# Pond Dynamics/Aquaculture Collaborative Research Support Program

# Seventeenth Annual Technical Report

1 August 1998 to 31 July 1999

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#### Acknowledgments

The Program Management Office of the Pond Dynamics/Aquaculture CRSP gratefully acknowledges the contributions of all the CRSP researchers and the support provided by the Secretaría de Agricultura y Ganadería, Honduras; the Escuela Agrícola Panamericana, Honduras; the Instituto de Investigaciones de la Amazonia Peruana, Peru; the Universidad Nacional de la Amazonia Peruana, Peru; the Universidad Juárez Autónoma de Tabasco, Mexico; the Department of Fisheries, Ministry of Natural Resources, Kenya; the Asian Institute of Technology, Thailand; and the Freshwater Aquaculture Center, Central Luzon State University, the Philippines.

This report addresses program accomplishments of the Pond Dynamics/Aquaculture Collaborative Research Support Program during the reporting period of 1 August 1998 to 31 July 1999. Program activities are funded in part by the United States Agency for International Development (USAID) under Grant No. LAG-G-00-96-90015-00.

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# PD/A CRSP Seventeenth Annual Technical Report

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Since the PD/A CRSPs inception in 1982, the annual report has presented, among many standard features (program overview, research background, staff and fiscal summaries, networking activities, report abstracts, and others), the full text of the technical reports that correspond to studies funded within the particular reporting period. Beginning in 1993, the sheer quantity of information generated by program research necessitated a two-volume annual report format—an administrative and a technical report. As with the Sixteenth Annual Report, owing to a combination of new technologies and fiscal constraints, we have again not published technical reports in a bound hard-copy volume. Technical reports will be available at the program's Internet website <www.orst.edu/dept/crsp/homepage.html>. In addition, recognizing that many people do not have access to electronic information, hard copies of individual technical reports are available on request to the Information Management and Networking Component, Oregon State University, 400 Snell Hall, Corvallis OR 97331, USA.



## POND SOIL CHARACTERISTICS AND DYNAMICS OF SOIL ORGANIC MATTER AND NUTRIENTS

Ninth Work Plan, Pond Dynamics Research 2 (9PDR2) Progress Report

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#### Abstract

Analyses of soil cores from the bottoms of three freshwater fish ponds in Thailand and three in Peru revealed the typical layering in bulk density, total carbon, total nitrogen, and other selected physical and chemical variables observed in ponds at other CRSP sites and at Auburn University. Thus, we are now confident that the system of dividing soil profiles into S horizon (surface mixed sediment), M horizon (unmixed bulk sediment), T horizon (transition layer), and P horizon (original pond bottom soil) is based on a general feature of pond soil profiles. The soils from Iquitos, Peru, were very highly weathered and low in concentrations of macronutrient cations and micronutrients. There was very weak development of horizons in ponds at Iquitos. Organic matter decomposition in pond soils at Iquitos appears to be nitrogen limited. Ponds at Sae Kaeo, Thailand, had more highly developed horizons than those at Iquitos, Peru. The pond soils at Sae Kaeo also were highly weathered, but higher in cations and micronutrients than the Iquitos pond soils. Respiration per unit carbon (mg CO<sub>2</sub> g<sup>-1</sup> C) was significantly different (P < 0.01) among layers in ponds from Auburn, Alabama, and Ecuador, and the highest respiration rate was obtained in the uppermost 1.0-cm layer. Higher respiration rate is attributed to a higher ratio of labile to refractive organic matter in the upper layers.

## INTRODUCTION

Data on the physical and chemical properties of pond soils and on nutrient dynamics of pond soils are needed to develop a theory of pond soil development, formulate a method of pond soil classification, and improve pond soil management procedures. This report contains data on soils collected from three freshwater ponds at the PD/A CRSP site in Iquitos, Peru, and from three freshwater ponds at the Sae Kaeo Fisheries Station near Sae Kaeo, Thailand. In addition, data on soil respiration in soil cores taken from freshwater ponds at Auburn University are included.

#### METHODS AND MATERIALS

#### Peru and Thailand

Three ponds in Peru were sampled on 1 and 2 August 1998 and three ponds at Sae Kaeo, Thailand, were sampled on 21 and 22 February 1999.

Soil cores were taken with a hand-operated, 5-cm diameter core sampler (Wildlife Supply Company, Saginaw, Michigan, USA). Procedures for separating the cores into successive 2-cm-long core segments were described by Munsiri et al. (1995). Core segments were dried at 102°C (moisture content and dry bulk density) or 60°C (other analyses) and transported to Auburn University for analyses.

Samples were analyzed for moisture content (gravimetry), dry bulk density (gravimetry), color (Munsell color chart), wet soil pH (Peru samples only; direct, glass electrode), dry soil pH (1:1 slurry of dry soil and distilled water, glass electrode), exchangeable acidity (Adams-Evans buffer method), total carbon and nitrogen (Leco CHN Analyzer), total phosphorus (Peru samples only; perchloric acid oxidation), total sulfur (Leco Sulfur Analyzer), and acid-extractable phosphorus and metal ions (extraction in a 0.075 N acid solution of 0.05 N hydrochloric acid plus 0.025 N sulfuric acid followed by plasma spectrophotometry). Particle size analyses of samples from Peru were made by the pipette method. All methods followed details provided by Munsiri et al. (1995).

Aerobic and anaerobic incubation methods were used to estimate potentially mineralizable nitrogen and carbon (aerobic incubation only) in soils sampled from 0 to 10 cm and 10 to 20 cm layers of pond bottoms in Peru. Soil samples were refrigerated at 5°C until incubation. Aerobic incubations were done following methods of Wood and Edwards (1992), while anaerobic incubations were done according to methods outlined by Keeney (1982). In aerobic incubations, respired carbon dioxide was trapped in a vial containing 8 ml of 1 M sodium hydroxide (Anderson, 1982).

Soil organic carbon and nitrogen, and inorganic nitrogen [NO<sub>3</sub>-N plus NO<sub>2</sub>-N (aerobic incubation only), and NH<sub>4</sub>-N] were measured before incubations were initiated. Soil

where

inorganic nitrogen and respired  $CO_2$ -C were measured upon termination of incubation. Soil organic carbon and nitrogen were determined via dry combustion with a LECO CHN-600 analyzer. Inorganic nitrogen was extracted with 2 M potassium chloride and analyzed via the microplate method (Sims et al., 1995). Carbon dioxide in sodium hydroxide traps was determined by titrating excess base with 1 M hydrochloric acid in the presence of barium chloride (Anderson, 1982).

Soil potential nitrogen mineralization was calculated as the difference between final and initial contents of inorganic nitrogen for each incubation. Potential carbon mineralization was calculated as the difference between the incubation base trap and the mean of four blanks.

## **Additional Soil Respiration Studies**

Soil for this study was obtained from six freshwater catfish ponds located on the Auburn University Fisheries Research Unit, Auburn, Alabama, and eight brackishwater shrimp ponds in Ecuador. Samples from the upper 5-cm sediment layer of each pond were collected with a 5-cm-diameter core liner tube. Freshwater and brackishwater sediment cores were then sliced into successive 0.5-cm and 1.0-cm-long segments, respectively, according to methods described in Masuda and Boyd (1994). The cores were obtained from 8 to 15 sites within each pond and combined to provide one sample of each depth layer. All samples were dried in a forced draft oven at 60°C, pulverized with a hammer mill-type soil crusher (Custom Laboratory Equipment Inc., Orange City, Florida, USA) to pass a 20-mesh screen, and stored in plastic bags for further analysis.

Soil moisture was determined by oven drying the samples at 105°C. Total carbon concentrations were measured with a LECO EC12 Induction Furnace Analyzer.

The technique used to measure soil respiration is described by Anderson (1982) and consists of trapping the carbon dioxide evolved during microbial respiration in 1 N sodium hydroxide solution placed inside an airtight chamber. Sodium carbonate resulting from the reaction of carbon dioxide and sodium hydroxide is precipitated with excess barium chloride, and the remaining alkali is then back-titrated with standard 1.00 N hydrochloric acid. The reactions are given in the following equations:

CO <sub>2</sub> + 2NaOH	=	$Na_2CO_3 + H_2O$
$Na_2CO_3 + BaCl_2$	=	$2NaCl + BaCO_3$

Respiration chambers were prepared in triplicate for each soil layer by adding 15 g of dry soil into 1-quart (946-ml) mason jars. Soils were moistened with bacterial enriched biological oxygen demand (BOD) dilution water. The bacterial inoculum Polyseed<sup>®</sup> was obtained from Polybac Corporation, Bethlehem, Pennsylvania, USA. Preparation of BOD dilution water and seed inoculum followed guidelines recommended by Eaton et al. (1995). PVC tubes (5 cm x 3.18 cm diameter) were inserted vertically into the soil to support 20-ml alkali containers. Support tubes had holes to allow passage of carbon dioxide formed in soil beneath alkali containers. Respiration chambers without soil were carried through the procedure as control blanks. Microcosms were tightly capped with lids. Soils were incubated in the dark for one week in an incubator at 25°C. Alkali solution containing sodium carbonate was precipitated with 3 N barium chloride in 25-ml centrifuge tubes. Supernatant containing remnant sodium hydroxide was separated from precipitate by centrifugation at 2,500 rpm. The amount of carbon dioxide evolved in soil respiration was estimated by the following equation:

$$CO_2 (mg g^{-1}) = (B - V) N 22/W$$

B = acid used to titrate sodium hydroxide in the blank (ml),

- V = acid used to titrate sodium hydroxide in treatment (ml),
- N = normality of hydrochloric acid (1.00 N),
- 22 = equivalent weight of carbon dioxide,
- W = dry weight of soil confined in respiration chamber (g).

## **RESULTS AND DISCUSSION**

#### Pond Soils at Iquitos, Peru

The moisture content of the soil in ponds at the Iquitos site decreased rapidly with depth, and dry bulk density increased with depth (Table 1). The S horizon was only 2 cm thick, and the M horizon also was thin, extending from 2 to 6 cm. The T horizon was between 6 and 10 cm depth. The P horizon had quite high dry bulk densities of 1.75 g cm<sup>-3</sup> and above. Soil color changed very little with depth (Table 2). Ponds at other CRSP sites including newly renovated ponds at Sagana, Kenya (Boyd et al., 1997, 1998), and on the Auburn University Fisheries Research Unit (Munsiri et al., 1995) had much more strongly developed profiles than the ponds at Iquitos.

Wet soil pH was very similar to dry soil pH in ponds at Iquitos (Table 3). At other CRSP sites and at Auburn University, wet soil pH usually has been slightly higher than dry soil pH in acidic soils, and wet soil pH has been slightly lower than dry soil pH in neutral or basic soils (Boyd et al., 1997, 1998). As no definite pattern can be established among wet and dry soil pH and most of the literature on soil pH is based on dry soil pH, we will not measure wet soil pH in future studies. The soil pH

Table 1. Profiles for moisture content and dry bulk density in soil cores from bottoms of aquaculture ponds in Iquitos, Peru. Moisture content averages and standard errors are given as percentages of dry weight. Dry bulk density averages and standard errors are given as gram dry soil per cubic centimeter (g cm<sup>-3</sup>). Each entry is the average of three ponds.

<b>Depth</b> (cm)	Moisture Content (%)	Dry Bulk Density (g cm <sup>-3</sup> )
0-2 2-4	$519.1 \pm 205.8$ $327.1 \pm 208.4$	$0.27 \pm 0.14$ $0.70 \pm 0.47$
$\frac{1}{4-6}$ 6-8	$95.6 \pm 54.8$ $33.9 \pm 12.9$	$0.97 \pm 0.32$ $1.44 \pm 0.22$
8 –10 10 –12	$\begin{array}{c} 26.2 \pm 5.2 \\ 17.9 \pm 0.4 \end{array}$	$\begin{array}{c} 1.60 \pm 0.14 \\ 1.78 \pm 0.04 \end{array}$
12–14 14–16	$16.2 \pm 0.4$ $16.1 \pm 0.2$	$\begin{array}{c} 1.83 \pm 0.05 \\ 1.78 \pm 0.04 \end{array}$
16–18 18–20	$16.6 \pm 0.5$ $15.3 \pm 0.6$ $15.6 \pm 0.7$	$1.81 \pm 0.05$ $1.90 \pm 0.10$ $1.90 \pm 0.01$
20-22 22-24 24-26	$15.6 \pm 0.7$ $16.1 \pm 0.5$ $18.0 \pm 0.8$	$1.80 \pm 0.01$ $1.75 \pm 0.10$ $1.75 \pm 0.04$
24–20 26–28 28–30	$17.1 \pm 0.8$ $17.4 \pm 0.9$	$1.86 \pm 0.07$ $1.92 \pm 0.14$

was greater in the S and M horizons at Iquitos than in deeper layers because of liming of pond bottoms. Exchangeable acidity of the soils ranged from 9.33 meq  $(100 \text{ g})^{-1}$  in surface soils to near 5 meq  $(100 \text{ g})^{-1}$  in deeper soils. The surface soils had a greater exchangeable acidity than deeper soils in spite of the higher pH of the surface soils. This phenomenon results because the surface soils were greatly enriched in organic matter (Table 4) and clay (Table 5) compared to deeper layers, and organic matter and clay increase the cation exchange

Table 2.Profiles for color in soil cores from bottoms of aquaculture<br/>ponds in Iquitos, Peru. Color values are given as standard<br/>Munsell Color Chart Units. Each entry is the value of a<br/>particular pond.

<b>Depth</b> (cm)	Pond 4	Pond 5	Pond 6
$\begin{array}{c} 0 - 2 \\ 2 - 4 \\ 4 - 6 \\ 6 - 8 \\ 8 - 10 \\ 10 - 12 \\ 12 - 14 \\ 14 - 16 \\ 16 - 18 \\ 18 - 20 \\ 20 - 22 \\ 22 - 24 \\ 24 - 26 \\ 26 - 28 \end{array}$	5Y 3/2 10YR 4/2 10YR 5/2 10YR 5/2	$\begin{array}{c} 2.5Y \ 3/3\\ 2.5Y \ 4/2\\ 2.5Y \ 4/2\\ 7.5Y \ 4/2\\ 7.5Y \ 4/1\\ 7.5Y \ 4/1\\ 7.5Y \ 4/1\\ 7.5Y \ 5/2\\ 7.5Y \ 4/2\\ 7.5Y \ 4/2\\ \end{array}$	10YR 4/2 10YR 4/2 10YR 4/2 10YR 5/2 10YR 4/1 10YR 5/2 10YR 5/2 10YR 5/2 10YR 4/1 10YR 5/2 10YR 4/1 10YR 5/2 10YR 5/2 10YR 5/2
28-30	10YR 4/2	7.5Y 4/2	10YR 5/2

Note: 2.5Y 3/3—Dark olive brown; 2.5Y 4/2—Dark grayish brown; 2.5Y 5/2— Grayish brown; 5Y 3/2—Dark olive gray; 7.5Y 4/1—Dark gray; 7.5Y 4/2—Brown; 7.5Y 5/2— Brown; 10YR 4/1—Dark gray; 10YR 4/2—Dark grayish brown; 10YR 5/2—Grayish brown; 10YR 5/3—Brown.

Table 3. Profiles for wet soil pH, dry soil pH, and exchangeable acidity in soil cores from bottoms of aquaculture ponds in Iquitos, Peru. The wet soil pH is directly measured in soil core and the dry soil pH is measured in 1:1 slurries of dry soil and distilled water. Averages and standard errors of pH are given as standard pH units. Exchangeable acidity averages and standard errors are given as milliequivalents per 100 grams dry soil (meq (100 g)<sup>-1</sup>). Each entry is the average of three ponds.

<b>Depth</b> (cm)	Wet Soil pH	Dry Soil pH	Exchangeable Acidity (meq (100 g) <sup>-1</sup> )
$\begin{array}{c} 0 - 2 \\ 2 - 4 \\ 4 - 6 \\ 6 - 8 \\ 8 - 10 \\ 10 - 12 \\ 12 - 14 \\ 14 - 16 \\ 16 - 18 \\ 18 - 20 \\ 20 - 22 \\ 22 - 24 \\ 24 - 26 \\ 26 - 28 \\ 28 - 30 \end{array}$	$\begin{array}{c} 6.5 \pm 0.1 \\ 6.5 \pm 0.1 \\ 6.2 \pm 0.2 \\ 5.8 \pm 0.5 \\ 6.3 \pm 0.2 \\ 6.6 \pm 0.1 \\ 6.1 \pm 0.4 \\ 5.6 \pm 0.4 \\ 5.6 \pm 0.2 \\ 5.7 \pm 0.4 \\ 5.9 \pm 0.2 \\ 5.9 \pm 0.3 \\ 5.9 \pm 0.0 \\ 5.5 \pm 0.7 \\ 5.9 \pm 0.4 \end{array}$	$\begin{array}{c} 6.3 \pm 0.2 \\ 6.3 \pm 0.3 \\ 6.2 \pm 0.1 \\ 6.1 \pm 0.1 \\ 5.9 \pm 0.3 \\ 6.0 \pm 0.3 \\ 5.8 \pm 0.3 \\ 5.9 \pm 0.3 \\ 5.8 \pm 0.3 \\ 5.8 \pm 0.2 \\ 5.8 \pm 0.3 \\ 5.7 \pm 0.3 \\ 5.7 \pm 0.3 \\ 5.7 \pm 0.3 \\ 5.7 \pm 0.3 \\ 5.5 \pm 0.3 \\ 5.5 \pm 0.3 \\ 5.5 \pm 0.3 \end{array}$	$\begin{array}{c} 9.33 \pm 2.71 \\ 7.20 \pm 3.33 \\ 5.07 \pm 2.18 \\ 5.33 \pm 0.53 \\ 5.33 \pm 1.92 \\ 6.40 \pm 3.33 \\ 4.80 \pm 2.40 \\ 5.07 \pm 2.54 \\ 5.33 \pm 2.67 \\ 5.07 \pm 2.54 \\ 5.33 \pm 2.67 \\ 5.47 \pm 2.53 \\ 5.60 \pm 2.40 \\ 5.73 \pm 2.68 \end{array}$

capacity (CEC) of soil. Soils with a higher CEC will have a greater exchangeable acidity at a given pH than will soils of a lower CEC.

There was a distinct layering of total carbon and total nitrogen in the soil profiles. The S horizon had 3.15% carbon and 0.08% nitrogen, the M horizon averaged about 1% carbon and 0.03% nitrogen, and deeper horizons were very low in carbon and nitrogen content (Table 4). The C/N ratios were 39 in the S horizon, 33 in the M horizon, and 29 to 62 in the T and P horizons. These are much lower C/N ratios than those found in ponds at other CRSP sites and at Auburn University (Boyd et al., 1997, 1998; Munsiri et al., 1995) where C/N ratios ranged between 7 and 20 with most values below 15. The wide C/N ratios in ponds at Iquitos suggest that nitrogen inputs to ponds in feeds and fertilizers have been much lower than at other CRSP sites.

Total sulfur concentrations (Table 4) were greater in the S and M horizons than in deeper soil, but there was very little sulfur in the soils at Iquitos. In fact, sulfur was undetectable in most samples from the P horizon. Three forms of phosphorus—total, dilute-acid-extractable, and water-extractable—were measured in the samples (Table 6). Total phosphorus concentrations were as high or higher than those measured at other CRSP sites (Boyd et al., 1997, 1998) and at Auburn University (Munsiri et al., 1995), but dilute-acid-extractable phosphorus concentrations were very low, and except in the S and M horizons, water-extractable phosphorus also was low.

The total phosphorus fraction is very difficult to measure because the phosphorus must be liberated from the soil by perchloric acid digestion. The water-extractable phosphorus also is difficult to measure because phosphorus concentrations often are near the lower detection limit. Also, total phosphorus concentrations often have little relationship to the concentrations of extractable phosphorus or water-extractable phosphorus (Boyd and Munsiri, 1996). Thus, for future analyses of pond soil cores, we have decided to measure only dilute-acidextractable and water-extractable phosphorus.

Table 4.Profiles of total carbon, total nitrogen, and total sulfur in soil<br/>cores from bottoms of aquaculture ponds in Iquitos, Peru.<br/>Averages and standard errors are given as percentages. Each<br/>entry is the average of three ponds.

<b>Depth</b> (cm)	Total Carbon (%)	Total Nitrogen (%)	Total Sulfur (%)
0-2	$3.15\pm0.77$	$0.08\pm0.03$	$0.04 \pm 0.02$
2 - 4	$1.28\pm0.55$	$0.03\pm0.01$	$0.02\pm0.01$
4 - 6	$0.77\pm0.23$	$0.02\pm0.01$	$0.01\pm0.01$
6 - 8	$0.58\pm0.09$	$0.01\pm0.00$	0.00
8-10	$0.61\pm0.19$	$0.01\pm0.003$	$0.01\pm0.01$
10 - 12	$0.62\pm0.23$	$0.01\pm0.003$	$0.01\pm0.00$
12 - 14	$0.44\pm0.10$	$0.01\pm0.003$	0.00
14 - 16	$0.39\pm0.06$	$0.01\pm0.00$	0.00
16 - 18	$0.37\pm0.06$	$0.01\pm0.00$	0.00
18 - 20	$0.49\pm0.12$	$0.01\pm0.003$	0.00
20-22	$0.36\pm0.08$	$0.01\pm0.003$	0.00
22 - 24	$0.29\pm0.06$	$0.01\pm0.003$	$0.01\pm0.00$
24-26	$0.31\pm0.04$	$0.01\pm0.00$	0.00
26-28	$0.29\pm0.02$	$0.01\pm0.00$	0.00
28-30	$0.32\pm0.04$	$0.01\pm0.00$	0.00

Table 5. Profiles for soil texture and particle size distribution in soil cores from bottoms of aquaculture ponds in Iquitos, Peru. Averages and standard errors for particle size distribution are given as percentages of dry weight. Each entry is the average of three ponds.

Depth (cm)	Soil Texture	Sand (%)	<b>Silt</b> (%)	<b>Clay</b> (%)
0.2	Loom	42 20 + 16 26	25.27 + 10.59	22 = 2 + 6 = 29
0 - 2	Loan	$42.20 \pm 10.30$	$55.27 \pm 10.38$	$22.33 \pm 0.36$
2 - 4	Sandy loam	$62.84 \pm 15.45$	$24.47 \pm 10.66$	$12.69 \pm 4.94$
4 - 6	Sandy loam	$68.37 \pm 5.85$	$21.10 \pm 3.32$	$10.53 \pm 3.26$
6 – 8	Sandy loam	$68.63 \pm 2.20$	$23.26 \pm 3.07$	$8.11 \pm 1.33$
8-10	Loamy sand	$82.70 \pm 5.01$	$11.81 \pm 2.93$	$5.49 \pm 2.20$
10-12	Loamy sand	$78.70 \pm 5.86$	$15.63 \pm 3.56$	$5.67 \pm 2.96$
12-14	Loamy sand	$79.04 \pm 5.61$	$15.83 \pm 4.14$	$5.13 \pm 2.34$
14-16	Loamy sand	$79.00\pm4.28$	$16.23\pm2.85$	$4.77\pm2.59$
16-18	Loamy sand	$81.33 \pm 3.37$	$13.71 \pm 1.55$	$4.96\pm2.15$
18-20	Loamy sand	$81.13 \pm 4.45$	$13.99 \pm 3.30$	$4.88 \pm 1.95$
20-22	Loamy sand	$82.20 \pm 5.11$	$13.49\pm3.78$	$4.31\pm2.11$
22-24	Loamy sand	$82.63 \pm 4.94$	$12.77 \pm 3.16$	$4.60\pm2.14$
24-26	Loamy sand	$79.64 \pm 3.32$	$15.03 \pm 2.55$	$5.33 \pm 1.69$
26-28	Loamy sand	$78.54 \pm 3.92$	$16.45\pm3.31$	$5.01 \pm 1.45$
28-30	Loamy sand	$81.14\pm3.38$	$13.93\pm2.52$	$4.93 \pm 1.50$

Table 6.Profiles for total phosphorus, dilute-acid-extractable<br/>phosphorus, and water-extractable phosphorus in soil cores<br/>from bottoms of aquaculture ponds in Iquitos, Peru.<br/>Averages and standard errors for total phosphorus and<br/>dilute-acid-extractable phosphorus are given as parts per<br/>million (ppm), and each entry is the average of three ponds.<br/>Concentrations for water-extractable phosphorus are given<br/>in milligrams per liter for a single pond at each location.

Total Phosphorus (ppm)	Dilute-acid- extractable Phosphorus (ppm)	Water-extractable Phosphorus (mg l <sup>-1</sup> )
$1,726 \pm 288$	$15.5 \pm 2.2$	0.238
$1,241 \pm 505$	$14.4\pm3.2$	0.123
$1,020 \pm 274$	$11.0\pm3.8$	0.081
$1,092 \pm 74$	$8.9\pm3.5$	0.074
$618 \pm 123$	$5.0 \pm 1.6$	0.007
$592 \pm 118$	$3.1 \pm 0.5$	0.035
$702 \pm 162$	$3.5 \pm 0.7$	0.006
$732 \pm 66$	$2.7\pm0.9$	0.023
$716 \pm 39$	$2.8\pm0.9$	0.005
$495\pm88$	$2.7\pm0.6$	0.020
$561 \pm 71$	$2.4 \pm 0.4$	0.005
$387 \pm 51$	$2.1 \pm 0.6$	0.018
$760 \pm 199$	$2.4 \pm 0.4$	0.004
$656 \pm 58$	$2.8\pm0.6$	0.015
$635\pm208$	$2.6\pm0.7$	0.004
	$\begin{array}{c} \textbf{Total}\\ \textbf{Phosphorus}\\ (ppm) \end{array} \\ \hline 1,726 \pm 288\\ 1,241 \pm 505\\ 1,020 \pm 274\\ 1,092 \pm 74\\ 618 \pm 123\\ 592 \pm 118\\ 702 \pm 162\\ 732 \pm 66\\ 716 \pm 39\\ 495 \pm 88\\ 561 \pm 71\\ 387 \pm 51\\ 760 \pm 199\\ 656 \pm 58\\ 635 \pm 208 \end{array}$	$\begin{array}{c} {\bf Total} \\ {\bf Phosphorus} \\ (ppm) \\ \end{array} \begin{array}{l} {\bf Dilute-acid- \\ extractable \\ {\bf Phosphorus} \\ (ppm) \\ \end{array} \end{array} \\ \begin{array}{l} 1,726\pm288 \\ 1,241\pm505 \\ 1,241\pm505 \\ 1,241\pm505 \\ 1,241\pm505 \\ 1,241\pm505 \\ 14.4\pm3.2 \\ 1,002\pm74 \\ 11.0\pm3.8 \\ 1,092\pm74 \\ 8.9\pm3.5 \\ 618\pm123 \\ 5.0\pm1.6 \\ 592\pm118 \\ 3.1\pm0.5 \\ 702\pm162 \\ 3.5\pm0.7 \\ 732\pm66 \\ 2.7\pm0.9 \\ 713\pm6.5 \\ 702\pm162 \\ 3.5\pm0.7 \\ 732\pm66 \\ 2.7\pm0.9 \\ 716\pm39 \\ 2.8\pm0.7 \\ 124\pm0.4 \\ 387\pm51 \\ 2.1\pm0.6 \\ 760\pm199 \\ 2.4\pm0.4 \\ 656\pm58 \\ 2.8\pm0.6 \\ 635\pm208 \\ 2.6\pm0.7 \\ \end{array}$

The soils at Iquitos were very low in concentrations of calcium, magnesium, sodium, and potassium (Table 7) when compared to soils from ponds at other CRSP sites and Auburn University (Boyd et al., 1997, 1998; Munsiri et al., 1995). Nevertheless, the S and M horizons had greater concentrations of these major ions as compared to the deeper horizons. This phenomenon is the result of additions of major ions to ponds in liming materials and fertilizers used in aquaculture. It is not surprising that the soils at Iquitos are low in concentrations of major ions, because soils in tropical areas with high rainfall and high temperature tend to be highly leached and low in bases.

Table 7.	Profiles for calcium, magnesium, potassium, and sodium in
	soil cores from bottoms of aquaculture ponds in Iquitos,
	Peru. Averages and standard errors are given as parts per
	million (ppm). Each entry is the average of three ponds.

$\begin{array}{cccccccccccccccccccccccccccccccccccc$	<b>Depth</b>	Potassium So	<b>Magnesium</b>	Sodium
	(cm)	(ppm) (	(ppm)	(ppm)
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\begin{array}{c} 0 - 2 \\ 2 - 4 \\ 4 - 6 \\ 6 - 8 \\ 8 - 10 \\ 10 - 12 \\ 12 - 14 \\ 14 - 16 \\ 16 - 18 \\ 18 - 20 \\ 20 - 22 \\ 22 - 24 \\ 24 - 26 \\ 26 - 28 \\ 28 - 30 \end{array}$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\begin{array}{c} 14 \pm 3 \\ 6.1 \pm 2.4 \\ 3.4 \pm 1.3 \\ 3.1 \pm 0.7 \\ 2.0 \pm 0.7 \\ 2.1 \pm 0.8 \\ 1.7 \pm 0.8 \\ 1.2 \pm 0.4 \\ 1.3 \pm 0.5 \\ 1.3 \pm 0.4 \\ 1.2 \pm 0.3 \\ 1.0 \pm 0.2 \\ 1.8 \pm 0.5 \\ 1.5 \pm 0.5 \\ 2.5 \pm 1.7 \end{array}$	$\begin{array}{c} 8.8 \pm 0.7 \\ 5.9 \pm 0.7 \\ 4.6 \pm 0.7 \\ 4.9 \pm 0.2 \\ 5.1 \pm 0.5 \\ 4.2 \pm 0.4 \\ 4.8 \pm 0.4 \\ 3.7 \pm 0.3 \\ 4.7 \pm 0.5 \\ 4.4 \pm 0.4 \\ 4.3 \pm 0.3 \\ 4.1 \pm 0.2 \\ 4.8 \pm 0.1 \\ 5.1 \pm 1.1 \\ 3.2 \pm 0.8 \end{array}$

Concentrations of micronutrients also were quite low in soils at Iquitos (Table 8). Manganese and zinc were higher in concentration in S and M horizons than in deeper layers, but iron and zinc did not exhibit layering.

Except for the surface 0 to 2 cm (S horizon), all other depths were very low in clay content (Table 5). The S horizon was a loam, the M horizon and upper part of T horizon (MT horizon) was a sandy loam, and deeper layers were loamy sand. Ponds at other CRSP sites contained considerably more clay than the Iquitos ponds (Boyd et al., 1997, 1998).

#### **Incubation Studies for Peru Samples**

Results of the soil incubations are provided in Table 9. The concentrations of total carbon were  $0.99 \pm 0.36\%$  and  $0.62 \pm 0.12\%$  in samples from depths of 0–10 cm and 10–20 cm, respectively. The total nitrogen values were  $0.03 \pm 0.01\%$  in samples from

l'able 8.	Profiles for iron, manganese, zinc, and copper in soil cores
	from bottoms of aquaculture ponds in Iquitos, Peru.
	Averages and standard errors are given as parts per million
	(ppm). Each entry is the average of three ponds.

<b>Depth</b> (cm)	<b>Iron</b> (ppm)	Manganese (ppm)	Zinc (ppm)	<b>Copper</b> (ppm)
0 - 2	$33 \pm 2$	$2.1\pm0.6$	$8.32 \pm 1.19$	$0.15\pm0.01$
2 - 4	$27 \pm 4$	$1.1 \pm 0.5$	$4.91 \pm 2.12$	$0.14\pm0.02$
4 - 6	$26 \pm 4$	$0.7 \pm 0.3$	$2.44 \pm 1.22$	$0.17\pm0.01$
6 – 8	$32 \pm 2$	$0.6 \pm 0.2$	$1.47\pm0.69$	$0.18\pm0.02$
8-10	$35 \pm 11$	$0.4 \pm 0.1$	$0.31 \pm 0.17$	$0.15 \pm 0.01$
10 - 12	$37 \pm 15$	$0.5 \pm 0.3$	$0.29 \pm 0.13$	$0.13\pm0.03$
12 - 14	$30 \pm 11$	$0.4 \pm 0.2$	$0.48 \pm 0.34$	$0.15\pm0.01$
14-16	$50 \pm 14$	$0.3 \pm 0.1$	$0.41 \pm 0.29$	$0.10\pm0.03$
16-18	$27 \pm 11$	$0.3 \pm 0.1$	$0.31 \pm 0.21$	$0.11\pm0.03$
18 - 20	$53 \pm 17$	$0.3 \pm 0.1$	$0.21 \pm 0.11$	$0.13\pm0.01$
20-22	$29 \pm 11$	$0.3 \pm 0.1$	$0.18 \pm 0.08$	$0.11 \pm 0.05$
22-24	$24 \pm 9$	$0.3 \pm 0.1$	$0.13 \pm 0.06$	$0.10 \pm 0.03$
24-26	$24 \pm 11$	$0.4 \pm 0.1$	$0.23 \pm 0.09$	$0.11 \pm 0.04$
26-28	$24 \pm 10$	$0.4 \pm 0.1$	$0.15 \pm 0.08$	$0.10 \pm 0.05$
28-30	$21 \pm 10$	$0.9 \pm 0.6$	$0.18 \pm 0.10$	$0.10 \pm 0.04$

Table 9.Net aerobic mineralization of nitrogen and carbon dioxide<br/>and net anaerobic mineralization of ammonia in soil from<br/>two depths from ponds in Iquitos, Peru.

<b>Depth</b> (cm)	Aerobic Incubation		Anaerobic Incubation
	(mg N kg <sup>-1</sup> )	$(mg CO_2 kg^{-1})$	(mg NH <sub>4</sub> -N kg <sup>-1</sup> )
0–10 10–20	$\begin{array}{c} -20.56 \pm 11.58 \\ -24.78 \pm 4.05 \end{array}$	$\begin{array}{c} 4.09 \pm 1.83 \\ 3.01 \pm 1.56 \end{array}$	$-0.75 \pm 1.57$ $-6.41 \pm 3.00$

Table 10. Profiles for moisture content and dry bulk density in soil cores from bottoms of aquaculture ponds, Thailand. Moisture content averages and standard errors are given as percentages of dry weight. Dry bulk density averages and standard errors are given as grams dry soil per cubic centimeter (g cm<sup>-3</sup>). Each entry is the average of three ponds.

<b>Depth</b> (cm)	Moisture Content (%)	<b>Dry Bulk Density</b> (g cm <sup>-3</sup> )
$\begin{array}{c} 0-2\\ 2-4\\ 4-6\\ 6-8\\ 8-10\\ 10-12\\ 12-14\\ 14-16\\ 16-18\\ 18-20\\ 20-22\\ 22-24\\ 24-26\\ 26-28\\ 28-30 \end{array}$	$\begin{array}{c} 273.2 \pm 75.1 \\ 175.0 \pm 44.6 \\ 139.8 \pm 28.0 \\ 123.5 \pm 19.3 \\ 86.0 \pm 12.6 \\ 78.3 \pm 15.0 \\ 75.5 \pm 16.1 \\ 73.5 \pm 13.5 \\ 65.7 \pm 16.1 \\ 61.6 \pm 16.3 \\ 50.9 \pm 7.4 \\ 36.6 \pm 4.9 \\ 33.3 \pm 4.0 \\ 27.2 \pm 1.2 \\ 26.4 \pm 3.4 \end{array}$	$\begin{array}{c} 0.33 \pm 0.06 \\ 0.47 \pm 0.07 \\ 0.54 \pm 0.09 \\ 0.59 \pm 0.09 \\ 0.77 \pm 0.09 \\ 0.82 \pm 0.12 \\ 0.85 \pm 0.15 \\ 0.82 \pm 0.11 \\ 0.94 \pm 0.17 \\ 0.97 \pm 0.19 \\ 1.04 \pm 0.11 \\ 1.29 \pm 0.13 \\ 1.29 \pm 0.06 \\ 1.44 \pm 0.04 \\ 1.51 \pm 0.08 \end{array}$

Table 11. Profiles for color in soil cores from bottoms of aquaculture<br/>ponds, Thailand. Color values are given as standard Munsell<br/>Color Chart Units. Each entry is the value of a particular<br/>pond.

<b>Depth</b> (cm)	Pond 1	Pond 2	Pond 3
$\begin{array}{c} 0-2\\ 2-4\\ 4-6\\ 6-8\\ 8-10\\ 10-12\\ 12-14\\ 14-16\\ 16-18\\ 18-20\\ 20-22\\ 22-24\\ 24-26\\ 26-28\\ 28-30 \end{array}$	5Y 7/4 5Y 7/4 5Y 7/4 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/3 5Y 6/2 5Y 6/2 5Y 6/2 5Y 6/2	10YR 5/4 10YR 5/4 10YR 4/1 10YR 4/1 10YR 4/1 10YR 4/1 10YR 4/1 10YR 4/1 10YR 4/1 10YR 4/1 10YR 4/1 5Y 5/2 5Y 5/2 5Y 5/2 5Y 5/2 5Y 5/2	$\begin{array}{c} 2.5Y5/3\\ 2.5Y5/3\\ 2.5Y5/3\\ 2.5Y5/3\\ 2.5Y5/3\\ 2.5Y5/3\\ 5Y4/1\\ 5Y4/1\\ 5Y4/1\\ 5Y4/1\\ 5Y4/1\\ 5Y5/1\\ 5Y5/1\\ 5Y5/1\\ 5Y5/1\\ 5Y5/1\\ 5Y5/1\\ 5Y5/1\\ 5Y5/1\end{array}$

Note: 2.5Y 5/3—Light yellowish brown; 5Y 4/1—Dark gray; 5Y 5/1—Gray; 5Y 5/2—Olive gray; 5Y 6/2—Light olive gray; 5Y 6/3—Pale olive; 5Y 7/4—Pale yellow; 10YR 4/1— Dark gray; 10YR 5/4—Yellowish brown.

0–10 cm and  $0.02 \pm 0.00\%$  in the 10–20 cm depth samples. Thus, C/N ratios were 33 in 0–10 cm samples and 31 in 10–20 cm samples. It is not surprising that nitrogen mineralization was negative (nitrogen was not mineralized but actually removed from the soil by microorganisms) under both aerobic and anaerobic conditions. However, similar results have been obtained in soils from other CRSP sites with lower C/N ratios. Thus, organic matter decomposition in pond soils must be either nitrogen-limited, which does not seem likely at other sites with a narrow C/N ratio, or nitrogen is denitrified after mineralization. The latter is probably the general situation, but ponds at Iquitos are likely also nitrogen-limited. We do not feel that the soil incubations are providing much useful information about nutrient dynamics in ponds, and we plan to stop conducting them in future work.

### Pond Soils at Sae Kaeo, Thailand

The moisture content and dry bulk density values (Table 10) for ponds at Sae Kaeo showed the typical layering found in ponds at other sites (Munsiri et al., 1995; Boyd et al., 1997, 1998). The S horizon was about 4 cm thick, the M horizon extended from 4 to 20 cm, and the T horizon occurred between 20 and 26 cm. The bulk density of the P horizon (original soil) was around 1.5 g cm<sup>-3</sup>. Soil color was darker in the S horizon and in the upper part of the M horizon than in deeper layers (Table 11).

The soil pH was higher (5.9 to 6.2) in the S horizon than in deeper layers (Table 12). This was the result of limestone applications to ponds in the past. The original soil had a pH of 5.3. Soil acidity did not change with depth and was usually around 5 meq (100 g)<sup>-1</sup> (Table 12).

The typical decrease in total carbon and total nitrogen concentrations with depth was observed in the ponds (Table 13). The S horizon and upper part of the M horizon had carbon concentrations slightly above 2%, and concentration decreased with depth to about 0.5% in the P horizon. Nitrogen concentrations declined from about 0.2% near the surface to 0.03% in the deepest layer. The C/N ratio was about 10 to 11 in the S horizon and upper part of the M horizon. This ratio then increased to 16.7 in the deepest layer. Total sulfur tended to be slightly higher (0.03 to 0.04%) in the upper 6-cm layer than in deeper layers (0.01 to 0.02%). Nevertheless, total sulfur concentrations were very low.

Dilute-acid-extractable phosphorus concentrations were different from other ponds that have been sampled.

Table 12. Profiles for dry soil pH and exchangeable acidity in soil cores from bottoms of aquaculture ponds, Thailand. The wet soil pH was directly measured in soil core and the dry soil pH was measured in 1:1 slurries of dry soil and distilled water. Averages and standard errors of pH are given as standard pH units. Exchangeable acidity averages and standard errors are given as milliequivalents per 100 grams dry soil (meq (100 g)<sup>-1</sup>). Each entry is the average of three ponds.

<b>Depth</b> (cm)	Dry Soil pH	Exchangeable Acidity (meq (100 g) <sup>-1</sup> )
0 - 2 $2 - 4$ $4 - 6$ $6 - 8$ $8 - 10$ $10 - 12$ $12 - 14$ $14 - 16$ $16 - 18$ $18 - 20$ $20 - 22$ $22 - 24$ $24 - 26$	$\begin{array}{c} 6.2 \pm 0.2 \\ 5.9 \pm 0.2 \\ 5.7 \pm 0.2 \\ 5.8 \pm 0.4 \\ 5.6 \pm 0.3 \\ 5.6 \pm 0.2 \\ 5.7 \pm 0.3 \\ 5.6 \pm 0.3 \\ 5.5 \pm 0.3 \\ 5.5 \pm 0.4 \\ 5.5 \pm 0.4 \\ 5.5 \pm 0.4 \\ 5.5 \pm 0.4 \\ 5.3 \pm 0.5 \end{array}$	$5.07 \pm 0.71$ $5.33 \pm 0.96$ $5.87 \pm 0.71$ $6.13 \pm 1.33$ $5.60 \pm 0.80$ $5.33 \pm 0.53$ $5.07 \pm 0.27$ $4.53 \pm 0.53$ $4.80 \pm 0.46$ $4.80 \pm 0.46$ $4.27 \pm 0.53$ $4.80 \pm 0.46$ $4.80 \pm 0.46$
26–28 28–30	$5.3 \pm 0.5 \\ 5.3 \pm 0.5$	$\begin{array}{c} 5.33 \pm 0.71 \\ 5.60 \pm 0.80 \end{array}$

Table 13. Profiles of total carbon, total nitrogen, and total sulfur in soil cores from bottoms of aquaculture ponds, Thailand. Averages and standard errors are given as percentages. Each entry is the average of three ponds.

<b>Depth</b> (cm)	Total Carbon (%)	Total Nitrogen (%)	Total Sulfur (%)
0.0	0.00 + 0.51	0.00	0.00 + 0.01
0 - 2	$2.32 \pm 0.51$	$0.22 \pm 0.05$	$0.03 \pm 0.01$
2 - 4	$2.14 \pm 0.47$	$0.20 \pm 0.06$	$0.04 \pm 0.01$
4 - 6	$2.15\pm0.61$	$0.20 \pm 0.07$	$0.03\pm0.01$
6 - 8	$2.11\pm0.57$	$0.19\pm0.07$	$0.02\pm0.01$
8 - 10	$1.74\pm0.51$	$0.13\pm0.06$	$0.03\pm0.00$
10 - 12	$1.55\pm0.51$	$0.12\pm0.06$	$0.02\pm0.01$
12 - 14	$1.49\pm0.52$	$0.11\pm0.05$	$0.03\pm0.01$
14 - 16	$1.54\pm0.65$	$0.16\pm0.08$	$0.02\pm0.01$
16 - 18	$1.32\pm0.61$	$0.10\pm0.05$	$0.02\pm0.00$
18 - 20	$1.24\pm0.58$	$0.09\pm0.05$	$0.02\pm0.01$
20-22	$0.98\pm0.39$	$0.08\pm0.05$	$0.02\pm0.02$
22-24	$0.91\pm0.44$	$0.09\pm0.05$	$0.03\pm0.00$
24-26	$0.73\pm0.37$	$0.06\pm0.03$	$0.01\pm0.01$
26-28	$0.67\pm0.29$	$0.06\pm0.02$	$0.01\pm0.00$
28-30	$0.50\pm0.16$	$0.03\pm0.01$	$0.01\pm0.01$

Concentrations were between 15 and 20 ppm in the surface 10-cm layer, from 22 to 33 ppm between 10 and 26 cm, and less than 20 ppm in the 26-to-30-cm layer (Table 14). No explanation is available, but we suspect that greater phosphorus inputs were made to the ponds in the past than at present. This would explain the greater phosphorus concentrations in the lower part of the M horizon. Water-extractable phosphorus concentrations were not closely related to dilute-acid-extractable phosphorus concentrations.

Concentrations of macronutrient cations (Table 15) and micronutrients (Table 16) are within the ranges of concentrations found in pond soils from other sites (Munsiri et al., 1995; Boyd et al., 1997, 1998). Calcium, magnesium, and potassium concentrations tended to be greater in the S horizon and the

Table 14. Profiles for dilute-acid-extractable phosphorus and waterextractable phosphorus in soil cores from bottoms of aquaculture ponds, Thailand. Averages and standard errors are given as parts per million (ppm) and each entry is the average of three ponds.

<b>Depth</b> (cm)	Dilute-acid-extractable Phosphorus (ppm)	Water-extractable Phosphorus (mg l <sup>-1</sup> )
$\begin{array}{c} 0-2\\ 2-4\\ 4-6\\ 6-8\\ 8-10\\ 10-12\\ 12-14\\ 14-16\\ 16-18\\ 18-20\\ 20-22 \end{array}$	$\begin{array}{c} 20.26 \pm 6.98 \\ 20.80 \pm 7.52 \\ 18.31 \pm 5.06 \\ 15.63 \pm 2.24 \\ 20.53 \pm 6.67 \\ 24.40 \pm 7.74 \\ 27.33 \pm 9.30 \\ 25.72 \pm 8.07 \\ 31.47 \pm 13.33 \\ 33.92 \pm 14.70 \\ 33.28 \pm 16.30 \end{array}$	$\begin{array}{c} 0.049 \pm 0.023 \\ 0.061 \pm 0.035 \\ 0.058 \pm 0.035 \\ 0.045 \pm 0.013 \\ 0.084 \pm 0.050 \\ 0.101 \pm 0.053 \\ 0.094 \pm 0.048 \\ 0.058 \pm 0.023 \\ 0.091 \pm 0.055 \\ 0.106 \pm 0.054 \\ 0.125 \pm 0.076 \end{array}$
22–24 24–26 26–28 28–30	$\begin{array}{c} 29.51 \pm 17.02 \\ 22.70 \pm 11.85 \\ 18.92 \pm 9.45 \\ 11.78 \pm 3.62 \end{array}$	$\begin{array}{c} 0.161 \pm 0.118 \\ 0.109 \pm 0.078 \\ 0.106 \pm 0.072 \\ 0.066 \pm 0.042 \end{array}$

Table 15. Profiles for calcium, magnesium, potassium, and sodium in soil cores from bottoms of aquaculture ponds, Thailand. Averages and standard errors are given as parts per million (ppm). Each entry is the average of three ponds.

<b>Depth</b> (cm)	<b>Calcium</b> (ppm)	<b>Magnesium</b> (ppm)	Potassium (ppm)	<b>Sodium</b> (ppm)
0 - 2	$1,077 \pm 173$	$187\pm42$	$90\pm16$	$37.3 \pm 7.1$
2 - 4	$886 \pm 154$	$165 \pm 57$	$74 \pm 14$	$35.8\pm1.0$
4 - 6	$105 \pm 105$	$178\pm50$	$73 \pm 3$	$34.4\pm2.4$
6 – 8	$1,375 \pm 518$	$167\pm49$	$68 \pm 1$	$36.6\pm4.2$
8-10	$846\pm236$	$143\pm53$	$52 \pm 10$	$32.8\pm1.9$
10-12	$821 \pm 254$	$135 \pm 50$	$46 \pm 10$	$32.1\pm2.0$
12 - 14	$832\pm305$	$138 \pm 52$	$43 \pm 11$	$30.7\pm4.4$
14 - 16	$911 \pm 463$	$127\pm46$	$40 \pm 11$	$33.8\pm3.9$
16-18	$748\pm329$	$131 \pm 48$	$40 \pm 10$	$30.3\pm2.8$
18 - 20	$612 \pm 236$	$119 \pm 44$	$35\pm8$	$24.5\pm1.6$
20-22	$494 \pm 157$	$108 \pm 42$	$32\pm 6$	$28.0\pm1.1$
22-24	$502 \pm 201$	$125 \pm 50$	$37 \pm 10$	$28.3\pm2.8$
24 - 26	$389 \pm 163$	$102 \pm 38$	$32\pm8$	$23.2\pm1.3$
26 - 28	$403\pm175$	$110 \pm 40$	$35\pm7$	$25.1 \pm 3.4$
28-30	$366\pm163$	$111\pm45$	$35\pm10$	$33.2\pm3.5$

upper part of the M horizon than in deeper layers. This no doubt reflects the inputs of calcium and magnesium in liming material and potassium in fertilizer. Sodium concentrations showed no appreciable change with depth. Iron and manganese concentrations tended to be greater in the middle part of the M horizon than at greater depths. We do not have an explanation for this phenomenon. Zinc appears to have accumulated slightly in the S and M horizons, but copper concentrations showed no apparent trends of layering.

#### **Soil Respiration Studies**

Freshwater samples were collected and analyzed for respiration before brackishwater samples. Differences between adjacent successive 0.5-cm soil layers of freshwater samples were not statistically different (P < 0.05). Therefore, brackishwater soil samples that were sampled a few months later were sampled at 1-cm intervals to reduce the number of samples to the lowest amount necessary.

Table 16. Profiles for iron, manganese, zinc, and copper in soil cores from bottoms of aquaculture ponds, Thailand. Averages and standard errors are given as parts per million (ppm). Each entry is the average of three ponds.

$\begin{array}{cccccccccccccccccccccccccccccccccccc$	(ppm)
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\begin{array}{c} 1.75 \pm 0.09 \\ 1.69 \pm 0.29 \\ 1.89 \pm 0.04 \\ 1.83 \pm 0.08 \\ 1.54 \pm 0.21 \\ 1.61 \pm 0.19 \\ 1.61 \pm 0.26 \\ 1.46 \pm 0.17 \\ 1.50 \pm 0.21 \\ 1.34 \pm 0.14 \\ 1.20 \pm 0.15 \\ 1.33 \pm 0.26 \\ 1.21 \pm 0.14 \\ 1.41 \pm 0.15 \\ 1.43 \pm 0.21 \end{array}$

Table 17. Average concentrations and standard errors of carbon, respiration rate, and respiration rate per unit carbon in successive 0.5-cm layers of six freshwater pond soils incubated in the dark at  $25^{\circ}$ C for one week. Soil moisture in all soils and layers was  $33 \pm 2\%$ .

<b>Layer</b> (cm)	Carbon (%)	<b>Respiration</b> (mg CO <sub>2</sub> g <sup>-1</sup> )	<b>Respiration</b> <b>per g of C</b> (mg CO <sub>2</sub> g <sup>-1</sup> )
$\begin{array}{c} 0.0-0.5\\ 0.5-1.0\\ 1.0-1.5\\ 1.5-2.0\\ 2.0-2.5\\ 2.5-3.0\\ 3.0-3.5\\ 3.5-4.0\\ 4.0-4.5\\ 4.5-5.0\\ \end{array}$	$\begin{array}{c} 2.49 \pm 0.10 \\ 2.36 \pm 0.11 \\ 2.35 \pm 0.13 \\ 2.26 \pm 0.13 \\ 2.14 \pm 0.12 \\ 1.98 \pm 0.09 \\ 2.01 \pm 0.07 \\ 1.97 \pm 0.08 \\ 1.90 \pm 0.09 \\ 1.82 \pm 0.10 \end{array}$	$\begin{array}{c} 4.85 \pm 0.25 \\ 4.45 \pm 0.35 \\ 4.11 \pm 0.41 \\ 3.73 \pm 0.38 \\ 3.33 \pm 0.36 \\ 2.87 \pm 0.33 \\ 2.69 \pm 0.31 \\ 2.59 \pm 0.37 \\ 2.28 \pm 0.29 \\ 2.04 \pm 0.25 \end{array}$	$195.0 \pm 7.0 \\189.2 \pm 11.5 \\169.6 \pm 11.8 \\164.7 \pm 16.7 \\155.2 \pm 17.0 \\143.7 \pm 15.1 \\132.6 \pm 14.4 \\130.3 \pm 16.3 \\120.0 \pm 15.3 \\110.1 \pm 10.8$

Carbon concentrations in both freshwater and brackishwater ponds were higher in the first layer and decreased with increasing soil depth (Tables 17 and 18). Within the 5-cm layer samples, average carbon reduction between the upper and lower layers was 0.67% and 0.49% for freshwater and brackishwater ponds, respectively. That is, there was approximately 1.4 and 1.3 times more carbon in the uppermost layer compared to the bottom layer. Respiration was also higher in upper layers compared to lower layers in both freshwater and brackishwater ponds as would be expected from the higher carbon concentration. Microbial respiration in the 0.0-to-0.5-cm layer of freshwater ponds was 2.4 times higher compared to the 4.5-to-5.0-cm layer. This proportional difference between amounts of carbon and microbial respiration among layers can be attributed to difference in organic matter composition. The amount of reactive carbon was greater in the first layers. To account for this difference in respiration between layers caused by different concentrations of carbon, the respiration rate was normalized by dividing the respiration rate in each layer by its respective carbon concentration. Highly significant statistical differences (P < 0.01) in soil respiration (mg CO<sub>2</sub> g<sup>-1</sup> C) were found among layers in freshwater pond soils. Hsu's MCB test revealed that all layers below 2.5-cm depth differed from the first 0.0-to-0.5-cm layer. Respiration per unit carbon was also higher in the first 0-to-1-cm layer of brackishwater soil, although no statistically significant differences were found among layers. Variability of soil organic matter composition within layers of different shrimp ponds was greater than the variation among layers. All freshwater soil samples came from different ponds of Auburn University Fisheries Research Unit, while brackishwater soils were obtained from shrimp farms at different locations, which probably explains the higher variability.

The amount of carbon lost in the uppermost layer of freshwater pond bottom soil during the one-week incubation period was estimated as 0.13% (4.85 mg CO<sub>2</sub> g<sup>-1</sup> x 12 ÷ 44 x 100 g x 1 g ÷ 1000 mg). Similarly, the percentage of carbon lost in the uppermost layer of brackishwater soil was 0.05%. Average soil respiration per unit carbon was higher in freshwater soils compared to brackishwater soils. These results reflect differences in organic matter composition and factors intrinsically related to experimental conditions such as pH, moisture, texture, and salt content in soils (Boyd and Pippopinyo, 1994). Freshwater samples were collected during the northern winter at water temperatures of 12 to 14°C, while soils from Ecuador were collected when temperatures were 24 to 28°C. Organic

Table 18. Average concentrations and standard errors of carbon, respiration rate, and respiration rate per unit carbon in successive 1.0-cm layers of eight brackishwater pond soils incubated in the dark at 25°C for one week. Soil moisture in all soils was  $30 \pm 4\%$ .

<b>Layer</b> (cm)	Carbon (%)	<b>Respiration</b> (mg CO <sub>2</sub> g <sup>-1</sup> )	<b>Respiration</b> <b>per g of C</b> (mg CO <sub>2</sub> g <sup>-1</sup> )
$\begin{array}{c} 0.0 - 1.0 \\ 1.0 - 2.0 \\ 2.0 - 3.0 \\ 3.0 - 4.0 \\ 4.0 - 5.0 \end{array}$	$\begin{array}{c} 2.00 \pm 0.25 \\ 1.69 \pm 0.25 \\ 1.65 \pm 0.28 \\ 1.53 \pm 0.25 \\ 1.51 \pm 0.27 \end{array}$	$\begin{array}{c} 1.90 \pm 0.27 \\ 1.44 \pm 0.15 \\ 1.20 \pm 0.14 \\ 1.07 \pm 0.14 \\ 1.42 \pm 0.15 \end{array}$	$132.3 \pm 32.2 \\ 116.4 \pm 25.3 \\ 92.2 \pm 15.7 \\ 81.3 \pm 12.5 \\ 83.0 \pm 15.4$

matter deposited in freshwater ponds had probably been decomposing at lower rates, therefore the ratio of labile to more refractive organic matter was higher in these ponds when the samples were taken.

A meaningful interpretation of the influence of soil organic carbon on the overlying water quality such as oxygen demand and transfer and absorption of nutrients and metabolites requires knowledge not only of its concentration but also of its composition. Labile organic matter is more reactive and will exert a higher oxygen demand. This study demonstrated that concentration of labile organic carbon decreased with increasing soil depth. This method should be considered only an index of decomposability, because the experiments were performed under aerobic conditions. Under waterlogged conditions, decomposition of organic matter below the first mm of the water-soil phase is anaerobically driven. However, it is suggested that for studies directed at correlating water quality with soil organic matter characteristics, the first 1 to 2 cm of soil should be sampled.

## **ANTICIPATED BENEFITS**

The results of soil analyses from the PD/A CRSP site at Iquitos, Peru, will be useful in explaining soil and water quality phenomena observed during investigations conducted in these ponds and other ponds at the site. The data from the ponds in Peru and Thailand add to the database that is needed for delineating soil profiles and using the characteristics of soil profiles in developing a system of pond bottom soil taxonomy. The soil respiration data confirm earlier observations based on chemical and physical properties that the surface few centimeters, probably not more than the upper 5-cm layer, is the most important part of the pond soil profile with respect to influence on pond water quality.

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## PD/A CRSP Seventeenth Annual Technical Report

## NEW SITE DEVELOPMENT AND CHARACTERIZATION

Eighth Work Plan, Kenya Research 1 (8KR1) Final Report

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#### Abstract

Site development and characterization activities for the new prime site at Sagana, Kenya, began on 31 March 1997. Major undertakings that were required to make the site suitable for CRSP research included modification of the existing ponds, refurbishment of the water quality laboratory, acquisition of suitable laboratory and farm supplies and equipment, installation of a weather monitoring and recording (datalogger) system, and acquisition of a new computer system and an appropriate four-wheel-drive vehicle. Pond and laboratory renovations proceeded rapidly, and the major portions of these tasks were complete by the end of September 1997. Four existing 4,000-m<sup>2</sup> production ponds were modified to create twelve 800-m<sup>2</sup> ponds of uniform size and shape for CRSP research. Extra soil from the pond renovation was used to make seven additional ponds, ranging from 800 m<sup>2</sup> to 1,500 m<sup>2</sup>, which will be used as holding ponds, for fry production, or for activities requiring the use of hapas. Three of the extra ponds have dimensions appropriate for experimental work. Farm and laboratory supplies and equipment arrived at Sagana in September 1997. A Land Rover was purchased from the United Kingdom and shipped to Kenya on 1 July, becoming available for project use in mid-September. Installation of the weather monitoring system was begun at the end of November, and weather data were recorded beginning the first week of December. In addition, observations on pond soil and source water chemistry and annual weather patterns were undertaken to allow characterization of the site. Initial pond soil samples were collected in October 1997, and water samples for source water characterization were collected starting in October 1997. Weather data recording was begun in December 1997. Solar radiation, photosynthetically active radiation (PAR), precipitation, relative humidity, wind speed, and air temperature were recorded hourly. Four temperature probes were suspended in one pond (D6) to record pond temperature at depths of 5, 25, 50, and 75 cm as of April 1998. Preliminary analyses of pond soil samples indicate that they are mainly of the "black cotton soils" variety, high in 2:1-type clay minerals (70 to 90% clay), with cation exchange capacities typical for that type of soil (30 to 55 meq 100 g<sup>-1</sup>), and pH values ranging from 5.4 to 7.5. Lime will be required to ensure that carbon is not limiting in fertilization experiments or during production cycles. Lime requirements of 5 to 10 t ha-1 have been calculated. The phosphorus adsorption capacity of these soils is quite high. Total alkalinity and total hardness levels of water provided to the Sagana ponds through the 2-km canal system are typically 10 to 20 mg l<sup>-1</sup> as CaCO<sub>3</sub>. Source water conductivity was measured at 0.05 mmho cm<sup>-1</sup>. Detailed characterization of the pond soils and source waters for the Sagana station, as well as a summary of the first year's weather, are included in this report.

#### INTRODUCTION

Maintaining a PD/A CRSP research site in Africa preserves the global range and environmental and socioeconomic diversity of CRSP efforts, giving greater applicability of database and model achievements and providing benefits for people in all of the major regions of the world. Following the tragic events in Rwanda in 1994, including the loss of the research site and facility at Rwasave Fish Culture Station, it was necessary to select a new site to carry on CRSP research in the African region. During 1994 and 1995, several sites in eastern and southern Africa were evaluated, and Sagana Fish Farm was recommended as the most suitable for CRSP research. A Memorandum of Understanding (MOU) between Oregon State

University (OSU) and the Fisheries Department (FD) of the Government of Kenya was negotiated in 1996 and signed in March 1997, establishing Sagana Fish Farm, Sagana, Kenya, as the focal point for CRSP activities in Africa.

The Kenya Research Group of the CRSP consists of researchers at Oregon State University (OSU), Auburn University (AU), the Fisheries Department (FD) of the Government of Kenya, and the University of Arkansas at Pine Bluff (UAPB). The Department of Fisheries and Wildlife of OSU serves as the lead US institution and coordinates all Kenya research activities. OSU and AU direct the research effort and other activities through a single on-site US researcher, who works closely with the Host Country Principal Investigator (PI) and Research Associate (RA). In-country activities of UAPB (mainly socioeconomic studies) are coordinated through OSU and the onsite researcher to ensure efficient use of facilities and coordination with the Host Country PI and RA. US and Kenyan PIs have met at least once each year since the beginning of the project to evaluate progress and to prepare work plan proposals and budgets.

In order to properly conduct CRSP research and related activities at Sagana, some features of the farm needed improvement, upgrading, or modification. Most notably, the existing ponds were too large for research and the existing chemistry laboratory was inadequately equipped for carrying out the full range of CRSP analyses. Lab supplies and equipment previously used at the site in Rwanda had been completely lost and needed to be replaced. Characterization of the pond soils and the source water of the new site was needed to provide a basis for comparison with other sites and for proper analysis of experimental results.

More efficient management strategies developed through research conducted at Sagana will benefit fish farmers in Kenya, and resulting increases in fish production will partially offset the high demand for this product. Kenyan fish farmers, extension workers, and consumers will thus be the initial beneficiaries of this effort. As research continues and regional efforts are increased, fish farmers, extension workers, and planning officials throughout the region will also benefit from improved management strategies and increased production. Ultimately, all users of global CRSP models and data will benefit.

#### METHODS AND MATERIALS

The objectives of this activity were to prepare the new site at Sagana for CRSP research and to characterize the site in terms of pond soil, source water chemistry, and climatic characteristics. Existing ponds were modified so that they would be of the appropriate size and shape for aquaculture research. The chemistry lab was upgraded, equipped, and supplied with reagents to become a functional laboratory capable of performing all CRSP analyses. Fish culture supplies were upgraded or increased sufficiently to handle the increased level of farm activity required for conducting CRSP research. A four-wheeldrive vehicle was purchased for site operation, and additional computer hardware and software were provided. Soil, water, and climate attributes were observed and monitored. Soil and water sampling was undertaken according to standard CRSP analytical methods, and a weather monitoring station similar to that in use at other CRSP sites was assembled and put into 11Se.

#### **RESULTS AND DISCUSSION**

Site development and characterization activities for the new prime site at Sagana, Kenya, began immediately upon arrival of the Africa Project's resident researcher, Karen Veverica, on 31 March 1997. Pond and laboratory renovations proceeded rapidly, and the major portions of these tasks were complete by the end of September 1997. Four existing 4,000-m<sup>2</sup> production ponds were modified to create twelve 800-m<sup>2</sup> ponds of uniform size and shape for CRSP research. Extra soil from the pond renovation was used to make seven additional ponds, ranging from 800 m<sup>2</sup> to 1,500 m<sup>2</sup>, which will be used as holding ponds, for fry production, or for activities requiring the use of hapas. Three of the extra ponds have dimensions appropriate

for experimental work. Farm and laboratory supplies and equipment, including a new desktop computer, laboratory instruments, and seines, were shipped from the US on 30 June and arrived at Sagana on 3 September 1997. A Land Rover was purchased from the United Kingdom and shipped to Kenya on 1 July, becoming available for project use in mid-September. The weather monitoring system was installed at the end of November and the recording of weather data was begun the first week of December.

With these changes, Sagana Fish Farm now has sufficient pond space of appropriate sizes and shapes for conducting pond research; adequate supplies and equipment for operating the farm; a well-equipped laboratory capable of performing the necessary analyses; a good weather monitoring station; computing equipment appropriate for recording data, conducting statistical analyses, and writing reports; and an efficient, reliable vehicle for transportation. The lab can and does analyze water samples brought in by farmers, and training of station personnel continues so that they can correctly interpret the results of water analyses and make recommendations to farmers.

In addition, observations on pond soil and source water chemistry and annual weather patterns were undertaken to characterize the new site. Initial pond soil samples were collected in October 1997, and water samples for source water characterization were collected starting in October 1997. Weather data recording was begun in December 1997. Solar radiation, photosynthetically active radiation (PAR), precipitation, relative humidity, wind speed, and air temperature were recorded hourly. Four temperature probes were suspended in one pond (D6) to record pond temperature at depths of 5, 25, 50, and 75 cm as of April 1998.

## **Pond Soils**

Analyses of pond soil samples indicate that they are mainly of the "black cotton soils" variety, high in 2:1 type clay minerals (70 to 90% clay), with cation exchange capacities typical for that type of soil (30 to 55 meq 100 g<sup>-1</sup>), and pH values ranging from 5.4 to 7.5. Lime will be required to ensure that carbon is not limiting in fertilization experiments or during production cycles. Lime requirements have been calculated to be in the range of 5 to 10 t ha<sup>-1</sup>. The phosphorus adsorption capacity of these soils is quite high.

Soil cores were collected from Sagana ponds as part of the Pond Soil Characteristics and Dynamics of Soil Organic Matter and Nutrients (8PDR1) study in September 1997 (Boyd et al., 1999). Although these samples will be used in long-term studies being undertaken by the soils group, they also provide information more fully characterizing Sagana's pond soils at the beginning of the CRSPs involvement there. Phosphorus concentrations were found to be low (0.03 to 0.07% total phosphorus and 13.39 to 17.34 ppm dilute acid extractable phosphorus), supporting our expectation that phosphorus adsorption capacities would be high. Carbon concentrations in these samples were in the 2 to 5% range, and carbon:nitrogen ratios were between 10 and 20. Concentrations of exchangeable bases were high, and soil pH values were in the near-neutral range (Boyd et al., 1999). Greater detail on the characteristics of these soil samples, including profile descriptions and comparisons with samples taken at other CRSP sites, are presented by Boyd et al. (1999).

#### Source Water Chemistry

Total alkalinity and total hardness levels of water provided to the Sagana ponds through the 2-km canal system are typically 10 to 20 mg  $l^{-1}$  as CaCO<sub>3</sub>. Conductivity was measured at 0.05 mmho cm<sup>-1</sup>.

#### Weather

Weather data recorded by earlier workers at Sagana from 1987 to 1992 indicated that average monthly air temperatures ranged from a July low of about 23.5°C to a high of about 29°C in March. Over this same period of time, average monthly sunshine ranged from about 110 h (August) to approximately 245 h (March). Monthly averages for rainfall were reported to range from lows of about 2.5 mm (February) and 4 to 5 mm (July to September) to highs of around 350 mm (April) and 230 mm (November), indicating that there are two rainy periods for the Sagana area (Vanlerberghe, pers. comm., 1996). Our observations during the first year of the CRSP presence at Sagana indicate that, for purposes of conducting "warmseason" or "cool-season" experiments, there is a short cool season from June to August, and that the remainder of the year should be considered warm.

Following is a brief summary of the weather patterns that were observed at Sagana during the PD/A CRSPs first year of experimental work there. Although site preparations were begun in late March 1997, installation of the weather monitoring system was not complete until late November. Detailed daily weather observations thus began on 26 November 1997. The data for this period of time (26 November 1997 through 26 November 1998) will be accessible at the CRSP Central Database by the end of 1999. The Internet address for the CRSP Database is <br/>biosys.bre.orst.edu/crspdb/>.

#### **Climate**

A few remarks about the climate of the Sagana area will provide the setting for weather patterns observed at Sagana Fish Farm during the CRSPs first year there. Sagana is located in Kenya's central highlands region (Central Province) near the southern slopes of Mt. Kenya. It lies at an elevation of 1,230 m, and is very close to the equator, at latitude 0°39'S and longitude 37°12'E. An official climate class designation is apparently not available for Sagana, but at least two other locations in the central highlands have been classified as having Cw climates ("warm with dry winter") in the Köppen classification system, due to the relatively cool average temperatures observed there. Both nearby Embu (30 km ENE from Sagana, at an elevation of 1,493 m) and Nairobi (90 km S, at 1,791 m elevation) fall into this category. Their cool average temperatures are associated with their relatively high elevations.

Sagana's climate is unique relative to the climates of these other locations, however, being noticeably warmer than both Embu and Nairobi. Rainfall and temperature data collected by the CRSP during its first year of involvement at the site suggest that Sagana's climate would be classified as an Aw ("tropical wet and dry" or "tropical savanna") climate in the Köppen system. According to Lutgens and Tarbuck (1992), Aw and Cw climates have similar rainfall patterns and the Cw climates are simply cooler variants of Aw climates. The cooler temperature regimes of Cw climates are usually due to higher elevations. The temperature boundary between A and C climates is at 18°C; in A climates the average temperature of the coldest month is greater than 18°, whereas in C climates the average temperature of at least one month drops below 18° (Lutgens and Tarbuck, 1992). In this case, the warmer temperature regime of Sagana is apparently due to the fact that it lies in a relatively low spot in the highlands. At an elevation 561 m lower than that of Nairobi, Sagana has mean monthly temperatures that average approximately 3 to 4°C higher than those of Nairobi. Similarly, Sagana is 263 m lower than Embu, and this results in temperatures about 1 to 2°C higher at Sagana than at Embu. Both Embu and Nairobi have at least one month each year with an average temperature below 18°C, which puts them over the boundary into the Cw class. During the CRSPs first year in Sagana, there was no month with an average temperature lower than 18°C. Although the average temperature observed during July 1998 was just 18.3°C, which suggests that Sagana's climate might be very close to the boundary between the A and C classes, local residents described the 1997-1998 year as being cooler than average. If over the long term Sagana's coldest month averages above 18°C, as we suspect it does, then it indeed has an Aw climate. Climatic diagrams showing temperature and precipitation patterns for Sagana, Embu, and Nairobi are shown in Figure 1. It should be noted, however, that the diagram for Sagana reflects a single year's data rather than long-term data.

Although Sagana's climate (Aw) is warmer than the Cw climates of Embu and Nairobi, it is still characterized by having mean annual temperatures that are slightly lower and annual temperature ranges that are higher than those in the "wet tropics," i.e., in the Af and Am climates of the Köppen system. Annual temperatures in Aw climates can vary by up to 10°C, as compared with typical variations of less than 3°C in Af or Am climates (Lutgens and Tarbuck, 1992). Indeed, Sagana's variation in monthly temperatures through the CRSPs first year exhibited a range of approximately 4.1°C (Figure 1). For comparison, Figure 1 also includes a climatic diagram for Mbandaka, in the Democratic Republic of the Congo (formerly Coquilhatville, Zaire), which has an Af climate. Mbandaka's average monthly temperatures are approximately 2.5°C warmer than those at Sagana, and the annual temperature variation at Mbandaka is only about 1.3°C.

#### Sagana's Weather During the 1997–1998 Year

Most of the 1997-1998 year at Sagana was warm but not hot, with daily average temperatures ranging from 19 to 23°C. The warmest period was from February through April, and the highest daily maximum temperature observed was 30.69°C, recorded on 8 April 1998. There was a distinct cool season between June and August, with average temperatures ranging from 17 to 19°C during this time. However, the lowest daily minimum temperature was 12.12°C, recorded on 1 October 1998. This seasonal pattern at Sagana is apparent from the air temperature data shown in Figures 1 and 2 and summarized in Table 1. Low average air temperatures from June through August were the result of low daily maximum temperatures; daily minimum temperatures were fairly constant during this period (Figure 2). The low daily maxima were due to low solar radiation levels (Figures 3 and 4). Even though there was not much rain during this period (Figure 5), the skies were overcast much of the day.

Photosynthetically active radiation (PAR) is much higher at this site than at the Rwanda site, which is only  $1.5^{\circ}$  farther south in latitude. At Butare, Rwanda, PAR rarely measured greater than 35 Einsteins m<sup>-2</sup> d<sup>-1</sup>, and readings in the 20s were



Figure 1. Climatic diagrams for four African sites located relatively close to the equator, including Sagana, Embu, and Nairobi, Kenya, and Coquilhatville, Zaire (now renamed Mbandaka, Democratic Republic of the Congo). In each diagram, the temperature scale is on the left and the precipitation scale is on the right. Vertical bars represent average monthly precipitation and curved lines represent average monthly temperatures. The dotted horizontal line at 18°C shows the temperature boundary between A and C climates. Note the narrow annual temperature ranges of the sites in Kenya (Aw and Cw climates), and the very narrow range at Coquilhatville (Af climate). Also note the similarity between the precipitation and temperature patterns of Sagana, Embu, and Nairobi (see text for discussion). Sagana data are from December 1997 to November 1998). Data for Nairobi and Coquilhatville are from Lutgens and Tarbuck (1992), and Embu data are from the WorldClimate website at <www.worldclimate.com/cgi-bin/grid.pl?gr=N00E037>.

Table 1.Approximate seasonal temperature ranges, in °C, at SaganaFish Farm during the period from December 1997 through<br/>November 1998. Temperature ranges are from five-day<br/>running averages recorded by the datalogger system.

	<b>Cool Season</b> (June to August)	Warm Season (September to May)
Daily Minima	14–16	15–19
Daily Maxima	20–24	23–30
Daily Averages	17–19	19–23

more common. At Sagana, Rwanda-like readings are common in cool season, but during the rest of the year, values of 30 to 50 Einsteins m<sup>-2</sup> d<sup>-1</sup> are more common (Figure 3). The relationship between solar radiation as MJ m<sup>-2</sup> d<sup>-1</sup> and PAR (Einsteins m<sup>-2</sup> d<sup>-1</sup>) for the reporting period is shown in Figure 6. In December 1997 and January and February 1998, temperatures were lower than usual, and rainfall was greater than usual. This was due to the El Niño rains, which started in October 1997 and continued into 1998 (Figure 5). Total rainfall for the interval from 26 November 1997 to 26 November 1998 was 1,385 mm, as compared with a total of 1,570 mm for 1997 and a 30-year average of 1,166 mm (Figure 5).

Evaporation was measured at the Ministry of Agriculture weather station, just behind the hatchery at the station. A rain gauge at the Ministry of Agriculture station was used in the evaporation calculations through August 1998, but the rain gauge was not functional from September on, with the result that no evaporation calculations could be made after 31 August 1998. During the 303 days that evaporation was measured, values ranged from 0 to 7.4 mm d<sup>-1</sup>, with an average of 3.2 mm evaporation d<sup>-1</sup>.



Figure 2. Five-day running averages of daily average, daily maximum, and daily minimum air temperatures at Sagana Fish Farm, Sagana, Kenya, between November 1997 and December 1998.



 Figure 3. Photosynthetically active radiation (PAR) in microEinsteins m<sup>-2</sup> d<sup>-1</sup> for the period 26 November 1997 through 3 December 1998 at Sagana Fish Farm, Sagana, Kenya.

Because the weather station is situated directly on the pond banks, maximum humidity near 100% was observed every night except during intervals when the ponds were drained. Average daily humidity observations for the year are shown in Figure 7. Average monthly humidity ranged from a high of 79.12% in July to a low of 63.35% in October.

Daily mean wind speed was lowest during the cool season (Figure 8). During the rest of the year, wind speed was greatest in the late afternoon (between 1600 and 1800 hours) on bright sunny days. Ponds mixed well to a depth of about 50 cm on windy days. Dissolved oxygen levels at 75 cm depth increased slightly after 1600 hours on windy days. Data on pond water temperatures and stratification will be presented elsewhere (Veverica et al., 2000).

#### **ANTICIPATED BENEFITS**

Establishing a new prime research site in Africa maintains the global range of environmental and socioeconomic diversity of the overall PD/A CRSP research effort. Developing the pond area at Sagana Fish Farm to include a complete set of research ponds of an appropriate size allows for proper replication of experimental treatments, and ensuring that the ponds are of uniform size and shape lends validity to results of research



Figure 4. Relationship between maximum air temperature (°C) and photosynthetically active radiation (PAR), in microEinsteins m<sup>-2</sup> d<sup>-1</sup>, at Sagana Fish Farm during the period 26 November 1997 through 3 December 1998.



Figure 5. Monthly precipitation at Sagana Fish Farm for 1997, 1998, and the 25-year average for the period from 1972 to 1997.



Figure 6. The relationship between solar radiation as MJ m<sup>-2</sup> d<sup>-1</sup> and photosynthetically active radiation (PAR), in Einsteins m<sup>-2</sup> d<sup>-1</sup>, at Sagana Fish Farm during the period 26 November 1997 through 3 December 1998.



Figure 7. Average daily humidity observations at Sagana Fish Farm, Sagana, Kenya, during the period 26 November 1997 through 3 December 1998 (average of daily maximum and minimum observations). Maximum humidity near 100% was observed most nights except during periods when ponds were drained.



