders will be trained on milkfish biology, current feeding practices, alternative feeding strategies with cost savings potential, seaweed and sea cucumber culture and their integration into milkfish cage culture, and the environmental benefits and additional income opportunities with integrated milkfish production systems. Additional training will focus on value-added processing of milkfish (deboning, smoking, marinating) and will be geared to women to enhance their continued participation in aquaculture and opportunity to improve household welfare through sale of products in local markets. Farmers will be provided questionnaires designed to assess their current milkfish feeding practices, stocking densities, size of operation, and willingness and constraints (*i.e.* costs) to the integration of other economically valuable species into their culture system. This questionnaire will provide basic information on milkfish culture practices over broader regions of the Philippines. It will also provide information on the potential cost constraints and policies or support that may be needed for implementing more environmentally friendly polyculture of milkfish. Overall, the proposed activities will broaden the outreach and impact of CRSP research to regions and communities of the Philippines not previously exposed. It also will provide ways to reduce feed inputs and wastage and improve sustainability of milkfish farming. The expected net effects are to preserve the biodiversity of coastal fisheries and to enhance income opportunities for small-scale fishers.

3. Assessing Impacts of Polyculture Training on Fish and Shrimp Farming in Aceh, Indonesia

We have previously conducted several workshops on the incorporation of seaweed polyculture in the production of shrimp and fish in tambaks (ponds) in Aceh, Indonesia. Several farms have incorporated seaweed into the culture of shrimp and fish, but the degree of adoption and its potential to improve household welfare have not been quantified. To ascertain the rate of adoption of this more sustainable coastal aquaculture technique and potential improvements to household welfare, we will survey the farming community of Aceh, using procedures similar to those outlined for assessing impacts of alternate feed reduction strategies for tilapia farming (See 1. above). For this activity, an Indonesian graduate student fluent in Bahasa (University of Arizona) and a socioeconomics investigator from the U.S. (U. Hatch) will travel to Aceh, Indonesia. These individuals will work with H. Hasanuddin of Ujung Batee Aquaculture Center and K. Fitzsimmons and R. Borski to develop a structured interview and questionnaire, in order to assess the percentage of farmers and communities that have incorporated seaweeds in the culture of fish and shrimp. Another objective of this survey work is to define whether seaweed culture as resulted in improved welfare in affected households (e.g., whether they have better nutrition from seaweed consumption, environmental health) or incomes. Maps, water hectareage, the number of farms, and other information will be collated from detailed data available at Ujung Batee Aquaculture Center, as

previously collected in cooperation with numerous international and Indonesian government agencies and NGO's during the recovery period following the 2004 Tsunami. These data, along with survey analyses, will allow a quantification of the impacts of polyculture training in Aceh, and possible future efforts to improve adoption rates. This assessment should provide information on farm criteria, e.g., physical and socio-economic attributes which might make them more amenable to the adoption of polyculture. Subsequent extension efforts could then focus on these designated populations to enhance future technology adoption.

4. Podcasting to Promote Impacts of AquaFish CRSP Research and Outreach Activities

Raw materials to be used in podcasts will be gathered through electronic contact between project participants at North Carolina State University (NCSU) and at the NOAA facility in Milford CT. Both U.S. and Host Country Principal Investigators from the Philippines, Indonesia, and other AquaFish CRSP sites (Africa: Kenya, Tanzania; Asia: China, Cambodia, Vietnam; and Latin America: Mexico, Guyana) will be asked to gather available photographs and data, digital videos, and (later) to assist editorially in the blocking out of working guidelines and the refinement of drafts for each of the 6 podcasts. Digital video cameras will be loaned among project principal investigators, with a request that they record approximately one hour of information displaying their research activities and facilities, aquatic farming in their area, commercial and educational activities, markets, educational events. The US Lead Investigator will travel for short information-gathering visits to selected AquaFish research sites for establishment of an overview of key research impacts/ accomplishments and activities to be illustrated in the podcast series. Project personnel and student interns at NCSU, Central Luzon State University (CLSU), and NOAA will be trained in the nuances of podcast production in order to increase the depth and range of podcasts to be made available. They will learn the methods of assembling, editing, refining, and uploading effective material. These individuals will contribute to the incorporation of video and other materials into the production of a thematically-integrated series of podcasts. Podcasts will be uploaded onto the AquaFish CRSP and NCSU websites, the latter of which provides a quantitative method for tracking podcasts hits and downloads and consequently a sense of its utility as an outreach and communication method. While these podcasts will be produced in English, we anticipate that several will subsequently be translated into languages of the host countries to better serve as outreach tools.

Schedule

January 2010 – November 2010

Study 1: Collate data for cost containment/feed reduction strategies for tilapia culture, produce extension fact sheet. Relevant data for participating tilapia farmers (location, pond area, contact information) will be collected. Survey instrument will be developed and reviewed.

Study 3: Collect data on aquaculture operations in Indonesia, begin developing surveys for assessment of adoption of seaweed polyculture

Study 4: Collect information for podcasts for Philippines/Indonesia, podcast training.

August 2010 – February 2011

Study 1: Modify survey instrument, meet with in-country project counterparts. Identify survey teams for specific regions. Prepare an introductory explanatory letter to accompany the survey. Modify survey instrument if necessary. Randomly select farmers to be surveyed. Conduct surveys and survey analyses.

Study 3: Modify survey instrument, meet with in-country project counterparts, conduct surveys, and survey analyses.

Study 4: Continue podcast training, produce two podcasts for activities/impacts in Philippines/Indonesia, two for CRSP activities/impacts at Latin American sites, collect information for development of podcasts at additional CRSP sites

January 2011 - June 2011

Study 1: Conduct workshop for tilapia farmers/feed manufacturers, present results of survey, modify podcast for extension/impacts.

Study 2: Collect data for questionnaire/survey of milkfish farmers; conduct workshops/questionnaire for milkfish/seaweed culture, integrated production systems and value-added processing; produce podcast for extension/impacts.

Study 3: Produce additional podcast for Indonesia site.

Study 4: Produce 2 podcasts for Southeast Asia/Asia CRSP sites, collect podcast information for additional sites.

July 2011 – September 2011

Studies 1, 2, 3, 4: Modify existing podcasts to include additional impacts and to improve extension. Produce two podcasts for Africa activities/impacts. Write technical reports and CRSP final report.

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TOPIC AREA
QUALITY SEEDSTOCK DEVELOPMENT



TRAINING PROGRAM IN PROPAGATION AND HATCHERY MANAGEMENT OF TILAPIA
(*OREOCHROMIS NILOTICUS*) AND CATFISH (*CLARIAS GARIEPINUS*) IN GHANA

Quality Seedstock Development / Activity / 09QSD05PU

Collaborating Institutions & Lead Investigators

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Objectives

The long-term goal of this project is to ensure a sustainable supply of tilapia (*Oreochromis niloticus*) and catfish (*Clarias gariepinus*) fingerlings for the growing aquaculture industry in Ghana and promote private small- and medium-scale commercial fish hatcheries. The specific objectives of the project are:

1. Provide training for fish propagation, hatchery management, and fingerling production.
2. Educate and train prospective, beginning and existing farmers on fish propagation and hatchery management.
3. Develop fish life cycle and reproductive strategies brochures for use as reference materials.

Significance

There is a policy of increased government support for aquaculture development in Ghana. Aquaculture has the potential to create new jobs and improve food security among poor households. Fish consumption per capita in Ghana is estimated to be as high as 20 kg compared to the global average of 13 kg per capita. Although fish is on high demand nationwide, consumption is greatly limited by insufficient production and supply. The Ghana government continues to import an additional US\$200 million worth of fish to meet the ever increasing domestic market. Technical problems associated with fish production, particularly the lack of quality fingerling supply has limited the supply of market size fish to meet increasing demand.

Some of the major setbacks to the production of fish to meet the increasing fish demand in Ghana are limited hatchery facilities, lack of proper management in hatcheries, and lack of quality fingerlings. The absence of quality fingerlings is seriously affecting small- and medium-scale commercial fish farms, with a considerable number of farmers continuing to rely heavily on fingerlings harvested from the wild. The few well established hatcheries are unable to cope with the rising demand for fingerlings. The hatcheries are either owned by the Ghana government or large commercial cage aquaculture operators, and they are focused on supplying

large tilapia cage operations on the Lake Volta through contract production. They do not have enough fingerlings to meet the demand from small- and medium-scale fish farmers. Thus, there is the need to train prospective, beginning and existing aquaculture farmers on fish early life cycle development and management to enable more investment into private small- and medium-scale commercial fish hatcheries to meet demand from that sector of the aquaculture industry.

This activity is also important because it is in pursuit of the national agenda to ensure responsible fisheries, food security and poverty reduction through fish farming to meet domestic fish consumption. Therefore, there is a need to focus on hatchery production and management, and fingerling production. This requires considerable education and hands-on training to expose fish farmers and potential fish farmers to early life cycle stages of fish.

It is anticipated that the training will bring together a body of fish farmers who will acquire scientific knowledge and information from experts in various aspects of hatchery location and design, construction, operation and management. It is further hoped that the knowledge acquired by the trainees (farmers) will be used to make considerable contributions to the production of fry and fingerlings for the benefit of the small- and medium-scale sector of the aquaculture industry in Ghana. The farmers will also have brochures that they can always use as a reference guide in their operations.

