## DEVELOPMENT AND EVALUATION OF POND INVENTORY METHODS

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## Reporting Period

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## PROJECT OBJECTIVES

1. Determine the most accurate and reliable methodologies for estimating ornamental fish density and size distribution in commercial ponds.
2. Determine the most accurate and reliable methodologies for estimating crawfish density and size distribution in experimental ponds and develop a means to estimate annual yield and harvest size from sampling methodologies.
3. Modify the Aquascanner Catfish SONAR system to size individual catfish collected from commercial catfish ponds.
4. Develop and evaluate several down-looking and low frequency side-scan sonar technologies to determine numbers of channel catfish in ponds.
5. Develop and evaluate a catfish trawl and portable computing technologiesto estimate size distribution and biomass in catfish production ponds.

## ANTICIPATED BENEFITS

Maintaining accurate inventory records in large earthen-pond aquaculture systems has always been problematic. Accurate biomass, headcount and size distribution information is critical for production management, business planning, accounting, and operation financing. This project will enhance current pond inventory methodologies and will
foster the development of novel techniques and/or equipment to objectively assess biomass, headcount and size distribution information in aquatic production systems. These technologies will help to improve the long-term sustainability of aquaculture production in the southeast.

## PROGRESS AND PRINCIPAL ACCOMPLISHMENTS

Objective 1. Determine the most accurate and reliable methodologies for estimating ornamental fish density and size distribution in commercial ponds.

University of Florida. Traditional methods for estimating inventories throughout the ornamental fish production cycle are primarily based on personal knowledge of number of fry per spawn (egglayers), general productivity of broodfish (live-bearers), general observations during growout, and historical production from individual ponds. It was hypothesized that these methods were extremely inaccurate and led to poor management tools within the industry. Very little, if any, record keeping is used. Business planning and comparing results of changes in production methods is, therefore, difficult. Improved techniques for estimating ornamental fish density and size distribution in commercial ponds are needed.

In this project, traditional inventory estimation methods were compared to actual inventory counts from experimental ponds located on 4 commercial facilities and at the University of Florida/IFAS Tropical Aquaculture Laboratory (TAL). Actual inventory counts included physical counts of fry and fish, volumetric estimates of number of fry at stocking, sub-sampling of inventory three times through grow-out using partial seine techniques, and physical counts of fish at harvest. A simple Excel spreadsheet was also employed to record stocking, production observations, and harvest data. This
study focused on three species of fish commonly produced in Florida; serpae tetras (Hyphessobrychon serpae); blue gouramis (Trichogaster trichopterus); and swordtails (Xiphphorus helleri).

For serpae tetras and blue gouramis, a similar protocol was developed for comparing estimation and actual counts of number of fry produced per female and number of fry in subsequent pond stocking. With both species, broodstock were selected for health, condition, and similar size. The trials with serpae tetras included 500, 2-gallon spawning tanks, in which one pair of fish were placed. At 96 hours, tanks were numbered and 50 tanks were randomly chosen. The blue gouramis trials included 60 individual 10 gallon spawning tanks, in which one pair of fish was placed. At 96 hours the tanks were numbered and six tanks were randomly chosen. In both cases the producer was not told which tanks were chosen. After tanks were labeled, the producer was asked to estimate the number of fry in each of the spawning tanks three times, using a commonly practiced method of holding the spawning tanks up to the light one by one and estimating the number of fry. Serpae tetra fry were then equally distributed into 28 larval rearing vats, except for the 50 randomly selected tanks, which were used to generate an exact count. Counts were conducted by first euthanizing
the fry in each spawning tank and transferring fry from one spawning tank at a time into a clean, white bucket. Fry were counted by carefully aspirating each fry into a $1-\mathrm{ml}$ pipette. For the blue gouramis, the six tanks which were randomly selected were euthanized and then hand counted using the same method. The actual count was then compared to the producer's estimates.