Date

Figure 8. Daily mean wind speeds, in km h<sup>-1</sup>, for the period 26 November 1997 through 3 December 1998 at Sagana Fish Farm, Sagana, Kenya.

conducted there. A fully functioning laboratory is an absolute necessity for carrying out the protocols established by the CRSP Technical Committee. With these physical improvements in place and the provision of a suitable vehicle for on-site operations and extension efforts and other farm operation tools, Sagana Fish Farm now has the physical capacity to conduct high-quality research; this in turn will contribute to the development of more efficient management strategies for use by fish farmers in the surrounding provinces, in the rest of Kenya, and in the region. Fish farmers will ultimately realize more efficient production in their ponds, and consumers will benefit from the resulting increases in the availability of food fish. Sagana Fish Farm also has the potential to become a center for practical training in the subjects of sex-reversal, pond construction, and laboratory techniques and could in this capacity serve not only Kenya but also other countries in the region.

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## EFFECT OF MUD TURBIDITY ON FERTILIZATION, AND AN ANALYSIS OF TECHNIQUES TO MITIGATE TURBIDITY PROBLEMS IN WET SEASON

Eighth Work Plan, Thailand Research 1 (8TR1) Final Report

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#### Abstract

The experiment was conducted in fifteen earthen ponds at the Asian Institute of Technology, Thailand, from June to November 1998 to assess the effects of various turbidity mitigation techniques on fish growth and water quality. The five treatments were: A) control; B) covering the upper 50 cm of pond dikes with black plastic to prevent turbidity from runoff; C) covering pond bottoms with green manure (terrestrial weeds) to alter soil texture; D) covering pond bottoms with small-mesh (1-cm) net to prevent turbidity from fish disturbance; and E) covering pond dikes with rice straw. All ponds were fertilized weekly with chicken manure at a rate of 500 kg ha<sup>-1</sup> (dry matter basis) supplemented with urea and triple superphosphate (TSP) to provide 28 kg N ha<sup>-1</sup> wk<sup>-1</sup> and 7 kg P ha<sup>-1</sup> wk<sup>-1</sup>. Sex-reversed all-male Nile tilapia (*Oreochromis niloticus*) were stocked at 2 fish m<sup>-2</sup> at a size of  $19.0 \pm 1.0$  g. No significant differences of fish survival were found among all treatments. The straw- and weed-covered treatments resulted in significantly higher fish growth and yield. In contrast, the edge- and bottom-covered treatments were not effective. The straw-covered treatment was the innovation of the experiment. Pond water in straw-covered ponds was green throughout the experimental period with low colloidal turbidity, which resulted in the highest fish yield among all treatments. The straw-covered treatment was probably the best mitigating technique in wet season.

#### INTRODUCTION

Mud turbidity is a global problem in fish ponds with heavy clay dikes and bottoms. Colloidal clay particles from the dikes and bottom (as well as from runoff and source water) suspended in the water column inhibit plankton growth by reducing light penetration and by binding with mineral nutrients from water as well as with plankton cells (Avinimelech et al., 1981, 1982). High mud turbidity usually causes acidity, low nutrient levels, and limited light penetration for photosynthesis (Boyd, 1990), and thus results in reduced primary production (Diana et al., 1991). With only fertilizer inputs, turbidity often limits production and growth of fish (Buck 1956; Banarjea and Ghosh, 1963). From these points of view, mitigation of mud turbidity is essential to enhance and allow normal phytoplankton growth in response to fertilization.

The purpose of this study was to evaluate several mud turbidity mitigation techniques in order to: 1) assess effects of different mitigation techniques on fish growth, and 2) find a suitable approach for turbidity mitigation during the rainy season.

#### METHODS AND MATERIALS

The experiment was conducted in fifteen 200-m<sup>2</sup> earthen ponds with an average depth of 1.0 m at the Asian Institute of Technology (AIT), Thailand. All ponds were fertilized weekly

with chicken manure at a rate of 500 kg ha<sup>-1</sup> (dry matter basis) supplemented with urea and triple superphosphate (TSP) to provide 28 kg N ha<sup>-1</sup> wk<sup>-1</sup> and 7 kg P ha<sup>-1</sup> wk<sup>-1</sup>. Sex-reversed all-male Nile tilapia (*Oreochromis niloticus*) were stocked at 2 fish m<sup>-2</sup> at a size of  $19.0 \pm 1.0$  g (mean  $\pm$  SE) on 24 June 1998.

The ponds were grouped randomly for five treatments, with triplicate ponds for each treatment. The five treatments were: A) control; B) covering upper 50 cm of pond dikes with black plastic to prevent turbidity from runoff (edge-covered); C) covering pond bottoms with green manure (terrestrial weeds) to alter soil texture (weed-covered); D) covering pond bottoms with small-mesh (1-cm) net to prevent turbidity from fish disturbance (bottom-covered); and E) covering pond dikes with rice straw (straw-covered).

Water quality analysis was conducted biweekly by taking integrated water column samples at 0900 h from walkways extending to the center of the ponds. Pond water samples were analyzed for dissolved oxygen (DO), temperature, pH, alkalinity, total ammonium nitrogen (TAN), nitrite nitrogen, nitrate-nitrite nitrogen, total Kjeldahl nitrogen (TKN), soluble reactive phosphorus (SRP), total phosphorus (TP), chlorophyll *a*, total suspended solids (TSS), and total volatile solids (TVS) using standard methods (APHA, 1980; Egna et al., 1987). Secchi disk depth was measured at 0900 h every two days in all the ponds throughout the experimental period. Diel measurements for temperature, DO, and pH were conducted monthly for each pond at three different depths (25 cm below water surface, middle, and 25 cm above pond bottom) at 0600, 0900, 1400, 1600, 1800, and 0600 h, and alkalinity and TAN were determined at 0900, 1600, and 0600 h.

Prior to stocking, DO concentration in the weed-covered ponds was monitored at 0600 h daily for two weeks to check the changes in DO concentration. Weeds grown at the bottom of those ponds were uprooted or cut to 10–20 cm high before flooding.

Bottom soil samples collected from nine points in each pond were air-dried, mixed, and homogenized. A subsample was taken from the homogenized sample for analyzing total nitrogen, total phosphorus, and organic matter at the beginning and end of the experiment.

To avoid causing turbidity, fish were not sampled during the entire 149-day experimental period, until harvest on 20 November 1998. Final biomass and numbers were determined. Daily weight gain (DWG, g fish<sup>-1</sup> d<sup>-1</sup>), yield (kg pond<sup>-1</sup>), and extrapolated yield (kg ha<sup>-1</sup> yr<sup>-1</sup>) were calculated.

Data were analyzed statistically by analysis of variance (ANOVA) using the SPSS 7.0 statistical software package. Differences were considered significant at an alpha level of 0.05. Results are reported with mean  $\pm$  1 standard error (SE).

#### RESULTS

Fish growth was best in the straw-covered treatment and poorest in the bottom-covered treatment (Table 1). Final mean weight, mean DWG, and net fish yield in the straw-covered treatment were significantly (P < 0.05) higher than those in the control, edge-, and bottom-covered treatments, but not significantly (P > 0.05) different from those in the weedcovered treatment. There were no significant (P > 0.05) differences of final mean weight, mean weight gain, or mean DWG among the control, edge-, bottom-, and weed-covered treatments although fish yields in the weed-covered treatment were significantly higher (P < 0.05). However, no significant

Table 1. Growth performance of Nile tilapia in the 149-day experiment.

Performance	Treatment					
	Control	Edge-covered	Weed-covered	Bottom-covered	Straw-covered	
Initial Biomass (kg pond <sup>-1</sup> ) Initial Mean Wt. (g fish <sup>-1</sup> ) Final Biomass (kg pond <sup>-1</sup> ) Final Mean Wt. (g fish <sup>-1</sup> ) Mean Wt. Gain (g fish <sup>-1</sup> ) Mean DWG (g fish <sup>-1</sup> d <sup>-1</sup> ) Net Fish Yield (kg pond <sup>-1</sup> ) Extrapolated Net Fish Yield (kg ha <sup>-1</sup> yr <sup>-1</sup> )	$\begin{array}{c} 7.6 \pm 0.1^{a} \\ 19.1 \pm 0.3^{a} \\ 22.2 \pm 11.5^{a} \\ 124.9 \pm 19.0^{a} \\ 105.8 \pm 9.1^{a} \\ 0.67 \pm 0.07^{a} \\ 14.6 \pm 5.3^{a} \\ 1,459 \pm 528^{a} \end{array}$	$\begin{array}{c} 8.0\pm0.4^{a}\\ 19.9\pm0.6^{a}\\ 26.8\pm7.4^{a}\\ 90.0\pm32.8^{a}\\ 70.1\pm16.0^{a}\\ 0.47\pm0.12^{a}\\ 18.8\pm3.6^{a}\\ 1,878\pm366^{a}\\ \end{array}$	$\begin{array}{c} 7.9\pm 0.3^a\\ 19.7\pm 0.7^a\\ 56.6\pm 16.4^b\\ 184.4\pm 61.0^{ab}\\ 164.8\pm 28.1^{ab}\\ 1.10\pm 0.21^{ab}\\ 48.5\pm 7.6^b\\ 4,853\pm 760^b\end{array}$	$\begin{array}{c} 7.9 \pm 0.3^{a} \\ 19.8 \pm 0.5^{a} \\ 20.6 \pm 11.7^{a} \\ 83.4 \pm 39.6^{a} \\ 63.6 \pm 19.1^{a} \\ 0.43 \pm 0.12^{a} \\ 12.7 \pm 5.5^{a} \\ 1266 \pm 554^{a} \end{array}$	$\begin{array}{c} 7.9\pm0.4^{a}\\ 19.8\pm0.5^{a}\\ 68.3\pm28.7^{b}\\ 235.5\pm123.1^{b}\\ 233.7\pm58.0^{b}\\ 1.57\pm0.37^{b}\\ 60.9\pm13.6^{b}\\ 6,090\pm1,366^{b} \end{array}$	
Survival (%)	$45.8\pm12.9^{\rm a}$	$75.8\pm8.2^{\rm a}$	$76.8\pm1.3^{\rm a}$	$64.8\pm13.9^{\rm a}$	$70.1\pm9.8^{\rm a}$	

\* Mean values with different superscript letters in the same row are significantly different (P < 0.05).

Table 2. Water quality parameters at the end of the experiment.

Water Quality			Treatment		
	Control	Edge-covered	Weed-covered	Bottom-covered	Straw-covered
DO at Dawn (mg $l^{-1}$ )	$0.9 \pm 0.1^{a}$	$18 \pm 01^{b}$	$25 \pm 0.3^{b}$	$0.8 \pm 0.3^{a}$	$1.2 \pm 0.3^{a}$
Temperature (°C)	$27.7 \pm 0.2^{a}$	$28.0 \pm 0.2^{a}$	$28.6 \pm 0.1^{a}$	$27.6 \pm 0.3^{\circ}$	$28.1 \pm 0.1^{a}$
Alkalinity (mg CaCO <sub>3</sub> $l^{-1}$ )	$130.0 \pm 19.1^{a}$	$99.3 \pm 8.4^{\text{a}}$	$90.7 \pm 13.9^{\text{ a}}$	$115.3 \pm 16.2^{a}$	$152.0 \pm 2.3^{a}$
pH	$7.8\pm0.4$ a	$7.7\pm0.2$ a	$8.1\pm0.1$ a	$8.0\pm0.5$ a	$8.2\pm0.3$ a
TKN (mg l <sup>-1</sup> )	$7.56\pm1.38$ a	$6.28\pm1.34^{\text{ a}}$	$5.74 \pm 0.59$ $^{\rm a}$	$5.99 \pm 0.50^{\mathrm{a}}$	$6.54\pm0.33$ a
TAN $(mg l^{-1})$	$0.57\pm0.25$ $^{\rm a}$	$1.22 \pm 0.12^{\mathrm{b}}$	$0.72\pm0.04$ a	$0.50\pm0.43$ a	$0.69\pm0.13$ a
Nitrite Nitrogen (mg l <sup>-1</sup> )	$0.11\pm0.05^{\rm a}$	$0.22\pm0.08$ a	$0.07\pm0.01$ $^{\rm a}$	$0.09\pm0.04^{\rm \ a}$	$0.13\pm0.04^{\rm \ a}$
Nitrate-Nitrite Nitrogen (mg l <sup>-1</sup> )	$0.69 \pm 0.30^{a}$	$0.73 \pm 0.05$ a	$0.21\pm0.05$ $^{\rm a}$	$0.69 \pm 0.36$ a	$0.32\pm0.13^{\mathrm{a}}$
Total Phosphorus (mg l <sup>-1</sup> )	$1.05\pm0.08$ $^{\rm a}$	$0.86\pm0.13^{\text{ a}}$	$0.48 \pm 0.04$ <sup>b</sup>	$0.85 \pm 0.15$ a	$0.91 \pm 0.06$ a
SRP (mg $l^{-1}$ )	$0.88\pm0.06^{\rm \ a}$	$0.38 \pm 0.06$ <sup>b</sup>	$0.09 \pm 0.09$ <sup>c</sup>	$0.64 \pm 0.12$ a	$0.60 \pm 0.19^{\mathrm{a}}$
Secchi Disk Depth (cm)	$8.3\pm0.3$ a	$12.7 \pm 0.7^{\mathrm{b}}$	$14.0 \pm 1.5^{\text{ b}}$	$10.0 \pm 2.6^{\text{ b}}$	$12.7 \pm 3.7^{\mathrm{b}}$
Chlorophyll $a (mg l^{-1})$	$153.2\pm79.4$ $^{\rm a}$	$203.8\pm19.6^{\rm \ a}$	$171.6 \pm 68.2$ a	$45.2\pm18.1^{\rm a}$	$93.3\pm64.9^{\mathrm{a}}$
TSS (mg $l^{-1}$ )	$201.9 \pm 29.8$ <sup>a</sup>	$98.3 \pm 19.9^{\mathrm{b}}$	$107.1 \pm 12.7$ <sup>b</sup>	$119.7 \pm 17.8$ <sup>b</sup>	$103.6 \pm 13.0^{\rm b}$
TVS (mg l <sup>-1</sup> )	$39.9\pm14.6^{\rm \ a}$	$20.4\pm5.6^{\rm \ a}$	$32.0\pm3.5{}^{\rm a}$	$31.9\pm13.6^{\text{ a}}$	$30.3\pm8.7^{\mathrm{a}}$

\* Mean values with different superscript letters in the same row are significantly different (P < 0.05).

Water Quality			Treatment		
	Control	Edge-covered	Weed-covered	Bottom-covered	Straw-covered
$DO$ at $Dawn$ (mg $l^{-1}$ )	$2.0 \pm 0.2^{a}$	$30 \pm 0.3^{b}$	$23 \pm 0.2^{a}$	$2.1 \pm 0.2^{a}$	$1.7 \pm 0.2^{a}$
Temperature (°C)	2.0 ± 0.2 29.7 ± 0.2 °	$30.0 \pm 0.3^{\circ}$	$2.5 \pm 0.2$ $30.7 \pm 0.1$ <sup>b</sup>	$2.1 \pm 0.2$ $29.9 \pm 0.2^{a}$	$1.7 \pm 0.2$ $30.1 \pm 0.2^{\circ}$
Alkalinity (mg CaCO <sub>3</sub> $l^{-1}$ )	$101.5\pm4.3^{\mathrm{a}}$	$90.8\pm2.6^{\rm \ a}$	$93.9\pm6.4^{\mathrm{a}}$	$91.5\pm3.9$ a	$158.0 \pm 6.8^{\mathrm{b}}$
pH	$7.7\pm0.1$ $^{\mathrm{a}}$	$7.8\pm0.1^{\rm \ a}$	$7.5\pm0.1$ a	$7.7\pm0.1$ $^{\mathrm{a}}$	$8.1\pm0.1$ <sup>b</sup>
$TKN (mg l^{-1})$	$5.26 \pm 0.50^{\mathrm{a}}$	$4.22\pm0.37^{\rm \ a}$	$4.17\pm0.46^{\mathrm{a}}$	$4.59 \pm 0.42^{\rm a}$	$4.81\pm0.40^{\mathrm{a}}$
TAN (mg $l^{-1}$ )	$1.26\pm0.16^{\mathrm{a}}$	$1.55\pm0.14$ $^{\rm a}$	$1.14\pm0.15^{\rm a}$	$1.37 \pm 0.17$ a	$0.78 \pm 0.12^{\mathrm{b}}$
Nitrite Nitrogen (mg l <sup>-1</sup> )	$0.14\pm0.03^{\mathrm{a}}$	$0.23 \pm 0.03^{\mathrm{b}}$	$0.07 \pm 0.02^{\mathrm{a}}$	$0.15\pm0.03^{\mathrm{a}}$	$0.12\pm0.03^{\mathrm{a}}$
Nitrate-Nitrite Nitrogen (mg l <sup>-1</sup> )	$0.69 \pm 0.20^{\mathrm{a}}$	$0.60 \pm 0.07$ $^{\rm a}$	$0.21 \pm 0.03^{\mathrm{b}}$	$0.79 \pm 0.19^{\mathrm{a}}$	$0.26 \pm 0.06$ <sup>b</sup>
Total Phosphorus (mg l <sup>-1</sup> )	$0.59 \pm 0.06$ a	$0.43\pm0.04^{\rm \ a}$	$0.27 \pm 0.02^{\mathrm{b}}$	$0.49\pm0.06^{\rm\ a}$	$0.64\pm0.03$ a
SRP (mg $l^{-1}$ )	$0.29 \pm 0.05^{\mathrm{a}}$	$0.18\pm0.03^{\rm \ a}$	$0.02 \pm 0.00^{\mathrm{b}}$	$0.25 \pm 0.05$ a	$0.24\pm0.04^{\mathrm{a}}$
Secchi Disk Depth (cm)	$11.4\pm0.3^{\rm a}$	$14.8\pm0.3^{\rm \ b}$	$26.1 \pm 0.9^{\text{ d}}$	$15.3 \pm 0.5^{\text{b}}$	$18.7\pm0.5^{ m c}$
Chlorophyll $a (mg l^{-1})$	$84.4\pm13.5^{\rm \ ab}$	$48.2\pm8.8^{\mathrm{a}}$	$104.5 \pm 11.8^{\mathrm{b}}$	$49.2\pm11.5^{\mathrm{a}}$	$121.7 \pm 11.3^{\mathrm{b}}$
TSS $(mg l^{-1})$	$111.4\pm10.9^{\rm \ a}$	$82.2 \pm 6.1^{\mathrm{b}}$	$162.9 \pm 42.2$ a	$103.4\pm21.25^{\rm a}$	$87.1 \pm 10.3^{\mathrm{b}}$
TVS (mg $l^{-1}$ )	$31.9\pm3.7^{\mathrm{b}}$	$22.1\pm1.9^{a}$	$28.4\pm3.0^{\mathrm{b}}$	$29.0\pm3.4^{\mathrm{b}}$	$36.9\pm4.6^{\mathrm{b}}$

Table 3. Mean water quality variables throughout the experiment.

\* Mean values with different superscript letters in the same row are significantly different (P < 0.05).

difference of survival (P > 0.05) was found among all treatments.

The final and overall means of water quality parameters are listed in Tables 2 and 3, respectively. Water temperature and pH throughout the experimental period in all ponds ranged from 27.0 to 38.4°C and 5.8 to 10.1, respectively. There was no significant difference of pH among treatments at the end of the experiment; the overall mean of pH in the straw-covered treatment was significantly (P < 0.05) higher than the mean of pH in other treatments. The measured DO concentrations at dawn fluctuated between 0.2 and 7.4 mg l<sup>-1</sup> over the entire culture period (Figure 1). The edge-covered treatment had significantly (P < 0.05) higher overall mean DO concentration than the mean DO concentrations in other treatments. Lowest overall mean DO concentration was observed in the strawcovered treatment, but not significantly (P > 0.05) different from the control, weed-, edge-, and bottom-covered treatments. Final DO concentration in the control, bottom-, and strawcovered treatments was significantly (P < 0.05) lower than DO concentration in the edge- and weed-covered treatments. In the straw-covered treatment the DO concentrations at dawn were low at the beginning of the experiment and were similar to other treatments after two months before dropping slightly at the end of the experiment (Figure 1). Un-ionized ammonia nitrogen concentrations were generally low and similar among all treatments (Figure 1). Overall mean alkalinity concentration over the experimental period in the straw-covered treatment was significantly (P < 0.05) higher than mean alkalinity concentration in other treatments (Table 3). No significant (P > 0.05) differences were found for overall mean TKN among all treatments. Overall mean nitrite nitrogen concentration was significantly (P < 0.05) higher in the edge-covered treatment than that in other treatments. Final nitrite and nitrate nitrogen concentrations were not significantly (P > 0.05) different among all treatments, but overall mean nitrate nitrogen concentrations in the control, edge-, and bottom-covered treatments were significantly (P < 0.05) higher than those in the other two treatments. Chlorophyll a concentrations increased over the experimental period in all treatments (Figure 1). The overall mean chlorophyll a concentration was significantly (P < 0.05) higher in the straw- and weed-covered treatments in

comparison with the other treatments (Table 3). Overall mean TSS concentrations in the straw- and edge-covered treatments were significantly (P < 0.05) lower than mean TSS concentrations in the control, bottom-, and weed-covered treatments





Figure 1. Changes in concentrations of dissolved oxygen (DO) at dawn, un-ionized ammonia nitrogen, alkalinity, and chlorophyll *a* in pond water during the 149-day experiment.





Figure 2. Changes in total suspended solids (TSS) and total volatile solids (TVS) in pond water during the 149-day experiment.

(Table 3). The straw-covered treatment had lower overall mean TSS, but higher TVS compared to the other treatments (Figure 2). Overall TSS was highest in the weed-covered treatment, but the control had the highest final TSS, which was significantly (P < 0.05) higher than the final TSS in other the treatments (Tables 2 and 3). Secchi disk depth declined over the experimental period (Figure 3). The overall mean value of Secchi disk depth in the weed-covered treatment was significantly (P < 0.05) higher than the overall mean value of Secchi disk depth in the weed-covered treatment was significantly (P < 0.05) higher than the overall mean value in the straw-covered treatment, which was further significantly (P < 0.05) higher than the overall mean value in the other treatments. The control had the lowest overall mean value of Secchi disk depth (P < 0.05).

#### DISCUSSION

Among the mitigation techniques, the straw- and weedcovered treatments, especially the straw-covered treatment, resulted in significantly (P < 0.05) higher fish growth rates and yields compared with the control ponds. DWG of fish and net fish yield in the straw-covered treatment were 2.3 and 4.2 times higher, respectively, than DWG and net fish yield in the control.



Figure 3. Changes in Secchi disk depth during the 149-day experiment.

Due to the decomposition of rice straw, DO concentration at dawn in the straw-covered treatment was rather low during the first two months, but increased towards the end of the experimental period. In contrast, DO concentration in the bottom-covered treatment decreased with time, from a relatively high level at the beginning to quite a low level at the end of the experiment. This probably indicated the accumulation of organic matter derived from fertilization with chicken manure and fish wastes on undisturbed pond bottoms. Decreasing DO concentration and probably low primary productivity, as indicated by low concentrations of chlorophyll *a* in these ponds, attributed to the retarded growth and low yield of fish.

In the bottom-covered treatment, netting material prevented turbidity caused by fish disturbance of pond bottoms; however, this may have reduced nutrient release from pond mud. Compared to the control, total phosphorus and chlorophyll *a* were relatively low in the bottom-covered treatment. Plankton growth is regulated to some extent by the concentration of phosphorus in the mud of natural waters (Boyd, 1990). However, compared to the control, ponds with covered bottoms did not evidence a significant reduction in fish growth, implying that Nile tilapia feed mainly on phytoplankton in the water column rather than graze on the pond bottom, which is consistent with the results obtained during the dry season in a previous experiment (Lin et al., 1999).

The edge-covered treatment was designed to prevent turbidity caused by dike runoff during the rainy season. In the dry season, there was no significant difference in most water quality parameters and fish growth performance between the control and the edge-covered treatment (Lin et al., 1999). In contrast, the overall TSS and Secchi disk depth in the edgecovered treatment were significantly different from TSS and Secchi disk depth of the control in the present experiment conducted during the wet season, indicating that the edgecovered treatment was effective in mitigating turbidity problems caused by runoff. However, the overall mean chlorophyll a level in the edge-covered treatment was apparently lower than the chlorophyll *a* level of the control, although there was no significant difference. Also the edge-covered treatment did not result in increased fish yields compared with the control. The results revealed that grazing on pond edges by Nile tilapia was less important than feeding on phytoplankton in the water column.

In a previous dry-season experiment, high fish mortality was observed in the weed-covered treatment due to DO depletion caused by the decomposition of terrestrial weeds in ponds during the first month of the experiment (Lin et al., 1999). However, there were no significant differences in survival among all treatments in the present experiment due mainly to the better timing of stocking fish. Ponds containing macrophytes usually had clearer water and, upon decay, dead vegetation increased the concentration of carbon dioxide, decreased pH, and resulted in the precipitation of colloidal clay (Irwin and Stevenson, 1951). In the present experiment, significantly higher Secchi disk visibility and lower TSS were observed in the weed-covered treatment at the end of the experiment compared with the control.

Reasonable Secchi disk visibility (20 to 30 cm) with a high level of chlorophyll *a* resulted in significantly greater fish yield in the straw-covered treatment. The decay of rice straw at the

beginning of the experiment, although it reduced DO concentration, provided additional nutrients for plankton growth. Meanwhile, turbidity from dike runoff was mitigated. The straw-covered treatment was probably the best mitigating technique in wet season. However, more research on physical and chemical changes of pond water caused by rice straw is needed.

## **ANTICIPATED BENEFITS**

The results generated in this study, in addition to similar studies in turbidity control at the other CRSP sites, will link bottom soil characteristics and water quality management for semi-intensive fish ponds. Turbidity problems prevail in many rain-fed ponds in Thailand, Cambodia, and Laos, where the available fertilizer input is reduced in effectiveness by turbidity, resulting in poor fish yields and a lack of interest in managing such ponds. The topic of turbidity control has been considered a priority by the Royal Thai Government Department of Fisheries and also by the Asian Institute of Technology outreach project.

## ACKNOWLEDGMENTS

The authors wish to acknowledge the Asian Institute of Technology, Thailand, for providing research, field, and laboratory facilities. Mr. Manoj Y. and Mr. Supat P. are greatly appreciated for their field and laboratory assistance.

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## PD/A CRSP Seventeenth Annual Technical Report

## INTENSIFICATION OF TILAPIA PRODUCTION: EFFECTS OF FEEDING AT DIFFERENT STOCKING RATES ON POND WATER QUALITY

Eighth Work Plan, Honduras Research 1 (8HR1) Final Report

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#### Abstract

Commercial production of tilapia is expanding rapidly in Central America, and hyper-intensive production systems often are being promoted to potential fish farmers. There are few or no sustainable technological packages for profitable tilapia production available to tilapia farmers in Central America. Commercial tilapia farms in Honduras routinely stock 5 to 7 fish m<sup>-2</sup>. The goals of the proposed research were to develop sustainable pond management practices for small- to mediumscale commercial tilapia farmers in Honduras by evaluating the effect of stocking rate on tilapia yield and production economics and on pond nutrient budgets. Tilapia stocking rates of 2, 5, or 8 fish m<sup>-2</sup> during a 240-d grow-out were to be tested in 0.1-ha earthen ponds at the Centro Nacional de Investigación Piscícola El Carao, Comayagua, Honduras. A total of 60,000 fish were needed for stocking experimental ponds. Research ponds were not available until 11 June 1998 because the Eighth Work Plan Global Experiment (8FFR1H) had to be extended beyond its programmed duration because fish growth continued. In May 1998 the well at the El Carao station failed, leaving the wet lab and fish transport facilities without water. Water to the wet lab and for fish transport had been restored partially by mid-July. In order to avoid further delays in Eighth Work Plan implementation it was decided to proceed, albeit with some risk, with the transfer of tilapia from nursery ponds to grow-out ponds to begin this experiment. Transfer took place on 21 July 1998. Unfortunately, approximately 40,000 of the more than 60,000 fingerlings in the nursery ponds did not survive the transfer process because of inadequate supply of water to the wet lab. Initiation of this experiment was delayed until December 1998 while a new group of fingerlings were reared to 50 to 100 g. Fortunately, there were adequate fingerlings in inventory at the El Carao station to allow the revised schedule to be met. On 30-31 October 1998 the torrential rains of tropical storm Mitch caused the El Carao station (as well as many other places in Honduras) to flood, which resulted in mass escape of fish in ponds. Thus, it became impossible to complete experiment 8HR1.

#### INTRODUCTION

In order to increase the profitability of tilapia culture, intensification of pond management systems is necessary to produce larger fish for urban markets (450 to 500 g) and export markets (500 to 700 g). Tilapia were stocked at 20,000 fish ha<sup>-1</sup> in semiintensive production with fertilizers and/or supplemental feed. Intensification involves increased stocking rates and use of high-quality formulated diets. Tilapia yields of 11,000 to 15,000 kg ha<sup>-1</sup> in five to eight months were reported in Thailand in fertilized and fed ponds stocked with 3 fish m<sup>-2</sup> (Diana et al., 1994; Diana et al., 1995a). Stocking rates of 3, 4.5, and 6 tilapia m<sup>-2</sup> were tested in Thailand; however, low survival complicated data analysis (Diana et al., 1995b). Past Honduras PD/A CRSP tilapia research has concentrated on semi-intensive production practices and response of water quality in fertilized and fed ponds stocked with 2 fish m<sup>-2</sup> (Teichert-Coddington et al., 1991; Green, 1992; Teichert-Coddington et al., 1992; Teichert-Coddington et al., 1993). The results of those studies were to serve as a point of reference for this study. High feeding rates required for intensive fish production lead to deterioration of pond water quality, which

can depress fish growth or result in fish kills (Boyd, 1990; Teichert-Coddington and Green, 1993). However, there is little information available in the literature on the effects of intensive tilapia production strategies on pond nutrient budgets and pond effluents.

Commercial production of tilapia is expanding rapidly in Central America, and hyper-intensive production systems often are being promoted to potential fish farmers. There are few or no sustainable technological packages for profitable tilapia production available to tilapia farmers in Central America. Only 12 of 41 production systems developed during the mid- to late-1980s by the PD/A CRSP were profitable and none yielded the larger-sized fish required by urban and export markets (Green et al., 1994). Commercial tilapia farms in Honduras routinely stock 5 to 7 fish m<sup>-2</sup>. The goal of the proposed research was to develop sustainable pond management practices for smallto medium-scale commercial tilapia farmers in Honduras. This study was proposed in the Eighth Work Plan. This report describes the circumstances that prevented the study from being completed.

## METHODS AND MATERIALS

This research was to be conducted in twelve 0.1-ha earthen ponds located at the Centro Nacional de Investigación Piscícola El Carao, Dirección General de Pesca y Acuacultura, Secretaría de Agricultura y Ganadería, Comayagua, Honduras. The experimental design called for 50- to 100-g male Nile tilapia (Oreochromis niloticus) to be stocked into ponds at rates of 2, 5, or 8 fish m<sup>-2</sup> for a 240-d grow-out period. A total of 60,000 fish were needed for stocking experimental ponds. Fish would be fed to satiation daily with a 25 to 30% protein extruded floating feed. Aeration (propeller-aspirator aerators, 20 HP ha<sup>-1</sup>) would be used to maintain pond dissolved oxygen concentration in excess of 20% of saturation. Fish were to be sampled by seine net at monthly intervals to measure growth. Early morning dissolved oxygen was to be measured in ponds daily. Total nitrogen, total ammonia nitrogen, nitrate-nitrite nitrogen, total phosphorus, soluble reactive phosphorus, Secchi disk visibility, chlorophyll a, and total alkalinity were to be determined biweekly and primary productivity was to be determined weekly according to standard protocol detailed in the PD/A CRSP Handbook of Analytical Methods (1992). Weather data were to be collected daily. Ponds were to be harvested by draining upon completion of the trial. Nutrient budgets were to be determined for each stocking rate; nutrients as inputs and outputs were to be measured. Full-cost enterprise budgets were to be developed for each stocking rate.

The null hypotheses to be tested in this completely randomized design experiment were:

- pond nutrient budgets are not affected by intensification of management system;
- 2) tilapia growth and yield are not affected by increased stocking rate; and,
- 3) production economics are not affected by intensification of management system.

Data analysis was to be by regression analysis and ANOVA with treatment differences determined by orthogonal contrasts.

## **RESULTS AND DISCUSSION**

In February 1998, nursery ponds were stocked with approximately 87,000 Nile tilapia fingerlings for growth to 50 to 100 g. Research ponds were not available until 11 June 1998 because the Eighth Work Plan Global Experiment (8FFR1H) had to be extended beyond its programmed duration because fish growth continued. In May 1998 the well at the El Carao station failed, leaving the wet lab and fish transport facilities without water. Water to the wet lab and for fish transport had been restored partially by mid-July. In order to avoid further delays in Eighth Work Plan implementation it was decided to proceed, albeit with some risk, with the transfer of tilapia from nursery ponds to grow-out ponds to begin experiment 8HR1. Transfer took place on 21 July 1998. Unfortunately, approximately 40,000 of the more than 60,000 fingerlings in the nursery ponds did not survive the transfer process because of inadequate supply of water to the wet lab. Initiation of the 8HR1 experiment was delayed until December 1998 while a new group of fingerlings were reared to 50 to 100 g. Fortunately, there were adequate fingerlings in inventory at the El Carao station to permit the revised schedule to be met. On

30–31 October 1998 the torrential rains of tropical storm Mitch caused the El Carao station (as well as many other places in Honduras) to flood, which resulted in mass escape of fish in ponds. Thus, it became impossible to complete experiment 8HR1.

#### **ANTICIPATED BENEFITS**

Results of this research would provide information on disposition of nutrients added as feed to intensively managed ponds; quality of pond effluents; growth, survival, and yield of tilapia at different stocking rates; and production economics of intensified pond management strategies.

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## Relative Contribution of Supplemental Feed and Inorganic Fertilizers in Semi-Intensive Tilapia Production

Eighth Work Plan, Kenya Research 3 (8KR3) Final Report

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#### Abstract

A 20-week experiment was conducted at Sagana Fish Farm, Kenya, to characterize the productive capacity of ponds at this new CRSP research site and to determine least-cost combinations of rice bran and inorganic fertilizer. Twelve 800-m<sup>2</sup> ponds were stocked with juvenile (32 g each) *Oreochromis niloticus* at 20,000 ha<sup>-1</sup> and *Clarias gariepinus* fingerlings (average weight 4.6 g) at 2,400 ha<sup>-1</sup>. Ponds contained about half sex-reversed and half mixed-sex tilapia, with an estimated ratio of approximately 75% males to 25% females at stocking. Four treatments were applied in triplicate as follows: 1) Urea and DAP to provide 16 kg N ha<sup>-1</sup> wk<sup>-1</sup> and 4 kg P ha<sup>-1</sup> wk<sup>-1</sup>; 2) Urea and DAP applied to give 8 kg N and 2 kg P ha<sup>-1</sup> wk<sup>-1</sup>, plus rice bran fed at 60 kg ha<sup>-1</sup> d<sup>-1</sup>; 3) Rice bran fed at 120 kg ha<sup>-1</sup> d<sup>-1</sup>; and 4) Rice bran as in Treatment 3 and fertilizer as in Treatment 2. Net fish yield averaged 1,127, 1,582, 1,607, and 2,098 kg ha<sup>-1</sup> for Treatments 1 through 4 respectively. Fish in ponds receiving rice bran (Treatments 2, 3, and 4) were still growing rapidly at harvest time, but the growth rate of fish in Treatment 1 was beginning to decrease near the end of the experiment. Treatment 1 was the most cost-effective, but Treatments 1, 2, and 4 all resulted in fairly similar net profits. Input costs for Treatment 1 may never reach market size at this stocking density. Fish had reduced growth towards the end of the culture period and resulting low final average weights, which were less than 100 g. If rice bran had cost 3.5 KSh or less per kilogram, profit for Treatment 3 would have surpassed that of Treatment 1. If rice bran had cost less than 5.8 KSh per kg, Treatment 2 would have been more profitable than Treatment 1.

#### INTRODUCTION

Aquaculture development in Kenya has been hampered by a lack of nutritionally complete feeds. The application of chemical fertilizers can enhance natural food production and indirectly provide protein to complement energy-rich rice bran (> 16 kcal digestible energy (DE) g<sup>-1</sup> protein; National Research Council, 1993). A 20-week experiment was conducted at Sagana Fish Farm, Kenya, to characterize the productive capacity of ponds at this new CRSP research site and to determine least-cost combinations of rice bran and inorganic fertilizer. In addition to the work reported here, samples of fish tissue, fish feed, soil, and plankton from the experiment were provided to researchers at the University of Arkansas at Pine Bluff for a companion study entitled "Nutritional Contribution of Natural and Supplemental Foods for Nile Tilapia: Stable Carbon Isotope Analysis." (See pp. 29-31 of this report)

#### METHODS AND MATERIALS

This experiment was conducted in twelve newly renovated CRSP research ponds at Sagana Fish Farm, Kenya, beginning on 31 October 1997 and ending on 25 March 1998 (144 days). Lime was applied to all ponds at a rate of 5 t ha<sup>-1</sup> prior to the experiment. The newest ponds received the lime treatment just prior to filling. Twelve 800-m<sup>2</sup> ponds were stocked with juvenile (32 g each) *Oreochromis niloticus* at 20,000 ha<sup>-1</sup> and *Clarias gariepinus* fingerlings (average weight 4.6 g) at 2,400 ha<sup>-1</sup>. Ponds contained half sex-reversed and half mixed-sex fish, with an estimated ratio of approximately 75% males to 25% females at stocking.

Four treatments were applied in triplicate as follows:

- Urea and diammonium phosphate (DAP) to provide 16 kg N ha<sup>-1</sup> wk<sup>-1</sup> and 4 kg P ha<sup>-1</sup> wk<sup>-1</sup>;
- Urea and DAP to provide 8 kg N and 2 kg P ha<sup>-1</sup> wk<sup>-1</sup>, plus rice bran fed at 60 kg ha<sup>-1</sup> d<sup>-1</sup>;

- 3) Rice bran fed at 120 kg ha<sup>-1</sup> d<sup>-1</sup>; and
- 4) Rice bran as in Treatment 3 and fertilizer as in Treatment 2.

Due to the relative newness of some ponds and a suspected high P adsorption capacity of newly exposed pond bottoms, the ponds were blocked according to the following criteria:

- Block 1: New ponds, never before filled and receiving lime just prior to filling.
- Block 2: Ponds that had been limed and were in production for less than a month; these were drained and refilled prior to this experiment.
- Block 3: Ponds that had been limed, filled, and in production (receiving feeds and fertilizers) for more than a month before the start of this experiment.

Ponds were assigned randomly in a split block design, with one replicate of each treatment in each block.

Dissolved oxygen concentration, temperature, and pH were measured weekly in the morning and afternoon. Total alkalinity, chlorophyll *a*, Secchi disk visibility, and total ammonia nitrogen concentration were measured every two weeks. Total nitrogen, mineral nitrogen, total phosphorus, and soluble reactive phosphorus were analyzed monthly. Samples for water chemistry were taken on Mondays, fertilizing was done on Tuesdays, and dissolved oxygen and temperature readings were done on Thursdays.

Ponds were sampled monthly for fish growth and drained completely after 20 weeks. Tilapia were separated by sex, counted, and weighed at draining. Tilapia reproduction was weighed and subsamples were counted. *Clarias* were counted and weighed. Two fish of each species were taken from each pond for proximate analysis at the beginning, in the middle (day 65), and at the end of the experiment. The whole fish, including viscera, was used for proximate analysis.

Rice bran was sampled as each lot arrived and combined into three batches. Each of the batches was analyzed for protein,

fiber, lipids, ash, and phosphorus. Proximate analyses of fish and feedstuff was done using the Kjeldahl method for protein, Soxhlet extraction for lipids, and muffle furnace for ash and phosphorus.

## **RESULTS AND DISCUSSION**

Although the Treatments 1, 2, and 3 were intended to be isonitrogenous, the rice bran contained less protein than expected. Total nitrogen and phosphorus inputs are summarized in Table 1. The first few lots of rice bran (all in Batch 1) contained excessively high quantities of fiber, and large amounts of husks were observed in these lots. Proximate analyses of the batches are given in Table 2.

Some of the ponds in Blocks 1 and 2 still had residual lime on their bottoms after draining; however, there were no significant differences in fish production between blocks. Block assignments were therefore not taken into account in the statistical analyses of other results. At harvest, the average weights of tilapia were 89, 106, 106, and 131 g, and *Clarias* weights were 110, 217, 236, and 296 g for Treatments 1, 2, 3, and 4, respectively (Table 3). Male tilapia and *Clarias* showed significantly different average weights among treatments but differences among female tilapia were significant only for Treatments 1 and 4. Survival ranged from 67 to 88%; there were no significant differences by treatment. Males made up 65 to 71% of total tilapia numbers at draining.

Fish began spawning during the first month of the experiment. However, due to the presence of *Clarias*, few fingerlings survived to harvest.

Net fish yield averaged 1,127, 1,582, 1,607, and 2,098 kg ha<sup>-1</sup> for Treatments 1, 2, 3, and 4, respectively (Table 3). Fish in Treatments 2, 3, and 4 were still growing rapidly at harvest time, but the growth rate of fish in Treatment 1 was beginning to decline near the end of the experiment (Figures 1 and 2).

Table 1. Weekly nitrogen and phosphorus inputs as chemical fertilizer or rice bran (feed) in 800-m<sup>2</sup> ponds for 20 weeks.

Treatment	I	<b>Nitrogen Input</b> (kg ha <sup>-1</sup> wk <sup>-1</sup> )		I	<b>Phosphorus Inpu</b> (kg ha <sup>-1</sup> wk <sup>-1</sup> )	t
	As Fertilizer	As Feed	Total N	As Fertilizer	As Feed	Total P
1	16	0	16.0	4	0	4.00
2	8	6.6	14.6	2	3.51	5.51
3	0	13.2	13.2	0	7.02	7.02
4	8	13.2	21.2	2	7.02	9.02

Table 2. Proximate analyses of rice bran batches (RB1–RB3) at the beginning, middle, and end of the study period (mean values presented unless noted otherwise).

Sample	Date	Water (%)	Protein (%)	Fiber (%)	Fat (%)	<b>Ash</b> (%)	Carbohydrate (NFE) (%)
RB1 RB2 RB3 RB Mean	31-Oct-97 03-Jan-98 23-Mar-98	10.35 10.23 19.38 13.32	8.60 10.15 10.73 9.83	19.65 16.93 14.30 16.96	9.68 3.38 6.12 6.39	17.77 7.00 6.47 10.41	33.95 52.32 43.00 43.09

Treatment	Original Tilapia Stock			Uncontro Repro	olled Tilapia oduction	Clarias (g)	<b>Net Fish Yield</b> (kg ha <sup>-1</sup> )
		Average Weight (g)		Number	Total Weight (kg)		
-	Males	Females	Mixed				
1 2 3 4	98 ª 121 <sup>b</sup> 125 <sup>b</sup> 155 <sup>c</sup>	61 <sup>a</sup> 70 <sup>ab</sup> 72 <sup>ab</sup> 77 <sup>b</sup>	89 ° 106 <sup>b</sup> 106 <sup>b</sup> 131 <sup>c</sup>	1,218 <sup>a</sup> 837 <sup>a</sup> 1,230 <sup>a</sup> 640 <sup>a</sup>	14.2 <sup>a</sup> 12.5 <sup>a</sup> 15.7 <sup>a</sup> 12.4 <sup>a</sup>	110 <sup>a</sup> 217 <sup>b</sup> 236 <sup>b</sup> 296 <sup>c</sup>	1,127 <sup>a</sup> 1,582 <sup>ab</sup> 1,607 <sup>ab</sup> 2,098 <sup>b</sup>

Table 3. Final average weights of original tilapia stock, number and total weight of uncontrolled reproduction, average weight of *Clarias*, and net total fish yield at harvest after 20 weeks by treatment.

<sup>a,b,c</sup> Values in a column followed by the same letter are not significantly different at the 95% level (Least Significant Difference; LSD).

Supply waters at Sagana typically have total alkalinity (TA) levels between 10 and 20 mg  $l^{-1}$  as CaCO<sub>3</sub>. TA levels in all ponds were higher than this throughout the experiment. The average TAs of ponds receiving rice bran rose to 70 mg  $l^{-1}$  or above by day 46 and remained relatively steady throughout the remainder of the experiment, whereas those of ponds receiving only chemical fertilizer descended to levels between about 35 and 55 mg  $l^{-1}$  after the first month (Figure 3).

Ponds in Treatment 1 had the highest average chlorophyll *a* concentrations (Tables 4 and 5). After December (month 2) the ponds in Treatment 3 (rice bran only) developed good algal blooms; however, prior to the second month they had little phytoplankton, and dissolved oxygen concentrations were frequently less than 1 mg l<sup>-1</sup> in the morning.

Ponds that received only inorganic fertilizer (Treatment 1) had significantly more suspended silt (inorganic matter) than other ponds (TSS - VSS in Tables 4 and 5), and suspended inorganic matter was linearly and negatively correlated with bran input rate (Figure 4). Nesting activities in monosex tilapia ponds can be a source of pond levee erosion. The high clay content of the pond soils at Sagana means they would be subject to erosion from activities such as nest building by male tilapia and from wind. Additions of organic matter such as chicken litter have been reported to reduce levels of suspended silt in ponds (Boyd, 1982; Teichert-Coddington et al., 1992).



Figure 1. Tilapia growth for Treatments 1 through 4 during the 144-day experiment to evaluate the relative contributions of supplemental feed and inorganic fertilizer in semi-intensive tilapia production.

Soluble reactive phosphorus concentrations were never higher than 0.05 mg l<sup>-1</sup> in any pond and were not greater than 0.03 mg l<sup>-1</sup> in ponds receiving rice bran (Figure 5). Total ammonia nitrogen never surpassed 0.5 mg l<sup>-1</sup> except on the final sampling date in Treatment 1 (Figure 6). Nitrate and nitrite were low throughout the experiment as well; average nitrite concentrations never surpassed 0.1 mg l<sup>-1</sup> NO<sub>2</sub>-N, and average nitrate concentrations always remained below 0.25 mg l<sup>-1</sup> NO<sub>3</sub>-N (Table 5). "Black cotton" soils, such as the ones in which the Sagana ponds are built have notoriously high phosphorus-adsorption rates. Assuming a clay content of 80% (ponds at Sagana are reported



Figure 2. *Clarias* growth for Treatments 1 through 4 during the 144-day experiment to evaluate the relative contributions of supplemental feed and inorganic fertilizer in semi-intensive tilapia production.



Figure 3. Average total alkalinity levels for ponds in Treatments 1 through 4 from day 17 through day 130 of the experiment.

Treatment averages for water quality parameter observations. Each parameter was measured at least five times during the experiment. Table 4. All values are in mg l<sup>-1</sup> except for chlorophyll *a* (uncorrected and corrected), which is in mg m<sup>-3</sup>, and total alkalinity, which is in mg l<sup>-1</sup> as CaCO<sub>3</sub>.

Treatment	Chl a <sup>1</sup> (uncorr)	Chl a <sup>2</sup> (corr)	TSS <sup>3</sup>	$VSS^4$	TSS - VSS⁵	Total N	TAN	NO <sub>2</sub> -N	NO <sub>3</sub> -N	Total P	SRP <sup>6</sup>	TA <sup>7</sup>
1	334	205	165 °	19	115 <sup>c</sup>	1 33	0.42 <sup>b</sup>	0.04	0 17 <sup>b</sup>	0.36	0 0 <b>22</b> °	18
2	184	115	105 132 <sup>ь</sup>	58	74 <sup>b</sup>	4.55 3.50	0.42 0.20 <sup>a</sup>	0.04	0.17 0.06 <sup>a</sup>	0.30	0.022 0.010 <sup>ab</sup>	40 75
3	93	61	79 <sup>a</sup>	44	35 ª	3.08	0.22 <sup>a</sup>	0.01	0.09 <sup>a</sup>	0.30	0.007 <sup>a</sup>	70
4	187	128	96 ª	59	37 <sup>a</sup>	4.39	0.20 <sup>a</sup>	0.02	0.07 <sup>a</sup>	0.41	$0.018 \ ^{bc}$	84

 $^{a,b,c}$  Values in a column followed by different letters are significantly different at P < 0.05.

Chlorophyll a (uncorrected)

Chlorophyll a (corrected)

Total suspended solids Volatile suspended solids 5 Total suspended solids minus volatile suspended solids-a measure of suspended inorganic matter

Soluble reactive phosphate

Total alkalinity

Averages of water quality parameters, by pond, for the experiment. Each parameter was measured at least five times during the Table 5. experiment. All values are in mg  $l^{-1}$  except for chlorophyll *a* (uncorrected and corrected), which is in mg m<sup>-3</sup>, and total alkalinity, which is in mg l<sup>-1</sup> as CaCO<sub>3</sub>.

Pond	Treatment	Chl <i>a</i> <sup>1</sup> (uncorr)	Chl a <sup>2</sup> (corr)	TSS <sup>3</sup>	VSS <sup>4</sup>	TSS - VSS⁵	Total N	TAN	NO <sub>2</sub> -N	NO <sub>3</sub> -N	Total P	SRP <sup>6</sup>	TA <sup>7</sup>
D05	1	206	131	162	50	111	4.32	0.51	0.06	0.23	0.37	0.02	43
D06	1	608	348	185	57	128	5.17	0.33	0.03	0.15	0.42	0.02	40
E05	1	187	136	147	41	106	3.50	0.42	0.02	0.14	0.28	0.02	60
D07	2	140	91	123	54	70	3.90	0.24	0.03	0.07	0.31	0.01	74
E07	2	186	129	127	52	75	2.78	0.16	0.01	0.06	0.33	0.01	91
E09	2	226	123	145	67	78	3.81	0.20	0.01	0.05	0.32	0.01	61
D08	3	90	55	77	42	35	2.74	0.21	0.01	0.08	0.26	0.00	55
E03	3	109	86	65	42	23	2.73	0.23	0.01	0.11	0.34	0.01	73
E04	3	79	41	95	48	47	3.75	0.22	0.01	0.07	0.29	0.01	83
D04	4	159	111	83	55	28	4.74	0.25	0.03	0.08	0.32	0.01	67
E06	4	168	114	97	55	42	3.78	0.20	0.01	0.07	0.43	0.02	99
E08	4	234	158	108	68	41	4.52	0.16	0.01	0.05	0.47	0.03	87

Chlorophyll a (uncorrected)

Chlorophyll a (corrected)

Total suspended solids Volatile suspended solids

Total suspended solids minus volatile suspended solids-a measure of suspended

inorganic matter

Soluble reactive phosphate

Total alkalinity



Figure 4. The relationship between bran input level (kg bran ha<sup>-1</sup> d<sup>-1</sup>) and suspended inorganic matter (mg l-1) in the ponds.



Figure 5. Average soluble reactive phosphorus concentrations for Treatments 1 through 4 during the experiment.

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Figure 6. Average total ammonia nitrogen (TAN) concentrations for Treatments 1 through 4 during the experiment.

to have as much as 90% clay soils), a phosphorus adsorption capacity of 350 kg ha<sup>-1</sup> was estimated using the formula proposed by Shrestha and Lin (1996). Only 80 kg ha<sup>-1</sup> was added to most ponds over the 20 weeks, so the phosphorus adsorption capacity was far from satisfied, and little difference was observed among blocks of ponds.

Nitrogen efficiency (kg net fish yield per kg N applied) averaged 3.52, 5.42, 6.09, and 4.95 for Treatments 1, 2, 3, and 4, respectively. The N:P ratio of the inputs was lowest for Treatment 3.

At Rwasave Fish Station, Rwanda, an experiment was conducted in which fish were fed rice bran at 5 g fish<sup>-1</sup> d<sup>-1</sup> in ponds stocked at a density of 2 male tilapia m<sup>-2</sup>. A mean net yield of 2,620 kg ha<sup>-1</sup> and a feed conversion ratio (FCR) of 7.6

were obtained after 192 days. No chemical fertilizers were applied, but small additions of chicken manure and grass were used as fertilizer (Verheust et al., 1992). The FCRs obtained in Treatments 3 and 4 of this experiment, in which rice bran was applied at 6 g tilapia<sup>-1</sup> d<sup>-1</sup>, are much higher (10.5 and 8 for Treatments 3 and 4, respectively) because the fish started out at a smaller size (32 g in this experiment versus 80 g in the Rwanda experiment) and they could not consume all the bran. Also, the ponds were harvested before market size was reached, thereby not allowing recovery from the overfeeding at the beginning of the experiment. A study that combined inorganic fertilization and feeding rice bran to tilapia (reaching a maximum application of 46 kg ha<sup>-1</sup> d<sup>-1</sup>) obtained a mean net yield of 1,160 kg ha<sup>-1</sup> after 159 days (Perschbacher and Lochmann, 1995). This is somewhat less than the net yield in Treatment 2 of this experiment (1,582 kg ha<sup>-1</sup>).

Fish of both species contained significantly higher lipid levels when fed rice bran (Treatments 2, 3, and 4) than when in the fertilizer-only Treatment 1 (Tables 6a and 6b).

Using the results of Treatment 1 (fertilizer only) as the baseline, partial enterprise budget analysis demonstrates that none of the other treatments resulted in greater returns to operating costs. However, in comparing Treatment 4 with Treatment 3, applying fertilizer in addition to the high bran application was definitely beneficial in the economic sense (Table 7).

#### CONCLUSIONS

Treatment 1 (fertilizer only) was the most cost-effective, but the net profits from Treatments 1, 2, and 4 were similar (Table 8). Input costs for Treatments 1 and 2 will definitely be of interest

Treatment	Date	Water (%)	Protein (%)	Fiber (%)	Fat (%)	<b>Ash</b> (%)
All	31-Oct-97	74.7	14.7	1.29	3.74	3.75
1	1-Mar-98	$78.6\pm2.77$ $^{\rm a}$	$15.1\pm3.45$ $^{\rm a}$	$0.7\pm0.72$ $^{\rm a}$	$0.9\pm0.72$ $^{\rm a}$	$3.8\pm1.03$ $^{\rm a}$
2	1-Mar-98	$73.8\pm3.41$ $^{\rm a}$	$14.6\pm1.03$ $^{\rm a}$	$0.5\pm0.16$ $^{\rm a}$	$7.2\pm0.73$ $^{\rm b}$	$3.2\pm0.19$ $^{\rm a}$
3	1-Mar-98	$71.2\pm2.66$ $^{\rm a}$	$15.8\pm1.26$ $^{\rm a}$	$0.3\pm0.22$ $^{\rm a}$	$8.4\pm1.04~^{\rm b}$	$3.3\pm0.67$ $^{\rm a}$
4	1-Mar-98	$73.7\pm1.55$ $^{\rm a}$	$16.7\pm0.75$ $^{\rm a}$	$1.0\pm0.56$ $^{\rm a}$	$9.2\pm2.01~^{\rm b}$	$3.0\pm0.72^{\rm a}$

Table 6a. Proximate analyses for tilapia at stocking and towards the end of the study period. Figures given are means ± SD for each treatment.

<sup>a,b</sup> Values in a column followed by the same letter are not significantly different at the 95% level (LSD).

Table 6b. Proximate analyses for Clarias at stocking and towards the end of the study period. Figures given are means ± SD for each treatment.

Treatment	Date	Water (%)	Protein (%)	Fiber (%)	Fat (%)	<b>Ash</b> (%)
All	31-Oct-97	68.13	12.42	1.07	0.62	2.94
1	1-Mar-98	$79.1\pm0.20$ $^{\rm b}$	$16.4\pm0.78$ $^{\rm a}$	$0.2\pm0.14$ $^{\rm a}$	$0.8\pm0.62$ $^{\rm a}$	$3.1\pm0.42$ $^{\rm a}$
2	1-Mar-98	$75.0\pm2.18$ $^{\rm a}$	$17.9\pm0.85$ $^{\rm a}$	$0.5\pm0.24$ $^{\rm a}$	$4.9\pm0.40$ $^{\rm b}$	$2.5\pm0.14$ $^{\rm a}$
3	1-Mar-98	$75.0\pm2.38$ $^{\rm a}$	$16.7\pm0.93$ $^{\rm a}$	$0.6\pm0.94$ $^{a}$	$5.0\pm0.48$ $^{\rm b}$	$2.9\pm0.22$ $^{a}$
4	1-Mar-98	$74.0\pm1.90$ $^{\rm a}$	$17.8\pm1.78$ $^{\rm a}$	$1.1\pm0.36$ $^{\rm a}$	$5.0\pm1.28$ $^{\rm b}$	$3.0\pm0.47$ $^{\rm a}$

<sup>a,b</sup> Values in a column followed by the same letter are not significantly different at the 95% level (LSD).

Table 7. Partial budget analysis in Kenya shillings (KSh) relative to Treatment 1 (fertilizer only). Cost of capital is calculated based on 12% interest (per annum interest is about 24%) of total increased operating costs. Figures are on a per-pond (800 m<sup>2</sup>) basis. At the time the experiment ended, the exchange rate was approximately 60 KSh to US\$1.

	Treatment			
	2	3	4	
A. CHANGE IN COSTS				
Feed Labor Cost of Capital Fertilizer	+3,828 +190 +482 -968	+7,656 +162 +938 -1,937	+7,656 +190 +942 -968	
Net Change in Costs	+3,532	+6,819	+7,820	
B. Additional Revenue from Fish	2,930	2,673	6,604	
C. NET CHANGE (A - B)	-602	-4,146	-1,216	

to fish farmers. Treatment 1 resulted in the highest profit (Table 8); however, it is possible that fish raised using only fertilizer at the rates in Treatment 1 may never reach market size at this stocking density, as evidenced by their reduced growth towards the end of the experiment (Figures 1 and 2).

In order to further increase production over that obtained using Treatment 2, applying additional fertilizer may be a better solution than increasing bran inputs. Diana et al. (1996) found that adding supplemental feed (floating pellets, 30% crude protein) after fish reached 150 g resulted in greater annual profit than either fertilization only or feeding right from the start. In this experiment, the bran can be considered to function partly as a feed and partly as an organic fertilizer.

Bran prices vary in Kenya. Rice bran can be purchased for as little as 3 KSh kg<sup>-1</sup> (by special arrangement with governmentowned rice mills), but 6 KSh kg<sup>-1</sup> is more common for farmers buying retail. At the price of 6 KSh kg<sup>-1</sup>, rice bran should be applied sparingly and not as a fertilizer. It is rumored that some unscrupulous retailers mix husks (obtained free) with bran. Proximate analyses indicate that some of this bran may have obtained early in the experiment. Wheat bran is available in greater quantities than rice bran and retails for 5 to 7 KSh kg<sup>-1</sup>. One batch was tested and resulted in 14.64% protein, 2.9% lipids, and 12.52% fiber. Wheat bran may present a better alternative for farmers and should be tested in future PD/A CRSP experiments. It is now being used at Sagana Fish Farm. Maize bran (8% protein) and maize germ (16% protein) are also available in even greater quantities than wheat bran, but prices are higher per unit protein. If rice bran had cost 3.5 KSh kg<sup>-1</sup> or less, then profit for Treatment 3 would have surpassed the profit of Treatment 1. If rice bran had cost less than 5.8 KSh kg<sup>-1</sup>, then Treatment 2 would have been more profitable than Treatment 1. Since this experiment was conducted, rice bran prices in the Sagana area have dropped to just over 2 KSh kg<sup>-1</sup>, making it much more attractive as a component in pond management regimes.

## **ANTICIPATED BENEFITS**

This experiment has provided data on the comparative value of inorganic fertilizers and low-cost supplemental feeds as pond inputs for semi-intensive tilapia production at Sagana, thus providing the basis for the development of more efficient production strategies for pond systems in Kenya and similar areas of Africa. In addition, determining pond productivity using high nutrient input levels at Sagana provides data for comparison of the Africa site with other CRSP sites in Southeast Asia, Central America, and South America.

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Table 8. Summary of costs and harvest revenues per pond, not counting fingerling costs of 7,200 KSh pond<sup>-1</sup> for all treatments. The price of adult fish is assumed to be 90 KSh kg<sup>-1</sup>. No value is attributed to fingerlings harvested.

Treatment	Fertilizer (KSh)	<b>Feed</b> (KSh)	Input Labor (KSh)	Adults Harvested (kg)	Revenue (KSh)	Net Profit (KSh)
1	1,937	0	1,220	133	11,970	8,813
2	969	3,828	1,410	166	14,900	8,693
3	0	7,656	1,382	163	14,643	5,605
4	969	7,656	1,410	206	18,574	8,539

Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics / Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]



## PD/A CRSP Seventeenth Annual Technical Report

## NUTRITIONAL CONTRIBUTION OF NATURAL AND SUPPLEMENTAL FOODS FOR NILE TILAPIA: STABLE CARBON ISOTOPE ANALYSIS

Eighth Work Plan, Kenya Research 3A (8KR3A) Final Report

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#### Abstract

Stable carbon isotope analysis can be used to obtain quantitative estimates of the relative contributions of different food sources to the nutrition of aquatic animals in ponds. Results can be used to make recommendations for feeding and fertilization practices that will minimize feed costs while maximizing fish production. This technique was used in conjunction with fish gut content analysis to obtain estimates of the contribution of natural and supplemental feeds to the nutrition of Oreochromis niloticus and Clarias gariepinus in ponds receiving different inputs in Sagana, Kenya. Four combinations of fertilizers and supplemental feed (rice bran) were used as experimental treatments: 1) Urea (16 kg N ha<sup>-1</sup> wk<sup>-1</sup>) + DAP (4 kg P ha<sup>-1</sup> wk<sup>-1</sup>); 2) Urea (8 kg N ha<sup>-1</sup> wk<sup>-1</sup>) + DAP (2 kg P ha<sup>-1</sup> wk<sup>-1</sup>) + Rice bran (60 kg ha<sup>-1</sup> d<sup>-1</sup>); 3) Rice bran (120 kg ha<sup>-1</sup> d<sup>-1</sup>); and 4) Rice bran (120 kg ha<sup>-1</sup> d<sup>-1</sup>) + Urea (8 kg N ha<sup>-1</sup> wk<sup>-1</sup>) + DAP (2 kg P ha<sup>-1</sup> wk<sup>-1</sup>). Samples of Oreochromis, Clarias, chemical fertilizers (DAP and urea), rice bran, plankton, and mud taken from ponds in Sagana at three times (initial, midpoint, final) during a 143-day feeding trial were analyzed for carbon isotope content. The most distinct trend in the isotope data was the more positive values for plankton, Oreochromis, and Clarias in treatment 1 versus treatments 2 through 4 for initial, midpoint, and final samples. The addition of rice bran to ponds in treatments 2 through 4 clearly increased fish production relative to ponds where the only inputs were DAP and urea. Gut content analysis indicated that the two most important food categories for Oreochromis and Clarias in treatments that included rice bran were plankton and rice bran. However, the isotope data did not allow further clarification of the relative nutritional importance of the two categories between treatments because the isotope ratios of plankton and rice bran were not isotopically distinct.

#### INTRODUCTION

Stable carbon isotope analysis is a useful technique to obtain quantitative estimates of the relative contributions of different food sources to the nutrition of aquatic animals in ponds (e.g., Schroeder, 1983; Anderson et al., 1987; Lochmann and Phillips, 1996). Presumably, isotope ratios of the fish will resemble those of the food(s) they assimilate most. In the present study, stable carbon isotope ratios of feeds, fertilizers, plankton, and mud were compared to those of *Oreochromis* and *Clarias* co-cultured in ponds receiving different inputs in Sagana, Kenya, during a 143-day feeding trial. We anticipate using the results to optimize nutrient utilization and minimize feed/fertilizer costs for *Oreochromis* production in Kenya and possibly other locations.

### METHODS AND MATERIALS

Several months prior to collection of initial samples, *O. niloticus* and *C. gariepinus* were fed a conditioning diet containing corn to increase the isotopic resemblance of the fish to corn (-14‰). Four combinations of fertilizers and supplemental feed (rice bran) were used as experimental treatments:

- 1) Urea (16 kg N  $ha^{-1}$  wk<sup>-1</sup>) + DAP (4 kg P  $ha^{-1}$  wk<sup>-1</sup>);
- Urea (8 kg N ha<sup>-1</sup> wk<sup>-1</sup>) + DAP (2 kg P ha<sup>-1</sup> wk<sup>-1</sup>) + Rice bran (60 kg ha<sup>-1</sup> d<sup>-1</sup>);
- 3) Rice bran (120 kg ha<sup>-1</sup> d<sup>-1</sup>); and
- 4) Rice Bran (120 kg ha<sup>-1</sup> d<sup>-1</sup>) + Urea (8 kg N ha<sup>-1</sup> wk<sup>-1</sup>) + DAP (2 kg P ha<sup>-1</sup> wk<sup>-1</sup>).

Each treatment was applied to three ponds. The major components of the pond system assumed to contribute to the nutri-

tional status of Oreochromis and Clarias were sampled monthly throughout the study. The components were: supplemental feed (rice bran), chemical fertilizers (DAP and urea), plankton, and mud. Samples from three periods (initial, midpoint, and final) were processed and subjected to isotope analysis. Initially, five individual Oreochromis and Clarias (a total of ten fish) were collected from a single pond at Sagana Fish Farm, Sagana, Kenya. Initial fish samples were not pooled to allow determination of variability in isotope ratios among individuals. Initial pooled samples of plankton and mud were collected from each of the 12 study ponds, as well as samples of DAP, urea, and rice bran. All samples were processed as described previously (Lochmann and Phillips, 1996), except that carbonates were removed from mud samples prior to lyophilization, and DAP and urea samples were not processed prior to analysis. Samples collected from the midpoint and final periods were the same as described for initial samples except that a pooled sample of Oreochromis and a pooled sample of Clarias (each pooled sample consisting of two individuals) was collected from each pond. All samples were sent to a commercial laboratory (Coastal Science Laboratories, Inc., Austin, Texas ) for stable carbon isotope analysis using a micromass isotope ratio mass spectrometer (Anderson et al., 1987).

#### RESULTS

The mean isotope ratio ( $\delta^{13}$ C) of initial plankton samples in treatment 1 was significantly more positive than that of plankton in treatments 2-4 (Table 1). The mean  $\delta^{13}$ C of initial

mud samples did not differ among Treatments (Table 1). The mean initial  $\delta^{13}$ C of *Clarias* was approximately 3‰ more negative than that of *Oreochromis*, and the *Clarias* data was more variable (Table 1). There were no differences in isotope ratios of initial mud samples among treatments. The  $\delta^{13}$ C of initial urea was -53.6‰ and those of DAP and rice bran were very similar: -26.8 and -27.8‰, respectively.

The mean  $\delta^{13}$ C of midpoint plankton samples in treatment 1 was significantly more positive than that of plankton in treatments 2 through 4 (Table 2), as in initial samples. The mean isotope ratios of *Oreochromis* and *Clarias* followed the same pattern as the plankton, although statistical differences were less pronounced

for *Clarias* (Table 2). There were no differences in isotope ratios of midpoint mud samples among treatments.

The mean  $\delta^{13}$ C of final plankton samples was significantly more positive than that of plankton in treatments 2 through 4 (Table 3), as in initial and midpoint samples. The mean isotope ratios of *Oreochromis* and *Clarias* from treatment 1 were significantly less negative than those of treatments 2 through 4. In addition, the mean isotope ratios of *Oreochromis* and *Clarias* from treatment 2 were significantly less negative than those from treatment 3, while isotope ratios of both species from treatment 4 were between those of fish from treatments 2 and 3 (Table 3). There were no differences in isotope ratios of final mud samples among treatments.

Table 1. Initial stable carbon isotope values ( $\delta^{13}$ C) of pond components in a feeding trial with *Oreochromis* and *Clarias* in Sagana, Kenya. Values for plankton and mud are means of three replicates. DAP (diammonium phosphate) and urea were used as chemical fertilizers. Rice bran served as an organic fertilizer and a supplemental feed. See text for full explanation of treatments.

Treatment (TX) Number and Description	$\delta^{13}C \pm S.D.$ (%)						
	Oreochromis	Clarias	Plankton	Mud			
1 DAP (4 kg P ha <sup>-1</sup> wk <sup>-1</sup> ) + Urea (16 kg N ha <sup>-1</sup> wk <sup>-1</sup> ) 2 $\frac{1}{2}$ (DAP + Urea) of TX 1 + $\frac{1}{2}$ Rice Bran of TX 3 3 Rice Bran (120 kg ha <sup>-1</sup> d <sup>-1</sup> ) 4 Rice Bran of TX 3 + $\frac{1}{2}$ (DAP + Urea) of TX 1	$\begin{array}{c} -16.6 \pm 0.7^c \\ -16.6 \pm 0.7^c \\ -16.6 \pm 0.7^c \\ -16.6 \pm 0.7^c \end{array}$	$\begin{array}{c} -19.8 \pm 1.2^{c} \\ -19.8 \pm 1.2^{c} \\ -19.8 \pm 1.2^{c} \\ -19.8 \pm 1.2^{c} \end{array}$	$\begin{array}{c} -17.3 \pm 0.5^{a} \\ -23.5 \pm 1.6^{b} \\ -26.3 \pm 2.5^{b} \\ -26.8 \pm 2.5^{b} \end{array}$	$-13.5 \pm 1.4$ $-13.1 \pm 0.6$ $-13.3 \pm 0.7$ $-12.3 \pm 0.5$			

 $^{,b}$  Means in columns with different letters are significantly different (P < 0.05) according to Fisher's Least Significant Difference test.

Initial isotope values for Oreochromis and Clarias are means of 5 individual fish taken from a group of fish fed a single conditioning diet in a common pond prior to stocking in individual ponds.

Table 2. Midpoint stable carbon isotope values (δ<sup>13</sup>C) of pond components in a feeding trial with *Oreochromis* and *Clarias* in Sagana, Kenya. Values are means of three replicates. DAP (diammonium phosphate) and urea were used as chemical fertilizers. Rice bran served as an organic fertilizer and a supplemental feed. See text for full explanation of treatments.

Treatment (TX) Number and Description	$\delta^{13}C \pm S.D.$ (%)					
	Oreochromis	Clarias	Plankton	Mud		
1 DAP (4 kg P ha <sup>-1</sup> wk <sup>-1</sup> ) + Urea (16 kg N ha <sup>-1</sup> wk <sup>-1</sup> ) 2 <sup>1</sup> / <sub>2</sub> (DAP + Urea) of TX 1 + <sup>1</sup> / <sub>2</sub> Rice Bran of TX 3 3 Rice Bran (120 kg ha <sup>-1</sup> d <sup>-1</sup> ) 4 Rice Bran of TX 3 + <sup>1</sup> / <sub>2</sub> (DAP + Urea) of TX 1	$\begin{array}{l} -17.9 \ \pm \ 0.9^a \\ -22.8 \ \pm \ 0.7^b \\ -22.9 \ \pm \ 2.4^b \\ -24.0 \ \pm \ 1.2^b \end{array}$	$\begin{array}{c} -22.4 \pm 2.1^{a} \\ -24.7 \pm 1.3^{ab} \\ -26.4 \pm 1.2^{b} \\ -26.8 \pm 0.6^{b} \end{array}$	$\begin{array}{l} -22.4 \ \pm 1.8^{a} \\ -27.0 \ \pm 1.2^{b} \\ -29.6 \ \pm 1.2^{b} \\ -27.8 \ \pm 1.8^{b} \end{array}$	$\begin{array}{c} -13.7\pm1.1\\ -14.6\pm1.1\\ -14.7\pm0.9\\ -15.4\pm0.9\end{array}$		

<sup>a,b</sup> Means in columns with different letters are significantly different (P < 0.05) according to Fisher's Least Significant Difference test.

Table 3. Final stable carbon isotope values ( $\delta^{13}$ C) of pond components in a feeding trial with *Oreochromis* and *Clarias* in Sagana, Kenya. Values are means of three replicates. DAP (diammonium phosphate) and urea were used as chemical fertilizers. Rice bran served as an organic fertilizer and a supplemental feed. See text for full explanation of treatments.

Treatment (TX) Number and Description		δ <sup>13</sup>		
	Oreochromis	Clarias	Plankton	Mud
1 DAP (4 kg P ha <sup>-1</sup> wk <sup>-1</sup> ) + Urea (16 kg N ha <sup>-1</sup> wk <sup>-1</sup> ) 2 $\frac{1}{2}$ (DAP + Urea) of TX 1 + $\frac{1}{2}$ Rice Bran of TX 3 3 Rice Bran (120 kg ha <sup>-1</sup> d <sup>-1</sup> ) 4 Rice Bran of TX 3 + $\frac{1}{2}$ (DAP + Urea) of TX 1	$\begin{array}{c} -17.9\pm0.7^{a}\\ -23.7\pm1.2^{b}\\ -25.8\pm0.1^{c}\\ -24.9\pm0.6^{bc} \end{array}$	$\begin{array}{l} -18.5\pm1.l^{a}\\ -24.7\pm0.2^{b}\\ -27.2\pm0.9^{c}\\ -25.9\pm1.1^{bc} \end{array}$	$\begin{array}{c} -20.6 \pm 3.4^{a} \\ -25.7 \pm 1.0^{b} \\ -29.3 \pm 0.5^{b} \\ -26.0 \pm 1.5^{b} \end{array}$	$\begin{array}{c} -14.0\pm0.5\\ -16.2\pm1.8\\ -16.1\pm1.1\\ -15.6\pm0.6\end{array}$

 $^{a,b,c}$  Means in columns with different letters are significantly different (P < 0.05) according to Fisher's Least Significant Difference test.

The mean isotope ratios of *Oreochromis* became substantially more negative between the initial and midpoint sampling periods for treatments 2, 3, and 4, and slightly more negative between the midpoint and final sampling periods (Tables 1 through 3). However, the mean  $\delta^{13}$ C of *Oreochromis* in treatment 1 changed very little during the study (Tables 1 through 3). The mean isotope ratios of *Clarias* became more negative between the initial and midpoint sampling periods for all treatments (Tables 1 and 2). The mean isotope ratios of *Clarias* changed only slightly between the midpoint and final sampling periods in treatments 2, 3, and 4 (Tables 2 and 3), while that of Clarias in treatment 1 became about 4‰ more positive (Tables 2 and 3). The mean isotope ratios of plankton became more negative in all treatments between the initial and midpoint sampling periods (Tables 1 and 2), then more positive between the midpoint and final periods (Tables 2 and 3). However, the magnitude of the change in plankton isotope ratios within treatments was small (approximately 3‰) for the whole study period. The mean isotope ratios of mud became more negative for all treatments throughout the study (Tables 1 through 3), although the changes were more pronounced for treatments 2, 3, and 4 than for treatment 1. As with the plankton, the magnitude of the changes in mean isotope ratios of mud within treatments was small (3% or less) for the whole study period.

#### DISCUSSION

The isotope technique is more effective in pinpointing nutritional inputs of an animal when the inputs are isotopically distinct from each other and from the animal itself (Anderson et al., 1987). Several months prior to collection of initial samples, *Oreochromis* and *Clarias* were fed a conditioning diet containing corn to increase the isotopic resemblance of the fish to corn (-14‰). Corn is isotopically distinct from the rice bran (-27.8‰) used as a supplemental feed in treatments 2, 3, and 4. The isotope ratios of the *Oreochromis* and *Clarias* before receiving the corn diet were unknown so the influence of the corn diet on the isotope ratios of fish at the initiation of the study is not certain. Although the isotope ratios of initial *Oreochromis* (-16.6‰) and *Clarias* (-19.8‰) were 3 to 6‰ different from that of corn, they were 8‰ or more different from that of rice bran.

The most distinct trend in the isotope data was the more positive values for plankton, Oreochromis, and Clarias in treatment 1 versus treatments 2 through 4 for initial, midpoint, and final samples. Treatment 1 did not include rice bran, whereas treatments 2 through 4 did. Gut content data of Veverica et al. (2000) indicated that both Oreochromis and Clarias in treatments 2 through 4 consumed rice bran directly, which contributed to the more negative isotope values of these fish compared to those in treatment 1. However, the isotope values of the plankton in treatment 1 were more positive initially than those of the plankton in the other treatments, which suggests that the result may be due to an undefined pretreatment effect. Veverica also observed significantly higher chlorophyll *a* concentrations, as well as lower weight gain of Oreochromis and Clarias in treatment 1 compared to the other treatments (Veverica et al., 2000).

The addition of rice bran to ponds in treatments 2 through 4 clearly increased fish weight gain relative to ponds where the

only inputs were DAP and urea (Veverica et al., 2000). Gut content analysis (Veverica et al., 2000) indicated that the two most important food categories for *Oreochromis* and *Clarias* in treatments 2 through 4 were plankton and rice bran. The isotope data did not allow further clarification of the relative nutritional importance of the two categories between treatments because the isotope ratios of plankton and rice bran differed by only 4‰ or less. The DAP (-26.8‰) was also isotopically similar to rice bran (-27.8‰), but DAP did not appear to have much influence on the isotope value of the plankton in treatment 1 (no rice bran).

The discriminating power of the isotope technique may be improved in future studies by fractionating the plankton into different categories before isotope analysis, as fish gut content analysis indicated that there were major differences in the relative use of zooplankton and phytoplankton for *Oreochromis* and *Clarias* in the different treatments. The effect of the different fertilization regimes on primary and secondary plankton production rates would be useful in optimizing nutrient utilization of co-cultured fish species with different trophic habits. In addition, the use of nitrogen isotope data might clarify the trophic relationships between *Oreochromis* and *Clarias* in different treatments. Gut content data were not useful in this respect, as *Oreochromis* guts contained an equal or higher number of fish scales than *Clarias* guts in treatments 2 through 4.