Quantifiable Anticipated Benefits

After participating in this activity, participants will be able to:

1. Name stages of the fish life cycle and identify the progression from egg, larval fish, fry, juvenile, to adult.
2. Describe natural and artificial reproductive strategies of fish
3. Contrast the reproductive strategies of tilapia and catfish.
4. Participants will have real-world, hands-on learning in fish early life cycle development management from spawning through the fingerling production stages.
5. Personnel from Purdue University will gain insights into international and intercultural fish farming experience that will inform their teaching and research in the US.

Research Design & Activity Plan

Activities will include 1) reviewing and synthesizing literature related to early life cycle of fish species, particularly tilapia and catfish, 2) conducting a training program in early life cycle development of tilapia (*Oreochromis niloticus*) and catfish (*Clarias gariepinus*), and 3) establishing the curriculum framework for continued training programs at WRI-ARDEC, Akosombo. The training program will cover the following cycle stages:

1. Spawning / Egg production: This will include natural and artificial reproduction strategies. Participants will also learn threats to eggs from changes in water temperature and oxygen levels, flooding or sedimentation, predators and disease.
2. Larval / Fry fish production and management.
3. Hatchery management from egg, larval fish, fry, juvenile, and consequently grow-out into market size adults.

The first step will be to identify subject area specialists and avenues for promoting the training program. Training will be done through a mix of theory-based and practical workshops. Two workshops will be held; the first will be held at the WRI-ARDEC, a government research facility

located at Akosombo for farmers from the Greater Accra, Eastern and Volta regions and the second will be held at the Pilot Aquaculture Center (PAC) in the Kumasi, which is the official government centre for Aquaculture training in Ghana for farmers from Brong-Ahafo, Ashanti and Western regions. The workshops will be offered by experienced aquaculturists and academics who know the challenges of early life stages of fish. The target audience is prospective, beginning and existing farmers (or investors) interested in establishing their own fish hatchery. The workshop setting will promote interactions among participants to allow them to benefit from the sharing of real-life fish farming experiences and also gain skills and ideas. It is also anticipated that other stakeholders such as aquaculture researchers, extension workers, administrators, scientists from governmental agencies, and representatives of NGOs will participate in the program.

The workshops will be co-sponsored by AquaFish CRSP, Kwame Nkrumah University of Science and Technology, Fisheries Directorate, and the Water Research Institute, in consultation with the National Aquaculture Farmers Association as well as Moi University in Kenya. These three institutions have suitably qualified and experienced personnel to handle the issues of hatchery and fry management and fingerling production. It is the hope that participants will cover their own expenses to participate in such a training program, but we recognize that some farmers cannot pay their travel expenses and will be supported from funds for this program. Government and industry sponsorships will also be sought to keep attendance costs as low as possible to allow maximum participation. Participation and sponsorship fees will be used to offset programming costs.

Impact Indicators

- A. Number of fish farmers receiving timely knowledge and management capabilities.
- B. Number of training participants (prospective, beginning and existing farmers, aquaculture researchers, extension workers, scientists from governmental agencies, and representatives of NGOs).
- C. Number of institutional collaborations on aquaculture in Ghana.

Schedule

Reviewing and synthesizing literature on early life cycle of tilapia and catfish	September – November 2010
Conducting a training program at WRI-ARDEC, Akosombo	November 2010
Conducting a training program at Pilot Aquaculture Center (PAC), Kumasi	March 2011

TOPIC AREA
FOOD SAFETY & VALUE-ADDED PRODUCT DEVELOPMENT



ASSESSING THE IMPACTS OF SUSTAINABLE FRESHWATER AQUACULTURE AND
SMALL-SIZED/LOW-VALUE FISHERIES MANAGEMENT IN THE LOWER MEKONG
BASIN REGION OF CAMBODIA AND VIETNAM

Food Safety & Value-Added Product Development/Study/09FSV03UC

Collaborating Institutions & Lead Investigators

University of Connecticut-Avery Point

Robert S. Pomeroy
Sylvain De Guise

Inland Fisheries Research and Development Institute (Cambodia)
Can Tho University (Vietnam)

So Nam
Le Xuan Sinh

Objectives

The objective of this study is to assess the impact of the investigations in the AquaFish CRSP project “Development of alternatives to the use of freshwater low value fish for aquaculture in the lower Mekong basin of Cambodia and Vietnam: implications for livelihoods, production and markets” on both the private and public sectors of Cambodia and Vietnam.

Significance

In the Mekong region, many capture fisheries resources have been largely overexploited and, as a result, development of aquaculture has been encouraged to provide the protein, income, employment and export earnings for some countries. Such a development trend implies that sufficient feed for aquaculture production will be available. One source of feed is low value/trash fish (Low value/trash is defined as fish that have a low commercial value by virtue of their low quality, small size or low consumer preference). There is increasing demand and trade in the lower Mekong region of Cambodia and Vietnam for low value/trash fish for (1) local consumption (e.g. fresh, dried); (2) direct feed (e.g. livestock, high value species aquaculture); (3) fish meal production (e.g. poultry, aquaculture); and (4) value-added products (e.g. fish sauce, fermented fish, fish paste, smoked fish).

There is an increasing conflict between the use of small-sized/low value fish for animals/fish and for human consumption. Supplies of small-sized/low value fish are finite, and as indicated by a recent increase in price, i.e. demand is outstripping supply. Small-sized/low value fish are important to the communities and aquaculture, as well as the ecosystems in Cambodia and Vietnam. There is a need to support the development of a policy and management framework to address aquaculture and capture fisheries interactions.

The larger research project, of which this investigation is an add-on, will produce a number of outcomes, including development of a plant based feed for snakehead fish, recommendations to government and the private sector for a sustainable snakehead aquaculture industry, value-added products from small-sized/low value fish such as fish paste and fish sauce, extension/outreach technologies, recommendations for

improvements in the marketing system for both capture and culture fish in the region, and recommended policies to improve management of small-sized/low value fish in the Mekong area. These outcomes will impact both the private and public sectors through improvements in technologies, commercialization of new products, sustainable aquatic resource management practices, and policies for aquaculture and capture fisheries. It is expected that the outcomes will have an impact on improving food security, livelihoods, poverty and economic development in the region.

Information is needed to identify and measure the direct and indirect impacts of the research, the communication/ dissemination of technologies, and human capacity development resulting from the project. .

Quantifiable Anticipated Benefits

- 10 undergraduate and graduate students in Cambodia and Vietnam will be trained and have experience in undertaking impact assessment of agricultural and resource management research.
- 10 scientists and academics in Cambodia and Vietnam will be trained and have experience in undertaking impact assessment of agricultural and resource management research.
- 8 provincial fisheries officers will be trained and have experience in undertaking impact assessment of agricultural and resource management research.
- 1 US graduate student from the University of Connecticut, Department of Agricultural and Resource Economics, will be trained and have experience in undertaking impact assessment of agricultural and resource management research.

Research Design & Activity Plan

- a. **Location of work:** The study will be implemented in project areas in both Cambodia and Vietnam.
- b. **Methods:** Agricultural research generates many types of outputs. These include technologies embodied in a physical object (e.g., improved feed), management tools and practices, information, and improved human resources. Impact assessment is a process of measuring whether or not research has produced its intended effect—that of meeting development objectives, such as increases in production and income and improvements in the sustainability of production systems. It is important to demonstrate that the changes observed are due to a specific intervention and cannot be accounted for in any other way. The effects can be measured at the household, target population, national, and regional levels. Impact assessment to be undertaken in this investigation will be of two types: ex-post and concurrent. The ex-post assessment refers to the evaluation made upon the completion of a project to determine achievements and to estimate the impact of research. Returns to investment in research and development are typically assessed using the ex-post concept. These studies also help to understand the process of disseminating technology and the constraints to its adoption. Concurrent assessment or evaluation is done to identify the impediments for larger adoption of the research outputs. The purpose of a concurrent evaluation is to correct the gaps and provide feedback for refining and tuning the technology as per the stakeholders' requirements. Often it is known as constraint analysis.

Four components determine the adoption of a technology: technology traits (e.g. duration, quality, etc.), policy environment (e.g. price support, procurement, etc.), institutional arrangements (e.g. seed supply sector, credit availability, etc.), and

infrastructure (e.g. markets, roads, power, clean water, processing facilities, etc.). Determining constraints for larger adoption forms a part of the impact assessment.

As reported above, this project will produce a number of outcomes. This project will have direct, indirect, and intermediate impacts in the two countries. Direct impact refers to the impact on the welfare of people and the environment as a result of adopting a technology. It is reflected mainly as an increase in productivity, a reduction in per unit cost of production, and/or reduced pressure on expansion into fragile ecosystems because of improvement in technology. Indirect impact includes flow-on impacts to other activities. Intermediate impact refers to increases in the knowledge base that could subsequently have a direct impact.

This investigation will be implemented through four activities:

1. Two trainings will be conducted on impact assessment methodologies for students, scientists, academics and fisheries officers (one each in Cambodia and Vietnam). The focus of the trainings will be on methods for conducting agricultural impact assessments, specifically adoption studies and economic studies (see below) to be utilized in the actual impact assessment of the investigations in the larger research project. The training will result in an understanding of the methods by the participants and a work plan to undertake impact assessment of the investigations in the project.
2. A team of 2-4 participants from the trainings will be assigned to undertake an impact assessment of one of the investigations for the larger research project in each country. The training will have resulted in a work plan that will be implemented over a five month period. The work plan will include steps for preparatory activities (goals, process, study area, parameters, stakeholder consultation); secondary and field data collection; and data analysis.
3. Following data collection and analysis, impact assessment reports will be prepared by the team on each investigation studied.
4. Impact assessment reports will be presented to USAID, AquaFish CRSP, government, academia and other interested groups. Where appropriate, scientific journal articles will be prepared and presentations made at professional meetings.