Fry from each remaining tank were then reared for 3 weeks before being stocked into two grow-out ponds ( 6 -feet deep, 0.04-acres). The producer was again asked to estimate the number of juvenile fish stocked into each of the two grow-out ponds, and selected 10 vats for each pond. To generate our count we randomly selected eight vats and counted each juvenile. For the blue gouramis, a simple reduction method was employed to enumerate the fry in two, 10 gallon tubs which are used to transport fry to the ponds. The water was gently mixed to maximize equal distribution of fry in the water volume, and three individual $50-\mathrm{mL}$ samples were counted. These reduction estimates were then compared to the producers visual tub estimates. The fry were then stocked into 2 growout ponds. All grow-out ponds were visually monitored twice daily by the producer and study staff during daily feedings and weekly for the duration of the 4-month grow-out cycle. Actual harvest numbers were supplied by the producer which reflected exact sales receipts. For the swordtail trials, two commercial producers and ponds at the Tropical Aquaculture Laboratory were used. Attempts to assess inventories on two participating commercial facilities were discontinued due to catastrophic losses which occurred on both farms during both summers during this study. Using data collected at the Tropical Aquaculture Laboratory, we were able to evaluate expected production of swordtails in open ponds. Based on previous data from tank studies, predicted production $=1.32$ fry $/$ female $/$ day. Five ponds were each stocked with 200 female and 50 male swordtails, and fed a standard diet for 12 weeks (note: 12 weeks is the typical time needed
to reach market size for swordtails).

Use of a simple Excel spread sheet to accurately record stocking dates, number stocked, regular observations, and harvest data has proven cost effective and beneficial to farms employing it. An unexpected result is that the local USDA Farm Service Agency is now using the Excel sheets developed as a handout to producers enrolling in the NAP program as an example of the data they are requesting during a disaster program. Numbers of fry generated in hatcheries for serpae tetras and blue gouramis using traditional methods of estimating by sight were grossly underestimated by 3.8 to 4.2 and 8.2 to 8.5 times, respectively. In individual tanks, the magnitude of the difference between actual counts and the producer's estimates increased dramatically as the total number of fish in the tank increased. However, the producer's estimate of the amount of juveniles stocked into each pond was more accurate, and exceeded the actual estimate only by $9.78 \%$. This showed that significant losses are incurred during the larval rearing stage. Better nutrition (i.e. simply increasing the amount of artemia fed to the larval rearing tanks) has led to increased survival at this stage and decreased man-hours in setting up breeding tanks. Similarly, the blue gourami trial showed that actual harvest data was significantly less than anticipated. Lowering the stocking rates has

## Results at a glance...

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& \text { Improved inventory methods based on } \\
& \text { this project are being applied by a } \\
& \text { commercial ornamental fish producer to } \\
& \text { more than } 60 \text { species of fish, resulting in } \\
& \text { significant improvement in production } \\
& \text { efficiency. Another producer reports } \\
& \text { increased revenues of almost } \$ 400,000 \\
& \text { using improved methods based, in part, } \\
& \text { on this project. }
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resulted in similar harvest numbers indicating that initial larval survival is possible and that fewer broodstock are required.

Physical counts of egg-layer fry are now recommended for all species, at least initially, as opposed to using visual estimates. Savings and increased production have resulted in a $50 \%$ reduction in the required broodstock, man-hours, and grow-out time for one farm participating, and they have now employed these methods to 60 varieties of fish. Another producer reports similar percentages and increased revenues of $\$ 392,000$ based on improved management. Several problems occurred with this project as designed. Periodic sampling of fish in open ponds, during production, using seine nets proved unacceptable and ineffective. Fish captured, counted, and returned to the pond experienced mortalities, and sampling was extremely inaccurate using a partial seine method. Increasing the portion of the pond seined to increase the accuracy resulted
in increased mortalities. This method is not recommended for farmers to assess standing crop during production of the species studied. The summer weather in west central Florida during both years of this study resulted in catastrophic losses on both of the commercial swordtail farms studied, which resulted in no usable data. However, work conducted at the Tropical Aquaculture Laboratory has been useful, as previous to this study there was no data available to accurately predict this production in open ponds. Ponds were harvested and all fish counted, resulting in an average total inventory of 20,292 fish, $90 \%$ of what was predicted. Based on this, farms can anticipate an average production of 1.18 fry/female/day in open ponds after 12 weeks of production for swordtails. Recommendations based on results of these studies are being implemented in Extension programs for ornamental fish producers.