## **ANTICIPATED BENEFITS**

Production efficiency of *Oreochromis* and *Clarias* can be optimized once the quantitative importance of different nutrients under defined experimental conditions is established using the isotope technique (perhaps multiple isotope tracers) in conjunction with comprehensive production data. Furthermore, the procedures used to define the importance of various components in this aquaculture production system may be modified and applied to other systems in other regions.

#### **ACKNOWLEDGMENTS**

The authors wish to thank the staff at Sagana Fish Farm for collection and processing of samples prior to isotope analysis.

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Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics / Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]


# PD/A CRSP Seventeenth Annual Technical Report

## GLOBAL EXPERIMENT: OPTIMIZATION OF NITROGEN FERTILIZATION RATE IN FRESHWATER TILAPIA PRODUCTION PONDS

Eighth Work Plan, Feeds and Fertilizers Research 1 (8FFR1K) Progress Report

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#### Abstract

Two experiments to determine the optimum nitrogen fertilization rates for freshwater tilapia production ponds at Sagana Fish Farm, Sagana, Kenya, were conducted during 1998 and 1999. Twelve 800-m<sup>2</sup> earthen research ponds managed by the PD/A CRSP at Sagana were used for the experiments. Diammonium phosphate and urea were used to apply nitrogen to the ponds at rates of 0, 10, 20, and 30 kg N ha<sup>-1</sup> wk<sup>-1</sup>. Triple superphosphate or diammonium phosphate (DAP) and sodium carbonate were applied to ponds to assure that phosphorus and carbon were not limiting. A completely randomized design was used, with three replicates for each of the four treatments. The experiment was conducted once during the 1998 cool season (May to October) and again during the warm season of 1998-1999 (November to March). In the cool-season experiment, ponds were stocked with sex-reversed Nile tilapia, Oreochromis niloticus, averaging 16.9 g at a rate of 1,000 kg ha<sup>-1</sup> and with Clarias gariepinus fingerlings averaging 37 g at a rate of 37 kg ha<sup>-1</sup>. In the warm-season experiment, all ponds were stocked with sexreversed O. niloticus averaging 90 g at 1,000 kg ha<sup>-1</sup> and with C. gariepinus juveniles averaging 166 g at 125 kg ha<sup>-1</sup>. Pond assignments were re-randomized prior to the second experiment. Ponds were drained when fish growth appeared to have stopped in all treatments. In both experiments, a highly significant (P < 0.01) quadratic relationship best described gross (as well as net) fish yield as related to weekly N input. Presence of Clarias had little impact on the relationship but it appeared the high nitrogen input rates had no negative effect on Clarias production. Increasing nitrogen input from 20 kg N ha<sup>-1</sup> wk<sup>-1</sup> did not result in increased tilapia yield. Total nitrogen and all mineral forms of nitrogen increased with increasing nitrogen input, as did chlorophyll a. Partial budget analysis indicated that greatest marginal returns were at the calculated rates of 19.9 and 16.0 kg N ha<sup>-1</sup> wk<sup>-1</sup> for the cool- and warm-season experiments, respectively. A carryover effect of the first experiment is suggested. Results from this experiment are similar to those obtained at the CRSP site at El Carao, Honduras.

#### INTRODUCTION

Nitrogen, phosphorus, and carbon availability are important considerations in the management of ponds for optimum fish production. Previous PD/A CRSP research has addressed increasing primary productivity through inorganic and organic nutrient additions to ponds, but findings on the optimum nitrogen, phosphorus, and carbon inputs required to improve fish yields at the PD/A CRSP sites appeared inconsistent and called for clarification. Higher nutrient inputs had resulted in increased fish production at all PD/A CRSP sites, but optimum inputs of nitrogen, phosphorus, and carbon had not been defined (see reports in Egna et al., 1990, 1991; Egna et al., 1992; Egna et al., 1993; Egna et al., 1994, 1995).

Fertilization rates in PD/A CRSP experiments were greater than rates reported for earlier pond fertilization research. In an often-cited series of fertilization experiments conducted in Malaysia, Hickling (1962) never used more than 1.1 kg P and 1.1 kg N ha<sup>-1</sup> wk<sup>-1</sup>. In Israel, the standard fertilizer dose was 2.3 kg P and 6.5 kg N ha<sup>-1</sup> wk<sup>-1</sup> (Hepher, 1962a, b). The highest rates of phosphorus and nitrogen used in most experiments at Auburn University were 1.26 and 3 kg ha<sup>-1</sup> wk<sup>-1</sup>, respectively (Swingle, 1947; Boyd, 1976, 1990; Boyd and Sowles, 1978; Murad and Boyd, 1987). Rates in Europe seldom exceeded 1 kg ha<sup>-1</sup> wk<sup>-1</sup> for nitrogen and phosphorus (Mortimer, 1954). Rates used in Malaysia, USA, Israel, and Europe were adequate to give dense phytoplankton blooms and good fish production. Also, in all of the studies cited above, phosphorus was the most important limiting nutrient.

This set of experiments was designed to determine the optimal application rates of nitrogen to attain the most profitable tilapia yields in tropical freshwater ponds at the PD/A CRSP site at Sagana, Kenya, and to provide data for the development of a full-cost enterprise budget for the most profitable fertilization rate. Trials were conducted during both the warm and cool seasons to determine seasonal effects.

## METHODS AND MATERIALS

## **Cool-Season Experiment**

These experiments were conducted in twelve CRSP-managed  $800\text{-m}^2$  earthen research ponds at Sagana Fish Farm, Sagana, Kenya. The cool-season experiment was conducted between May and October 1998. Triple superphosphate (TSP) was applied to each pond at a rate of 250 kg P ha<sup>-1</sup> prior to filling. Each pond was also limed at a rate of 2.5 t ha<sup>-1</sup>. The lime and TSP were raked into the bottom of the pond. Inorganic fertilizer, either as TSP or as diammonium phosphate (DAP) and urea, was applied weekly beginning two weeks prior to stocking (first application on 29 April 1998). Soda ash (97% sodium carbonate), obtained from Magadi Soda (the world's largest soda mine), was applied weekly to ponds with total alkalinity less than 75 mg l<sup>-1</sup> as CaCO<sub>3</sub>.

DAP and urea were used to apply nitrogen to the ponds at rates of 0, 10, 20, and 30 kg N ha<sup>-1</sup> wk<sup>-1</sup>. A completely randomized design was used, with three replicates for each of the four treatments. The final fertilizer and soda ash applications were on 6 October 1998. The total quantities of inputs used are shown in Table 1.

All ponds were stocked with sex-reversed tilapia averaging 16.9 g at a rate of 1,000 kg ha<sup>-1</sup> on 13 May 1998. This resulted in a stocking rate of approximately 60,000 fish ha<sup>-1</sup>. Each pond also received 80 Clarias averaging 37 g, resulting in an additional 37 kg ha<sup>-1</sup>. Ponds were sampled at biweekly intervals by seining. All fish caught in the seine were separated by species, counted, and weighed. No attempt was made to obtain enough Clarias for a sample, so no Clarias were caught on some sampling dates, and growth curves were not made for this species. Ponds were drained on 16 October 1998 after it was determined that fish in all treatments had ceased to grow ("no growth" was defined as weight increases of less than 10% on two successive sampling occasions). Fish were separated by species, counted, and weighed. Of the fish harvested, 25,000 tilapia were sold to a cage culture operation for 120 KSh kg<sup>-1</sup>, 10,640 tilapia and 720 Clarias were used to restock ponds for the next experiment, and 4,000 tilapia and the remaining Clarias were used to stock ponds in a supplementary carryingcapacity experiment.

Temperature and dissolved oxygen (DO) were measured weekly at four depths (5, 25, 50, and 75 cm below surface) at the pier near the drain in the morning and the afternoon. Total alkalinity (TA) and pH were measured weekly using column samples collected at three places in each pond. Chlorophyll a (corrected and uncorrected), TAN, nitrite, nitrate, and soluble reactive-P were measured biweekly (a total of 11 measurements for each parameter). Total N, total P, and total hardness (TH) were measured every four weeks, with a total of eight total N and total P measurements made during the experiment. Methods used were as indicated in the CRSP Handbook of Analytical Methods (PD/A CRSP Technical Committee, 1992), with the exception of nitrate analysis, which was performed using the NAS method. Diurnal oxygen and temperature readings were also made on these occasions to calculate primary productivity using the whole-pond method. Water sampling and analysis was done on Mondays (all were usually finished the same day or by Tuesday), and fertilizers and soda ash were applied on Tuesdays. Fish sampling was done on Wednesdays.

Ponds' water levels were recorded daily. Ponds were topped off to replace evaporation and seepage losses weekly (Mondays or Tuesdays). Night watchmen occasionally added water to some ponds when fish deaths occurred, but these ponds never overflowed, so effects on water quality are thought to have been negligible.

On occasions of fish death, dissolved oxygen, TAN, and nitrites were measured as soon as possible after fish deaths were reported. Most fish die-offs occurred on weekends (about four days after fertilization). Dead fish were counted and weighed. Only the ponds receiving the highest nitrogen input were affected.

## Warm-Season Experiment

The warm-season experiment was conducted in the same 12 ponds between November 1998 and March 1999. At the end of the first (cool-season) experiment, residual lime and TSP could be seen on the bottoms of most ponds. Prior to refilling, ponds that had had TH less than 75 mg l<sup>-1</sup> at the end of the cool-season experiment were relimed. Liming rates varied according to TH at the end of the first experiment. No further

Table 1.Total quantities of inputs used during cool season (147-day duration) and warm season (133-day duration) Global Experiment on<br/>optimum nitrogen fertilization at Sagana, Kenya.

Weekly N Input (kg ha <sup>-1</sup> )	$\frac{\mathbf{DAP}}{(\mathrm{kg ha}^{-1})}$	<b>Urea</b> (kg ha <sup>-1</sup> )	TSP (kg ha <sup>-1</sup> )	<b>Lime</b> (kg ha <sup>-1</sup> )	<b>Soda Ash</b> (kg ha <sup>-1</sup> )
COOL SEASON					
0	0	0	1,585	2,500	283
10	960	146	625	2,500	492
20	960	668	625	2,500	229
30	960	1,190	625	2,500	254
WARM SEASON					
0	0	0	840	1,875	75
10	840	128	0	417	258
20	840	584	0	625	321
30	840	1.041	0	2.083	704

additions of TSP were deemed necessary because soluble phosphate levels in all ponds had remained high in the first experiment. Treatments for the warm-season experiment were the same as those for the cool-season experiment: DAP and urea were again used to add nitrogen at rates of 0, 10, 20, and  $30 \text{ kg N} \text{ ha}^{-1} \text{ wk}^{-1}$ .

Ponds were re-randomized and the first fertilizer application was made on 4 November 1998. Male tilapia averaging 90 g were used (left from the previous experiment) to stock all ponds at 1 t ha<sup>-1</sup> on 18 November. Sixty Clarias averaging 166 g were added to each pond (125 kg ha<sup>-1</sup>) on 23 November. This resulted in a stocking density of 11,000 tilapia and 750 Clarias ha-1. Two tilapia and two Clarias were removed from each pond once a month to examine stomach contents. The removed fish were counted as live when survival was calculated but their weight was not used in calculating gross fish yield, on the assumption that the fish remaining in the pond increased in weight to compensate for those that had been removed. Ponds were drained on 29 March 1999 after it was concluded that all treatments had less than 5% weight gain on two successive samplings. Fish were separated by species, counted, and weighed. Tilapia were separated by sex, and fingerling weight was noted. The tilapia and Clarias were sold at 120 KSh kg-1 to a Nairobi fish-out operator.

Water quality parameters were measured at the same frequency as in the cool-season experiment. Fertilizers and soda ash were applied on Tuesdays. Some ponds in the experiment exhibited very high morning pH and their hardness was beginning to diminish, so soda ash applications were suspended for ponds that had a morning column pH greater than 8.5. These ponds typically increased in pH to more than 10 before noon, so any additional carbonate would not have been available for photosynthesis. Chlorophyll *a* (corrected and uncorrected), TAN, nitrite, nitrate, and soluble reactive-P (a total of 9 measures for each parameter) were measured biweekly. Total N, total P, and TH were measured every four weeks. A total of five total N and total P measurements were made during the experiment.

Data for each experiment were analyzed statistically by regression analysis using Statgraphics Plus for Windows software (Statistical Graphics Corp. 1997). Models that resulted in the lowest *P* values (not necessarily the highest  $r^2$  values) were used.

## RESULTS

Preliminary observations are outlined in this progress report. A complete, final report will be submitted after completion of the data analysis for these experiments.

#### **Site Observations**

The Sagana site has very cool surface waters (usually 19 to 21°C) with low alkalinity and hardness compared to the El Carao station in Honduras and the AIT station in Thailand. TA runs about 16 to 28, and TH is 13 to 28, with higher values in dry months. There is not much difference in average air temperature in the so-called cool season. Minimum air temperatures tend to be the same but maximum temperatures are lower in the cool season due to increased cloud cover. Solar radiation during the cool season is much lower than during the warm season. Average solar radiation during the first experi-

ment was 24.64 Einsteins  $m^{-2}$  (7.56 MJ  $m^{-2}$ ), whereas it was 41.91 Einsteins  $m^{-2}$  (12.34 MJ  $m^{-2}$ ) during the warm-season experiment. The beginning and end of the cool-season experiment actually fell into warm-season months. There was a slight difference in pond water temperatures between the cool- and the warm-season experiments.

Another seasonal difference has to do with winds. In the cool season, wind is less and the ponds mix only in the night to early morning. In the warm season, especially December through February, high winds occur every afternoon and the ponds mix twice, once in the evening and once in the early morning.

During the cool season, ponds in the 20 and 30 kg N treatments had dense blooms of euglenophytes (*Trachelomonas, Euglena*, and *Phacus* spp.) These blooms did not occur in the warmseason experiment. The lower solar radiation tends to favor the euglenophytes during the cool season (these blooms have occurred each cool season since the CRSP began work at Sagana). Surface blooms of any kind cannot resist the intense solar radiation experienced at Sagana from December through February.

## Fish Growth

Figures 1 and 2 show average fish weight by nitrogen input rate for each experiment. In the cool season, the high nitrogen input rate showed high growth after about 25% of the fish died in two of the ponds.



Figure 1. Growth of tilapia under four different nitrogen input rates during the cool-season trial of the PD/A CRSP Global Experiment conducted at Sagana Fish Farm, Sagana, Kenya.



Time (d)

Figure 2. Growth of tilapia under four different nitrogen input rates during the warm-season trial of the PD/A CRSP Global Experiment conducted at Sagana Fish Farm, Sagana, Kenya.

Table 2.Mean (± SE) tilapia yields, Clarias yields, average weights by species, and survival for cool and warm season of the Global Experiment on<br/>optimum nitrogen fertilization at Sagana Fish Farm, Sagana, Kenya.

Week (kg ha	d <b>y N Input</b> a <sup>-1</sup> )		Yie (kg l	2 <b>ld</b> na <sup>-1</sup> )			Averag (	<b>e Weight</b> (g)	<b>Tilapia</b> <b>Reproduction</b> (kg ha <sup>-1</sup> )
		Tilapia	Clarias	Gross	Net		Tilapia	Clarias	_
Cool	Season								
$0$ 10 20 30 Best F $r^2$	<sup>7</sup> it Model	$1,015 \pm 113$ $2,602 \pm 107$ $2,953 \pm 296$ $2,510 \pm 348$ quadratic $82\%^{**}$	$208 \pm 34$ $270 \pm 59$ $250 \pm 22$ $495 \pm 24$ linear $57\%^*$	$1,297 \pm 116$ $2,949 \pm 120$ $3,229 \pm 278$ $3,043 \pm 371$ quadratic $82\%^{**}$	$251 \pm 117$ 1,908 ± 120 2,191 ± 280 2,004 ± 371 quadratic 82%**		$\begin{array}{c} 23.1 \pm 1.6 \\ 48.1 \pm 1.9 \\ 60.4 \pm 3.8 \\ 70.5 \pm 5.9 \\ \text{sq. root } x \\ 92\%^{**} \end{array}$	$\begin{array}{l} 156.1 \pm 2.9 \\ 263.2 \pm 40.6 \\ 275.1 \pm 28.2 \\ 375.7 \pm 45.3 \\ \text{reciprocal-y} \\ 78\%^{**} \end{array}$	$74 \pm 20.6$ $7 \pm 19.3$ $30 \pm 3.5$ $37 \pm 4.8$ linear $43\%^*$
WARM	M SEASON								
0 10 20 30 Best F <i>r</i> <sup>2</sup>	Fit Model	$1,119 \pm 115$ $1,672 \pm 156$ $1,720 \pm 100$ $1,520 \pm 160$ quadratic $68\%^{**}$	$\begin{array}{c} 154 \pm 23 \\ 164 \pm 6 \\ 181 \pm 89 \\ 183 \pm 16 \\ NS \end{array}$	$1,272 \pm 137$ $1,837 \pm 161$ $1,901 \pm 95$ $1,703 \pm 45$ quadratic 67%**	$296 \pm 162$ $883 \pm 169$ $957 \pm 91$ $761 \pm 28$ quadratic $67\%^{**}$	c	$\begin{array}{c} 106 \pm 8.3 \\ 162 \pm 12.4 \\ 181 \pm 4.3 \\ 168 \pm 6.5 \\ quadratic \\ 86\%^{**} \end{array}$	$\begin{array}{c} 251.3 \pm 29.5 \\ 262.9 \pm 3.8 \\ 283.2 \pm 6.7 \\ 283.1 \pm 16.4 \\ \text{NS} \end{array}$	$43 \pm 7.4$ 29 ± 2.3 34 ± 11.5 17.5 ± 0.9 exponential 41%*
** highly * signific	significant ( $P < 0.$ cant ( $P < 0.05$ )	.01)	<u>models used in Tal</u> linear: exponential: sq. root y:	bles 2 and 4: y = a + bx y = exp(a + b) $y = (a + bx)^2$	x) sq qu	. root x: ciprocal-y iadratic:	y = a + y = (a + y) = a + y	$bx^{1/2} + bx)^{-1} - bx + cx^2$	
	4000 J y =	Cool season -4.5968 $x^2$ + 193.07 $x$ - $r^2$ = 0.818	+ 1342.6			4000	$y = -249.17x^2 - r^2 = r^2$	ll season + 2600.9x - 3373.6 = 0.7929	•
	3500 -			•		3500			
a-1)	3000 -	•	•	•	-	3000			-
ield (kg h		-	•		l (kg ha <sup>-1</sup> )	2500 -			
fish y	2500 -	/			yield	2000			
Gross	2000 -		<b>A</b>	•	Gross fish	1500 -			•
		/		Å	U	1000		IAT- une	
	1500	y = -1.90	Warm season $29x^2 + 70.622x + r^2 = 0.6743$	+ 1285.1		500 -		$y = -86.349x^2 + 8$ $r^2 = 0$	27.19x - 20.978 .7287
	1000					0	1 2	3 4	5 6 7
	U	N input (kg k	$20 20$ $10^{-1}$ wk <sup>-1</sup> )	30 33			Ave	erage total N (m	g l <sup>-1</sup> )

Figure 3. Gross fish yield (GFY) as a function of nitrogen (N) input for the cool- and warm-season trials of the PD/A CRSP Global Experiment conducted at Sagana Fish Farm, Sagana, Kenya.

Figure 4. Relationship between gross fish yield (GFY) and average total nitrogen (N) content in pond waters during the cooland warm-season trials of the PD/A CRSP Global Experiment conducted at Sagana Fish Farm, Sagana, Kenya.

The relationship between gross fish yield (GFY) and nitrogen input rate was best described by a quadratic model. It was highly significant (P < 0.01) in both experiments but better correlated in the cool-season experiment (Table 2 and Figure 3). Fish deaths occurred in the treatment with the highest nitrogen application rate on at least two occasions during the cool-season experiment, on 12–13 June in pond E09 and on 22 June in pond D04. Dead tilapia were picked up occasionally during the warmseason experiment but they never totaled more than two or three on any day. No *Clarias* died on these occasions. In fact, the relationship between *Clarias* yield and nitrogen input rate was linear, suggesting that they are less affected by the high nitrogen.

Taking the first derivative of the regression equation gives the nitrogen input rate at which maximum GFY would be obtained: 21.0 kg N ha<sup>-1</sup> wk<sup>-1</sup> for the cool-season experiment and 18.6 kg N ha<sup>-1</sup> wk<sup>-1</sup> for the warm-season experiment. A highly significant relationship exists between GFY and average total N in ponds as well (Figure 4). Differences in initial fish size and density do not permit a valid comparison of growth and productivity as a function of season.

At draining of the cool-season experiment, every pond had at least two large *Clarias* (> 1 kg). These fish were probably left over from the previous experiment. Some ponds contained many more *Clarias* than were stocked, and these extra fish may have entered with the tilapia fingerlings. It was easier to separate out *Clarias* during stocking in the second experiment because fish were fewer in number and larger.

The weight of tilapia reproduction decreased with increasing nitrogen input (Table 2). Size of tilapia females, *Clarias* size, or adverse water quality due to high nitrogen input rate could have been causes.

## Water Quality

During the warm season the ponds did have higher water temperatures and lower morning dissolved oxygen concentrations than they did during the cool season (Table 3). If the difference between afternoon and morning dissolved oxygen is used as an indicator of net primary production, then primary production was a bit higher in

Table 3.Dissolved oxygen (DO) and temperature observations in ponds during the cool- and warm-season trials of the Global Experiment on<br/>optimum nitrogen fertilization conducted at Sagana Fish Farm, Sagana, Kenya.

Ponds	Weekly N Input (kg ha <sup>-1</sup> )	Dissolved Oxygen (mg l <sup>-1</sup> )							
		Morning					After	rnoon	
		5 cm	25 cm	50 cm	75 cm	5 cm	25 cm	50 cm	75 cm
COOL									
D7, D8, E6 D6, E4, E5 D5, E7, E8 D4, E3, E9	0 10 20 30	4.6 4.3 3.5 2.5	4.5 4.1 3.3 2.2	4.3 4.0 3.2 2.0	3.7 3.7 2.8 1.9	12.1 14.7 16.5 15.9	10.6 13.6 15.3 13.7	5.3 6.9 7.2 7.0	2.9 3.7 3.1 3.3
WARM									
D4, D8, E9 D7, E3, E6 E4, E5, E7 D5, D6, E8	0 10 20 30	3.0 2.1 2.0 1.8	3.0 2.0 1.9 1.8	3.0 2.0 1.9 1.8	2.9 1.9 1.9 1.7	9.5 10.9 11.9 13.4	9.3 10.1 10.5 12.7	6.5 7.4 7.1 8.7	3.3 3.1 2.9 3.9

Ponds	Weekly N Input (kg ha <sup>-1</sup> )	Temperature (°C)							
		Morning					Afteri	10011	
	-	5 cm	25 cm	50 cm	75 cm	5 cm	25 cm	50 cm	75 cm
COOL									
D7, D8, E6 D6, E4, E5 D5, E7, E8 D4, E3, E9	0 10 20 30	22.5 22.9 22.8 22.9	22.5 22.9 22.8 22.8	22.5 22.9 22.8 22.8	22.5 22.9 22.8 22.8	26.4 26.3 26.4 26.1	25.9 26.0 26.0 25.7	24.0 24.6 24.5 24.7	23.4 23.8 23.8 23.9
WARM									
D4, D8, E9 D7, E3, E6 E4, E5, E7 D5, D6, E8	0 10 20 30	23.5 23.5 23.6 23.6	23.5 23.5 23.6 23.3	23.5 23.5 23.6 23.3	23.5 23.5 23.6 23.3	28.2 27.8 27.6 28.0	28.0 27.6 27.5 27.9	26.4 26.5 26.5 26.6	24.7 25.1 25.0 24.7

the cool season than the warm season. This relationship will be examined in more detail in the final report for this experiment.

Observations for TH, TA, corrected and uncorrected chlorophyll *a*, soluble reactive phosphorus, pH, total N, ammonia-N, nitrite-N, and nitrate-N for both the cool- and warm-season experiments are shown in Table 4. In both

experiments, it can be concluded that phosphate was not limiting in any way. Attempts to keep TA high were successful in the first experiment but not in the second. Decreasing total hardness meant that there was less buffering capacity, which probably kept pH high in ponds receiving large amounts of sodium carbonate. Although TA averaged less than 60 mg l<sup>-1</sup>, chlorophyll *a* levels were higher in the highest nitrogen input treatment.

Table 4.Mean (± SE) pond water quality observations during the cool- and warm-season Global Experiments on optimum nitrogen fertilization at<br/>Sagana Fish Farm, Sagana, Kenya.

<b>Weekly N Input</b> (kg ha <sup>-1</sup> )	<b>Total</b> <b>Alkalinity</b> (mg l <sup>-1</sup> as CaCO <sub>3</sub> )	<b>Total Hardness</b> (mg l <sup>-1</sup> as CaCO <sub>3</sub> )	<b>Corrected</b> <b>Chlorophyll</b> <i>a</i> (mg m <sup>3</sup> )	<b>Uncorrected</b> <b>Chlorophyll</b> <i>a</i> (mg m <sup>3</sup> )	Total Phosphorus (mg l <sup>-1</sup> )	Soluble Reactive Phosphorus (mg l <sup>-1</sup> )
COOL SEASON						
0 10 20 30 Best Fit Model Correlation Std. Error Est.	89.7 ± 15.9 73.8 ± 5.5 74.8 ± 8.5 77.9 ± 3.6 NS	$75.5 \pm 18.3 \\ 60.0 \pm 7.2 \\ 52.1 \pm 5.4 \\ 60.0 \pm 8.0 \\ NS$	$60.2 \pm 2.7$ $85.0 \pm 8.3$ $156.5 \pm 43.8$ $185.4 \pm 15.5$ reciprocal-y $86^{**}$ 0.00277	$107.5 \pm 10.6$ $171.5 \pm 14.5$ $233.0 \pm 52.3$ $277.6 \pm 20.2$ exponential $.84^{**}$ 0.243	$\begin{array}{c} 2.0 \pm 0.72 \\ 1.9 \pm 0.29 \\ 2.4 \pm 0.12 \\ 1.3 \pm 0.24 \end{array}$ NS	$\begin{array}{c} 1.0 \pm 0.46 \\ 1.1 \pm 0.20 \\ 1.2 \pm 0.06 \\ 0.6 \pm 0.14 \\ \\ NS \end{array}$
0 10 20 30 Best Fit Model Correlation Std. Error Est.	$84.8 \pm 6.3$ $69.6 \pm 0.9$ $64.0 \pm 1.6$ $55.3 \pm 0.8$ reciprocal .95** 0.0008	$80.2 \pm 7.2$ $59.2 \pm 3.4$ $47.5 \pm 4.3$ $30.8 \pm 3.6$ sq. root-y $94^{**}$ 0.511	$89.5 \pm 37.6$ $109.3 \pm 8.8$ $182.2 \pm 78.4$ $198.5 \pm 16.9$ exponential .63* 0.514	$89.9 \pm 23.5$ $147.3 \pm 7.0$ $252.3 \pm 97.8$ $271.0 \pm 25.7$ exponential $.76^{**}$ 0.412	$\begin{array}{c} 1.5 \pm 0.06 \\ 3.0 \pm 1.88 \\ 2.6 \pm 0.47 \\ 3.7 \pm 0.42 \\ \text{reciprocal} \\75^{**} \\ 0.129 \end{array}$	$0.7 \pm 0.14$ $1.4 \pm 0.39$ $1.3 \pm 0.08$ $1.5 \pm 0.23$ exponential 65* 0.357
<b>Weekly N Input</b> (kg ha <sup>-1</sup> )	рН	<b>Total Nitrogen</b> (mg l <sup>-1</sup> )	$\begin{array}{c} \textbf{Ammonia-N} \\ (mg \ l^1) \end{array}$	<b>NO<sub>2</sub>-N</b> (mg l <sup>-1</sup> )	<b>NO<sub>3</sub>-N</b> (mg l <sup>-1</sup> )	Soda Ash Added to Ponds (kg ha <sup>-1</sup> )
COOL SEASON						
0 10 20 30 Best Fit Model Correlation Std. Error Est.	$7.5 \pm 0.13 7.8 \pm 0.16 8.0 \pm 0.25 7.5 \pm 0.07 NS$	$\begin{array}{c} 2.5 \pm 0.27 \\ 3.5 \pm 0.16 \\ 5.7 \pm 0.35 \\ 6.4 \pm 0.75 \\ \text{linear} \\ .96^{**} \\ 0.529 \end{array}$	$\begin{array}{c} 0.8 \pm 0.06 \\ 0.9 \pm 0.12 \\ 1.1 \pm 0.07 \\ 1.3 \pm 0.09 \\ \text{linear} \\ .82^{**} \\ 0.140 \end{array}$	$\begin{array}{c} 0.0 \pm 0.00 \\ 0.0 \pm 0.02 \\ 0.3 \pm 0.08 \\ 0.7 \pm 0.20 \\ linear \\ .83^{**} \\ 0.196 \end{array}$	$\begin{array}{c} 0.0 \pm 0.01 \\ 0.1 \pm 0.02 \\ 0.3 \pm 0.14 \\ 0.3 \pm 0.06 \\ \text{sq. root-y} \\ .79^{**} \\ 0.143 \end{array}$	$254 \pm 242$ $229 \pm 67$ $491 \pm 256$ $283 \pm 136$ NS
WARM SEASON						
0 10 20 30 Best Fit Model Correlation Std. Error Est.	$7.7 \pm 0.31 \\ 8.4 \pm 0.13 \\ 8.8 \pm 0.30 \\ 8.8 \pm 0.20 \\ sq. root-x \\ .77^{**} \\ 0.388$	$\begin{array}{c} 2.1 \pm 0.33 \\ 3.3 \pm 0.16 \\ 5.0 \pm 0.69 \\ 6.1 \pm 0.52 \\ \text{sq. root-y} \\ .92^{**} \\ 0.179 \end{array}$	$\begin{array}{c} 0.5\pm 0.05\\ 0.5\pm 0.10\\ 0.6\pm 0.09\\ 0.8\pm 0.08\\ \text{linear}\\ .67^{*}\\ 0.144 \end{array}$	$\begin{array}{c} 0.0 \pm 0.00 \\ 0.0 \pm 0.01 \\ 0.1 \pm 0.05 \\ 0.3 \pm 0.11 \\ \text{sq. root-y} \\ .90^{**} \\ 0.101 \end{array}$	$\begin{array}{c} 0.0 \pm 0.03 \\ 0.1 \pm 0.03 \\ 0.2 \pm 0.08 \\ 0.3 \pm 0.01 \\ \text{linear} \\ .77^{**} \\ 0.119 \end{array}$	$75 \pm 38$ $258 \pm 33$ $321 \pm 17$ $704 \pm 115$ sq. root-y $.89^{**}$ 1.05

\*\* highly significant (P < 0.01)

\* significant (P < 0.05)

Table 5. Cost of inputs and revenue per hectare for the optimum nitrogen fertilization experiments (Global Experiment) conducted at Sagana Fish Farm, Sagana, Kenya, during 1998 and 1999. Costs include initial phosphorus-saturation inputs of TSP applied to ponds for the coolseason experiment. (US\$1 = 60 KSh)

Treatment				Weekly (kg	<b><sup>•</sup> N Input</b> ha <sup>-1</sup> )			
		Cool	Season			Warm	ı Season	
-	0	10	20	30	0	10	20	30
Costs (KSh ha <sup>-1</sup> )								
DAP Urea TSP Lime Soda Ash Stockers Total Cost	- 39,625 10,000 2,796 124,440 176,861	24,000 2,776 15,625 10,000 2,521 124,440 179,362	$\begin{array}{c} 24,000\\ 12,688\\ 15,625\\ 10,000\\ 5,408\\ 124,440\\ 192,161 \end{array}$	24,000 22,601 15,625 10,000 2,521 124,440 199,186	- 36,625 7,500 825 135,000 179,950	21,000 2,429 - 1,667 2,842 135,000 162,937	21,000 11,102 - 2,500 3,529 135,000 173,131	21,000 19,775 - - 8,333 7,746 135,000 191,855
REVENUES (Ksh)								
Tilapia Revenue Clarias Revenue Total Revenue	121,800 24,960 146,760	312,240 32,400 344,640	354,360 30,000 384,360	301,200 59,400 360,600	134,280 18,480 152,760	200,640 19,680 220,320	206,400 22,080 228,480	182,400 21,960 204,360
GROSS PROFIT								
KSh \$US	-30,101 -502	165,278 2,755	192,199 3,203	161,414 2,690	-27,190 -453	57,383 956	55,349 922	12,505 208





## **Economic Analysis**

The costs of inputs and revenues per hectare for these experiments are shown in Table 5. The highest marginal return (increase in revenues less increased costs) was obtained at the 20 kg N ha<sup>-1</sup> wk<sup>-1</sup> rate in both experiments. Maximum marginal returns were calculated at input rates of 19.9 and 16.0 kg N ha<sup>-1</sup> wk<sup>-1</sup> for the cool- and warm-season experiments, respectively (Figure 5).

## **Carry-Over Effect**

Table 6 was assembled to examine the possible effects of previous treatments on the outcome of the second experiment.

Table 6.Performance of ponds in the second trial in relation to the<br/>nitrogen input level received in the first trial of the Global<br/>Experiment on optimum nitrogen fertilization, Sagana,<br/>Kenya.

<b>Weekly N Input</b> <b>in Second Expt.</b> (kg ha <sup>-1</sup> )	Performance in Second Expt.	Weekly N Input in First Expt. (kg ha <sup>-1</sup> )
0	Best pond: D04	30
	Worst pond: D08	0
10	Best pond: E03	30
	Worst pond: E06	0
20	Best pond: E05	10
	Worst pond: E07	20
30	Best pond: D06	10
	Worst pond: E08	20



Figure 6. Combined net fish yield in relation to total nitrogen input for the cool- and warm-season trials of the Global Experiment conducted at Sagana Fish Farm, Sagana, Kenya.

Ponds that received no nitrogen during the first experiment were the worst performers in the lower nitrogen input rates of the second experiment and the best performers in the high-N treatments of the second experiment. Ponds that received high nitrogen inputs were better performers (in terms of fish yields) in the no nitrogen input treatments of the second experiment and the worst performers in the high nitrogen input treatments. It therefore appears that overall production over the two experiments was related to cumulative nitrogen input. Figure 6 shows this relationship. Combined net fish yields for the cool- and warm-season experiments were highly correlated to combined nitrogen inputs over the two experiments. Unfortunately, no pond received two consecutive highest nitrogen input rates. The calculated maximum net fish yield would occur at 1,183.5 kg ha<sup>-1</sup> N input over 45 weeks or 26.3 kg N ha<sup>-1</sup> wk<sup>-1</sup>. This relationship was the most highly correlated of any fish yield relationship examined.

#### DISCUSSION

The observations of water quality parameters (chlorophyll *a* and total N), which usually correlate well with high tilapia production, were very similar in both experiments. They definitely do not explain why fish production was so low in the warm-season experiment. Production was lower in the second experiment at the El Carao site as well but no explanation was given (Green et al., 1999). A lack of significant differences in fish yields in the second experiment at El Carao was attributed to carry-over effects of the nitrogen from the preceding experiment. The results from Sagana also suggest some carry-over effect or possibly the effect of differences in initial stocking size and density.

In addition to conducting the first round of the Global Experiment on optimum nitrogen, researchers at the Asian Institute of Technology (AIT) in Thailand conducted a complementary study, in which they compared production from ponds that had been stocked with three different sizes of tilapia. They concluded that stocking with medium-sized fish (averaging 10 g in size) resulted in greater production than stocking with either small tilapia (averaging 4.6 g in size) or larger fingerlings (averaging 21 g in size). The fish used to stock ponds in the first round of the Sagana experiment were in between the medium and large sizes used in the AIT study.

The results obtained at Sagana are more similar to those obtained at the El Carao site than to those from the AIT site.

Increasing nitrogen input rates to 30 kg ha<sup>-1</sup> wk<sup>-1</sup> did not result in increased fish yields at either El Carao or Sagana. The high water temperatures that are typical at the AIT site (29 to 37°C) are a plausible explanation for a higher optimum nitrogen input rate there. Hatchery technicians who work with recirculating systems know that the capacity of a biofilter to process nitrogen is affected by temperature, with maximum nitrogen loading rates being higher at higher temperatures.

The two experiments conducted at Sagana took much longer than at the other two CRSP sites. The experiment was terminated after 91 days at AIT and after 121 and 107 days at El Carao. In contrast, the cool-season experiment at Sagana lasted 147 days and the warm-season experiment lasted 133 days.

These results and questions led us to qualify our recommendations for farmers. A chemical fertilizer-only treatment will probably not form part of the extension recommendations to farmers in Kenya because of problems in controlling alkalinity. Organic matter inputs seem to stabilize the pond system and maintain total alkalinity. However, the optimum nitrogen input rate will be used in setting input recommendations and in estimating carrying capacity for certain input combinations. Farmers in cooler areas will be cautioned to not exceed 16 kg ha<sup>-1</sup> wk<sup>-1</sup> of chemical nitrogen.

#### **ANTICIPATED BENEFITS**

The results from this experiment were valuable additions to the formulation of extension recommendations for tilapia culture in the East Africa region. Combining the discussions from the three PD/A CRSP sites that conducted the Global Experiment can lead to some additional research topics as well: Fish size as a variable in carrying capacity estimates and temperature as the main reason for different optimum N input rates. The temperature question has been answered for the most part but the question of fish size versus carrying capacity merits further work, especially for systems using filter feeders and relying on natural production.

#### **ACKNOWLEDGMENTS**

The work of Mr. James Karuri, lab technician, and Thomas Ndegwa, lab assistant, was invaluable. Paul Wamwea Wabitah helped out in keeping data for the pond sampling. Our super seine crew demonstrated that 12 ponds can indeed be sampled in under two hours—a feat never thought possible at Sagana. Mr. John Kogi was a highly reliable pond manager and recordkeeper.

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## GLOBAL EXPERIMENT: OPTIMIZATION OF NITROGEN FERTILIZATION RATE IN FRESHWATER TILAPIA PRODUCTION PONDS DURING COOL SEASON

Eighth Work Plan, Feeds and Fertilizers Research 1T (8FFR1T) Final Report

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## Abstract

An experiment was conducted in twelve 200-m<sup>2</sup> earthen ponds at the Asian Institute of Technology, Thailand, for 91 days from 11 September to 11 December 1998. The experiment was designed to determine the optimal rate of nitrogen fertilization in cool season, to determine which of the nitrogen fertilization rates evaluated to produce Nile tilapia had the greatest profitability, and to develop a full-cost enterprise budget for the fertilization level that resulted in greatest profitability. Treatment ponds were fertilized with TSP at a rate of 8 kg ha<sup>-1</sup> wk<sup>-1</sup>, and with urea at 0, 10, 20, and 30 kg N ha<sup>-1</sup> wk<sup>-1</sup>, respectively. Sex-reversed male Nile tilapia were stocked at 1,000 kg ha<sup>-1</sup> at a size of 23.1 to 25.5 g in all ponds (4.1 fish m<sup>-2</sup>). Sodium bicarbonate was applied to all ponds weekly to attain and maintain the minimum alkalinity (75 mg l<sup>-1</sup> as CaCO<sub>3</sub>) based on weekly measurement of alkalinity in pond water. The experiment showed that greater nitrogen inputs resulted in better growth performance of Nile tilapia. Growth in the treatment without N inputs declined after the first fish sampling, which was earlier than the decline (around day 70) in treatments with N inputs. During the entire culture period, the estimated fish biomass was highest in the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup>, intermediate in the treatments with 10 and 20 kg N ha<sup>-1</sup> wk<sup>-1</sup>, and lowest in the treatment without N inputs. The highest gross yield of Nile tilapia was obtained in the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> (1,938 ± 257 kg ha<sup>-1</sup>, mean ± SE), intermediate in the treatments with 10 and 20 kg N ha<sup>-1</sup> wk<sup>-1</sup>  $(1,628 \pm 190 \text{ and } 1,755 \pm 190 \text{ kg ha}^{-1}$ , respectively), and the lowest in the treatment without N inputs  $(818 \pm 109 \text{ kg ha}^{-1})$ . The partial budget analysis indicated that the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> was most profitable. The full-cost enterprise budget showed that US\$2.1 net return could be produced from a 200-m<sup>2</sup> pond in this treatment during a three-month culture period.

#### INTRODUCTION

Nile tilapia (Oreochromis niloticus) are commonly grown in semi-intensive culture using fertilizers to increase primary production and fish food (Boyd, 1976; Diana et al., 1991). There is voluminous literature on pond fertilization, documenting many conflicting and inconsistent results based on various types of fertilizer, rates of input, and methods and frequency of application (Coleman and Edwards, 1987). An efficient production system requires optimal use of nutrient inputs. Among a large number of nutrients required to stimulate phytoplankton growth, nitrogen, phosphorus, and occasionally carbon are the most common limiting nutrients in natural water and fish ponds (Lin et al., 1997). The research of the Pond Dynamics/ Aquaculture Collaborative Research Support Program (PD/A CRSP) has addressed enhancement of primary productivity in ponds through additions of inorganic and organic fertilizers. However, the findings on optimal nitrogen, phosphorus, and carbon inputs required to improve fish yields at the PD/A CRSP sites appear to be inconsistent, and further research is needed. Higher nutrient inputs increased fish production at all PD/A CRSP sites, but optimal inputs of nitrogen, phosphorus, and carbon were not well defined (Lin et al., 1997).

Fertilization rates in PD/A CRSP experiments were much greater than rates reported for earlier pond fertilization research (Lin et al., 1997). In an often-cited series of fertilization experiments conducted in Malaysia, Hickling (1962) used less than 1.1 kg P and 1.1 kg N ha<sup>-1</sup> wk<sup>-1</sup>. In Israel, the standard fertilizer dose was 2.3 kg P and 6.5 kg N ha<sup>-1</sup> wk<sup>-1</sup> (Hepher, 1962a, b). The highest rates of phosphorus and nitrogen used in most experiments at Auburn University were 1.26 and 3 kg ha<sup>-1</sup> wk<sup>-1</sup>, respectively (Swingle, 1947; Boyd, 1976, 1990; Boyd and Sowles, 1978; Murad and Boyd, 1987). Rates in Europe seldom exceeded 1 kg ha<sup>-1</sup> wk<sup>-1</sup> for nitrogen and phosphorus (Mortimer, 1954). However, rates used in Malaysia, USA, Israel, and Europe gave low fish production. Also, in all of the studies cited above phosphorus was the most important limiting nutrient.

Therefore, the purposes of this study were to:

- Determine the optimal rate of nitrogen fertilization (in the presence of adequate phosphorus and carbon) to obtain optimum primary productivity and optimum yields of Nile tilapia in freshwater production ponds;
- Determine which of the nitrogen fertilization rates evaluated to produce Nile tilapia in freshwater production ponds had the greatest profitability;

3) Develop a full-cost enterprise budget for the fertilization level that results in greatest profitability.

## METHODS AND MATERIALS

An experiment was conducted using a randomized complete block design in twelve 200-m<sup>2</sup> ponds at the Asian Institute of Technology, Thailand. The experiment involved the culture of Nile tilapia using four nitrogen fertilization rates.

All ponds were fertilized with triple superphosphate (TSP) at a rate of 8 kg ha<sup>-1</sup> wk<sup>-1</sup> and nitrogen (N), as urea, at 0, 10, 20 and 30 kg ha<sup>-1</sup> wk<sup>-1</sup>. All treatments were in triplicate. Initial pond fertilization took place two weeks prior to stocking of fish. Sodium bicarbonate was added weekly to attain and maintain minimum alkalinity (75 mg l<sup>-1</sup> as CaCO<sub>3</sub>) based on weekly measurement of alkalinity in pond water. Sex-reversed male Nile tilapia were stocked at 1,000 kg ha<sup>-1</sup> at a size of 23.1 to 25.5 g in all ponds on 11 September 1998. The stocking density averaged 4.1 fish m<sup>-1</sup> in all ponds.

Water depth in all ponds was maintained at 1 m throughout the experiment by adding water weekly to replace evaporation and seepage losses.

Most parameters of pond water quality were analyzed with column water samples taken from the center of each pond. Parameters of pond water quality, including total nitrogen (TN), total ammonium nitrogen (TAN), total phosphorus (TP), Secchi disk visibility, total alkalinity, and chlorophyll *a*, were analyzed at 1000 h during the second week (15 September 1998), midway (29 October 1998), and final week (8 December 1998) of the experiment using Standard Methods (APHA, 1980) modified by Egna et al. (1987). Dissolved oxygen (DO), pH, and temperature measurements were made on these three sampling dates at 0600, 1000, 1600, 1800, and 0600 h the following morning at 5-cm, 25-cm, 50-cm, and 75-cm depths in the water column. Total alkalinity and total hardness were determined weekly at 1000 h for calculating the amount of sodium bicarbonate required to maintain the minimum alkalinity defined as above.

During the experiment approximately 10% of the initial stock was seined, counted, and weighed en masse biweekly for each

pond. All fish were harvested on 11 December 1998 after 91 days of culture. Daily weight gain (g fish<sup>-1</sup> d<sup>-1</sup>), yield (kg pond<sup>-1</sup>), and extrapolated yield (kg ha<sup>-1</sup>) were calculated. Fish biomass on the sampling dates was calculated by the measured mean fish weight from sampling and the estimated number of fish surviving. It was assumed that surviving fish number decreased linearly from the beginning to the end of the experiment.

Data from the experiment were analyzed statistically by regression analysis and analysis of variance (ANOVA) using the SPSS 7.0 statistical software package. Differences were considered significant at an alpha level of 0.05. All means were given with  $\pm$  1 standard error (SE).

A partial budget analysis was conducted to determine which fertilization rate yielded the greatest profitability, and a full-cost enterprise budget was developed for the fertilization rate that yielded the greatest profitability (Shang, 1990). The economic analyses were based on the current local market prices expressed in US dollar (US\$1 = 40 baht) in Thailand. Prices of urea and TSP were \$0.200 and \$0.325 kg<sup>-1</sup>, respectively. Market value of Nile tilapia fingerlings around 30-g size was \$1.50 kg<sup>-1</sup>. To simplify the analyses the prices for stocked and harvested fingerlings were fixed to US\$1.50 kg<sup>-1</sup>. Total fixed cost in the full-cost enterprise budget was derived from a previous study (Engle and Skladany, 1992).

## RESULTS

Growth performance of Nile tilapia was better with higher N inputs (Table 1; Figures 1, 2, and 3). Survival rates were not significantly different (P > 0.05) among all treatments. Differential growth of Nile tilapia among all treatments was observed at the first fish sampling (Figure 1). Growth in the treatment without N inputs ceased after the first fish sampling, while growth in the treatments with N inputs continued steadily through the experimental period (Figure 1). Final mean weight, mean daily weight gain, and gross and net fish yields were significantly higher (P < 0.05) in treatments with N inputs. However, there were no significant differences (P > 0.05) for those growth parameters among the treatments with N inputs (Table 1).

Table 1.Growth performance of Nile tilapia in ponds fertilized with different N rates (0, 10, 20, and 30 kg ha<sup>-1</sup> wk<sup>-1</sup>) in the 91-day experiment.<br/>Mean values with different superscript letters in the same row are significantly different (P < 0.05).

Performance	Treatment						
	0	10	20	30			
Initial Biomass (kg pond <sup>-1</sup> )	$20.0\pm0.0^{\text{a}}$	$20.0\pm0.0^{\text{a}}$	$20.0\pm0.0^{\text{a}}$	$20.0\pm0.0^{\text{a}}$			
Initial Mean Weight (g fish <sup>-1</sup> )	$24.3\pm0.6^{\rm a}$	$23.4\pm0.3^{\rm a}$	$24.3\pm0.4^{\rm a}$	$24.7\pm0.5^{\rm a}$			
Final Biomass (kg pond <sup>-1</sup> )	$16.4\pm2.2^{\rm a}$	$32.6\pm3.1^{\rm b}$	$35.1\pm3.8^{\rm b}$	$38.8\pm5.1^{\rm b}$			
Final Mean Weight (g fish <sup>-1</sup> )	$28.2\pm0.8^{\rm a}$	$46.5\pm1.6^{\rm b}$	$53.9\pm6.5^{\rm b}$	$61.8\pm8.0^{\rm b}$			
Mean DWG (g fish <sup>-1</sup> $d^{-1}$ )	$0.04\pm0.01^{\text{a}}$	$0.25\pm0.02^{\rm b}$	$0.33\pm0.07^{\rm b}$	$0.41\pm0.08^{\rm b}$			
Net Fish Yield (kg pond <sup>-1</sup> )	$-3.6\pm2.2^{\rm a}$	$12.6\pm3.1^{\rm b}$	$15.1\pm3.8^{\rm b}$	$18.8\pm5.1^{\rm b}$			
Extrapolated Net Fish Yield (kg ha <sup>-1</sup> )	$\textbf{-181.7} \pm 109.4^{a}$	$628.3 \pm 155.3^{\rm b}$	$755.0\pm190.0^{\text{b}}$	$938.3\pm256.7^{\text{b}}$			
Gross Fish Yield (kg pond <sup>-1</sup> )	$16.4\pm2.2^{\rm a}$	$32.6\pm3.1^{\rm b}$	$35.1\pm3.8^{\rm b}$	$38.8\pm5.1^{\rm b}$			
Extrapolated Gross Fish Yield (kg ha <sup>-1</sup> )	$818.3\pm109.4^{\rm a}$	$1,628.3 \pm 155.3^{b}$	$1,\!755.0\pm190.0^{\rm b}$	$1,938.3 \pm 256.7^{\mathrm{b}}$			
Survival (%)	$70.5\pm9.0^{\text{a}}$	$81.9\pm5.9^{\rm a}$	$79.4\pm2.6^{\rm a}$	$77.3\pm1.7^{\rm a}$			

During the entire culture period, the estimated fish biomass was highest in the treatment with 30 kg N, intermediate in the treatments with 10 and 20 kg N, and lowest in the treatment without N inputs (Figure 2). The estimated fish biomass decreased after the first fish sampling (day 14) in the treatment without N inputs, resulting in negative net fish yields, while estimated fish biomass in the treatments with various N inputs kept increasing until the later part of the experimental period (around day 70) (Figure 2). Gross and net fish yields increased with increasing N inputs. The highest gross yield of Nile tilapia was obtained in the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup>  $(1,938 \pm 256 \text{ kg ha}^{-1})$ , intermediate in the treatments with 10 and 20 kg N ha<sup>-1</sup> wk<sup>-1</sup> (1,628  $\pm$  190 and 1,755  $\pm$  190 kg ha<sup>-1</sup>, respectively), and the lowest in the treatment without N inputs  $(818 \pm 109 \text{ kg ha}^{-1})$  (Table 1; Figure 3). The relationship between net fish yield and N inputs (Figure 4) can be expressed as Y = 34.87 X + 12.00 ( $R^2 = 0.60$ ,  $\tilde{P} < 0.05$ ), which shows that net fish yield increases with increasing N inputs.



Figure 1. Growth of Nile tilapia in treatments with 0, 10, 20, and 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> during the 91-day experiment.



Figure 2. Estimated fish biomass in treatments with 0, 10, 20, and  $30 \text{ kg N} \text{ ha}^{-1} \text{ wk}^{-1}$  during the 91-day experiment.

Water quality parameters varied among treatments in the experiment (Table 2). There were no significant differences (P > 0.05) for all measured parameters except TN at the initial measurements. Water temperature and pH ranged from 26.0 to 36.8°C and 6.0 to 10.3, respectively, throughout the experimental period in all ponds. The DO concentration at dawn fluctuated over the entire culture period, but there were no significant differences (P > 0.05) among treatments. TN and TAN concentrations increased with increasing N inputs for all three measurements during the experiment. The final TN concentrations were significantly higher (P < 0.05) in treatments with N inputs than those in the treatment without N inputs. The final TAN concentrations in treatments with 20 and 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> were significantly (P < 0.05) higher than that in the treatment without N inputs. However, there were no significant differences (P > 0.05) in final TAN concentrations between the treatments with 10 kg N ha<sup>-1</sup> wk<sup>-1</sup> and without N inputs. TP concentration was not significantly different (P > 0.05) among all treatments during the entire experimental period. To maintain the minimal alkalinity, the total amount of sodium bicarbonate added to ponds was  $21.1 \pm 0.9$ ,  $22.2 \pm 1.9$ ,  $21.0 \pm 3.3$ , and  $19.6 \pm 2.5$  kg for the treatments with 0, 10, 20, and 30 kg N, respectively. Alkalinity fluctuated between 39 and 97 mg  $l^{-1}$  for all treatments during the culture period (Figure 5). There were no significant differences (P > 0.05) in alkalinity concentrations among all treatments during the entire culture

2500 Cr-PH SD 1500 1500 1000 0 -500

Figure 3. Gross and net fish yields in treatments with 0, 10, 20, and  $30 \text{ kg N} \text{ ha}^{-1} \text{ wk}^{-1}$  in the 91-day experiment.



Figure 4. The relationship between net fish yield and nitrogen input rate in the 91-day experiment.

period. The concentrations of chlorophyll *a* were generally higher with increasing N inputs. The final concentrations of chlorophyll *a* were significantly higher (P < 0.05) in the treatment with N inputs than the treatment without N inputs. The treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> had significantly higher (P < 0.05) final chlorophyll *a* concentration than the treatments with lower N inputs (10 and 20 kg N ha<sup>-1</sup> wk<sup>-1</sup>), but there were no significant differences (P > 0.05) between the latter two.

The partial budget analysis indicated that the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> was most profitable. However, the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> resulted in a high ratio of added income to added cost (Table 3). The full-cost enterprise budget showed that a \$2.10 net return could be produced from a 200-m<sup>2</sup> pond in the treatment with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> during a 3-month culture period (Table 4).

## DISCUSSION

The addition of nitrogen fertilizer significantly increased Nile tilapia yields. Greater N inputs resulted in higher phytoplankton productivity, giving higher tilapia yields. The nitrogen inputs at rates of 10, 20, and 30 kg ha<sup>-1</sup> wk<sup>-1</sup> in ponds fertilized with 8 kg P ha<sup>-1</sup> wk<sup>-1</sup> brought the N:P ratio to 1.25:1, 2.5:1, and 3.75:1, respectively. The best fish growth performance was achieved in the treatment with the highest N input and N:P ratio. The result is consistent with that of a similar experiment done in the warm season (Lin et al., 1999). This finding confirms the previous results of CRSP experiments which showed optimal rates of 28 kg N ha<sup>-1</sup> wk<sup>-1</sup> and 7 kg P ha<sup>-1</sup> wk<sup>-1</sup> giving a 4:1 N:P ratio (Knud-Hansen et al., 1991; Lin et al., 1997). Nile tilapia ceased to grow starting around the day 70 in all treatments with various N inputs, indicating that the pond

Table 2. Mean values of water quality parameters measured at the initial, midway, and final week in ponds fertilized with different N rates (0, 10, 20, and 30 kg ha<sup>-1</sup> wk<sup>-1</sup>) in the 91-day experiment. Mean values with different superscript letters in the same row are significantly different (P < 0.05).

Parameter		Treat	ment	
	0	10	20	30
DO AT DAWNI (mg $l^{-1}$ )				
Initial	$2.53 \pm 0.29^{a}$	$2.05\pm 0.26^{a}$	$2.60 \pm 0.16^{a}$	$272 \pm 0.21^{a}$
Midway	$3.55 \pm 0.19^{a}$	$2.08 \pm 0.44^{a}$	$2.73 \pm 0.34^{a}$	$1.57 \pm 0.46^{a}$
Final	$4.48 \pm 0.09^{a}$	$3.07 \pm 0.24^{a}$	$4.12 \pm 0.48^{\rm a}$	$4.42 \pm 0.72^{a}$
рН				
Initial	7.1–9.6	7.1-10.3	7.2-9.9	7.0-10.3
Midway	6.0-9.1	6.2-9.0	6.1–9.5	6.0-9.8
Final	6.6–9.9	6.2–10.0	6.5–10.3	6.6–10.2
TOTAL NITROGEN (mg l <sup>-1</sup> )				
Initial	$1.01 \pm 0.05^{a}$	$1.37{\pm}~0.18^{ab}$	$1.70\pm0.04^{bc}$	$2.19 \pm 0.29^{\circ}$
Midway	$1.95\pm0.32^a$	$3.40\pm0.53^{ab}$	$3.90\pm0.36^{bc}$	$5.30 \pm 0.59^{\circ}$
Final	$2.01\pm0.22^a$	$4.64{\pm}~1.40^{b}$	$6.75\pm0.16^{bc}$	$8.98 {\pm}~ 0.54^{\texttt{C}}$
TAN (mg l <sup>-1</sup> )				
Initial	$0.42 \pm 0.37^{a}$	$0.49 {\pm}~0.49^{a}$	$0.91\pm0.24^a$	$0.75 {\pm}~0.75^a$
Midway	$0.13 \pm 0.13^{a}$	$0.26 {\pm}~0.02^a$	$0.74\pm0.22^a$	$0.71 {\pm}~ 0.71^a$
Final	$0.36 \pm 0.15^{a}$	$0.65{\pm}~0.36^{ab}$	$2.20\pm0.23^b$	$2.37 {\pm}~0.55^{b}$
Total Phosphorus (mg $l^{-1}$ )				
Initial	$0.22 \pm 0.01^{a}$	$0.31{\pm}~0.04^a$	$0.24\pm0.02^a$	$0.31{\pm}~0.04^a$
Midway	$0.21 \pm 0.02^{a}$	$0.37 {\pm}~0.07^a$	$0.35\pm0.07^a$	$0.44 \pm 0.10^{a}$
Final	$0.34 \pm 0.03^{a}$	$0.55 {\pm}~0.15^a$	$0.45\pm0.05^a$	$0.75 \pm 0.10^{a}$
TOTAL ALKALINITY (mg $l^{-1}$ as CaCO <sub>3</sub> )				
Initial	$54\pm5.3^{a}$	$51\pm3.3^{a}$	$52\pm5.0^a$	$53 \pm 4.1^{a}$
Midway	$53 \pm 1.8^{a}$	$59\pm7.3^{a}$	$55\pm9.6^a$	$54\pm3.1^{a}$
Final	$82\pm5.0^{\mathrm{a}}$	$72\pm 8.1^{a}$	$74\pm7.0^{a}$	$97 \pm 18.4^{a}$
CHLOROPHYLL A (mg m <sup>-3</sup> )				
Initial	$30 \pm 4.6^a$	$54\pm11.0^{a}$	$34\pm11.5^a$	$56\pm10.0^a$
Midway	$16 \pm 2.6^{a}$	$49 \pm 11.2^{a}$	$52\pm18.0^{a}$	$108{\pm}~39.4^{a}$
Final	$19\pm3.8^{a}$	$51 \pm 14.9^{b}$	$61\pm9.0^{b}$	$158 \pm 54.4^{c}$



Figure 5. Change in alkalinity concentration in the treatment with 0, 10, 20, and 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> over the 91-day experiment.

carrying capacity was near 2,000 kg ha<sup>-1</sup> in the cool season, compared with 2,000 to 2,500 kg ha<sup>-1</sup> in the warm season. With chemical fertilization alone, maximum fish production is about 15 kg ha<sup>-1</sup> d<sup>-1</sup> (Boyd, 1990), which is similar to the highest net fish yield (15.3 kg ha<sup>-1</sup> d<sup>-1</sup>) achieved in this 91-day experiment. However, the net fish yield would reach 19.9 kg ha<sup>-1</sup> d<sup>-1</sup> and much higher profit could be produced if tilapia had been harvested before growth ceased around the day 70, suggesting the importance of timing for fish harvest in optimization of pond production systems.

The optimal N and P input rates determined in PD/A CRSP experiments are much higher than those used in most previous pond experiments, ranging from 1.0 to 6.5 kg N and 1.0 to 2.3 kg P ha<sup>-1</sup> wk<sup>-1</sup> (Swingle, 1947; Mortimer, 1954; Hickling, 1962; Hepher, 1962a, b; Boyd, 1976, 1990; Boyd and Sowles, 1978; Murad and Boyd, 1987). The annual fish yields in those studies were generally below 1,000 kg ha<sup>-1</sup>. In addition to higher fertilizer inputs, the fish stocking rates were also much higher in most PD/A CRSP experiments (10,000 to 20,000

Table 3. Partial budget analysis for Nile tilapia cultured in ponds fertilized with different N rates (0, 10, 20, and 30 kg ha<sup>-1</sup> wk<sup>-1</sup>) in the 91-day experiment (budget items in US\$ pond<sup>-1</sup>).

Item Income (Selling Fish) Added Income (A) Cost for Urea Added Cost from Urea (B1) Cost for NaHCO	Treatment					
	0	10	20	30		
Income (Selling Fish) Added Income (A) Cost for Urea Added Cost from Urea (B1) Cost for NaHCO <sub>3</sub> Added Cost from NaHCO <sub>3</sub> (B2) Ratio of Added Income to Added Cost Profit (A - B1 - B2)	24.60 0 16.35 	54.45 29.85 1.30 1.30 17.20 0.85 13.9 27.70	52.65 28.05 2.60 2.60 16.28 -0.07 11.1 25.52	58.20 33.60 3.90 15.20 -1.15 12.2 30.85		

Table 4. A full-cost enterprise budget for Nile tilapia cultured in ponds fertilized with 30 kg N ha<sup>-1</sup> wk<sup>-1</sup> in the 91-day experiment.

Item	Unit	Price (US\$)	<b>Quantity</b> (kg pond <sup>-1</sup> )	Value (\$ pond <sup>-1</sup> )
GROSS REVENUE (A) Harvested Tilapia	ko	1.50	38.8	58 20
Cost	10	1.00	00.0	00.20
Variable Cost				
Fingerlings Urea	kg kg	$1.50 \\ 0.20$	20.0 19.6	30.00 3.90
TSP	kg	0.33	12.0	3.90
$NaHCO_3$ Interest on Operating Capital	kg vr	$0.78 \\ 12\%$	0.25	15.20
Total Variable Cost	5	7 -		54.60
Pond Depreciation and Equipment	ha*vr	133.75	0.02*0.25	0.68
Interest on Fixed Capital	yr	12%	0.25	0.80
Total Fixed Cost				1.48
Total Cost (B)				56.08
NET RETURNS (A - B)				2.13
BREAK-EVEN PRICE	kg	1.45		

fish ha<sup>-1</sup>) with extrapolated annual fish yields of 3,000 to 5,000 kg ha<sup>-1</sup> (Lin et al., 1997). The highest extrapolated net yield obtained in the present study is around 3,750 kg ha<sup>-1</sup> yr<sup>-1</sup> and could have reached 5,000 kg ha<sup>-1</sup> yr<sup>-1</sup>, if fish had been harvested around the day 70. These yields are lower than those (5,500 and 7,000 kg ha<sup>-1</sup> yr<sup>-1</sup>, respectively) obtained in the warm season.

With the development of Nile tilapia cage culture in rivers in northeast Thailand, there are strong demands for large fingerlings (30 to 50 g) to stock cages. Small-scale farmers nurse Nile tilapia fry to such sizes and sell them at around \$1.50 kg<sup>-1</sup>, which is much higher than the price of marketable adult Nile tilapia. Farmers commonly nurse fry in fertilized ponds supplemented with artificial feed. In the present study, however, Nile tilapia were nursed at a very high density in ponds with fertilizer only, resulting in high yields. This study implies that moving fish from a high to low density when fish growth ceases or pond carrying capacity is reached could be a good strategy. Results of the present study may provide small-scale farmers with a technically and economically effective strategy to optimize resource utilization and maximize profits.

## **ANTICIPATED BENEFITS**

This is the first in a series of experiments to determine optimal rates of nitrogen, phosphorus, and carbon additions to ponds for fish production. Results of these trials will determine N, P, and C application rates to obtain fish yields with the greatest profit. Development of a full-cost enterprise budget for the fertilization rate that results in the greatest profitability will assist host-country and international economists and planners in their evaluation of fish culture systems. Identification of optimal nutrient application rates would reduce the environmental impact of pond effluents.

## ACKNOWLEDGMENTS

The authors wish to acknowledge the Asian Institute of Technology, Thailand, for providing the research, field, and laboratory facilities. Mr. Manoj Y. and Mr. Supat P. are greatly appreciated for their field and laboratory assistance.

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# PD/A CRSP Seventeenth Annual Technical Report

## TIMING OF THE ONSET OF SUPPLEMENTAL FEEDING OF TILAPIA IN PONDS

Ninth Work Plan, Feeds and Fertilizers Research 4 (9FFR4) Progress Report

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## Abstract

Supplemental feeding has been a common practice among tilapia farmers in the Philippines. However, the timing of the start of supplemental feeding is variable. This study aims to demonstrate efficient supplemental feeding strategies for tilapia production in fertilized ponds through on-farm trials. Seven farmer cooperators were enlisted for this experiment. Two ponds of nearly the same size were stocked with all-male Nile tilapia fingerlings of the Genetically Improved Farmed Tilapia (GIFT) strain at a stocking rate of 4 fish m<sup>-2</sup>. The ponds were fertilized weekly with urea and 16-20-0 fertilizer at a rate of 4 kg N ha<sup>-1</sup> d<sup>-1</sup> and a N:P ratio by weight of 5:1. Supplemental feeding commenced 45 days after stocking in one pond while in the other pond, supplemental feeding started 75 days post-stocking. Preliminary results show a trend of better growth of fish in ponds fed early during the culture period.

## INTRODUCTION

Earlier PD/A CRSP studies showed that initiation of feeding *Oreochromis niloticus* after 80 days in the pond produced the same yield as initiation of feeding at 38 days. Later feeding also increased growth rates and yields to the target levels, but with some delay over earlier feeding (Diana et al., 1996). This means that fish do not need to be fed immediately after stocking, but can be supported by the pond ecosystem. The objective of this study is to demonstrate efficient supplemental feeding strategies for tilapia production in fertilized ponds through on-farm trials.

## METHODS AND MATERIALS

Seven farmers from the province of Nueva Ecija were enlisted for this trial; pond systems of farmers ranged from extensive to semi-intensive. Two ponds from each farm were assigned each of the two treatments—supplemental feeding beginning at 45 and 75 days after stocking.

The pond size ranged from 416 to  $3,500 \text{ m}^2$  with water depth maintained at 1 m. The ponds were stocked with sex-reversed Nile tilapia (*Oreochromis niloticus*) of GIFT strain. The prevalent size commercially available was size 24 (less than 0.2 g in average weight). This size was used at a stocking rate of 4 fish m<sup>-2</sup>. All ponds were fertilized weekly with urea and 16-20-0 fertilizer at a rate of 4 kg N ha<sup>-1</sup> d<sup>-1</sup> and a N:P ratio by weight of 5:1.

All ponds are analyzed monthly for water quality. Water samples are obtained for the analysis of dissolved oxygen, pH, total alkalinity, total ammonia, and soluble reactive phosphorus. Analyses are done at the Freshwater Aquaculture Center (FAC) laboratory according to standard methods (Boyd, 1979; APHA, 1980).

The fish are fed with prepared feeds consisting of 50% rice bran and 50% fish meal at 5% fish body weight per day (BWD) during the first 15 days of feeding and 4% BWD thereafter up to 30 days. For the remaining period prior to harvest, the fish will be fed at the rate of 3% BWD. Random samples of 50 fish are obtained in each pond every two weeks to measure average weights of the fish.

The ponds will be harvested by seining or complete draining at 150 days in pond. Total number of fish will be counted and bulk weighed. A sample of 100 fish per pond will be individually measured for length and weight. Total yield, feed conversion rates, and survival rates will be obtained at the end of this study.

## RESULTS

The mean weights of tilapia taken at different sampling days are shown in Table 1. As can be gleaned from the table, in four out of the seven farms, the preliminary results showed that there was better growth of fish in ponds which received supplemental feeds at 45 days after stocking. The ranges of water quality data taken on a monthly interval are summarized in Table 2.

## ANTICIPATED BENEFITS

Feeding practices have been variable among tilapia farmers in the Philippines. This study will provide technical guidance to farmers on efficient feeding practices that will optimize tilapia production.

Mean weights (g) of tilapia fed at 45 and 75 days after stocking in ponds, Philippines. Table 1.

Farm	TreatmentMean WeightPond(g)								
		Day 0	Day 30	Day 45	Day 60	Day 75	Day 90	Day 105	Day 120
1	А	0.06	9.17	13.44	21.05	33.47	44.15	61.78	68.62
2	B A	0.06 0.05	9.29 10.52	18.84	33.20 35.60	44.18 67.52	51.01 86.35	54.79 104.46	60.63
3	B A	0.05 0.07	15.91 17.13	21.44	46.76 37.16	73.97 56.97	107.23 75.25	146.30 113.78	
4	B	0.06	15.50 16.24	24.00	50.68	52.39 97.25	77.88	106.56	
4	B	0.15	10.24	54.00	53.47	61.49	90.65		
5	A B	0.15 0.15	16.46 14.15	28.86	52.20 50.69	72.85 70.13	96.37 90.62		
6	A B	0.19 0.19	18.72 11.55	39.80	59.52 36.90	94.77 48.08			
7	A B	0.12 0.12	14.14 11.19	24.85	51.49 56.32	10.00			

Pond A – early feeding Pond B – delayed feeding Note:

Table 2.	Ranges of water c	uality	parameters i	n feeding	g trial	ponds in	Philippines.
	()			(	,		

Farm	Treatment Pond				Parameter			
	_	Secchi Disk Visibility (cm)	D.O. (mg l <sup>-1</sup> )	Temperature (°C)	рН	Alkalinity (mg l <sup>-1</sup> )	Ammonia (mg l <sup>-1</sup> )	Phosphorus (mg l <sup>-1</sup> )
1	А	23–7	4–10	28–31	7.8–8.9	36–95	0.080-0.172	0.185–0.805
	В	28-8	3–9	28-31	7.6–9.9	42-87	0.030-0.33	0.295-0.560
2	А	58–9	9–15	30-35	8.6-9.1	112-156	0.117-0.359	0.060-0.850
	В	60-14	11–16	31-38	8.4–9.6	92-220	0.102-0.300	0.060-0.615
3	А	30-15	11–12	30-31	8.4-10.3	49-81	0.022-0.261	0.075-0.125
	В	54-15	8-13	29-31	8.0-10.3	56-93	0.048-0.320	0.065 - 0.445
4	А	32-12	9–12	30-32	7.9–9.7	150-162	0.102-0.348	0.265-0.365
	В	22-10	9–11	29-31	8.0 - 10.4	115-140	0.030-0.374	0.025-0.295
5	А	65–9	6–16	29-32	8.0-9.9	127-175	0.048-0.248	0.140-0.675
	В	65-11	5-14	29-32	8.0-9.5	119-230	0.026-0.352	0.105-0.345
6	А	50-8	14-20	31-32	7.7–9.6	112-210	0.113-0.280	0.175-0.765
	В	46-12	15-20	31-33	7.7–9.5	86-207	0.048-0.235	0.085-0.630
7	А	40-8	4-15	29-33	8.4–9.7	239-302	0.063-0.215	0.040-0.510
	В	28–9	5–12	29–33	8.4–9.7	271-345	0.093-0.102	0.095-0.565

Note:

Pond A – early feeding Pond B – delayed feeding

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Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics / Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]



# PD/A CRSP Seventeenth Annual Technical Report

## METHODS FOR ANDROGENESIS TECHNIQUES APPLICABLE TO TILAPIA

Eighth Work Plan, Reproductive Control Research 1B (8RCR1B) Final Report

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## Abstract

Control of reproduction is vital to aquaculture and includes artificial propagation as well as management of unwanted recruitment. Developments in manipulation of the reproductive system provide options to enhance production. Nile tilapia, *Oreochromis niloticus*, spawning was managed by photoperiod and temperature manipulation. A controlled light cycle of 20L:4D and water temperature of  $26 \pm 2^{\circ}$ C directed spawning to a predictable time frame. A developmental rate ( $\tau_0$ ) relationship was described and applied to chromosome manipulation. Blond Nile tilapia are homozygous recessive for a color mutation that was used as a phenotypic marker in the development of protocol for androgenetic induction. Androgenotes were produced by neutralizing the female genome of normal color Nile tilapia (600 J m<sup>-2</sup> UV dose), activating the egg with sperm from blond males, and diploidizing with cold shock ( $11 \pm 0.5^{\circ}$ C for 60 min) applied at various times after incubation at  $28 \pm 0.2^{\circ}$ C. Shock applied at 69 min post-activation produced greater numbers of androgenotes than shocks applied at 59 or 79 min post-activation. Optimization for shock type and associated parameters will be required for production of practical numbers of androgenotes for YY-male breeding programs.

## INTRODUCTION

Management of fish reproduction is important to aquaculture; it may involve production of seedstock through artificial propagation or, conversely, through development of limits to recruitment. Enhancement of spawning can be by habitat manipulation or through direct hormonal intervention, while reproductive control also can be direct or based on more complicated modifications (Shelton, 1989; Dunham, 1990). The basic techniques and capacity for control vary between species. Tilapias do not respond well to hormonal induction for spawning control, but unwanted recruitment can be effectively managed by monosexing through direct hormonal treatment. While this direct control is practical, growing concern with the use of steroids in food suggests the desirability of other options. Chromosome manipulation, which includes ploidy alteration or euploidy induction with single-parent genome contribution, can provide alternative control mechanisms. Ploidy manipulation includes the induction of triploids through polar body retention or tetraploidy through interference with first zygotic mitosis. Euploid alterations include gynogenetic (gynogenote) diploidization of the maternal genome through polar body retention (meiogynote) or mitotic interference (mitogynote). Diploidization of the paternal genome (androgenote), or androgenesis, is accomplished by interference with first mitosis for eggs that have been genome neutralized before activation. This report describes the initial development of protocol for the production of androgenetic progeny of Nile tilapia, Oreochromis niloticus.

Chromosome manipulation involves one or two basic treatments after obtaining fresh gametes (Thorgaard and Allen, 1986). For androgenesis, the first treatment is the deactivation of the female genome. Ultraviolet irradiation is preferred for simplicity and safety, but also because it dimerizes the DNA rather than fragmenting it. Egg activation with untreated spermatozoa then requires diploidization of the haploid zygote

by some form of shock to interrupt the first mitotic division. Shock is most often physical, e.g., thermal (cold or hot) or pressure. Thermal treatment is usually preferred because of the ease of application and equipment simplicity. In order to prevent chromosome segregation, the shock must be timed to coincide with a cytological event, such as disruption of the spindle fibers during metaphase to prevent karyokinesis, or interference with the cell duplication during cytokinesis. Thus, shock type and intensity, duration, and time of application must be optimally combined into a protocol for maximum yield of diploid progeny. Further, because the rate of development is inversely temperature dependent, either the preshock incubation temperature must be standardized or the shock time must be calibrated to account for the temperature effect. Absolute shock time (minutes post-activation) can be transformed with reference to an index of development rate or mitotic interval ( $\tau_0$ ), also in minutes (Dettlaff and Dettlaff, 1961). Shock time  $(\tau_s)$  can be related to tau  $(\tau_s/\tau_0)$  to report shock protocol in a dimensionless index which is temperature compensated (Dettlaff, 1986). A tau curve for the Nile tilapia was described during an earlier segment of this study (Shelton, 1999). Finally, these requisite gametic treatments (UV and shock) are near lethal, increasing mortality, and the diploidization of the genome increases homozygosity, thereby reducing fitness and survival.

## METHODS AND MATERIALS

## Broodstock

The University of Oklahoma stock of *O. niloticus* used in the earlier study segments originated in Ivory Coast and was obtained from Auburn University in 1982. The blond stock was derived from a population in Lake Manzala, Egypt (Scott et al., 1987; McAndrew et al., 1988), and was obtained from Swansea University, UK, in 1987. During various studies the Ivory Coast population became hybridized to an unknown level with the blond stock. Since this is the phenotypic marker that was to be used in our study, a different stock was obtained for the final series of experiments. Nile tilapia from a Ghana source were obtained from Auburn University in 1998, but the fish in this founder stock were small. They were transferred to ponds for the summer to mature, then overwintered. Fish from this population were first used in experiments beginning in January 1999. Each stock (Ghana, blond, and Ghana × blond) was progeny tested for color phenotype at developmental stages from hatching to swim-up.

## Spawning

Spawning of Nile tilapia was managed by photoperiod manipulation, as results of earlier efforts to induce ovulation with gonadotropic injections (Gissis et al., 1988) were inconsistent. Several females (four to six) were stocked with one male in four large aquaria (550 l) or paired in 200-l aquaria. Water was aerated and was circulated at the rate of one turnover per day, and temperature was maintained at  $26 \pm 2^{\circ}$ C. The light cycle was controlled by adjusting a timer to turn on the overhead lighting at 0100 hours; the natural sunset regulated the end of the photoperiod for a total of about 20L:4D. Tilapia spawn about 8 to 10 h after the beginning of the light cycle (Myers and Hershberger, 1991). Ovulation and spawning readiness was judged by courtship behavior, coloration, and papilla erection (Rothbard and Pruginin, 1975; Rothbard, 1979). Females were stripped after initiation of spawning or as indicated by other characteristics that signal ovulation.