The specific impact assessment methods to be utilized in this investigation are adoption studies and economic studies. These are both considered to be partial impact assessment studies. Adoption studies look at the effects of new technology on farm productivity and farmers' welfare. Economic studies look beyond mere yield and crop intensities to the wider economic effects of the adoption of new technology. These studies generally estimate the economic benefits produced by research in relation to associated costs and estimate a rate of return to research investments.

Adoption studies generally trace the results of plant and animal innovations from the research station or on-farm trials through networks of adopters. This type of evaluation will analyze the underlying patterns of adoption and the use of new practices and will utilize statistical and econometric tools. Adoption surveys of snakehead farmers will be used to identify adopters, to see if farmers are using (or not using) improved technology and to look at its effects on farm production, to measure client satisfaction (or dissatisfaction) of research results, to measure threats to disadoption, to identify relative attributes of the new technology that point to trade-offs in sources of benefits, and to determine how research activities can be reoriented to make technologies more useful (CIMMYT 1993). The adoption study will attempt to determine why the feed technology is or is not being used and to

compare the benefits of old (small fish) versus new technologies. An excellent extract of the relevant literature on the theoretical concepts and factors underlying adoption of improved technology and the process of innovation has been put together by Kuby (1999). The data and results of the adoption study will provide the baseline data for the evaluation of technology impacts on productivity, income, environment, equity and other goals. Analysis of constraints to technology adoption are also important in increasing research efficiency, as well as in designing policy, programs and institutional reforms to enhance technology adoption. Aside from generating the raw material for estimates on adoption and contributing information on benefit estimation, adoption surveys can uncover human interest stories that have potential use in raising public awareness.

Economic studies include studies that estimate economic benefits and measure economic rates of return associated with innovations coming out of research investment. An economic study at the research program level helps to identify what the technical and economic outcomes of the program were and help identify why such outcomes were achieved; and assess the welfare effect of the program by comparing it with the level of welfare that would have arisen in the absence of the research project. Cost-benefit studies will be utilized in this investigation since they are most suitable at the level of this type of individual research program (Evenson 1999). The economic study will strive to assess all the benefits and costs associated with the research program including: (1) direct financial impacts through changes in input costs (quantity and/or prices), output price (including quality effects) and quantity produced; (2) the costs and revenue of products or activities replaced by the new technology; (3) indirect financial and economic impacts through other spillovers; (4) externalities through benefits and costs imposed on others; and (5) environmental benefits and costs.

Schedule

This study is planned to be implemented as below:

Activity	Beginning	Ending
Training on impact assessment methods: Training 1: Can Tho 14 participants (5 students, 5 scientists and academics, 4 fisheries officers) Training 2: Phnom Penh 14 participants (5 students, 5 scientists and academics, 4 fisheries officers)	10/1/2010	12/30/2010
Implementation of impact assessment projects	1/1/2011	05/30/2011
Impact assessment report preparation	05/30/2011	08/30/2011
Publication and presentation of impact assessment reports	08/30/2011	09/29/2011

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TOPIC AREA
TECHNOLOGY ADOPTION & POLICY DEVELOPMENT



EFFECTS OF ACRSP AND AQUAFISH CRSP INITIATIVES AND ACTIVITIES ON
AQUACULTURE DEVELOPMENT IN KENYA

Technology Adoption & Policy Development /Study/09TAP07PU

Collaborating Institutions & Lead Investigators

Purdue University

Kenyatta University

Kwamena Quagrainie

Kenneth Foster

Charles C. Ngugi

Objectives

Activities proposed in this study will take an in-depth ex-post assessment of the impact of ACRSP-funded activities at the micro (farm and household) level as well as the macro (governmental and policy) level in terms of practices and policy recommendations, i.e., technology- and policy-oriented assessments. Qualitative ex-post impact assessment methods will be conducted from the perspective of cost-benefit analysis using feasible empirical indicators of influence or adoption. ACRSP intervention strategies should also translate into increased efficiency in fish farming and resource productive capacity. The specific objectives of the study are:

1. Assess the technical efficiency of fish farms adopting ACRSP knowledge and technologies
2. Assess the determinants of adoption of ACRSP technologies
3. Assess the extent to which ACRSP's initiatives and activities meet the objectives of contributing towards poverty alleviation in rural communities, protein enhancement, and reducing pressure on capture fisheries.

Significance

Until recently, aquaculture in Kenya was a little known and young industry in spite of fish culture practices that had existed for decades. Kenya's aquaculture consists mainly of small-scale commercial tilapia (*Oreochromis niloticus*), and also catfish (*Clarias gariepinus*), producing only about 1,000 metric tons annually until the early 2000. In 2007, however, the government reported annual production from aquaculture of about 4,220 metric tons of fish valued at KSh 500 million. The Government of Kenya has recognized aquaculture as a sub-sector with great potential to contribute towards poverty alleviation in rural communities, dietary protein enhancement, and reducing pressure on capture fisheries. The government is supporting the industry with KSh 1.1 billion under a program called 'Fish Farming Enterprise and Productivity Program.'

Improving fish farming technology is important to the economic growth and development of rural communities. Improved technologies are necessary to help fish producers respond to changing circumstances and raise productivity and household incomes. Technical change with the adoption of new technologies in fish farming is

particularly important in Africa to enable sustainable economic growth and increased production with minimal resources.

Earlier ACRSP work recognized that increased production from fish farming required appropriate technical support and intervention strategies in the form of extension services and research technologies to increase productivity. Improving technical knowledge and the efficiency at the farm level can increase fish production in a more cost-effective way than introducing new technologies. One intervention strategy that ACRSP adopted in Kenya in 2000 was addressing the constraints of availability of quality juvenile fish for stocking private ponds. The use of poor quality juveniles usually resulted in poor survival at the early stages of fish propagation. Juvenile mortality was as high as 90%. ACRSP also focused on addressing the availability of farm records for use in farm business planning. Lack of adequate records on fish farming prevented lending institutions from taking serious interest in investing in aquaculture.

ACRSP and AFCRSP embarked on a series of training programs that taught record keeping and various hatchery techniques and nursery management that could be adopted by farmers for producing fish for the food market as well as for producing baitfish for the commercial fisheries. Results from this study would be useful for maximizing aquaculture production without necessarily adopting new technologies. The information obtained will both demonstrate the importance of agricultural research to achieving development outcomes, and also help to target future resources to achieve greater impact.

Quantifiable Anticipated Benefits

1. Empirical assessment of farm level production with improved knowledge and technologies from AFCRSP activities.
2. Identification of factors influencing fish production and economic efficiency of AFCRSP technologies.
3. Insights into efficiencies in fish production that will enable further targeted interventions by AFCRSP and government policy.
4. Insights into any need for policy and improvement in fisheries extension to enhance delivery of improved technologies to fish farmers.
5. Support 2-3 graduate students from a Kenya partner institution to conduct thesis research on technical and economic efficiency of fish farms.
6. Personnel from Purdue University will gain insights into international and intercultural fish farming experience that will inform their teaching and research in the US.

Research Design & Activity Plan

Adoption of ACRSP knowledge and technologies may be influenced by a number of factors such as local economic, social and environmental factors, as well as spatial characteristics. For example, Quagraine et al (2009) reported that farmers in the Western province have a 19% more probability of using credit facilities for their fish farming operations than farmers from the Rift Valley, Central and the Eastern provinces. Market proximity, transport infrastructure, land and labor availability, and other local ecological characteristics could also help explain adoption practices and consequently, productivity (Nelson 2002, Mertens et al 2002).

Techniques for the measurement of technical efficiency of production can be broadly categorized into two approaches: parametric and nonparametric. The most popular among empirical analysts are the parametric stochastic frontier production function (SFP) approach (Aigner, Lovell and Schmidt, 1977) and the nonparametric mathematical programming approach, also called the data envelopment analysis (DEA) (Charnes, Cooper and Rhodes, 1978; Färe et al., 1989; Färe, Grosskopf and Lovell, 1994). The main strengths of the SFP approach are that it deals with stochastic noise and permits statistical tests of hypotheses pertaining to production structure and degree of inefficiency. However, the methodology requires imposing an explicit parametric form for the underlying technology and distributional assumption for the inefficiency term (Coelli, 1995; Sharma, Leung and Zalleski, 1999). The main advantages of the DEA approach are that it avoids parametric specification of technology and any distributional assumption for the inefficiency term. However, DEA is deterministic and attributes all the deviations from the frontier to inefficiencies, consequently a frontier estimated by DEA is likely to be sensitive to measurement errors and other noise in the data (Coelli, 1995).

In assessing the extent to which ACRSP's initiatives and activities have contributed towards poverty alleviation in rural communities and protein enhancement, the study will assess changes in farmers' incomes and revenues of selected aquaculture-related businesses to reflect changes in the welfare of rural households. In addition, other indicators such as the wealth, and the level of consumption expenditures for nutritious food, housing, vehicles, and other items will be used to assess the economic well-being of rural households.