Objective 2. Determine the most accurate and reliable methodologies for estimating crawfish density and size distribution in experimental ponds and develop a means to estimate annual yield and harvest size from sampling methodologies.

## Louisiana State University Agricultural Center.

Crawfish farming in Louisiana depends upon natural reproduction from indigenous or supplemented broodstock to populate ponds. This subjects growers to great variation in yield and harvest size due to large natural variations in adult survival and reproductive success from year to year and pond to pond. Furthermore, these problems are exacerbated by a lack of predictability and a reliable means of assessing pond inventory. Currently, there is no reliable means of accurately determining the success or failure of young-of-the-year recruitment. Without a means of determining population density and structure prior to initiation of harvesting, economic and business planning and implementation of corrective measures are not viable tools for the producer. Therefore, this project attempted to
eliminate natural recruitment and instead accomplish the task of populating ponds with stocking of hatchlings at known numbers. This was done so that systematic sampling efforts could be employed with the intent of establishing some kind of relationship between sampling (with different gear) and known populations, and furthermore, to determine if harvest results could be relatively associated with sampling outcomes.

A rice crop was established during the summers of 2006 and 2007 in 12, 1-acre experimental plots at the LSU AgCenter's Rice Research Station. Following rice harvest, the pond was managed for crawfish production according to typical rice-crawfish rotational practices in the region. Ponds were not stocked with brood crawfish and fields were

## Results at a glance...

- Sampling with test traps, dip-net sweeps, and a passive experimental sampler generally were good indicators of relative crawfish recruitment density and potential yield. The most accurate pond inventory methodologies were those that sampled around the pond margin rather than in the pond interior and occurred several months after the simulated recruitment period.
additionally treated with a pyrethroid insecticide prior to the permanent flood to eliminate migrant crawfish into the field. Ponds were completely drained and re-flooded with fresh water after 3 days. Crawfish populations were subsequently established by stocking of hatchlings, spawned under laboratory conditions, at known densities and at predetermined timing post flooding. Treatments
consisted of a low ( 3 crawfish $/ \mathrm{m}^{2}$ ) or high ( 6 to 7 crawfish $/ \mathrm{m}^{2}$ ) stocking rate and either single or multiple age classes (biweekly over two months) in a factorial arrangement of treatments.

Systematic population sampling was conducted prior to initiation of harvests and consisted of employing four sampling gear: large mesh traps, consisting of standard 0.75 -inch square mesh pyramid traps, small mesh traps, consisting of common 0.25 -inch wire mesh minnow traps with 1.25 -inch funnel openings at each end, long handle dip nets ( 3 -mm mesh), and specially constructed drop sampling devices $\left(0.5-\mathrm{m}^{2}\right.$ surface area; Figure 1). The drop sampler consisted of a metal cylinder that was rigged to slide up and down on three legs with a trigger that allowed the unit to be "set" in the up position with 50 feet of rope, whereby the unit could be placed in the pond some distance and triggered from the levee to prevent disturbing of crawfish during sampling. When "dropped," the sampler formed an enclosure entrapping any crawfish that were captured within the interior of the cylinder. Water was pumped out and crawfish counted and sized. Crawfish catch,

total and by size category, were noted for each sampling effort.

Annual yield of market-size crawfish averaged 194 pounds/acre for year 1 and 360 pounds/acre for year 2 , which was lower than the state wide average of about 600 pounds/acre. Average overall capture rate based on number of crawfish stocked was $12.8 \%$ in year 1 and $26.2 \%$ in year 2 . This seems low, but without a means to accurately assess recruitment density in commercial ponds, it is unknown how well these numbers represent the percent recovery of commercial operations. Correlation coefficient is a measure of how well one group of data corresponds to a second group of data. Sampling
efficacy was highly variable from year to year and from gear to gear in this study; but in general, sampling results using baited large-mesh traps (late in the season) and the passive drop sampler were well correlated to both stocking density and yield.