## Fertilization

For progeny tests of body color and for developmental rate experiments, eggs were collected in a clean container, milt was expressed over the eggs, and water was added directly to initiate activation (Rana, 1988). Fertilized eggs were placed in incubation chambers submerged in a water bath within 2 to 3 min, temperature was regulated closely and 20 to 30 eggs were examined under magnification at intervals of 5 min to record cleavage rate. Developmental rate ( $\tau_0$ ) is defined by the duration of one mitotic cycle during early synchronous cleavage (Dettlaff and Dettlaff, 1961). Tau curves have been used to facilitate chromosome manipulation studies in various fishes (Shelton and Rothbard, 1993; Shelton et al., 1997). The tau curve of Nile tilapia was reported in an earlier progress report (Shelton, 1999); it was described from mean time intervals between the initiation of the first and third mitoses in 5 to 10% of the eggs over a range of temperatures (20 to 30°C). Subsequently, eggs were incubated at  $28 \pm 0.2$ °C in upwelling units of 1-l capacity with sufficient flow to gently tumble the developing embryos. Observations on pigment development were recorded at prehatching, posthatching, and swim-up stages for Ghana, blond, and Ghana x blond. Fish occasionally spawned outside of periods of observation, but fertilized eggs were collected from the brooding females, moved to incubators, and pigmentation development observed.

## **Chromosome Treatment**

Freshly ovulated eggs were stripped from normally pigmented Ghana females (not carrying the blond gene) and allocated into four subsamples. Two to four hundred eggs (about 2 ml) were placed in each of four 10-cm petri dishes with enough 28°C water (10 ml) to provide slight buoyancy. Three subsamples were placed in a UV-crosslinker (FisherBiotech, FB-UVXL- 1000) and exposed to 600 J m<sup>-2</sup> UV; this dosage was determined in an earlier study as sufficient to dimerize the female genome (Shelton, 1999). The UV-treated eggs were activated with freshly collected spermatozoa from a blond male, then maintained at 28°C on a rotating shaker table; the untreated eggs (control) were fertilized with milt from the same male. After 30 min all eggs were transferred from the petri dishes to screened, individual incubator chambers which were submerged in a 28°C water bath. Treated eggs were subsequently plunged into water in a thermal bath at  $11 \pm 0.5$ °C for 60 min; one subsample was cold-shocked at 59 min post-activation, the second at 69 min, and the third at 79 min. The first shock time coincided with the initial mitotic metaphase, the second just prior to cleavage furrow formation (cytokinesis), and the third immediately after cell division. After shock, all eggs were returned to the water bath and then they were transferred, including controls, into individual flow-through incubators. Development was monitored through hatching and swim-up.

#### **RESULTS AND DISCUSSION**

## Spawning and Progeny Testing

Photoperiod manipulation permitted spawning activity to be forced to mid-day. Females ovulated/spawned between about 9 and 14 h (mean = 11.8 h) after the light-on cycle. Ovulation based on the light cycle was a reasonable means of predicting stripping time. Hormonal induction of ovulation would seem to be a logical extension, but cichlids have responded poorly to gonadotropic therapy (Rana, 1988). During the summer of 1998 while the Ghana broodstock were in ponds, progeny testing of blond stocks continued, then beginning in January 1999, the Ghana stock was incorporated in the spawning efforts. No spawning occurred among the Ghana stock until April, then progeny testing was initiated. Five spawns were sufficient to establish the developmental pattern of pigment for this race. The appearance of melanophores in larval tilapia from normally pigmented broodstock is clearly differentiated from that for blond progeny.

The sequence of melanophore development in four areas is useful for identifying the parental source of progeny; these areas are the yolk, eye, brain surface, and lateral body along the notochord. Progeny of normally pigmented stocks (Ghana) developed melanophores first on the yolk surface-pigment cells could be seen 1 to 2 d prior to hatching. Eye color appeared about 12 h prior to hatch, but was initially diffuse, then developed as a gradual blending of melanin and gold pigmentation. At hatching, scattered chromatin-filled melanophores were present on the dorsal surface of the brain and laterally on the body adjacent to the notochord and near the yolk sac, but subsequently progressed caudad, gradually increased in abundance. In contrast, progeny of blond stocks totally lacked pigmentation in these areas until posthatching, and then coloration was limited to a gold cast in the eyes. Body color did not develop during the larval stages and only nonpigment-bearing melanophores could be seen on the yolk and brain surfaces. These appeared as shadowy cellular outlines that contained no melanin. Finally, hybrids between pigmented females (Ghana) and blond males developed pigmentation in areas and in the sequence described for the Ghana stock. Thus, progeny that have dark coloration in the first several days post-hatching (until the mouth becomes functional) possess genomic contribution from one or both pigmented parents, but not exclusively from blond parent(s); the latter are nonpigmented throughout larval states. Androgenotes derived from blond male parents can therefore be easily identified from prehatching through swim-up stages.

## **Chromosome Manipulation**

Fertilization rate was variable, with no development in controls (artificial fertilization) of 6 of 17 batches of ova stripped from Ghana females. In the 11 other strip-spawned groups of the control that were artificially fertilized, hatch rate ranged from only 4.0 to 11.3% (mean 10.9%) (Table 1). Motility of spermatozoa was evaluated microscopically and milt was used only if more than 90% of cells were viable. Poor quality ova are most often the primary cause of developmental failures. Initiation of egg resorption is common among tilapias and, although it does not prevent ovulation of the bad eggs, fertilization and hatching are poor (Peters, 1983). Physiological characteristics of tilapia gametes provide advantages in chromosome manipulation. Eggs retain high fertility for 3 to 6 h post-immersion in water (Myers et al., 1995), and sperm remain motile in water for several hours in contrast to most fishes (Yeheskel and Avtalion, 1986). Some of the low rate of development may also be attributable to the type of incubation. Hatch rate in the upwelling type of incubator is not as good as in a down-flow system (Rana, 1986); our incubators incorporate this preferred down-flow design.

Hatch rates for UV-treated and cold-shocked ova were significantly lower than those of controls, i.e., never more than a fraction of a percent. Relative to the controls, hatch rate ranged from about 3% of the control for the shock that started

at 59 min after activation to about 22% of the control for the shock that started at 69 min (Table 1). Increased mortality resulting from the UV dose is expected due to cell damage. Successful diploidization varied depending on the proximity of shock to the optimal induction "window." Optimization was not the primary objective during the Eighth Work Plan and only three shock times were tested. The late shock protocol for tetraploid induction that was successfully developed by Don and Avtalion (1988a, 1988b) and Don (1989) was used as the basis for induction of androgenesis. The cold shock has a wider time of application (60 min) compared to heat or pressure shock (2 to 5 min) and intuitively should provide a greater likelihood of some successful diploidization. Further, the shock application time used by the Israeli scientists is based on cytological correlation that relates well to time factors as described by tau adjustments (Saat, 1993; Shirak, 1996; Shirak et al., 1998). This is in sharp contrast to the empirical approach of Hussain et al. (1993), Mair (1993), and Myers et al. (1995), where shock time application had little relationship to cytological events.

Hatch rates of control groups in 11 trials indicated reasonably good quality ova (Table 1). Induction of androgenetic progeny (UV-treated Ghana ova, activated with sperm from blond males and diploidized by cold shock) was tested by cold-shocking at three post-activation times (59, 69, and 79 min at 28°C) which correspond to 0.75, 0.87, and 1.0 T (T = time of first mitosis) or 1.8, 2.1, and 2.5  $\tau_0$ , respectively. The former (0.75 T) is the approximate time for first mitotic metaphase (Saat, 1993; Rubinshtein et al., 1997), while the second (0.87 T) is just prior to first mitotic cytokinesis and the optimal induc-

Table 1. Induction of and rogenotes of Nile tilapia (Ghana females  $\times$  blond males) using cold shock on eggs artificially fertilized (11 ± 0.5°C for 60 min to eggs UV-treated with 600 J m<sup>-2</sup>).

Trial	Eggs (#)	Control (#)		Treatment (#)	Androgenote (#)	
			Hatch (Survia	val to Swim-up)		Survival to Swim-up
			59 min	69 min	79 min	
1 2	510 Inviable	25 (12)	1 (0)	2 (1)	0	1
3	400	10 (3)	0	1 (0)	0	0
4	650	35 (10)	1 (0)	4 (1)	2(0)	1
5	Spawn					
6	Inviable					
7	Inviable					
8	Inviable					
9	Spawn					
10	Inviable					
11	Inviable	((0)	<b>a</b> (a)		. (-)	_
12	800	55 (40)	2(0)	10 (4)	4(1)	5
13	Spawn		4 (1)	01(11)	10(2)	15
14	/50	85 (62) 42 (21)	4(1)	21 (11)	10(3)	15
15	625 Caracian	42 (21)	1 (0)	13 (5)	7(2)	1
10	Spawn					
17	120	20(11)	0	5 (2)	4(1)	3
10	420 850	20(11) 75(43)	$\frac{1}{2}$ (0)	$\frac{3(2)}{21(10)}$	$\frac{4(1)}{7(2)}$	12
20	710	63(24)	$\frac{2}{4}(0)$	16(5)	$\frac{7}{2}(0)$	5
20	410	33(21)	$\frac{1}{1}(0)$	$\frac{10}{8}(2)$	2(0) 2(1)	3
22	530	25(10)	0	2(0)	0	Ő
Sum	6.655	257 = 3.9%	16 = 3.4%	103 = 22%	38 = 8.1%	0
Culli	0,000	_0, 0.070	of control	of control	of control	

tion time for late cold-shock in tilapia as determined by Shirak et al. (1998). The highest hatch rate of androgenotes (nonpigmented larvae) was at the 69-min post-activation shock time; the other two shock times had lower yields. Induction success for androgenotes to hatching does not ensure survival to later stages. From a total of 157 androgenotes that hatched, only 57 survived to swim-up or yolk absorption (Table 1). Additional mortality during the juvenile stages further reduces the number that will be available for progeny testing to verify the production of YY males. As of this report, only five of these androgenotes have survived to the end of July.

Optimization of shock protocol is affected by the asynchrony in cell division; at 28°C, the time from initiation of first cleavage to end of the last encompasses about 20 min for the Nile tilapia (Shirak et al., 1998). Thus, the shock application and duration will affect only a small percentage of the cells at an optimal induction time. Further, even adjusting for the incubation temperature effect, optimization of treatment time for shock is complicated by apparent differences in effectiveness of various types of shock. Palti et al. (1997) clearly demonstrated a difference in optimal shock time for gynogenetic induction with heat and pressure shock for rainbow trout, Oncorhynchus mykiss. Similarly, Hussain (1995) demonstrated differential optima between cold, heat, and pressure shock in Nile tilapia. Shirak et al. (1998) suggest that cold shock interferes with cell division, while the mechanism for heat shock has been considered to be disruption of the spindle fibers during karyokinesis (Mair, 1993).

The most effective type of shock must be determined and must be optimized based on the induction mechanism. Thermal shocks will be used for practical reasons and because effectiveness is as good as or better than pressure shock. Don and Avtalion (1988a) used cold shock for tetraploid induction based on comparative effectiveness for triploid induction (Don and Avtalion, 1988b). However, the timing for shock reported by Myers et al. (1995) at 27 min post-activation (28°C) markedly contradicts the optimal shock time of 92 min post-activation (26°C) reported by Don and Avtalion (1988a) and Don (1989), even when adjusted for the different incubation temperatures. Therefore, these reported induction times and shock types will be examined for optimization in the context of developmental rates ( $\tau_0$ ).

### **ANTICIPATED BENEFITS**

Control of unwanted reproduction in tilapia culture has been one of the primary considerations affecting successful food production. Various approaches have been developed and utilized. Most recently, efforts to develop systems that will allow breeding programs to produce monosex cultures have been sought. Androgenesis offers a mechanism that in theory can directly produce YY-males that can be used as broodstock to breed all-male progeny. Demonstration of the induction technology, with its multiple pitfalls, is the first step. Optimization of treatments must be the next achievement so as to increase the numbers of viable androgenotes for practical survival expectations to test the theoretical basis for sex determination and demonstrate that breeding for a monosex can be accomplished.

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# PD/A CRSP Seventeenth Annual Technical Report

## DETECTION OF MT IN POND WATER AFTER TREATMENT WITH MT FOOD

Eighth Work Plan, Reproduction Control Research 3B (8RCR3B) Final Report

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## Abstract

The objective of the study was to determine if methyltestosterone (MT) can be detected in the treatment environment and, if so, for how long after treatment. Oreochromis niloticus fry with a mean initial length of 9.5 mm were stocked into adjacent cages in an earthen pond at Auburn University, Alabama, at 2,000 fry cage<sup>-1</sup> and fed for 28 days a feed containing 60 mg MT kg<sup>-1</sup> or a non-treated feed. At the end of the controlled feeding period both sets of fish were harvested, and growth and survival were determined. Fish were returned to their respective hapas and fed a non-hormone treated feed for additional growth until a mean total length of approximately 5 cm was reached. A sample of 100 fish from each hapa was preserved in 10% formalin and the sex was determined. Water and soil samples from the treatment pond were taken prior to, during, and after the hormone administration period. Water samples (collected approximately 10 cm below the surface) were taken weekly from within the cage receiving hormone-treated feed and the cage receiving non-treated feed. At the same time intervals water samples were taken at 2, 5, and 10 m from the cage receiving hormone-treated feed. Soil samples were taken of the upper 5 cm of soil at the same locations at which water samples were taken; samples were collected from under the cages and from the pond bottom at the distances specified above. Soil and water samples were analyzed at Oregon State University. There was no evidence that MT altered the sex ratio of non-target tilapia held in the same pond as and confined near fish receiving MT. The treated population was 91% male, 5% female and 4% intersex. Fish held in an adjacent cage approximately 50 cm away and fed a nonhormone-treated diet had a sex ratio of 48% males and 52% females. Mean MT concentration in the water sampled within MTtreated or non-treated cages did not differ (P = 0.14). Pretreatment MT concentration in the water column was  $8.0 \pm 5.7$  pg g<sup>-1</sup>, and values within the treatment cage were similar except for one sample during the treatment period. The radioimmunoassay when used with soil cross-reacted with other materials in addition to MT. Pretreatment soil samples from the pond, which had no previous history of MT administration, had a concentration of  $875 \pm 147$  pg g<sup>-1</sup>. The highest concentration of MT indicated (1,417 pg g<sup>-1</sup>) was from a soil sample beneath the cage receiving the non-treated feed.

## INTRODUCTION

The use of all-male populations of tilapia for culture offers several important advantages, including enhanced growth (males grow faster and larger than females) and prevention of unwanted reproduction, which diverts energy away from somatic growth. Treatment with  $17\alpha$ -methyltestosterone (MT)-impregnated food has been shown to be an effective means of producing all-male tilapia populations. However, there exists the possibility of steroid loss into the treatment environment from uneaten or unmetabolized food and a risk of unintended exposure of fish or other non-target organisms.

Budworth and Senger (1993) reported on fish-to-fish testosterone transfer in a two-tank recirculating-water system. MTexposed non-target fish had significantly higher plasma testosterone concentrations than non-exposed control fish during weeks 1 to 3, and no significant difference was detected between MT-exposed and control fish during week 4. In another recirculating system study, common carp (*Cyprinus*  *carpio*) in one tank were fed an androgen-treated diet, while fish in another tank within the same system were fed an androgen-free diet. Both sets of fish developed skewed sex ratios (Gomelsky et al., 1994). It is clear that unmetabolized MT and metabolites of MT, both excreted by the target fish, can accumulate in the water of recirculating systems. The degree of accumulation appears to depend on the frequency and dose of MT administered to the target fish.

The fate of MT in other settings is less clear. In an outdoor tank setting, Phelps et al. (1992) used small hapas placed approximately 30 cm apart in a static water 20-m<sup>2</sup> tank to hold fish given an MT- or fluoxymesterone-treated feed or a non-treated feed. The treatments were randomly assigned within the tank. There was no evidence that hormone leaching affected the sex ratio of non-treated fish. Soto (1992) used a similar experimental setup with eighteen 0.12-m<sup>2</sup> hapas distributed in a 20-m<sup>2</sup> tank to evaluate the androgenic potential of mestanolone to sex-reverse *O. niloticus*. Non-hormone treated fish did not have a skewed sex ratio even though they were surrounded by

sets of fish that had been given the hormone-treated feed and were successfully sex-reversed. Abucay et al. (1997) found that reusing static water aquaria that had been used to hold tilapia fry that were given an MT-treated feed for 25 days could alter sex ratios. When a second group of fish were stocked into such aquaria and given a non-hormone-treated feed, the sex ratio was skewed. They also found that when an all-female set of fry were stocked into a cage in an aquarium and an MT-treated feed was added to the bottom of the aquarium where the fish had no access to it, the sex ratio became skewed to males.

In a laboratory setting, Fitzpatrick et al. (1999) detected MT in the water and soil of containers in which tilapia were fed an MT-treated feed during a treatment period of four weeks. Concentrations of MT in the water had decreased to within background levels by one week after the end of treatment, while measurable levels above background in the soil persisted for four weeks post-treatment.

MT is susceptible to breakdown when exposed to light or high temperatures (McEvoy, 1997). Both fungi and bacteria can metabolize exogenous steroids. Many different steroid metabolism reactions, including the metabolism of MT, are possible in bacteria (Schubert et al., 1972; Jankov, 1977), as well as metabolism of steroids to  $CO_2$  and  $H_2O$  (Sandor and Mehdi, 1979). In an outdoor pond setting where fish are held in cages, the combination of light, temperature, and microbial degradation may result in a rapid breakdown of MT. This study evaluated the concentrations of MT in soil and water when tilapia were given a diet containing MT for a 28-day period when cultured in cages in an outdoor earthen pond. The effect on the sex ratio of non-treated tilapia held in an adjacent cage was also examined.

#### METHODS AND MATERIALS

Oreochromis niloticus fry were obtained from a mass spawning of brooders in an earthen pond and graded to obtain fish with a mean initial length of 9.5 mm. Fish were stocked into adjacent cages in a 400-m<sup>2</sup> earthen pond at Auburn University, Alabama, at 2000 fry cage<sup>-1</sup> and fed for 28 days a feed containing 60 mg MT kg<sup>-1</sup> or a non-treated feed. Fish were fed at 25% body weight the first week; thereafter, feeding was reduced by 5% of body weight each week. Fish were sampled weekly during the feeding period, average weights were determined, and feeding rates were adjusted daily based on observed growth rates. At the end of the controlled feeding period both sets of fish were harvested, and growth and survival were determined. Fish were returned to their respective hapas and fed a non-hormone-treated feed for additional growth until a mean total length of approximately 5 cm was reached. A sample from each hapa of 100 fish was preserved in 10% formalin and dissected. Gonads were removed, lightly stained with fast green, squashed to facilitate microscopic examination, and the sex was determined. The entire gonad was examined and those containing both ovarian and testicular tissue were classified as intersex; gonads having only one form of tissue were classified as male or female, corresponding to the type of tissue observed.

Water and soil samples from the treatment pond were taken prior to, during, and after the hormone administration period. Water samples (collected approximately 10 cm below the surface) were taken weekly within the cage receiving hormonetreated feed and the cage receiving the non-treated feed. At the same time intervals, water samples were taken at 2, 5, and 10 m from the cage receiving hormone-treated feed. Soil samples were taken of the upper 5 cm of soil at the same locations at which water samples were taken; samples were collected from under the cages and from the pond bottom at the distances specified above. All soil and water samples were frozen soon after collection and held frozen for shipment.

Soil and water samples were analyzed at Oregon State University following the procedures of Fitzpatrick et al. (1999).

## **RESULTS AND DISCUSSION**

There was no evidence of MT altering the sex ratio of non-target tilapia held in the same pond and confined near fish receiving MT. Treatment of *O. niloticus* fry for 28 days with MT at 60 mg kg<sup>-1</sup> of diet was effective in altering the sex ratio. The resultant population was 91% male, 5% female, and 4% intersex. The control group of fish held in an adjacent cage approximately 50 cm away and given a non-hormone-treated diet had a sex ratio of 48% males and 52% females. The lack of intersex fish in the non-treated fish is further evidence that MT was not released into the environment at a rate to alter sex ratios.

There was little to no evidence of MT escapement from the treatment cage environment. Mean MT concentrations in the water sampled within MT-treated or non-treated cages did not differ (P = 0.14). Pretreatment MT concentration in the water column was  $8.0 \pm 5.7$  pg g<sup>-1</sup>, and values within the treatment cage were similar during the treatment period with the exception of one sample. A concentration of  $161 \pm 28$  pg MT g<sup>-1</sup> was detected in this sample, and it is thought that this was caused by a particle of feed collected in the sample. MT concentrations in the water from the treatment cage for the 20-day post-treatment period averaged  $9.7 \pm 4.1$  pg g<sup>-1</sup>. The mean concentration of MT detected 2 to 10 m from the treatment cage was 9.7 pg g<sup>-1</sup>. Concentrations of MT within the control cage averaged 9.2 pg g<sup>-1</sup> for the study period. Concentration of MT in water or soil did not increase in relation to the increased MT input over time associated with feeding (Figure 1). On the last day of hormone treatment, 43.3 g of feed



Figure 1. Concentration of methyltestosterone (MT) determined from weekly sampling of soil and water collected from below the treatment cage and within the cage, respectively, and cumulative quantity of MT added to the feed during the treatment period.

containing approximately 2.6 mg MT was given, but water from within the treatment cage contained less than 5 pg MT g<sup>-1</sup>.

The radioimmunoassay used for soil samples cross-reacted with other materials in addition to MT. Pretreatment concentrations of soil from the pond, which had no previous history of MT administration, had a concentration of  $875 \pm 147 \text{ pg MT g}^{-1}$ . The highest concentration of MT measured (1,417 pg g<sup>-1</sup>) was from a soil sample collected beneath the cage of fish receiving the non-treated feed. The mean concentration of MT measured from soil samples beneath the cages was greater under the non-treated cage (*P* = 0.007).

The radioimmunoassay may have also cross-reacted with material in the water column since a pretreatment level of 8.0 pg g<sup>-1</sup> was measured, and values within the control cage and up to 10 m from the treatment cage ranged up to 34.9 pg g<sup>-1</sup>. The assay appeared to detect a general background level of approximately 9 pg MT g<sup>-1</sup>. All assays, except for one sample within the treatment cage, gave values less than 40 pg MT g<sup>-1</sup>.

The lack of accumulation of MT in the pond environment is to be expected. Tilapia are aggressive feeders and would consume all MT-treated feed offered under normal circumstances. MT ingested by the fish is metabolized, and MT and metabolites are excreted. If it is assumed that tilapia, like rainbow trout (Cravedi et al., 1993), excrete 14.8% of the administered MT dose as unmetabolized MT, then in this study the daily release of unmetabolized MT may have been as high as 192 mg l<sup>-1</sup> on the last day of feeding, while the level detected in the water was < 5 mg l<sup>-1</sup>. Fitzpatrick et al. (1999) found that MT concentrations in the water column returned to background levels within one week after hormone administration.

Both fungi and bacteria can metabolize exogenous steroids. The biodegradability of MT and its degradation by high light intensity and temperature contribute to the rapid decline of MT in the environment. In a pond setting the breakdown of MT is rapid with little to no effect on non-target fish.

## **ANTICIPATED BENEFITS**

The environmental fate of steroids is a poorly understood area, but it is of concern in settings where non-target animals may be exposed. The use of methyltestosterone is of particular concern because it is a key component of a successful tilapia aquaculture industry. This study has shown that in a pond setting the life of the hormone is short. This study indicated that fish confined close to fish being treated with MT were not affected; concentrations of MT detected in the environment were not significantly above background levels. These data will be very useful for regulators assessing the impacts of producing male tilapia by MT administration. The study illustrates the difficulties associated with this field of study and the need for greater understanding of steroids and similar compounds in the environment before definitive statements can be made about any specific steroid.

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## PD/A CRSP Seventeenth Annual Technical Report

## STRAIN VARIATIONS IN SEX RATIO INHERITANCE

Eighth Work Plan, Kenya Research 2 (8KR2) Final Report

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## Abstract

Three distinct subspecies of Oreochromis niloticus have been identified in Kenya, including O. niloticus eduardianus, O. niloticus baringoensis, and O. niloticus vulcani. O. niloticus vulcani is cultured at the Sagana Fish Farm, Kenya, the PD/A CRSP prime site in Africa. During 1997 and early 1998, well over 50 individual pair spawns of tilapia from the Sagana stock of O. niloticus vulcani were attempted in support of a larger CRSP study designed to evaluate the sex ratios of offspring from a number of strains of O. niloticus from collaborating CRSP sites. Pond D3 at Sagana Fish Farm was dedicated to this activity and equipped with hapas. All fry produced in spawning hapas were transferred to rearing hapas. Although more than 100 fry were obtained from most spawns, survival to 5 cm was very low in the rearing hapas, and usually fewer than 25 fingerlings per spawn were obtained. This number was too low to complete the proposed Eighth Work Plan protocol and the fingerlings were discarded. Survival of about 80% is obtained during sex reversal in similar hapas in a similar pond. The only procedural difference is that during sex reversal fry are reared at higher densities. Only six batches of single-spawn fingerlings with adequate survival beyond a length of 5 cm were obtained. These were initially reared in hapas, followed by three weeks in the hatchery. However, these batches still contained no more than 60 fish, which was an insufficient number for this study. In mid-1998 a blower was installed in the hatchery and a complete diet became available, so the probability of success in rearing fry to 5 cm in the hatchery was greatly improved. However, information obtained in mid-1998 suggests that the population of tilapia at Sagana is not a pure strain of O. niloticus vulcani as originally believed, but is contaminated with O. spirulis and perhaps other species. If true, this greatly reduces the value of conducting further pair spawns. In consultation with the principal investigators of the parent study, it was decided not to conduct additional pair spawns. Provided funding is available, blood samples from this population and, if possible, from other Kenyan strains (e.g., O. niloticus baringoensis, from Lake Baringo) will be sent to Auburn University to assess the purity of these populations by electrophoretic analysis before undertaking any possible related followon activity.

## INTRODUCTION

Although populations of *Oreochromis niloticus* give mean population sex ratios of 50:50, considerable variation in sex ratios is observed when the offspring of individual pairs are considered. The source of this variation is unknown, but may be due to autosomal influence on sex determination in the strain of *O. niloticus* used. The Reproduction Control Research study "Methods for Strain Variations in Sex Ratio Inheritance" (8RCR1A) was included in the PD/A CRSP Eighth Work Plan to address this question (PD/A CRSP, 1997). In this study, the sex ratios of offspring from pair spawns from at least five distinct strains of *O. niloticus* were to be evaluated.

In Kenya there are three distinct subspecies of *O. niloticus* that have been identified: *O. niloticus eduardianus, O. niloticus baringoensis,* and *O. niloticus vulcani* (Seyoum and Kornfield, 1992). *O. niloticus vulcani* is presently cultured at the Sagana

Fish Farm, the PD/A CRSP prime in Africa. A minimum of 50 individual pair spawns were proposed to be carried out at Sagana for the purpose of determining the sex ratios of *O. niloticus vulcani* offspring in support of the larger study mentioned above. This document is the final report on work carried out at Sagana under this Eighth Work Plan activity.

## METHODS AND MATERIALS

This work was carried out at the at the Sagana Fish Farm, Sagana, Kenya. One pond and hapas for spawning and fry rearing were dedicated to the activity. Tilapia from the Sagana Fish Farm, reported to be of the *vulcani* strain of *O. niloticus*, were used for the pair spawns. Well over 50 pair spawns were attempted in hapas during 1997 and the first half of 1998. Fry produced by each pair were collected and reared as individual sets for determination of sex ratios by gonad examination after rearing to a minimum length of 5 cm.

## **R**ESULTS AND **D**ISCUSSION

The proposed protocol stipulated that sex ratios in each pair spawn would be based on at least 100 fish. Although more than 100 fry were obtained from most spawns, survival to 5 cm was low in the rearing hapas, and usually less than 25 fingerlings per spawn were obtained. This number was too low to complete the proposed protocol and the fingerlings were discarded. To date, only six batches of single-spawn fingerlings with good survival beyond a size of 5 cm have been obtained. These fingerlings were initially reared in hapas, followed by three weeks in the hatchery. However, due to low fry survival from the spawns, these batches still contained no more than 60 fish.

The cause of low survival during rearing to 5 cm is unclear. Survival of about 80% is obtained during sex reversal in similar hapas in a similar pond. The only difference is that during sex reversal fry are reared at higher densities.

In mid-1998 a blower was installed in the hatchery and a complete diet became available, so the probability of success-fully rearing fry to 5 cm in the hatchery as a follow-on activity during the next warm season was greatly improved. However, information obtained from regional tilapia experts in mid-1998 suggests that the population of tilapia at Sagana is not a pure strain of *O. niloticus vulcani* as originally believed, but rather that it is contaminated with *O. spirulis* and perhaps also with other species (Thys, pers. comm., 1998; Balarin, pers. comm., 1998). If true, this would greatly reduce the value of conducting follow-on pair spawns. In consultation with the principal investigators of the parent study, it was decided not to conduct additional pair spawns. Provided funding is available, blood

samples from this population and, if possible, from other Kenyan strains (e.g., *O. niloticus baringoensis*, from Lake Baringo) will be sent to Auburn University for electrophoretic analysis to assess the purity of these populations before undertaking any possible related follow-on activity.

## **ANTICIPATED BENEFITS**

Identification of strains of *O. niloticus* in which individual pairs consistently produce offspring with sex ratios of 50:50 would be a significant contribution towards the development of populations of YY males and reliable programs for the mass production of non-hormone-treated all-male tilapia fingerlings for stocking in rearing ponds. Production from ponds stocked only with male tilapia would be far more productive than ponds stocked with mixed-sex fish or with less than 100% males. Increased production would result in increased income for the farmers and greater supplies of food fish in the markets. These benefits would accrue to fish farmers throughout the tropics, including Africa, Southeast Asia, and Latin America.

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## EFFECT OF TREATMENT TIMING AND DOSE ON MASCULINIZATION WITH TRENBOLONE ACETATE

Ninth Work Plan, Reproduction Control Research 5A (9RCR5A) Final Report

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## ABSTRACT

Preliminary studies in our laboratory showed that the synthetic androgen trenbolone acetate (TA) is a good candidate for masculinizing Nile tilapia fry using short immersions. In this study, we investigated the effects of treatment timing and treatment dose on the masculinizing potential of TA. Our results suggest that maximum masculinization can be achieved by short-term immersion on 13 and 14 days post-fertilization. Immersion prior to and after these days resulted in less or no masculinization. We tested the effects of dosage by using the traditional single factor experiment as well as a novel approach: the fractional factorial experiment. In one experiment, immersion in all doses (500, 750, and 1,000 µg l<sup>-1</sup>) of TA resulted in significant masculinization with no differences observed between doses. In a subsequent experiment with fry from a different brood, none of the doses resulted in significant masculinization. The fractional factorial experiment was designed to simultaneously examine the effects of treatment dose, treatment duration, and density of fish. Significant masculinization occurred in some treatments; however, no clear pattern of interaction emerged among these factors. Nevertheless, this experimental approach holds great promise for gaining rapid screening results which will be useful in designing follow-up experiments.

## INTRODUCTION

Masculinization of tilapia for the production of male-biased populations continues to be an important tool for aquaculturists to prevent unwanted reproduction (which shunts energy away from growth towards gamete production) and to produce the sex with the larger growth potential (Green et al., 1997). Previous work in our laboratory has shown that shortterm immersion in androgenic steroids can result in masculinization of Nile tilapia (Gale et al., 1995, 1999; Contreras-Sánchez et al., 1997). These studies showed that immersion in androgen has the potential to be an alternative to dietary treatment with steroids for the masculinization of tilapia. A variety of androgens-especially synthetic androgens-are effective masculinizing agents (Hunter and Donaldson, 1983); however, there may be differences in their potency. We have shown that a single immersion in the non-aromatizable synthetic androgen 17α-methyldihydrotestosterone (MDHT) on 13 days postfertilization (dpf) at 28°C, which corresponds to 364 Celsius Temperature Units (CTU), is as effective as two immersions in MDHT at 280 and 364 CTU (Fitzpatrick et al., 1999a). Both treatments resulted in significantly more males being produced than in the controls.

Trenbolone acetate (TA) has been reported to be an effective masculinizing agent when fed to tilapia (Galvez et al., 1996). Because we have had variable success masculinizing tilapia by immersion in  $17\alpha$ -methyltestosterone (MT) or MDHT, we

decided to examine the efficacy of immersion in TA. Trenbolone acetate has been widely used in the cattle industry for growth enhancement and is considered a potent androgenic and masculinizing agent (Galvez et al., 1996). To determine the best treatment conditions for masculinization by immersion, studies must be conducted on the factors that are believed to play a critical role in determining efficacy. These major factors are: type of hormone, timing of treatment (relative to fish development), hormone dosage, duration of exposure, and density of fish during immersion. We have found that the best density for achieving significant masculinization by single immersion is 33 fish l-1 (Contreras-Sánchez et al., 1997; Fitzpatrick et al., 1999b). Previous studies in our laboratory showed that significant masculinization can be achieved by single immersion in MDHT if the fry are exposed to the hormone at 364 CTU (13 dpf). In the present study we determined if the results obtained with MDHT could be improved by using the synthetic steroid TA.

Fish density, hormone dosage, and length of exposure are factors that may interact to influence the degree of masculinization; therefore, a factorial design is needed to establish the minimum dosage, maximum density, and the shortest exposure needed for successful treatment. One approach is to conduct a single experiment in which all factors are examined simultaneously at different levels; however, this would require large numbers of tanks and more tilapia fry than can be produced from a single spawning. Therefore, our approach up to this time has been to examine one factor at a time while holding all others constant. We will describe our studies on the effects of day of exposure and hormone dose, which were based on the best treatment conditions obtained thus far. However, this approach limits the amount of information that can be gained on the interactions among the various factors. To investigate these interactions, we carried out a fractional factorial design (Kuehl, 1994) to examine the influence of multiple factors (hormone dosage, exposure duration, and fish density) on the efficacy of TA. This design allows information to be obtained on factors of interest in the early stages of experimentation when the number of treatments exceeds the resources (Kuehl, 1994). Results of fractional factorial experiments can then be utilized in designing follow-up studies.

## METHODS AND MATERIALS

Breeding families of Nile tilapia, Oreochromis niloticus, were placed in 200-l aquaria (one male to three females). The temperature was maintained at  $28 \pm 1^{\circ}$ C. Time of spawning was monitored every two hours. All spawning occurred between 1600 and 1900. Once breeding occurred, the other fish were removed and the brooding female was left to incubate the progeny. At 10 dpf (280 CTU), fry were removed from the tank and randomly assigned to experimental groups. Development of the fry was expressed in CTU (mean water temperature in °C x the number of days since fertilization). The fry used in each experiment came from an individual female. Each replicate was housed in a 3.8-1 glass jar with dechlorinated tap water. The water in all treatments was maintained at  $28 \pm 1^{\circ}$ C under constant aeration. Treatments consisted of immersions in either steroid or ethanol, which were mixed before addition of fry. Steroids were obtained from Sigma Chemical Company (St. Louis, Missouri) and stored in stock solutions of ethanol  $(1 \text{ mg ml}^{-1})$  at  $4 \pm 1^{\circ}$ C.

The number of treatments and replicates included in each experiment was based on the number of fry obtained from a single spawning. Fry were collected after each immersion, jars were thoroughly cleaned, and then fish were reallocated in fresh dechlorinated tap water. Seven days after the final immersion, fry were transferred to Oregon State University's Warm Water Research Laboratory, Corvallis, Oregon, and reared in 75-1 fiberglass tanks in a recirculating system. For the fractional factorial experiment, fish were reared in 25-1 plexiglas chambers in a recirculating system. In all systems temperature and pH were monitored daily; ammonia, nitrites, and dissolved oxygen were checked weekly (Table 1). Water temperature in the grow-out system was maintained at  $28 \pm 1$  °C. At 70 to 80 dpf, sex ratios were determined by microscopic examination (10 and 40X) of gonads using squash preparations in Wright's stain (Humason, 1972). The weights of sampled fish were recorded at this time.

## Effect of Treatment Timing on Masculinization with TA

#### Experiment 1

Fry were immersed for three hours at 336, 364, or 392 CTU (12, 13, or 14 dpf) in 500  $\mu$ g l<sup>-1</sup> of TA at densities of 33 fish l<sup>-1</sup> in each replicate. Fish in the EtOH control group were immersed at 364 CTU at a density of 33 fish l<sup>-1</sup>. Each experimental group was triplicated.

#### Experiment 2

Fry were immersed for three hours at 308, 336, 364, 392, 420, or 448 CTU (11, 12, 13, 14, 15, or 16 dpf) in 500  $\mu$ g l<sup>-1</sup> of TA at densities of 33 fish l<sup>-1</sup> in each replicate. Fish in the control groups were immersed in EtOH at either 364 or 420 CTU at a density of 33 fish l<sup>-1</sup>. Each experimental group was triplicated.

#### Experiment 3

Fry were immersed for three hours at 336, 364, 392, or 420 CTU (12, 13, 14, or 15 dpf) in 500  $\mu$ g l<sup>-1</sup> of TA at densities of 33 fish l<sup>-1</sup> in each replicate. Fish in the control group were immersed in EtOH at 364 CTU at a density of 33 fish l<sup>-1</sup>. Each experimental group was triplicated.

#### Effect of Dose on Masculinization with TA

#### Experiment 4

Based on results obtained from the previous experiments, fry were immersed for three hours at 364 and 392 CTU (13 and 14 dpf) in 500, 750, or 1,000  $\mu$ g l<sup>-1</sup> of TA; 500, or 1,000  $\mu$ g l<sup>-1</sup> of MDHT; or EtOH vehicle (control group) at densities of 33 fish l<sup>-1</sup>. Because of brood size, treatments were not replicated.

#### Experiment 5

Fry were immersed for three hours at 364 and 392 CTU (13 and 14 dpf) in 500, 750, or 1,000  $\mu$ g l<sup>-1</sup> of TA or in EtOH vehicle (control group) at densities of 33 fish l<sup>-1</sup>. Because of brood size, treatments were not replicated.

## Effect of Dose, Exposure Time, and Fish Density on Masculinization with TA

## Experiment 6: Fractional Factorial

Fry were immersed at 308, 336, 364, 392, and 420 CTU (11, 12, 13, 14, and 15 dpf) in TA or EtOH. Fry densities were 10, 19, 38, 75, or 150 fish  $l^{-1}$ ; hormone dosages were 62.5, 125, 250, 500, or 1,000 µg  $l^{-1}$ ; exposure duration was 0.75, 1.5, 3, 6, or 12 hours. Because we decided to use a fractional factorial design, only certain combinations of treatment conditions were used (Table 2a). To choose the combination of treatment factors to be used, a model was generated using Statistical Analysis Systems for Windows, release 6.10 (SAS Institute Inc., Cary, NC). Under this model, only replication around the middle treatment level for each factor is recommended (38 fish  $l^{-1}$ ; 250 µg  $l^{-1}$ ; 3 h). The

 Table 1.
 Mean ± SE values for temperature, pH, dissolved oxygen (DO), nitrites, hardness, alkalinity, and ammonia for all immersion experiments.

Experiment	Temperature	pН	DO	Nitrites	Hardness	Alkalinity	Ammonia
1 (Time)	29.95 ± 0.14	$7.70 \pm 0.06$	$6.29 \pm 0.09$	0.01 + 0.02	150 40 + 15 04	20.00 ± E.66	0.17 + 0.05
1(111110)	$26.65 \pm 0.14$	$7.79 \pm 0.06$	$6.26 \pm 0.06$	$0.01 \pm 0.08$	$159.40 \pm 13.04$	$69.00 \pm 3.00$	$0.17 \pm 0.03$
3 (11me)	$26.80 \pm 0.20$	$6.56 \pm 0.36$	$6.24 \pm 0.06$	$0.07 \pm 0.03$	$160.40 \pm 12.26$	$55.20 \pm 4.80$	$0.70 \pm 0.46$
4 & 5 (Dose)	$28.72\pm0.14$	$7.02\pm0.98$	$6.58\pm0.07$	$0.02\pm0.004$	n.a.	n.a.	$0.23\pm0.05$
6 (Frac. Fact.)	$28.25\pm0.16$	$7.83\pm0.84$	$6.30\pm0.23$	$0.04\pm0.09$	n.a.	n.a.	$0.13\pm0.35$

Experimental Design	<b>Density</b> (fish 1 <sup>-1</sup> )			Dos (119	sage		
Design	(110111)	0	62.5	125	250	500	1.000
0.375 н	10 19 38 75			120	1		1,000
0.75 н	10 19 38 75 150			1 1		1 1	
1.5 н	10 19 38 75 150	1	1		1 6 1		1
3н	10 19 38 75 150			1 1		1 1	
6н	10 19 38 75 150	1			1		

Table 2a. The combinations of factors tested for effects on masculini-

Factorial.

zation of Nile tilapia are shown for Experiment 6: Fractional

fractional factorial design is effective in screening studies to check on many factors, under the assumption that only a few effects are important. However, the fractional factorial design carries the caveat that follow-up experiments must be conducted using suitable replication once the levels for the various factors are chosen.

## **Statistical Analysis**

For experiments 1 and 3, data were pooled from replicate tanks, because there was no evidence of tank effects within treatments (Chi-square test). Pairwise comparisons for sex ratio and mortality data were analyzed using Fisher's exact test with exact p-values (a more conservative test than the Chi-square test for small sample sizes) estimated in GraphPad Prism<sup>TM</sup>. The mean final weights of sampled fish were analyzed for differences between groups using one-way ANOVA including mortality as a possible confounding variable. For all analyses, differences were considered statistically significant when the p-value (*P*) was less than 0.05. For experiment 2, the data were not analyzed statistically because of the sex ratio bias (see Results). Experiments 4 and 5 had only one experimental unit

#### RESULTS

## **Effects of Treatment Timing**

### Experiment 1

Significant masculinization of tilapia fry was obtained with single immersions in TA (Figure 1). The percentage of males in the groups immersed for 3 h on 12, 13, and 14 dpf was significantly higher than that in the control group (46.5, 72.3, and 69.3% versus 18.1%, respectively;  $P \le 0.0001$ ). The percentage of males in the 13 and 14 dpf treatments were not significantly different from each other (P = 0.26). The proportion of males in the 12 dpf immersion treatment was significantly higher than the control, but significantly lower than the 13 and 14 dpf treatments (P < 0.001 in all cases).

#### Experiment 2

All control groups (immersed in EtOH) and treatment groups contained 100% males. Therefore, the data were not analyzed further.

#### Experiment 3

Significant masculinization of tilapia fry was obtained with single immersions in TA (Figure 2; P = 0.008). No significant differences were found between the two control treatments (EtOH immersions on 12 or 15 dpf; P = 0.76). The percentages of males in the groups immersed for 3 h on 13 or 14 dpf were significantly higher than that in the control group (41.5 and 41.9 versus 22.6%, respectively;  $P \le 0.01$ ). The percentages of







Figure 2. Effect of single immersion in trenbolone acetate (TA) on 12, 13, 14, or 15 days post-fertilization on masculinization of *Oreochromis niloticus* fry. Figure depicts mean + SE percentage of males in each treatment with n = 18–98 per treatment (pooled triplicates). Fish were immersed for 3 h at a density of 33 fish l<sup>-1</sup>. Statistically significant differences from controls are represented by asterisks (\*\* P < 0.01).

males in the 12 and 15 dpf treatments were not significantly different from the control groups nor between each other (P = 0.08, 0.09, and 0.89, respectively).

For all single immersion experiments, mortality and final weights were not significantly different among treatment groups. Water quality in rearing tanks was maintained close to the optimal values for tilapia culture (Table 1).

## **Effects of Dose**

#### Experiment 4

Immersion of tilapia fry in 500, 750, and 1,000 µg l<sup>-1</sup> of TA and 1,000 µg l<sup>-1</sup> of MDHT resulted in significant masculinization of tilapia fry (Figure 3a;  $P \le 0.0001$ ). Control fish were 45% males. Fry immersed in 1,000 µg l<sup>-1</sup> of TA showed the highest percentage of males (94.0%), followed by those in 500 µg l<sup>-1</sup> of TA (80%), 1,000 µg l<sup>-1</sup> of MDHT (76%), and 750 µg l<sup>-1</sup> of TA (70%); however, the group treated with 500 µg l<sup>-1</sup> of MDHT (47%) did not show significant masculinization. All significant differences had P < 0.05. High mortality was observed in treatments groups immersed in 1,000 µg l<sup>-1</sup> of MDHT and in 500 and 1,000 µg l<sup>-1</sup> of TA.

## Experiment 5

All treatments in this experiment had similar percentages of males (46 to 54%; Figure 3b) which did not differ from the control group (49%).

Some fish in experiments 4 and 5 showed malformed heads and mouths, incomplete development of the operculum, and segmented dorsal fins.

## Effect of Dose, Exposure Time, and Fish Density on Masculinization with TA

#### **Experiment 6: Fractional Factorial**

Significant masculinization was achieved by immersion of tilapia fry in TA. The best results obtained ranged between 72.1





Figure 3. Effect of dose of trenbolone acetate (TA) and  $17\alpha$ methyldihydrotestosterone (MDHT) on masculinization of *Oreochromis niloticus* fry. Fish were immersed for 3 h at a density of 33 fish l<sup>-1</sup> on 13 and 14 days post-fertilization. Graph depicts the percentage of males in each treatment with samples sizes of 15 to 49 for (a) and 45 and 48 for (b). Statistically significant differences from controls are represented by asterisks (\* *P* < 0.05; \*\* *P*< 0.01; \*\*\* *P* < 0.001).

and 83.3% males in comparison to 51.1% in controls. These results included a wide variety of values for dosage, density, and duration of exposure. No clear pattern of interaction between the three factors tested was apparent (Table 2b).

Some fish in this experiment showed white spots in their eyes as well as malformed heads and mouths.

### DISCUSSION

Masculinization of tilapia fry can be achieved through shortterm immersion in TA. Our results indicate that significant masculinization occurs when fish are immersed in 500  $\mu$ g l<sup>-1</sup> of TA for three hours at 420 or 448 CTU (13 or 14 dpf) at a density of 33 fish l<sup>-1</sup>. In an earlier study, we obtained 92% males in fry immersed in 500  $\mu$ g l<sup>-1</sup> of TA for two hours on 11 and 13 dpf, while the controls had only 14% males (Contreras-Sánchez et al., 1997). This suggests that the window of sensitivity for masculinizing tilapia is short and may vary between broods, which in turn supports the idea that there are other major factors that influence susceptibility to androgen-induced masculinization. These differences among broods may be due to differences in embryo developmental stages, sensitivity to the steroid used, and/or inbreeding.

The single factor dose experiments suggest that increases in the percentage of males produced cannot be achieved through minor increases in concentration of hormone (e.g.,  $500 \ \mu g \ l^{-1}$ ) versus 1,000  $\ \mu g \ l^{-1}$ ). However, due to small brood sizes and high mortality rates, our results are still inconclusive. Since water quality in our experiments has been carefully monitored and shown to be excellent (Table 1), we speculate that the mortality and malformations observed may indicate inbreeding problems. These trials will need to be repeated with outbred populations in order to draw more substantive conclusions on dose effects.

The fractional factorial design presented an opportunity to explore the influence of several major factors and their interactions. However, despite significant masculinization results, this technique needs refinement and perhaps larger experimental units. We feel confident that a larger sample size

Table 2b. The percentage of males in the treatment groups is for Experiment 6: Fractional Factorial. Statistically significant differences from controls are depicted with \* (P < 0.05).

Results	<b>Density</b> (fish l <sup>-1</sup> )	Dosage (µg l <sup>-1</sup> )					
	-	0	62.5	125	250	500	1,000
0.375 н	10 19 38 75 150				52.4		
0.75 н	10 19 38 75 150			73.7 72.1*		53.3 69.6	
1.5 н	10 19 38 75 150	52.2	58.6		$81.8$ $58.7 \pm 4.1$ $66.7$		55.2
3н	10 19 38 75 150			83.3* 62.2		60.0 77.8*	
6н	10 19 38 75 150	50.0			73.7		

will provide better results for this design. Once again, the use of new broodstock will help to clarify this.

Sex ratios deviating substantially from the theoretical 1:1 were obtained in the control groups of some of the experiments. Progeny sex ratios have ranged from 15 to 100% males and seem to be linked to specific adult crosses. We have documented this result previously (Fitzpatrick et al., 1999c) and it has also been reported in Shelton et al. (1983).

## **ANTICIPATED BENEFITS**

We have successfully refined the technique for masculinizing Nile tilapia by immersion in masculinizing steroid. This latest development defines the times at which the highest masculinization can be achieved. Trenbolone acetate is a good candidate for successful masculinization of tilapia. The use of fractional factorial design and larger sample sizes will allow us to refine this technique to increase effectiveness and consistency. The optimization of this technique may enable farmers to masculinize tilapia with androgens while minimizing the risk of pond contamination with MT.

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# PD/A CRSP Seventeenth Annual Technical Report

## AQUACULTURE POND MODELING FOR THE ANALYSIS OF ENVIRONMENTAL IMPACTS AND INTEGRATION WITH AGRICULTURE: MODEL EVALUATION AND APPLICATION TO THE ECOLOGICAL ANALYSIS OF INTEGRATED AQUACULTURE/AGRICULTURE SYSTEMS

Eighth Work Plan, Aquaculture Systems Modeling Research 1A (8ASMR1A) Final Report

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## Abstract

A model developed to analyze the environmental impacts of aquaculture and the productivity and ecological function of integrated aquaculture/agriculture systems was evaluated using sensitivity analysis and model validation methods. The validated model was used to identify priority areas for future research in integrated aquaculture/agriculture systems and to study the flow of nitrogen in these systems. Sensitivity analysis results showed that the model was most sensitive to maximum photosynthetic rate, aerobic sediment depth, oxygen threshold for aerobic conditions, water infiltration rate, and organic matter sedimentation rate. Model validation was established by the successful replication of observed patterns for individual fish weight, dissolved oxygen, total ammonia nitrogen, sediment organic matter, sediment nitrogen, chlorophyll *a* biomass, and corn grain yield. Application of a qualitative evaluation of research priorities that combined sensitivity analysis and parameter availability identified stocking practices, sediment processes, and water management as priority areas for future research in integrated aquaculture/agriculture systems. Based on the simulation results, the model appears to be appropriate for analyzing the management of organic matter and nitrogen in integrated aquaculture/agriculture systems. The model is also useful for identifying research areas that may be important in the scientific understanding of integrated aquaculture/agricu

## INTRODUCTION

The development of an integrated aquaculture/agriculture model was based on existing models of aquaculture ponds and of agricultural crops. Both models were modified extensively to achieve an integrated model that could be executed with data such as those collected under the PD/A CRSP. The integrated model was developed for analyzing the function of integrated aquaculture/agriculture systems and the impacts of aquaculture on its surrounding environment.

Previous progress reports describe various model components. The primary objective in this final report is to summarize the results of model testing (sensitivity analysis and validation) and discuss the implications of simulation results obtained in a set of modeling studies. The focus in the modeling studies was the examination of the effect of cycling pathways on system nitrogen retention and productivity. Details of the model are available in previous progress reports (Jamu and Piedrahita, 1998, 1999) and in Jamu (1998), where sediment changes over long-term (10 year) simulations are also discussed.

## SENSITIVITY ANALYSIS

Sensitivity analysis involves making changes to model rate coefficients singly or in combinations and determining the resulting changes in model output. Sensitivity analysis results are used to identify the most important model parameters, areas for future research, and the level of precision required for measuring system input variables (Kitchell et al., 1977). Sensitivity analysis can also be used to test the robustness of a model. The integrated aquaculture/agriculture model was subjected to a sensitivity analysis by varying rate coefficients based on the observed variability (±1 SD) for the selected rate coefficients and parameters (initial values). Where such information was not available, a constant  $\pm$  50% adjustment was used. The percentage adjustment was determined on the basis of sensitivity results for aquaculture ponds reported in the literature (e.g., Lanhai, 1997; Schaber, 1996) and the observed variability of the different parameters in the PD/A CRSP database. The response variables for the sensitivity analysis were selected to reflect the objectives of the research with respect to nitrogen cycling and yield productivity. Response variables were also selected on the basis of their utility in aiding the interpretation of the sensitivity analysis results. The sensitivity analysis consisted of varying 22 parameters and determining the response for the following 11 state variables: water column nitrogen, sediment organic matter, nitrogen retention index (N output/N input) (also known as the nitrogen output to input ratio (DeAngelis, 1992)), individual fish weight, crop soil nitrogen, chlorophyll a concentration, pond dissolved oxygen, water column organic matter, crop biomass, and terrestrial soil organic matter.

The sensitivity analysis results are presented as a percent change and as a normalized sensitivity coefficient (NS). The normalized sensitivity coefficient is calculated as (Fasham et al., 1990):

$$NS = \frac{\left(\frac{\Delta V}{V_b}\right)}{\left(\frac{\Delta P}{P_b}\right)}$$

where

 $DV = (V_b - V)$  = average change of a response variable (e.g., fish weight) over the simulation period

- $V_{b}$  = average value of a response variable for the base run
- *V* = average value of a response variable for the sensitivity analysis run
- $DP = (P_{h} P) =$ change in model parameter
- $P_{h}$  = baseline value of model parameter and
- *P* = value of model parameter for the sensitivity analysis run

To determine the most sensitive parameters, the normalized sensitivity coefficients were first ranked in descending order based on the absolute value of NS. The determination of the cut-off point between the most sensitive and less sensitive parameters is usually arbitrary. However, in this work, the cutoff point was identified with the aid of quantile plots or Q-Q plots (Swartzman and Kaluzny, 1987). This procedure involved plotting the effects (normalized sensitivity coefficients) against quantiles from a normal distribution. If the plots come from the same normal distribution the quantile plots should be a straight line and any sensitive parameters deviate from the straight line. The quantile plots were not used in a quantitative manner because this required that the data be normally distributed—an assumption that could not be met by the simulated values.

#### **MODEL VALIDATION**

The integrated aquaculture/agriculture systems model was validated by graphically comparing model output to observed time series data from experiments conducted in Honduras, Thailand, and Malawi. Where adequate time series data were not available, tabular comparisons were used. The data used for model validation were independent from the data used for model calibration.

## **Agriculture Component Model**

The agriculture component model was validated using data from irrigation trials conducted at the Kasinthula Agricultural Research Station, Malawi (Kauta and Kadwa, 1993). The Kasinthula Agricultural Research Station located at 16°S and 34°E is part of the Malawi National Agricultural Research System. This station is used for research on irrigated tropical agriculture. The data for validating the terrestrial crop model were from trials designed to investigate the effect of water and nitrogen application rates on the yield of corn (Zea mays). The treatments consisted of nitrogen fertilization rates as the main factor. Nitrogen was applied as urea at 80 and 160 kg ha<sup>-1</sup> at planting and 35 days after planting. Water application rates were the second factor in the factorial experiment. Water was applied as a percentage (20%, 40%, 80%, 100%, or 120%) of the base amount (100% of total crop evapotranspiration ( $Et_0$ )). The corn was planted during the winter (May to September) and was harvested after 121 days. Malawi crop data were used because of the unavailability of comparable data in the PD/A CRSP database that could be used to validate the behavior of the agriculture model when nitrogen and water rates were varied. Validation of the agriculture model under these conditions is crucial since transfer of nitrogen from the pond and the use of pond water for irrigation are the major pathways for integration.

#### **Aquaculture Component Model**

Validation of the aquaculture component model was done using data from PD/A CRSP experiments conducted in Honduras (Third Work Plan, Experiment 3) and Thailand (Fourth Work Plan, Experiment 4) and from a pilot integrated

aquaculture/agriculture system in Malawi. The variables selected for validation were individual fish weight, chlorophyll a concentration, water column total ammonia nitrogen (TAN), dissolved oxygen concentration (DO), pond sediment organic matter, and pond sediment nitrogen. Details of site descriptions, experimental protocols, and sampling procedures for the PD/A CRSP data are given in Bowman and Clair (1996) and Egna et al. (1995). In summary, the Honduras experiment (PD/A CRSP Third Work Plan, Experiment 3) was designed to investigate the effects of four chicken manure application rates (125, 250, 500, and 1,000 kg<sup>-1</sup> ha<sup>-1</sup> wk<sup>-1</sup>) on fish production and pond dynamics. The experiment at the Bang Sai Fisheries Station, Ayutthaya, Thailand (PD/A CRSP Fourth Work Plan, Experiment 4) was designed to investigate the effects of fertilizing ponds with chicken manure and different levels of supplemental nitrogen fertilization. Three different chicken manure application rates were used and nitrogen was added to maintain an input N:P ratio of 5:1.

The experiments conducted in Malawi were designed to investigate the effects of applying farm plant wastes to ponds and determining the effect on fish yield and sediment organic matter quality and accumulation. The fish species used was the macrophytophagous *Tilapia rendalli* stocked at 2 fish m<sup>-2</sup>. The experiment was part of an integrated aquaculture/agriculture systems study designed to investigate pond utilization of agricultural wastes, nitrogen cycling, and productivity of integrated aquaculture/agriculture systems. The treatments were as follows: corn bran x leucaena leaf supplement, corn bran x leucaena leaf supplement x plant wastes at 24 kg<sup>-1</sup> ha<sup>-1</sup> d<sup>-1</sup>, and corn bran x leucaena leaf supplement x plant wastes at 42 kg<sup>-1</sup> ha<sup>-1</sup> d<sup>-1</sup>. The treatments follow guidelines for input types and rates used on low-input integrated fish ponds in Malawi (Brummett and Noble, 1995).

The time series data for experiments with three or more replicates were plotted as mean  $\pm 1$  SD. The standard deviation was selected for evaluating the model fit to data because it is a fairly robust statistic for representing data spread around the mean. In addition, use of the standard deviation does not require the assumption of conditions that invalidate the use of other statistical methods, e.g., regression for model validation. The model was considered to be adequate or reliable when the model output followed the pattern of observed values and the model simulations were generally within one standard deviation of the replicate data.

## MODEL APPLICATION

One of the major objectives for developing ecosystem models is their application as analysis tools for complex systems which otherwise would be difficult to analyze using conventional tools such as field experimentation, inventory monitoring, etc. Of more relevance to integrated aquaculture/agriculture systems is the identification of priority areas for future research and the identification of important processes that could be managed to increase/optimize productivity and efficiency of the system. In this report, the application of the model to identification of priority areas for future research and the use of the model to conduct a modeling study are demonstrated.

#### **Identification of Priority Areas for Future Research**

The parameters that would benefit from future research were identified using a qualitative method that combined the rankings of sensitivity analysis results and parameter availability to arrive at the priority ranking of a given parameter/ process for future research (Kitchell et al., 1977). Parameter availability considers the observed variability of the parameter, ease of measurement, and source of values for the parameter, i.e., whether parameters were estimated for the species or system under consideration or were extrapolated from another species/system. For example, a parameter that had high variability in field measurements would be ranked as having low availability. If the response variable was sensitive to this parameter then one would rank it high on the priority list for future research. The priority rankings were in three categories that represented parameters with high, medium, and low potential for future research. The parameters that were used in the qualitative evaluation were the most sensitive parameters with respect to nitrogen cycling index, fish weight, crop grain yield, pond (water column and sediment) nitrogen, and organic matter.

## Ecological Analysis of Integrated Aquaculture/ Agriculture System

The modeling study consisted of simulations of different scenarios designed to study the effects of different cycling pathways on system productivity and nitrogen retention. This provided information necessary to identify the most important linkages between an aquaculture and an agricultural crop. The input scenarios that were simulated for three hypothetical integrated aquaculture/agricultural systems are shown in Table 1. The basic input scenario for the aquaculture component was either chicken manure or crop waste applied as a pond fertilizer. Cycling pathways were added to integrate the aquaculture and the agriculture components until all possible combinations of linkages were achieved (Table 1).

The model was run for a 120-day period using dry season input data from PD/A CRSP experiments conducted at El Carao, Honduras. Model runs simulated tilapia and corn production in the aquaculture and agriculture components, respectively. System variables for the simulation runs were selected to reflect both the ecological performance (biomass yield and nitrogen retention) and impact on marketable product (individual fish weight). The variables selected to assess the effect of cycling pathways were total system nitrogen input (kg N ha-1); gross fish yield (kg ha-1); corn yield (kg ha<sup>-1</sup>); individual fish weight (g wet weight); total nitrogen yield (kg N ha<sup>-1</sup>); and nitrogen retention index. The total system nitrogen yield was calculated on the assumption that the agriculture and aquaculture components occupied equal proportions of land totaling one hectare. For testing the hypothesis that integration of aquaculture and agriculture

Table 1.	External input scenarios for three different hypothetical
	integrated aquaculture/agriculture systems.

Farm System	Agriculture Component	Aquaculture Component
1	Inorganic fertilizer @ 160 kg N ha <sup>-1</sup>	Chicken manure @ 500 kg ha <sup>-1</sup> wk <sup>-1</sup>
2	None	Chicken manure @ 500 kg ha <sup>-1</sup> wk <sup>-1</sup>
3	Inorganic fertilizer @ 160 kg N ha <sup>-1</sup>	None

increases system nitrogen retention, the nitrogen retention index appears to be the most appropriate measure of nutrient cycling. The behavior of the nitrogen retention index (DeAngelis, 1992) relative to other ecological measures of nitrogen cycling is known for natural systems.

Biological nitrogen fixation was not considered in the nitrogen input term because it was assumed that this process constituted an insignificant nitrogen source relative to other inflow sources. In addition, nitrogen input through wet and dry deposition was also considered to be insignificant. The output term included nitrogen losses through leaching and denitrification. Volatilization was not considered to be a significant nitrogen sink. It was assumed that all harvests were consumed within the farm ecosystem. In order to compare the efficiency of nitrogen utilization between different simulated scenarios and that of agro-ecosystems, the nitrogen retention index was recalculated so that only yield output was considered as a nitrogen output. The recalculation of the nitrogen retention index reflects the shortage of information on estimates of nitrogen leaching losses in agro-ecosystems (e.g., Dalsgaard, 1996).

## RESULTS

Because of the large number of response variables used in the sensitivity analysis, the results for the different response variables were combined and only the most sensitive (absolute values) parameters are summarized in Table 2. The results indicate that the model is very sensitive to the water infiltration rate, the maximum photosynthetic rate, the oxygen threshold for aerobic conditions, the organic matter sedimentation rate, and the aerobic sediment depth. The model sensitivity results suggest that these parameters require accurate estimation and/or calibration.

The results on the identification of important processes that are likely to affect system productivity and nitrogen retention are presented in Table 3. These results are based on: a) a comprehensive sensitivity analysis (summarized in Table 2 for the ten most (absolute) sensitive parameters); b) literature reviewed during the development of the model; and c) observations from field experiments. The processes and management activities associated with the initial fish weight, aerobic sediment depth, non-phytoplankton light extinction coefficient, crop irrigation rate, water infiltration rate, and mineral soil organic matter decomposition rate coefficient were identified as the most likely to benefit from future research on integrated aquaculture/agriculture systems. Processes related to the organic matter sedimentation rate coefficient and initial sediment organic matter were found to be of medium research priority. The parameters with a low priority ranking for future research are the oxygen threshold for aerobic conditions, nitrogen mineralization rate coefficient, and the maximum photosynthetic rate. The oxygen threshold for aerobic conditions, despite having a high sensitivity is ranked low on future research priorities because of the high availability of basic information in the literature on this parameter for a wide range of microorganisms and aquatic environments. In general, these results show that research on stocking practices, sediment processes, and water management should be considered a priority for enhancing the scientific understanding of the function of the integrated aquaculture/agriculture system.

Model validation results show that the integrated model was able to replicate the pattern and behavior of observed data for the selected parameters (Jamu, 1998). The results of the crop growth simulations show that simulated maize yield responded to different water and nitrogen application rates. The simulated maize yields were similar to observed values. However, the model did not capture the pattern of phytoplankton dynamics for Ayutthaya, Thailand, and it also failed to capture the decrease in corn yield observed in treatments where nitrogen and water were applied at 160 kg N ha<sup>-1</sup> and 120% Et<sub>0</sub> respectively. In addition, the model failed to capture the increase in yield with increasing water application rates in fields that were fertilized at 80 kg N ha<sup>-1</sup>.

The unsatisfactory results for Thailand can be attributed to failure of the model to account for factors such as zooplankton control on phytoplankton populations or preference of phytoplankton species by fish. It was assumed in the model that the phytoplankton biomass consisted of a single assemblage of species with similar physiological and biochemical characteristics. Improving the phytoplankton model to include zooplankton grazing, variability in phytoplankton palatability, and modeling the dynamics of phytoplankton species composition may increase the accuracy of the phytoplankton simulations. The model accurately replicated the pattern of fish

Table 2. The effects of  $\pm$  50% or  $\pm$  1 SD in different parameter values on different state and output variables. The results show the ten most sensitive parameters ranked according to absolute magnitude of the normalized sensitivity values (NS).

Parameter	Sensitivity Variable	Baseline Value	Sensitivity Value	Parameter Change	Baseline Simulation	Sensitivity Simulation	% Change	NS
Water Infiltration Rate (m d <sup>-1</sup> )	Water column nitrogen	0.007	0	-1 SD	39.89	346.24	767.9	-7.68
Maximum Specific Phytoplankton Production	Chlorophyll <i>a</i>	0.83	0.91	+1 SD	1.4090	0.9376	-33.46	3.47
Rate per Unit of Carbon $(mg O_2 (mg C)^{-1} h^{-1})$	Chlorophyll <i>a</i> biomass	0.83	0.75	-1 SD	1.4090	1.8697	32.70	3.39
O <sub>2</sub> Threshold for Aerobic Conditions (mg l <sup>-1</sup> )	Nitrogen retention index	0.2	0.1	-50%	0.0962	0.2243	133.18	-2.66
	Water column nitrogen	0.2	0.1	-50%	39.89	13.51	-66.12	1.32
Organic Matter Sedimentation Rate k (d <sup>-1</sup> )	Water column organic matter	0.05	0.025	-50%	4,307.40	6,925.77	60.79	-1.22
Maximum Specific Phytoplankton Production	Dissolved	0.83	0.91	+1 SD	14.86	13.35	-10.18	1.06
Rate per Unit of Carbon $(mg O_2 (mg C)^{-1} h^{-1})$	Crop soil nitrogen	0.83	0.91	+1 SD	151.86	137.46	-9.48	-0.98
Aerobic Sediment Depth (m)	Water column nitrogen	0.001	0	-50%	39.89	2.29	-94.26	0.94
Phytoplankton Digestibility Coefficient (Dimensionless)	Individual fish weight	0.55	0.42	-1 SD	170.73	133.47	-21.62	0.92

Table 3. Summary of a sensitivity analysis of model parameter evaluations and their projected impact on future research on nitrogen cycling and productivity of integrated aquaculture/agriculture systems. Parameters were evaluated qualitatively based on the observed variability of measurements, availability of data for the system, and sensitivity.

Parameter	Sensitivity	Availability	<b>Research Priority</b>
Organic Matter Sedimentation Rate	High	Medium	Medium
Maximum Photosynthetic Rate	High	High	Low
Initial Fish Weight	High	Low	High
Aerobic Sediment Depth	High	Low	High
Non-phytoplankton Light Extinction Coefficient	High	Low	High
Irrigation Rate	High	Low	High
O <sub>2</sub> Threshold for Aerobic Conditions	High	High	Low
Water Infiltration Rate	High	Low	High
N Mineralization Rate	High	High	Low
Initial Sediment Organic Matter	High	Medium	Medium
Mineral Soil Organic Matter Decay Rate	High	Low	High

growth, and overall simulations of individual fish weight were within the range of observed data.

The results of the modeling study on the effects of introducing different organic matter and nitrogen cycling pathways on system productivity and nitrogen retention for the three system management scenarios are presented in Table 4. Recycling of plant wastes to the aquaculture pond in Systems 1 and 2 had a more appreciable effect on the nitrogen retention index, final individual fish weight, and gross fish yield than in System 3 (Table 4). Plant waste recycling also reduced the nitrogen retention index by 11% in Systems 1 and 2 and by 5% in System 3. In System 1, where the corn crop was assumed to be fertilized inorganically, recycling sediment to the agriculture component did not increase the corn yield. In System 2, corn yield increased with successive addition of linkages, and the simulated yield attained when both plant wastes and pond

sediments were recycled was equivalent to that of inorganically fertilized corn (Table 4b).

Addition of a cycling pathway for plant wastes from the agriculture to the aquaculture component reduced the nitrogen retention index and increased final fish weight and gross fish yield from 223 to 254 g and 3,793 to 4,320 kg ha<sup>-1</sup>, respectively. Recycling plant wastes to the aquaculture component in System 3 led to a slight reduction in the nitrogen retention index without a concomitant increase in gross fish yield (Table 4c). Linking the aquaculture and agriculture components through pond sediment and plant waste recycling led to a larger decrease in the nitrogen retention index compared to the recycling of plant wastes alone (Table 4a–4c).

Table 5 compares the simulated nitrogen yield efficiencies (system nitrogen yield/system nitrogen input) for various integrated systems scenarios with observed data reported in

Table 4a. Productivity and nitrogen retention for a hypothetical integrated aquaculture/agriculture farm. Corn crop fertilized at 160 kg N ha<sup>-1</sup>, chicken manure input to aquaculture component at 500 kg ha<sup>-1</sup> wk<sup>-1</sup> (System 1).

ention Gross Fish Yield Corn Yield N Yield lex (kg ha <sup>-1</sup> ) (kg ha <sup>-1</sup> ) (Fish + Grain)
$/N_{in}$ ) (kg ha <sup>-1</sup> ) <sup>a</sup>
92 3,793 2,880 57.5   71 4,320 2,880 63.5   99 3,791 2,880 57.5
5

<sup>a</sup> Assumes that fish and corn occupy equal land area in a 1-ha integrated farm.

Table 4b. Productivity and nitrogen retention for a hypothetical integrated aquaculture/agriculture farm. Fish ponds fertilized with chicken manure at 500 kg ha<sup>-1</sup> wk<sup>-1</sup>, no external nitrogen input to agriculture component (System 2).

Cycling Pathway	Ecosystem Performance Indicator							
	N Input (kg ha <sup>-1</sup> )	N Retention Index (N <sub>loss</sub> /N <sub>in</sub> )	Gross Fish Yield (kg ha <sup>-1</sup> )	Corn Yield (kg ha <sup>-1</sup> )	N Yield (Fish + Grain) (kg ha <sup>-1</sup> ) <sup>a</sup>			
Irrigation Water Irrigation Water x Plant Waste Irrigation Water x Pond Sediment Irrigation Water x Plant Waste x Pond Sediment	278 278 278 278 278	3.11 2.75 3.11 2.79	3,793 4,322 3,793 4,320	2,680 2,680 2,703 2,880	55.1 57.5 56.5 63.5			

<sup>a</sup> Assumes that fish and corn occupy equal land area in a 1-ha integrated farm.

Table 4c. Productivity and nitrogen retention for a hypothetical integrated aquaculture/agriculture farm. Corn crop fertilized at 160 kg ha<sup>-1</sup>, no external input to aquaculture component (System 3).

Cycling Pathway	Ecosystem Performance Indicator							
	N Input (kg ha <sup>-1</sup> )	N Retention Index (N <sub>loss</sub> /N <sub>in</sub> )	Gross Fish Yield (kg ha <sup>-1</sup> )	Corn Yield (kg ha <sup>-1</sup> )	N Yield (Fish + Grain) (kg ha <sup>-1</sup> ) <sup>a</sup>			
Plant Wastes	160	3.75	3,513	2,880	54.4			
Irrigation Water x Plant Waste	160	3.56	3,513	2,880	54.4			
Irrigation Water x Pond Sediment	160	3.56	3,513	2,880	54.4			
Irrigation Water x Plant Waste x Pond Sediment	160	3.67	3,513	2,881	54.4			

<sup>a</sup> Assumes that fish and corn occupy equal land area in a 1-ha integrated farm

the literature for various agro-ecosystems. Simulated nitrogen yield efficiency for the different integrated systems scenarios in this study ranged from 0.29 to 0.87. Integrated systems that recycled both pond sediments and plant wastes had the highest simulated yield efficiency (0.87) among the integrated farm scenarios modeled. This value was similar to that observed in diversified integrated rice-fish farms (Dalsgaard, 1996). Nitrogen yield efficiencies for integrated systems that were linked through irrigation water only were generally similar to those observed for rice monoculture systems and integrated rice-livestock-fish systems.

## DISCUSSION

The largest normalized sensitivity value was obtained when the water infiltration rate was reduced to zero. It has been observed that water infiltration rates in ponds are variable both over time and within any given single site (Teichert-Coddington and Claros, 1994; Chikafumbwa, 1996). Other factors that influence infiltration rates are pond inputs and soil type (Teichert-Coddington and Claros, 1994). In the model, the pond water infiltration rates were assumed to be constant. Although this assumption simplifies the incorporation of the water infiltration rate term in the model, it is incorrect and could result in significant errors in the simulation of water column nitrogen.

One of the uses of the analysis to determine which are likely to be the research areas with the greatest potential benefits is in designing field experiments. The highest priority areas identified are: processes and management activities associated with the initial fish weight, aerobic sediment depth, non-phytoplankton light extinction coefficient, crop irrigation rate, water infiltration rate, and mineral soil organic matter decomposition rate coefficient. PD/A CRSP experiments have addressed some of these questions in the past, while other areas have not received much attention. A possible experiment that could be conducted would look at the effect of pond water removal on pond water quality and fish yield. In such an experiment, water removal timing and amount would be based on predicted irrigation needs for a coupled agricultural crop, as opposed to pond considerations. Another possible experiment would vary the fish stocking density and fish size at stocking. A third experiment could vary the pre-stocking practices of a pond to improve food availability at the time of stocking.

The recycling of agricultural plant wastes to fertilize fish ponds produced the highest (absolute terms) system nitrogen retention index followed by the recycling of both pond sediments and plant wastes. Pond sediment input to inorganically fertilized corn did not lead to further increases in yield. The recycling of pond sediments to the agriculture component had a positive effect on yield of the agriculture component only when the corn crop in the agriculture component was not inorganically fertilized.

It was hypothesized that system nitrogen retention and productivity would increase as more cycling pathways between the aquaculture and agriculture components were introduced. However, the simulation results showed that an increase in nitrogen cycling pathways increased the system nitrogen retention and productivity only when one or both component(s) were nitrogen-limited. Since the incorporation of all possible cycling pathways did not improve the nitrogen retention index and productivity, the number of cycling pathways can not be interpreted as a measure of the improvement in the system's nutrient utilization efficiency. Similarly, lack of corn yield response in inorganically fertilized corn, and the corn yield response to increased organic matter in unfertilized corn, lend support to this hypothesis.

The reduction of the nitrogen retention index when plant wastes were applied to the fish pond could be attributed to a series of feedback processes that ultimately led to the efficient utilization of nitrogen. Presumably, the application of plant wastes enhanced the detrital and phytoplankton food webs that ultimately increased fish growth rates. Increased fish growth rates resulted in higher grazing rates, which stimulated phytoplankton productivity and nitrogen uptake. Also, application of plant wastes enhanced sediment organic matter accumulation rates as the results showed higher sediment organic matter concentrations in ponds receiving plant wastes than in ponds receiving chicken manure and artificial feed inputs alone. The increased sediment organic matter accumulation rates were due to both the high application rates and the large fraction of moderately decomposable organic matter that occur in corn stover relative to chicken manure or artificial feed (Gohl, 1981; Wang et al., 1985). Sediment organic matter has a high cation exchange capacity and therefore more sites for adsorption of cations like ammonium (Boyd, 1995). Therefore, sediment organic matter accumulation translates into higher

Table 5. Simulated and observed nitrogen retention for various agro-ecosystems.

Farm Type	Nitrogen Retention	Source of Data	Reference
Diversified Integrated Rice-Fish Farm, Philippines	0.78–1.36	Field measurements	Dalsgaard, 1996
Extensive Wheat, Straw Returned to Soil	1.00	Various	Frissel, 1978
$IRR \times PS \times F^1$	0.87	Model	This work, Table 4c
IRR x F	0.87	Model	This work, Table 4c
Intensive Wheat, Straw Returned to Soil	0.66	Model	Frissel, 1978
Mixed Arable Crop Livestock, South America	0.53	Model	Frissel, 1978
CM x IRR x PW	0.45	Model	This work, Table 4b
CM x IRR x PS	0.40	Model	This work, Table 4b
Extensive Carp Culture, India	0.14 - 0.47	Pond N mass balance	Chatterjee et al., 1997
CM x IRR x PŴ x F	0.33	Model	This work, Table 4a
CM x IRR x PS x F	0.33	Model	This work, Table 4a
Integrated Rice-Livestock-Fish, Philippines	0.22	Model	Schaber, 1996
Rice Monoculture, Philippines	0.17–0.18	Field measurements	Dalsgaard, 1996

<sup>1</sup>CM = chicken manure; IRR = irrigation; PW = plant wastes; PS = pond sediments; F = inorganic N fertilizer.

capacity for ammonia adsorption. Since sediment-adsorbed ammonia is not available for nitrification in the water column (Messer and Brezonik, 1983), the net effect of increased sediment organic matter concentration is a reduction in the amount of ammonia available for leaching and nitrification. Further, the increased ability of ponds receiving plant wastes to retain nitrogen may have been due to the development of the sediment anaerobic layer which decreased overall sediment nitrification rates. A reduction of sediment nitrification rates results in low nitrate seepage and nitrification-coupled denitrification losses (Blackburn and Blackburn, 1992).

## **ANTICIPATED BENEFITS**

Results of the sensitivity analysis, validation, and application show that the model is able to capture the general behavior of important variables in an integrated agriculture/aquaculture system. The model can also be used in research and management of integrated aquaculture/agriculture systems. The model has proven useful in identifying parameters that require accurate field measurements, priority areas of future research, and processes that are important to the scientific understanding of integrated systems. The model can also be used in planning experiments with integrated aquaculture/agriculture systems designed to improve sediment management and overall management of nitrogen and organic matter in aquaculture ponds.