The data for this study will come from a survey that will be jointly designed and administered by the Ministry of Fisheries Development, Moi University, Kenya and Purdue University, USA. Smallholder fish farmers who have participated in the ACRSP programs in the Western, Central, Eastern, and Rift valley provinces of Kenya will be surveyed. These regions produce much of the farm raised fish in Kenya because of relatively better annual rainfall and distribution. A sampling frame of all fish farmers in the regions will be generated randomly using a simple random sampling technique. The survey questionnaire will be administered face-to-face by visiting the farmers. The questionnaire will solicit information on their practices prior to participating in any ACRSP program as well as their current practices relating record keeping, brood stock handling, hatchery, nursery, stocking, feeding, pond management, harvesting, and marketing. Data will also be collected on production levels, input use, socio-economic characteristics, and average input and output prices in the region. A structured questionnaire will be used to collect these data.

Impact Indicators

- a) Percentage increases (or decrease) in fish farming productivity with adoption of ACRSP knowledge and technologies.
- b) Number of fish farm that adopted ACRSP knowledge and technologies and their distribution of technical efficiencies.
- c) Number of determinants of economic efficiency of fish farms.
- d) 2 - 3 M.Sc students will graduate.

Schedule

Literature Review, Survey Design and Pre-tests	January – April 2010
Survey pretests and refinements	May - June 2010
Survey administration	July – November 2010
Data analysis	December – May 2011
Reports & Distribution	June - July

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TRAINING TRAINERS FOR LONG TERM AND SUSTAINED IMPACT OF POND
AQUACULTURE IN AFRICA

Technology Adoption & Policy Development/ Activity/09TAP08AU

Collaborating Institutions & Lead Investigators

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Jeff Terhune

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Claude Boyd

Karen Veverica

Kenyatta University (Kenya)

Charles C. Ngugi

Network of Aquaculture Centres in Asia-Pacific (Thailand)

Yuan Derun

Objectives

1. Develop a team of 6 highly-qualified trainers who have a wide range of field experience.
2. Make CRSP research results more accessible and applicable to the African context through the production of fact sheets based upon CRSP technologies.
3. Conduct basic principles and practical applications training programs in selected African countries for technical advisors, prospective fish farm managers and advanced farmers (total 60 individuals)

Significance

The sustained growth and development of pond aquaculture in Africa depends on a well-prepared cadre of researchers and consultants who are conversant with technical fundamentals and best practices (Machena and Moehl 2001; Molnar et al. 1996). Ensuring and renewing the skills of aquaculture professionals will advance productivity and protect the investment made by private landowners, governments, and donors in fish farm development. Most nations in which aquaculture plays a significant role in poverty alleviation recognize that human resource capacity renewal is a key to sustained development of the sector. The types and levels of training required must focus on basic principles. Yet the need to increase specialized training in the aquaculture sector will grow as the sector develops (De Silva et al. 2001).

In the past, extension workers were expected to have specialized knowledge in technological aspects, such as the artificial propagation of an aquatic species. The training for this, however, tended to be relatively short term, lacking a holistic approach. Such training was frequently driven more by technology interests than by development needs or the needs of the farmers. A holistic approach to aquaculture training is, and will continue to be, an essential ingredient of sustainable aquaculture development (De Silva et al. 2001; Veverica and Molnar 1997).

During the First International Meeting for the Education in Fisheries and Aquaculture that took place in November of 2009 in Bangkok, Thailand, a non-degree training program that could provide a global view of aquaculture founding principles was identified. Auburn University had just embarked upon creation of a distance learning program called Certification of Aquaculture Professionals (CAP). With this demonstrated need, AU has invested over \$100,000 in its production. The program, based upon the lectures from Auburn's Aquaculture Training Program, was delivered to

fish farming extensionists and farm managers in Puebla State, Mexico. Participant assessments indicate that the program offered much more than expected and is very practical. The most frequent criticism is that the program is not accompanied by a practical training program in the field.

Several subjects covering the fundamentals of aquaculture have been divided in modules. The complete certification consists of ten modules and a total of 136 segments with an average duration of 30 minutes each (see Appendix 1. for detailed content):

Module	Instructor	Segments
Principles of Aquaculture	Leonard Lovshin	10
Water Quality	Claude E. Boyd	16
Physiology	Imad P. Saoud	9
Hatchery Management	Ronald Phelps	20
Aquatic Animal Nutrition	D. Allen Davis	12
Genetics and Breeding	Rex Dunham	17
Aquatic Health	Jeff Terhune / Karl Hayden	17
Aquaculture Production	Masser / Daniels / Veverica	21
Extension Methods	John Jensen	5
Aquaculture Economics	Terril R. Hanson	9
TOTAL		136

Aside from a computer and internet connection, no special technology is required to gain access to the CAP modules. The participant is assigned a user name and selects a password. The student can access the learning modules at any time. Users with slower internet connections will be able to view the slides, listen to the lectures in English and view the translated lecture transcripts. Those with faster internet connections will be able to view the lecturer as well.

Trainers in Africa are mostly university or technical school personnel who have higher degrees in subjects other than aquaculture or who have had course work in aquaculture without ever actually producing any fish. Few teaching institutions in Africa have access to field facilities that allow for further development of aquaculture skills of a level that would be useful for development of commercial aquaculture.

This activity seeks to provide to a selected group of current trainers (termed Master Trainers), 1) a complete online course that provides the fundamentals of aquaculture for their further use in developing their own courses, as well as a Certificate of Aquaculture Professional, and 2) hands-on field experience in pond-based fish production to accompany the CAP and to allow the Master Trainers (MT) to experience all of the techniques they will be teaching others. As the CAP program is completed by the prospective MT, a 1 month field exercise will be conducted in the US.

The hands-on field experience will be conducted at Auburn, EW Shell North Auburn Fisheries Research Station, of 800 hectares, with more than 300 ponds, totaling 100 ha of water surface. Karen Veverica will coordinate the activities. A set of 400 sq meter ponds will be allocated to set up a small feed trial, a tilapia spawning pond for production of uniform-age fry, and fry grow-out. Hatchery facilities will also be made available. In addition, the trainees will be able to get fish marketing experience, fish harvesting

experience for small (<1 ton) and medium scale (>1 ton per harvest), fish transport experience, hatchery experience and general pond management practice. The MT will come to Auburn as “visiting scientists” and will be allocated the “Aquaculture training room” where they will have computer access. Time will be provided for the review of CRSP research publications and the production of fact sheets. Each MT will produce 2 or more fact sheets that will be reviewed and printed. Auburn Professors Boyd, Phelps, Terhune and others will conduct one-hour question and answer sessions with the visiting scientists.

Following this Master Trainer program, the trainers will be contracted to hold one-week sessions in their own country, at a location they select. Each will be given a budget with which to finance the training. They will be assisted in the training by K. Veverica, Dr Charles Ngugi of Kenyatta University and Yuan Derun of the Network of Aquaculture Centres in Asia-Pacific.

Additive buy-ins may pay the marginal costs of additional program participants and are being considered by other agencies. FAO may sponsor a trainer and the World Bank may provide for further training of prospective fish farm managers. Namibia is considering adding candidates to the master trainer program (CAP plus field work at Auburn). An announcement of the Master Trainer opportunity for training will be sent out via ANAF and SARNISSA and to Aquafish CRSP collaborators.

Selection of the MT candidates will be based upon recommendations from: CRSP collaborators; department heads at Legon University, the Kwame Nkrumah University Of Science and Technology, and the Ministry of Fisheries in Ghana; Sokoine University. Tanzania Fisheries Research Institute, and the Ministry of Fisheries in Tanzania; and from Makerere University, the Fisheries Training Institute, and Gulu University in Uganda.

The selection process will be competitive as it would be for scholarships. The basis for selection of the 2 candidates per country will be the long-term training potential of the candidate, either through their appointment as instructor in aquaculture or their continued training of fish farmers and managers. Final approval will come from the AquaFish CRSP management entity.

A maximum of 2 Master Trainer candidates from Ghana, 2 from Tanzania and 2 from Uganda will be considered for the scholarships. Private consultants acting as fish farmer advisors will be allowed to compete for the scholarships. The in-country trainees will be selected by the Master Trainers, with owner-operators and those currently employed in fish farm management being given priority.

Quantified Anticipated Benefits

- The 6 “Master Trainers” will be selected individuals who conduct research and/or teach students in aquaculture or who currently train farmers, and who would train future farm managers.
- The 2-week in-country trainees will be selected from the population of serious and competent individuals who provide technical assistance and farm management advice to pond-based fish farmers in Ghana, Uganda, and Tanzania. The training activities will foster practical hands-on knowledge of pond construction, water management, fertilization, feeding, fish health management, harvest, record-keeping and marketing for those working with commercial fish farmers in medium- and small-scale production enterprises.
- At least 12 aquaculture training fact sheets will be prepared by the MT under the supervision of Karen Veverica during the program at Auburn and will be used to

conduct the training events in Africa. These materials will be made available through the ANAF website sponsored by FAO and the Aquaculture and Fisheries CRSP web sites. The fact sheets will be duplicated and distributed during the training sessions as well. Each fact sheet will list reference materials for further reading that is readily available to African readers who have access to the internet.

Suggested indicators are:

- Number of Master Trainers receiving training in the United States.
- Number of people trained by Master Trainers in host countries (disaggregated by gender and by type: university student, technical school student, fish farm manager, fish farm owner)
- Number of fact sheets published

These are indicators that can be measured by the end of September 2011. However, there will be further benefits beyond this time, including the potential revision of university courses in aquaculture, further training funds being made available from other sources, based upon the feedback generated from the in-country training events, and measurable increases in fish farm productivity or profitability.