While these findings provided a basic foundation forbetter understanding of the relationships between initial recruitment numbers, surviving population density and resulting yields in crawfish aquaculture suggests that timely sampling has merit for assessing relative population inventories. Further research is needed to develop applied management options and recommendations for maximizing profits based on sampling outcomes.

Objective 3. Modify the Aquascanner Catish SONAR system to size individual catfish collected from commercial catish ponds.

National Center for Physical Acoustics at the University of Mississippi. Pond production of channel catfish is the largest sector of domestic aquaculture. Pond management strategies can be compromised because of inaccurate inventories resulting in continuous culture for several years without a total harvest. Research was proposed to use an acoustic system to provide the size distribution of a pond to aid management decisions which require inventory information. A current method to determine the size distribution of a pond is to pull a sub-sample seine net across a pond to obtain a sample of the fish and manually weigh them in a time-consuming process. The work presented here still uses a sub-sample seine net to collect a sample of fish but uses acoustic backscatter from the fish as they swim back to the pond to determine the size distribution of the fish in lieu of the manual weight measurements.

A prototype acoustic measurement system (Figures 2 and 3) was assembled and tested in various commercial and research ponds in the Mid-south. The system consisted of a SONAR system operating at

460 kHz placed on top of a PVC floatation system which was attached to a 6 -inch PVC pipe extending from the float into the pond. The pipe allowed fish to swim back into the pond after being seined and also acted to restrict fish movement relative to the SONAR's active element, thereby reducing variation in sound reflection due to fish orientation. As fish pass through the pipe, they are pinged by acoustic pulses and the return echo amplitude is recorded and stored on the unit for future analysis. Measurements were made in pens and ponds at the University of Arkansas Pine Bluff as well as commercial ponds at Wilmot, AR. In the preliminary tests, both the acoustic reflections as well as the actual fish weights were collected, on a fish-by-fish basis, to develop an empirical prediction routine relating fish weight to acoustic reflection, also known as the target strength. This prediction routine was later used in blind tests on ponds in Wilmot, AR, Pine Bluff, AR (UAPB) and the National Warmwater Aquaculture Center (NWAC) in Stoneville, MS. The results from the acoustics predictions were compared against actual measured weights to determine efficacy.


Figure 2. The catfish sizing system with electronics and battery mounted on board and bubbler system in place to provide calibrated signal in data.


Measurements show a trend ofincreasing reflectivity, or target strength, with increasing weight of fish, but the variances between predicted and measured weights for individual fish are larger than expected. For instance some small fish ( 0.5 pound) returned echo amplitudes comparable to larger ( 5 pound) brood fish. It is supposed that fish movement, changes in orientation or entrained air bubbles as fish
swim back into the pond from the flotation device may be responsible for the variations. The data also showed a point of diminishing returns in that larger and larger fish return a diminishingly smaller increase in target strength (Figure 4). Echoes from fish weighing 8 pounds, for example, were are not markedly different than 4-pound fish. Such a scenario, in an inversion process which uses the acoustic echo

to predict the weight of the fish, makes electronic errors problematic in potentially predicting large fish weights. The use of the data, in a statistical sense however, may have value. While predicted fish weights differ from actual weights for individual fish (mitigating the technology for use in say vaccine delivery), the errors may cancel so that predictions for a large enough sample, may be useful.

Two blind tests were taken at Pine Bluff and Wilmot, AR. The model predicted average fish weights of 1.42 pounds/fish and 2.15 pounds/fish for populations that had measured average weights of 1.43 and 2.34 pounds/fish, respectively, giving errors of $-0.5 \%$ to $-8 \%$. Four additional blind measurements were later taken at Stoneville, MS. These tests consistently under-predicted measured fish weight by an average of $30 \%$. It is unclear if this discrepancy is due to local pond-specific issues, the distribution of sizes in the fish population, an equipment-specific issue or some other factor. The prediction model used was developed to minimize errors of both the average weight and standard deviation in weight of a sample. It may well be that
improvements can be made in estimating the average weight of a sample but presumably this will come at the expense of the accuracy of the standard deviation. It is also possible that improvements (and consistency) in how the fish move through the pipe may reduce variations in the reflectivity of the fish which should improve predictions. Data and the prediction algorithm are being investigated further as part of a graduate thesis.