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# PD/A CRSP Seventeenth Annual Technical Report

## AQUACULTURE POND MODELING FOR THE ANALYSIS OF ENVIRONMENTAL IMPACT AND INTEGRATION WITH AGRICULTURE: MODELING OF TEMPERATURE, DISSOLVED OXYGEN, AND FISH GROWTH RATE IN STRATIFIED PONDS USING STOCHASTIC INPUT VARIABLES

Eighth Work Plan, Aquaculture System Modeling Research 1B (8ASMR1B) Final Report

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## Abstract

A model has been developed for the prediction of water temperature, dissolved oxygen (DO), and fish growth using stochastically generated input weather variables. The model has been calibrated and validated using data from pond sites in Thailand, Honduras, and Rwanda. The model includes modules for the generation of weather parameter values, and for the calculation of water quality and fish growth. The weather parameters generated include hourly solar radiation, air temperature, wind speed, and wind direction. The water quality variables modeled include water temperature, DO, total ammonia nitrogen, and phytoplankton (in terms of chlorophyll *a*). For modeling purposes, the water column is divided into three layers, each of which is considered to be fully mixed. Temperature and DO are calculated separately for each of the three layers resulting in simulations of stratified ponds. Given the stochastic nature of the weather input variables, the model must be run a number of times for a given set of pond management conditions. Typically, the model is run 20 times for each data set. The probability distributions for water quality and fish yield can be calculated from the simulation results, providing the basis for the estimation of probability distributions that can be of use to pond managers, planners, researchers, and teachers.

## INTRODUCTION

Computer models developed previously for aquaculture systems use deterministic approaches. Under a deterministic simulation, a single set of model results is obtained for a particular run. While this approach is satisfactory for many situations, it fails to provide information related to potential changes in model results caused by stochastic variations in parameters or processes. An aquaculture pond is exposed to weather parameters that undergo stochastic changes. These changes result in fluctuations in model outcomes and real system conditions. The model described here is a first attempt at generating probability distributions of critical model variables based on a deterministic model run with stochastically generated weather parameter values. The model includes a module for the stochastic generation of weather parameter values (wind, solar radiation, wind velocity, wind direction, air temperature) (Lu et al., 1998). The module was developed to make use of the weather data available for PD/A CRSP sites. Although weather data have been collected at PD/A CRSP sites for a number of years, the magnitude of the data base for a given site is limited relative to data sets required for previous stochastic weather parameter generation models.

The weather generation module is linked to a model used to simulate pond water temperature, dissolved oxygen (DO), phytoplankton, total ammonia nitrogen (TAN), and fish biomass. Water temperature and DO are simulated using deterministic models based on previously developed PD/A CRSP models (+Culberson, 1993). TAN and chlorophyll *a* are simulated based on mass balance calculations (Lee et al., 1991). Fish growth is calculated using a bioenergetic model (Bolte et al., 1995; Jamu and Piedrahita, 1996) that can account for

growth based on natural food (phytoplankton) as well as on artificial feed and other food sources.

The model accounts for water quality stratification, and three layers are considered: top, middle, and bottom. Conditions within each layer are assumed to be uniform. Fish distribution in the water column is assumed to be uniform as long as DO concentration is above a critical value. If DO drops below the threshold in a given pond layer, the fish move to an adjacent layer in search for higher DO. If DO is below the threshold in all layers, the fish congregate in the surface layer.

This final report provides a general description of the model and each of its components. The description is followed by a presentation of sample results and a discussion of how these can be used.

## WEATHER GENERATION MODULE

The model has been constructed based on the statistics of monthly daily clearness indices ( $K_{\nu}$  defined as the ratio of the actual solar radiation to the clear-sky radiation) (Lu et al., 1998). The cumulative frequency distribution (CFD) of the daily clearness index for each month was been obtained from the PD/A CRSP database for each of the Honduras, Rwanda, and Thailand sites. The correlation of the CFD and monthly average daily clearness index was examined for each of the sites to test the procedure for the generation of hourly solar radiation values.

After the CFD curves of  $K_t$  are obtained from the historical data, the procedure for the generation of the hourly solar radiation values has two steps. The first step consists of generating solar radiation values for each day based on the

CFD of  $K_t$  for each month. In the second step, a series of hourly values is obtained by dis-aggregation of the daily value using a first order autoregressive model.

The data sets used to calculate the daily clearness index probability distributions cover periods of eight years (1984 to 1991), six years (1986 to 1991), and six years (1990 to 1995) for Honduras, Rwanda, and Thailand, respectively. However, the data sets are not complete, with data missing from periods of up to three months. The extraterrestrial solar radiation was calculated using the equations of Duffie and Beckman (1991), to allow the calculation of  $K_t$  and  $\overline{K_t}$  (monthly average clearness index value).

Since the monthly CFD curves are of similar shapes, a single equation form was selected for all the CFD curves after exploratory analysis of the data (TableCurve<sup>TM</sup>). The equation was selected on the basis of the quality of fit as indicated by the correlation coefficient ( $R^2$ ). To reduce the number of equations used while also maintaining the accuracy of the model, it was decided to combine similar CFD curves into a single equation as long as the  $R^2$  for the combined equation could be maintained above 0.98. The CFD curves were then normalized (Amato et al., 1986; Graham et al., 1988), and daily solar radiation values were generated from the normalized equations using an autoregressive model in which the clearness index value on a given day depended on the value for the previous day and on a random term (Lu et al., 1998).

Hourly values for solar radiation were generated from the daily values determined as indicated above using the approach proposed by Knight et al. (1991). The approach essentially breaks down the daily values according to the sunset hour angle and the hour angle (Duffie and Beckman, 1991) using an autoregressive model similar to the one used for estimating daily values.

## WATER QUALITY MODEL

Water quality parameters modeled were temperature, chlorophyll *a*, DO, and TAN. The model used is based on a previous model (Culberson, 1993) in which temperature and DO were simulated for a stratified pond (three layers) over 24-h periods. The model was expanded so that additional water quality parameters could be simulated (chlorophyll *a* and TAN), and simulations could be carried out for a complete growing season while maintaining a time step of one hour. Weather variables upon which water quality parameter calculations are based were generated using the Weather Generation Module described above.

## Temperature

Water temperature in the model is calculated based on energy balances for each of the water layers (Culberson, 1993). Potential energy inputs are through solar and atmospheric radiation and through sensible heat transfer from the air over the pond. Heat losses are through back radiation to the atmosphere, through the latent heat of evaporation of water from the pond, and through sensible heat transfer to the air over the pond. Heat may also be lost from the pond bottom layer by transfer to the pond sediment. Distribution of the heat energy through the water column is modeled with processes of diffusion (molecular, turbulent, and convective) and radiation. Distribution of radiant heat energy through the water column is modeled using the Beer-Lambert Law in which the light extinction coefficient is estimated from chlorophyll *a* values. The relationship between chlorophyll *a* and light extinction coefficient was developed from PD/A CRSP data sets (Jamu et al., 1999) and accounts for phytoplankton and non-phytoplankton contributions to light extinction.

## Chlorophyll a

Chlorophyll *a* was chosen as the variable to represent phytoplankton biomass. Phytoplankton growth rate was considered to be a function of light intensity, nutrient concentration (TAN), temperature, and chlorophyll *a* concentration. Non-optimal conditions of light intensity, nutrients, and temperature result in a reduction in the growth rate of phytoplankton below its optimum level (maximum specific growth rate). The effect of light intensity on photosynthetic rate at any particular time is considered to be dependent on the light intensity to which phytoplankton are exposed for the three previous days. Phytoplankton "sink" terms include respiration, grazing by fish, settling to the sediments, and non-predatory mortality.

## **Dissolved Oxygen**

Dissolved oxygen calculations were based on mass balances in which production, consumption, and transfer rates were simulated. Oxygen production was due to photosynthesis by phytoplankton. Consumption terms considered include phytoplankton respiration, fish respiration, organic matter oxidation, nitrification, and sediment respiration. Transfer terms include diffusion across the air-water interface, and diffusion between adjacent layers (molecular, turbulent, and convective diffusion as for the temperature calculations).

## Total Ammonia Nitrogen

TAN calculations were based on a mass balance similar to that developed for dissolved oxygen. Potential TAN sources include fertilizers, fish excretion, and the breakdown of organic nitrogen in the water column or the sediments. Processes that contribute to a reduction in TAN include uptake by phytoplankton and nitrification. Diffusion to the atmosphere was neglected in the model and TAN was assumed to be uniformly distributed in a pond.

## FISH GROWTH MODEL

The fish growth model uses bioenergetics calculations and is based on models proposed by Liu and Chang (1992), and later revised by others (Bolte et al., 1995; Jamu and Piedrahita, 1996). In the model, fish growth is estimated from the difference between the energy content of food intake and energy outputs from the fish. Food intake is considered to be a function of environmental parameters (temperature, DO, and TAN), fish characteristics (species, size, and feed preferences), and food concentration and composition (three types of food are considered: phytoplankton, non-phytoplankton, and supplied feed). The energy output is estimated as a single term representing excretion, wastes, and catabolism (Liu and Chang, 1992).

## SAMPLE RESULTS

The process of model calibration and validation was carried out with data from Honduras, Rwanda, and Thailand. After initial testing, the model was run for Thailand data, and results were used to adjust model coefficients in the process of calibration (Table 1). Subsequent runs for other sites used the calibrated values obtained from the Thailand simulations, in addition to site-specific parameters and initial conditions. Initial conditions used for a given run were the mean values of measurements from replicate ponds. In all cases, the simulations were carried out for 150-day growing seasons, and each simulation run was repeated 20 times to obtain probability distributions for the outcome variables. Sample results presented below include water quality and fish growth variables.

## Water Quality

The simulated water quality variables include temperature, chlorophyll *a*, DO, and TAN. Diel measurements of temperature and dissolved oxygen were collected at approximately two-week intervals, and those values were used to determine the accuracy of the simulations. The figures show the simulated values only for the days for which diel measurements

Table 1. Calibrated parameter values for the water quality model.

Parameter	Thailand	Rwanda	Honduras
Slope of PI Curve (mg C/ (mg chla *(µmol m <sup>-2</sup> ))	0.05	0.04	0.03
Oxygen Consumption in Organic Matter Oxidation (mg O <sub>2</sub> /mg OM)	3.0	3.0	1.08
Water Column $O_2$ Respiration Rate (mg $O_2$ m <sup>-2</sup> h <sup>-1</sup> )	100	100	200
Non-phytoplankton Light Extinction Coefficient (m <sup>-1</sup> )	3.6	4.1	8.5
Chla Content (mg l <sup>-1</sup> Chla/ mg l <sup>-1</sup> dry cell)	0.01	0.012	0.01



Figure 1. Simulated (curves) and observed (data points) temperature of the surface layer for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.

were available for comparison. Temperature and DO simulations are presented here. Chlorophyll results have been presented previously (Lu and Piedrahita, 1999).

Simulation results can also be presented as frequency distributions. The frequency distributions are constructed by determining the frequency of obtaining a simulated value within a given range (after 20 simulations, each 150 days long). Frequency distributions are constructed for each depth and for each time of the day for which measurements are available, namely 0600 h, 1000 h, 1400 h, 1800 h, and 2200 h.

The impact of the stochastic variation in the weather variables can be seen in the variation of the simulation results obtained. In addition, the accuracy of the simulations in predicting measured conditions varied between sites and between experimental treatments.

#### **Temperature**

Temperature simulations are shown for the Thailand site (Figures 1 through 4). The data correspond to Experiment 4, Cycle 4, designed to test the effects of different fertilization rates on fish growth and water quality. The treatments used included fertilization with 44, 100, and 200 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure. The manure was supplemented with urea to maintain the ratio of carbon to nitrogen (C:N) at 5:1. Figures 1 through 3 show the temperature at the three depths for the 100 kg ha<sup>-1</sup> wk<sup>-1</sup> manure fertilization treatment. This treatment was used to calibrate the model. Figure 4 shows the surface water temperature for the 200 kg ha<sup>-1</sup> wk<sup>-1</sup> treatment. In general, there were substantial differences between the temperatures at the three depths, but there was little difference between the temperatures for the different fertilization treatments.

Differences between the three depths are easily observed with the frequency distribution curves (Figures 5, 6, and 7). In general, the distributions tend to be much tighter for the bottom layer than for the top layer. This is in agreement with observations of bottom conditions being more stable than surface or mid-water conditions. There is also an increased frequency (and probability) of excessively high temperatures in the surface layer.



Figure 2. Simulated (curves) and observed (data points) temperature of the middle layer for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.

## Dissolved Oxygen

In general, dissolved oxygen simulations were less accurate than those for temperature, as illustrated in Figures 8 and 9. This reflected the difficulties in simulating the processes associated with oxygen production (photosynthesis) and consumption (respiration by phytoplankton, fish, sediments, etc.). Frequency distributions (Figure 10) illustrate the usefulness of the stochastic simulations in quantifying the probability of having DO values outside a prescribed range needed by the fish. The frequency distribution (Figure 10) shows that during the evening there is a high probability of low DO even in the surface layer, but this probability is reduced considerably during the middle of the day. Comparison of the simulated and measured probability distributions (Figure 11) shows very good agreement. The results on Figure 11 are aggregated for the daylight hours (0600 h to 1800 h) due to the low number of measured values available in the database.



Figure 3. Simulated (curves) and observed (data points) temperature of the bottom layer for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 4. Simulated (curves) and observed (data points) temperature of the surface layer for the Thailand site under fertilization with 200 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 5. Frequency distribution curve for temperature of the surface layer for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 6. Frequency distribution curve for temperature of the middle layer for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 7. Frequency distribution curve for temperature of the bottom layer for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 8. Simulated (curves) and observed (data points) DO of the surface layer for the Thailand site with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 9. Simulated (curves) and observed (data points) DO of the surface layer for the Honduras site with 500 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 10. Frequency distribution curve for DO of the surface layer for the Honduras site under fertilization with 500 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.

## Fish Growth

Fish growth proved to be the most difficult parameter to simulate. Accuracy of the simulations varied considerably between sites and between pond treatments. In addition, data from some replicate ponds showed very high variability (Figure 12). This suggests that there are subtle differences between ponds receiving replicate treatments that are difficult to identify and that result in substantial differences in fish growth. These differences in fish growth between replicates were more noticeable for Thailand than for the other sites. For example, a simulation for the Rwanda site shows less variation between replicate ponds, and better agreement between the measured and simulated values (Figure 13).

## DISCUSSION

The generated weather variables constituted satisfactory stochastic data sets based on the available measurements for each PD/A CRSP site. However, agreement between simulated and measured values of water quality and fish growth varied considerably. The quality of the simulations varied between



Figure 11. Frequency distributions for the daytime (0600 to 1800 h) measured and simulated surface DO for the Honduras site



Figure 12. Fish growth simulation for the Thailand site under fertilization with 100 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure.



Figure 13. Fish growth simulation for the Rwanda site under fertilization with 500 kg ha<sup>-1</sup> wk<sup>-1</sup> of chicken manure and fresh cut grass.

sites and between treatments. Although the stochastic simulations account for much of the variability caused by weather fluctuations, there are variations between replicate ponds that could not be simulated with the model. These variations between ponds were particularly evident for the Thailand site, and their cause is unknown. Identifying the causes of variation between replicate ponds, and developing models to be able to predict the magnitude of that variation should be a goal for future research efforts.

Even with the limitations noted, the model developed can provide useful information on potential water quality and fish yields. The model can be used with data from different sites, and the results obtained are useful in identifying possible probability distributions for water quality variables and fish growth. The relatively modest data requirements for the weather generation submodel make possible the generation of stochastic weather values for sites for which limited data are available. As a result, the model can be run for potential sites as part of an analysis of possible yields from aquaculture ponds under a variety of treatment options.

## **ANTICIPATED BENEFITS**

The model provides the possible ranges of water quality and fish growth in ponds using statistically generated weather values as

inputs. The weather values are generated based on historical records for a particular site. The variability of water quality and fish yield for short and long terms also can be studied for varying feeding and fertilization regimes, size of fish at stocking and harvesting, pond location, and date of fish stocking and harvesting. The model will be useful in the planning of fish ponds, management of water quality, selection of pond site, and analysis of alternative pond management strategies.

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# PD/A CRSP Seventeenth Annual Technical Report

## Culture of Mixed-Sex Nile Tilapia with Predatory Snakehead

Ninth Work Plan, New Aquaculture Systems/New Species Research 2 (9NS2) Abstract

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## Abstract

An experiment was begun in eighteen 200-m<sup>2</sup> earthen ponds at the Asian Institute of Technology, Thailand, during May and will terminate in October 1999. This experiment will assess the efficiency of snakehead (*Channa striata*) in controlling overpopulation of mixed-sex Nile tilapia (*Oreochromis niloticus*) in ponds. Also, the growth and production characteristics of Nile tilapia in monoculture and polyculture with snakehead will be analyzed. The six treatments were: (A) monoculture of sexreversed tilapia; (B) monoculture of mixed-sex tilapia; (C) polyculture of mixed-six tilapia and snakehead at 20:1 ratio; (E) polyculture of mixed-six tilapia and snakehead at 40:1 ratio; and (F) polyculture of mixed-six tilapia and snakehead at 80:1 ratio. All ponds are fertilized weekly with urea and TSP at rates of 28 kg N and 7 kg P ha<sup>-1</sup> wk<sup>-1</sup>. Sex-reversed all-male and mixed-sex Nile tilapia were stocked at 2 fish m<sup>-2</sup> at sizes of 42.3 ± 1.0 g and 31.0 ± 0.5 g, respectively. Fish growth performance will be evaluated for different treatments. Partial budget analysis will be conducted to estimate input costs and fish value.

Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics/Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]



## ESTUARINE WATER QUALITY MONITORING AND ESTUARINE CARRYING CAPACITY

Eighth Work Plan, Honduras Research 2-1 (8HR2-1) Final Report

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## Abstract

Water quality was monitored in estuaries of the shrimp-producing regions of southern Honduras. This project is a collaborative effort of universities, the private sector, and the public sector, with each group contributing time and resources to the overall effort. The project goal is to provide a scientific basis for estuarine management and sustainable development of shrimp culture in Honduras. Specific objectives are to: a) detect changes in estuarine water quality; b) formulate and validate predictive models for estuarine water quality; and c) estimate assimilative capacity of selected estuaries in the shrimp-producing region of southern Honduras based on water quality, farm chemical budgets, and estuarine fluid dynamics. Samples were collected from October 1996 to October 1998; during 1997-1998 data were collected from 20 sites on 12 estuaries. Nutrient sources for riverine estuaries include nutrient load in river discharge and rainfall or irrigation runoff from the watershed, and shrimp farm discharge. Changes in land-use patterns in the Gulf of Fonseca watershed also will affect estuarine water quality because of changes in runoff patterns and volumes. Water quality in riverine estuaries continues to be influenced directly by seasonal variation in river discharge and watershed runoff, while embayments of the Gulf of Fonseca experience less seasonal variation in water quality. The impact of the 1997-1998 El Niño in Honduras were delayed and reduced rains, which resulted in higher observed salinity, total nitrogen and chlorophyll a concentrations at sampling sites along riverine estuaries in comparison to 1996-1997. Embayment water quality was less affected by the El Niño. No trends for total nitrogen or total phosphorus enrichment were evident in riverine estuaries or embayments during the period 1993–1998. Total nitrogen and total phosphorus concentrations in riverine estuaries were reduced by 10-30% during the rainy season because of river discharge and watershed runoff.

## INTRODUCTION

A long-term water quality monitoring project in estuaries of the shrimp-producing regions of Honduras was initiated in 1993 as part of the Honduras project of the Pond Dynamics/Aquaculture Collaborative Research Support Program (Teichert-Coddington, 1995; Green et al., 1997a). This project is a collaborative effort of universities, the private sector, and the public sector, with each group contributing time and resources to the overall effort. The goal of this monitoring effort is to provide a scientific basis for estuarine management and sustainable development of shrimp culture in Honduras. Specific objectives of the water quality monitoring are to: a) detect changes in estuarine water quality over time; b) formulate and validate predictive models for estuarine water quality; and c) estimate assimilative capacity of selected estuaries in the shrimp-producing region of southern Honduras based on water quality, farm chemical budgets, and estuarine fluid dynamics. Since 1993 the project has generated a continuous, long-term, systematic database on estuarine water quality in shrimp-producing areas of southern Honduras.

Estuarine water quality was monitored at 13 sites on 6 different estuaries when the project began in 1993. During 1998, 20 sites on 12 estuaries were monitored. The number of sites sampled has varied from 13 to 20 on 6 to 12 estuaries. Variation in participation is attributed in part to farms closing for the dry season, farms going out of business, change of farm ownership, change in managers or technical staff responsible for collection and delivery of water samples to the lab, logistical difficulties (e.g., no transport available) or distraction caused by crisis situations on farm. There is an ongoing effort to maintain continuous participation in the project and to incorporate additional farms.

This report summarizes results of water quality monitoring conducted under the PD/A CRSP Eighth Work Plan. Hurricane Mitch made landfall in Honduras at the end of October 1998 and resulted in massive flooding and losses to shrimp farmers (Green, 1999). As a consequence, estuarine water quality monitoring was suspended. Modeling work on assimilative capacity of selected estuaries will be reported separately (Ward, 2000).

## MATERIALS AND METHODS

Estuarine water samples were collected from pump discharge on individual farms within plus-or-minus one hour of high tide. It is assumed that the water samples collected represented a mixed water column sample of the estuary at the pump station because of the superficial vortex caused by the 60- to 90-cm-diameter pump intakes, which are located near the bottom of the estuary. Samples were placed on ice and transported to the water quality laboratory where analysis began within 12 hours of collection. The Choluteca River also was sampled weekly at La Lujosa, which is located downstream from the city of Choluteca and upstream from tidal influence.

Samples were analyzed for total settleable solids (APHA, 1985), nitrate-nitrogen by cadmium reduction to nitrite (Parsons et al., 1992), total ammonia-nitrogen (Parsons et al., 1992), filterable reactive phosphorus (Grasshoff et al., 1983), chlorophyll *a* (Parsons et al., 1992), total alkalinity by titration to pH 4.5 endpoint, salinity and BOD<sub>2</sub> (APHA, 1985), and reactive silicate (Strickland and Parsons, 1977). Total nitrogen and total phosphorus are determined by nitrate and phosphate analysis, respectively, after simultaneous persulfate oxidation in a strong base (Grasshoff et al., 1983).

Data collected from October 1996 through October 1998 were tabulated by sampling site. Time-series graphs (1993 through 1998) of total nitrogen and total phosphorus concentrations were made using data from El Pedregal estuary (illustrative of riverine estuaries) and embayment estuaries (pooled data).

## RESULTS

Results of water quality analyses by site are summarized in Table 1. Water quality in riverine estuaries is influenced directly by seasonal variation in river discharge and watershed runoff, while embayments of the Gulf of Fonseca experience less seasonal variation in water quality. Nutrient concentrations in riverine estuaries follow a cyclical trend that is controlled by season with higher concentrations of total nitrogen and total phosphorus occurring during the dry season and lower concentrations occurring during the rainy season. Rains in southern Honduras generally begin in May, remain strong through June, taper off during July and August, and resume during September and October. However, the effects of the 1997–1998 El Niño in Honduras were delayed and reduced rains. As a result, observed salinity, total nitrogen, and chlorophyll *a* concentrations were higher at sampling sites along riverine estuaries in comparison to 1996–1997 (see Green et al., 1998). Nutrient concentrations in embayments were not affected noticeably by the El Niño.

During the rainy season heavy watershed runoff and river discharge quickly flush out riverine estuaries and reduce salinity to zero or nearly zero parts per thousand, while embayment salinities drop only moderately. Concentrations of other nutrients in riverine estuaries decrease, but not to the degree observed with salinity because of the nutrient load carried by the increased river discharge and watershed runoff. Nutrient concentrations in embayments were lower and less affected by season than in riverine estuaries; data from 1996–1997 are shown to illustrate effect of season on water quality (Figure 1).

No trends for long-term total nitrogen or total phosphorus enrichment were evident in the El Pedregal estuary or embayments of the Gulf of Fonseca (Figures 2 and 3). Data from all



Figure 1. Comparison of median total nitrogen and total phosphorus concentrations in embayments (pooled data) and a riverine estuary, El Pedregal, using box plots. Data are from June 1997 to June 1998. Mid-June through mid-December was classified as the rainy season. The notches represent the 95% confidence limits around the median.

embayment estuaries are pooled because of the small number of sampling sites. Trends in nutrient concentrations in other riverine estuaries are similar to those shown in Figures 2 and 3.

#### DISCUSSION

Water quality in riverine estuaries is related to season. Global climatic events such as El Niño, which provoked drought conditions in Honduras, exacerbate water quality decline in riverine estuaries of the Gulf of Fonseca. In normal years, seasonal rains increase river discharge and watershed runoff, which serve to dilute nutrient concentrations in riverine estuaries. While salinity in riverine estuaries may be reduced to near zero during the rainy season because of massive freshwater inflow, total nitrogen and total phosphorus concentrations decrease only by 10 to 30% because of nutrient load in inflow. Changes in land-use patterns in the Gulf of Fonseca watershed also will affect estuarine water quality because of changes in runoff patterns and volumes. Catastrophic climatic events, such as tropical storm Mitch, provoked massive flooding in the estuarine areas of the Gulf of Fonseca. In addition, two important rivers, the Choluteca and Negro Rivers, experienced changes in their courses as a result of tropical storm Mitch. Reduction of river flow in the pre-Mitch channels reportedly was moderate to severe. If these two rivers do not return completely to their pre-Mitch channels during the next rainy season, it is possible that water quality in these



Figure 2. Mean total nitrogen concentration in the riverine El Pedregal estuary and embayments of the Gulf of Fonseca from 1993 to 1998. Total nitrogen concentration trends in other monitored riverine estuaries were similar to El Pedregal estuary. Solid horizontal line in each graph is the grand mean concentration during this period.

riverine estuaries will be negatively impacted, as could shrimp farms located along these riverine estuaries.

Nutrient concentrations in riverine estuaries increase during the dry season because of evaporation and evapotranspiration, reduced river discharge, the absence of watershed runoff, and shrimp farm discharge. Although river discharge drops dramatically and nutrient concentrations increase significantly during the dry season, total nutrient discharge by rivers is significantly lower during the dry season. In fact, river discharge can drop to zero during very dry years. Water quality in the upper reaches of riverine estuaries deteriorates during the dry season, making maintenance of water quality very difficult on farms located in these regions. The deterioration in water quality in riverine estuaries during the dry season is exacerbated because water exchange with the Gulf of Fonseca decreases rapidly with distance upstream (Teichert-Coddington, 1995). Some farms located in the upper reaches of estuaries close during the dry season, probably because of slow growth attributed to lower water temperatures (Teichert-Coddington et al., 1994) and impaired water quality.

Water quality in embayments is less variable because embayments have better exchange with the Gulf of Fonseca, which is low in nutrients. In addition, the Gulf of Fonseca has a high tidal range (1.5 to 3.5 m) which promotes water exchange and nutrient dilution with the Pacific Ocean. River discharge and watershed runoff result in lower salinities, but not as low as those observed in riverine estuaries. Mean total nitrogen and total phosphorus concentrations increase from 11 to 25% during the rainy season because of nutrient load in river discharge and watershed runoff.



Figure 3. Mean total phosphorus concentration in the riverine El Pedregal estuary and embayments of the Gulf of Fonseca from 1993 to 1998. Total phosphorus concentration trends in other monitored riverine estuaries were similar to El Pedregal estuary. Solid horizontal line in each graph is the grand mean concentration during this period.

# Table 1.Summary of estuarine water quality at shrimp farm pump station sites in southern Honduras from October 1996 to October 1998. Sites<br/>are labeled as "riverine" or "embayment" based on whether or not a river discharges directly into the estuary. Reactive silicate analyses<br/>began January 1998.

Variable	Oc	tober 1996-	-October 1	.997	October 1997–October 1998			
	Mean	SD	Min	Max	Mean	SD	Min	Max
Aquacui tura Fonseca - Riverine								
Salinity (ppt)	13.75	12.12	0.50	39.00	18.12	13.18	0.40	48.00
Total Ammonia N (mg l <sup>-1</sup> )	0.25	0.12	0.01	0.51	0.19	0.11	0.00	0.41
I otal Nitrogen (mg I <sup>-</sup> )	1.31	0.50	0.24	2.29	1.46	0.47	0.03	2.19
Total Phosphorus (mg $1^{-1}$ )	0.23	0.24	0.02	0.79	0.15	0.10	0.00	0.74
Soluble Reactive Phosphate (mg 1 <sup>-1</sup> )	0.14	0.06	0.04	0.25	0.12	0.06	0.04	0.22
Total Alkalinity (mg l <sup>-1</sup> )	123.80	45.28	39.20	181.26	130.41	49.79	41.62	202.98
Chlorophyll $a$ (mg m <sup>-3</sup> )	27.61	14.13	4.51	61.05	36.20	24.04	0.00	92.61
$BOD_2 (mg l^{-1})$	2.74	1.42	0.00	6.40	2.61	1.15	0.70	5.90
Settleable Solids (mg l <sup>-1</sup> )	1.45	2.33	0.00	12.80	0.52	0.63	0.00	2.50
Reactive Silicate (mg l <sup>-1</sup> )					4.69	1.71	0.85	9.65
Acuacultivos #1 - Riverine								
Salinity (ppt)	14.53	12.14	0.00	34.00	16.59	11.97	0.10	41.00
Total Ammonia N (mg l <sup>-1</sup> )	0.12	0.80	0.00	0.38	0.13	0.10	0.02	0.46
Total Nitrogen (mg l <sup>-1</sup> )	1.06	0.40	0.32	2.21	1.17	0.39	0.48	2.19
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.38	0.26	0.02	1.26	0.34	0.32	0.01	1.80
Total Phosphorus (mg l <sup>-1</sup> )	0.17	0.06	0.09	0.36	0.20	0.05	0.08	0.30
Soluble Reactive Phosphate (mg I <sup>-1</sup> )	0.12	0.04	0.05	0.26	0.11	0.04	0.04	0.21
$\frac{10}{10} \frac{10}{10} 10$	20.28	39.05 16.77	0.00	197.01	199.25	41.03	2.00	198.65
BOD <sub>2</sub> (mg $1^{-1}$ )	20.28	1 02	2.41	5 40	1 91	0.77	2.00	4 40
Settleable Solids (mg $1^{-1}$ )	1.82	3.06	0.05	16.00	1.10	1.54	0.00	9.00
Reactive Silicate (mg $l^{-1}$ )					5.24	2.84	1.62	11.77
Acuacultivos #2 - Riverine								
Salinity (ppt)	1.80	4.46	0.00	20.50	3.76	9.74	0.00	36.50
Total Ammonia N (mg l <sup>-1</sup> )	0.06	0.07	0.01	0.30	0.07	0.09	0.01	0.54
Total Nitrogen (mg $l^{-1}$ )	0.78	0.52	0.24	2.55	1.07	0.58	0.18	2.82
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.24	0.34	0.00	1.22	0.46	0.58	0.00	2.30
Total Phosphorus (mg l <sup>-1</sup> )	0.22	0.08	0.11	0.41	0.25	0.06	0.14	0.38
Soluble Reactive Phosphate (mg $l^{-1}$ )	0.18	0.06	0.04	0.32	0.16	0.07	0.04	0.33
Total Alkalinity (mg $\Gamma$ )	129.05	43.93	46.00	217.50	125.65	66.31	41.62	300.72
Chlorophyll $a$ (mg m°) BOD (mg 1 <sup>-1</sup> )	24.44	27.48	0.00	165.44 5.10	31.76	36.19	0.00	125.80
$DOD_2$ (Ing I) Settleable Solids (mg l <sup>-1</sup> )	2.31	1.57	0.00	5.10 2.80	2.30	1.27	0.60	5.60 12.00
Reactive Silicate (mg $\Gamma$ )	0.01	0.40	0.00	2.00	8.78	4.25	1.36	21.50
BIOMAR - RIVERINE								
Salinity (ppt)	21.88	9.74	0.50	34.50	23.56	10.66	1.90	46.00
Total Ammonia N (mg l <sup>-1</sup> )	0.04	0.04	0.00	0.15	0.07	0.06	0.01	0.33
Total Nitrogen (mg l <sup>-1</sup> )	0.63	0.17	0.27	1.13	0.86	0.38	0.42	2.29
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.20	0.12	0.00	0.41	0.18	0.10	0.00	0.41
Total Phosphorus (mg l <sup>-1</sup> )	0.14	0.06	0.06	0.38	0.20	0.18	0.06	1.30
Soluble Reactive Phosphate (mg $l^{-1}$ )	0.08	0.03	0.00	0.16	0.09	0.04	0.04	0.24
Total Alkalinity (mg $1^{-3}$ )	113.46	25.95 14 22	29.92	150.48	120.99	28.86 18.04	52.38	177.80
BOD <sub>2</sub> (mg $l^{-1}$ )	0.08	0.56	∠.37 0.00	2 35	20.23 1 87	10.94	0.00	7 35
Settleable Solids (mg l <sup>-1</sup> )	0.66	1.35	0.00	7.00	0.64	1.20	0.00	6.00
Reactive Silicate (mg l <sup>-1</sup> )					3.54	2.19	0.00	8.81
č								

Variable	Oc	tober 1996–	October 19	997	Oc	tober 1997	–October 1	oer 1998		
	Mean	SD	Min	Max	Mean	SD	Min	Max		
CADELPA - RIVERINE										
Salinity (ppt)	14.82	11.93	0.00	39.00	18.30	15.16	0.30	51.00		
Total Ammonia N (mg l <sup>-1</sup> )	0.22	0.12	0.03	0.50	0.14	0.10	0.01	0.40		
Total Nitrogen (mg l <sup>-1</sup> )	1.30	0.51	0.00	2.23	1.62	0.60	0.64	3.32		
Nitrates + Nitrites (mg $l^{-1}$ )	0.20	0.22	0.00	0.78	0.07	0.11	0.00	0.47		
Total Phosphorus (mg $l^{-1}$ )	0.21	0.05	0.09	0.33	0.27	0.11	0.14	0.64		
Soluble Reactive Phosphate (mg I <sup>-1</sup> )	0.15	0.06	0.00	0.29	0.14	0.08	0.02	0.41		
$\frac{1}{2} \int \frac{1}{2} \int \frac{1}$	130.99	39.85	53.93	182.16	131.07 51.46	53.58 26.08	31.47	215.11		
BOD $(mg^{1-1})$	3 15	27.09	0.11	119.04 8.70	3 54	30.00 2.04	0.10	104.24 9.35		
Settleable Solids (mg $l^{-1}$ )	0.40	0.61	0.00	3.50	0.37	0.42	0.00	2.00		
Reactive Silicate (mg l <sup>-1</sup> )	0.10	0.01	0.00	0.00	4.97	1.83	0.95	9.45		
CRIMASA - RIVERINE										
Salinity (ppt)	23.23	11.74	1.50	47.00	21.46	11.88	1.90	46.00		
Total Ammonia N (mg l <sup>-1</sup> )	0.10	0.09	0.00	0.31	0.11	0.08	0.02	0.29		
Total Nitrogen (mg $l^{-1}$ )	1.07	0.35	0.56	2.15	1.09	0.28	0.71	1.89		
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.23	0.18	0.00	0.64	0.18	0.12	0.01	0.45		
Total Phosphorus (mg l <sup>-1</sup> )	0.19	0.05	0.08	0.29	0.18	0.05	0.12	0.28		
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.13	0.05	0.04	0.23	0.10	0.04	0.04	0.18		
Total Alkalinity (mg $l^{-1}$ )	129.06	28.35	57.90	174.58	120.90	29.89	67.90	182.58		
Chlorophyll $a (mg m^3)$	23.40	29.73	0.00	124.32	26.36	18.18	2.95	85.72		
$BOD_2 (mg 1^2)$	1.81	1.03	0.00	5.70	1.80	0.81	0.20	4.55		
Reactive Silicate (mg l <sup>-1</sup> )	0.04	0.71	0.00	5.00	0.48 4.29	0.37 1.91	2.39	2.00 9.35		
Culcamar - Embayment										
	07.01	( 01	0.00	25.00	25 50	1.05	01 50	07.00		
Salinity (ppt)	27.01	6.31	0.00	35.00	25.58	1.35	21.50	27.00		
Total Ammonia N (mg $1^{-1}$ )	0.01	0.01	0.00	0.05	0.02	0.02	0.00	0.04		
Nitrates + Nitrites (mg $l^{-1}$ )	0.49	0.00	0.14	4.11	0.57	0.12	0.20	0.07		
Total Phosphorus (mg l <sup>-1</sup> )	0.01	0.01	0.00	0.05	0.01	0.01	0.00	0.02		
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.00	0.00	0.00	0.01	0.02	0.07	0.00	0.30		
Total Alkalinity (mg l <sup>-1</sup> )	108.22	11.34	85.88	124.97	119.77	9.76	103.79	133.62		
Chlorophyll $a (mg m^{-3})$	8.03	7.62	2.05	43.50	11.73	13.17	1.70	41.78		
$BOD_2 (mg l^{-1})$	1.04	0.63	0.00	2.60	1.27	0.58	0.30	2.50		
Settleable Solids (mg l <sup>-1</sup> )	0.01	0.02	0.00	0.10	0.00	0.00	0.00	0.00		
Reactive Silicate (mg l <sup>-1</sup> )					1.47	1.30	0.27	4.26		
Cumar - Riverine										
Salinity (ppt)	7.10	6.81	0.50	20.00	16.12	14.88	0.10	51.00		
Total Ammonia N (mg l <sup>-1</sup> )	0.15	0.11	0.02	0.34	0.12	0.09	0.01	0.44		
Total Nitrogen (mg l <sup>-1</sup> )	0.61	0.61	0.00	1.67	1.30	0.48	0.62	3.14		
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.23	0.13	0.11	0.53	0.21	0.13	0.00	0.53		
Total Phosphorus (mg l <sup>-1</sup> )	0.20	0.07	0.12	0.37	0.25	0.13	0.14	0.98		
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.13	0.04	0.09	0.23	0.15	0.07	0.05	0.34		
Total Alkalinity (mg $\Gamma^{-1}$ )	108.37	20.32	84.45	138.38	130.03	50.50	50.44	222.36		
Chiorophyli <i>a</i> (mg m <sup><math>\circ</math></sup> )	25.26	16.60	4.65	47.86	31.11	24.26	0.00	106.80 E 40		
$DUD_2 (\text{mg I})$ Sattlashla Solida (mg $1^{-1}$ )	2.52 0.82	0.97	1.40	4.33	2.34 0.66	1.04	1.05	5.40 4.00		
Reactive Silicate (mg $1^{-1}$ )	0.05	1.70	0.10	5.50	5 37	2.09	2.00	4.00 10.90		
reactive official (fing 1 )					0.07	2.20	2.20	10.70		

Variable	October 1996–October 1997			October 1997–October 1998				
	Mean	SD	Min	Max	Mean	SD	Min	Max
El Faro - Riverine								
Salinity (ppt)	16.55	13.46	0.00	49.00	22.34	15.08	0.20	54.00
Total Ammonia N (mg l <sup>-1</sup> )	0.18	0.12	0.02	0.60	0.12	0.09	0.01	0.42
Total Nitrogen (mg $l^{-1}$ )	1.15	0.47	0.00	2.24	1.55	0.59	0.67	2.95
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.26	0.22	0.00	0.79	0.12	0.19	0.00	0.95
Total Phosphorus (mg l <sup>-1</sup> )	0.23	0.08	0.08	0.48	0.32	0.12	0.14	0.65
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.18	0.07	0.08	0.42	0.19	0.09	0.04	0.38
Total Alkalinity (mg $l^{-1}$ )	141.01	48.63	0.00	223.24	169.80	63.10	60.14	280.50
Chlorophyll $a$ (mg m <sup>-3</sup> )	15.50	15.01	0.00	66.63	56.91	46.99	0.00	177.72
$BOD_2 (mg l^{-1})$	2.14	1.35	0.00	6.30	2.84	1.44	0.70	5.30
Settleable Solids (mg l <sup>-1</sup> )	4.10	8.72	0.00	32.00	2.09	4.71	0.00	24.00
Reactive Silicate (mg l <sup>-1</sup> )					5.26	1.90	0.00	10.46
Finca Sur #1 - Riverine								
Salinity (ppt)	9.37	7.94	0.00	25.00	17.22	11.90	1.10	44.00
Total Ammonia N (mg l <sup>-1</sup> )	0.09	0.11	0.01	0.42	0.08	0.11	0.01	0.56
Total Nitrogen (mg l <sup>-1</sup> )	1.38	0.83	0.43	3.59	1.81	0.67	0.74	3.65
Nitrates + Nitrites (mg $l^{-1}$ )	0.05	0.11	0.00	0.37	0.04	0.08	0.00	0.25
Total Phosphorus (mg l <sup>-1</sup> )	0.26	0.09	0.10	0.44	0.35	0.09	0.18	0.51
Soluble Reactive Phosphate (mg $l^{-1}$ )	0.13	0.06	0.03	0.29	0.15	0.08	0.03	0.29
Total Alkalinity (mg $l^{-1}$ )	123.51	39.76	51.94	216.00	173.31	60.76	76.31	280.50
Chlorophyll $a (mg m^{-3})$	60.20	42.15	6.01	124.75	79.70	60.89	2.01	197.03
$BOD_{2}$ (mg $l^{-1}$ )	3.99	2.15	0.00	8.70	4.01	1.99	1.45	7.90
Settleable Solids (mg l <sup>-1</sup> )	1.51	4.65	0.00	22.00	0.70	1.37	0.00	6.50
Reactive Silicate (mg l <sup>-1</sup> )					5.30	1.95	1.20	9.35
FINCA SUR #2 - RIVERINE								
Salinity (ppt)	10.10	8.53	0.50	27.50	16.91	11.12	0.80	47.00
Total Ammonia N (mg l <sup>-1</sup> )	0.07	0.09	.00	0.36	0.08	0.08	0.01	0.31
Total Nitrogen (mg $l^{-1}$ )	1.39	0.78	0.00	2.84	1.76	0.58	0.66	3.16
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.02	0.04	0.00	0.12	0.03	0.09	0.00	0.48
Total Phosphorus (mg l <sup>-1</sup> )	0.29	0.08	0.12	0.45	0.34	0.09	0.12	0.54
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.14	0.08	0.00	0.31	0.15	0.07	0.04	0.27
Total Alkalinity (mg $l^{-1}$ )	122.47	29.24	80.36	177.00	173.86	60.08	84.39	277.44
Chlorophyll $a (mg m^{-3})$	68.24	32.14	24.31	147.62	66.61	46.50	0.94	205.63
$BOD_2 (mg l^{-1})$	4.49	1.86	0.00	8.30	3.70	1.71	0.80	6.95
Settleable Solids (mg l <sup>-1</sup> )	0.17	0.23	0.00	1.00	0.20	0.23	0.00	0.90
Reactive Silicate (mg l <sup>-1</sup> )					4.96	1.47	3.07	8.44
GMSB #1 - Riverine								
Salinity (ppt)	17.10	10.95	0.00	33.50	17.75	10.74	1.00	38.50
Total Ammonia N (mg l <sup>-1</sup> )	0.12	0.08	0.00	0.28	0.13	0.07	0.03	0.32
Total Nitrogen (mg $l^{-1}$ )	0.96	0.36	0.50	1.88	1.10	0.31	0.60	1.90
Nitrates + Nitrites (mg $l^{-1}$ )	0.29	0.15	0.10	0.67	0.29	0.21	0.01	1.08
Total Phosphorus (mg l <sup>-1</sup> )	0.16	0.05	0.04	0.32	0.19	0.04	0.12	0.26
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.11	0.03	0.03	0.16	0.10	0.04	0.04	0.23
Total Alkalinity (mg l <sup>-1</sup> )	113.71	32.59	25.00	156.42	118.96	37.45	50.44	210.12
Chlorophyll $a$ (mg m <sup>-3</sup> )	19.46	16.01	2.57	80.59	25.94	18.99	0.00	77.38
$BOD_2 (mg l^{-1})$	1.49	0.78	0.00	3.75	1.73	0.72	0.65	3.30
Settleable Solids (mg l <sup>-1</sup> )	1.12	1.64	0.00	8.00	0.56	0.78	0.00	3.50
Reactive Silicate (mg l <sup>-1</sup> )					4.73	2.20	1.79	9.24

Variable	October 1996–October 1997			997	October 1997–October 1998			
	Mean	SD	Min	Max	Mean	SD	Min	Max
GMSB #1 - Riverine								
Salinity (ppt)	19.04	11.40	0.00	37.00	20.29	11.34	0.20	42.00
Total Ammonia N (mg l <sup>-1</sup> )	0.40	2.20	0.00	14.48	0.10	0.06	0.01	0.24
Total Nitrogen (mg l <sup>-1</sup> )	0.91	0.28	0.45	1.79	1.06	0.57	0.29	4.08
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.34	0.18	0.10	0.94	0.24	0.11	0.00	0.42
Total Phosphorus (mg l <sup>-1</sup> )	0.17	0.05	0.03	0.28	0.19	0.05	0.10	0.34
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.12	0.03	0.07	0.18	0.12	0.05	0.01	0.27
Total Alkalinity (mg l <sup>-1</sup> )	122.57	28.46	52.00	160.27	125.42	36.64	56.26	225.42
Chlorophyll $a (mg  m^{-3})$	14.11	14.53	0.00	72.45	19.76	14.60	0.00	57.82
$BOD_2 (mg l^{-1})$	1.38	0.81	0.00	5.25	1.71	0.87	0.35	5.05
Settleable Solids (mg l <sup>-1</sup> )	0.48	1.01	0.00	5.00	0.43	0.56	0.00	2.50
Reactive Silicate (mg l <sup>-1</sup> )					4.51	2.58	1.87	10.90
La Jagua - Riverine								
Salinity (ppt)	14.33	11.51	0.00	34.00	15.23	12.52	0.10	42.00
Total Ammonia N (mg l <sup>-1</sup> )	0.10	0.09	0.00	0.34	0.15	0.11	0.02	0.48
Total Nitrogen (mg l <sup>-1</sup> )	1.01	0.41	0.00	2.21	1.35	0.40	0.70	2.35
Nitrates + Nitrites (mg $l^{-1}$ )	0.38	0.20	0.02	1.01	0.35	0.35	0.05	1.76
Total Phosphorus (mg l <sup>-1</sup> )	0.18	0.04	0.11	0.28	0.21	0.04	0.13	0.35
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.14	0.05	0.07	0.40	0.11	0.04	0.00	0.18
Total Alkalinity (mg $l^{-1}$ )	119.56	40.44	0.00	171.72	122.00	43.93	50.88	196.45
Chlorophyll $a (mg m^{-3})$	25.11	19.50	2.45	83.15	35.86	24.46	0.00	99.57
$BOD_2 (mg l^{-1})$	2.37	1.46	0.00	7.50	2.62	1.46	0.00	8.00
Settleable Solids (mg l <sup>-1</sup> )	1.99	2.96	0.00	14.00	1.35	1.89	0.00	8.50
Reactive Silicate (mg l <sup>-1</sup> )					5.84	2.74	2.07	11.33
Las Arenas - Embayment								
Salinity (ppt)	26.97	5.20	9.00	34.00	27.61	4.15	18.80	35.50
Total Ammonia N (mg l <sup>-1</sup> )	0.03	0.04	0.00	0.18	0.04	0.03	0.00	0.11
Total Nitrogen (mg l-1)	0.30	0.11	0.00	0.54	0.37	0.19	0.16	0.98
Nitrates + Nitrites (mg $l^{-1}$ )	0.03	0.04	0.00	0.26	0.02	0.02	0.00	0.06
Total Phosphorus (mg l <sup>-1</sup> )	0.02	0.01	0.00	0.08	0.03	0.03	0.01	0.20
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.00	0.01	0.00	0.04	0.01	0.02	0.00	0.16
Total Alkalinity (mg l <sup>-1</sup> )	105.43	12.79	58.87	122.76	111.12	13.15	80.19	139.74
Chlorophyll $a (mg m^{-3})$	3.54	1.77	0.00	7.46	4.72	2.90	0.00	20.92
$BOD_2 (mg l^{-1})$	0.87	0.35	0.10	2.25	0.98	0.77	0.00	3.50
Settleable Solids (mg l <sup>-1</sup> )	0.01	0.05	0.00	0.30	0.00	0.01	0.00	0.05
Reactive Silicate (mg l <sup>-1</sup> )					1.42	2.16	0.25	12.06
Lorette #1 - Embayment								
Salinity (ppt)	24.50	5.32	9.50	33.00	24.16	6.33	2.90	39.50
Total Ammonia N (mg l <sup>-1</sup> )	0.04	0.04	0.00	0.10	0.04	0.04	0.00	0.14
Total Nitrogen (mg l <sup>-1</sup> )	0.46	0.26	0.00	0.90	0.63	0.32	0.27	1.44
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.03	0.05	0.00	0.15	0.01	0.02	0.00	0.08
Total Phosphorus (mg l <sup>-1</sup> )	0.07	0.04	0.01	0.18	0.08	0.03	0.04	0.16
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.04	0.05	0.00	0.14	0.01	0.02	0.00	0.07
Total Alkalinity (mg l <sup>-1</sup> )	130.42	17.28	99.72	169.00	128.57	17.69	93.12	169.32
Chlorophyll $a (mg  m^{-3})$	9.24	8.55	0.00	28.73	15.58	15.59	0.00	71.11
$BOD_2 (mg l^{-1})$	2.04	1.33	0.65	6.05	1.69	1.01	0.20	4.60
Settleable Solids (mg $l^{-1}$ )	0.06	0.06	0.00	0.20	0.05	0.05	0.00	0.20
Keactive Silicate (mg I <sup>-1</sup> )					3.12	1.40	1.61	6.28

Variable	October 1996–October 1997			October 1997–October 1998				
	Mean	SD	Min	Max	Mean	SD	Min	Max
Lorette #2 - Embayment								
Salinity (ppt)	23.09	5.31	10.00	32.50	23.91	5.54	15.00	39.50
Total Ammonia N (mg l <sup>-1</sup> )	0.03	0.03	0.00	0.08	0.03	0.02	0.00	0.10
Total Nitrogen (mg l <sup>-1</sup> )	0.49	0.25	0.25	1.25	0.56	0.32	0.23	1.73
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.04	0.05	0.00	0.16	0.02	0.03	0.00	0.16
Total Phosphorus (mg l <sup>-1</sup> )	0.05	0.04	0.02	0.18	0.06	0.03	0.03	0.16
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.02	0.03	0.00	0.14	0.01	0.01	0.00	0.05
Total Alkalinity (mg l <sup>-1</sup> )	110.04	11.91	91.58	130.70	113.86	20.82	77.60	159.14
Chlorophyll $a (mg m^{-3})$	15.44	36.21	0.00	150.00	18.15	29.01	0.03	150.00
$BOD_2 (mg l^{-1})$	2.08	1.73	0.70	7.00	1.71	1.12	0.40	4.40
Settleable Solids (mg l <sup>-1</sup> )	0.18	0.42	0.00	1.50	0.15	0.33	0.00	1.50
Reactive Silicate (mg l <sup>-1</sup> )					2.49	1.14	1.18	5.04
CHOLUTECA RIVER AT LA LUJOSA								
Salinity (ppt)	0.00	0.00	0.00	0.00	0.02	0.05	0.00	0.20
Total Ammonia N (mg l <sup>-1</sup> )	0.04	0.06	0.00	0.40	0.05	0.03	0.00	0.12
Total Nitrogen (mg l <sup>-1</sup> )	0.83	0.60	0.31	4.23	1.05	0.55	0.38	2.98
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.30	0.44	0.00	2.51	0.53	0.49	0.00	2.16
Total Phosphorus (mg l <sup>-1</sup> )	0.26	0.13	0.10	0.63	0.25	0.09	0.11	0.43
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.20	0.09	0.10	0.41	0.19	0.09	0.06	0.35
Total Alkalinity (mg $l^{-1}$ )	113.80	34.93	43.75	166.00	109.57	42.14	43.75	183.28
Chlorophyll $a (mg m^{-3})$	25.08	20.31	0.00	79.98	13.54	15.66	0.00	92.03
$BOD_2 (mg l^{-1})$	2.39	1.51	0.00	8.25	1.87	0.78	0.60	3.60
Settleable Solids (mg l <sup>-1</sup> )	0.51	1.45	0.00	9.40	0.30	0.51	0.00	2.00
Reactive Silicate (mg l <sup>-1</sup> )					11.47	5.45	0.93	27.48
Sea Farms #1 - Embayment								
Salinity (ppt)	28.20	3.77	16.50	35.50	27.78	4.05	15.20	34.50
Total Ammonia N (mg l <sup>-1</sup> )	0.04	0.04	0.00	0.12	0.05	0.03	0.00	0.17
Total Nitrogen (mg l <sup>-1</sup> )	0.27	0.08	0.11	0.44	0.31	0.12	0.17	0.84
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.02	0.01	0.00	0.06	0.02	0.03	0.00	0.21
Total Phosphorus (mg l <sup>-1</sup> )	0.05	0.02	0.00	0.13	0.05	0.02	0.02	0.13
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.03	0.02	0.00	0.10	0.03	0.02	0.01	0.11
Total Alkalinity (mg l <sup>-1</sup> )	113.74	7.93	97.00	127.71	115.19	10.88	68.01	135.66
Chlorophyll $a (mg m^{-3})$	4.85	4.55	0.00	25.39	5.43	3.50	0.00	22.53
$BOD_2 (mg l^{-1})$	0.94	0.36	0.00	1.60	0.92	0.68	0.15	4.45
Settleable Solids (mg l <sup>-1</sup> )	0.00	0.01	0.00	0.05	0.02	0.03	0.00	0.10
Reactive Silicate (mg l <sup>-1</sup> )					1.72	0.95	0.13	4.68
Sea Farms #2 - Embayment								
Salinity (ppt)	28.36	2.89	23.00	35.50	28.17	3.57	23.00	35.00
Total Ammonia N (mg l <sup>-1</sup> )	0.03	0.03	0.00	0.13	0.05	0.04	0.00	0.16
Total Nitrogen (mg l <sup>-1</sup> )	0.27	0.08	0.00	0.41	0.33	0.13	0.02	0.84
Nitrates + Nitrites (mg l <sup>-1</sup> )	0.01	0.02	0.00	0.12	0.02	0.02	0.00	0.08
Total Phosphorus (mg l <sup>-1</sup> )	0.03	0.02	0.00	0.11	0.03	0.01	0.01	0.05
Soluble Reactive Phosphate (mg l <sup>-1</sup> )	0.01	0.01	0.00	0.04	0.01	0.01	0.00	0.02
Total Alkalinity (mg $l^{-1}$ )	109.96	7.13	92.64	120.78	110.14	9.86	91.18	130.56
Chlorophyll $a \pmod{m^{-3}}$	4.57	4.75	0.00	28.09	4.44	3.63	0.00	23.74
$BOD_2 (mg l^{-1})$	1.01	0.46	0.00	2.50	1.02	0.67	0.30	4.00
Settleable Solids (mg l <sup>-1</sup> )	0.00	0.02	0.00	0.10	0.01	0.02	0.00	0.10
Reactive Silicate (mg l <sup>-1</sup> )					0.96	1.39	0.24	8.30

Nutrient sources for riverine estuaries include nutrient load in the form of river discharge and rainfall or irrigation runoff from the watershed and shrimp farm discharge. Shrimp farmers must be acutely aware of estuarine water quality, as often the same estuary serves both as the source of water for production ponds and as the repository of production pond effluents. Nutrient concentration in shrimp farm effluents is the only source of estuarine nutrients that can be controlled by the farmer. The principal methods to achieve reduction of shrimp farm effluent nutrient load in Honduras are reducing exogenous nutrient inputs, i.e., feeds and fertilizers into ponds and controlling development both in terms of new pond area and intensification of production systems. Significant progress has been achieved in terms of feed use: feed conversion ratios have decreased from a mean of 3.2 in the early 1990s to 1.5 to 2.0 currently (Teichert-Coddington et al., 1991; Teichert-Coddington and Rodriguez, 1995; Teichert-Coddington et al., 1996; Green et al., 1997b). Results of PD/A CRSP research have demonstrated that feed protein content and daily feed ration can be decreased during the dry season without affecting yield (Teichert-Coddington and Rodriguez, 1995; Green et al., 1997b). Research on chemical fertilizer use and lower protein content diets is being conducted by some farms. Reduced use of exogenous nutrients in shrimp production during the dry season should reduce environmental impact of shrimp farm effluents. The development of assimilative capacity models for selected estuaries will provide information necessary in the formulation of strategies and regulations governing future development of shrimp farming.

## **ANTICIPATED BENEFITS**

The estuarine water quality database serves to track long-term trends in estuarine water quality in shrimp-producing regions of southern Honduras. It also serves to increase awareness among shrimp farmers of their relation to the environment and encourages them to pursue sustainable production strategies. In the aftermath of tropical storm Mitch, it is critical that the water quality monitoring effort be sustained as the shrimp industry is reconstructed given the dramatic effects the storm had on river flow patterns. Unfortunately, this work could not be continued beyond the Eighth Work Plan because of severe budget reductions and the de-emphasis of shrimp production research by the PD/A CRSP. Fortunately, the Honduran National Association of Aquaculturists (ANDAH) assumed full ownership of the program and is committed to continuing the estuarine water quality monitoring program and operation of La Lujosa water quality laboratory. This occurrence demonstrates the significant development impact of the Honduras project of the PD/A CRSP. Data from this study will support development of models of assimilative capacity for selected estuaries. The assimilative capacity models will provide information necessary in the formulation of strategies and regulations governing future development of shrimp farming.

## ACKNOWLEDGMENTS

We thank Jaime Lopez and farm personnel responsible for sample collection and transport to the La Lujosa Lab for their collaboration. This study was made possible by the collaboration of the General Directorate of Fisheries and Aquaculture (DIGEPESCA), Ministry of Agriculture and Livestock, and the Honduran National Association of Aquaculturists (ANDAH).

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Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics/Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]



## EVALUATION OF SHRIMP FARMING IMPACTS IN GOLFO DE FONSECA REGION, HONDURAS

Eighth Work Plan, Honduras Research 2-2 (8HR2-2) Final Report

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## Abstract

An intensive data collection and modeling study has been underway for the past several years addressing two of the channel estuaries draining into the Gulf of Fonseca, namely Estero El Pedregal and Estero San Bernardo. Data have been compiled on the shrimp farm configurations, exchange rates, and effluent chemistry. Temperature/salinity/dissolved oxygen profiles have been measured in the estuary channels in both rainy and dry seasons. Physiographic, hydrographic, and meteorological data have been obtained to supplement the estuary data. This report examines the assimilative capacity of these estuaries with respect to dissolved oxygen (DO). The oxygen demand of organics is measured by biochemical oxygen demand (BOD). Shrimp farm BOD loadings were estimated from effluent data and exchange. A transport model for salinity and DO in the estuaries was applied to predict the tidal-mean, section-mean concentrations of salinity and DO. The model predictions of DO given 1995 BOD loadings were satisfactory. Future loadings based upon full shrimp farm development along these two estuaries were then input to determine the resulting DO under these conditions. It was found that the 1995 configuration is already pressing the carrying capacity of both systems, and the DO will be worsened at full development. Shrimp farms placed farther upstream than about 20 km from the mouth will most likely have excessive impact on the DO in the estuary. The impact is exacerbated under dry season conditions. Negative impacts of a specific farm can be ameliorated by reducing or eliminating pond discharges during the dry season, and by reducing the level of water exchange employed. This work needs to be extended to address additional water-quality parameters and to incorporate larger spatial scales, especially to establish the interaction between different estuaries draining into Fonseca.

## INTRODUCTION

The Gulf of Fonseca, a large estuarine embayment on the Pacific coast of Central America, has become the focus of the shrimp aquaculture industry in Honduras, which represents the third most important export of the country. Much of the industry, representing a total of over 14,000 ha of operational pond area in 1998, is situated along the distributaries of the deltaic region in the eastern arm of Fonseca, designated Monypenny Bay in this report. The phenomenal growth of the shrimp industry in the past decade has raised concerns that the combined effluent from the ponds might accumulate in such concentration as to contaminate the estuary waters as a source of exchange influent. This self-limiting concentration is referred to here as the "carrying capacity" of the system, and corresponds to the concept of "assimilative capacity" in environmental engineering.

While it is tempting to think of "carrying capacity" as a single number, like passenger capacity of a bus, it is important to recognize that it is in fact a function of position, both in terms of the region of the watercourse in which acceptable water quality must be maintained and in terms of the actual locations of the sources of contamination. For this reason, there is not a unique value for carrying capacity, because different distributions in space in combination with different effluent loads can result in the same impact on water quality. On the other hand, if the combined effect of a collection of effluent loads is to reduce water quality below its acceptable value, then it can be said that the carrying capacity of the system has been exceeded.

This study addressed the effect of shrimp aquaculture on the single parameter of dissolved oxygen (DO) in two channel estuaries in the Monypenny Bay region that are the sites for the greatest development of shrimp farming operations in the country, the Estero El Pedregal and the Estero San Bernardo (Figure 1). During the past five years, data collection efforts have been underway in Honduras to quantify pond metabolism, receiving water quality, and basic hydrography of the distributaries of the Monypenny Bay region, especially Pedregal and San Bernardo. This work was accomplished under the aegis of the Auburn University/Honduras PD/A CRSP and involved collaboration among universities, the private sector, and the public sector. A full description of the program is given in Green et al. (1997). Since 1995, annual summaries of estuarine water quality findings have been published (Teichert-Coddington, 1995; Teichert-Coddington et al., 1997; Green et al., 1998; Green et al., 1999).

## HYDROGRAPHY AND HYDROLOGY

The Gulf of Fonseca is a tectonobay (e.g., Ward and Montague, 1996), a flooded coastal indentation formed by faulting and vulcanism. It has a free connection with the Pacific of some 30 km in width and 20 m average depth. The inland reaches of the Gulf of Fonseca exhibit features of a drowned-river-valley type estuary, with extensive mud shoals, and deltaic-like shoal areas, especially its eastern arms. Its coastal physiography consists of tidal flats, tidally flushed mangrove swamps fringing largely unvegetated tidal flats, and low-relief "sweetland" punctuated by steep igneous formations.



Figure 1. Location map of study area.

Riverine inflow affects the hydrography of the estuary by establishing a gradient of salinity across the system and further influences the water quality by its associated influx of constituents of terrestrial origin, including human wasteloads. Several major rivers drain into Monypenny Bay (Figure 1). The primary riverine inflow to this region from Honduras is the Río Choluteca, but there are also several other major rivers mainly from Nicaragua, most prominent of which are the Estero Real, whose watershed extends to Lago de Managua, and the Río Negro, which enters the head of the San Bernardo. Numerous distributaries lace this deltaic region, most carrying freshwater throughflow deriving from local runoff, but each in itself is a channel estuary.

The climatology of Honduras is characterized by two distinct seasons in the year, the dry season and the rainy season. These seasons are keyed to the annual movement and intensity of the Intertropical Convergence Zone. The winter dry season typically extends from November through May, during which the region becomes quite arid. The rainy season, typically extending from May through October, is in fact interrupted in July by a brief dry period, known as the *canícula* in Honduras. This produces a characteristic bimodal shape to the annual pattern of precipitation. Daily precipitation measurements from Marcovia and Yusgüare were compiled for the period 1973–96 and analyzed to provide insight into the flows of ungauged rivers. The estuarine data analyzed in this paper were collected in 1995. The rainfall data demonstrate that the rainy season in 1995 was exceptional, the regional precipitation being the highest in the 1973–96 record. August 1995, in particular, logged the highest rainfall for any month in this entire period, by a substantial factor.

The only one of the inflows for which we have gauged data is the Río Choluteca. The annual pattern of flow in the Río Choluteca, based upon 1979–90 gauged data, is clearly bimodal, with two high-flow seasons, spring and fall, separated by dry seasons of winter and summer. Other inflows to the study area had to be estimated from the precipitation data at Marcovia and Yusgüare, using the approximate drainage

Table 1. Estimated BOD loads for 1995 shrimp farms.

areas of the tributaries and a coefficient of runoff (e.g., Larsen and Concepción, 1998).

Hydrology is dominated by the terrestrial environment. Hydrography, on the other hand, is decidedly marine. Most important is the effect of tides. The effect of the coastal bight and the morphology of the Gulf is to amplify the Pacific tide within the Gulf, so that the mean amplitude is over 3 m in many of the coastal inlets and channel estuaries. Moreover, this is a semidiurnal tide (period 12.4 hours), so the movement of water with the flood and ebb is substantial. Coastal pilots note tidal currents in the Estero Real exceeding 3 knots (e.g., Admiralty, 1951).

## SHRIMP FARMS

Because the field data employed in this analysis were collected mainly in 1995, it was necessary to characterize the state of shrimp farm operation at this time. A concerted effort was made by the CRSP personnel in Honduras to obtain data on pond area, exchange, and management from the various farm operators with installations on the El Pedregal or San Bernardo systems. Details on the physical facilities at each of the farms were compiled by this effort. From the standpoint of determining the impact of shrimp farm operations on receiving water quality, the important data are the flow out of the pond, its oxygen-demanding constituents, and the point at which pond effluent enters the watercourse.

The biochemical oxygen demand (BOD) was employed as a gross measure of the oxygen-demanding potential of the effluent. Samples from both influent and effluent of GMSB were analyzed for ultimate BOD (BODu), as the asymptote of an oxygen depletion series (Teichert-Coddington et al., in prep.), so the incremental BODu added by the pond to that already in the estuary water could be isolated. Detailed data from GMSB were scaled to the other farms in the El Pedregal/San Bernardo study area according to their individual exchange rates. The product of the resulting effluent BODu concentration and the discharge flow (with appropriate units conversions) is the BODu load (mass per unit time), tabulated in Table 1.

Farm	Pond Area	Exchange Throughflow		BODu	BOD Load		
	(ha)	(% d <sup>-1</sup> )	$(Mm^3 d^{-1})$	(mg l <sup>-1</sup> )	(kg d <sup>-1</sup> )	(lb d <sup>-1</sup> )	
Aquacultura Fonseca	682	2	0.136	4	546	1,200	
Aquacultivos de Honduras	600	8	0.480	16	7,680	16,896	
BIMAR	47	10	0.0470	20	940	2,068	
CADELPA	312	3	0.0936	6	562	1,236	
CAYDESA	110	10	0.110	20	2,200	4,840	
CRIMASA	1,068	6	0.641	12	7,690	16,917	
CUMAR	600	7	0.420	14	5,880	12,936	
EXMAR	149	8	0.1192	16	1,907	4,196	
GMSB	1,987	6	1.192	12	14,306	31,474	
HONDU-ESPECIES	357	(Not operating in 1995)					
HONDUFARM	350	10	0.350	20	7,000	15,400	
INMAR	238	12	0.286	24	6,854	15,080	
La Jagua	203	3	0.0609	6	365	804	
PROMASUR	378	6	0.227	12	2,722	5,988	

\* For pond volume calculations, mean pond depth was estimated to be 1.0 m.

## Hydrography and Water Quality of the Channel Estuaries

Hydrography of the channel estuaries was addressed through analysis of field data and application of a tidal hydrodynamic model. The hydrographic influences include channel morphology (bathymetry and cross sections), freshwater inflow, density structure, which is dominated by salinity, and forcing from the Gulf of Fonseca, most importantly tides. In the early work on these estuaries (Ward, 1995), there was virtually no information on channel geometry, so these parameters (depth, width, and cross section area) had to be estimated from inspection of topographic maps and from limited field excursions on the estuaries. The field work of the past five years obtained much better data on estuary depths and widths, much improving the physiographic depiction of the channel estuaries. An equally important feature is the large tidal flats which communicate with the main tidal channel through small scoured tidal passes through the mangrove fringe. These tidal flats have the capacity to store a great amount of water on the rising tide and release that water back to the tidal channel as the tide stage falls. The extent to which the flats are regularly flooded therefore is an important feature of the tidal functioning of this system.

One concern expressed early in this work (e.g., Ward, 1995) was the possibility that installation of shrimp pond levees might reduce the tidal prism and thereby diminish tidal flushing, contributing to degraded water quality in the tidal estuaries. In effect, these shrimp farms could eliminate the tidal flats, hydraulically isolating these areas by enclosure within levees to create their shrimp ponds. This potential was evaluated by determining the extent of regular inundation of the tidal flats before shrimp pond construction and locating as precisely as possible the actual extent of the farm levees, accomplished through examination of topographic maps and satellite photographs and through the efforts of Sr. Felix Wainwright (1996), who has studied these areas extensively. As matters turn out, the practice of most of the shrimp farms of placing the levees inside the mangrove fringe follows approximately the extent of normal tidal inundation, so the impact on tidal prism reduction is limited. The exception is for those farms in the upstream sections of the estuaries, but in these regions the more important control on water quality is the volume and quality of inflows.

The salinity structure is an important aspect of the hydrography of these channel estuaries because it is a direct demonstration of the movement of water in response to tides and because it provides an index to density stratification. Analysis of the 1995 field data on salinity indicate the tidal excursion in the lower reach of the estuaries to be at least 25 km in the Pedregal and 10 km in the San Bernardo. Moreover, within the salinityintrusion zone, there is also substantial vertical stratification, especially on the flooding tide. This is due to the longitudinal salinity gradient created by the freshwater inflow, and to the tidal influx of more saline water from Monypenny Bay. Such dramatic stratification appears to be a phenomenon only of the combination of transient conditions during the high inflow freshet of the rainy season. With the stabilization of inflows in July, the longitudinal salinity gradient mixes out more, and the vertical stratification is much less pronounced.

A tidal hydrodynamic model was applied to the combined Pedregal and La Jagua system and the combined San Bernardo and Berberia system following the formulation of Dronkers (1964). Because of the importance of the tidal flats to the hydrography of this system, special provision for these is made in the numerical treatment (Hauck and Ward, 1980). The area of the tidal flat is an input to the model that must be determined from survey data, in this case from the tidal inundation areas provided by Wainwright (1996). Modeled tidal excursions appear to be on the correct order compared to observations of Currie (1994) and compared to the field observations of tidal excursion in salinity structure analyzed in this project (see above). The most important use of the tidal calculations is to provide a basis for estimating longitudinal dispersion in the transport modeling (see below) based upon tidal excursion and for computing oxygen reaeration, which is a strong function of current speed.

A total of 13 data runs were made on El Pedregal and 10 on the San Bernardo in 1994–96, in both the dry and rainy seasons. Only a portion of the data collected has been analyzed and employed in the present study. The measurements consisted of vertical profiles at 0.5-m intervals from surface to bottom of temperature, dissolved oxygen, and salinity, using an electrometric probe and deck-readout.

A numerical transport model was applied to each of these estuaries to model salinity and DO. The model treats the longitudinal variation of properties, i.e., the tidal-mean, vertical-mean concentrations, and assumes an equilibrium profile, i.e., steady-state, as a balance between transports and kinetics. Each estuary was depicted by a network of computational nodes running up the channel to a point well above the region of shrimp farm development. The objective of the modeling is to determine the DO response to shrimp farm loading. The main purpose of modeling salinity is to provide some assurance that the transport terms are approximately correct in the model. The field data were used to analyze the behavior of the estuaries and to validate the salinity and DO models.

The model application to the Pedregal salinities for July 1995 (rainy season) inflows is shown in Figure 2a. For comparison, the tidal-mean vertical-mean observed salinities are shown also. (The vertical bars represent observed tidal range for that



Figure 2a. Rainy season salinity, modeled and observed, in El Pedregal, July 1995.

sample run.) Generally, the agreement between model and observed salinities is considered satisfactory. Figure 2b shows El Pedregal DO profiles for the same conditions and the corresponding model prediction of DO. The total shrimp-farm BOD load to El Pedregal for 1995 conditions is about 16,000 kg d<sup>-1</sup>. The data show decreasing DOs with distance upstream from the mouth. The model predicts a substantial zone of low oxygen, the combined result of the BOD loads from tributary runoff from the lowlands and the load from the shrimp farms. The relation between the locations of the shrimp farm loads (shown as arrows) and the region of low DO should especially be noted. All of the measured (vertical-mean) DOs upstream from about 13 km from the mouth are below 3 mg l<sup>-1</sup> and some are less than 1 mg l<sup>-1</sup>. The model results substantiate that these low DOs result in part from the BOD loads from the shrimp farms. These two facts indicate that the 1995 shrimp farm development is already approaching the carrying capacity of El Pedregal.

A problem in the modeling of the San Bernardo is the inflow of the Río Negro, which enters the head of the San Bernardo from its watershed in Nicaragua. This appears to be a major source of inflow for San Bernardo; moreover, it is evidently water of good oxygen content, originating in a mountainous basin in Nicaragua. Because we have no data on this inflow, it is a major source of error and had to be indirectly estimated in the present work. The model does a fair job of predicting a DO depression like that depicted in the data, but the location of the low point of the sag is some 5 to 10 km too far up the estuary. We also conclude that the San Bernardo is not as seriously stressed as the Pedregal under these inflow conditions. However, this estuary receives a total BOD load (in 1995) of some 24,000 kg d<sup>-1</sup>, nearly twice that of the Pedregal. It seems certain that it is the extraordinarily high inflows during August 1995 that prevented occurrence of depressed DOs.

## SHRIMP FARM IMPACTS AND CARRYING CAPACITY ANALYSIS



The present model development permits a reliable estimation of the combined impacts of the regional shrimp farms under various scenarios of increased development. Information was compiled on shrimp farm enlargements and new operations as of 1998, the additional pond area that could be developed on

Figure 2b. Rainy season dissolved oxygen, model and observed, in El Pedregal, July 1995.

operating farms, and the concessions capable of future development. This information was used to construct a "full-development" scenario. The drain locations and corresponding loads were implemented in the BOD-DO model using the same 1995 rainy season conditions, so that the 1995 model runs (and associated field data) could be used as a baseline for comparison. The resulting DO profiles under rainy season conditions predicted by the model with these loads are shown in Figure 3a for El Pedregal and Figure 3b for San Bernardo. The result could be anticipated: even lower DOs in the main sag zones. For El Pedregal, this looks particularly crucial, as a reach of some 10 km results with *average* DOs less than 1 mg l<sup>-1</sup>. For the San Bernardo, there is a reach with DOs below 2 mg l<sup>-1</sup>, but with a higher assumed flow in the Río Negro this reach would not occur.

The data and the model suggest that under these rainy season conditions, well-aerated waters are brought into the channel estuaries from the adjacent Gulf of Fonseca on the flooding tide and poorly oxygenated waters enter the estuary channels from the tidal flats and tributaries draining the tidal lowlands. Even if shrimp farms did not exist, degraded water quality would result as a result of the latter intermixing with the former.



Figure 3a. Model prediction of DO in El Pedregal, 1995 rainy season conditions, full shrimp farm development.



Figure 3b.Model prediction of DO in San Bernardo, 1995 rainy season conditions, full shrimp farm development.

## **ANTICIPATED BENEFITS**

For systems such as Estero el Pedregal and San Bernardo, these studies establish that there is a level of development at which the estuary can become so degraded as to prohibit economical aquaculture, i.e., shrimp farming can become self-limiting. The field data and modeling allow quantification of this condition, and, more importantly, provide a tool to evaluate alternative shrimp farm development scenarios to minimize impacts.

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# PD/A CRSP Seventeenth Annual Technical Report

## WATER EXCHANGE TO RECTIFY LOW DISSOLVED OXYGEN

Eighth Work Plan, Honduras Research 4 (8HR4) Final Report

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## Abstract

In Central America semi-intensive shrimp production technology is used by many producers. Semi-intensive production technology is characterized by final stocking rates of 5 to 11 shrimp m<sup>-2</sup>, daily water exchange at  $\leq 10\%$  of pond volume, and use of 20 to 25%-protein feeds. The role of water exchange in semi-intensive shrimp culture is being evaluated in Honduras. A recent study "Influence of daily water exchange volume on water quality and shrimp production" (HR3) indicated that daily or emergency water exchange did not affect significantly shrimp production, but that water quality was better in ponds that received daily water exchange. However, differences in water quality generally did not become pronounced until the latter half of the 12 to 16-wk production cycle. Producers may find unacceptable the risk associated with utilizing an emergency-only water exchange policy. However, it appears that the current standard practice of initiating water exchange beginning the fourth week post-stocking is not the most efficient was exchange strategy. This experiment builds on the previous experiment by investigating the effects of time of initiation of water exchange on early morning dissolved oxygen, water quality, and shrimp production in ponds. The objectives of this experiment are to evaluate the effect of time of initiation of water exchange on pond dissolved oxygen, water quality, and shrimp production. Nine 0.93-ha ponds located on a commercial shrimp farm in southern Honduras were used for this completely randomized design study to test time of initiation of water exchange. Water was exchanged at 10% of pond volume per day, six days per week beginning four, seven, or ten weeks after stocking. The rainyseason experiment was initiated and was to be repeated during the dry season. Ponds for the rainy-season experiment were stocked with hatchery-spawned post-larval (PL) P. vannamei at 145,000 PL ha<sup>-1</sup> (14.5 PL m<sup>-2</sup>) on 15 August 1998. Shrimp were fed six days per week beginning three weeks after stocking. On 30–31 October 1998 the torrential rains of tropical storm Mitch resulted in massive flooding of farms and enormous losses to shrimp farmers in southern Honduras. Data were collected up until the ponds were flooded. Treatment effects on pond water quality appeared to begin to manifest themselves in those treatments where water exchange had been initiated (the four- and seven-week treatments). Shrimp growth appeared to be affected by treatment as shown by the divergence of growth curves in Figure 1, but because there are no harvest data available it is impossible to draw conclusions regarding effects of treatment on shrimp growth and yield. Shrimp farms suffered infrastructural damage and very large economic loss as a result of the flooding caused by tropical storm Mitch. Given this situation it was not possible to repeat the rainy season experiment nor conduct the dry-season experiment.