Activity Plan

1. Selection of the 6 participants in the Master Trainer Program with counsel from CRSP investigators in Ghana, Tanzania, and Uganda and final decision reserved for the CRSP management entity
2. Participants begin the online CAP program in April 2011. If they could not complete all of the units before the US training takes place, they will need to spend some of the evenings on these units while in Auburn because the field training will build upon the CAP.
3. Trainet formalities completed for each of the 6 MT participants, by beginning May 2011.
4. 6 prospective Master Trainers travel to Auburn AL, where they will assemble the CRSP information into fact sheets and conduct field exercises geared towards increasing their experience in fish handling, sampling, harvesting, water quality analyses, fish health management, feed evaluation and management and hatchery management.
5. Each MT will prepare at least 2 fact sheets on CRSP-related research while at Auburn.
6. Master Trainers return to home country and organize training events. They select the training site, make up the schedule and prepare the budget.
7. Training events organized and carried out in Ghana, Uganda and Tanzania. Two one-week sequenced training events to consolidate and reinforce the technical capacity of aquaculture trainers. Successful participants will receive a certificate either of achievement or attendance if they cannot meet the knowledge and performance objectives of the training activity.

Staffing for in-country training

- Karen Veverica (Auburn University) will serve as training coordinator in Auburn and as primary training advisor for the events in each country. Two Auburn graduate students will assist the Auburn training activity.

- Charles Ngugi (Kenya) will provide expertise in pond construction and fish handling for the first training program in each country. He will be retained via personal services contract.
- Yuan Derun will provide training expertise in the area of rice-fish culture, and in fertilizer-based aquaculture production for the second training program in each country. He will be retained via personal services contract.

In-Country Training Week One: Basic Principles of Pond Aquaculture

- **2 Days prior:** Trainers arrive, planning meeting, review of local arrangements, course management and strategy discussions
- Day 1: Travel and assembly at training site, participant arrival day.
- Day 2: Principles of pond construction and renovation Pond Design and construction. Pond management –carrying capacity.
- Day 3: How to select the fish production system (ponds or tanks, liners or no, aeration or not). Tilapia biology and cage culture .
- Day 4: Carrying capacity at different production levels.
- Day 5: How to sample and harvest fish. Pond record keeping.
- Day 6: Basics of fish handling and transport.
- Day 7: Basic principles of fish marketing, Assessment by training team; participants depart.

In-Country Training Week Two: Basic Principles of Pond Aquaculture

- Day 0: Trainers arrive, planning meeting, review of local arrangements
- Day 1: Travel and assembly at training site
- Day 2: Water quality and fish production
- Day 3: Pond fertilization for zooplankton and for tilapia production
- Day 4: Feeding and feed management, Catfish Hatchery Management
- Day 5: Economics of feeding, and a comparison of fertilizer-based and feed-based fish production options.
- Day 6: Preparation of farm management plans. Revisit fish marketing and production planning.
- Day 7: Assessment by training team; participants depart.

In-Country Training Participation

The selection process also will seek to ensure that bona fide technical advisors, farmers and farm managers are included. The trainees will have to find their own transport to the training site. Lodging and meals will be provided but no daily allowances will be given.

The training materials will be developed as part of activity 4 and during the field training of technicians; the project will provide training materials to accompany the lectures, demonstrations, and practical exercises in farm ponds. Participants will have durable reference materials to guide pond construction, seed stock development, production, and marketing processes. The training modules will be made available on CD-ROM, and on CRSP, Auburn, and ANAF websites.

Regional and Global Integration

Karen Veverica will provide leadership and coordination in organizing all training sessions and in adaptation of training materials from the Auburn University CAP. Charles Ngugi and Yuan Derun will serve as trainers in Africa, preparing PPT training materials, and employing other resources developed under AquaFish CRSP projects.

The training events will contribute to strengthening AquaFish CRSP contacts in countries where we have previously interacted and provide opportunities for new contacts in countries that have not had prior relations with the AquaFish CRSP. Dr. Joseph Molnar will lead the monitoring and reporting of project activities, and the integration of the activity into the overall project. Mr. Billy Earle will conduct the financial administration of project activities. Dr. Claude Boyd will serve as alternative trainer in the event that key parties are not available and will review fact sheets that are related to water quality and pond soils. Dr. David Rouse will serve as a trainer and is the Head of the Auburn University Department of Fisheries and Allied Aquacultures. Dr. Jeff Terhune, and Dr Ron Phelps will assist in training at Auburn for the Master Trainer part of the program.

Schedule

	YR1	Project Year 2				Project Year 3			
		1-3	4-6	7-9	9-12	1-3	4-6	7-9	9-12
Recruiting and processing participants for Master trainer						x			
CAP program for trainers						x	x	x	
Auburn field training plus Preparation and adaptation of modules (June 2011)							x		
Recruiting and processing participants for in-country training						x	x		
Conducting training events in Ghana								x	
Conducting training events in Tanzania								x	
Conducting training events in Uganda								x	
Process & report participant assessments								x	

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Appendix 1. Detailed Topics for the Auburn University Certification of Aquaculture Professionals (CAP)

PRINCIPLES OF AQUACULTURE	WATER QUALITY MANAGEMENT	AQUACULTURE PRODUCTION	PHYSIOLOGY	HATCHERY MANAGEMENT	NUTRITION AND FEEDING	AQUATIC HEALTH	GENETICS AND BREEDING	EXTENSION METHODS
Fisheries and Aquaculture Statistics	Physical Properties of Water	Concept of Commercial Fish Production	Introduction	Introduction	Introduction	Disease Dynamics I Knowing the Fish and the Culture System	Goals Plans and environmental variations	Introduction
Aquaculture Definitions	Hydrology	Facilities/Types of Production Systems	Environmental Stressors	Gonad Development	Digestive Morphology	Disease Dynamics II Knowing the Diseases	Qualitative and Basic Genetics	Goals
Primary productivity in water	Dissolved Oxygen and Redox Potential	Site/Species/System Evaluation	Homeostatic control	Brood Stock Management	Digestive Process	Non-infectious diseases (Water Quality)	Strain Evaluation and Selection	Adoption/Diffusion
Carrying capacity in water: nutrient inputs	pH, Carbon Dioxide and Alkalinity	Production Planning-I	Receptor mechanisms	Environmental Control of Reproduction	Energy	Non-infectious diseases (Nutrition Deficiencies)	Inbreeding and Random Genetic Drift	Program Development
Carrying capacity in water: water quality	Microorganisms and Water Quality	Water Budgets	Gas exchange	Hormone Induced Spawning	Protein	Sample Collection and Preservation	Intraspecific Crossbreeding	Demonstration of how to do a Demonstration
Carrying capacity	Oxygen Production and Demand	Pond Preparation-Liming	Metabolism	Ovulation and Spawning	Lipids	Diagnostic techniques-I	Interspecific Hybridization	
Carrying capacity in water: species and polyculture	Nitrogen and Phosphorus	Pond Preparation-Fertilizing	Osmoregulation	Sex Manipulation	Minerals	Diagnostic techniques-II Video	Selection and Heritability	AQUACULTURE ECONOMICS
Growth rate	Sulfur and Trace Elements	Pond Preparation-Insect & Unwanted Fish Control	Excretion	Egg Development	Vitamins	Treatment theory	Correlated Response, Indirect Selection and Multiple trait selection	Budget Analysis
Yield and economics	Pond Liming	Aquatic Weed Management	Measures Stress.	Egg Handling	Dietary components	Vaccines	Polyploidy	Cost Return Analysis
Classification systems for aquaculture	Pond Fertilization	Water Quality Monitoring/Maintenance-DO ₂		Egg Incubation	Grow-out diets	Warm Water Bacterial Pathogens	Sex reversal and breeding	Economic Analysis
	Mechanical Aeration	Water Quality -pH, ammonia, etc.		Larval Management	Maturation and Larval Diets	Cold Water Bacterial Pathogens	Genetic Markers	Cash Flow Analysis
	Feed BOD and Aeration Requirements	Feeds and Feed Management		Intensive Larval Feeding - Rotifers	Feed processing and Feeding programs	Finfish Viral Pathogens	Marker Assisted Selection	How to Price your Product
	Off Flavor and Toxic Algae	Handling/Grading/Transportation/Harvesting		Intensive Larval Feeding - Algae		Invertebrate Pathogens	Conservation and Population Genetics	Record Keeping
	Miscellaneous Treatments	Flow-Through System -Tanks & In-Pond Raceways		Intensive Larval Feeding - Artemia		External Parasites	Genetic Engineering	Partial Enterprise Budgeting
	Aquaculture and the Environment	Cage Culture- Types of Cages & Construction		Intensive Larval Feeding - Daphnia		Internal Parasite	Environmental Risk, Fitness and transgenic sterilization of GE fish	Project Analysis
	Best Management Practices for Aquaculture	Cage Culture-Cage Placement & Management		Intensive Larval Feeding - Artificial Diets		Biosecurity Summary	Public Education and Concerns in Biotechnology	Marketing
		Recirculation System Production Considerations		Pond Management			Combining Genetic Enhancement Programs	
		Production Planning-II		Nursery Ponds				
		AMR's-Aerated Microbial reuse systems		Practical Example				
		Record Keeping		Practical Example II				
		Project start up						

TOPIC AREA
WATERSHED & INTEGRATED COASTAL ZONE MANAGEMENT



IMPROVED CAGES FOR FISH CULTURE COMMERCIALIZATION IN DEEP WATER LAKES

Watershed & Integrated Coastal Zone Management/Experiment/09WIZ03UM

Collaborating Institutions & Lead Investigators

University of Michigan
Shanghai Ocean University
Huazhong Agricultural University
Wuhan University

James S. Diana
Liu Liping
Wang Weimin
Song Biyu

Objectives

To compare water quality parameters during the grow-out phase of traditional and improved cages

1. To estimate sedimentation rates from both cage systems
2. To evaluate fish production in each cage type
3. To develop a mass balance model to provide a first estimate of the carrying capacity for these cages in a reservoir system.