## Results at a glance...

- An acoustic backscatter system has been built to measure the target strength of individual fish from a harvested population. A relationship between fish weight and acoustical target strength was developed into a model that can be used to predict the population weight distribution of the fish harvested.

Objective 4. Develop and evaluate several down-looking and low frequency side-scan sonar technologies to determine numbers of channel catfish in ponds.

Mississippi State University. Previous work with custom-fabricated side-scanning SONAR showed that the technology may have application for pondscale counting of fish as part of inventory assessment in catfish farming. Advances in the private sector on other applications of side-scanning SONAR showed further promise, with the possibility of not only assessing fish numbers, but also individual fish weights, which could then be combined to provide population size distributions and total fish biomass.

Initial research focused on testing a 997c Humminbird ${ }^{\circledR}$ side-imaging SONAR. The transducer has down-looking as well as side-scanning capabilities. The side-imaging SONAR must be moving to operate properly; therefore, it was mounted on a boat with a trolling motor. Testing showed that the side-scan mode produced excellent images of the pond bottom and various structural features in both shallow and deep water but must be operated at 455 kHz in shallow water because the higher frequency saturates the water column making imaging impossible. The down-looking mode worked well in deeper water but not in water as

## Results at a glance...

- The DIDSON sonar with its sophisticated software can detect, identify, and measure catfish cultured in shallow, turbid waters. Catfish as small as 4 inches and as large as 30 inches have been detected, imaged, and measured using sonar data files collected at multiple sampling sites in culture ponds stocked with mixed sizes of fish. Such data could be used to develop population size distributions for commercial ponds.
shallow as most catfish ponds. This preliminary work showed that side-scanning SONAR can be used to image fish, and therefore may have the potential to be calibrated to obtain data on fish sizes.

Further work used a DIDSON 300M unit with associated hardware and software. The DIDSON 300 M can be operated at either a high- (1.8 MHz) or low- (1.1 MHz) frequency. The DIDSON unit was deployed using a custom-made, adjustable system that allows quick deployment to the desired depth and rapid re-deployment for multiple sampling. Details of the deployment system and data-processing options for the DIDSON unit can be obtained from Dr. C.D. Minchew at Mississippi State University.

The strength of the DIDSON sonar for studying catfish in shallow production ponds is its ability to record recognizable images of fish in turbid ponds and process that data to obtain estimates of fish size. Useable images have been taken with both the lowand high-frequency beams. Under ideal conditions, ranges of the high- and low-frequency beams are 1 to 15 m and 1 to 35 m , respectively. However, in the present study, the useful range of the two sonar beams was limited to about 8 m (high frequency) and 16 m (low frequency). While the low frequency beam has a longer range, it is limited in its ability to image small fish. Therefore, the high-frequency setting was used to image obtain the following images. Figure 5 shows a large and small catfish swimming together. Fish were sized using the DIDSON "measure tool." The fingerling was 16 cm (6.3 inches) and the larger fish was 58 cm (22.8 inches).

Although not a part of this study, it is clear that the DIDSON has the potential to be useful in studying catfish behavior. It could be used to observe the behavior of pond cultured catfish in response to

Figure 5. A sonar image of a large $(58 \mathrm{~cm})$ and small ( 16 cm ) catfish swimming near each other. The sonar image was taken at a sampling station at Delta Research and Extension Center on August 3, 2009. The pond had recently been stocked with two size classes of catfish as a part of a growth study.

seining, grading, low oxygen, and feeding. For example, Figure 6 shows an image collected while catfish were being fed fed using a tractor-pulled feeder. Additional images were collected during a
severe dissolved oxygen depletion. Both events (feeding and response to low oxygen) are more impressive when examined while running the video of each event rather than as still pictures.

Figure 6. Raw data image of feeding fish.


Objective 5. Develop and evaluate a catfish trawl and portable computing technologies to estimate the size distribution and biomass in catfish production ponds.