## INTRODUCTION

Results of previous research on water exchange in shrimp production in Honduras indicated that daily or emergency water exchange did not affect significantly shrimp production, but that water quality was better in ponds that received daily water exchange (Green et al., 1999). However, differences in water quality generally did not become pronounced until the latter half of the 12 to 16-week production cycle. Producers may find unacceptable the risk associated with utilizing an emergency-only water exchange policy. However, it appears that the current standard practice of initiating water exchange beginning the fourth week post-stocking is not the most efficient water exchange strategy. This experiment was designed to build on the previous experiment "Influence of daily water exchange volume on water quality and shrimp production" (Green et al., 1999) by investigating the effects of time of initiation of water exchange on pond early morning dissolved oxygen, water quality, and shrimp production.

## METHODS AND MATERIALS

Nine 0.93-ha (± 0.04 ha SD) ponds located on a commercial shrimp farm on a riverine estuary of the Gulf of Fonseca, Honduras, were used for this completely randomized design study to test three water exchange regimes. Water was exchanged at 10% of pond volume per day, six days per week beginning four, seven, or ten weeks after stocking. No water exchange occurred during the first three weeks of culture. In all water exchanges, water first was discharged and then added to refill ponds.

Ponds for the rainy season experiment were stocked with hatchery spawned post-larval (PL) *P. vannamei* at 145,000 PL ha<sup>-1</sup>

(14.5 PL m<sup>-2</sup>) on 15 August 1998. Stocking of ponds for the dryseason experiment was scheduled for December 1998. A survival rate of 50% was assumed because of Taura Syndrome effects on hatchery-produced larvae. Most of the mortality was assumed during the first month following stocking. Shrimp in the rainy-season experiment were scheduled to be harvested in late November 1998. Dry-season experiment ponds were scheduled to be harvested in March 1999. However, on 30–31 October 1998 the torrential rains of tropical storm Mitch resulted in massive flooding of farms and enormous losses to shrimp farmers in southern Honduras (Green, 1999). Thus, it was impossible to complete this experiment.

Shrimp were fed a 20% protein commercially formulated ration. Shrimp were fed six days per week beginning three weeks after stocking for the rainy-season experiment. Feed rate for all ponds was based on the theoretical feeding curve for *P. vannamei*:

 $Log_{10}Y = -0.899 - 0.56Log_{10}X$ where Y = feed rate as a percent of biomass and<math>X = mean shrimp weight in grams.

Daily feed rate was calculated for individual ponds and then averaged so that all ponds received the same quantity of feed on a daily basis. Feed was offered once daily. Shrimp growth was monitored weekly by cast net samples of each pond's population. Feed rate was adjusted weekly based on shrimp samples.

Water quality variables in each pond were measured weekly in pond and intake water. Intake water was sampled from supply canals, while pond water was sampled by pooling a minimum of six column samples collected at random within the pond. Pond water and replacement water samples were obtained with a column sampler. Water samples were analyzed for pH measured potentiometrically, nitrate-nitrogen by cadmium reduction (Parsons et al., 1992), total ammonia-nitrogen (Parsons et al., 1992), soluble reactive phosphorus (SRP) (Grasshoff et al., 1983), chlorophyll a (Parsons et al., 1992), total alkalinity by titration to pH 4.5 endpoint, salinity, 2-d biochemical oxygen demand (BOD<sub>2</sub>) at 20°C (APHA, 1985), and reactive silicate (Strickland and Parsons, 1977). Total nitrogen and total phosphorus were determined by nitrate and phosphate analysis, respectively, after simultaneous persulfate oxidation (Grasshoff et al., 1983). Dissolved oxygen (DO) concentration and temperature were measured in ponds twice daily (0400 and 1600 h) at 25 cm below the water surface.

## RESULTS

Data were collected up until the ponds were flooded. The data are presented in this report, but the reader is reminded that it is impossible to draw conclusions as the experiment was never harvested. Water exchange had begun in the four- and sevenweek treatments and was about to begin in the ten-week treatment when the hurricane struck. Shrimp growth through week 10 averaged 0.65 g wk<sup>-1</sup> (Figure 1). Total nitrogen (Figure 2), total phosphorus (Figure 3), chlorophyll *a* (Figure 4), and BOD<sub>2</sub> (Figure 5) concentrations in ponds began to increase several weeks after ponds were stocked but then declined in treatments where water exchange had been initiated. Mean water quality variable concentrations for the ten-wk period are shown in Table 1.

## DISCUSSION

Treatment effects on pond water quality seemed to manifest themselves in those treatments where water exchange had been initiated (the four- and seven-week treatments). Water exchange reduced water quality variable concentrations in ponds because inlet water had lower nutrient concentrations and diluted pond water (Table 1). Shrimp growth appeared to be affected by treatment as shown by the divergence of growth curves in Figure 1, but because there are no harvest data



Figure 1. Growth of shrimp in 1-ha earthen ponds during the first ten weeks of the rainy-season experiment in Honduras to test the effects of beginning water exchange four, seven, or ten weeks after stocking.



Figure 2. Mean total nitrogen concentrations in ponds during the first ten weeks of the rainy-season experiment in Honduras to test the effects of beginning water exchange four, seven, or ten weeks after stocking.



Figure 3. Mean total phosphorus concentrations in ponds during the first ten weeks of the rainy-season experiment in Honduras to test the effects of beginning water exchange four, seven, or ten weeks after stocking.

available it is impossible to draw conclusions regarding effects of treatment on shrimp growth and yield.



Figure 4. Mean chlorophyll *a* concentrations in ponds during the first ten weeks of the rainy-season experiment in Honduras to test the effects of beginning water exchange four, seven, or ten weeks after stocking.



Figure 5. Mean two-day biochemical oxygen demand in ponds during the first ten weeks of the rainy-season experiment in Honduras to test the effects of beginning water exchange four, seven, or ten weeks after stocking.

Shrimp farms suffered infrastructural damage and very large economic loss as a result of the flooding caused by hurricane Mitch. Given this situation it was not possible to repeat the rainy-season experiment nor conduct the dry-season experiment.

#### **ANTICIPATED BENEFITS**

Results of this research would have continued to contribute to the refinement of techniques for exchanging water efficiently in shrimp ponds managed semi-intensively.

## **ACKNOWLEDGMENTS**

We thank Jaime Lopez and farm personnel responsible for sample collection and transport to the La Lujosa Lab for their collaboration. This study was made possible by the collaboration of the General Directorate of Fisheries and Aquaculture (DIGEPESCA), the Ministry of Agriculture and Livestock, and the Honduran National Association of Aquaculturists (ANDAH).

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- Table 1. Mean water quality variable concentrations in ponds and inlet water during the ten-week period before ponds were flooded because of tropical storm Mitch. Water exchange (10% pond volume d<sup>-1</sup>) in 0.1-ha earthen ponds stocked with 14.5 *P. vannamei* m<sup>-2</sup> began four, seven or ten weeks after ponds were stocked.

Variable	Inlet Water	Water Exchange Regime				
		4 Weeks	7 Weeks	10 Weeks		
Salinity (g l <sup>-1</sup> )	9.79	11.45	11.37	12.25		
Total Ammonia N (mg l <sup>-1</sup> )	0.04	0.04	0.05	0.04		
Total Nitrogen (mg l <sup>-1</sup> )	1.15	1.50	1.75	1.85		
Total Phosphorus (mg l <sup>-1</sup> )	0.14	0.19	0.22	0.27		
Soluble Reactive P (mg $l^{-1}$ )	0.02	0.04	0.02	0.06		
Total Alkalinity (mg $l^{-1}$ as CaCO <sub>3</sub> )	79.54	99.12	114.38	111.28		
Chlorophyll $a (mg m^{-3})$	53.38	52.08	63.40	70.73		
$BOD_2 (mg l^{-1})$	3.61	4.78	5.37	6.03		
Reactive Silicate (mg l <sup>-1</sup> )	3.81	3.83	3.43	3.73		

Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics/Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]


# Use of Pond Effluent for Irrigation in an Integrated Crop/Aquaculture System

Ninth Work Plan, Effluents and Pollution Research 1 (9ER1) Progress Report

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#### Abstract

Kenya's annual production from aquaculture is estimated at 1,100 Mg, the largest portion of this being harvested from fish ponds. The value of pond effluents in flood irrigation of crops has been demonstrated, but little research has addressed use of pond water in more efficient systems such as drip irrigation of high-value vegetable crops. A field experiment was conducted on a vertisol at Sagana, Kenya, to determine the suitability of polyculture (tilapia (Tilapia aureus) and African catfish (Clarias gariepinus)) fish-pond effluent for drip irrigation of french bean (Phaseolus vulgaris cv. Samantha). Treatments included nonirrigated, unfertilized (-I-F); nonirrigated, fertilized (-I+F); irrigated with canal water, unfertilized (+I-F); irrigated with canal water, fertilized (+I +F); irrigated with fish pond effluent, unfertilized (+P -F); and irrigated with equal parts canal and pond water, unfertilized (+IP-F). Canal water supplied to polyculture production ponds and to treatments +I-F, +I+F, and +P -F contained 0.49 and 0.04 mg l<sup>-1</sup> of nitrogen (N) and phosphorus (P), respectively. For treatments utilizing fish-pond effluent, water was transferred from nearby polyculture ponds that received 20 kg N ha $^{-1}$  wk $^{-1}$  and 8 kg P ha $^{-1}$  wk $^{-1}$  over a 17-week cycle. Pond water contained higher concentrations of N (6.03 mg kg<sup>-1</sup>) and P (3.89 mg kg<sup>-1</sup>) than canal water. French bean harvest began 46 days after planting and continued for 28 days. Significant differences were observed among treatments  $(P \le 0.001)$  with -I -F yielding 1.2 Mg fresh beans ha<sup>-1</sup>, and +I +F providing the highest yields: 9.1 Mg fresh beans ha<sup>-1</sup>. Irrigation alone (+I -F) resulted in 7.7 Mg fresh beans ha-1 with stepwise yield decline as fish-pond water was substituted for canal water: +IP -F and +P -F yielded 6.1 and 4.3 Mg fresh beans ha-1, respectively. Yield decline with increasing amounts of pond effluent may be owing to particulates that clog drip line emitters. The 41% yield decline from pond-water substitution represents an economic loss of KSh 89,850 ha<sup>-1</sup> (≈US \$1,404). Given the potentially high N and P concentrations of fish-pond effluent, its direct discharge into water bodies should be discouraged. Either pre-treatment filtration or alternative irrigation methods are required before advantage may be obtained from application of nutrient-enriched pond water.

#### INTRODUCTION

In Kenya there are approximately 46,000 fish ponds producing about 1,100 Mg of fish annually (Government of Kenya, 1997). Fertilizers are applied to ponds to increase inorganic nutrient concentrations that favor phytoplankton growth, enhancing production of fish and crustaceans (Boyd, 1990). During harvesting, ponds are drained to levels where fish can be recovered via nets. A result of pond draining is effluent discharge (Sumari, 1982). Such effluents are often allowed to run into natural waterways. Effluents from fertilized ponds have relatively high nutrient concentrations and can be potential sources of pollution and eutrophication for receiving waters.

Pond effluents have been applied to crops as irrigation water (Prinsloo and Scoonbee, 1987; Al-Jaloud et al., 1993; Hussein

and Al-Jaloud, 1995). Hussein and Al-Jaloud (1995) report wheat grain yields ranging from 770 to 5,010 kg ha<sup>-1</sup> with well water and 2,140 to 5,790 kg ha<sup>-1</sup> with aquaculture effluent. Improved water use efficiency (WUE) was also reported with aquaculture-effluent-irrigated crops having a WUE of 11 to 30 kg ha<sup>-1</sup> mm<sup>-1</sup>, whereas well-water treatments had a WUE of 7 to 22 kg ha<sup>-1</sup> mm<sup>-1</sup>. Grain yield and WUE obtained with well water combined with 75 to 100% of the nitrogen requirement as fertilizer were comparable to treatments irrigated with aquacultural effluents combined with 25 to 50% of the nitrogen requirement. These results imply that application of 150 to 225 kg N ha-1 for well-water irrigation and 75 to 160 kg N ha<sup>-1</sup> for aquaculture-effluent irrigation containing 40 mg N l-1 is sufficient for optimum grain yield and WUE. Similar results were obtained by Al-Jaloud et al. (1993).

When pond effluents are applied in arid and semi-arid environments, greater crop returns may be obtained through more efficient application methods. In Kenya, where farm ponds can also serve as water reservoirs for irrigation, drip irrigation could be profitable. Drip irrigation is a technique whereby water and fertilizers can be placed directly over the root zone through use of emitters that are calibrated for low flow rates. Drip irrigation appears most promising when water and fertilizer application is split into several events over a cropping season.

Little work has been conducted in East Africa on the use of fish-pond effluent as a source of irrigation water for high-value crops. A study was undertaken to determine the effects of irrigation with polyculture (tilapia and catfish) pond water on French bean yields.

# METHODS AND MATERIALS

The study was conducted at Sagana Fish Farm in the Central Province of Kenya. The farm lies at an elevation of 1,231 m above sea level. Rainfall at the farm ranges from 1,332 to 1,612 mm yr<sup>-1</sup>, and daily average air temperatures range from 16.3 to 26.9°C. The water supply to the farm comes from the Ragati River. Soils at the farm are "black cotton soils" (vertisols) of volcanic origin.

One fish pond from Sagana Fish Farm was selected to supply effluent for this experiment. The pond was fertilized with 8 kg P ha<sup>-1</sup> as diammonium phosphate (DAP) during a 17-week period prior to stocking. The pond was then stocked with tilapia (*Tilapia aureus*) and African catfish (*Clarias gariepinus*). Subsequently, nitrogen was supplied to the pond weekly at a rate of 10 kg ha<sup>-1</sup>.

Eighteen field plots measuring  $10 \times 6$  m were prepared on land previously under star grass (*Digituara scalarum*). Plots were hand-tilled and hand-harrowed sufficiently for planting French bean. In October 1998, plots were planted with French bean (var. Samantha) at a spacing of  $0.6 \times 0.1$  m.

The experiment design was an incomplete factorial arranged as a randomized complete block with six treatments replicated three times. Treatments consisted of: nonirrigated, unfertilized (-I -F); nonirrigated, fertilized (-I +F); irrigated with canal water, unfertilized (+I -F); irrigated with canal water, fertilized (+I +F); irrigated with fish-pond effluent, unfertilized (+P -F); and irrigated with equal parts canal and pond water, unfertilized (+IP -F). At planting, DAP (200 kg ha<sup>-1</sup>) was applied to treatments receiving fertilizer. These treatments received an additional 200 kg ha<sup>-1</sup> of calcium-nitrate as top dressing after bean emergence. Plots receiving irrigation water were fitted with garden drip irrigation systems. A 10-l distribution bucket suspended on a post held water (canal or pond) to irrigate individual plots receiving irrigation treatments. Irrigated treatments received 0.03 mm water d<sup>-1</sup> over a growing season of 74 days.

French bean harvest began 46 days after planting, and continued for 28 days. Fresh and dry weight of bean pods and total biomass dry weight (dry pods, leaves, and stems) were recorded. Analyses of variance were performed to determine variation in French bean fresh and dry biomass owing to treatments.

# RESULTS

Table 1 provides information on nitrogen and phosphorus concentrations in water used for irrigation from the canal and the polyculture pond. Upon introduction of canal water into the pond and subsequent fertilization, the nitrogen and phosphorus contents in the pond water increased.

French bean was in the field from early November to early February. A total of 28.8 m<sup>3</sup> water was withdrawn from the pond for irrigation of French bean plots. With an irrigation addition of 0.03 mm d<sup>-1</sup>, 2.4 kg N ha<sup>-1</sup> and 1.6 kg P ha<sup>-1</sup> were supplied to the root zone for the whole season. This input was extremely low and equivalent to 4.8% and 3.2% of the recommended rates for nitrogen and phosphorus, respectively.

Fresh and dry weight of bean pods and above-ground dry matter yield of bean plants are shown in Table 2. Yields were modified by treatment ( $P \le 0.001$ ).

Table 1.Nitrogen and phosphorus concentrations of canal and fish-<br/>pond water used for irrigation treatments on French beans<br/>at Sagana, Kenya, in 1998. Numbers in parentheses are<br/>standard errors.

Source	Ν	Р
	(mg	$(1^{-1})$
Canal Pond	0.49 (0.13) 6.03 (0.58)	0.04 (0.03) 3.89 (1.06)

Table 2. Fresh bean yield, dry bean yield, and dry biomass (stem + leaf) yield of French beans as affected by irrigation and fertilizer treatments at Sagana, Kenya, in 1998. Numbers in parentheses are standard errors.

Treatment	Fresh Bean Weight	Dry Bean Weight	<b>Biomass Yield</b>
		(kg ha <sup>-1</sup> )	
Nonirrigated, Unfertilized	1,249 (701)	116 (66)	272 (176)
Irrigated, Fertilized	7,284 (1,881) 7,704 (357)	680 (175) 719 (33)	1,494 (569) 1,214 (490)
Irrigated with Canal Water, Fertilized Irrigated with Fish Pond Effluent, Unfertilized	9,144 (2,419) 6,051 (1,935)	853 (226) 565 (181)	1,567 (475) 785 (277)
Irrigated with Equal Parts Canal and Pond Water, Unfertilized	4,289 (851)	400 (79)	640 (144)

# DISCUSSION

The amount of water withdrawn from the fish pond and applied to the French bean plots was relatively small compared to losses from evaporation and seepage. Pond water volume reduction related to irrigation of horticulture crops can apparently be accommodated in an integrated fish/crop system.

The control treatment (-I -F) yielded 1.2 Mg ha<sup>-1</sup> of fresh beans; the inadequate rainfall and soil fertility may explain the low yield of this treatment. The highest bean yield, 9.1 Mg ha<sup>-1</sup>, was observed where canal water was used for irrigation in combination with fertilizer application. Adequate amounts of moisture were attained with irrigation, while the crop received sufficient nutrition from fertilizer. Application of pond water without fertilization resulted in a yield of 4.3 Mg ha<sup>-1</sup>. The amount of nutrients supplied by pond water over the growing season was not adequate to produce high yields. In addition, filters fitted on the drip system receiving water from the pond clogged frequently and were cleaned more often than those fitted on systems receiving water from the canal. Clogging was due to high total solids and algae in pond water. Pond water was poorly distributed along drip irrigation lines due to irregular clogging of emitters.

As pond water was substituted for canal water, the effects of the accompanying high algae and total solids concentrations increased (Table 2). There was a gradual decline in yield, with the 1:1 ratio of pond:canal water giving an intermediate yield of 6.1 Mg ha<sup>-1</sup>. A 41% yield decline from substituting pond water for canal water resulted in an economic loss of KSh 75,000 ha<sup>-1</sup> if French beans are valued at KSh 30 kg<sup>-1</sup>.

# ANTICIPATED BENEFITS

Application of chemical fertilizers in ponds and activities of fish increase the nutrient concentration of pond water. Application of pond water to crops during fish grow-out is feasible, but filtration will be required if it is to be delivered through a drip irrigation system. Nutrient enrichment of pond water during aquaculture production is insufficient to meet crop nutrient demand unless the water is applied in larger amounts. Larger amounts of water may cause negative effects including impaired soil aeration.

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# PD/A CRSP Seventeenth Annual Technical Report

# FATE OF METHYLTESTOSTERONE IN THE POND ENVIRONMENT: DETECTION OF MT IN SOIL AFTER TREATMENT WITH MT FOOD

Ninth Work Plan, Effluents and Pollution Research 2A (9ER2A) Final Report

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## Abstract

This study examined the persistence of  $17\alpha$ -methyltestosterone (MT) in the environment after its use for masculinizing Nile tilapia. Fry were treated with a masculinizing dose of MT (60 mg kg<sup>-1</sup>) for four weeks beginning at the initiation of feeding in model ponds which consisted of 60-1 tanks that contained either 5 kg of soil, gravel, or no soil. Water and soil samples were taken before the onset of treatment and weekly beginning on the last day of treatment (water samples were also taken weekly during the four-week treatment period). Concentrations of MT were determined by radioimmunoassay, which revealed that the levels of MT in the water peaked at approximately 3.6 ng ml<sup>-1</sup> at 28 days after the onset of feeding. Concentration of MT in water decreased to background level by 35 days after the onset of feeding (one week after the end of treatment with MT-impregnated food) in the tanks with soil or gravel, but remained above background through 49 days in the tanks without soil. The levels in the soil at between 2.8 and 2.9 ng g<sup>-1</sup> after 84 days (eight weeks after ending treatment with MT-impregnated food). In tanks with gravel or no soil, MT was detected at higher levels in a fine sediment that formed after the end of dietary treatment. These results demonstrate that MT persists in soil for up to eight weeks after cessation of MT treatment, which raises the possibility that unintended exposure to MT may occur.

#### INTRODUCTION

Treatment of tilapia fry with  $17\alpha$ -methyltestosterone (MT)impregnated food to produce all-male populations has become a common aquaculture practice. All-male populations are desirable because no energy is shunted toward reproduction and no competition with younger fish for food occurs (Green et al., 1997). Nevertheless, uneaten or unmetabolized food may leak significant amounts of MT into the pond environment, posing the risk of unintended MT exposure to hatchery workers as well as to aquatic and terrestrial organisms. Therefore, to increase the safety and efficacy of MT use for masculinization, we have set about to determine the fate of MT in semi-closed systems such as ponds. This study was undertaken to extend and expand earlier work (Fitzpatrick et al., 1999) which demonstrated persistence of MT in soil for nearly a month after cessation of treatment.

# METHODS AND MATERIALS

Breeding families of Nile tilapia, *Oreochromis niloticus*, were placed in 200-l aquaria (one male to three females) where water temperature was maintained at  $28 \pm 1^{\circ}$ C. Once breeding occurred, the other fish were removed and the brooding female was left to incubate the progeny. The female was forced to release the fry from her mouth at 280 Celsius Temperature

Units (CTU) or 10 days post-fertilization (dpf). Fry were removed from the tank and randomly assigned initially to 3.8-1 jars until 420 CTU (15 dpf) when they were placed in the model ponds at a stocking rate of 200 fry tank<sup>-1</sup> (1 fry per 9.0 cm<sup>2</sup>). This value corresponds to one-third of the recommended stocking rate (by area) for masculinization of 3,000 fry m<sup>-2</sup> (Popma and Green, 1990). However, the volume used in model ponds in this study was limited by the tank height, conferring a stocking rate by volume of 4 fish l<sup>-1</sup> (1 fish l<sup>-1</sup> more than the recommended stocking rate). Model ponds were set up two days before the expected time of fry release.

Water quality and survival data obtained from previous experiments suggested the need for modifying the original experimental design proposed for this study. The proposed design included the use of 3.8-l glass chambers; instead, 60-l aquaria were used. Two aquaria contained 5,000 g (approximately 3 cm) of packed soil, which was obtained from a meadowed hill north of Corvallis, Oregon; two other aquaria contained gravel or no soil, respectively. Each model pond contained 50 l of dechlorinated tap water. To determine the fate of MT in sediments of model ponds, the following experimental groups were included: MT-fed fish in an aquarium containing soil; control-fed fish in an aquarium containing soil; MT-fed fish in an aquarium containing gravel; and MT-fed fish in an aquarium with no soil. The latter two treatments were

included to minimize soil effects on water quality that could affect survival and/or efficacy of treatment. Because the number of fry obtained from the spawning was limited, the experiment treatments were not replicated. MT-impregnated food was made by spraying crushed flaked food with MT dissolved in EtOH; control food was made by spraying crushed flaked food with EtOH. Fry were initially fed with Hatchfry Encapsulon<sup>™</sup> (Argent Chemical Laboratories) in the jars because in this system active feeding does not initiate until about 15 dpf. Delaying treatment in this instance also allowed the fry to reach the initial size proposed by Popma and Green (1990) for treatment. Fry were fed MT (60 mg kg<sup>-1</sup>) or control diet for 4 weeks (from 15 to 43 dpf). Water temperature was maintained at  $28 \pm 1^{\circ}$ C in the jars and at  $26.5 \pm 1^{\circ}$ C in the model ponds. Temperature was monitored daily; pH, ammonia, nitrites, and dissolved oxygen were checked weekly. Feeding rate was at 20% per calculated body weight for the first 23 days of treatment and then 10% per calculated body weight through day 28 of treatment (Popma and Green, 1990). Water lost by evaporation from the model ponds was restored twice weekly.

After 28 days of dietary treatment (on 44 dpf), fry were transferred to the Oregon State University's Warm Water Research Laboratory, Corvallis, Oregon, and reared in 75-1 fiberglass tanks in a recirculating system. Water temperature in the grow-out system was maintained at  $28 \pm 1$  °C and water quality parameters described above were also checked. At 80 to 90 dpf, sex ratios were determined by microscopic examination (10 and 40X) of gonads using squash preparations in Wright's stain (Humason, 1972). The weights of sampled fish were recorded at this time.

Water samples (12 ml) were collected with pipets into 15-ml polypropylene tubes and stored at -20°C until analysis for MT. Soil core samples were collected with 0.5-cm-diameter plastic pipes, placed in whirl pak bags, excess water poured off, and stored at -20°C until analysis. In tanks with gravel and no substrate, a film of fine sediments was formed. This material was collected with a pipet and stored at -20°C until analysis. Fine sediments were precipitated by centrifugation, and a 1-ml subsample was oven-dried at 50°C. For analysis of MT concentration, 1.0 ml of each water sample, 0.2 g of each soil sample, and 0.2 g of each fine sediment sample were extracted in 8 ml of diethyl ether. The organic phase of each sample was collected in a new tube after the aqueous phase was snapfrozen in liquid nitrogen. The extraction procedure was repeated and the ether extracts were pooled for each sample and dried down in a SpeedVac. Each dried extract was reconstituted in 1 ml of phosphate-buffered saline containing gelatin. Aliquots of the reconstituted extracts were removed to 12×75 mm tubes for determination of MT concentration by radioimmunoassay (RIA). The RIA methods followed the procedure outlined in Fitzpatrick et al. (1986; 1987). Antisera specific to MT were purchased from UCB-Bioproducts SA, and <sup>3</sup>H-MT (Amersham) was generously donated by Dr. Gordon Grau of the Hawaii Institute of Marine Biology. Standards of known concentration of MT were made in EtOH and used in each assay to generate a standard curve. The assay was validated by demonstration of parallelism between serial dilutions of several samples and the standard curve, and by demonstration of low cross-reactivity with testosterone and 11-ketotestosterone. Furthermore, soil samples were subjected to analysis by HPLC after extraction (as above), filtering, and reconstitution in MeOH to search for possible metabolites of

MT. Extraction efficiency for MT for the RIA was checked by adding a known amount of <sup>3</sup>H-MT to water and soil, (n = 5 for each), and then extracting the samples as described above. Once each of these tubes was reconstituted in 1 ml of phosphate-buffered saline containing gelatin, 0.5 ml was removed from each and the amount of radioactivity was counted by scintillation spectroscopy (extraction efficiencies were 73.3% for water and 71.4% for soil).

Sex ratio and mortality data were analyzed using Fisher's exact test with exact p-values (a more conservative test than the chisquare test for small sample sizes) estimated in GraphPad Prism<sup>TM</sup>. Intersex fish were counted as females for the purposes of analysis in order to be conservative. Concentrations of MT in water, soil, and interface at the various sample times were not compared statistically because of the limited sample size (n = 1 or 2 per date) and because the goal of the study was descriptive (presence/absence). The mean final weights of sampled fish were analyzed for differences between groups using one-way ANOVA including mortality as a possible confounding variable. For all analyses, differences were considered statistically significant when the p-value (P) was less than 0.05.

#### RESULTS

The percentages of males in the MT-treated groups with soil (47.0%) and gravel (48.4%) were significantly higher than the control group (10.4%; P < 0.0001). The MT-treated group with no substrate had significantly more males (19.3%) than the control group (P = 0.02), but significantly less males than the treatments with substrate (P < 0.0001; Figure 1). Mortality values ranged from 6 to 10%, and showed no significant differences between treatments (P = 0.52).

Water samples had a background level of MT between nondetectable (nd) and 0.02 ng ml<sup>-1</sup>. During the treatment period, the levels of MT in the water varied from 0.04 to 3.61 ng ml<sup>-1</sup> in





MT-treatment with soil (MT-soil); 0.02 to 0.50 ng ml<sup>-1</sup> in MT treatment with gravel (MT-gravel); and from 0.05 to 4.97 ng ml<sup>-1</sup> in MT-treatment with no soil (MT-no soil; Figure 2a). After removal of the fish and cessation of MT treatment, MT levels in water declined rapidly in MT-soil and MT-gravel tanks; however, MT remained about 10 times the background in water from the MT-no soil tank through week 12.

A background level of MT was detected in soil samples collected at the beginning of the experiment (mean =  $0.5 \text{ ng g}^{-1}$ ). In the MT-soil tank, MT increased to  $6.1 \text{ ng g}^{-1}$  at the conclusion of dietary treatment (Figure 2b), remained near this level for two more weeks, and then decreased to about 3 ng g<sup>-1</sup> through the end of the experiment (Eight weeks after the cessation of dietary treatment). Concentrations of MT in the fine sediment in the MT-gravel and MT-no soil tanks were considerably higher than MT levels in soil or water (Figure 2c). One week after cessation of feeding, the level of MT in fine sediment was 168.7 ng g<sup>-1</sup> in the MT-gravel tank, MT ranged from 22.9 to 99.2 ng g<sup>-1</sup> of fine sediment throughout the eight weeks following cessation of treatment.



Figure 2. Levels of  $17\alpha$ -methyltestosterone (MT) in the environment during and after dietary treatment of *Oreochromis niloticus* fry. Fish were fed either a control diet or a diet containing  $60 \text{ mg kg}^{-1}$  of MT through week 4. Graph depicts MT levels in (a) water, (b) soil, and (c) fine sediments.

#### DISCUSSION

These experiments confirm previous studies using smaller containers (Fitzpatrick et al., 1999) which demonstrated that considerable amounts of MT can be found in the pond environment during and after dietary treatment with MT. MT levels in water peaked at the end of dietary treatment and consistently returned to background levels in tanks with substrate. However, when no substrate was present, MT levels remained above 1 ng ml<sup>-1</sup> for two weeks after the end of treatment. The accumulation and persistence of MT in soil was demonstrated to extend through eight weeks after the end of dietary treatment. Our results suggest that sediments act as a trap for MT while in tanks with no substrate, MT remains in suspension for longer periods of time until finally binding to fine particles to form a film at the bottom of the tank.

A significant level of masculinization was obtained from dietary treatments with MT tanks with substrate; however these values are below what has been reported by other researchers (see Green et al., 1997). Interestingly, the fish fed with MT but kept in tanks with no substrate had significantly lower masculinization values than those with either soil or gravel. MT in the water and in the fine sediments was high in the MT-no soil tank; therefore, the fish may have been exposed to higher doses of MT. Since MT can be converted to estrogenic compounds by the enzyme aromatase, the low level of masculinization in the MT-no soil group may be the result of paradoxical feminization. Previous results showed that lower levels of masculinization are obtained when tilapia fry are fed twice the recommended dose of MT (Fitzpatrick et al., 1999).

The inability to obtain 95 to 100% masculinization is troubling but is not uncommon in production facilities around the world. The concentrations of MT were measured in the diets (data not shown) and established to be at the target dosage of 60 mg kg<sup>-1</sup> of food. Therefore, the lack of masculinization was not due to improper diet preparation. One possible explanation is that because water filtration was not provided during treatment, water quality deteriorated within the tanks to limit the effectiveness of MT by stressing the fish.

These data confirm previous results (Fitzpatrick et al., 1999) showing that treatment of tilapia with MT results in leakage of this anabolic steroid into the environment. This study extends previous work by showing that MT may persist in the soil for at least two months after cessation of treatment with no indication that the levels will decline thereafter. Thus, MT persistence may pose an exposure risk to humans and other organisms.

#### **ANTICIPATED BENEFITS**

The anabolic steroid  $17\alpha$ -methyltestosterone (MT) can be detected in the sediments of model ponds for up to two months after its use for masculinizing tilapia fry. MT dissipates from water a few days after fish are taken out of the treatment tanks. Removal of fish from the system allows suspended material to precipitate carrying with it MT and/or its metabolites. However, MT is persistent in sediments for up to eight weeks after the end of treatment. These findings raise concerns about the potential impact of such residual MT with regard to unintended exposure of pond workers as well as other fish and organisms. Further research is needed to determine the nature of the background levels of MT detected in the radioimmunoassay in soil samples, as well as possible metabolites produced after MT treatment.

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# PD/A CRSP Seventeenth Annual Technical Report

# Fate of Methyltestosterone in the Pond Environment: Detection of MT in Pond Soil from a CRSP Site

Ninth Work Plan, Effluents and Pollution Research 2B (9ER2B) Abstract

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# Abstract

The following study will examine if  $17\alpha$ -methyltestosterone (MT) persists in the environment after its use for masculinizing Nile tilapia at one or more PD/A CRSP sites. Experiments are currently underway at the Universidad Juárez Autónoma de Tabasco, Mexico. Fry have been treated with a masculinizing dose of MT (60 mg kg<sup>-1</sup>) for four weeks beginning at the initiation of feeding. Water and soil samples were taken from the pond before the onset of treatment and one day after the end of treatment; samples will also be taken at four weeks after the end of treatment. Concentrations of MT will be determined by radioimmunoassay. If possible, a similar sampling design will be applied to the Sagana Station, Kenya, with subsequent analysis of samples at Oregon State University.

Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics/Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_.]



# NONPARAMETRIC ESTIMATION OF RETURNS TO INVESTMENT IN HONDURAS SHRIMP RESEARCH

Eighth Work Plan, Marketing and Economic Analysis Research 1 (8MEAR1) Final Report

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### Abstract

Economic returns to the investment in shrimp research in Honduras between 1993 and 1997 by PD/A CRSP researchers were estimated using a nonparametric approach. A survey of shrimp growers in Honduras provided data on yield, input application, and prices for their first year of production and for the year 1997. Research investment data included funding from both public and private sectors. Results showed that total factor productivity indices increased from 1995 to 1997, indicating technical progress due to research. When both private and public investment were considered, the internal rate of return to the investment in research was 45%. However, the internal rate of return to public-sector investment alone was above 6,352%. This indicated that the public funds invested in shrimp research in Honduras have been leveraged effectively with private-sector capital to generate technological progress.

# INTRODUCTION

Assessment of the effects of agricultural research investment has become increasingly important to justify public-sector funding to research administrators and government policymakers. The United States Agency for International Development (USAID) has provided public-sector funding for shrimp research in Honduras. Using these resources, researchers from Honduras and the US, under the auspices of the Pond Dynamics/Aquaculture Collaborative Research Support Program (PD/A CRSP) (a USAID-funded program involved in global pond aquaculture research), have been conducting experiments to improve shrimp production efficiency in Honduras since 1993. This study estimated the economic returns to this investment, particularly the effects of research on shrimp-farm productivity.

The shrimp industry, with US\$77 million in farmgate sales in 1996, is one of the largest and fastest growing sources of foreign exchange for Honduras (ANDAH, 1997). Improving production efficiency for this industry could contribute significantly to economic development of this and other shrimp-producing nations. Auburn University's Memorandum of Agreement with ANDAH called for member farms to provide ponds and production inputs to contribute to this research program. Granjas Marinas San Bernardo (GMSB) is one of the largest Honduran shrimp farms, and, because of its infrastructure and desire to collaborate, it supported PD/A CRSP research by 1) making multiple ponds available for experiments and 2) absorbing shrimp production costs in those ponds by supplying feed, post larvae (PL), labor, energy, and all other production inputs.

Although there is a rich history of returns to research investment studies in agriculture, there is no record of this type of analysis for aquaculture research. Hence, this paper fills a void in the literature with the first attempt to estimate returns to aquaculture research. The remainder of this paper is divided into sections that 1) briefly review the existing literature on measuring returns to research investment, 2) describe the theoretical and empirical models, 3) summarize the data, and 4) discuss results. The paper ends with a concluding section that summarizes key results from the study.

# MEASURING RETURNS TO RESEARCH INVESTMENT

Typically, three techniques exist in estimating returns to agricultural research investment: parametric methods, nonparametric methods, and index-number methods. Parametric methods, which have been the traditional approach, involve explicit functional form specifications that link inputs to outputs. In the primal parametric technique, functional forms for the production technology are specified. Research impacts on production are measured by either directly including research expenditures as exogenous shifts to the production function or by measuring technical change using proxy variables such as a time trend and attributing the technical change to research-based information. Other methods include estimating a production function by conditioning the technical coefficients on research investment (Alston et al., 1998). Examples of the primal approach exist in Griliches (1964) and Evenson (1967).

The dual parametric technique estimates the impact of research on profit or cost functions of a firm. After a functional form specification for the profit or cost function, output supply and input demand functions can be derived using standard microeconomic principles. Hence, research expenditure variables included in the profit or cost functions could also be components of the output supply and input demand functions. Joint econometric estimation of output supply and input demand functions measure the research impacts on a firm's profit or cost functions. An example of this type of analysis exists in Huffman and Evenson (1989).

If the analysis uses a single-product technology, the direct estimation of a supply response to research investment model could also be an alternative to the primal approach. Here, "direct estimation" means that the supply-response model is not a derivative of a pre-specified profit or cost function. This technique allows the dynamics of supply response to prices and research investments to be modeled with greater flexibility than in primal and dual models (Alston et al., 1998). An example of this method appears in Fox et al. (1987). Once the research impacts on production have been estimated, the remaining step is to evaluate the resulting benefits. The underlying methodology used in this step is based on, either explicitly or implicitly, the economic surplus concept. Typically, this involves allocating the research benefits to different productive surplus (e.g., producer factors) and different consumer surplus (e.g., consumer groups).

The striking feature of the above methodologies is the *a priori* constraints in the empirical models stemming from an assumed parametric form of the technology. The primal approach usually restricts the technology structure, and the dual approach imposes behavioral constraints (Alston et al., 1998). The nonparametric approach dispenses with the use of functional forms altogether. Here, the data are checked for consistency with cost-minimization or profit-maximization behavior (weak axiom of cost minimization and weak axiom of profit maximization developed by Varian (1984)). If the data are inconsistent with the above axioms, the minimum adjustments to the data necessary to restore consistency are determined. These adjustments refer to the quantity changes in outputs and inputs that can be attributed to an input-using, input-saving, or neutral technical change (Chavas and Cox, 1992). By incorporating research expenditure in the analysis, output and input changes that are not attributable to changes in output and input prices or production scale are used to measure the effects of research (Alston et al., 1998).

The nonparametric algorithms do not restrict input substitution possibilities, joint estimation of the production technology, technical change, or the effects of research on technical progress using disaggregate inputs (Alston et al., 1998). This approach requires only a standard linear programming algorithm. Given its flexibility, this approach is adopted in this study to measure the impacts of public and private research expenditures on Honduran shrimp production.

The index-number approach is useful in developing partial factor and total factor productivity measures that summarize the growth in agricultural output. This method is often used in conjunction with the above techniques to determine research-induced technical changes as exhibited in the pre-change and post-change input and output quantities. An example of this method exists in the Christensen and Jorgenson total productivity index discussed in Christensen and Jorgenson (1970) and Diewert (1976). Partial factor and total factor productivity indices are also used in this paper.

# THEORETICAL AND EMPIRICAL MODEL

The nonparametric approach of evaluating the returns to research investment is based on Varian's (1984) weak axiom of profit maximization (WAPM). This hypothesis indicates that a profit-maximizing agent's production decisions are such that profit from any time period is at least as large as the profit that could have been obtained using production decisions from any other time period. Mathematically, this is expressed by the inequalities in Equation 1, which are developed for a single output (y) technology with two variable inputs (x1, x2) such that the following firm-level data are available for time periods t1 and t2:  $(y_{tl'} x 1_{tl'} x 2_{tl'} p_{tl'} w 1_{tl'} w 2_{tl})$  and  $(y_{t2'} x 1_{t2'} x 2_{t2'} p_{t2'} w 1_{t2'} w 2_{t2})$ , where  $p_{ti'} w 1_{ti'}$  and  $w 2_{ti}$  are prices during time ti (i = 1, 2) of the output and inputs x1 and x2, respectively.

$$\begin{array}{l} p_{t1} y_{t1} - w 1_{t1} x 1_{t1} - w 2_{t1} x 2_{t1} \ge p_{t1} y_{t2} - w 1_{t1} x 1_{t2} - w 2_{t1} x 2_{t2} \\ p_{t2} y_{t2} - w 1_{t2} x 1_{t2} - w 2_{t2} x 2_{t2} \ge p_{t2} y_{t1} - w 1_{t2} x 1_{t1} - w 2_{t2} x 2_{t1} \end{array}$$
(1)

Cox and Chavas (1990) have shown that the WAPM inequalities form necessary and sufficient conditions for profit maximization, i.e., in every time period t, if price-taking producers apply inputs to maximize profit:  $p_t y_t - w_t x_1 - w_t x_2$ , where y is a function of x1 and x2, then the inequalities in Equation 1 would be satisfied, and vice versa.

Technical change is incorporated in the above framework by structuring WAPM behavior on effective inputs and output. Effective inputs and output are the result of the augmentation effect of technical change on actual inputs and output. The underlying concept is that, due to an input-saving (or -using) technical change, a single unit of an input would be equivalent to a larger (or smaller) amount of the same input had the technical change never occurred. Thus, the ith effective input (Xi) is a function of both the actual input quantity (xi) and a technological index (Bi), i.e., Xi = Xi(xi, Bi) (Chavas and Cox 1992). For empirical tractability, Xi(xi, Bi) is expressed as Xi = xi+ Bi. The technological indices for the inputs (Bi) capture the input bias effects of technical change. If Bi > (or <) 0, the technical change is said to be input-saving (or -using) in the ith input. If all Bis are zero, the marginal rates of substitution between the two inputs are unaffected by the altering technology, indicating a Hicks neutral technical change (Chambers, 1988). Similarly, a technological index (A) captures the outputaugmenting effect of technical change by inter-relating effective (Y) and actual (y) output: Y = y - A or y = Y + A. Thus, higher values of A are associated with higher productivity. If A is positive, the technical change is said to be progressive; negative A values indicate a regressive technical change. The augmented WAPM hypothesis discussed by Chavas and Cox (1992), Cox and Chavas (1990), and Cox et al. (1997) explains producer behavior under technical change by expressing the WAPM inequalities in Equation 1 in terms of effective inputs and output.

$$\begin{array}{l} p_{t1} \left( y_{t1} - A_{t1} \right) - w \mathbf{1}_{t1} \left( x \mathbf{1}_{t1} + B \mathbf{1}_{t1} \right) - w \mathbf{2}_{t1} \left( x \mathbf{2}_{t1} + B \mathbf{2}_{t1} \right) \geq \\ p_{t1} \left( y_{t2} - A_{t2} \right) - w \mathbf{1}_{t1} \left( x \mathbf{1}_{t2} + B \mathbf{1}_{t2} \right) - w \mathbf{2}_{t1} \left( x \mathbf{2}_{t2} + B \mathbf{2}_{t2} \right) \\ p_{t1} \left( y_{t1} - A_{t2} \right) - w \mathbf{1}_{t1} \left( x \mathbf{1}_{t1} + B \mathbf{1}_{t2} \right) - w \mathbf{2}_{t1} \left( x \mathbf{2}_{t1} + B \mathbf{2}_{t2} \right) \geq \\ p_{t1} \left( y_{t2} - A_{t1} \right) - w \mathbf{1}_{t1} \left( x \mathbf{1}_{t2} + B \mathbf{1}_{t1} \right) - w \mathbf{2}_{t1} \left( x \mathbf{2}_{t2} + B \mathbf{2}_{t1} \right) \\ \end{array} \right)$$

The idea behind Equation 2 is that although producers maximize profit, WAPM (as shown in Equation 1) may not be satisfied due to technical change. The technological indices in Equation 2 measure the extent by which producer behavior has deviated from WAPM. In doing so, they measure the input and output bias effects of technical change.

Since the technological indices ( $A_t$  and  $Bi_t$ ) capture the effects of technical change, their relationship to research expenditures is of interest. For any year t, assume that past research expenditures (with a minimum one-year lag) could potentially influence technology (and hence, the technological indices). Since the focus of this study is to evaluate research investment impacts on productivity, only the  $A_t$ s are expressed in terms of lagged research expenditures. (3)

$$A_{t} = a_{1} R_{t-1} + a_{2} R_{t-2} + a_{3} R_{t-3} + \dots + a_{T} R_{t-T}$$

where

 $\begin{array}{ll} R_{t-\tau} \ = \ research \ expenditure \ made \ in \ the \ (t-\tau)^{th} \ year \ and \\ T \ = \ maximum \ lag \ length. \end{array}$ 

Following Chavas and Cox (1992), the  $a_t s$  in Equation 3 are restricted to a linear spline function of t by the following process: (I) the space {0, 1, 2, 3,..., T, T+1} is partitioned into s subsets {0,...,k<sub>1</sub>}, {k<sub>1</sub>,..., k<sub>2</sub>}, {k<sub>2</sub>,..., k<sub>3</sub>}, {k<sub>s-1</sub>,..., T+1} and (II) each  $a_{\tau}$  is defined to be equal to  $(\alpha_t + \beta_j \times \tau)$  if  $\tau \in \{k_{j-1},..., k_j\}$ , j = 1...s, such that  $\alpha_1 = 0 = \alpha_s + \beta_s \times (T+1)$  and  $(\alpha_j + \beta_j \times k_j) = (\alpha_{j+1} + \beta_{j+1} \times k_j)$ . The  $a_{\tau}s$  measure of the marginal impact on current output of research expenditures made  $\tau$  years ago (i.e.,  $R_{\tau}$ ). Hence, as T is the maximum allowed lag in Equation 3,  $a_{T+1} = \alpha_s + \beta_s \times (T+1) =$ 0. Therefore, the linear spline restrictions allow the  $a_{\tau}s$  to vary while imposing some degree of continuity or smoothness, so that the marginal research impacts on output are not erratically distributed over time.

For a given farm, if production price and quantity data were available for two time periods, two augmented WAPM inequalities could be specified as shown in Equation 2. Assuming such data are available for several farms that share identical technology and technical change, the empirical model is specified by incorporating the pairs of inequalities in a mathematical programming model. In doing so, Equation 2 is rewritten so that only unknown parameters and their coefficients appear on the left-hand side.

$$\begin{aligned} (A_{t2} - A_{t1}) &- \frac{w1_{t1}}{p_{t1}} (B1_{t1} - B1_{t2}) - \frac{w2_{t1}}{p_{t1}} (B2_{t1} - B2_{t2}) \geq \\ &- [(y_{t1} - y_{t2}) - \frac{w1_{t1}}{p_{t1}} (x1_{t1} - x1_{t2}) - \frac{w2_{t1}}{p_{t1}} (x2_{t1} - x2_{t2})] \\ (A_{t1} - A_{t2}) - \frac{w1_{t2}}{p_{t2}} (B1_{t2} - B1_{t1}) - \frac{w2_{t2}}{p_{t2}} (B2_{t2} - B2_{t1}) \geq \\ &[(y_{t1} - y_{t2}) - \frac{w1_{t2}}{p_{t2}} (x1_{t1} - x1_{t2}) - \frac{w2_{t2}}{p_{t2}} (x2_{t1} - x2_{t2})] \end{aligned}$$

Here, the unknown parameters are the technological indices for output (the A<sub>ti</sub>s are subject to Equation 3 and two variable inputs (B1<sub>ti</sub> and B2<sub>ti</sub>). Since the input technological indices could be positive, zero, or negative, it is empirically convenient to express them as a difference of two positive parameters, i.e.,  $B1_{ti} = B1_{ti}^{+} - B1_{ti}^{-}$  and  $B2_{ti} = B2_{ti}^{+} - B2_{ti}^{-}$ . Hence, the empirical model consists of a mathematical program with pairs of constraints specified for each farm for which data are available (as shown in Equation 4) and an appropriate objective function so that a solution can be derived for the  $a_{\tau}s$ ,  $B1_{ti}$ ,  $B1_{ti}$ ,  $B2_{ti}$ ,  $B2_{ti}$ and B2<sub>ti</sub>'s that jointly satisfy the augmented WAPM hypothesis. This assumes the availability of production data for at least two time periods from one or more farms that share nearly identical technology and experience similar technical change. Details of data used in the empirical model are discussed in the next section.

Given the "greater-than-or-equal-to" inequalities in Equation 4 and restrictions that all unknown parameters are non-negative, the objective function minimizes a linear combination of the unknown parameters with positive coefficients in order to keep the solution bounded. The coefficients of the unknown parameters in the objective function are chosen to be large positive numbers so that the solutions give the smallest values for the technological indices. Following Chavas and Cox (1992) and Cox and Chavas (1990), the coefficients of the a<sub>t</sub>s in the objective function are taken to be a large positive number (q in Equation 5). The coefficients of the Bs (in Equation 5) are the square of q. Consistent with this layout, changing the values of q did not alter the solution of the empirical model.

#### Empirical Model:

$$\underset{t_{2}}{\text{Min } q a_{1} + q a_{2} + q a_{3} + \ldots + q^{2} B1_{t_{1}} + q^{2} B1_{t_{1}} + q^{2} B1_{t_{2}} + q^{2} B1_{t_{2}}$$

Subject to: Augmented WAPM constraint pairs (Equation 4) for each farm

$$\begin{array}{l} a_{1'}, a_{2'}, a_{3'}, \dots, B1_{t1}^+, B1_{t1}^-, B1_{t2}^+, B1_{t2}^-, \dots, \\ B2_{t1}^+, B2_{t1}^-, B2_{t2}^-, B2_{t2}^-, \dots \ge 0 \end{array}$$
(5)

# Data

Shrimp production data from Honduras were collected from a survey of 21 farms that represented 48.25% of the Honduran shrimp industry. Each survey questionnaire elicited information from each farm on production practices (including input application intensity and shrimp yield) for their first production year and for 1997 (survey year). Feed and PL are key variable inputs in shrimp production (Dunning, 1989; Stanley, 1993) and were used in the empirical model (Expressions 4 and 5).

Price data from the survey provided prices of shrimp tails for the first production year and for the year 1997. Estimates of PL prices were provided by Ralph Parkman of Grupo Granjas Marinas (Parkman, pers. comm.). Data on shrimp feed prices were available for 1997 only. Shrimp feed price estimates for other years were obtained by adjusting contemporaneous catfish feed prices (Engle and Kouka, 1996) in the same proportion as the 1997 ratio of shrimp feed prices to catfish feed prices. This assumes that shrimp and catfish feed prices exhibit similar fluctuations that are determined by world grain market prices. All Honduran price data in lempiras were converted to US dollars using exchange rates provided by the Banco Central de Honduras. All price time series in US dollars were deflated by a US Purchasing Power Index (USDA, 1996).

Table 1 summarizes information from 13 of the 21 farms related to yield of shrimp tails (kg ha<sup>-1</sup> yr<sup>-1</sup>) and the partial factor productivity indices (average product) for stocking and feeding rates for the first year of operation and the year 1997. Farms that returned incomplete questionnaires and new farms were excluded. Farms were selected for the empirical analysis after evaluation of the potential for constraint redundancy (or degeneracy) problems. These problems arise in mathematical programming models when coefficients for multiple constraints are nearly identical (McCarl, 1977).

PD/A CRSP research investment data were obtained from PD/A CRSP budgets for shrimp research in Honduras from the work plans covering the period 1993-94 to 1996-97 (Seventh, Interim, and Eighth Work Plans) (PD/A CRSP, 1993, 1995, 1996). The PD/A CRSP data came from four annual investment periods, which are illustrated in Table 2; Table 2 also contains a list of shrimp-related research projects conducted during this time-period. Specific research studies targeted primarily pond management studies (to improve production efficiency) and estuarine water-quality studies (to determine whether or not pond management strategies should involve the exchange of pond water). Pond water exchange could also affect optimal feeding and stocking strategies in shrimp ponds. PD/A CRSP research investment per hectare of shrimp farms was calculated for each time period by dividing the cumulative budgeted Table 1. Partial factor productivity (PFP) for stocking and feeding rate. The PFP of an input is the ratio of the output over the input (i.e., average product) (Alston et al., 1998). "First yr" indicates the first year of operation; "Last yr" indicates the most recent year of operation (1997).

Farm	First Year	Last Year	Yield (Shrimp Tails)		Pa	Partial Factor Productivity		
			kg ha	<sup>-1</sup> yr <sup>-1a</sup>	Stockir	ıg Rate <sup>b</sup>	Feeding	Rate <sup>c</sup>
			First yr	Last yr	First yr	Last yr	First yr	Last yr
$1^d$	1982	1997	499	386	0.00126	0.00088	0.130	0.220
2	1987	1997	538	579	0.00354	0.00318	0.320	0.340
3 <sup>d</sup>	1982	1997	445	265	0.00125	0.00049	e	0.160
4	1993	1997	907	726	0.00179	0.00239	0.159	0.163
5	1992	1997	817	776	0.00336	0.00274	0.384	0.531
6	1986	1997	572	454	0.00188	0.00224	0.117	0.357
7	1993	1997	690	828	0.00284	0.00292	0.324	0.567
$8^{d}$	1995	1997	818	744	0.00367	0.00193	0.411	0.375
9 <sup>d</sup>	1995	1997	1,021	889	0.00336	0.00231	0.466	0.462
10	1990	1997	680	454	0.00336	0.00149	0.429	0.286
11	1995	1997	953	1,216	0.00262	0.00297	0.411	0.524
12 <sup>d</sup>	1993	1997	686	714	0.00242	0.00177	0.216	0.321
13	1985	1997	363	544	0.00199	0.00245	0.171	0.446

<sup>a</sup> Average per-year yield computed from aggregating dry and wet season yields.

<sup>b</sup> The PFP of stocking rate is measured in kg of shrimp tails per PL ha<sup>-1</sup> yr<sup>-1</sup>.
<sup>c</sup> The PFP of feeding rate is measured in kg of shrimp tails per kg of feed ha<sup>-1</sup> yr<sup>-1</sup>.

<sup>d</sup> Farms used in the empirical analysis.
<sup>e</sup> Farm 3 reported no feed applied during their first year of operation.

Table 2. PD/A CKSP shrimp research studies from 1993 to 19	Table 2.	PD/A CRSP	shrimp	research	studies	from	1993	to	199
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Years	Study
1993–1994 Seventh Work Plan	<ol> <li>Estuarine water quality</li> <li>Farm nutrient budgets</li> <li>Reduction of feed input by inorganic fertilizers</li> <li>Influence of frequency and quantity of water exchange on water quality and shrimp production</li> </ol>
1994–1995 Seventh Work Plan	<ol> <li>Estuarine water quality</li> <li>Farm nutrient budgets</li> <li>Reduction of feed input by inorganic fertilizers</li> <li>Influence of frequency and quantity of water exchange on water quality and shrimp production</li> <li>Relationship among stocking density, mean shrimp size, survival, and carrying capacity during wet and dry seasons of Honduras</li> </ol>
1995–1996 Interim Work Plan	<ol> <li>Effect of diet protein on feed conversion and nitrogen discharge during semi-intensive production of <i>Peneaus vannamei</i></li> <li>Estuarine water quality</li> <li>Tidal effects on nutrient, oxygen, temperature and salinity profiles in estuaries of two major shrimp-producing areas in southern Honduras</li> </ol>
1996–1997 Eighth Work Plan	<ol> <li>Estuarine water quality monitoring and estuarine carrying capacity</li> <li>Influence of daily water exchange volume on water quality and shrimp production</li> <li>Water exchange to rectify low dissolved oxygen</li> </ol>

Source: Green, pers. comm.

expenditure on all shrimp projects by the total Honduran shrimp farm area. However, PD/A CRSP research expenditures did not include the pond costs of raising shrimp for experiments conducted in GMSB ponds. For these experiments, GMSB absorbed all variable costs of the studies, including PLs, feed, labor, and energy expenses as well as the sales revenue from the harvested shrimp. Due to the unavailability of historical cost data from GMSB, it is assumed that costs per hectare per year of shrimp cultivation remained approximately constant (\$1,792 ha<sup>-1</sup>) from 1993 to 1997 (Valderrama, unpubl. data). This cost includes expenses for PL, feed, labor, fuel, interest for operating loans, and machinery depreciation. Given that the research was initiated by publicsector (PD/A CRSP) funds, our intention was to estimate the returns from the total research investment. The total research expenditure, which combines both public- and private-(GMSB) sector investments, represents the research costs that would have to have been incurred by the public sector, had there been no private-sector support. Hence, the outputenhancing technological indices (A<sub>t</sub> in Equation 3) included research expenditures (R<sub>t-t</sub>) without adjustment for the GMSB income from sales of shrimp grown in the experimental ponds. Given that PD/A CRSP shrimp research expenditure per hectare varied from \$5.39 (in the 1993-94 time period) to \$7.46



Figure 1. Average shrimp yield in Honduras from 1985 to 1997.

(in the 1995-96 time period), it is clear that GMSB assumed most of the research expenses. Evaluating the returns to PD/A CRSP investment alone without accounting for the pond production costs associated with the experiments would inflate the internal rate of return to the total investment in research. Hence, the empirical model was analyzed by 1) considering the total public and private research investment and 2) exclusively considering the public research investment.

In March 1994, the Taura Syndrome Virus (TSV) affected shrimp cultivation in Honduras (Teichert-Coddington, 1999). The impact of the virus surfaced during the decline of pond productivity (of shrimp tails) from 1,032 kg ha<sup>-1</sup> yr<sup>-1</sup> in 1993 to 774 kg ha<sup>-1</sup> yr<sup>-1</sup> in 1994 (Figure 1) (ANDAH, 1997). Although production declined dramatically at the initial onslaught of the TSV, the industry was able to partially compensate, and production increased after 1995 (Figure 1). In this framework, the empirical model was analyzed by first using survey data that reflect TSV-related yield reduction and second, using survey data with 1995 and 1997 yield levels adjusted by factors of 1.7 and 1.25, respectively, as a sensitivity analysis to evaluate the effects of the TSV-related yield declines on the results of this study (Teichert-Coddington, pers. comm.).

# **RESULTS AND DISCUSSION**

Table 3 reports the technological indices that solve the empirical model (Equation 5) under four scenarios: with (or without) considering the private research expenditures and with (or without) considering the effect of TSV. These results correspond to a minimum one-year lag in determining the research impacts on production. This minimum lag specifies that research conducted cannot begin to impact farm technology until the succeeding year. All data analyses were conducted on a per-hectare basis. The available data did not allow estimation of any spill-in effects (i.e., impacts of information obtained from non-PD/A CRSP research efforts) or spillover effects (i.e., impacts of PD/A research on non-Honduran shrimp production) of research. Since PD/A CRSP shrimp research investment began with the 1993-94 time period, output and input technological indices for all years prior to 1994 were taken to be zero (i.e.,  $A_t$ ,  $B1_t$ , and  $B2_t$  were zero for all t less than 1994). With respect to the "research years" (1994 to 1997), since data exist for only 1995 and 1997, Table 3 reports values for (A<sub>95</sub>, B1<sub>95</sub>, B2<sub>95</sub>) and (A<sub>97</sub>, B1<sub>97</sub>, B2<sub>97</sub>). (6)

$$\begin{array}{l} A_{95} = a_1 \; R_{94\cdot93} \\ A_{97} = a_1 \; R_{96\cdot95} + a_2 \; R_{95\cdot94} + a_3 \; R_{94\cdot93} \end{array}$$

The parameters  $a_1$ ,  $a_2$ , and  $a_3$  provide information on the lag relationship between research and productivity (Chavas and

Cox, 1992). Since  $a_3 \ge a_2 \ge a_{1'}$  research from the more distant past has a higher productivity impact than more recent research. Technically, the parameters  $a_{1'}, a_{2'}$  and  $a_3$  indicate the marginal impacts of research expenditure on output (per hectare). Hence, it is clear from Table 3 that the marginal research impacts are higher if output is inflated to correct for TSV. Further,  $a_{1'}, a_{2'}$  and  $a_3$  are larger in magnitude if private research investments are excluded from computing the R<sub>4</sub>s.

Table 3 also gives the values of the estimated input bias effects of research. The input bias parameters  $B1_{95}$  and  $B2_{95}$  indicate that the technical changes from past years to 1995 are neutral for stocking rate but input-saving for feed (Cox et al., 1997). Given that the results are on a per-hectare basis, Table 3 shows that shrimp producers have benefited from the 1993–94 time period by reducing feeding rates for equivalent stocking rates. Further evidence of this technological improvement in feed use from the pre-research years (prior to 1994) to the post-research years (after 1994) appears in the feed partial factor productivity (PFP) indices shown in Table 1. Except for farms 8 and 9, the PFP of feed improved in the post-research years in all farms, implying that a lower feeding rate in the post-research years resulted in at least the same yield as in the pre-research years.

From 1995 to 1997, the negative input bias indicated by  $B1_{97}$  and  $B2_{97}$  showed that the technical changes were input-using in both inputs. This result can be interpreted by considering the consequences of TSV in 1994. Reports from the PD/A CRSP shrimp research indicate that, although TSV had a considerable effect in reducing pond productivity in 1994, producers were "able to quickly adjust management to partially compensate for low survivals [of PLs] by avoiding the nursery stage and doubling or tripling the stocking rates" (Teichert-Coddington, 1999). This increase in stocking rate for a less-than-proportional increase in yield (yield actually fell due to TSV) explains the negative value of  $B1_{97}$ . In the empirical model, production data from 1995 and 1997 were used from only two farms

Table 3. Technical indices satisfying the augmented weak axiom of profit maximization (WAPM) hypothesis.

Without TSV <sup>a</sup> Correction	With TSV Correction
PUBLIC AND PRIVATE RESEARCH	Investment <sup>b</sup>
$A_1 = 0.068, a_2 = 0.136,$	$A_1 = 0.1618$ , $a_2 = 0.3236$ ,
$a_3 = 0.3472^{\circ}$	$a_3 = 0.3236$
$B1_{95} = 0$ , $B2_{95} = 152.45^{d}$	$B1_{95} = 0, B2_{95} = 9,876.32$
$B1_{07} = -535.064.$	$B1_{07} = -605.736.$
$B2_{97} = -6,780.78$	$B2_{97} = -7,496.56$
PUBLIC RESEARCH INVESTMENT A	ALONE
$A_1 = 23.15, a_2 = 46.3,$	$a_1 = 49.31, a_2 = 98.62,$
$a_3 = 123.52$	$a_3 = 98.62$
$B1_{95} = 0, B2_{95} = 118.16$	$B1_{95} = 0, B2_{95} = 10,071.24$
$B1_{97} = -535,064,$	$B1_{97} = -605,738,$
$B2_{97} = -6,780.78$	$B2_{97} = -7,496.56$
<i></i>	<i>.</i>

<sup>a</sup> TSV: Taura Syndrome Virus

<sup>&</sup>lt;sup>b</sup> Public research investment (ha<sup>-1</sup> yr<sup>-1</sup>): \$5.39 (1993–1994 work plan), \$5.63 (1994–1995 work plan), \$7.46 (1995–1996 work plan). Private research investment (ha<sup>-1</sup> yr<sup>-1</sup>) assumed to be \$1,792 from 1993 to 1997.

<sup>&</sup>lt;sup>c</sup> "a<sub>t</sub>" represents the marginal impact on current output of research completed  $\tau$  years ago. <sup>d</sup> (B1<sub>1</sub> - B1<sub>2</sub>) > (<) 0 indicates input-saving (-using) bias due to technical change in the PL stocking input, measured in number of PL ha<sup>-1</sup>, from year t to year s (Cox et al., 1997). Similarly, (B2<sub>t</sub> - B2<sub>s</sub>) gives the bias effect due to technical change for the feeding input measured in kg ha<sup>-1</sup>.

(farms 8 and 9 in Table 1). Survey data indicate that both these farms did not reduce feeding rates over the 1995-97 time period. However, the remaining farms in the empirical analysis had been reducing feeding rates over time. The low TSVrelated yield during 1995-97 and the nondecreasing feeding rate for farms 8 and 9 explain the negative feed-input bias B2<sub>97</sub>. These results are further supported by the stocking and feeding PFPs for farms 8 and 9 in Table 1. Data from both farms indicate that the PFP for PLs and feed declined from 1995 to 1997, implying more inputs were being used during 1997 for less than a proportionate increase in output. Hence, the negative input bias indicated by B1<sub>97</sub> and B2<sub>97</sub> reflects 1) higher stocking rates adopted by producers to reduce TSV-related losses and 2) nondecreasing feeding rates of farms 8 and 9 over the 1995-97 time period, respectively.

The Total Factor Productivity index (TFP) measures, at any point in time, the total output produced by a unit of an aggregate input (Alston et al., 1998). The nonparametric TFP index, which can be derived from  $a_1$ ,  $a_2$ , and  $a_3$  indicates the extent of technical progression (or regression) over time. The nonparametric TFP index at time t2 using time t1 as a reference is given by

$$\left[1 + \frac{(A_{t2} - A_{t1})}{y_{t1}}\right]$$
 where

 $y_{t1}$  is the output level in time period t1 (Chavas and Cox, 1992).

Using 1990 as a base year, nonparametric TFP indices are reported in Table 4 for 1995 and 1997 only, because A<sub>t</sub>s are zero for all other years. The increasing value of the TFP index from 1995 to 1997 gives evidence of technical progress due to research investment.

The internal rate of return (IRR) is a parameter that is often used to evaluate the profitability of a research program. The research program is said to be profitable if the internal rate of return is greater than the rate of return associated with the opportunity costs of the research funds (Alston et al., 1998).

Table 4. Nonparametric total factor productivity (TFP) indices for the years 1995 and 1997. Given a base year of 1990, the nonparametric TFP index for 1995 measures the relative shift in the production function in 1995 when compared to 1990. Therefore, the 1995 TFP index measures the proportionately higher output produced in 1995 than in 1990 due to technological improvement, keeping the amount of effective inputs identical in both years. Similar interpretation is applicable for the 1997 TFP index.

Source of Research Funds	Without TSV <sup>a</sup> Correction	With TSV Correction
PUBLIC AND PRIVATE	Research Investment	
1995	1.08	1.19
1997		1.97
PUBLIC RESEARCH IN	VESTMENT ALONE	
1995	1.08	1.18
1997	1.73	1.97

a TSV: Taura Syndrome Virus

Given that  $a_{\tau}$  is the marginal impact of research on output, i.e.,

$$a_{\tau} = \frac{\partial y_t}{\partial R_{t-\tau}},$$

it could also be interpreted as the marginal physical product of the "research" input. Hence, following Chavas and Cox (1992) and Bredahl and Peterson (1976), the IRR can be expressed as:

$$\sum_{\tau=1}^{T} \left[ p \frac{\partial y_{t+\tau}}{\partial R_{\tau}} / (1 + IRR)^{\tau} \right] = 1$$
(7)

where

T = maximum lag length.

IRRs are reported for the four empirical scenarios in Table 5. The estimated IRR to the total research investment (i.e., combined public- and private-sector investments) was 18% (or 45%, with the adjustment for TSV-related shrimp losses). Both these IRR estimates indicate a favorable return to the investment in research. If the private-sector research costs are excluded and returns to the public-sector research expenditures alone are calculated, the resulting IRRs are very high. This is due to the high percentage of research costs that have been borne by the shrimp industry. Indeed, these results illustrate a very effective leveraging of US federal research dollars with private-sector capital.

# CONCLUSIONS

Relevant conclusions from this study include evidence of technological progress in Honduran shrimp production from the middle to the late 1990s. The nonparametric TFP indices showed that yields have been proportionately higher in the post-research years. The technological innovations from the 1980s to the middle/late 1990s allowed producers to apply shrimp feed more efficiently (i.e., with higher PFPs). Thus, equivalent yields were obtained at lower feed rates and lower costs. Other evidence indicated that between 1995 and 1997 producers were induced to stock at higher rates without receiving proportionately higher returns in output. This change is attributed to the onset of TSV in Honduras due to the reported tendency of producers to significantly increase PL stocking rates to allow for the high mortality rate that is typical of TSV infestation. However, in spite of TSV, TFP indices increased from 1995 to 1997, indicating continued technical progress.

The private-sector investment was considerably higher than the public-sector investment, which can be an indicator of the importance that commercial shrimp farms place on the PD/A CRSP. This sharing of public- and private-sector funding responsibilities supports the notion that the research benefits

Table 5.Internal rates of return for public and private research<br/>investment and public research investment alone with and<br/>without Taura Syndrome Virus (TSV) correction.

Source of	Without TSV	With TSV
Research Funds	Correction	Correction
Public and Private Research Investment Public Research Investment Alone	18% 6,352%	45% 13, 412%

spill over both spatial and temporal boundaries, and the beneficiaries often include the producers, consumers, and scientists who perhaps do not live in the same region, country, or time (Alston et al., 1998). Taking into account both private and public research investment, the IRR was 18% without accounting for TSV and 45% with adjustments for TSV. Extremely high rates of return to public research expenditures alone reflected effective leveraging of public research with private research funds to generate technological progress in shrimp production in Honduras.

# **ANTICIPATED BENEFITS**

This study indicated a positive and profitable return on the investment of the PD/A CRSP in shrimp research in Honduras. This level of return, especially since it was measured only a few years after the actual investment should encourage bi- and multi- lateral agencies to fund aquaculture research initiatives.

#### **ACKNOWLEDGMENTS**

The authors wish to thank Bart Green and David Teichert-Coddington for numerous discussions on study assumptions and the factors that affect management of shrimp ponds. Green provided invaluable assistance with the logistics of conducting the survey. Ing. Alberto Zelaya and the Asociación Nacional de Acuicultores de Honduras (ANDAH) provided support and assistance with data collection. Ari Mwachofi, Nathan Stone, Bart Green, and David Teichert-Coddington provided valuable review comments. This study was supported in part with financial support from the Pond Dynamics/Aquaculture Collaborative Research Support Program. This paper is published with the approval of the Director, Arkansas Agricultural Experiment Station, manuscript # 99064.

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# PD/A CRSP Seventeenth Annual Technical Report

# **RISK ANALYSIS OF SHRIMP FARMING IN HONDURAS**

Eighth Work Plan, Marketing and Economic Analysis Research 2 (8MEAR2) Final Report

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# Abstract

Honduras has established itself as the leading producer of pond-raised shrimp in Central America. Although this activity already represents the third staple of the national economy, relatively few economic analyses have been conducted to date. For this study, data on production of farm-raised shrimp were collected from 21 farms. Data are from the year 1997. Information was collected on technical aspects of shrimp culture (stocking densities, feeding rates, FCRs) as well as on financial performance of the farms (production costs, farm revenue) during the considered period. A risk analysis was carried out from the resulting data. Three scenarios were defined according to farm size and a fourth was created to aggregate farms with uncommonly high yields. Scenarios were defined in order to identify possible differences in management strategies. Simulations for this study were run with commercially available risk analysis software. Results indicated that farms of the last scenario have developed a major potential for profit, far greater than that of those farms adopting more conservative approaches. Risk is more associated with low yields than with high production costs. Regardless of size, farms should target a minimum acceptable yield. Annual production of less than 450 kg ha<sup>-1</sup> is connected with a large potential for loss.

# INTRODUCTION

Honduras is the major producer of farm-raised shrimp in the Central American region with 12,000 metric tons (live weight) produced on 14,000 ha of shrimp farms during 1998 (Rosenberry, 1998). The first commercial shrimp operations in Honduras began in 1980 (Weidner, 1991), and the industry entered into a period of rapid growth in 1987 when new farms were opened and existing operations were expanded. Annual shrimp exports increased steadily for several years, but production declined in 1994 when the first incidences of Taura Syndrome Virus (TSV) were reported in Honduras. Some analysts (ANDAH, 1997) point out that the industry may have already entered a stage of maturity and that profitability of operations in the future will depend largely on the efficient use of available resources.

Shrimp farming has grown at an accelerated pace worldwide. The rapid growth of the industry has been accompanied by fluctuating prices and quantities supplied that have contributed to an unstable market. The collapse of the shrimp farming industries in China, the Philippines, and Taiwan due to diseases caused by overstocking and the lack of rotation of ponds further contributed to the instability in the shrimp market.

Continued problems with disease and environmental concerns have encouraged shrimp farmers to evaluate traditional management practices and to seek to reduce dependence on external resources such as abundant supplies of wild seed and clean estuarine waters. There is a recognized need for tools that quantify the uncertainties and risks associated with shrimp production.

Optimization models have been developed for shrimp farming in Panama (Pérez, 1986), Ecuador (Dunning, 1989), and

Honduras (Stanley, 1993). Using linear programming, these models have been used to determine profit-maximizing stocking densities and production scheduling for representative farms. Hatch et al. (1987) evaluated shrimp production in Panama with a MOTAD (Minimization of Total Absolute Deviations) risk model that considered the worst possible outcomes (e.g., shrimp are hit by a disease and the final survival rate is 0%).

The primary objective of this study was to analyze the profitability of shrimp farming in Honduras under conditions of risk and uncertainty. Specifically, enterprise budgets were developed for typical farm situations and management options without accounting for risk. Finally, the effect of risk on profitability was evaluated through Monte Carlo simulation.

Results of the risk analysis are intended to provide the farm manager-decision maker with a compendium of possible outcomes that could be obtained under different scenarios, which are modeled according to the characteristics of the Honduran shrimp industry. This information can be used to redirect current practices to either minimize the impact of operation failures or improve the chances of an increased profit.

# MATERIALS AND METHODS

A direct, personal survey of shrimp farms was conducted in Honduras to obtain information on production costs and technical aspects of shrimp culture such as stocking densities, feeding rates, and FCRs. A stratified random sample of 21 farms was drawn from the sampling universe of 67 commercial shrimp growers identified by the National Aquaculture Association of Honduras (ANDAH). Farms were stratified by size based on previous studies (Aguirre and Torres, 1991) and

Farm Size	Farm	S	Area		
	Number	%	ha	%	
10 to 150 ha 150 to 400 ha	44 17	66 25	2,710.13 4,605.75	22 37	
Total	67	9	12,405.16	41	

#### Table 1. Size distribution of shrimp farms in Honduras.

Source: ANDAH (1997).

by the current size distribution of shrimp farms in Honduras (Table 1). Farms of similar size were substituted for those selected in the original sample that refused to participate. Survey data were entered into spreadsheets, summarized, and cross-tabulated. Figure 1 displays the proportion of shrimp farms in the sample in terms of the number of farms and the production area.

Representative enterprise budgets were developed for each farm size group based on the survey data using standard budgeting techniques (Kay and Edwards, 1994). Values used in the enterprise budgets were means for a given parameter for each respective farm size. A fourth enterprise budget was developed for some large- and medium-sized farms having yields greater than 1,250 kg ha<sup>-1</sup> yr<sup>-1</sup>. Stocking and feeding rates and production costs are also higher for this group of farms. These differences in production, management, and cost characteristics made it necessary to develop a separate enterprise budget. To facilitate denomination of scenarios for the risk analysis, the last group of farms are called "intensive," even though in shrimp aquaculture this term is applied worldwide to units producing more than 4,500 kg ha<sup>-1</sup> yr<sup>-1</sup> (Fast, 1992). Operations of this type are rare in Latin America since a high capital input is needed in response to constraints in availability of land, water, and cheap labor. Although by definition the term "intensive" is not used correctly in this study, it serves to illustrate differences among scenarios.

The risk analysis was conducted as a stochastic simulation using Crystal Ball<sup>™</sup> software. This is a spreadsheet add-in program that allows the incorporation of uncertainty in risk analysis models. Previous applications of this program to aquaculture situations include the stochastic model of a summer flounder firm developed by Zucker and Anderson (1999). In the simulations, ranges of values that individual variables or parameters may take are defined by probability distributions instead of the single mean values used in standard enterprise budgets. Monte Carlo simulation techniques (using 500 iterations) are used to generate values for individual cost and quantity parameters based on the probability distributions. Results present the entire range of possible outcomes and the likelihood of achieving them.

Different distribution forms were selected for different parameters based on availability of data and with input from professionals with long-term experience with the shrimp industry in Honduras. Table 2 summarizes the choice of distributions for each item in the enterprise budgets and the correspondent values selected as distribution parameters.



Figure 1. Distribution of shrimp farms in Honduras by size range: small (10 to 150 ha), medium (150 to 400 ha), and large farms (more than 400 ha). Bar height indicates number of farms and total production area for each size group. Upper portion of the bars represents number and production area of the farms included in the survey.

Production area (ha) within each farm size category was defined with a uniform distribution: all values between the minimum and maximum occur with equal likelihood, and minimum and maximum values correspond to the limits for each size range. Normal distributions were used to define shrimp yields and prices. These parameters are highly variable and influenced by many factors. Yield is determined by stocking densities, feeding rates, cycle length, and overall survival, but is also influenced by weather patterns that fluctuate randomly. Farm prices depend on average shrimp size, marketing strategies, and supply-demand interactions in the international market at the moment of harvest. These uncertain variables can be described by a normal distribution. Within each farm size category, the mean and standard deviation values for yields and shrimp prices were used to define the probability distributions.