Significance

Many lakes, reservoirs, and slower flowing rivers in Asia are used for cage culture operations. These cages are often very productive, because they can use the larger volume of water in the system to improve water quality of cages, allowing a higher carrying capacity in the cages and intensive feeding. However, they also can be problematic because cages allow their solid and liquid wastes to disperse into the receiving waters, often causing pollution and habitat degradation. Costa-Pierce (1998) found that cage culture in lakes of Indonesia often exceeded the assimilative capacity of the water, resulting in poor water quality and even fish kills in the lakes. Diana (2009) hypothesized that cage culture often becomes a problem in lakes because of easy access to the culture location, low cost of operation, and lack of government control on the carrying capacity of cages. This appears to be particularly true in freshwater cage culture. Environmental problems related to cage culture include excessive nutrient release to the water and sedimentation smothering benthic habitats below the cages.

Management of cage culture for better sustainability requires improvements both in the cage systems and in regulating carrying capacity of cages in a water body. Professor Yang Yi, CRSP Host Country Principal Investigator until his death in August 2009, had a long history of CRSP research on the use of cages in natural waters as well as cage-cum-pond culture to recover nutrients from the waste produced by fish in cages. He recently worked with the Tongwei Group of China to develop a cage system for freshwater that includes two improvements (Figure 1). The first is a collecting system at the cage bottom to retain solid waste and allow its removal for safe disposal or use on land. The second is to include a second surrounding cage to allow for the culture of fish that will eat suspended waste solids, phytoplankton, and zooplankton produced by the higher nutrient levels of the cage, as well as periphyton on the cage surface. With these two

improvements, sediment discharge should be dramatically reduced from an individual cage, and more of the dissolved nutrients could be retained in harvested fish, thus producing less eutrophication effect from an individual cage into the receiving waters.

In order to produce a cage culture system that is more environmentally benign, not only does the cage system have to improve to reduce waste inputs per cage, but also knowledge of the nutrient release from these cages must be estimated in order to understand the eutrophication effects on the water body. Avery et al. (2005) developed a eutrophication model for Indonesian reservoirs and predicted that eutrophication of receiving waters would be fairly small if the FCR of the culture system was below 1.3, since most of the applied food would be used in fish growth rather than in waste. However, even at low conversion rates, it is important to evaluate the nutrient release from a cage system in order to estimate carrying capacity of cages in the overall reservoir. This is true whether the cage system produces lower nutrient release and sedimentation or not.

We propose to cooperate with Tongwei Group to evaluate the environmental efficiency of improved cages for use in deep reservoirs. Cages designed by the group are large and deep, and plan to be used in deep water reservoirs throughout Asia. Our initial testing will be done in China due to nearby location of the Tongwei facilities as well as an agreeable experimental site for the research. Future testing, assuming good performance of the cages, could be done in other parts of Asia, Africa, and even America, using species that are more commonly cultured in those countries. In fact, we have already made tentative arrangements with a culturist in Hawaii to field test the cages in a deep quarry there, assuming results are promising in the China tests. This experimental design would follow the earlier progress on cage-cum-pond systems in Asia, where initial systems developed in Thailand were exported to Vietnam and Bangladesh for further testing with local species. Deeper water in reservoirs allows more distance to the bottom to allow for decomposition of suspended solids before they settle out of the water, as well as a larger volume of water to assimilate nutrients produced by the cages. The individual cage design is shown in Figure 1. We will operate an experimental system of these cages, along with traditional cages, in order to test the relative efficiency of traditional and improved cage systems. Tongwei Group will fund cage construction and positioning, as well as feed and labor for cage maintenance. We intend to investigate the effects of each type of culture system on the nutrient release and sediment loss from the cages through a grow-out period. Using this information, we would also be able to produce nutrient dynamic models of the reservoir to make a first prediction of carrying capacity for both traditional and improved cages.

Quantifiable Anticipated Benefits

Expansion of cage culture is a very important means of increasing fish production in many countries, but the systems that exist are often the cause of much pollution burden. The proposed system should reduce the pollution burden considerably, and coupled with nutrient dynamic models, should be managed to maintain reasonable water quality along with major fish production in these deep waters. Measureable benefits will include the number of new cage systems tested, the increase in fish production provided by polyculture in the experimental systems, and the reduction in nutrient and sediment input to the lake resulting from the improved cage design. These benefits apply to North America as well, where organizations like SEA Vision (Sustainable Ecological Aquaculture) are using similar cages to produce sablefish more sustainably. Application in the U.S. will likely require development of cages for marine fishes, and modification of cage design. Deliverables on this research will include at least the presentation of two papers at aquaculture meetings and the publication of two theses or peer-reviewed papers. In

addition, at least one article will be produced in *Aquanews* to extend the results to a more general audience. Upon completion of the cage project, a workshop will be held in China to extend cage culture techniques, as well as the special results of this new cage culture system. The workshop will target practicing culturists, with a planned gender ratio of 50% female.

Research Design & Activity Plan

Location: China

Research methods:

1. Cage details: two series of 8 cages, with two rows and four cages per row, will be set up in the reservoir in May 2010. The location chosen is Changtan reservoir, a 30,000 ha hydropower reservoir near Guizhou. The water is used for hydropower and has a limited but measureable flow, so the cages will be set up with the rows perpendicular to the flow. Eight cages in one set will include the improved design cages with collecting cones (mesh size 0.25 cm) and surrounding nets. These cages will be 10x10 m (mesh size 4 cm) with another 1-m cage (mesh size 5 cm) surrounding the main culture cage. The second series of eight cages will be similar but only include the 10x10 m cages, with no collecting cones or surrounding cages. Grow out will continue for 6 months, when the catfish should reach a market size of 1 kg.
2. Fish stocking: traditional cages will be stocked at 100 channel catfish (*Ictalurus punctatus*) per m², or a total of 10,000 fish per cage, with fish of 100 g initial size. Experimental cages will have a similar stocking density in the inner cage, but also another combination of 7 bighead carp (500 g each), 3 silver carp (200 g), and 6 tench (50 g) per m² in the outer cage.
3. Cage management: cages will be managed by feeding the inner cage with pelleted feeds (X% CP) at manufacturer recommended levels. Feeding will be done at least twice daily—in morning and afternoon—by hand. No feed or other materials will be added to the outer cages.
4. Water quality measurement: water quality will be monitored using regular routine measurements and more infrequent detailed measurements.
 - 4.1. Regular routine measurements will be performed weekly, and will include DO, pH, conductivity, and temperature of the water using a data sonde. These measures will be taken in the cages and in outside water. Measures in the cages will be done in 3 locations: at 1x1 m inside the cage corner of the first cage in the upstream group, at the center of the cage cluster, and at 1x1 m inside the downstream edge of the last cage in the downstream group. In addition, water quality measures will be made surrounding the cages. For upstream and side locations, water quality will be measured at 1m from the edges and at the middle of the 4x4 cage cluster, approximately 5 m from the cages. At the downstream side, water quality will be measured at the same locations but at 5, 10, and 15 m from the cages. At all locations, measurements will be made at 0.5, 7, and 14 m in depth.
 - 4.2. Detailed water quality measurements will be made monthly after stocking at approximately 12 PM. Grow-out duration is six months. These measurements will include total phosphorus, soluble reactive phosphorus, total nitrogen, NO₃, NO₂, TAN, TVS, and TSS. In addition, phytoplankton and zooplankton samples will be collected with the water samples. Water will be collected in the cages at the same locations as regular measurements, and in the outside water only in

middle location of each side, using the same distances from the cages as regular measurements. Similar depth distributions of collections will also be used. Diel sampling of DO, temperature, and pH will also be done monthly at a location away from the cages.

5. Other measurements: Fish size will be evaluated monthly by collecting a sample of at least 100 fish for catfish only. Sedimentation rates will be estimated by placing sedimentation chambers 1 m below the bottom of cages in the same locations as in cage water samples, and removing them to determine the quantity of sediment deposited there on a monthly basis. Since the water depth will be 30-50 m, it will not be feasible to evaluate sedimentation rate on the lake bottom with this relatively few number of cages in operation, especially for the improved cages. Periphyton growth on the cages of each type will be estimated using digital photography on a monthly basis.

6. Null hypothesis, statistical analysis:

Different cage types have no effect on nutrient dynamics or sedimentation rates of the cages, and fish production will be similar in each cage type.

Data will be analyzed using ANOVA, t-test, correlation, and regression.

- 6.1. Economics: Cost-benefit analyses will be done on each cage type. Costs evaluated will include cage construction, feed, and stocked fish. Revenues will include money generated by the sale of fish. Estimates will be done for each eight cage unit as a whole. Costs and revenues will use current prices in the study locale as inputs.