University of Arkansas at Pine Bluff. Estimating biomass and size distribution of catfish in earthen production ponds has always been difficult. Accurate inventory estimates are often needed for management considerations, business planning, and operation
financing. New inventory tools and methodologies are needed to help catfish producers obtain accurate inventory data from large earthen pond systems. The objective of this study was to develop a commercial-scale sampling apparatus and then
determine if it could be used to collect accurate inventory data.

We started with two standard pieces of equipment that are commercially available; a hydraulic seine reel, and a standard "otter trawl." The idea was to devise a mechanism that would allow us to lay the otter trawl on the levee on one side of pond, and then pull it through the pond with a rope to the opposite levee to collect our sample. Previously published data indicated that the trawl would need to move through the pond at about 5 feet/second in order to catch catfish consistently. With that in mind, we made several modifications to the seine reel to ensure the trawl would travel at that speed. First, we outfitted the seine reel with a hydraulic system that could be powered by the $540-\mathrm{rpm}$ PTO of any $30-\mathrm{hp}$ (or larger) tractor. This was necessary because the standard hydraulic systems on most fish farm tractors were not designed to deliver the amount of power necessary for this application. We also increased the center drum diameter by welding cross bars to the inside of the spool and decreased the size of the primary drive sprocket to increase the rotational speed. We used an otter trawl, which is a funnelshaped net with a 30 -foot-wide mouth, and "otter boards" that are designed to pull the net open as it moves through the water. Several modifications were made to the original trawl design, including reducing the size and weight of the otter boards, adding mud rollers to the mud line, and removing the trap at the head of the cod end (Figures 7 and 8). Detailed equipment design specifications for the catfish sampling rig can be obtained from David Heikes, Aquaculture and Fisheries Center, University of Arkansas at Pine Bluff.

The sampling trawl was tested in a series of trials conducted in large commercial ponds in Arkansas and Mississippi and also under controlled, replicated conditions at the National Warmwater Aquaculture Center, in Stoneville, MS. In the first commercial pond trials, we wanted to document the performance of the trawl both with and without the use of feeding
to attract fish to the trawls path. Each pond was pulled one time without feeding in the morning and then pulled again after feeding in the afternoon. Following the trawl events in each pond, the entire pond was seined three times using a small mesh fingerling seine to determine the actual inventory. Fish captured by the trawl were returned to the pond before seining in each trial. Fish caught by the trawl were individually weighed and the size distribution compared to samples taken from the actual population caught in the seining events. Results of this trial showed us that the frequency distribution of sizes of catfish caught in the trawl pulls was not significantly different from that obtained from seining. A second trial with a similar protocol was conducted in six, 4-acre ponds at Stoneville. While trawling with feeding caught more fish in each pull, there was no difference in average weight or size distribution between the two trawling methods. These results also indicated that a single trawl pull, either with or without feeding, resulted in an accurate size distribution. However, a single trawl pull, which is similar to the technique known in fisheries as the "swept area method", was inadequate for estimating total pond biomass. These results led to a third trial in the same 4-acre ponds at Stoneville to determine if multiple trawl pulls (without feeding) could lead to a better estimate of total biomass. When the inventory was estimated from pooling the catches

## Results at a glance...

> The sampling trawl techniques developed in this project have been adopted commercially on a limited basis. Using fish-size distribution data generated from fish captured by trawling, managers are able to more accurately identify ponds that are ready for harvest and to schedule harvest dates for ponds that are not currently ready.
from more than one trawl pull (and using the total area swept from the pulls in the calculation), the estimated inventories were similar to that of the base inventory. This indicates that at least under these conditions, the catfish sampling trawl can be used to generate size distribution information as well as an estimate of total biomass.

An additional study was conducted to systematically evaluate the traditional methods of estimating pond inventories that are based on daily feeding response of catfish. Daily feed data from experimental (0.25-
acre and 4-acre) and commercial (10-acre) ponds were used to characterize feed response of catfish and to compare the accuracy and precision of feeding response methods used to estimate fish inventories. Daily feed consumption of catfish was highly variable. Inventories estimated with feed response methods had errors of $16 \%$ to $37 \%$ in single-batch production and $28 \%$ to $49 \%$ in multiplebatch production. Inventories estimated with feed response methods were too inaccurate and variable for reliable use in management, by lenders, or by the court system.


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