Costs were described by triangular distributions based on the most likely value (which was used in the enterprise budget) and minimum and maximum values determined from the original data for each scenario. In general, minimum values were calculated by assuming that the smallest farm of each size group has the lowest usage rate of a given resource and pays the lowest possible price for it. Likewise, maximum values are obtained under the assumption that the largest farm of each size group will have the highest rate of input usage and will acquire resources at the highest price. Occasionally, minimum values were zero (e.g., fertilizer and electricity costs) since at least one of the farms within each scenario did not report the usage of the respective input. Blank spaces in Tables 3 through 6 indicate that no corresponding information was obtained from any of the farms within the respective farm group. An additional cost variable ("others") was included to account for costs such as security expenses, insurance payments, and estimates of losses by bird depredation and poaching. Most farm managers expressed that expenses of this type can be considerable. The variable ("feed costs") was divided into two categories (feed quantity and feed price) since a negative correlation was found between farm size and price paid per feed unit. Correlations between variables need to be defined before running the simulations for the risk analysis. Crystal Ball<sup>™</sup> normally calculates values independently of other values. Therefore, results could be biased if existing dependencies between variables are not accounted for. In addition to feed price (r = -0.65), production area was found to be correlated with six other variables: seed costs (r = 0.94), feed quantity (r = 0.98), full-time labor (r = 0.71), diesel costs (r = 0.98), debt payment (r = 0.65), and infrastructure depreciation (r = 0.75). These costs increased as a function of farm size. Correlation coefficients were calculated from data of the 19 semi-intensive farms and included in the model for each scenario. Other costs such as fertilizer, part-time labor, and electricity were not related to farm size, but varied according to factors such as management strategies, natural fertility of pond water, and electricity availability.

Correlation coefficients could not be calculated for the intensive farm scenario due to the low number of observations. Crystal Ball<sup>™</sup> generated random numbers for each cell independently of the values used for others.

The likelihood of achieving profit (positive net returns) and the distribution of outcomes for total revenue, total costs, breakeven yield, and breakeven price were calculated for each farm scenario. Overlay and trend charts were developed to compare the distribution of outcomes among farm scenarios and to draw overall conclusions from the risk analysis.

# **RESULTS AND DISCUSSION**

# **Enterprise Budgets**

Tables 3 through 6 present the enterprise budgets developed for each farm scenario: small, medium, large, and intensive farms. Across the four scenarios, yields from the surveyed farms ranged from 250 to 1550 kg ha<sup>-1</sup> yr<sup>-1</sup>. These production levels are characteristic of semi-intensive systems in shrimp aquaculture (Fast, 1992).

It is observed from Table 5 that large farms (more than 400 ha) typically produce less than 450 kg ha<sup>-1</sup> yr<sup>-1</sup> and have the lowest cost per hectare and break-even yield (Lps. 34,248 ha<sup>-1</sup> and

Table 2.Assumptions used for the risk analysis of shrimp farming in Honduras. Parameter distributions are given for each variable and scenario.Estimates are based on production data of 21 farms for the year 1997.

Variable	Unit	Distribution	Parameter	er Farm-Size Scenario			Intensive Farms
			-	10–150 ha	150–400 ha	> 400 ha	
Production Area	ha	Uniform	Minimum	20	150	400	150
			Maximum	150	400	2000	800
Yield	kg head-off	Normal	Minimum	0	0	0	0
	shrimp ha <sup>-1</sup>		Maximum	1,510	1,202	680	1,860
	yr-1		Mean	675	724	410	1,426
			Standard Deviation	279	152	159	142
Shrimp Price	Lps kg <sup>-1</sup>	Normal	Minimum	57	66	66	84
•	1 0		Maximum	154	139	121	157
			Mean	106	103	92	120
			Standard Deviation	15	11	7	11
Seed Costs	Lps kg <sup>-1</sup>	Triangular	Minimum	20,000	150,000	320,000	1,575,000
	1 0	0	Maximum	6,000,000	19,200,000	48,000,000	25,600,000
			Likeliest	1,241,791	5,697,705	10,592,895	11,679,531
Feed Quantity	MT	Triangular	Minimum	21	170	635	680
- ,		0	Maximum	354	816	4,536	5,806
			Likeliest	119	453	1,651	3,003
Feed Price	Lps MT <sup>-1</sup>	Triangular	Minimum	6,614	4,409	3,748	5,666
	1	0	Maximum	8,818	7,937	4,850	6,922
			Likeliest	7,684	6,083	4,365	6,283
Fertilizer	Lps	Triangular	Minimum	0	0	0	-
Costs	1	0	Maximum	165,000	420,000	1,300,000	-
			Likeliest	46,400	192,324	141,522	-
Chemical Costs	Lps	Triangular	Minimum	, 0	, 0	0	-
	1	0	Maximum	79,500	88,000	260.000	-
			Likeliest	11.520	40,524	26,568	-
Labor, Part-	Lps	Triangular	Minimum	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	0	0	-
time	1	0	Maximum	495,000	1,104,000	9,000,000	-
-			Likeliest	54,202	141,136	2,077,097	-

Table 3. Annual enterprise budget of a 73-ha shrimp farm (representative of the smallest size category, 10 to 150 ha) in Honduras. Estimates are based on production data of eight farms during 1997. US\$1 = 13 Lps.

Item	Unit	Quantity	Price	Total
Yield Shrimp Price Gross Receipts	kg head-off shrimp ha <sup>-1</sup> yr <sup>-1</sup> Lps kg <sup>-1</sup> Lps	675	106.00	5,223,150
VARIABLE COSTS (VC)				
Seed Lab PL Wild PL Total Feed Fertilizer Chemicals Labor, Part-time Labor, Full-time Diesel Gas Electricity Equipment Repairs	1,000 1,000 MT kg kg h annual salary l l kWh Lps yr <sup>-1</sup>	$9,432 \\ 15,718 \\ 25,150 \\ 119 \\ 14,018 \\ 320 \\ 8,672 \\ 23 \\ 31,583 \\ 1,572$	$\begin{array}{r} 65.00\\ 40.00\\ 7,684.00\\ 3.31\\ 36.00\\ 6.25\\ 21,797.22\\ 4.23\\ 6.08\end{array}$	613,057 628,734 1,241,791 935,816 46,400 11,520 54,202 501,336 133,596 9,558 - 93,714
Levee Repairs	Lps yr <sup>-1</sup>			289,293
Fixed Costs (FC)				3,317,226
Interest on Operating Capital Depreciation Concession Others	Lps yr <sup>-1</sup> Lps yr <sup>-1</sup> Lps ha <sup>-1</sup> Lps yr <sup>-1</sup>	88	17.45	908,403 564,319 1,529 86,748
Total FC				1,560,999
TOTAL COSTS (TC)				4,878,225
Cost per Hectare Net Returns (NR) Net Returns per Hectare Ratio NR/TC (%)	Lps ha <sup>-1</sup> Lps Lps ha <sup>-1</sup>			66,825 344,925 4,725 7
dreak-even price Break-even Yield	Lps кg kg ha <sup>-1</sup> yr <sup>-1</sup>			630

372 kg ha<sup>-1</sup> yr<sup>-1</sup>, respectively). Given the large number of production units (ponds), profitability of these operations is mainly dictated by keeping costs low. This indicates that managers of these farms have a marked preference for conservative strategies that minimize the potential for losses. If presented alternative production strategies, these managers are expected to assume risk-averse attitudes.

Yields are higher for medium farms (150 to 400 ha) than for large farms (Table 4). On average, these farms produce over 700 kg ha<sup>-1</sup> yr<sup>-1</sup>. Costs per hectare increase (Lps. 57,796 ha<sup>-1</sup>), but so do net returns (from Lps. 3,472 ha<sup>-1</sup> for large farms to Lps. 16,776 ha<sup>-1</sup> for medium farms). On average, yields for small farms (less than 150 ha) are about the same (675 kg ha<sup>-1</sup> yr<sup>-1</sup>) as those for medium farms, but greater variability was found in the survey data. Some of these operations are owned by small, independent producers. These farmers are not financially secure and are very cautious when presented alternative, riskier strategies. They do not benefit, as larger farms do, from the effects of the economies of scale and vertical integration. In general, large farms rely on their own processing plants, hatcheries, and broker services overseas. Undoubtedly, this represents a competitive advantage over small, family-owned operations.

Total costs of intensive farms in Honduras are higher by 40% than those of the largest farms, even though the former are about half the size of the latter (Tables 5 and 6). Differences are even larger in terms of revenue. Gross receipts of intensive farms (Lps. 79,741,920) are more than double those of the largest farms (Lps. 36,437,520). Obviously, costs per unit area and break-even yields are higher for the intensive farms, but net returns, both total (Lps. 33,937,849 ha<sup>-1</sup>) and per unit area (Lps. 72,828 ha<sup>-1</sup>), also increase in an appreciable way, as a function of the high production input.

Data from the enterprise budgets suggests that farmers should intensify activities within the reach of their capabilities. Medium and intensive farms invest more per unit area than larger farms do, so they obtain higher returns per hectare, and Table 4.Annual enterprise budget of a 293-ha shrimp farm (representative of the medium farm-size category, 150 to 400 ha) in Honduras.Estimates are based on production data of eight farms during 1997. US\$1 = 13 Lps.

The second	TT */		Datas	T-1-1
Item	Unit	Quantity	Price	Total
Yield Shrimp Price Gross Receipts	kg head-off shrimp ha <sup>-1</sup> yr <sup>-1</sup> Lps kg <sup>-1</sup> Lps	724	103.00	21,849,596
VARIABLE COSTS (VC)				
Seed Lab PL Wild PL Total Feed Fertilizer Chemicals Labor, Part-time Labor, Full-time Diesel Gas Electricity Equipment Repairs Levee Repairs	1,000 1,000 MT kg kg h annual salary l l kWh Lps yr <sup>-1</sup> Lps yr <sup>-1</sup>	72,62524,42797,05245358,1041,22817,64250191,8728,33277,381	$\begin{array}{c} 65.00\\ 40.00\\ \end{array}\\ 6,083.00\\ 3.31\\ 33.00\\ 8.00\\ 42,857.00\\ 4.04\\ 6.07\\ 1.40\\ \end{array}$	4,720,625 977,080 5,697,705 2,755,599 192,324 40,524 141,136 2,142,850 775,163 50,575 108,333 448,952 548,538 12,901,699
Total VC				
Interest on Operating Capital Depreciation Concession Others	Lps yr <sup>-1</sup> Lps yr <sup>-1</sup> Lps ha <sup>-1</sup> Lps yr <sup>-1</sup>	352	14.63	1,958,296 1,385,979 5,150 683,153
Total FC				4,032,578
TOTAL COSTS (TC)				16,934,277
Cost per Hectare Net Returns (NR) Net Returns per Hectare Ratio NR/TC (%) Break-even Price Break-even Yield	Lps ha <sup>-1</sup> Lps Lps ha <sup>-1</sup> Lps kg <sup>-1</sup> kg ha <sup>-1</sup> yr <sup>-1</sup>			57,796 4,915,319 16,776 29 80 561

the ratio between net returns and total costs is more favorable (10, 29, and 74% for large, medium, and intensive farms, respectively; Tables 4, 5, and 6). However, costs per unit area are high for small farms but the ratio of net returns to total costs is low (7%; Table 3). Several factors account for this: some of the small farms reported losses in the survey, mostly because of low survival related to disease outbreaks. In addition, these farms pay a higher price per unit of feed. Feed is usually the most expensive component of the cost structure of shrimp farming (Shang, 1992). From the survey data, it is evident that feed mills offer discounts in feed price that vary with the amount of feed ordered. This is an arrangement that represents a considerable reduction of costs for large farms, but from which small farms do not derive any benefit whatsoever. The correlation between farm size and feed price was included when defining the variables for the risk analysis.

High yields of intensive farms appear to be connected with moderate to high stocking densities along with strong reliance on supplemental feed (Table 6). Most of the stocked post larvae (PL) came from hatcheries, which is more expensive than wild PL. There is a considerable degree of risk associated with these practices, as diseases and other mortality factors can decimate shrimp stocks at any time. It is possible that the financial performance of these farms could be reflecting a case of isolated success.

# **Risk Analysis**

Table 6 summarizes the results of the simulations. The entire range of possible outcomes is given for each scenario (Range minimum – Range maximum). Certainty levels indicate the likelihood of achieving values within a specific range. In the case of net returns and net returns ha<sup>-1</sup>, certainty levels indicate the probability a farm has of achieving profit (positive net returns) given the scenario conditions. Certainties for total costs, break-even price, and break-even yield refer to these values' likelihood of being lower than the mean values of the enterprise budgets. As to gross receipts, the certainty level expresses this value's probability of being higher than the Table 5.Annual enterprise budget of a 966-ha shrimp farm (representative of the largest size of shrimp farms, more than 400 ha) in Honduras.Estimates are based on production data of three farms during 1997. US\$1 = 13 Lps.

Item	Unit	Quantity	Price	Total
Yield Shrimp Price Gross Receipts	kg head-off shrimp ha <sup>-1</sup> yr <sup>-1</sup> Lps kg <sup>-1</sup> Lps	410	92.00	36,437,520
VARIABLE COSTS (VC)				
Seed Lab PL Wild PL Total Feed Fertilizer Chemicals Labor, Part-time Labor, Full-time Diesel Gas Electricity Equipment Repairs Levee Repairs	1,000 1,000 MT kg kg h annual salary l l kWh Lps yr <sup>-1</sup> Lps yr <sup>-1</sup>	78,927 $136,566$ $215,493$ $1,651$ $42,756$ $738$ $259,637$ $82$ $856,894$ $16,302$ $197,087$	65.00 40.00 4,365.00 3.31 36.00 8.00 47,685.00 3.78 6.08 1.40	5,130,255 5,462,640 10,592,895 7,206,615 141,522 26,568 2,077,096 3,910,170 3,239,059 99,116 275,921 144,675 213,928
Total VC				27,927,565
FIXED COSTS (FC)				
Interest on Operating Capital Depreciation Concession Others	Lps yr <sup>-1</sup> Lps yr <sup>-1</sup> Lps ha <sup>-1</sup> Lps yr <sup>-1</sup>	1,160	3.54	2,443,736 2,166,578 4,109 541,667
Total FC				5,156,090
TOTAL COSTS (TC)				33,083,655
Cost per Hectare Net Returns (NR) Net Returns per Hectare Ratio NR/TC (%) Break-even Price Break-even Yield	Lps ha <sup>-1</sup> Lps Lps ha <sup>-1</sup> Lps kg <sup>-1</sup> kg ha <sup>-1</sup> yr <sup>-1</sup>			34,248 3,353,865 3,472 10 84 372





Figure 2. Probability distribution of net returns for a small (10 to 150 ha) shrimp farm in Honduras. Likelihood of achieving profit = 13%. Values between arrows are positive (> 0) and indicate certainty of achieving profit.



Table 6. Annual enterprise budget of a 466-ha intensive shrimp farm in Honduras (production level of more than 1,250 kg ha<sup>-1</sup> yr<sup>-1</sup>). Estimates are based on production data of two farms during 1997. US\$1 = 13 Lps.

tem Unit		Quantity	Price	Total
Yield Shrimp Price Gross Receipts	kg head-off shrimp ha <sup>-1</sup> yr <sup>-1</sup> Lps kg <sup>-1</sup> Lps	1,426	120.00	79,741,920
VARIABLE COSTS (VC)				
Seed Lab PL Wild PL Total Feed Fertilizer Chemicals Labor, Part-time Labor, Full-time	1,000 1,000 MT kg kg h annual salary	146,091 7,689 153,780 3,003 80 (28.10)	78.00 37.00 6,283.00 43,609.00	11,395,038 284,493 11,679,531 18,867,849 - - 3,488,720
Diesel Gas Electricity Equipment Repairs Levee Repairs	l kWh Lps yr <sup>-1</sup> Lps yr <sup>-1</sup>	638,106 43,830	5.02	3,203,292 254,652 - 2,260,624 1,242,599
Total VC				40,997,267
Fixed Costs (FC)				2,508,306
Interest on Operating Capital Depreciation Concession Others	Lps yr <sup>-1</sup> Lps yr <sup>-1</sup> Lps ha <sup>-1</sup> Lps yr <sup>-1</sup>	1,050	25.00	1,074,331 26,250 1,197,917 4,806,804
Total FC				
TOTAL COSTS (TC)				45,804,071
Cost per Hectare Net Returns (NR) Net Returns per Hectare Ratio NR/TC (%) Break-even Price Break-even Yield	Lps ha <sup>-1</sup> Lps Lps ha <sup>-1</sup> Lps kg <sup>-1</sup> kg ha <sup>-1</sup> yr <sup>-1</sup>			98,292 33,937,849 72,828 74 69 819



Figure 4. Probability distribution of net returns for a large (> 400 ha) shrimp farm in Honduras. Likelihood of achieving profit = 11%.

mean value of the budgets. In summary, Table 6 indicates the likelihood a farm has of presenting an adequate financial performance (positive net returns, high receipts, and low costs).

The objective of the simulations is to quantify the potential for profit given the characteristics of every farm scenario. Extreme values were generated for every variable with the goal of incorporating the worst and the most optimistic conditions under which a farm would operate, according to information from the survey. Results strongly favor the intensive farm scenario. The certainty for achieving profit is 74%, well above certainties for medium (24%), small (13%), and large farms (11%; Figures 2 through 4). Additionally, the intensive scenario is the only one where mean net returns (overall and per hectare) are positive (Figure 5). Potential for loss is clearly greater for the other three farm scenarios, as indicated by the negative mean net returns and the higher absolute value of range minima over range maxima. Certainty levels for attaining break-even prices and yields less than or equal to the actual value from the enterprise budgets are also greater for the

intensive farms (in the case of break-even yields, 37%, compared to 1% for the other three scenarios).

Figures 2 through 5 display probability distributions for net returns under different scenarios. All values within 2.6 standard deviations are included, which represents approximately 99% of the forecast values. Certainty levels are also indicated in the charts. A series of overlay charts is displayed in Figure 6. These charts superimpose frequency data from each forecast under the different farm-size scenarios. Data are grouped in reverse cumulative distributions. These charts show how distributions among farm scenarios are related to



Figure 5. Probability distribution of net returns for an intensive (> 1,250 kg ha<sup>-1</sup> yr<sup>-1</sup> yield) shrimp farm in Honduras. Likelihood of achieving profit = 74%. Values between arrows are positive (> 0) and indicate certainty of achieving profit.

each other. Distribution shape for intensive farms is clearly dominant (located farther to the right) for the gross receipts (Figure 6a) and net returns (Figure 6c) forecasts. This suggests that managers can expect to generate more revenue and increased net returns by adopting the strategies chosen by intensive farms. Likewise, it would be reasonable to assume that the range of total costs will be wider than for other scenarios. However, it is observed in Figure 6b that costs are likely to be higher for the large-farm scenario. This explains in part why the likelihood of profit is so low for large farms: under current conditions, operational costs could escalate at a faster pace than gross receipts, since there is a reduced potential for increasing yields.

Although cost distribution for intensive farms is greater than that for small and medium farms at any point, break-even prices are lower for the former (Figure 6e). This is due to the elevated yields of intensive farms. Break-even price is a function of total costs, yield, and area. Assuming that total costs will increase with increases in farm area, yielding more per unit area (the case of intensive farms) will determine a lower break-even price. Break-even yields are, however, high both for small and intensive farms (Figure 6f). Break-even yield is a function of total costs, price, and area. The distributions for small and intensive farms are similar since the difference in shrimp prices between these scenarios is reduced more than the difference in farm yields (Tables 3 and 6).

The net returns chart (Figure 6c) shows the small-farm curve



Figure 6. Reverse cumulative distributions for a) gross receipts, b) total costs, c) net returns, d) net returns ha<sup>-1</sup>, e) break-even price, and f) breakeven yield. Distributions from all four farm-size scenarios are shown in each chart.

located to the right of the curve for large farms. The relative position of these distributions changes in Figure 6d (net returns ha<sup>-1</sup>). This is due to the large potential for loss implicit in both scenarios. Dominance of the net return distribution for small farms in the first chart does not precisely imply that overall returns can be higher, but that losses can be smaller. However, on an areal basis (Figure 6d), it is clearly observed that small farms cope with larger losses.

Figure 7 provides another view of the forecasts. Certainty ranges are displayed in a series of patterned bands. These charts illustrate how the range of possible outcomes and their associated probabilities vary from scenario to scenario. The potential for profit of the intensive farms is related to their capacity to generate higher revenues at the same time that costs are restricted in range (Figures 7a and b). Large farms have in turn a wider distribution of costs (Figure 7b) even though the total cost value is lower for large farms in the enterprise budgets (Tables 5 and 6). Since intensive farms are in actuality producing shrimp at high costs, the model determines that there is not much room for additional expenses, which results in a narrower distribution of costs. Total costs for large farms can amount to more than 120 million Lps., while gross receipts do not surpass this number (Figures 7a and b). This situation occurs in part because of the potential for elevated costs as farm size increases and also reflects the nature of the data as one of the large farms reported losses in the survey.

The net returns chart in Figure 6c illustrates that returns are in general higher for small and medium farms than for large farms. However, the trend chart for net returns in Figure 7c

shows the range of positive net returns for large farms is greater than for small and medium farms. This suggests that large farms have a latent potential for obtaining a notable increase of profits by moderately intensifying production.

Potential for loss is magnified for the small- over the mediumand large-farm scenarios when the distribution of net returns is considered on an areal basis (Figure 7d). As mentioned before with Figure 6d, this is because there is a major likelihood that the distribution of overall net returns across the three scenarios will take on negative values.

Mathematical characteristics of the model determine the occurrence of extraordinarily high values for break-even prices in the small-farm scenario, over 10,500 Lps. kg<sup>-1</sup> (Figure 6e; Table 7). This is the case when a very small farm (e.g., 20 ha) has high production costs but the final yield is close to zero. Results of this type are generated in response to the procedure used to define the variables of the model. In consequence, the use of trend charts is impractical for making comparisons among scenarios for the break-even price parameter.

Net returns is the most common criterion under which management decisions are evaluated in business enterprises, shrimp aquaculture included. Nevertheless, net returns are rarely known with certainty, but are often associated with a distribution of possible outcomes that may include the potential for loss (Hatch et al., 1987). Commonly, strategies targeted to achieve a large profit also contemplate loss possibilities and may look less attractive than conservative choices associated with a narrower distribution of returns and a reduced potential for loss. Decisions depend ultimately on



Figure 7. Certainty ranges displayed in series of bands for a) gross receipts, b) total costs, c) net returns, and d) net returns ha<sup>-1</sup> across all four farmsize scenarios.

the manager's attitudes toward risk. However, results of the analysis indicate that under current strategies followed by farm managers in Honduras, there is more risk associated with adopting conservative practices and a low-cost-per-hectare approach than intensifying shrimp culture by increasing stocking densities and feeding rates. All farms, regardless of size, should target a minimum yield of 450 kg ha<sup>-1</sup> yr<sup>-1</sup>. As some of the costs are fixed, farmers could reduce the potential for loss by opting for more intensive strategies. This applies especially to the large farms characterized by low yields. Small farms are more affected by cost factors, such as the higher price of feed. Strategies for these farms should include alternatives that decrease the cost of production per unit area. Cooperatives or other types of agreements between small producers could be implemented to acquire feed at bulk price. Benefits of discounts would then cover a wider range of farms.

Some annotations should be made before stating final conclusions. Enterprise budgets were developed to describe the functioning of "typical" farms during 1997. There are two main production cycles every year in Honduras. These cycles are related to the alternation of dry and wet seasons. There are marked differences of productivity between cycles (Teichert-Coddington et al., 1994). The wet season is recognized as a more desirable period for shrimp grow-out. These differences were not accounted for in this study since the risk analysis is intended to examine the financial performance of the farms for an entire year.

Results of the model should not be considered as definitive. Low return levels for the first three scenarios do not necessarily imply that a new shrimp enterprise adopting similar strategies is doomed before initializing operations. The

Table 7.Forecast parameters and statistics generated from Monte Carlo simulations (500 iterations) of shrimp farming in Honduras.US\$1 = 13 Lps.

Forecast	Unit	Parameter	]	Farm-Size Scenario		Intensive Farms
			10–150 ha	150–400 ha	>400 ha	
Gross Receipts	Lps	Mean	6,022,939	20,715,118	43,193,907	79,680,829
-	-	Range minimum	27,234	6,250,731	2,351,653	23,305,090
		Range maximum	22,189,054	50,222,637	116,653,535	165,801,837
		Certainty level	52%	41%	57%	46%
		Certainty range	5,223,150 to ∞	21,849,596 to ∞	36,437,520 to ∞	79,741,920 to ∞
Total Costs	Lps	Mean	8,918,464	24,911,289	63,838,732	52,267,982
	_	Range minimum	1,894,472	7,343,892	21,650,616	25,9*86,764
		Range maximum	19,348,762	47,993,751	120,653,610	77,376,799
		Certainty level	11%	18%	7%	23%
		Certainty range	-∞ to 4,878,225	-∞ to 16,934,277	-∞ to 33,083,655	-∞ to 45,804,072
Net Returns	Lps	Mean	(2,895,525)	(4,196,171)	(20,644,825)	27,412,846
		Range minimum	(12,500,000)	(24,610,039)	(105,833,371)	(45,680,242)
		Range maximum	7,500,000	16,611,078	34,586,701	118,248,910
		Certainty level	13%	24%	11%	74%
		Certainty range	0 to ∞	0 to ∞	0 to ∞	0 to ∞
Net Returns	Lps ha <sup>-1</sup>	Mean	(42,064)	(14,751)	(18,985)	34,847
		Range minimum	(188,323)	(62,700)	(71,238)	(245,553)
		Range maximum	87,471	51,794	25,911	168,189
		Certainty level	13%	24%	11%	74%
		Certainty range	0 to ∞	0 to ∞	0 to ∞	0 to ∞
Break-Even Price	Lps kg <sup>-1</sup>	Mean	251	130	173	96
		Range minimum	64	44	55	25
		Range maximum	10,651	220	1,734	307
		Certainty level	12%	2%	5%	41%
		Certainty range	-∞ to 99	-∞ to 80	-∞ to 84	-∞ to 69
Break-Even Yield	kg ha <sup>-1</sup> yr <sup>-1</sup>	Mean	1,094	878	603	1,137
		Range minimum	528	468	319	263
		Range maximum	2,182	1,310	1,037	3,890
		Certainty level	1%	1%	1%	37%
		Certainty range	-∞ to 630	-∞ to 561	-∞ to 372	-∞ to 819

assumptions included very pessimistic estimates (extreme high costs, poor yields) that strongly influenced the results of the simulations. This is especially true of large and small farms. For both scenarios, Crystal Ball<sup>™</sup> recalculated the spreadsheet incorporating a certain likelihood of zero yields. Even though it was also considered, the probability of achieving a zero yield is negligible for the medium and intensive scenarios. This is due to the shape of the normal distributions, defined from the survey data.

In any case, the model is categorical as to establishing the wisdom of intensifying shrimp culture. Differences among scenarios are clearly identified and quantified by the analysis, and is here where the usefulness of the model should be found.

# **ANTICIPATED BENEFITS**

This study shows the importance of including risk in economic analyses. Without accounting for risk, management strategies recommended might not be the most appropriate.

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# DEVELOPMENT OF CENTRAL AMERICAN MARKETS FOR TILAPIA PRODUCED IN THE REGION

Ninth Work Plan, Marketing and Economic Analysis Research 3 (9MEAR3) Progress Report

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# Abstract

Marketing studies will be conducted in Honduras in Year 1 of this project to identify and characterize existing market channels for fish and seafood in Honduras. Profiles will be developed of the types of markets that currently sell tilapia in Honduras, and the factors related to increased tilapia sales will be determined. The factors that affect the likelihood of other markets adding tilapia products also will be determined. The survey instrument for the supermarket survey has been developed and is currently under review. The pre-test of the survey instruments will be conducted in August 1999. Contacts have been made with Escuela Agrícola Panamericana (Zamorano) and the Universidad Tecnológica de Honduras to identify a potential graduate student and enumerators to assist with the project. Interviews with candidates will be conducted in August 1999, and the surveys will be conducted from September through December 1999.

# INTRODUCTION

Tilapia culture was initiated in Honduras in the late 1970s (Teichert-Coddington and Green, 1997). In the early years, tilapia production was primarily a small-scale, family operation that was managed either extensively or semi-intensively as a supplemental agricultural activity. In 1995, Sarmiento and Lanza Nuñez found a total of 113.6 ha of small-scale, familylevel fish ponds (2,738 ponds) operated in every department (province) of Honduras.

Export-oriented production of tilapia began in 1990 and has grown rapidly since 1991-1992 (Teichert-Coddington and Green, 1997). In 1997, there were 15 tilapia farms with a total water surface of 185.3 ha that produced for export and domestic markets. These farms produce tilapia exclusively and are owned by individuals, local investors, and international investors (Green and Engle, in press). Exports of tilapia to the US from Honduras have grown consistently since 1992.

The rapid growth in tilapia production is expected to generate supply that could be available domestically in Honduras. The development of a strong domestic market for tilapia in Honduras could diversify market opportunities for tilapia growers and serve to stabilize this young aquaculture industry from the external shocks common in export-oriented markets. Furthermore, the development of a domestic market could enhance the income-generating potential of small-scale tilapia production.

A limited amount of work has been done on markets for finfish in Central America. The few studies that have been carried out focused on the catch from commercial fisheries in Panama (Matton, 1981) and in Costa Rica (Schied and Sutinen, 1979). Head et al. (1994) developed market guidelines for saltwatercultured Florida red tilapia in Puerto Rico. Several studies conducted in the US have examined the potential to develop markets for tilapia (Crawford et al., 1978; Nelson et al., 1983; Galbreath and Barnes, 1981). More recently, Swanson (1995) described US market requirements for tilapia. Engle (1997b) interviewed intermediate seafood buyers in the US to determine the potential to increase sales of fresh and frozen tilapia fillets in the US. However, virtually no work has been done on the potential to develop domestic markets in Central America for tilapia. Engle (1997a) describes the domestic markets that have emerged in Colombia for Colombian- and Ecuadorian-produced tilapia.

# METHODS AND MATERIALS

The survey instrument for the supermarket survey has been developed and is currently under review. The pre-test of the survey instruments will be conducted in August 1999. Contacts have been made with Escuela Agrícola Panamericano (Zamorano) and the Universidad Tecnólogica de Honduras to identify a potential graduate student and enumerators to assist with the project. Interviews with candidates will be conducted in August 1999, and the surveys will be conducted from September through December 1999.

Profiles will be developed of the types of markets that currently sell tilapia in Honduras, and the factors related to increased tilapia sales will be determined. The factors that affect the likelihood of other markets adding tilapia products also will be determined.

Analyses planned include logit and ordered probit analyses of maximum likelihood factors and variables, although the survey response rate may dictate the analyses used. The use of direct personal interviews by Honduran economists and their students is expected to generate an acceptable response rate that will allow us to perform logit and ordered probit analyses.

Descriptive reports summarizing key results of the crosstabulations conducted will be prepared in both English and Spanish.

# **R**ESULTS AND **D**ISCUSSION

We are entering the data collection phase of the project and there are, as yet, no results to be reported.

#### **ANTICIPATED BENEFITS**

This activity will provide guidelines to develop domestic markets for tilapia in Central America and reduce market risk by developing more diverse marketing strategies. More comprehensive market information will provide opportunities for Central American tilapia producers to take advantage of reduced transportation, storage, and handling costs by understanding volume, packing, and labeling requirements of the most important domestic market channels. This activity will provide information that will allow the tilapia industry in Central America to develop and access new markets, increase the volume of goods sold, diversify marketing strategies, and improve financial success of tilapia growers. Results of this activity will be published in English and in Spanish.

The primary direct beneficiary of this study will be the Central American tilapia industry. Tilapia growers will benefit from the market information made available to them and from the training on marketing strategies that will be provided through this activity. At the present time, growers are struggling to develop and maintain export markets without the benefit of comprehensive market information and in competition with other tilapia-exporting countries. This is particularly true for small-scale growers who have greater difficulty in meeting the volume and size requirements of export markets. The proposed training workshop will teach tilapia growers, small and large, how to develop and maintain markets so that they will be capable of adapting to changing market conditions. While the primary focus is on the Central American industry, information on how to develop a domestic tilapia market will also be of interest to Thailand, other Asian countries, and African nations wishing to develop domestic markets for new or non-traditional species as well as tilapia. The PD/A CRSP will be a secondary beneficiary because improvements in market development will result in industry expansion that will allow for greater adoption and need for PD/A CRSP research results. The overall impact of the PD/A CRSP will be greater as the industry grows in Central America. Honduran university students will also benefit from the experience and skills gained through participating in this project.

# **ACKNOWLEDGMENTS**

We thank Dan Meyer and Bart Green for helpful reviews of the survey instruments.

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# ECONOMIC AND SOCIAL RETURNS TO TECHNOLOGY AND INVESTMENT IN THAILAND

Ninth Work Plan, Marketing and Economic Analysis Research 4 (9MEAR4) Progress Report

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# Abstract

A survey will be conducted in northeastern Thailand to measure rates of adoption of CRSP-developed technologies. The rates of adoption will be used in a quantitative model to measure the internal rate of return to investment in aquaculture research in Thailand. This project follows work of the Eighth Work Plan that measured these returns to research for CRSP shrimp research in Honduras. Funding for this project was available as of July 1999.

#### INTRODUCTION

The Pond Dynamics/Aquaculture CRSP is a global research activity directed toward improving the reliability and efficiency of pond aquaculture production. The ultimate benefit of this effort will be the economic and social returns that represent the impact from farmers adopting new technologies developed by the PD/A CRSP.

Technical progress has been modeled as a lagged function of research expenditures (Chavas and Cox, 1992). This study identified and measured the length of time required to fully translate public research expenditures into economic benefits and estimated internal rates of return for research expenditures. In the Chavas and Cox model, there were no restrictions on substitution possibilities among inputs, joint estimation of the production technology, technical change, and the effects of research on technical progress using very disaggregate inputs. This approach required only a standard linear programming algorithm. Ayer and Schur (1972), Ardito-Barletta (1971), and others estimated social rates of return to the investment in public research.

Fischer et al. (1996) used a random-effects model within a Bayesian framework to analyze the effect of adoption of new wheat varieties in South Australia. Results showed that not all pieces of information added equally to knowledge about the innovation. It further showed that the acquisition of information was much slower than had been suggested by previous Bayesian models and could also explain laggards and partial adoption. Huang and Sexton (1996) developed a general imperfect competition model to evaluate returns to a costreducing innovation. In an imperfectly competitive market structure, this study showed that farmers' incentives to adopt a mechanical harvester for tomatoes in Taiwan were attenuated because the benefits were reduced by oligopsony power of processors.

Fuglie (1995) developed a multimarket model to explore equity and efficiency implications of improving crop storage

technologies. The rate of return to research on potato storage in Tunisia was estimated to be between 44 and 75%.

Dasgupta and Engle (2000) utilized nonparametric estimation techniques to estimate returns to the PD/A CRSP investment in shrimp research in Honduras to be from 18 to 45% for the combined public and private research investment.

# METHODS AND MATERIALS

This project began in the month that this report was written. To date, the authors have reviewed all PD/A CRSP reports on research conducted in Thailand and have compiled a list of the most important technological advances emanating from the CRSP-funded research initiatives. This list of technologies has been circulated to investigators who have been actively involved in the PD/A CRSP Thailand research program for ranking. When the most important technologies have been selected, the survey instrument will be developed, pre-tested in Thailand and the survey implemented. Final decisions on the form of the analytical model used will be based, in part, on the response rate and the number of usable observations obtained from the survey.

# **ANTICIPATED BENEFITS**

Results of this study will be useful for the PD/A CRSP to justify continued funding by quantifying benefits and impacts of the research effort. This study will provide the first estimates of the global social and economic returns generated by the PD/A CRSP. The results of this project will document the contribution that the PD/A CRSP research has made and will continue to make over time in both social and economic terms. This is essential to justify continued funding for the CRSP in the US and for host country support.

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# FISH CULTURE IN THE PERUVIAN AMAZON: PRODUCER PERCEPTIONS AND PRACTICES IN THREE RIVER SYSTEMS

Eighth Work Plan, Adoption/Diffusion Research 1-1 (8ADR1-1) Final Report

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#### Abstract

The Instituto de Investigaciones de la Amazonia Peruana (IIAP) is the leading governmental organization working in aquaculture and fisheries research in the Peruvian Amazon region. In addition, IIAP produces fingerlings, offers training courses, and works with nongovernmental organizations (NGOs) endeavoring to promote fish culture. This report summarizes fieldwork and survey results from rural communities in the Iquitos area of the Amazon served by NGOs assisted by IIAP. The researchers visited rural communities and interviewed fish farmers, community residents, and public and private agency officials to discover the strategies and approaches to small-scale, community-based aquaculture employed in the Peruvian Amazon. Subsequently, data were collected from a sample of 146 practicing fish farmers in the Napo, Tamishiyacu, and Tahuayo river systems areas north and south of Iquitos, as well as in the Iquitos-Nauta Road area directly south of the city. Fish farmers were identified in selected communities provided technical assistance in aquaculture by CARE/Peru and several other nongovernmental organizations. Results portray the species cultured, marketing strategies employed, and the perceived impact of fish culture on families and farming systems. The data show that fish farmers are in an advantageous situation for fish culture. They encounter few barriers to building ponds, obtaining fingerlings, feeding their fish, or marketing the product. Fruits and other forest-based fish foods are widely available to support extensive production systems. A number of NGOs are providing regular farm visits and advice on fish culture. The natural cycle of the Amazonian river systems ensures a market period of relatively high prices for farm-reared fish. Additional attention is needed on identifying and communicating production practices that will reduce risk and enhance the benefits of aquaculture.

#### INTRODUCTION

Loreto, Peru's largest department, is entirely forested except for water bodies and urban areas. Iquitos is a city of more than 300,000 people accessible only by boat or airplane. No roads connect the city in northeast Peru to Lima on the western coast. Founded during an earlier period of rubber boom (Barnham and Coomes, 1996), Iquitos is now a regional trade center that serves the many small communities that line the Amazon and its tributaries (Chibnik, 1994). Many humanitarian and environmental nongovernmental organizations (NGOs) operate in the Loreto area and are based in Iquitos (Rainforest Conservation Fund, 1999). The Peruvian Amazon has been subjected to large-scale commercial exploitation for the last two centuries. As Nauta, Tamishiyacu, and then Iquitos grew during the late 1800s, they became centers of urban consumption and international export. While fine rubber-jebe fino (Hevea brasiliensis)-was recognized worldwide, Peru produced large amounts of weak rubber-jebe debil-from upland varieties of Hevea sp. Coomes (1992b) provides an analysis of the rubber trade and the difficulties involved in the business. Over 3,000 metric tons was exported annually from the Peruvian Amazon between 1902 and 1917 (Villarejo, 1988). This export economy crashed after the Second World War, but the tire industry in Lima rejuvenated the rubber trade during

the 1960s and early 1970s. Petroleum-based tires have effectively ended the rubber trade in the northeastern Peruvian Amazon.

The Amazon River fishery plays a fundamental role in the livelihoods and survival of rural populations in this region (McDaniel, 1997). Fishing is by far the most important source of animal protein in the Amazon Basin and the main generator of cash for people living along the river. Araujo-Lima and Goulding (1997) argue that fishing is the most promising means for increasing animal protein within the Amazon Basin with a minimum of environmental degradation. Aquaculture plays a unique and dynamic role in the forest-and-river-based farming system of the Peruvian Amazon or Selva (Tomich et al., 1995; Pinedo-Vasquez et al., 1992).

The number of fish species in the Amazon hydrographic basin has been estimated at 2,000. Only 1,400 of these have been described scientifically. These represent approximately 10% of the planet's ichthyofauna (Rainforest Conservation Fund, 1999). The percentile distribution of the species in the main fish families is as follows: Sirulydeous 44%, Characoids 42%, Cichlidae 6%, and other species comprising 8%. Araujo-Lima and Goulding (1997) maintain that while *Colossoma macropomum* (called gamitana in Peru) is not the only Amazon fish species deserving of special attention, it is the first species about which enough is known to both manage wild stocks and develop aquaculture.

# **Fishing Strategies**

Successful fishing depends on the fisherman's knowledge of fish biology and physiology, of the peculiarities of the river or lake, and of water level variations. This knowledge and the necessary tools, which vary according to the species to be caught, have been largely developed by the Indians and inherited by the *ribereños*—the multi-ethnic river community dwellers. These native fishing techniques, which are carried out with bow and arrow, harpoon, spear, hooks, nets, traps, or even by poisoning parts of the floodplains with a plant poison (of the *Lonchocarpus, Phyllanthus*, and *Tephrosia* genera, which paralyzes the fish gill, forcing them to come up to breathe) do not damage the fishing stock significantly (Rainforest Conservation Fund, 1999; Hiraoka, 1986, 1989).

In Amazonia, the introduction of dip nets and their use in the flooded parts of the forest, the use of ice to store the fish, and the improvements in refrigeration and transport systems all led to increased exploitation of river fisheries. In turn, these technical innovations were a response to the increasing demand from foreign markets and facilitated exports. Boats with large-scale gear operated by urban interests are often a threat to stocks in local rivers and oxbow lakes. Moreover, fish with skin—traditionally not consumed by Indians and *ribereños*—began to be included in the number of edible fish. Consequently, predatory practices tended to replace traditional fishing techniques (Rainforest Conservation Fund, 1999).

The Amazon's fishing potential, previously regarded as inexhaustible, was overestimated. Though most waters in the Iquitos area are brown, black and clear water rivers are poor in fish. The fishing output of the Amazonian basin in 1980 was estimated at 150,000 t yr<sup>-1</sup>, but the fishing potential could be approximately twice this figure, that is, 300,000 t. This estimate encompasses all species and sizes, not only those with commercial value (Rainforest Conservation Fund, 1999).

Under these circumstances, it is foreseeable that fish supplies for the rapidly growing Amazonian population cannot be guaranteed for long. Management of fish for subsistence use or local consumption is done by the inhabitants of the areas around the lakes, whose interests in preservation conflict with those of the professional fishermen. The river people supplement fishing with subsistence agricultural activities, jute plantations, and extraction of wood and other products. Now aquaculture is widely perceived as a farm-based activity that complements traditional sources of food and livelihood.

# Aquaculture in the Selva

There is no fish-breeding tradition in Amazonia. The aboriginal populations kept fish, manatees, and turtles in large corrals for periodic consumption, but no techniques for reproduction in captivity were developed. Previous efforts by government agencies, NGOs, missionaries, and others, however, have led to a certain level of indigenous knowledge and interest in aquaculture.

There is a unique relationship between aquaculture and fisheries in many parts of the Selva (Hall, 1997). The abun-

dance of large, rapidly growing fish species supports an extensive capture fishery in the Amazon, its tributaries, and a large number of oxbow lakes. The fishery, however, is cyclic, as fishing is more difficult during the high water period of December through March. At this time, fish prices for some species are as much as twice the low water period price. This cyclical deficit in the supply of fish coupled with a widespread perception that river and lake fish stocks have declined and will continue to do so are the primary motivations for fish culture in the Selva. Commercial-scale fishers using large-scale fishing gear have depleted fish stocks in many oxbow lakes, further encouraging pond-based fish production.

Abundant supplies of warm water, generally available pond inputs, and easily obtainable grow-out stock are some of the favorable conditions for fish culture in the Amazon river system. Fingerlings can be obtained through individual effort in rivers or oxbow lakes or through purchase from fishermen. Longstanding efforts by the Department of Fisheries and other agencies that provided technical assistance in aquaculture has stimulated interest in fishponds. Current NGO efforts build on previous government efforts in some communities. In others, NGOs are introducing pond aquaculture as a new activity.

For example, the upper Tahuayo is mainly *várzea* forest with abundant and productive aquatic habitat. Nonetheless, many farmers on this other river systems construct the regionally popular *piscigranjas* (small dams on creeks or wetlands). While fish are scarce during some parts of the year and often difficult to catch in others, the flooding of the forest limits where farmers can build ponds.

An environmental NGO (Rainforest Conservation Fund) reported building a small pond project with some of its staff and members of the Chino Lakes ranger (vigilante) group. The environmental group's conclusions emphasized the risks involved in fish farming (theft, dike breakage, predators, etc.) and the high labor costs. It concluded that the construction of *piscigranjas* (fishponds) was unproductive on the Tahuayo. Nonetheless, CARE/Peru and other agencies are undertaking a program of pond construction in this and similar locales. Our fieldwork suggests that ponds are popular among farmers and do contribute to income and food security in these communities.

Fishing is not an important activity on the Tamishiyacu due to the lack of lakes and natural habitat for fish. While fish are abundant there, little commercial fishing occurs due to the lack of an accessible market. On the upper Tahuayo River, however, fishing is a very important economic and subsistence activity (Rainforest Conservation Fund, 1998).

On the upper Tahuayo, scores of ponds and swampy areas as well as large lakes and the river itself make fishing a primary economic activity. In this part of the buffer zone, seven lakes are presently, and have historically been, of utmost importance to the people here. These lakes, as well as the Tahuayo River, are blackish in color and support species considered to be "blackwater fish," but the pH and nutrient content of the water is similar to "white water" (see Coomes, 1992a, 1992b).

Rainforest Conservation Fund (1998) identifies a number of uses for the oxbow lakes. Charo *Cocha* (*cocha* is the word for "lake" in the local Quechua dialect) is over 500 ha in size, and supplies residents of Esperanza and the village of Charo with fish and income. Cunshicu *Cocha*, lying within the jurisdiction
of Buena Vista, has an area of 65 ha, and provides subsistence and income to Buena Vista and the village of Cunshicu. The NGO reports that some 100 families use these two lakes, with often-considerable pressure from non-locals who come from as far as the Amazon River and Iquitos.

The local people have long contested the use of freezer boats, illegal nets, traps, poisons, and explosives. Just upriver, five small lakes, ranging from 1 to 10 ha in size, are of vital importance to the village of Chino (population 310). Besides being subjected to the same problems as Charo and Cunshicu *Cocha*, their small size makes them extremely sensitive to environmental conditions (water levels, temperature, predators, etc.). The strong drop in the Tahuayo River's water levels during July to November nearly eliminates fish from these lakes. Consequently, the community of Chino has implemented an effective vigilance and management system since 1986 (Penn and Alvarez, 1990). These conditions underscore the importance of fishponds as a means for countering the cycles of the river.

The reciprocal relationship between fisheries and aquaculture in the Peruvian Amazon is further enhanced by the wellestablished patterns of fish marketing present in the region. Fish are a central part of the *ribereños'* diet, many species are accepted for consumption, and fish sales seem to be readily accomplished locally or at market centers. Tello's (1998) study of fish landings in Iquitos illustrates the diversity of fish in the markets and the centrality of *Prochilodus nigricans* (boquichico) as the most heavily harvested fish. *Colossoma macropomum* (gamitana) has had a steady, albeit slightly declining level of reported fish landings over the extended period of data that were available (see also Alcántara, 1994).

#### METHODS AND MATERIALS

#### Sample and Data Collection

Fish farmers were identified in selected communities that were provided technical assistance in aquaculture by CARE/Peru and several other NGOs in the Napo, Tamishiyacu, and



Figure 1. Study area near Iquitos, in the Department of Loreto in the Peruvian Amazon.

Tahuayo river systems that combine to form the Amazon, as well as in the Iquitos–Nauta Road area south of Iquitos (Figure 1). Structured interviews were conducted with a sample of 146 fish farmers having accomplished at least one harvest in the past two years (Casley, 1988; Townsley, 1996). The sample was drawn from NGO program participants in selected communities provided technical assistance in aquaculture by CARE-Peru and other selected NGOs.

The survey instrument was adapted from previous research conducted by Molnar et al. (1996) in five PD/A CRSP countries—Honduras, Thailand, the Philippines, Rwanda, and Kenya. The Peru survey, however, reflects the unique conditions and context of Amazonian fish culture, the diversity of species, and the singular relationship of aquaculture to the river fishery in the region. Ponds were identified in communities on three river systems north and south of Iquitos as well as the Nauta Road area south of Iquitos. Data collection took place in early 1999 and was conducted by graduate students from the Department of Fisheries at the Universidad Nacional de la Amazonia Peruana.

#### Analysis

The analysis tabulates the survey responses across the three locations—north, south, and central to Iquitos—where data were collected for the study. From this information, central patterns of comparison and difference in practice and approach to fish production and technology utilization can be discerned.

#### RESULTS

#### **Respondent Characteristics**

Table 1 describes the individual and household characteristics of study respondents. Women comprised about 45% of the respondents in Nauta Road, but only about one-tenth of the Napo River and Tamishiyacu fish farmers who we contacted. The Napo River farmers were somewhat older and the Tamishiyacu farmers slightly younger than those in the other locations.

Women and children are central beneficiaries of enhanced protein availability, income, and food security associated with fish ponds in the Selva. While pond construction labor seems primarily to be the province of men, it was sometimes reported that women provided pond inputs and participated in fish harvest.

Over 75% of the study households had children under age ten. Tamishiyacu families had the most children under age 10 and the smallest proportion with children over age 18. Tamishiyacu farmers also had the largest households, as 72% reported six or more members. About two-thirds of the respondent households in the other locations were that large.

Most respondents in the study were associated with individual family ponds. In some communities, NGO technicians worked with groups of families that shared pond construction labor and pond management responsibilities. About 14% of the study respondents were associated with such group ponds.

Few group or collective ponds were reported or encountered during our field visits, but the survey data show that group

Table 1.	Respondent characteristics,	fish farmers in t	the Peruvian Amazon, 1999.
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	Napo River $(N = 87)$	Nauta Road (N = 20)	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)
			(%)	
Gender of Respondent				
Male Female	92 8	55 45	87 13	86 14
Age of Respondent (yr)				
Less than 25 25 to 34 35 to 44 45 to 54 55 to 64 65 or Older	3 17 30 23 22 5	5 15 35 25 5 15	8 15 44 18 13 3	5 16 34 22 17 6
RESPONDENTS WITH CHILDREN				
Under Age 10 Age 10 to 18 Over Age 18	76 70 94	70 70 100	82 72 87	77 71 93
Two or Less Three to Five Six or More	5 31 64	0 45 65	8 20 72	3 30 67
Individual or Group Pond				
One or Two Families Three to Seven Families Eight or More Families	84 5 11	95 5 0	85 15 0	86 7 7

 Table 2.
 Land ownership of fish farmers in the Peruvian Amazon, 1999.

	Napo River (N = 87)	Nauta Road (N = 20)	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
NUMBER OF PIECES OF LAND IN FARM				
One to Two Parcels Three to Nine Parcels Ten or More Parcels	96 0 4	100 0 0	90 8 2	95 2 3
Amount of Land Owned				
< 1 ha 1 to 10 ha > 10 ha	0 50 50	25 15 60	3 27 70	5 36 59
LAND OWNED IN COMPARISON WITH OTHE	ER FARMERS			
More About the Same Less	17 15 68	15 20 65	10 24 66	15 18 67

ponds are an important mechanism for introducing aquaculture into poor rural communities. CARE/Peru officials indicated that they used community groups to construct an initial demonstration pond in some locales, but that family-based ponds were the preferred strategy. In some communities, groups of families cooperate in a *minga*—a labor exchange arrangement—to jointly construct a fishpond on one family's land. *Minga* is a Quechua word for the collaboration of communities on specific tasks (e.g., harvesting, sowing, and house building). The completion of the pond is typically followed by a small celebration hosted by the beneficiary family. Subsequent group efforts may build additional fish ponds on other farms.

### Landholding

Table 2 profiles the landholding of study respondents. Nearly all owned one or two parcels of land. Napo River holdings tended to be divided between the two larger categories, but 70% of the Tamishiyacu farmers reported holding more than 10 ha. Holders of multiple parcels were primarily pond groups organized for constructing and maintaining a fishpond.

Nauta Road farmers were most diverse in size as one-fourth reported holdings less than 1 ha in size. About two-thirds of the farmers in each location indicated that they owned less land than other farmers did.

# **Farm Enterprises**

The farm enterprises maintained by fish farmers are portrayed in Table 3. Chickens were the most commonly reported animal enterprise, followed by pigs, ducks, and cattle. In terms of cash income, 60% of the Nauta Road farmers cited fish as their primary source. Furthermore, Nauta Road farmers were more likely to integrate animal production with fish culture. Fish was identified as the primary source of cash income by about

 Table 3.
 Farm enterprises of fish farmers in the Peruvian Amazon, 1999.

20% of the farmers in the other locales. Chickens were the primary source of cash income for most farmers in the study.

More Nauta Road farmers raised animals near or with their fishponds than farmers from other areas. Such integrated operations recycle the undigested feed and other nutrients from penned animals.

#### Pond Location and Water Source

More than 85% of the fish farmers had but a single pond, as shown in Table 4. In contrast, about 45% of the Nauta Road farmers had more than one pond. None of the Napo ponds were located near the farm residence, but one-third of the Nauta Road ponds were. The location of the fishpond relative to the household is significant; ponds near households are easier to monitor. Family members can attend to the pond as well as give regular surveillance to deter theft. About 19% of the fishponds were located near the house. Farmers primarily filled their ponds from springs using gravity flow methods. No farmers experienced problems obtaining water for their fishponds.

# **Species Cultured**

Table 5 shows that *Prochilodus nigricans* (boquichico) was the most frequently cultured fish in the Peruvian Amazon, grown by about 75% of the farmers. *Colossoma macropomum* (gamitana) was the next most frequently culture fish. *Piaractus brachypomus* (pacu) was grown by more group operations (71%) than on individual farms (58%), the largest difference in the comparison. Similar proportions grew *Brycon* sp. (sabalo) and *Leporinus* sp. (Lisa), but 9% more individual farmers favored *Semaprochilodus insignis* (yaraqui) and 11% more individual farmers reported growing *Astronotus ocellatus* (Oscar or acarahuazú). Alcántara (1994) presents a detailed analysis of

	Napo River $(N = 87)$	Nauta Road $(N = 20)$	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)
			(%)	
Animals Raised				
Cows	33	5	13	24
Pigs	66	30	62	60
Chickens	91	90	90	90
Ducks	63	55	49	58
ENTERPRISES THAT PROVIDE MOST OF CASH	Income			
Rice	17	25	23	17
Bananas	24	30	31	27
Fruits	18	60	38	29
Fish	23	60	18	27
Sugar Cane	3	20	0	5
Cows	20	5	5	14
Corn	14	25	13	15
Chickens	63	10	31	47
RAISE ANIMALS WITH FISHPOND				
No	74	40	59	65
Yes	26	60	41	35

	Napo River $(N = 87)$	Nauta Road $(N = 20)$	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
Number of Ponds Owned				
One	91	55	95	86
Two	7	25	5	9
Three or More	2	20	5	5
DISTANCE TO THE MAIN ROAD OR RIVER				
Next to House	0	33	27	19
< 1 km	63	33	33	42
2 to 3 km	24	34	33	31
> 3 km	13	0	7	8
PROBLEMS GETTING ENOUGH WATER				
No	100	100	100	100
Yes	0	0	0	0
WATER SOURCE FOR PONDS				
Well	0	0	3	1
Spring	71	100	77	77
River or Stream	1	0	5	2
Lake or Reservoir	0	0	5	1
Irrigation Canal	25	0	10	17
Collected Runoff	3	0	0	2
WATER SUPPLY TO POND				
Pumped	0	0	0	0
Gravity Flow	100	100	100	100
Combination	0	0	0	0

 Table 4.
 Pond location and water source reported by fish farmers in the Peruvian Amazon, 1999.

Table 5. Species cultured by fish farmers in the Peruvian Amazon, 1999. See also Alcántara (1994).

Type of Fish Raised	Napo River $(N = 87)$	Nauta Road $(N = 20)$	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
Colossoma macropomum (gamitana)	66	63	61	64
Piaractus brachypomus (paco)	61	53	63	60
Prochilodus nigricans (boquichico)	76	63	82	76
Brycon sp. (sábalo)	54	74	26	49
Leporinus sp. (lisa)	38	37	13	31
Astronotus ocellatus (acarahuazú)	17	21	24	19
Cichla monoculus (tucunaré)	14	11	5	11
Cichlasoma amazonarum (bufurqui)	39	42	37	39
Semaprochilodus insignis (yaraqui)	45	16	47	42

	Napo River $(N = 87)$	Nauta Road (N = 20)	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)
			(%)	
Things Most Often Fed*				
Kitchen Waste	42	95	74	58
Fresh Vegetation	10	65	62	31
Rice Bran	6	15	3	7
Dead Animals	1	5	6	3
Slaughter Waste	8	60	21	19
Fruits	92	50	59	77
VISITS TO PONDS				
Several Times a Day	10	70	37	25
Daily	37	15	42	36
Almost Daily	25	10	13	20
Several Times Weekly	23	5	3	15
Once a Week	3	0	5	3
Several Times a Month	2	0	0	1
Number of Feedings				
Several Times a Day	4	45	22	14
Weekly	29	20	22	26
Several Times Monthly	35	25	22	30
Monthly	20	10	19	18
Less Often	6	0	6	5
Never	6	0	8	6
TIME USUALLY SPENT AT PONDS				
Less than an Hour	76	55	35	62
About an Hour	21	15	16	19
Two or Three Hours	3	15	19	9
More than Three Hours	1	15	30	10
	-			

 Table 6.
 Feeding practices of fish farmers in the Peruvian Amazon, 1999.

\* Multiple responses possible.

Table 7. Pond management practices of fish farmers in the Peruvian Amazon, 1999.

	Napo River (N = 87)	Nauta Road $(N = 20)$	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)
			(%)	
Pond Fertilizer Used*				
None	40	84	13	38
Other	0	0	10	3
Chicken Manure	1	16	21	8
Cattle Manure	58	0	54	48
Compost	1	0	10	3
PONDS LIMED LAST YEAR				
No	99	85	97	96
Yes	1	15	3	4

\* Multiple responses possible.

fish landings at Iquitos ports and the species that are brought to market.

# **Fish Feeding**

Farmers in the four locations fed their fish a variety of different items reflecting differences in the species cultured and items available (Table 6).

About 73% of the Napo farmers fed their fish several times a day. The greater incidence of integration with poultry and duck production that requires multiple daily feedings literally spills over to the fish crop. Poultry houses are typically located directly over the fishpond, so feed and litter are nearly continuously deposited into the pond.

Nauta Road resident farmers reported a high level of attentiveness to their ponds, but more Tamishiyacu farmers spent long periods at their ponds when they visited them. All the Nauta Road farmers fed their fish several times a week or more often. Three-quarters of the Nauta Road farmers spent less than an hour on each visit as they were making more visits to attend to their fish. Feeding refers primarily to the provision of fruits, household scraps, and other items directly consumed by the fish. Patterns of fish feeding parallel patterns of pond visitation.

# Pond Management

Table 7 profiles some of the pond management practices used in the Peruvian Amazon. Nauta Road farmers tended to not fertilize their ponds, whereas half the farmers at the other sites used cattle manure. Curiously, 15% of the Nauta Road farmers limed their ponds, a fundamental fish culture management practice not followed by many farmers in the other areas. These farms had truck delivery access to Iquitos port facilities where lime and other bulk agricultural chemicals are more readily available. Lime increases the alkalinity (pH) of the pond and fosters primary productivity. Tamishiyacu farmers spent the most time with their ponds when they visited them, Napo farmers the least.

#### **Stocking Practices**

Table 8 suggests that fingerlings are typically acquired from local fisherman, and few farmers are using farmproduced fingerlings from private or government sources. About two-thirds of the farmers experienced some problems finding fingerlings when they wanted to restock their ponds. Most were stocking fingerlings between 3 and 10 cm in size.

Table 8.Stocking practices of fish farmers in the Peruvian Amazon, 1999.

	Napo River $(N = 87)$	Nauta Road $(N = 20)$	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
Source of Fingerlings				
Research Station Private Dealer From a Neighbor Other (Collected from River)	0 7 1 92	10 50 5 35	3 11 3 83	2 14 2 82
PROBLEMS FINDING FINGERLINGS FOR RESTO	CKING			
No Yes	34 66	55 45	27 73	35 65
Size of Fingerlings Stocked				
< 3 cm 3 to 5 cm 5 to 10 cm > 10 cm	4 34 57 5	10 55 35 0	14 57 29 0	7 43 48 2

Table 9. Farm equipment owned by fish farmers in the Peruvian Amazon, 1999.

Equipment on Farm	<b>Napo River</b> (N = 87)	Nauta Road (N = 20)	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
Fish Net Water Quality Test Kit Scale Wheelbarrow	15 1 17 99	29 0 86 64	48 0 24 70	26 1 27 87

Nauta Road Tamishiyacu-Tahuayo River Napo River All  $(\bar{N} = 87)$ (N = 20)(N = 39)(N = 146)(%) LABOR HIRED TO HARVEST FISH No, Self 37 0 33 28 59 No, Family 85 63 65 2 No, Laborers 10 4 4 2 Buyer Harvested Pond 5 0 3 TROUBLE GETTING ENOUGH HELP TO HARVEST No. Labor Used 77 80 77 72 No Problems 21 20 23 21 5 Yes, Difficulty 2 0 2 AVERAGE SIZE HARVESTED 0 5 10 3 < 20 cm 20 to 45 cm 84 90 85 86 > 45 cm16 5 5 11 AVERAGE WEIGHT HARVESTED 0 0 14 3 < 120 g 120 to 249 g 2 0 2 5 27 250 to 499 g 41 0 18 20 17 500 to 749 g 18 6 37 94 45 51 >750 g

#### Table 10. Harvest practices of fish farmers in the Peruvian Amazon, 1999.

#### **Farm Equipment**

Table 9 shows the farm equipment reported by the surveyed farmers. Wheelbarrows were owned by about 87% of the sample. About one-quarter had a scale and a similar proportion a fish net.

#### **Harvest Practices**

Farm labor for harvesting fish was usually supplied by family members, particularly in the Tamishiyacu-Tahuayo communities, as shown in Table 10. Few operators employed laborers or used the services of a buyer to harvest their ponds. Few reported difficulties obtaining labor. Farmers harvested large fish, most favoring sizes 20 to 45 cm in length and greater than 750 g. Thus, most operators achieved only one cycle of production each year.

#### Marketing

Only a small proportion of each sample reported fish harvested solely for home consumption or barter, i.e., not sold for cash (Table 11). Most sold some fish for cash and only a few said that they sold less than half the harvest for cash. About 80% of the Napo River respondents sold all their harvest for cash, but only 21% of the Tamishiyacu did. In Nauta River, 80% said they sold more than half the harvest for cash. Nearly 100% did so in Tamishiyacu-Tahuayo Rivers.

Middlemen purchased fish from about 41% of the farmers overall. In the Napo River 56% used a middleman. Only 18% of Tamishiyacu farmers sold to middlemen. Few farmers sold fish to restaurants. Tamishiyacu farmers were less likely to sell fish in a community marketplace than farmers from the Napo River and Nauta Road regions. Direct marketing by way of pond bank sales was most common for farmers of Tamishiyacu-Tahuayo Rivers. The most common marketing method for farmers from all regions was pond bank sales to neighbors and others coming to the ponds at harvest. Word-of-mouth knowledge about prospective harvests or the willingness to partial-harvest for immediate sale remain primary means for marketing fish for most small- and medium-size farmers.

#### **Marketing Problems**

Table 12 shows that very few farmers experienced trouble marketing their fish. Few reported price problems, though onefourth of the Nauta Road farmers did so. Most felt they could move their product at a lower price. Most respondents felt that larger fish are easier to sell.

Fingerling sales to other farmers were most common in Napo River, where more than half the respondents reported such transactions. Private fingerling sales among farmers are an important indicator of sustainability, especially where government services are unreliable or unavailable in much of the locale. The respondents did not frequently report fingerling sales. Most were growing species that do not naturally reproduce in ponds, so fingerlings would not be an expected by-product of their ponds.

#### Impacts on Households

Table 13 shows a series of questions profiling the impacts of fish culture on households. Almost none of the farmers thought that

there were points in the annual farm cycle when the pond was too much work. Few respondents noted problems associated with the fish enterprise in taking care of other crops, taking care of the family, or completing other household work. Most thought the pond fit well with other activities of the household. Previous work suggests that women are much more likely to report these difficulties (Molnar et al., 1996). About 84% of the respondents noted the benefits of additional cash for their families as something associated with the fish crop.

# **Pond Conflicts**

Table 14 shows respondent experiences with a series of problems sometimes encountered by fish farmers. No farmers noted problems concerning conflict with others about water resource available for fish crops. Tamishiyacu operators had the most problems with predators eating their fish, but this was also an issue for many farmers in each of the other locations.

Theft was a concern for about two-thirds of the Napo River and Nauta Road farmers, but much less so on the Tamishiyacu-Tahuayo Rivers. Napo farmers were most likely to agree that fish were easier to steal than other crops, though one-third of the other respondents thought so as well.

Table 11. Marketing practices of fish farmers in the Peruvian Amazon, 1999.

In our field visits, farmers reported few specific production problems. Nutria was most frequently mentioned as a source of production losses, with some occasional mentions of alligators and birds. In general, farmers seemed happy with the fish enterprise, several were in the process of planning or constructing additional ponds, and we encountered only a few unused or abandoned ponds. These were typically small ponds with water supply problems. Ponds that fail to keep water during the dry season thwart the counter-cyclical possibilities of fish culture.

#### **Prospects for the Pond**

Nearly all respondents thought their fish pond produced enough to be worth the work they put into it, though a few Nauta Road respondents were skeptical (Table 15). Nearly all respondents thought fish was the best use of the land it occupied. Nauta Road and Tamishiyacu residents were most likely to report themselves as planning to build new ponds (over 90%). In Napo River, only 72% thought so. Nearly all were happy with fish as a crop to raise, but less so on the Tamishiyacu River (84%). Overall, 86% of the farmers surveyed felt that fish was more profitable than other farm activities, but 92% of the Napo River farmers felt this way.

	<b>Napo River</b> (N = 87)	Nauta Road $(N = 20)$	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
CASH LAST HARVEST				
Yes No	90 10	70 30	67 33	80 20
Amount of Fish Sold				
None for Cash Less than Half Half for Cash More than Half All for Cash	9 2 2 7 80	7 7 47 32	36 11 11 21 21	16 5 5 18 56
Middleman Purchased Fish				
No Yes, Some of it Yes, All of it	44 19 37	72 17 11	82 12 6	59 17 24
FISH SOLD TO RESTAURANTS				
No Yes, Some of it Yes, All of it	94 6 0	95 5 0	100 0 0	96 4 0
FISH SOLD IN THE MARKET				
No Yes, Some of it Yes, All of it	48 30 22	55 25 20	70 25 5	55 27 17
FISH SOLD TO OTHER BUYERS				
No Pond Bank Sales	44 56	45 55	39 61	43 57

	Napo River $(N = 87)$	Nauta Road $(N = 20)$	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)
			(%)	
TROUBLE SELLING FISH				
No Yes	98 2	93 7	87 13	94 6
PROBLEMS SELLING AT DESIRED PRICE				
No Yes	85 15	73 27	79 21	81 19
FISH SOLD AT A LOWER PRICE (IF DESIRED PRICE )	NOT OBTAINED)			
No Yes	67 33	33 67	31 69	53 47
LARGER FISH EASIER TO SELL				
No Yes	7 93	12 88	3 97	7 93
FINGERLINGS SOLD TO OTHER FARMERS				
No Yes	100 0	90 10	100 0	98 2
TROUBLE SELLING FINGERLINGS				
No, Did Not Sell No Problems Yes, Problems	100 0 0	90 0 10	100 0 0	98 0 2

#### Table 12. Marketing problems of fish farmers in the Peruvian Amazon, 1999.

#### **Technical Assistance**

Aquacultural extension services were making frequent contacts with farmers on the Napo and Tamishiyacu Rivers, but less so on the Nauta Road. In each location, NGO technicians were working in aquaculture in one way or another. Most farmers reported some kind of extension contact in the past year. Most farmers received regular visits if they wanted them. Nearly all respondents indicated that they wanted extension help in the future.

#### **C**ONCLUSIONS

There are few obstacles to at least limited success with fish culture in Amazonia. We observed a high degree of variability in the quality of pond construction and culture conditions, as well as in the type of species employed and level of management applied. Farmers typically stock a polyculture of three to five different species of wild–caught fry or juveniles for growout. Farmers typically reported counts of the number of fish of each species that was stocked.

Peru is in an advantageous situation for fish culture. The data show that farmers encounter few barriers to building ponds, obtaining fingerlings, feeding their fish, or marketing the product. Fish farmers in the Peruvian Amazon are interested in and receptive to technical assistance. Fruits and other forest-based fish foods are widely available to support extensive production systems. A number of NGOs are providing regular farm visits and advice on fish culture. The natural cycle of the Amazonian river systems ensures a market period of relatively high prices for farm-reared fish. The research agenda is appropriately focused on enhancing the availability of hatchery-reared fry. Nonetheless, additional attention is needed on identifying and communicating production practices that will reduce risk and enhance the benefits of aquaculture for the many small- and medium-scale farms in the Selva.

#### **ANTICIPATED BENEFITS**

The communication process linking experimental pond practice to farm practice involves several layers of translation and transmission (Cernea, 1991a, 1991b). Many factors interact to affect the nature and extent of impact that PD/A CRSP scientists and research programs have on national aquacultural institutions and farm practice (Huisman, 1990). Experimental findings are at base, experimental; that is, they reflect controlled conditions and careful measurement of a focused set of factors. Farm conditions reflect variable physical and management situations that often mitigate the impact of effects identified by repeated experimental trial. The data presented here provide empirical specification of the needs and preferences of the actual intended beneficiaries of PD/A CRSP activities in Peru.

	<b>Napo River</b> (N = 87)	Nauta Road $(N = 20)$	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)		
			(%)			
Pond is Sometimes Too Much Work						
No Yes	97 3	100 0	100 0	98 2		
FISH FIT WELL WITH OTHER FARM ACTIVITIES C	F HOUSEHOLD					
No Yes	1 99	0 100	3 97	1 99		
Pond Makes it Harder to Care for Other Crops						
No Yes	98 2	100 0	100 0	99 1		
Pond Makes it Harder to Take Care of Family						
No Yes	98 2	95 5	100 0	98 2		
Pond Makes it Harder to Complete Other Household Work						
No Yes	97 3	100 0	100 0	98 2		
CASH FROM FISH MAKES IT EASIER TO BUY THINGS FOR FAMILY						
No Yes	23 77	6 94	4 96	16 84		

Table 13. Fish pond impacts on households of fish farmers in the Peruvian Amazon, 1999.

Table 14. Pond conflicts experienced by fish farmers in the Peruvian Amazon, 1999.

	<b>Napo River</b> (N = 87)	Nauta Road (N = 20)	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
CONFLICTS OVER WATER				
No Yes	100 0	100 0	100 0	100 0
Birds or Other Animals Eating Fish from F	ONDS			
No Yes	38 62	42 58	19 81	34 66
PEOPLE STEALING FISH				
No Yes	37 63	32 68	57 43	42 59
FISH EASIER TO STEAL THAN CROPS				
No Yes	38 62	68 32	61 32	61 39

	<b>Napo River</b> (N = 87)	Nauta Road (N = 20)	Tamishiyacu-Tahuayo River (N = 39)	<b>All</b> (N = 146)
			(%)	
Fish Produce Enough to Be Worth th	e Work			
No	0	5	3	1
Yes	100	95	97	99
Fishpond Best Use of Land it Uses on t	THE FARM			
No	1	0	8	3
Yes	99	100	92	97
Planning to Build More Ponds				
No	28	11	8	20
Yes	72	90	92	80
GENERALLY HAPPY WITH FISH AS A CROP	TO RAISE			
No	0	0	19	4
Yes	100	100	84	96
FISH PROFITABILITY COMPARED TO OTHER	ACTIVITIES			
More Profitable	92	76	76	86
About the Same	8	18	24	13
Less Profitable	0	6	0	1

 Table 15. Prospects for the pond reported by fish farmers in the Peruvian Amazon, 1999.

Table 16. Sources of technical assistance reported by fish farmers in the Peruvian Amazon, 1999.

	Napo River $(N = 87)$	Nauta Road (N = 20)	<b>Tamishiyacu-Tahuayo River</b> (N = 39)	<b>All</b> (N = 146)
			(%)	
LAST EXTENSION CONTACT				
Never Contacted	5	55	11	13
In Past Month	83	20	76	73
Month to Year	7	5	3	6
More than a Year	5	20	0	8
LAST FISH STATION CONTACT				
Never Contacted	92	80	80	88
In Past Month	7	10	6	7
Month to Year	0	0	6	1
More than a Year	1	10	8	4
WANT EXTENSION HELP				
No	1	15	5	4
Yes	99	85	95	96

As such, they provide a baseline or template for interpreting the cumulative impact of PD/A CRSP and NGO partner activities, as well as a starting point for identifying new directions and emphases that will help realize the promise of aquaculture for farmers and their families in developing countries.

#### **ACKNOWLEDGMENTS**

We thank the several PD/A CRSP Principal Investigators, Host Country counterparts, and NGO colleagues who facilitated our work and advised us on the many problems and issues. In particular we are grateful to Juan Guerrero of CARE/Peru for his help in obtaining the interview data. Without this assistance, our work would have been much narrower in scope.

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Cite as: [Author(s), 2000. Title.] In: K. McElwee, D. Burke, M. Niles, X. Cummings, and H. Egna (Editors), Seventeenth Annual Technical Report. Pond Dynamics/Aquaculture CRSP, Oregon State University, Corvallis, Oregon, [pp. \_\_\_\_]



# Sources of Technical Assistance for Fish Farmers in the Peruvian Amazon

Eighth Work Plan, Adoption/Diffusion Research 1-2 (8ADR1-2) Progress Report

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#### Abstract

The Institute for Investigation of the Peruvian Amazon (IIAP), a PD/A CRSP host country institution in Peru, is the leading governmental organization working in aquaculture and fisheries research in the Peruvian Amazon region. In addition, IIAP produces fingerlings, offers training courses, and works with nongovernmental organizations (NGOs) endeavoring to promote fish culture. This report summarizes data collected from a sample of 146 practicing fish farmers in the Napo, Tamishiyacu, and Tahuayo River systems areas north and south of Iquitos, as well as in the Iquitos-Nauta Road area directly south of the city. Fish farmers were identified in selected communities that were provided technical assistance in aquaculture by CARE/Peru and several other NGOs. Results address perceptions of need for technical assistance based on gender and the type of operating arrangements for aquaculture. Results suggest women and group farmers have distinctive sets of experiences and preferences for technical assistance. About 38% of the women had no contact with extension versus 9% of the men. All the women respondents indicated that they desired extension contact in the future, but 5% of the men did not.

#### INTRODUCTION

#### Aquaculture in the Selva

There is a unique relationship between aquaculture and fisheries in many parts of the Selva (Hall, 1997). The abundance of large, rapidly growing fish species supports an extensive capture fishery in the Amazon, its tributaries, and a large number of oxbow lakes. The fishery, however, is cyclic, as fishing is more difficult during the high water period of December through March. At this time, fish prices for some species are as much as twice the low-water-period price. This cyclical deficit in the supply of fish coupled with a widespread perception that river and lake fish stocks have declined and will continue to do so are the primary motivations for fish culture in the Selva. Commercial-scale fishers using large-scale fishing gear have depleted fish stocks in many oxbow lakes, further encouraging pond-based fish production. On the other hand, the annual population growth rate is 3.1% (Bayley et al., 1992). About 46% of the protein for human consumption in the Selva comes from fish (Eckmann, 1983).

Abundant supplies of warm water, generally available pond inputs, and easily obtainable grow-out stock are some of the favorable conditions for fish culture in the Amazon River system. Fingerlings can be obtained with cast-nets in rivers and in oxbow lakes or through purchase from fishermen or governmental organizations like IIAP and Fondo Nacional de Desarrollo Pesquero—Acción Promotora para el Desarrollo de la Acuicultura (FONDEPES). These institutions produce fingerlings with induced reproduction in controlled environments. Longstanding efforts by the Ministry of Fisheries, IIAP, and other agencies that were provided technical assistance in aquaculture have stimulated interest in fish ponds. Current NGO efforts in some communities build on previous government efforts. In others, NGOs are introducing pond aquaculture as a new activity.

# Nongovernmental Organizations Working in Aquaculture

In 1992, CARE/Peru began an effort to increase food security and raise incomes by targeting families in nine villages along the Napo River. The Napo is a tributary of the Amazon about 20 km north of Iquitos. In 1995, a parallel effort was begun in six villages along the Tamishiyacu River and another six villages along the Tahuayo River about 30 km south of Iquitos. At each of the 21 villages an initial pond was established for training and demonstration purposes. Subsequently, farmers dug about 250 ponds, between 10 and 30 per village. Aquaculture was part of a broader strategy of community development, health education, and food security improvement.

The CARE/Peru project also provided fingerlings, nets, small loans for pond construction costs, and continuing technical support for aquaculture. One aquaculture technician worked with Napo River villages, while five others provided technical assistance to the Tamishiyacu and Tahuayo River regions. When cultured fingerlings were available, ponds were stocked with *Colossoma macropomum* (gamitana). When cultured fingerlings were not available, farmers used wild-caught fry and juveniles or delayed restocking until they could obtain seed stock. Most CARE/Peru ponds were operated by a single family, but about 14% of study respondents participated in aquaculture through community or group ponds. The Spanish NGO Agencia Española de Cooperación Internacional (AECI) supported the services of a technician who advised about 75 pond operators located primarily along the as-yet-uncompleted Iquitos-Nauta Road. In 1998, about 15 ponds had achieved at least one harvest. The remaining were growing their first crop of fish. The aquaculture technician was one of about five staff members working in various aspects of agricultural and community development in the Iquitos area. Farmers were provided technical assistance in pond construction and instruction in production management. An Italian NGO—Terra Nuova—supported by European Community funds was extending the work initiated by AECI in this area. Terra Nuova has begun working recently in cooperation with IIAP in a food security program primarily in the as-yetuncompleted Iquitos-Nauta Road and in the year 2000 will work in the Tigre River with Indian communities.