Schedule

1 Jan 2010 to 30 August 2011 Report submission: no later than 29 September 2011.

Schedule includes:

Cage stocking – 1 July 2010

Fish and water sampling – monthly from 1 July 2010 through 1 January 2011

Harvest of cages – 1 January 2011

Evaluation of water chemistry and fish production data – through 1 June 2011

Development of loading models for the reservoir – through 1 July 2011

Preparation of final report and theses – through 29 September 2011

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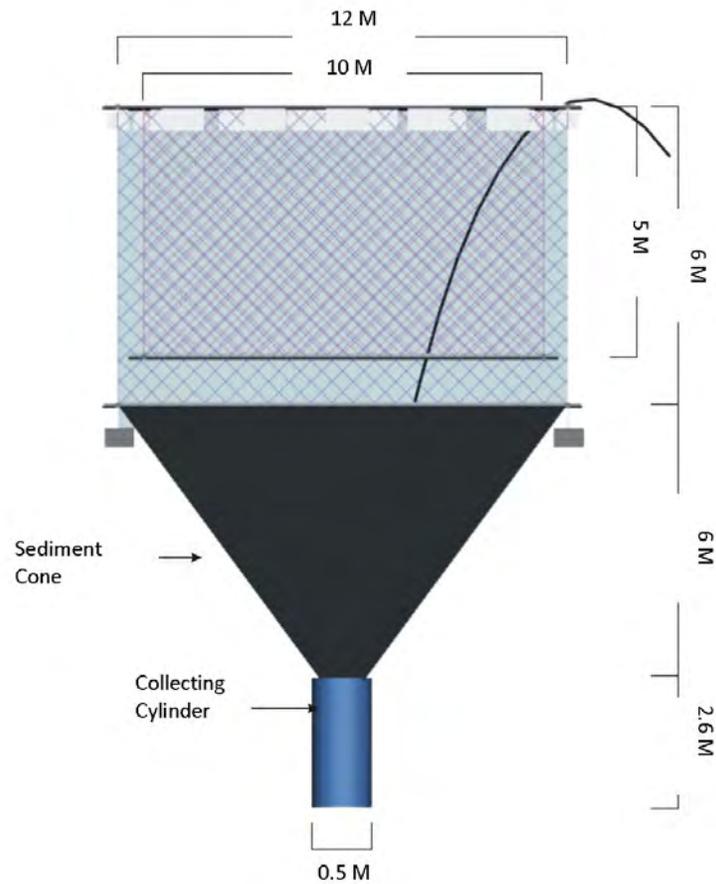


Figure 1. Diagram of the improved cages with sediment collection from the bottom and an outer cage for nutrient retention.

TOPIC AREA
MITIGATING NEGATIVE ENVIRONMENTAL IMPACTS



REACHING THE FARMS THROUGH AQUAFISH CRSP TECHNOLOGY TRANSFER:
ELIMINATION OF MT FROM INTENSIVE MASCULINIZATION SYSTEMS USING
BACTERIAL DEGRADATION

Mitigating Negative Environmental Impacts/Study/09MNE07UA

Collaborating Institutions & Lead Investigators

University of Arizona (USA)
Universidad Juárez Autónoma
de Tabasco (Mexico)

Kevin Fitzsimmons
Wilfrido M. Contreras-Sánchez

Objectives

The general objective of this project is to transfer AquaFish CRSP-generated technology to the private sector, ensuring the development of sustainable tilapia aquaculture in Southeastern Mexico and Central America. The specific objectives are:

1. Develop written materials for the dissemination of current MT degrading technology using bacteria protocols.
2. Transfer current MT technologies to a commercial hatchery.
3. Train tilapia farmers in the use of bacterial bio-flocks as MT-degrading entities and probiotics.
4. Continue evaluation of combinations of bacteria that best degrade MT in tilapia masculinization systems with on-farm trials
5. Assessment and evaluation of MT and hatchery technologies.

Significance

Agricultural and industrial activities generate a significant number of pollutants that are released to the environment through sewage water, which reach the superficial and underground water reservoirs. Many of these substances are highly toxic and do not degrade easily in nature. As a result of years of negligence, the levels of these substances in rural and industrial areas in Latin America are dramatically high. In order to reduce risks to human health and the environment, water treatment systems have been implemented (Valladares, 1995). Water treatments aim at 1) eliminating waste, floating fats and oils, sand, and all other coarse elements water may contain; 2) eliminating decantable materials, both organic and inorganic; 3) eliminating biodegradable organic matter dissolved in water; and 4) stabilizing and disposing mud extracted as a result of those processes (Crites and Tchobanoglous, 2000).

Aquaculture is an area in which Advanced Oxidation Processes (AOP's) could be applied to effluent treatment. The tilapia aquaculture industry uses steroidal hormones to manipulate phenotypical sex (Yamamoto, 1969). If these chemicals are adequately treated in the effluent, their use is beneficial and has no adverse environmental effects. However, there seems to be grounds for concern regarding water overuse. In particular, a legitimate international concern is the lack of up-to-date quantitative information regarding the chemicals being used and the lack of efforts oriented to studying and

treating the polluting by-products generated by this industry --steroids being one of them. Hormones used in aquaculture are among a list of compounds which environmental effects and possible treatments have been prioritized for research by GESAMP (FAO's Joint Group of Experts in the Scientific Aspects of Marine Environmental Protection).

Innovations in hormone applications in aquaculture include sex control, thus producing the so-called mono-sex lines, which result in significant production improvement (GESAMP, 1997). In particular, the molecule 17 α - Methyltestosterone (MT) has been widely used as an androgenic agent in masculinization of tilapia (Contreras-Sánchez, 2001). After the hormonal treatment (28 days), the hormone is released to the water through urine and feces and could become a powerful pollutant capable of producing harmful effects in wild species when effluents are released. In prior studies (Contreras-Sánchez, 2001), we found that masculinization of fry through dietary treatment with MT resulted in the accumulation of MT in sediments which produced both intersex fish and females with altered ovarian development. In systems where substrate was not present, there were higher concentrations of MT in the water and lower (sometimes null) masculinization rates than in systems with either soil or gravel. We found that charcoal filtration of water from systems where substrate was not present lowered the amount of MT in water to almost background levels and the treatment resulted in almost complete masculinization of all three broods tested (100, 98 and 100% males, respectively). Apparently, the recommended dose of MT for masculinize tilapia is higher than needed and a significant portion of it separates from the food and remains either in suspension in the water for the short term or persists in the sediments over the long term (Contreras-Sánchez, 2001). In the cited study, we recommended the use of filtration systems to eliminate excess MT to increase masculinization, and to prevent potential risks to humans or non-target fishes of unintended exposure to MT due to contamination of water and soils in farms. In this regard, research has been published on photocatalytic hormone elimination (Tanizaki, 2002). However, the published studies have focused on sewage water estrogen elimination due to the harmful biological effects of the presence of small levels of steroidal estrogens in water (Ike, 2002; Coleman et al., 2000 ; Carballa, 2004). Nevertheless, the elimination of MT in aquaculture using advanced oxidation processes has not been addressed.

A possible approach to eliminating MT from masculinization systems involves the use of bacterial degradation since it has been reported that some bacteria are capable of degrading steroids (Voishvillo et al, 2004). From this information and results from the previous investigations, we hypothesized that MT was being eliminated from water by solar irradiation and/or bacterial degradation within the filtration system. In our latest experiments (Contreras et al., 2010) we have isolated specific bacteria colonies from the biological filters of MT treatment tanks. We were also capable of eliminating up to 99% of the MT added in bacteria culture media using our bacteria isolates. Even more, the results with the strain of bacteria evaluated (*Pseudomonas aeruginosa*) indicate that these bacteria acted as probiotics improving survival and growth of the masculinized fish. In Southern Mexico, the use of bio-flocks has not been developed despite positive results in the shrimp and tilapia industries. Only one farm (Pucte del Usumacinta) has shown interest in the use of probiotics in their facilities and they are very interested in using bacteria to degrade MT in their masculinization tanks. In this hatchery up to 750,000 fry are masculinized monthly and the elimination of MT is a big concern for them. We propose to tune-up the methodology we have developed through AquaFish CRSP support and initiate technology transfer using the farm "Pucte del Usumacinta" as a model. Training sessions will help disseminate the methodology in such a way that more farmers will implement this method in their facilities.

Quantifiable Anticipated Benefits

New methods for elimination of MT will bring areas of opportunity for farmers that currently face MT in their effluents. Authorities will also perceive that MT can efficiently be eliminated from tilapia hatcheries. If successful, these methods can be transferred to tilapia hatcheries that play an important role supplying masculinized tilapia fry in the states of Tabasco and Chiapas, México as well as other countries in Central America and US based tilapia hatcheries that also could use this technique. The use of reliable and efficient masculinizing methods in the hatcheries will benefit thousands of small-scale fish farmers who currently see their productivity negatively affected by the use of mixed-sex populations of tilapia. A series of training workshops will be developed and offered to different audiences to ensure that these methodologies are effectively transferred to its final users. Technical workshops will target hatchery managers, extension agents and university students (many of whom will become workshop instructors over time). An UJAT student receiving a graduate degree with a focus on environmental microbiology would have many job opportunities on returning to Mexico. Involvement with this project as a potential thesis topic would strengthen the overall effort.

The reduction or elimination of MT from any potential effluents from the farm is an added benefit both for the environment and for potential US (and Mexican) consumers of the fish. Hatchery effluents are sometimes directed to other portions of farms or could pass to an adjacent farm. Reduction or elimination of MT in production fish which might be sold to US consumers is something we want to achieve as well.

Any excess MT leftover from the trials that is not broken down by the bacteria in the bioreactors will be oxidized with UV lamps. This technique has been shown to be effective, but is considerably more expensive than biodegradation. The assessment survey will provide us with valuable information regarding how the technologies have been applied and how the respondents perceive the costs and benefits have changed their behaviors and well being.