Caritas Internationalis is a Catholic assistance agency that targets poor communities not presently served by government programs or other NGOs. The Iquitos office is one of four in Loreto Department. It has seven technicians—including one specialist in aquaculture—who spend two months in target communities followed by one week in Iquitos. With the exception of some lowland area communities where flooding makes fish culture impossible, most Caritas communities have families with functioning fish ponds. These villages are located on rivers largely west of Iquitos and more distant than the communities served by other organizations in this discussion.

# **Governmental Organizations Working in Aquaculture**

Two government agencies are particularly important for aguaculture in the Selva. The Peruvian Ministry of Fisheries (Ministerio de Pesqueria, Dirección de Acuicultura) is a regional enforcement and fisheries development agency. The regional headquarters is in Iquitos and ten other offices are in Loreto Department. Each has a staff of between two and ten persons. Eleven aquaculture specialists, including those at the Iquitos headquarters, provide technical assistance and training programs throughout the region. In the past it has offered a program of technical assistance on pond construction and production techniques in many communities surrounding Iquitos. Recently efforts have been directed to protein-deficient areas near the Ecuador frontier, largely populated by indigenous tribal people. These populations are in great need of assistance and the government needs to reinforce the political allegiance of populations in the disputed border area with Ecuador.

Fisheries stations are a source of fingerlings in some locales, but IIAP and FONDEPES are the largest public fingerling suppliers in the Iquitos area. IIAP is a governmental organization that produces Colossoma macropomum (gamitana) and *Piaractus brachypomus* (paco) fingerlings the last 12 years for the entire Peruvian Amazon. It operates a 2.5-ha fingerling production station near Iquitos and another 2.0-ha fingerling production station near Pucallpa, about 1,000 km south of Iquitos. In addition, IIAP produces fingerlings in Tarapoto in cooperation with the Ministry of Fisheries, as well as at the Ahuashiyacu station, south of Tarapoto. In Iquitos, the IIAP station has produced about 130,000 fingerlings per year since 1985. Last year the Pucallpa station produced nearly 80,000 fingerlings. IIAP continues to offer training courses for the CARE and AECI extensionists and for farmers, especially in the Iquitos area.

FONDEPES is a national fisheries development agency. It operates a 10-ha fingerling production station near Nuevo Horizonte, 35 km southwest of Iquitos. The agency projects a shortfall in the stock of wild fingerlings and plans to engage in production of grown fish for the market as well. Plans are to produce fingerlings for a variety of species, but this agency's strategy features *Prochilodus nigricans* (boquichico). This species requires a less intensive level of cultivation, but is mainly suitable for home consumption and local markets. This species was the single most frequently reported kind of fish grown by farmers operating the ponds we visited during our fieldwork.

Each of these organizations is presently or potentially a partner with PD/A CRSP activities. There is a clear need for expanded understanding of gamitana and paco reproduction and growth processes. New technology for increasing the yield of current breeding techniques and expanding the period during which breeding is possible will yield clear and widespread benefits for aquaculture producers in the Selva (Kohler et al., 1999).

# METHODS AND MATERIALS

#### Sample and Data Collection

Structured interviews were conducted with a sample of 146 fish farmers having accomplished at least one harvest in the past two years (Casley, 1988; Townsley, 1996). The sample was drawn from fish farmers who were NGO program participants in selected communities that were provided technical assistance in aquaculture by CARE/Peru and several other selected nongovernmental and governmental organizations, primarily the Fisheries Ministry and IIAP, in the Napo, Tamishiyacu, and Tahuayo River systems that combine to form the Amazon, as well as in the Iquitos-Nauta Road area south of Iquitos.

The survey instrument was adapted from previous research conducted by Molnar et al. (1996) in five PD/A CRSP countries—Honduras, Thailand, the Philippines, Rwanda, and Kenya. The Peru survey, however, reflects the unique conditions and context of Amazonian fish culture, the diversity of species, and the singular relationship of aquaculture to the river fishery in the region. Ponds were identified in communities on three river systems north and south of Iquitos as well as in the Nauta Road area south of Iquitos. Data collection took place in early 1999 and was conducted by graduate students from the Department of Fisheries at Universidad Nacional de la Amazonia Peruana.

# Analysis

The analysis tabulates the survey responses by gender and type of organization—individual or group ponds. Group ponds were those operated by three or more families; individual ponds were those operated by one or two families. From this information, central patterns of comparison and differences in access and interest in technical assistance can be discerned.

#### RESULTS

# Women and Men

Table 1 shows a series of questions profiling level of contact with various sources of technical assistance. No formal governmental aquacultural extension services are organized in

Gender **Operating Arrangement** Survey Response Men Women Individual Group (N = 125)(N = 21)(N = 21)(N = 125)(%) (%) HAD CONTACT WITH EXTENSION? 9 38\* 15 5 No Yes 91 62 85 95 LAST EXTENSION CONTACT? 9 38\* 15 5 Never Contacted 75 In Past Month 76 57 73 Months to Year 7 0 5 10 7 More than a Year 8 5 10 WANT EXTENSION HELP? 5 5 No 0 4 95 100 95 Yes 96 LAST FISH STATION CONTACT? Never Contacted 87 90 88 81 In Past Month 7 5 6 14 Months to Year 2 0 2 0 More than a Year 4 5 4 5 CONTACT WITH UNIVERSITY TECHNICIAN? 95 95 96 91 No Yes 5 5 4 9 HAPPY GROWING FISH AS A CROP? 5 10 No 4 3 96 95 97 Yes 90

Table 1. Sources of technical assistance by gender and operating arrangement among fish farmers in the Peruvian Amazon, 1999.

 $^*\!\mathrm{X}^2p < .05$ 

the Selva. Consequently, the questions concerning extension are interpreted as contacts with CARE/Peru and other NGO representatives working in rural communities. It is a matter of happenstance that there were 21 group farms and 21 women respondents. Only three respondents were both women and from group farms.

The data show that men reported significantly more contact with extension, as 91% had had extension contact versus 38% of the women respondents. Among those who had contact with extension, there was little difference between women and men in the recency of that contact.

All the women respondents indicated that they wanted extension help in the future, but about 5% of the men said no. Few differences by gender were found for contacts with government stations or university technicians. About 5% of each gender indicated that they were unhappy with growing fish as a crop.

#### **Group and Individual Ponds**

Some NGOs utilize group ponds as a means of introducing aquaculture in poor communities. Group ponds often are products of a *minga* or cooperative ring organized to dig and stock a pond shared by a community or subset of families in a community. About 15% of the sample respondents represented ponds operated by three or more families. Group ponds have a

number of problematic aspects that often discourage participation, including but not limited to the difficulty of apportioning the harvest in accordance with labor or other inputs provided to the group. Often the yield from a pond is not sufficient to motivate the large number of people that may participate in a group endeavor (Molnar et al., 1985; Schwartz et al., 1988).

Group farmers reported slightly more contact with extension than did individual farmers, 95% versus 85%, but individual farmers were more likely to have had recent contacts with extension. There were no significant differences in contact with university technicians. Overall, members of pond groups were the least content with fish as a crop, as 10% indicated that they were not happy with growing fish.

#### CONCLUSION

Fish farmers in the Selva have few natural obstacles to at least limited success with fish culture in Amazonia. Poverty and lack of access to capital are basic problems in poor rural communities and can be obstacles to intensification and a higher level of practice of aquaculture. One important means of raising the productivity of aquaculture as a farm enterprise is through technical assistance to guide pond construction, reproduction, feeding, and fertilization. The data suggest that fish farmers in the Peruvian Amazon are quite receptive to education and practical advice in fish culture, but that the demand far outstrips the supply of these services. The several NGOs that work with poor rural communities feature aquaculture as part of their repertoire of interventions for which they provide assistance. Thus, NGOs are a logical audience for PD/A CRSP research findings. NGO fieldworkers have regular contact with villages. They can provide information about local experiences and problems with aquaculture that would not otherwise be available. NGOs can provide venues for IIAP scientists to serve as resource persons for training programs, field days, and other types of farmer meetings.

Government agencies provide technical assistance and supply fingerlings to fish farmers in many locales. In Peru, as in most other PD/A CRSP countries, budget limitations, competing demands, and other factors constrain the ability of agencies to provide technical assistance to fish farmers on a regular and widespread basis. Where government extension and fingerling production centers exist, they are important PD/A CRSP partners. The experience in Peru suggests that NGOs can be productive and enduring mechanisms for supporting familybased fish culture in rural areas.

#### **ANTICIPATED BENEFITS**

The central benefits of this study accrue to an improved understanding of the knowledge system for aquaculture in the Peruvian Amazon. The many species cultured there and the ready availability of fingerlings and natural foods enhance the possibilities for small- and medium-scale aquaculture in poor rural communities. The data reported here are providing important insights into needs and problems experienced by fish farmers for use by the staff of IIAP as well as the several governmental and nongovernmental actors working in aquaculture in the Amazon.

#### ACKNOWLEDGMENTS

We thank the several PD/A CRSP Principal Investigators, host country counterparts, and NGO colleagues who facilitated our

work and advised us on the many problems and issues. In particular we are grateful to CARE/Peru Regional Director Juan Guerrero for his organization's help in obtaining the interview data. Without this assistance, our work would have been much narrower in scope.

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# IMPACTS OF INTEGRATED FISH CULTURE ON RESOURCE-LIMITED FARMS IN GUATEMALA AND PANAMA: AN EX-POST EVALUATION

Eighth Work Plan, Adoption/Diffusion Research 2 (8ADR2) Final Report

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#### Abstract

The study evaluated the status of fish pond projects initiated in the 1980s on resource-poor farms in Guatemala and Panama. In both places, the host country and the United States Agency for International Development (USAID) provided financial assistance and Auburn University provided technical support to the respective governments. The study examined the impact of aquaculture technology, extension services, and local socioeconomic conditions on the projects. The evaluation team (an aquaculturist, an agricultural economist, and a social anthropologist) had a rare opportunity to evaluate sustainability of two different types of fish farming projects. Other ex-post evaluations of aquaculture projects occur shortly after external support has ended, rather than after 14 and 9 years as was the case in Panama and Guatemala. In both Guatemala and Panama, the projects were designed to improve the nutrition and increase the income of poor farmers, and participants were to become selfsufficient pond managers by the end of the project. The critical difference between the two projects is that in Guatemala fish ponds were managed by individual families on their farms, while in Panama more complex fish pond modules were managed by organized groups of farmers. In central and eastern Guatemala, the team visited 37 family and 2 cooperative fish pond projects between 9 and 19 June 1998. After the team left, a household survey was administered to these 37 families and another 9 families. So far as was possible, households were randomly selected from a list of 651 farm families known to have had functioning fish ponds when external financing was withdrawn in 1989. The team found that 39% of the ponds were abandoned, 48% were under-utilized; and 13% were well-managed. The fish did not have the intended impact on household nutrition and income for a combination of technical, domestic, economic, social, and broad political reasons. These include problematic water supplies to the ponds, lack of sufficient nutrients entering ponds to increase fish yield, theft, inconsistent technical assistance because of civil unrest and changing policy environments, and changing participant priorities linked to changes in household needs over the years. In Panama, the team visited 21 cooperative fish pond projects between 20 June and 3 July 1998. After the team left, a household survey was administered to 115 current or former project members. The team found that 6 projects had been completely abandoned, and 15 were being used to grow rice and /or fish. Only two projects still in use were well-managed. Fish did not have the intended impact on household nutrition and income for a combination of technical, domestic, economic, social, and broad political reasons. These include too little water to maintain pond water level during the dry season, lack of sufficient nutrients entering ponds to increase fish yield, inconsistent technical assistance related to changing government strategies, a lack of managerial and business skills on the part of project group leaders, overdependence on local elites and/or government for various types of assistance, and macrosocial and political changes. Typically, abandonment or poor performance results from a combination of technical, economic, and social factors, each playing on and amplifying the others. In both countries, many project participants who maintained their ponds did so to irrigate gardens, water animals, or serve as flooded rice paddies. Thus, although the projects did not meet intended goals related to fish culture, participants found ways to profit from the existence of the ponds. In Panama 15 of 21 cooperatively managed pond projects and in Guatemala 28 of 46 individual household pond projects were still used at some level of proficiency.

#### INTRODUCTION

The purpose of this study is to evaluate the current status of tilapia pond projects initiated in the 1980s by the governments of Guatemala (GOG) and Panama (GOP), with financial support from the US Agency for International Development (USAID). This ex-post evaluation is unusual in that most evaluations of aquaculture projects occur within several years

after external support has ended, rather than after 14 and 9 years as are the cases for Panama and Guatemala, respectively. This study is an attempt to determine the technological, economic, and social factors that influenced the success or failure of integrated fish culture projects in Panama and Guatemala. The role of women in managing the fish pond projects and the impact of the projects on communities in which they are located was investigated.

In Guatemala the integrated fish pond project was initiated in 1982 and external funding ended in 1989 (Castillo et al., 1992). The project was a collaborative effort, involving the National Directorate for Livestock Services (DIGESEPE), Cooperative for Assistance and Relief Everywhere (CARE), USAID, and the Peace Corps. Auburn University provided technical assistance in fish culture to the government of Guatemala and CARE. The budget for the project was US\$953,000, not including Peace Corps contributions and the salaries DIGESEPE paid for 32 local promoters (extension agents) and 7 part-time supervisors. DIGESEPE also provided logistic and administrative support. The Peace Corps assigned 73 volunteers to the project, though not all were in the field at the same time. Volunteers worked directly with project participants and identified local people who could be employed by DIGESEPE as local extension agents (Castillo et al., 1992). The project was designed to improve nutrition and income for poor farm families in eastern, coastal, and northern Guatemala. To do so, the project promoted small-scale fish culture on small, individually owned farms. On many farms, 100- to 200-m<sup>2</sup> hand-dug ponds were integrated with livestock. The manure was used to fertilize the pond waters to increase fish yields. The nutrientrich pond mud also could be used to fertilize gardens adjacent to the ponds. New ponds were initially stocked at no cost to participating farmers, but later they had to buy their own fingerlings. Most ponds were stocked with mixed-sex Nile tilapia, Oreochromis niloticus, and common carp. Participants were taught to produce their own tilapia fingerlings by retaining offspring, spawned in the fattening pond, at harvest for restocking to produce the next crop. Common carp fingerlings were purchased from government hatcheries for restocking ponds (Castillo et al., 1992). By 1989, 1,200 ponds had been built or renovated, about 15% of the ponds were integrated with animals (usually poultry in enclosures suspended over the ponds), and 21% were integrated with vegetable gardens. On average, a pond of 120 m<sup>2</sup> produced about 48 kg (4,000 kg ha<sup>-1</sup>) of fish annually, of which about 48% was consumed by the household, 42% sold, and 10% given to neighbors or used for restocking ponds (Castillo et al., 1992).

In Panama the integrated fish pond project was initiated in 1980 and external funding ended in 1984. USAID granted the Government of Panama US\$1,420,000 to mount a four-year pilot fish culture project in 21 communities. The Panamanian National Directorate of Aquaculture (DINAAC), a bureau in the Ministry of Agriculture and Livestock Development (MIDA), implemented the project. Auburn University provided technical assistance in fish culture to DINAAC. All the extension persons were government employees, most of them from DINAAC. The project was designed to teach organized groups of poor farmers how to manage integrated modulesassemblages of two, three, or four machine-dug ponds and animals and, in some places, gardens and trees-by themselves. Project participants were trained to produce their own Nile tilapia seed in small spawning and nursery ponds for stocking into the grow-out pond. Most projects were stocked with male tilapia, but some ponds were stocked with mixedsexes and the predacious guapote tigre, Cichlasoma managuense, to control offspring in the grow-out ponds. Various types of carp were added to the fish ponds to increase fish production. Because carp are difficult to reproduce, fingerlings were obtained from government hatcheries. Average annual fish yield from grow-out ponds averaging 2,600 m<sup>2</sup> and fertilized with pig, chicken, duck, or cattle manure was 2,177 kg ha<sup>-1</sup>.

External technical support was to continue for about 24 months, after which the groups were to be largely selfsufficient, with minimal support from extension. Production of fish, garden produce, livestock, and trees benefited the groups by improving their nutrition and by providing them with additional income (Lovshin et al., 1986; Schwartz et al., 1988).

#### METHODS AND MATERIALS

During June and July 1998 the authors visited 37 family and 2 cooperatively managed fish ponds in Guatemala, and 1 church-managed and 20 cooperatively managed fish pond projects in Panama. Host country personnel who had been involved with the projects in the 1980s coordinated the on-site visits. In Guatemala, families with fish ponds were selected from a list compiled by CARE of 651 families known to have had functioning fish ponds when the project ended in 1989. The Guatemalan extension person in charge of providing technical assistance to fish pond owners in each province assisted with the selection of families visited. The evaluation team made a rapid evaluation of the pond site and attempted to interview either the husband or wife at each site to obtain information on species cultured, source of small fish for stocking, fish care, harvest and utilization, and reasons for pond and animal husbandry abandonment. Ponds were classified as abandoned, under-utilized, or well-utilized for fish culture in Guatemala. Abandoned ponds had no water and bottoms overgrown with grasses and weeds, or were partially filled with water but full of aquatic weeds. Under-utilized ponds contained water and a few fish but were poorly cared for as evidenced by clear or muddy water color, pond banks overgrown with weeds, little noticeable fish activity on the water surface or along the pond margin, and general lack of interest in the pond voiced by owners during the visit. Wellutilized ponds had a green water color, generally well-kept pond banks, observable fish activity in the pond, and the pond owner showed pride and a knowledge of fish culture during the interview. Other observations included the integration of animal husbandry and vegetable gardens with the fish pond and secondary utilization of the pond water for irrigating crops or watering livestock.

In Panama, the team made a rapid evaluation of the pond sites and attempted to interview at least one, and often more, participants or ex-participants to obtain information on fish species cultured, source of small fish for stocking, fish care, harvest and utilization, and reasons for pond abandonment. Projects were classified as abandoned or utilized. Abandoned ponds contained no water and their bottoms were overgrown with grasses and weeds or were partially filled with water but full of aquatic weeds. They were considered utilized if at least one pond was used for growing fish or an agricultural crop, even if remaining ponds were abandoned. Utilized ponds were further classified into three groups: a) culture of rice only in at least one pond, b) culture of fish only in at least one pond, and c) fish culture integrated with animal husbandry.

The number of fish harvests and weight of fish harvested was not recorded by farmers or extension agents in either Panama or Guatemala. Thus, neither a comparison of fish yields at project termination and at the time of this evaluation nor an analysis of economic benefits to project participants was possible.

#### **RESULTS AND DISCUSSION**

#### Guatemala

The team found 14 projects abandoned, 20 under-utilized, and 5 well-utilized. Mixed-sex tilapia were stocked in 24 of the 25 projects with well-utilized and under-utilized ponds. None of the projects reported stocking only male tilapia to reduce or eliminate tilapia reproduction. Of 25 projects still growing fish, 9 reported stocking guapote tigre to permit stocked fish to reach a larger harvest weight. However, a number of fish farmers reported that they did not like guapote tigre in their ponds because they ate all the tilapia offspring and none remained for restocking. Lack of tilapia fingerlings did not seem a major constraint to growing fish.

Fish did not have the impact on family nutrition and financial well-being envisioned when the project was planned. The final report to USAID and the Guatemalan agencies participating in the project (Castillo et al., 1992) did not reflect this circumstance. However, nine years after the project was terminated, most fish ponds evaluated were abandoned or under-utilized. Whatever the reasons for abandonment or under-utilization, in most cases fish ponds were not well-cared-for, and this suggests that fish do not or may not play an important role in family nutrition or financial well-being. Incentives to properly manage the fish ponds are not present. Theft of tilapia from ponds not located close to the household was a problem. Integrating animals with fish ponds to improve fish yields failed. For reasons not fully understood but likely related to unprofitability, broiler and layer chicken production associated with fish ponds was abandoned. Without a consistent source of manures to fertilize fish ponds, producers resort to kitchen and table scraps and on-farm by-products to feed fish. Most families with fish ponds are resource-limited and do not have enough feed to provide the fish with a nutritious diet, and this results in slow fish growth and low yields. Even farmers with the financial means to purchase feed are reluctant to purchase sufficient quantities to adequately feed their fish. Fish simply do not provide the nutritional or financial return to justify the expense of purchased feeds. The most beneficial aspect of the fish ponds appears to be their ability to store water during the dry season to irrigate vegetables and water cattle. Many of the ponds used to store water are not well utilized for growing fish but do play an important role in the nutritional and financial well-being of their owners in other ways. Without the fish pond, farmers would be unable to plant a garden or raise cattle during the dry season.

Since the Guatemalan project did not involve group effort, the survey dealt with division of labor within the household. Women seem to play a much larger role in pond management in Guatemala than in Panama. In Guatemala, women played a significant role in pond management in about 50% of the households. Gender differences between Panama and Guatemala are difficult to account for. However, differences in project scale appear to account for much of the difference in gender role. Women may play larger roles in small, familysized ponds than in the more complex, multiple-pond system used in Panama.

Abandoning or under-utilizing a small-scale family pond may be an artifact of changes in the domestic cycle. As children become adults and move away from home, particularly if they find relatively well-paying jobs in urban areas and remit funds home, project participants simply have less need for the ponds. In this context, it is useful to recall that most participants entered the project to secure more food. As households become smaller they have less need for additional food, especially in cases where adult children help to support parents and/or in cases where the older person is not as healthy or as strong as she/he was when the project began. The ponds, having done their job, now become part of the past.

Extension agents provide not only technical support but also may be or may become useful economic or political connections for farmers, and lack of extension continuity and constancy may dampen commitment to the ponds. Simply being able to touch base, so to speak, with an extension agent may maintain interest in the ponds. The employment of local farmers to act as extension agents may have been beneficial when the project was active. However, the GOG failure to continue to pay local extension agents after the project terminated and the exit of Peace Corps volunteers left farmers without technical assistance except for government workers located at distant fish hatcheries.

#### Panama

Six projects were considered abandoned. The remaining fifteen projects still grew tilapia or rice in at least one pond. Four projects had abandoned fish culture and only planted paddy rice in some of their fish ponds. Eleven projects still had tilapia stocked in at least one pond. Of the eleven projects still growing fish, nine also had at least one pond planted with paddy rice and two had not added rice as a component of their integrated project. Of the eleven projects culturing fish, four projects said they stocked mixed-sex tilapia, while seven projects stocked male tilapia. Eight projects stocked carps, and seven projects stocked guapote tigre with their tilapia to control tilapia offspring. Surprisingly, nine projects obtained male tilapia for pond stocking only from government hatcheries even though project members had been instructed in methods to produce mixed-sex fingerlings or male fingerlings by visual selection on-farm. Only one project did not get any fingerlings from the government while one project got fingerlings from both the government and on-farm. Most project participants thought that producing male tilapia fingerlings by visual selection was too difficult. However, participants thought that inadequate availability of fingerlings in a timely manner hindered efficient use of fish ponds. Nine of eleven projects growing fish had an animal husbandry activity associated with the fish pond. Pigs were found on eight projects, chickens on five projects, and ducks and goats on one project. More than one animal was raised on four projects. None of the projects growing fish reported feeding their fish or fertilizing pond waters with chemical fertilizers. Only three projects obtained off-farm manures to fertilize pond waters. Of the fifteen projects still operating, seven had vegetable gardens when the project ended in 1984 (Lovshin et al., 1986). At the time of the survey, only two of these seven projects continued to plant vegetables. However, paddy rice had displaced gardening and was found on 13 of 15 operating projects. Two projects were culturing rice and fish together in the same pond, while the remaining eleven projects mono-cultured rice in refurbished fish ponds. Trees were planted on five of the fifteen projects still in use, and the trees had grown to maturity and provided income, building materials, and fuel.

As in Guatemala, most fish ponds in Panama were abandoned or poorly utilized for growing fish. Fish culture did not have the anticipated economic and nutritional impact on participant families. Levels of fish, animal, and vegetable production recorded in 1984 (Lovshin et al., 1986) were apparently not sustained. Whatever the reasons for pond abandonment or poor utilization, in most cases fish ponds were not well-cared-for, and this suggests that fish do not or may not play an important role in family nutrition or financial well-being. Incentives to properly manage the fish ponds are not present. Extension assistance by the GOP was not maintained, and contacts between extension agents and project groups were infrequent.

Tilapia continued to be the principal culture fish, although carps were appreciated by some project members. Few project groups were able to learn or were sufficiently motivated to produce their own male tilapia fingerlings. A government hatchery was able to provide fingerlings for most projects still growing fish, though supply was often sporadic. Most project members preferred to purchase fingerlings or to receive them free from the government. Self-sufficiency in tilapia fingerling production was not accomplished, though this was a principle goal of the project. Most projects still growing fish continued to raise animals close to their fish ponds. Although the number of animals raised was below recommended levels for good fish yield, animal manures were the only source of nutrients entering fish ponds. Project members did not provide fish with on-farm or purchased feeds. Although the concept of integrating animals with fish ponds was retained, lack of cash and difficulty obtaining loans from banks, government, and nongovernmental organizations hindered the ability of project members to raise animals in a manner that would effectively fertilize the fish pond to increase fish yields. In only three projects had animal husbandry developed into a self-sustaining activity. As in Guatemala, fish ponds were used in a manner unforeseen when the project was designed and implemented. Many ponds had been adapted to plant irrigated rice. The Panamanian government actively promoted the conversion of fish ponds into rice paddies. In many cases, rice has provided a greater benefit to project members than fish and has replaced fish as the primary project activity. Benefits to project members from trees are probably equal to or greater than any other activity. If participants are willing to wait until trees reach maturity, trees make a good addition to community projects as they provide participants with both environmental and economic benefits.

A puzzling question is who does the work of maintaining the fish ponds. To judge from responses to survey questions, in no more than 11.5% of households do women participate. Yet when one spends some time in at least certain communities, it is apparent that women have a more active role than is reflected on the survey. To be sure, there are some places where women play no role in fish-pond maintenance, and there are some tasks with which they may not be involved in any communities. Yet observations made in 1985 indicated that women played a larger role in pond management than the 1998 survey indicates.

As for the number of beneficiaries of the project, there has been a constant drop in numbers. Figures given here refer to 14 communities for which there were complete data in the 1980s. In 1981, when the project began, 353 heads of household enrolled as members (*socios*) in the project in these 14 communities, each head of household representing a family of about six persons at that time. Across the 14 communities, the average number of socios per project was 25.2, with a median of 24.0, and a range of 20 to 38. Average size of the fattening pond in the 14 communities was 2,830 m<sup>2</sup> so that each of the 25 socios had 113 m<sup>2</sup> of fattening pond to grow fish for his family. By 1984, the total number of socios had dropped 31% and was 244, with an average of 17.4 socios per project, a median of 16.5 and a range of 8 to 37. In 1985, a year after external funding for the project ended, there was another decrease, with 172 socios, and an average of 12.3 socios per project group, a median of 11.5 socios, and a range of 1 to 29. In 1998, there were 113 socios in 9 projects because 5 had been abandoned, with an average of 8.1 socios per project group, a median of 6.0, and a range of 1 to 21. Average size of the fattening pond in the 9 projects was 3,009 m<sup>2</sup> and each socio had 376 m<sup>2</sup> of fattening pond from which to harvest fish. Thus, between 1984 and 1998, membership in the project groups in 14 communities had decreased 69%, and between 1985 and 1998 the decrease was 34%. In the 9 ongoing projects mentioned above, 2 had become the property of a single owner, 5 were more or less owned and operated by several related families, and 2 were operated by unrelated community members.

More time identifying communities whose social system was compatible with the demands of rural producers' cooperatives prior to pond construction was needed. Since this is not the place for an extended discussion of donor agency policy, perhaps it will suffice to note that donor agencies operate with fixed, relatively short-term time scales, about three years in the case under study. Moreover, donors require specified objectives to be achieved by carefully budgeted and regularly scheduled activities and expenditures. This is entirely proper given the need for accountability and the organizational culture of donor agencies. This reasonable-enough way of doing things, nonetheless, puts pressure on consultants and recipient agencies like DINAAC to concentrate efforts on the most readily monitored components of a project, such as formally organized groups, numbers of people trained, physical infrastructure, and so on.

However understandable the focus on deliverables may be, it probably makes more sense to invest time up front on social analysis. The cost of making sociocultural studies secondary to technical ones can be high. As an analysis of ex-post evaluations of World Bank and USAID projects indicated, the average economic rate of return for rural development projects which have incorporated sociocultural analysis was more than double that for projects which had been poorly appraised from a sociological viewpoint (Cernea, 1991; Schwartz and Deruyttere, 1996). Sociocultural analyses are not easy, they are always sitespecific, and they take time. But they should be carried out prior to building infrastructure rather than concurrently with that activity. The up-front time invested in such analyses turns out not to increase total project length because potential problems (particularly social ones) are identified and resolved early in the project rather than being allowed to fester beneath the surface only to erupt later on. Another reason for conducting sociocultural studies prior to building infrastructure is that no two communities in this or any other project were or are exactly alike. Prior understanding of the particularities of each community, though this takes time, might have made some problems more manageable.

#### General

Finally, most development projects must not be seen primarily as solely social, cultural, technical, economic, or political, but

rather as a subtle interplay of all these factors. Generally, the team found that project failure was due to a combination of linked causes, rather than to any single cause. Of course, there are exceptions to this statement. For example, even if participants have excellent managerial skills and are genuinely committed to and knowledgeable about fish farming, if a pond simply loses its supply of water, then the project will fail. But more typically, abandonment or poor performance results from a combination of technical, economic, and social factors, each playing on and amplifying the others.

#### **ANTICIPATED BENEFITS**

The goal of PD/A CRSP research is to improve animal protein sources to LDCs. Results of PD/A CRSP research must be transferred to farmers to increase fish protein supplies in developing countries. An understanding of the elements that insure that new technology will be accepted and sustained by target farmers and lessons learned from past small-pond fish culture projects will assist the PD/A CRSP and host country governments to design appropriate research and outreach activities.

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# PD/A CRSP Seventeenth Annual Technical Report

# **REGIONAL OUTREACH IN AFRICA**

Eighth Work Plan, Kenya Research 5 (8KR5) Final Report

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#### Abstract

Regional outreach activities were undertaken under the Eighth Work Plan as a means of disseminating information developed through CRSP research; giving CRSP researchers opportunities to learn about fish culture practices, research priorities, and research activities in other parts of Africa; encouraging efforts to create linkages between research and extension activities in the region; and in general continuing the process of making contacts and regionalizing CRSP efforts in Africa. CRSP researchers in Kenya attended meetings of District Fishery Officers of Central Province and a meeting for Provincial Fisheries Officers (Kenya). During these meetings the PD/A CRSP was described, pond management recommendations were outlined, proposed on-farm trials were discussed, pond census forms were distributed, information was provided on sex-reversed tilapia, and the results of a feeds and fertilizer experiment at Sagana Fish Farm were presented. Students doing research at Sagana in connection with that experiment also presented short summaries of their research findings. Several regional meetings were attended by CRSP personnel during the reporting period. The first was the 5th Session of the Organization of African Unity's Scientific, Technical, and Research Commission (OAU/STRC) Inter-African Committee and Symposium on Oceanography, Sea and Inland Fisheries, Mombasa, Kenya, 4-8 May 1998. The meeting was hosted by Fred Pertet, member of the OAU/STRC and host country Principal Investigator for CRSP research in Kenya. Karen Veverica and Bethuel Omolo also attended this meeting, which provided an excellent opportunity to publicize the CRSP and to present Sagana Fish Farm as an ideal aquaculture training site. Veverica and Omolo also attended the 8th Annual East African Environmental Network (EAEN) conference, 29-30 May 1998, Nairobi, where they presented an invited paper entitled "An overview of aquaculture practices in East Africa: Potential environmental impacts and prospects for sustainable livelihoods." CRSP participants from Kenya and the US attended the PARADI/FISA conference held in Grahamstown, South Africa, 13-19 September 1998. Nine aquaculture and fisheries presentations (oral and poster) were made by CRSP or Kenya Fisheries Department participants. CRSP PIs helped organize and conduct a workshop (Aquaculture in Africa – Quo Vadis) to examine what has previously been done to promote aquaculture in Africa, to look at successes and failures among those efforts, and to discuss how the sub-Saharan region might become an important player in aquaculture in the future. Through contacts made at these meetings and conferences, CRSP researchers, collaborating scientists, and students are developing a better understanding of the research and extension needs for aquaculture development in Africa. Other participants are also gaining a better understanding of the research and extension needs of the region, as well as learning about the CRSP. Linkages have been established that will enhance further correspondence and exchanges of ideas on these issues and on how future programs can be more effective.

#### INTRODUCTION

Regional outreach activities were planned under the Eighth Work Plan as a way to disseminate information derived from CRSP research; to give CRSP researchers opportunities to learn about fish culture practices, research priorities, and research activities in other parts of Africa; to encourage efforts to create linkages between research and extension activities in the region; and in general to continue the process of making contacts and regionalizing CRSP efforts in Africa. This document is the final report on CRSP regionalization efforts undertaken in Africa under the Eighth Work Plan.

#### MATERIALS AND METHODS

Plans for regional outreach included teaching short courses for extension agents at the Naivasha Fisheries Officer Training Center and at the Lake Basin Development Authority in Kisumu (both in Kenya), attending meetings at the Southern Africa Development Community (SADC) fisheries coordination center, and actively participating in meetings of the Fisheries Society of Africa (FISA).

#### **RESULTS AND DISCUSSION**

#### **Extension Agent Short Courses**

The initial plan was to participate in courses given at the Naivasha Training Center for fisheries officers and to contact the Lake Basin Development Authority (LBDA) to offer short courses for extension agents. However, after submission of the Eighth Work Plan, the Naivasha Training Center was handed over to the Kenya Wildlife Service, and fisheries officers are no longer trained there. Also the LBDA in Kisumu has not pursued aquaculture activities during the last two years, so it could not be used for training. In an effort to disseminate information to extension personnel, meetings of District Fishery Officers of Central Province were attended, and a meeting was held for Provincial Fisheries Officers.

During the first meeting, held 25–26 November 1997, Karen Veverica presented an overview of pond management recommendations and presented the PD/A CRSP to attendees. The proposed on-farm trials were discussed and pond census forms were distributed.

For the second meeting, held 7–8 April 1998, Veverica presented information on sex-reversed tilapia and offered collaborating farmers all-male fingerlings to compare with mixed sex fingerlings on their farms. Guidelines for record keeping and pond management were handed out. The results of the experiment on rice bran and fertilization (Veverica et al., 1999a) were presented. Each of the students doing research at Sagana (Veverica et al., 1999b) presented a short summary of his or her research findings.

#### **Attendance at Regional Meetings**

The following regional meetings were attended by Veverica and Omolo:

1. Fifth Session of the OAU/STRC Inter-African Committee and Symposium on Oceanography, Sea and Inland Fisheries, Mombasa, Kenya, 4–8 May 1998. Fred Pertet, member of the OAU/STRC (and Host Country Principal Investigator for the Africa Project) hosted this session. Representatives from Cameroon, Ghana, Kenya, Mauritania, Nigeria, Senegal, Tunisia, the Food and Agriculture Organization of the United Nations (FAO), and the Southeast Asian Programme in Ocean Law, Policy and Management (SEAPOL) attended.

Karen Veverica and Bethuel Omolo attended the meeting and set up a table with CRSP publications and descriptions of the PD/A CRSP program. This was an excellent opportunity to publicize the CRSP to attendees from several countries and to present Sagana Fish Farm as an ideal training site.

2. Karen Veverica and Bethuel Omolo attended the Eighth Annual EAEN (East African Environmental Network) conference, 29–30 May 1998, at the Louis Leakey Memorial Hall, National Museums of Kenya, Nairobi. They presented an invited paper entitled "An overview of aquaculture practices in East Africa: Potential environmental impacts and prospects for sustainable livelihoods." Member countries of the EAEN are Burundi, Djibouti, Eritrea, Ethiopia, Kenya, Rwanda, Somalia, Sudan, Tanzania, and Uganda. The EAEN is a regional non-profit NGO committed to sustainable development in Eastern Africa. Papers presented at this conference are published in a proceedings volume.

## Participation in Fisheries Society of Africa (FISA) Meetings

The first and only FISA meeting to be scheduled during the Eighth Work Plan took place in Grahamstown, South Africa, 13–19 September 1998. The conference was organized by two distinct African fisheries associations—the PARADI Association and the Fisheries Society of Africa (FISA). CRSP participants who attended this meeting included Karen Veverica, Wilson Gichuri, Jim Bowman, Dr. Luc De Vos (representing Mr. Fred Pertet), and Tom Popma. In addition, partial sponsorship for conference attendance was provided to three Kenya Fisheries Department (FD) officers: Mr. Booker Odour (Deputy Director of Fisheries and treasurer of FISA), Ms. Nancy Gitonga (Assistant Director of Fisheries and FISA secretary), and A.L. Aloo (Lecturer at Kenyatta University). Nine presentations (oral or poster) were made by these CRSP and FD participants:

- Semi-intensive *Oreochromis niloticus* (Cichlidae) and *Clarias gariepinus* (Clariidae) polyculture in ponds receiving rice bran and chemical fertiliser in Kenya, by W.M. Gichuri, K.L. Veverica, J.G. Omondi, P.N. Mwau, P.I. Bilal, and K.N. Mavuti (poster presented by W.M. Gichuri).
- Use of grasses from pond levees to promote *Oreochromis niloticus* (Cichlidae) production: Application rates, methods and resulting water quality, by K.L. Veverica and E. Rurangwa (oral presentation by K.L. Veverica).
- A review of lime requirement determination methods for aquaculture ponds, by J.R. Bowman (oral presentation).
- Current status and problems of fisheries in Kenya, by F. Pertet (poster).
- Present status of the fish fauna and fisheries of Lake Baringo, Kenya, by L. De Vos, F. Pertet, K. Vanlerberghe, S. Nuguti, and M. Ntiba (oral presentation by L. De Vos).
- Fish diversity in the Lake Tanganyika system with emphasis on the Rusizi basin in the north and the Lufubu system in the south, by L. De Vos (oral presentation).
- Factors influencing oviposition, larval growth and mortality of blowflies found in smoked fish in Kenya, by N.K. Gitonga (poster).
- Investigation into the effect of salt treatments in reduction of post harvest losses of Nile perch (*Lates niloticus*) during smoking and storage, by N.K. Gitonga (poster).
- Parasitic fauna of tilapia species from Lake Naivasha and the Oloidien Bay, Kenya, by P.A. Aloo (oral presentation).

A book of abstracts was printed and distributed to conference attendees at registration, but full papers presented at the conference will not be published.

In addition, the CRSP contacted the conference organizers in February to inquire about the possibility of including a special discussion session on the status, constraints, and priorities of aquaculture in Africa in the conference. This was accepted and Jim Bowman was asked to serve as rapporteur for the session, which was called *Aquaculture in Africa*—*Quo Vadis*. The purpose of this session was to examine what has previously been done to promote aquaculture in Africa, to look at successes and failures among those efforts, and to discuss how the sub-Saharan region might become an important player in aquaculture in the future. The discussion session was organized by Veverica, Bowman, Dr. Peter Britz, a senior lecturer at the Department of Ichthyology and Fisheries Sciences, Rhodes University (host institution for the conference), and Dr. John Balarin, who facilitated the discussion. This small organizing group identified a set of development objectives and strategies and presented them to the larger group for discussion. Participants voiced a wide range of views on the reasons for past failures of aquaculture development efforts in Africa. It rapidly became clear that the subject area was far too complex and the number of participants far too large to reach consensus within the time allotted  $(1\frac{1}{2})$ hours) for the workshop. Balarin, Britz, Veverica, and Bowman plan to draft a discussion document that will be based on the objectives and strategies presented for discussion as well as the responses and experiences that were voiced in the workshop.

Discussions with potential collaborators at the conference regarding the establishment of a companion site were postponed pending final Ninth Work Plan funding decisions. Attendance at the conference did, however, give CRSP team members the opportunity to learn about research and extension efforts in many other African countries and to make the contacts necessary for future collaboration. CRSP publications were distributed to people from the following countries at the FISA conference: Cameroon, Eritrea, Gabon, Mali, Mozambique, Nigeria, Tanzania, Uganda, and Zambia.

Conference participants showed a lot of enthusiasm regarding uniting the PARADI Association and FISA, two previously independent fisheries societies, to create a dynamic new organization. The CRSP will support this move by cosponsoring the next meeting of the new, combined African fisheries society if possible. Such sponsorship would be consistent with plans laid out under the *Human Capacity Development* section of the *Continuation Plan* and would contribute to the further sharing of CRSP research results in the region.

#### **ANTICIPATED BENEFITS**

Through contacts made at these meetings and conferences in Kenya and other African countries, CRSP researchers and CRSP-sponsored students are developing a better understanding of the research and extension needs for aquaculture development in Africa. Other participants are learning about the CRSP, its research and other activities, and the results of its efforts. It is intended that they are also gaining a better understanding of the research and extension needs of the region. In any case, linkages have been established that will enhance further correspondence and exchanges of ideas on these issues and on how future programs can be more effective.

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# AQUACULTURE TRAINING FOR KENYAN FISHERIES OFFICERS AND UNIVERSITY STUDENTS

Ninth Work Plan, Adoption and Diffusion Research 3 (9ADR3) Progress Report

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#### Abstract

A lack of technical training was cited as a major reason for the low output of fish ponds in Kenya. The need for training was observed at all levels, from the lowest level extension agent through university levels. The training program undertaken by PD/A CRSP researchers in Kenya seeks to improve training and to provide a cadre of trainers who have extensive practical fish production experience. Stipends for student research have allowed undergraduate university students to remain longer at Sagana Fish Farm and gain valuable field experience. A small research projects program has allowed the station staff to further their professional development and carry out their own research, which can have a positive impact on station management. Following requests from farmers, a program of farmer education days was developed. During the first half of 1999, five farmer education days were held, in which 107 farmers and 40 extensionists participated. All districts in the Central Province were covered and one district each from the Eastern and Rift Valley Provinces was included. The farmer education days are being continually improved, following feedback from farmers. Programs for more specialized training are planned, as well as demonstration visits held at farmers' ponds.

#### INTRODUCTION

A lack of technical training was cited as a major reason for the low output of fish ponds in Kenya. The need for technical training was observed at all levels, from the lowest level extension agent through university levels. The training program undertaken by PD/A CRSP researchers in Kenya seeks to improve training and to provide a cadre of trainers who have extensive practical fish production experience. This activity was originally planned to include training only for university students and fisheries officers at all levels, but has been expanded to include farmer training as well.

The following were the objectives for the training program:

- To increase the pond management skills of fisheries personnel currently involved in aquaculture extension activities in Kenya, and
- To enhance the research and extension capabilities of Kenyan university students likely to be employed in the aquaculture sector.

#### **UNIVERSITY STUDENT TRAINING**

Three MS students from the University of Nairobi conducted their thesis research at Sagana with supervision by the CRSP US Principal Investigator, Ms. Karen Veverica. They were Mr. Paul Izaru, Mr. Wilson Gichuri, and Ms. Patricia Mwau. All three handed in the final drafts of their theses in November and December 1998. Theses are sent to outside readers, after which a defense is scheduled. To date, none of the three theses have been scheduled for defense. Students have no choice but to wait for a reaction from their department. Mr. Paul Izaru elected to stay on at Sagana to write a proposal for Ph.D. research. Mr. Wilson Gichuri returned to his job as a Fisheries Officer and is awaiting assignment to Sagana. In the meantime, he has submitted a proposal for further research to the International Foundation for Science. The third, Patricia Mwau, is seeking employment. Graduate-level training conducted in 1998–1999 is shown in Table 1.

Undergraduates at Kenyan universities are required to do a six-week "attachment" in which they learn the practical aspects of station work and often take on a special subject for their Attachment Report. If the student can stay longer, they often conduct their Senior Project research, for which a report is due at the end of their senior year. Student stipends from the CRSP have made it possible for some students to remain at Sagana for the whole break between their junior and senior years, thus allowing them to complete a senior project. Names and senior project subjects for these students are presented in Table 1.

#### FISHERIES OFFICER TRAINING

A key part of the overall training activity has been the selection of the candidate for training to fill the Fisheries Department's Research/Extension Liaison position. The selected candidate would be eligible to receive an MS scholarship sponsored at least in part by the PD/A CRSP. This training thus relates to both university student training and fisheries officer training. Mr. B. Omolo was selected from a short list of seven candidates for the scholarship. His maturity and experience were the major factors in the selection, which was conducted by a training committee made up of seven people. Mr. Omolo has applied to two graduate programs and is currently arranging to take the necessary entrance exams. His MS program will be at least partially supported by the CRSP.

Fisheries officers assigned to Sagana Fish Farm have asked for assistance in conducting small research projects. Assistance in experimental design and in materials is provided by the CRSP (Table 2). These projects further the professional development of the officers and provide background information and experience for writing proposals for more detailed research. Some of the projects, such as those looking at *Clarias* fingerling production techniques, have immediate impact on station management. One of the fisheries officers, Mr. Felix Lagat, has won a scholarship to Belgium for an MS program in aquaculture at University of Ghent. He will be leaving in September 1999.

Two staff members at Sagana Fish Farm were selected for further professional training, fully financed by the PD/A CRSP. John Maina Kamau followed an intensive computer course at Kenyatta University. He has completed his exams and is awaiting certificates for the units completed. Mr. James Karuri has received a scholarship for a three-year diploma program in Applied Biology at the Murang'a College of Technology. He began his first year in January 1999.

#### FARMER AND EXTENSION AGENT TRAINING

A planning meeting was conducted for district fisheries officers, in which the latest PD/A CRSP research results were presented and during which farmer and extension agent training sessions were planned. Quarterly meetings such as this with district fisheries officers are needed, but budgetary constraints often mean that meetings occur only once a year. PD/A CRSP funds can help increase the frequency of fishery officer meetings.

Table 1. Participants, university affiliation, and project of students who attend the university-level training conducted in 1998-1999. Mr. Meso's stipend was paid under the PD/A CRSP Effluents and Pollution Research project.

Name	University / Department	Project	Date Started
Attachments			
Paul Wamwea Wabitah	Kenyatta University/ Zoology	Senior project in comparison of tanks and hapas for sex reversal of tilapia	Intermittent, as of May 1998
David Mirera William Nyaga	Moi University/Fisheries Moi University/Fisheries	Senior project on primary productivity indicators Senior project on treatments to enhance survival of goldfish larvae	May 1999 May 1999
Cosmas Munga	Moi University/Fisheries	Senior project on Clarias larvae feeding strategies	May 1999
GRADUATE STUDENTS			
Daniel Oenga Nyanchiri	Moi University/Fisheries	Largemouth bass introductions and fingerling production	July 1998
Bernard Meso	University of Nairobi/ Soil Sciences	Irrigation of horticulture crops with pond water effluents	September 1998
Paul Bilal Izaru	University of Nairobi/ Zoology	Phytoplankton dynamics at different nitrogen and phosphorus input levels	October 1998
Paticia Nduku Mwau	University of Nairobi	Nitrogen and phosphorus budgets in tilapia/ <i>Clarias</i> polyculture ponds receiving different combinations of rice bran and chemical fertilizer	
Wilson Maina Gichuri	University of Nairobi	Comparison of chemical fertilizer and rice bran in different combinations as inputs for tilapia/Clarias polyculture	

Table 2. Fisheries officers assigned to Sagana who have worked on their own research projects, with advice and materials provided by the PD/A CRSP.

Name	Project Title	Status	
Charles Gatune Felix Lagat and Stephen Njao	Production of ornamental fish Feed electivity indices for <i>Clarias</i> and tilapia in fertilized ponds	Report in progress. Data collected; now in analysis.	
Rapĥael Mbaluka	Construction and testing of fish graders <i>Clarias</i> fingerling production technologies	Data collected. Data collected on first experiment; second experiment to begin August 1999.	

With reduced government spending, the extension service cannot be expected to disseminate information to all farmers. Therefore a program of farmer training, in which as many farmers as possible receive some information, seems to be the best option. To assist the extension service a program of farmer education days was developed by Mr. Omolo, K. Veverica, and the Social Development Officer, Judith Amadiva. Short (oneday) training sessions were planned because it is difficult for farmers (especially women) to leave their homes for long periods. The first farmers' education day was requested by the newly formed Mt. Kenya Fish Farmers' Association. Education day packages consisting of binders containing fact sheets are

Table 3. Subjects covered in the farmers' education day package. Fact sheets were printed for each of the subjects and presented to farmers in a binder. Two fact sheets have not yet been prepared: fish harvesting methods and fish preservation. Translation into Kiswahili is being considered but the fact sheets still need some editing. Students on assignment at Sagana accompany the trainees and help with translations into Kikuyu.

Subject	Instructor
Pond Management—Especially Water Flow Control and Weed Control	Veverica
Feeds and Feeding	Lagat or Gichuri
Fertilizing Options and Rates	Lagat
Predator Control	Oenga or Veverica
Parasites and their Prevention	Njau
Requirements and Biology of Tilapia, Catfish, and Carp	Omolo
Stocking Rates as They Relate to Carrying Capacity and Desired Size	Mbaluka
Tips on Pond Construction and Pond Maintenance	Veverica
Integration of Farm Practices with Fish Ponds	Meso or Njau
Fish Drying and Smoking	Wasane
Fish Harvesting Techniques	Kibe or Makau

prepared for the trainees at each education day; subjects covered in these packages are described in Table 3. As the field days continue, new subjects are added and new handouts are developed. More specialized programs will be offered in the future. Feedback from farmers has been very positive and encouraging. A few simple ideas such as water inflow control and pond fertilization were totally new to the majority of farmers. Although meetings can be held elsewhere, most are held at Sagana, which is an ideal place because farmers accept information on pond management more readily when they actually see a pond with static water in which fish are feeding actively. One on-site demonstration on fish harvesting and preservation was held at a large, seven-acre dam in Kerugoya. Farmer training/education programs that have were held in 1999 are summarized in Table 4. More of these programs are foreseen, because they seem to be more easily attended by women and they allow for additional involvement of onlookers and school teachers.

#### **Observations on Farmer and Extension Agent Training**

Extensionists were selected by their superiors based on the likelihood that they would use their new information to the benefit of the farmers. The selection process seems to have worked fairly well. Most extensionists were very eager to learn more and complained about never having received training. Only a few showed little interest in the subjects. Extensionists who were the most eager to learn will be invited to a longer session later in 1999. They will receive a certificate after the one-week training.

The farmers were asked what kinds of new things they learned at the education days. The following is a list of their answers with the more frequent appearing first:

- 1) Flowing cool water through the pond is not a good thing for tilapia and *Clarias* production.
- 2) Inputs do not have to be purchased but many things available on the farm can be used as feeds or fertilizer.
- 3) Chemical fertilizers can be used in ponds.
- 4) Clarias catfish is a good second species to grow with

Table 4. Farmer/Extension Agent training programs in 1999. The term fisheries officer is used loosely; it indicates Fisheries Department staff mostly fisheries officers, assistant fisheries officers, or fisheries assistants. Except as noted, education days were held at Sagana Fish Farm.

Date	Farmers		Fisheries Officers		Origin	Subjects
	Women	Men	Women	Men		
11 March	5	13	1	1	Kiambu	Introduction to fish farming
18 May	5	25	1	9	Nyeri, 7 divisions	Farmers education package
24–25 June	0	0	0	5	Central Province	Research results of CRSP on-farm trials
11 June*	7	28	0	0	Kerugoya	Fish harvesting and processing
8 July	0	10	0	5	Kirinyaga District	Farmers education package
5 5	0	5	0	0	Muranga DST	Farmers education package
20–21 July	2	13	1	3	Thika and Kiambu Districts	Farmers education package
5 5	1	5	0	6	Nyandarua District	Farmers education package
29 July	2	9	2	6	Nyeri District	Farmers education package
5	2	6	1	1	Nyanyuki District	Farmers education package
	0	6	1	1	Embu District	Farmers education package
	0	0	0	1	Murang'a District	Farmers education package

\* Held at a reservoir site in Kerugoya.

tilapia and it consumes those tadpoles and frogs that are such a bother.

- 5) Just about anybody can smoke and dry fish for their own use; it is not so difficult.
- 6) Some of the algae growing on the pond surface, like *Euglena*, are not a problem for tilapia and are consumed by them.
- 7) Fish can taste really good (even catfish) if prepared correctly.
- For optimal water regulation, it is better to capture a spring and divert its water to the pond when needed instead of growing fish right in the cool, flowing spring.

The water flow question appears to be the single most important issue in improving fish growth and production. Most extensionists were previously taught to continually flow water through ponds. Unfortunately, most surface waters in the Central Province are very cold and have very low total alkalinity and hardness. This is a recipe for disaster in low-input warm water fish culture ponds. But simply *telling* farmers that static water is better was not enough—the farmers needed to *see* ponds in which no new water had been added during the previous five months, except for topping off to replace water lost to evaporation. At Sagana the farmers saw such ponds and the fish produced in them. They also saw evidence that Sagana Fish Farm's leakiest ponds are consistently the poorest producers. As the education days start to include farmers from greater distances, the program will have to be lengthened to allow farmers transport time and to make the relatively high transport costs pay off. Alternatively, other training centers can be established. However, it is imperative that good pond management techniques be observed and not just talked about, so any other sites considered for training will need to have appropriate examples in the vicinity.

#### **ANTICIPATED BENEFITS**

This activity is providing university students, fisheries officers (including those involved in extension efforts), and farmers with improved fish handling and pond management skills. Short training courses are improving technical confidence and morale among extensionists. Linkages between research and extension activities in Kenya are being strengthened. Ultimately, better pond management by farmers will lead to increased fish production, increased farm income, increased amounts of fish available to communities and markets, and increased employment opportunities. Support and hands-on guidance of graduate students in aquaculture will strengthen their degree programs and ultimately help promote productive and sustainable aquaculture growth in Kenya and in the region.



# PD/A CRSP Seventeenth Annual Technical Report

# ESTABLISHMENT OF COMPANION SITES IN THE AFRICA REGION

Ninth Work Plan, Adoption and Diffusion Research 4 (9ADR4) Progress Report

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#### Abstract

The establishment of one or more companion sites in the Africa Region was proposed as a way of verifying the results of CRSP research at its prime site and of expanding the regional effort of the CRSP by assisting with the conduct of needed research at other sites in the region. The objectives specifically listed for this effort in the Ninth Work Plan were: 1) to identify and establish one or more companion sites for the Africa Region and 2) to define and implement investigations at the companion site in support of PD/A CRSP and companion site goals. The first of these two objectives was to be achieved during the first year of the Ninth Work Plan. Sites previously identified for possible companion sites included Bunda College of Agriculture (Malawi), Kingolwira Aquaculture Center (Tanzania), and Akosombo Aquaculture Research and Development Center (Ghana), but investigation of additional sites could also be undertaken. During this first year of the Ninth Work Plan (late 1998 and 1999), discussions have focused on collaboration at sites in Malawi, resulting in a recent decision to propose companion site efforts in collaboration with ICLARM at the National Aquaculture Center (Zomba) and Bunda College, near Lilongwe

#### INTRODUCTION

The establishment of one or more companion sites in the Africa Region was proposed as a way of verifying the results of CRSP research at its prime site and of expanding the regional effort of the CRSP by assisting with the conduct of needed research at other sites in the region. The objectives specifically listed for this effort in the Ninth Work Plan were:

- 1) To identify and establish one or more companion sites for the Africa Region and
- 2) To define and implement investigations at the companion site in support of PD/A CRSP and companion site goals.

The goal for the first year under the Ninth Work Plan was to meet the first of these two objectives, i.e., to identify and establish at least one companion site in the region. The first investigation under this activity would be conducted during the second year.

# **PROGRESS TO DATE**

Discussions on the possibility of conducting companion site research in Malawi were begun when the sites at Bunda College and the National Aquaculture Center (NAC), managed by the International Center for Living Aquatic Resources Management (ICLARM), were visited during site evaluation visits by Wayne Seim, Karen Veverica, Jim Bowman, and Tom Popma in 1995. Although several other possible sites have also been under consideration, these discussions have continued through the present time, both through personal visits and through email and telephone communications, and it was concluded that the initial companion site efforts are most likely to be successful if conducted in Malawi. Recent (Spring 1999) discussions culminated in a visit by ICLARM/Malawi Project Leader Daniel Jamu to Sagana Fish Farm to meet with the CRSP US Principal Investigator, Karen Veverica, to consider a three-way collaboration between the PD/A CRSP Kenya Project, Bunda College, and ICLARM/ Malawi. Several possible experiments were discussed and the protocols used in CRSP experiments were reviewed. A visit to Malawi by Veverica was considered unnecessary at this time because the companion site discussion has been going on quite some time between ICLARM staff and Bunda College representatives. On returning to Malawi, Dr. Jamu will consult with representatives from Bunda College and present a proposal for one or more experiments to the CRSP Africa Project.

Under the most likely arrangement, students from Bunda College will conduct research at the NAC, with much of the limited research funding going to student stipends. Working in Malawi under such an arrangement seems most suitable because:

- Malawi is the seat of the Fisheries Office for the Southern African Development Community (SADC), and Bunda College is the training site for Fisheries and Aquaculture for the SADC countries,
- The Domasi station (NAC) has sufficient facilities to carry out research on the small budget allocated,
- There is ample opportunity to include university students from Bunda College as well as from around Africa in the research,
- Collaboration between ICLARM and the PD/A CRSP would be strengthened, and
- The CRSPs regional linkages would be enhanced through collaboration with Bunda College.

Funds for companion site work become available in December 1999, and the first investigation can begin at that time. Contacts with other companion site candidates will be maintained, in the event that further funding is available.

#### **ANTICIPATED BENEFITS**

Research at a companion site will address areas of research that are mutually useful, i.e., that will advance lines of investigation already being pursued by the CRSP and also address the needs of fish farmers in the vicinity of the companion site. Students in aquaculture programs will receive training through their involvement in the research. CRSP findings from prime sites may be verified under the differing conditions of the companion site, and performance testing of tilapia species or strains not present at the CRSP prime site can be undertaken. The CRSP Central Database will be broadened through the inclusion of data from additional sites in Africa. Companion site researchers will benefit from data collected during the course of experiments, and improved fish farming methods resulting from the experiments will be available for adoption by fish farmers in the area around the companion site. Ultimately, fish farmers in new areas will experience increased fish yields, and greater amounts of fish will be available for consumption in communities and markets in those areas.



# PD/A CRSP Seventeenth Annual Technical Report

# **REGIONAL OUTREACH IN AFRICA**

Ninth Work Plan, Adoption and Diffusion Research 5 (9ADR5) Progress Report

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#### Abstract

Personnel involved with PD/A CRSP research in Kenya attended a conference entitled "Shallow Water Bodies in the Tropics," held in Naivasha, Kenya, from April 12 to 16 1999. Four presentations were made, of which two were based on CRSP Eighth Work Plan feeds and fertilizers research ("Relative Contribution of Supplemental Feed and Inorganic Fertilizers in Semi-Intensive Tilapia Production"). Attendance is also planned for the 17th Conference and Silver Jubilee of the Soil Science Society of East Africa, to be held in Kampala, Uganda, 6 to 10 September 1999 and a conference on the Lake Victoria Basin, to be held in Jinja, Uganda, 23 to 26 November 1999. If possible, CRSP personnel will also attend meetings of the Southern Africa Development Community (SADC) and the Fisheries Society of Africa (FISA). Participation in these meetings is part of the CRSPs effort to promote communication and the establishment of linkages among aquaculture research and extension workers and to better understand the needs and constraints in Kenya and surrounding regions.

#### INTRODUCTION

The intent of this activity is to promote contact and communication among aquaculture research and extension personnel and organizations throughout the region. This is to be achieved mainly through participation at regional meetings, not only by presenting papers but also through participation in planning and organizing the meetings and in helping to develop and implement plans to increase participation in them. Through this effort, research results from current and previous CRSP activities can be shared, other African researchers can be met and encouraged, CRSP workers can learn about research and extension efforts in other parts of the region, and linkages for future collaboration can be established. Examples of regional meetings that might be included are annual meetings of the Southern Africa Development Community (SADC) Inland Fisheries Sector Technical Coordination Unit and of the Fisheries Society of Africa (FISA), but other meeting opportunities will also be taken advantage of.

The objectives specifically listed for this effort are to:

- 1) Promote the dissemination of information emanating from PD/A CRSP research results;
- Learn about fish culture practices and research priorities and activities in Kenya and neighboring countries in Africa; and
- 3) Encourage the establishment of regional linkages between research and extension programs in Africa.

This activity continues from a similar activity in the Eighth Work Plan.

#### **PROGRESS TO DATE**

Karen Veverica, three fisheries officers (Bethuel Omolo, Stephen Njao, and Felix Lagat), and four university students (Patricia Mwau, Paul Bilal, Daniel Oenga, and Wilson Gichuri) attended the conference entitled "Shallow Water Bodies in the Tropics," held in Naivasha, Kenya, 12 to 16 April 1999. Four presentations were made, of which three were based on CRSP Eighth Work Plan feeds and fertilizers research ("Relative Contribution of Supplemental Feed and Inorganic Fertilizers in Semi-Intensive Tilapia Production") and one on bass introductions in Kenya. The presentations made were as follows:

- 1) Nitrogen and phosphorus budgets in polyculture fish ponds. P.N. Mwau, K.M. Mavuti, P.I. Bilal, and K.L. Veverica
- 2) Plankton dynamics in tilapia (*Oreochromis niloticus*) and catfish (*Clarias gariepinus*) polyculture ponds in Central Kenya. P.I. Bilal, K.M. Mavuti, J.G. Omondi, and K.L. Veverica
- 3) The history of largemouth bass *Micropterus salmoides* introduction and transfers in East Africa. D. Oenga Nyanchiri, B. Wangila, M. Muchiri, and K.L. Veverica
- Relative condition factors (K<sub>n</sub>) for *Oreochromis niloticus* (Cichlidae) and *Clarias gariepinus* (Clariidae) in small managed ponds. W.M. Gichuri, J.G. Omondi, and K.L. Veverica

The United Nations Educational, Scientific and Cultural Organization (UNESCO) will print a one-page extended abstract of each presentation. Veverica was also able to hand out several publications (some from CRSP) of interest to researchers working in the area of aquatic ecology.

Bethuel Omolo attended the East African Environmental Network (EAEN) conference, 28 to 29 May 1999 in Nairobi.

Attendance is also planned for a conference on the Lake Victoria Basin to be held in Jinja, Uganda, 23 to 26 November 1999. Two abstracts for presentations to be given have been submitted. An abstract based on the first season of the pond effluent use for crop irrigation research (9ER1) was also sent to the 17<sup>th</sup> Conference and Silver Jubilee of the Soil Science Society of East Africa, to be held in Kampala, Uganda, 6 to 10 September 1999.

Karen Veverica maintains active contact with officers of the Fisheries Society of Africa (FISA), which is headquartered in Nairobi, and attended the FISA executive officers meeting on 22 April 1999. The CRSP will help by getting the NAGA subscription form out to all fisheries officers with encouragement to subscribe. Others present at the meeting included E. Okemwa, the president of FISA; N. Githonga, secretary; Randall Brummett, ICLARM/Africa; Mike Kittivo, Tana and Athi Rivers Development Authority; and Betty Nyandat, District Fisheries Officer for Kiambu.

## **ANTICIPATED BENEFITS**

Contact with researchers and extension personnel in Kenya and in other countries in the region will result in a better understanding of research needs and enhanced researchextension linkages. Extension services in Kenya and other African countries will benefit by being more closely linked with research institutions and African researchers will have an enhanced understanding of research needs. Ultimately, fish producers throughout the region will benefit, because these linkages will enable extension services not only to more easily convey farmers' needs to researchers, but also to extend new research results back to the farmers.



# ENHANCING THE POND<sup>©</sup> Decision Support System for Economics, Education, and Extension

Ninth Work Plan, Decision Support Systems Research 3 (9DSSR3) Progress Report

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#### Abstract

Decision support systems (DSSs) are potentially valuable tools for assessing the economic and ecological impacts of alternative decisions on aquaculture production. This report discusses the latest design, functional modules, and application areas of POND<sup>®</sup>, a decision tool that has been developed to allow analysis of pond aquaculture facilities by the use of a combination of simulation models and enterprise budgeting. The software makes use of a simulation framework to provide much of the generic simulation, data handling, time flow synchronization, and communication features necessary for complex model-based DSSs. POND<sup>®</sup> contains representations for manipulating pond aquaculture and utilizes a series of mini-databases, a number of knowledge-based components ("experts"), models of the pond ecosystem, and various decision support features (e.g., assembling alternate management scenarios, economic analysis, and data visualization). A typical POND<sup>®</sup> simulation consists of assembling a number of appropriate objects or entities (e.g., multiple ponds and fish lots) and their management settings together with appropriate experts (e.g., an aquaculture engineer, an aquatic biologist, and an economist), and projecting changes in the facility over time. Most recent efforts have focused on improving the economic analysis capabilities of POND<sup>®</sup> and improving the usefulness of the software for addressing specific needs of the education and extension community.

#### INTRODUCTION

Ponds used for aquacultural production are typically complex systems which can be driven by a wide range of inputs and interactions. This complexity can make designing and managing these systems challenging; successful fulfillment of these tasks can often be assisted by the application of tools which capture important system drivers and their interactions. These drivers can be both ecological and economic in nature. Within the realm of pond aquaculture planning and management, decisions must be made regarding site locations, target fish species, and appropriate practices such as fish feeding, pond fertilization and liming, stocking densities, aeration, and water exchange (Boyd, 1979; Colt, 1986; Hepher, 1988). These decisions typically have considerable effects on resource use efficiency and therefore the economics of an aquaculture facility. The decisionmaking process typically requires some expertise on the part of the planner, manager, or extension agent. Such expertise includes an understanding of the principles of pond aquaculture and the implications of various decisions on facility-level economics (Shang, 1981). In certain situations, it may also be necessary to address socioeconomic issues such as receptivity of farmers to new technology and alternative uses of available resources (Molnar et al., 1996). Decisionmakers usually acquire the required knowledge via a combination of formal education and experience.

Often, the immediate need for pond aquaculture technology may cause decisionmakers to apply or recommend manage-

ment practices developed and tested at one location to a new site, without first assessing the appropriateness of the technology. A technology found to be suitable for one location may very well be inadequate when applied elsewhere (Colt, 1986). This may be due to differences in fish production potential caused by the variability in climate, water, and soil characteristics among sites and differences in the availability and cost of resources used in pond production (Shang, 1981). For example, a decision as specific as the calculation of feed requirements for a pond requires consideration of fish biomass, natural food availability, and water temperature, which vary both with time and among different locations (Hepher, 1988). Similarly, calculation of fertilizer application rates requires a basic understanding of soil and water chemistry, both of which also vary among different sites. In both cases, availability and cost of appropriate inputs should be factored into the decisionmaking process (Shang, 1981).

The complexity of decisionmaking for an aquaculture facility resulted in the development of POND<sup>©</sup>, a software decision support tool for analyzing and projecting important cultural and economic aspects of warmwater aquaculture production systems. POND<sup>©</sup> has been through several iterations of development, with effort during the first year of the Ninth Work Plan focused on completing POND<sup>©</sup> Version 4.0. Previous iterations of POND<sup>©</sup> have been documented in PD/A CRSP Annual Technical Reports. Here, we provide a current overview of POND<sup>©</sup> and discuss the changes in design incorporated into Version 4.

# The POND<sup>©</sup> Architecture

In developing POND<sup>©</sup>, we utilized an existing simulation framework (Bolte et al., 1993), which provides a wide range of simulation services for managing collections of interacting simulation objects. These services include:

- a) Basic time-flow synchronization of system components;
- b) Data storage, collection, display, and output;
- c) Linear programming tools for optimization; and
- d) Parameter estimation methods (for determining best-fit model parameters).

The framework also provides a generic simulation object class, which was subclassed into specific simulation components relevant to pond modeling and decision support. The framework also relieves the developer of much of the management of simulation details and instead allows focus on the specific components of the (physical) system to be modeled. This approach has proven to be an effective and powerful approach for model and/or DSS development and has provided the ability to effectively share simulation objects between applications. Because the underlying framework takes care of simulation details, the primary task in developing POND<sup>©</sup> was to specify important factors controlling the dynamics and decisionmaking processes of an aquaculture facility and to define a corresponding set of simulation objects. It was important that these components allow simulation of pond dynamics at both the individual pond level, as well as at the facility level. Addressing this need involved providing capabilities for simulating processes within a pond as well as allowing the definition of multiple ponds and multiple fish lots (i.e., a population of fish stocked in a pond), each with their own characteristic data. Simulation of dynamic pond process requires expertise from a number of domain areas, include aquatic biology, aquatic chemistry, fish biology, fish culture, aquacultural engineering, and economics. In an aquaculture facility, each of these domain areas is typically represented by well-defined entities; a facility is a collection of these entities, operating under a particular management context to allocate resources and produce fish.

POND<sup>®</sup> contains a series of mini-databases, which are accessible to the various objects in the software. For instance, databases are maintained for each lot and pond in a facility as well as for simulation settings, economic information, soil types, fertilizers, feeds, liming materials, site information, and weather characteristics. The software also has an experimental database that allows users to specify the combination of the above databases to be used in a model experiment. This feature has proven useful for quickly assembling and executing relatively complex scenarios of alternate pond management practices.

Additional objects representing "experts" managing the facility were defined. These experts include 1) an aquatic chemist, with the ability to perform a wide range of water chemistry calculations, 2) an aquatic biologist, with the ability to perform functions related to fish growth and algal dynamics, 3) a weather manager, with the ability to estimate weather conditions for specific sites, 4) an aquacultural engineer, with the ability to perform heat and water balance calculations, among others, and 5) an economist, capable of performing enterprise budget analyses and managing costs of various facility operations.

The various experts in POND<sup>©</sup> have capabilities for simulating different aspects of production. These areas include fish

performance, water temperature, water quality dynamics, and primary and secondary productivity. Models in POND<sup>©</sup> are organized hierarchically into two levels, allowing users to perform different kinds of analyses based on data availability and output resolution requirements. Level 1 models are fairly simple, require minimal data inputs, and are intended for applied management and rapid analysis of pond facilities. At this level, the variables simulated are fish growth (based on a bioenergetics model) and water temperature. Consumption of natural food by fish is assumed to be a function of fish biomass and appetite. Fertilizer application rates are typically userspecified, but the model optionally generates supplementary feeding schedules.

Level 2 models provide a substantially more sophisticated view of pond dynamics, allowing prediction of phytoplankton, zooplankton, and nutrient dynamics (carbon, nitrogen, and phosphorus) in addition to fish growth and water temperature. This modeling level is intended for detailed pond analysis, management optimization, and numerical experimentation. Fish can feed from natural and/or artificial food pools. Consumption of natural food (phytoplankton and zooplankton pools) by fish is predicted on the basis of a resource competition model and also depends on fish appetite. At this level, a constant, user-specified concentration of pond nitrogen, phosphorus, and carbon is assumed. Mass balance accounting for each of these variables is maintained, allowing estimation of fertilizer requirements necessary to maintain steady state levels. Level 2 models generate both fertilization and feeding schedules. Further details regarding the models in POND<sup>©</sup> and their verification can be found elsewhere (e.g., Nath, 1996).

#### Economics

POND<sup>©</sup> allows the incorporation of economic analyses of facilities in the form of enterprise budgets. Enterprise budgets allow for the accumulation of various types of cost and income streams, summarized and coupled with interest and depreciation expressions, to assess the overall economic viability of a particular production enterprise. POND<sup>©</sup> supports three cost categories: 1) fixed, 2) depreciable, and 3) variable costs. Fixed costs are those costs that do not change over the course of facility operation (e.g., construction cost for a pond, a one-time cost that does not vary over time). Related to fixed costs are depreciable costs, which typically are used for items that require up-front expenditures, but which may have some backend redeemable value after some period of time. POND® incorporates depreciation schedules describing the loss of value of the depreciable asset over time. An example of a depreciable cost is a tractor, which has an initial cost as well as a resale value after some period in use at the facility. Variable costs are those costs that are not fixed or depreciable and typically vary according to the scale of production (e.g., labor costs, fertilizer and feed costs, and fuel and electricity costs).

To generate an enterprise budget, income sources are also required. POND<sup>©</sup> allows the specification of any number of income sources, based on either a per unit area, per unit of production, or per facility basis. The facility simulator provides income sources relating to fish production. Additionally, interest rates used for calculating fixed and variable investment costs are required. After specifying each cost by an amount—a cost type (fixed, depreciable, or variable), basis (per unit area, per unit of production, or per facility), and other related information—the economics module in POND<sup>©</sup>
summarizes costs on an areal, per unit of production or per facility basis, balances those costs against income, and reports the results in a tabular form. By including and/or excluding particular costs/incomes, or adjusting cost/income details, one can quickly "experiment" to determine alternative possibilities for the economic viability of various facility and management configurations. Version 4 expands on previous enterprise budget capabilities in several important areas. The first of these is an ability to specify market prices for different sizes of a particular target species, thus determining a) at what point in the culture period returns from harvest begin to exceed fixed and variable costs (where the facility becomes profitable) and b) where the optimal harvest time occurs, based on maximizing marginal returns relative to marginal costs. A second enhancement is the ability to schedule periodic costs, allowing more accurate representation of annual cycles of production and providing the ability to conduct long-term facility analyses.

## **EXPERIENCE GAINED**

The design, development, and implementation of the POND<sup>©</sup> software has provided useful lessons in several areas. As the user base for POND<sup>©</sup> has grown, we have had the opportunity to solicit feedback on how well POND<sup>©</sup> is addressing user needs. The first lesson is that a diverse group of users has used POND<sup>©</sup> to address a diverse set of needs. Although we originally anticipated a research-focused audience, our largest group of users has in fact been commercial aquaculture facility managers. The primary focus of this group has been on economic analyses, with the utility of the biological models contained within POND<sup>©</sup> of secondary importance. An additional audience has been educators using POND<sup>©</sup> in the classroom as a tool for examining pond dynamics, where the biological models play a more important role. Each of these groups has a different set of interests and a different user interface requirement, and the "one size fits all" approach that we initially used has not been optimal in addressing their respective needs.

Feedback from POND<sup>®</sup> users primarily pertained to the ease of use of the software. Although we have spent considerable effort in developing a modern user interface for POND<sup>®</sup>, because of the underlying complexity of the models POND<sup>®</sup> employs and our desire to fully expose these models, the user interface proved to be burdensome for many users. Exposure of the underlying models is helpful to those focused on understanding the detailed biological dynamics of these systems, but it is less helpful or irrelevant to those focused primarily on economic analyses of facility operations.

We have started to address this issue in the latest release of POND<sup>©</sup> in two ways. First, the focus of POND<sup>©</sup> Version 4, from a user interface perspective, is more on decision support and less on models of the underlying biophysical system. The underlying models continue to be an essential tool for supporting decisionmaking but are less apparent to the user. In other words, they operate in the background but play a secondary role to higher-level decision-making processes. Second, we have introduced a series of "wizards" into POND<sup>©</sup>. These wizards are software tools that "walk" the user through specific and frequently used tasks (e.g., estimating feed, fertilizer, and water requirements and associated economic implications). The wizards hide much of the complexity of these tasks and provide immediate help to users in accomp-

lishing their goals. We anticipate continued development of these wizards to address specific needs of different user groups and to improve ease of use of the program.

The development of specific wizards was driven by 1) identifying frequently used application areas within POND<sup>©</sup> and 2) identifying areas which were sufficiently complex to warrant additional user support. This resulted in the development of the following wizards, which have been incorporated into Version 4:

- A *pond setup* wizard to enable users to define new ponds at their facility;
- A *lot setup* wizard to enable users to define new lots that are associated with specific ponds;
- A *fertilizer* wizard to generate routine pond fertilization recommendations;
- A *liming* wizard to estimate lime requirements for ponds associated with specific soil types;
- A *feed optimizer* wizard to generate feed schedules that minimize the amount of feed needed to reach a specified fish target weight;
- A *water balance* wizard to conduct water balance calculations and estimate water requirements;
- An *economics* wizard to assist in calculating enterprise budgets and optimize economic performance; and
- A *simulation* wizard to conduct facility-level simulations at a given site and view simulation results in graphical and tabular formats.

The target audience for POND<sup>©</sup> continues to evolve. The bulk of our requests come from commercial producers looking for tools to improve their ability to design and manage their facilities. We are examining the possibility of developing an additional product specifically oriented towards routine management of aquaculture ponds. It is expected that some of the components developed and tested in POND<sup>©</sup> will be reused for this product. Another audience from whom we have had numerous requests is small commercial entrepreneurs, often new to aquaculture, who are looking for tools to help them explore the financial feasibility of launching an aquaculture venture. Both of the above groups tend to focus primarily on economic analyses but require some basic understanding and consideration of the biophysical processes underlying facility operation. Aquaculture educators represent a final audience for POND<sup>®</sup>. Their requirement is for readily accessible tools that students can use in class to enhance their understanding of the biophysical processes controlling aquaculture ponds, as well as to complete specific design tasks related to facility management.

## **ANTICIPATED BENEFITS**

The work accomplished in this study will provide improved analytical tools for managing warmwater aquaculture facilities and increase understanding of the economic implications of various facility configurations and management options. Additionally, it will provide tools for educators to integrate the biological and economic aspects of pond dynamics in a readily accessible format appropriate for students to explore important issues of pond production.

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