Gender Inclusivity: Four women will be involved in this project: Rosa Martha Padrón-López (Microbiologist); Lucero Vazquez-Cruz (technician) and two undergraduate students. We will encourage women participation in our workshops; however women involvement in tilapia masculinization at the farms is low. The volunteer biofloc workshop leader is a woman from Florida with extensive experience in microbial systems for aquaculture.

Research Design & Activity Plan

1) Written Materials development. Based on our results a manual and a brochure containing the information for bacterial degradation of MT and/or probiotics usage will be constructed. These materials will focus on simple description of concepts and methods, in such a way that they will be easily understood by farmers.

2) Technology transfer. The method that has been developed will be scaled up to fit the system characteristics present at the partner hatchery.

3) Capacity building in the use of bacterial bio-flocks as MT-degrading entities and probiotics. A seminar and workshop would be organized which will include experts from UJAT and a commercial hatchery in Florida to train the farmers and hatchery technicians on bacterial bioflocs and their use as MT-degrading entities. Two additional

workshops will be conducted: one at the national level in Mexico City and one at the Central America level in either Honduras or Guatemala. One student will be awarded an assistantship for graduate study at the University of Arizona. The student will focus on Environmental Microbiology as it applies to aquaculture.

4) Conduct On-Farm trials to evaluate combinations of bacteria that best degrade MT in commercial-scale tilapia masculinization systems.

a. Use of bacterial degradation using single and combined strains of bacteria.

Adaptive research is needed in order to tune-up our method to offer even better results to the hatchery owners. Work will be conducted at a partner hatchery and replicated at the Tropical Aquaculture and Microbiology Laboratories at UJAT to confirm the efficacy of using selected bacteria species and combination of bacteria species to eliminate MT in the hatchery environment.

Four bacteria species will be used: *Pseudomonas aeruginosa*, *Pseudomonas fluorescens*, *Bacillus cereus* and *Bacillus spp* (under current evaluation).

To evaluate the MT degradation capacity based on its use as carbon source, single and mixed strains will be cultured on MT enriched mineral media based on Perez et al., (2006) containing 40 mg of 17 α -Methyltestosterone (Argent Labs) for each 100 mL of culture media.

Based on this, a complete randomized; one factor experimental design will be constructed, consisting of four treatments:

- 1) Control: No bacterial colonies added.
- 2) *Pseudomonas aeruginosa* + *Pseudomonas fluorescens*.
- 3) *Bacillus subtilis* + *Bacillus spp*.
- 4) *Pseudomonas aeruginosa* + *Pseudomonas fluorescens* + *Bacillus subtilis* + *Bacillus spp*.

All treatments will be run using mineral culture media enriched with 40 mg/100 mL of MT and evaluated in triplicate. Erlenmeyer flasks containing 100 mL of culture media will be inoculated with 2 mL of bacterial suspension containing 15×10^8 CFU/100 mL (0.5 McFarland turbidimetric units). Flasks will be placed in a temperature controlled shaker bath and maintained to constant temperature and agitation (30° C @ 175 rpm) during a culture period of 26 days. Daily bacterial growth will be checked by plate counting microbiological procedures at 30°C to establish the growing stages. Samples for MT determination will be collected depending on bacterial culture lifetime.

General sampling days will be for all treatments on day: 0 (before bacteria added), 2, 6, 10, 16 and 20. All samples will be frozen (-20°C) and preserved until processing. Samples will be diluted in 40 % methanol (Cromasolv™ Sigma-Aldrich) and the MT concentration will be obtained by Methyltestosterone EIA Ridascreen™ determination kit (R3601, r-Biopharm) and data will be processed by RIDA®SOFT Win software to obtain the final concentration.

Colony formation units for each species will be compared using a Kruskal- Wallis test. Final MT concentrations among treatments will be evaluated by means of One-way ANOVA using the initial MT concentration as covariate. Differences between initial and final MT quantity will be analyzed by Kruskal- Wallis test.

An extra culture will be grown in the lab to test a large volume production system. This culture will be transferred to 2,000 mL Kettler Jar bioreactor (1100 mL working volume).

The jar system is equipped with an air inlet and a high-pressure valve. Media agitation and filtered air will be supplied using an aquarium compressor connected to a 0.45 µm Millipore™ membrane air filter. FCU counts will be obtained by normal plate culture count procedure at 30°C.

Polyethylene floating biospheres (450 units) will be disinfected by immersion during 48 h in a 10 % (v/v) solution prepared with commercial sodium hypochlorite and washed with distilled water. Biospheres will be transferred to a glass container containing 10 L of similar starting culture media and then inoculated with approximately 5.7 E+12 cells/mL and maintained for a period of 15 days with one culture media renovation at day 7. Presuming this system is successful it will be applied to the commercial hatchery trial.

b. Use of bacterial degradation of MT in masculinizing systems.

At the commercial hatchery, an experiment will be conducted for safely scaling-up the bacterial degradation technology. All treatments will be evaluated in triplicate. Treatments will be as follows: 1) Fry fed with MT at 60 mg/kg of food for 28 days; water recirculated through biofilter without bacterial inoculation (BA). 2) Fry fed with MT at 60 mg/kg of food for 28 days; water recirculated through biofilter with bacterial inoculation (BP).

Best combination of bacteria will be selected for running this experiment, based on MT-elimination efficiency. Mass production will be achieved using a laboratory-scale bioreactor. Starting cultures will be prepared in 250 mL Erlenmeyer flasks containing 100 mL of liquid media (two-fold concentration); cultures will be performed in a static system for 12 to 24 h at 35°C in a temperature-controlled oven.

Starting culture will initiate with approximately 3.27 E+07 cells/mL of each strain. This culture will be transferred to 2,000 mL Kettler Jar bioreactor (1100 mL working volume). The jar system is equipped with an air inlet and a high-pressure valve. Media agitation and filtered air will be supplied using an aquarium compressor connected to a 0.45 µm Millipore™ membrane air filter. FCU counts will be obtained by normal plate culture count procedure at 30°C. Polyethylene floating biospheres (450 units) will be disinfected by immersion during 48 h in a 10 % (v/v) solution prepared with commercial sodium hypochlorite and washed with distilled water. Biospheres will be transferred to a glass container containing 10 L of similar starting culture media and then inoculated with approximately 5.7 E+12 cells/mL and maintained for a period of 15 days with one culture media renovation at day 7.

Experimental units will consist of 8,000 L concrete tanks, equipped with a 1/2 HP centrifugal water pump (10 gpm, Aquapack AP-5X™) for water recirculation. Water will be pumped to a 200 L vertical tank containing the biofiltration unit. The biofilter will contain a 35 cm bottom layer made of coarse river gravel (~1" diameter) and an upper layer containing polyethylene biospheres (Aquatic ecosystems). Water will be pumped from the masculinization tank, will be supplied to the biofilter by a water inlet at the top of the filter spraying the water over the biospheres, then, by gravity, water will be passed by the biospheres and gravel to return to the masculinization tank by a water outlet located near the biofilter bottom. Each tank will be stocked with 5,000 tilapia fry (*Oreochromis niloticus*) obtained from our hatchery and then will be fed for 28 days with MT-enriched feed with at 60 mg/kg food.

After 7 days of fish treatment, bacterial biomass (liquid suspension with biospheres) will be inoculated into the biofilters for treatment BP, in case of no bacterial inoculation only

biospheres will be added. Air will be supplied to each biofilter for a three day-period and then water recirculation will be restarted. Water samples for MT determination will be collected the day of inoculation and at days 14 and 28 of MT treatment. All samples will be preserved frozen (-20°C) until processing. Samples will be processed directly or diluted in 40 % methanol — depending on MT concentration — (Cromasolv™; Sigma-Aldrich).

MT concentrations by date, and final growth for total length and weight will be compared using a Kruskal-Wallis test. Sex ratios and final survival will be compared by a Chi-square test to determine efficacy of MT treatment and possible bacterial effects on survival. Impact of bacterial flocks as probiotics will be tested comparing growth and survival results.

6) Survey for assessment and evaluation. We will conduct a survey of hatchery owners and personnel to evaluate the efficacy of the MT reduction system and of the overall tilapia and native cichlid hatchery training that has been provided in this and prior A/F CRSP training and technology transfer. We will develop a survey document and interview approximately half of the present and prior participants in the MT and other hatchery techniques trainings that we have conducted with A/F CRSP support. The survey will include questions on how the training has changed behaviors and attitudes on use of MT, elimination of MT from hatchery effluent, and which technology is most likely to be implemented. It will also examine the implementation of other hatchery methods and fish used. Selection of strain of tilapia and / or native cichlids will be evaluated as participants will be queried on why they selected a particular fish and on the economic benefits and costs. Finally, the survey will include general economic and enterprise budget questions regarding the costs of the various techniques and strains and the benefits that have been achieved.

Impact Indicators

- a) Implementation of clean technology for masculinization of tilapia fry.
- b) Number of technologies transferred
- c) Number of persons participating in workshops.
- d) Number of linkages with other organizations completed.
- e) Number of technical reports and journal articles.

Schedule

Sept-December 2010. Bacterial culture.

Sept 2010 Biofloc workshop at UJAT

Oct. 2010-Jan 2011. Bacterial MT degradation experiments.

Jan-Feb 2011. MT determination by RIA.

Oct 2010-May 2011. Technology transfer to hatchery “Pucté del Usumacinta”.

Sept 2010-Jul 2011. Workshops.

May – July 2011. Survey and assessment.

July-Sep 2011. Data analysis and preparation of reports and publications.

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