Growth, Condition, and Daily Ring Validation of a Cichlid in Puerto Rico

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Abstract: We validated otolith daily rings of *Cichlasoma managuense* using immersion in alizarin complexone to provide a known-age reference mark. Cichlids were stocked at 38 fish/cage into 3 121-liter cages with 5-mm mesh size within an experimental pond in Puerto Rico. The smallest 78% immediately escaped the cages and were at large in the ponds. Initial total length of cage cichlids averaged 26.5 mm compared to 20.4 mm for cichlids at large in the pond, although pond cichlids quickly overcame this deficit and realized growth rates 2 times faster than confined cichlids (1.42 mm/day vs. 0.69 mm/day). By the final sample at day 60, pond cichlids were 47% longer and 234% heavier than caged cichlids, with significantly higher values of condition. There were no differences in ring formation between the 2 groups, and rings were found to be accurate estimators of daily age. Slight under-ageing by the day 60 sample corresponded to true fish ages averaging 110 days, suggesting that the maximum accuracy would be obtained for cichlids <100 days old. We concluded that temperate strategies of daily ageing using otoliths could be applied to this tropical cichlid species up to 100 days old.

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Otoliths have been used to determine individual age of many species of fish in different regions of the world (Summerfelt and Hall 1987). Daily otolith rings have been widely used to estimate the ages of many species of age-0 fishes (Jones 1986), providing valuable information on early life history and factors affecting recruitment (Jones 1993). Daily age information is essential for determination of hatching times, growth, and mortality rates in young fishes (Miller and Storck 1984, Essig and Cole 1986, Graham and Orth 1987, Isely and Noble 1987, Pepin 1989, Jones 1992). It is necessary, however, to validate the temporal periodicity of otolith increment formation because not all species deposit rings daily (Geffen 1992), and increment formation may not occur at the same time in different species (Jones 1986).

In temperate regions, otolith annuli and daily rings have been validated for many species of fish. In tropical systems, however, validation of otolith rings has occurred with variable success. For example, adult largemouth bass *(Micropterus salmoides)* in Puerto Rico fail to produce annual growth rings (Neal et al. 1997), but daily growth rings have been validated and are widely used for this species (Churchill et al. 1995, Neal et al. 1999). Use of daily rings for cichlids has proven more difficult, thus complicating research and management of these species (Neal et al. 1999).

One common method of validation applied to daily otolith ring formation is the use of chemical marks applied by group marking (Blom et al. 1994). This mark serves as a reference point for establishing the periodicity of increment formation following mark incorporation. Fish are generally marked in confinement, and then placed into cages, small ponds, or released into larger systems to be sampled at a later time. Although evaluation of fish released into the system for which validation is required is preferable, this generally requires more marked fish and greater collecting effort. Hence, cage and small pond studies are frequently employed in validation of otolith daily rings, as well as other studies where time and money issues warrant more control and fewer experimental fish.

The primary goal of this study was to determine if rings observed on otoliths of a cichlid species in Puerto Rico are produced on a daily basis. We used guapote tigre *(Cichlasoma managuense),* an exotic cichlid species established in some Puerto Rico pond communities. Additionally, we analyzed growth, condition, and survival of this little-studied species.

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Methods

We collected small (15–27 mm total length TL) cichlid juveniles from an unnamed farm pond in Lajas, Puerto Rico, using small-mesh seines. These fish were immediately transported to Caribe Fisheries aquaculture facility for marking and stocking. We immersed 115 fish for 24 hours in an 18.9-liter bucket containing 123 mg/liter of alizarin complexone (AC) buffered to a pH of 7 using sodium bicarbonate. We chose AC as the marking agent because Neal and Noble (1998) found that it could be used successfully to mark otoliths of a related cichlid species. Only 2 mortalities occurred during our marking process.

Following removal from the marking solution, we measured (TL mm) and weighed (g) each fish, and then stocked 38 fish into each of 3 cages. Cages were placed into a 30 × 10-m pond containing zooplankton, insect, and poeciliid prey. Cages were made from 121-liter plastic containers with 4 48 × 25 cm windows of 5-mm mesh in the sides, and 2 39-cm long crescent-shaped mesh windows in the lid. Mesh was attached to cage walls and tops using hot melt glue. Cage tops were attached firmly using plastic wire wraps. The 3 cages were placed standing in water <1 m deep so that the lids were accessible above the water line.

Inspection of the cages the following day revealed that 78% of the cichlids had escaped through the cage mesh and were at large in the pond. We assumed that the remaining 25 fish were the largest 22% usable to escape the cages, and recalculated starting sizes accordingly. Fish remaining in the cages at this point averaged 6 mm longer than pond fish (26.5 mm TL vs. 20.4 mm TL).

Cichlids were held for 60 days during which water temperatures ranged 25–27 C. Cage and pond cichlids were sampled 10, 30, and 60 days after marking. On the first 2 sampling dates, we targeted 5 fish from cages and 10 fish from the ponds using dip netting and seining. On the day 60 sample, all remaining cichlids were removed from the cages, and the pond was drained for collection of cichlids at large. All fish collected were weighed and measured, and both saggital otoliths were removed.

Otoliths were stored dry until preparation, when right or left sagittae were randomly selected and affixed to a glass microscope slide using thermaplastic adhesive before being ground and polished to the primordia on one side. Otoliths were viewed using a compound microscope with reflected fluorescent light at 100×. Estimated age of each AC mark was given by the number of visible rings outside but not including the mark. For otolith terminology we followed Campana (1992).

We used *t*-tests to compare differences in growth for TL and weight of each group, as well as differences in condition. Condition was measured using a Fulton-type condition factor (K) calculated as $[(W/L^3) \times 100{,}000]$ where W is weight and L is the length of the fish. We used regression to fit the weight-length relationship power function ($W = aL^b$), and linear regression for comparison of K values and of ageing accuracy. All statistical tests were 2-tailed.

Results and Discussion

A total of 84 of the original 113 cichlids (74.3%) were recovered during the 60-day study (Table 1). Recovery rates were only slightly lower for cichlids at large in the pond than caged cichlids (73.9% vs. 76.0%), although it is likely that additional fish went undetected in the thick sediment of the drained pond. Hence, we believe that survival of each group through the 60-day study period did not differ significantly, even though pond fish were smaller at the onset of the study.

Cichlids at large in the pond quickly overcame the size disadvantage, and realized much faster growth than cichlids confined in cages (Fig. 1), with pond fish significantly longer ($t = 2.43$, DF = 13, $P = 0.03$) and heavier ($t = 2.60$, DF = 13, $P = 0.01$) by the day 30 sample. The overall mean daily growth rate for pond fish (1.42 mm/day) was twice as high as cage fish (0.69 mm/day). By the day 60 sample, mean TL of pond fish was 47% higher and mean weight was 234% higher than that of caged cichlids. Condition was higher for pond fish than for cage fish (Fig. 2; $t = 2.51$, DF = 35, $P = 0.02$) of similar size. Statistical comparison of K was limited to similar-sized fish (26–86 mm TL) due to possible biases associated with size differences. Comparison

Table 1. Numbers, mean total length (TL) and weight (Wt), and mean daily growth (DG) rate (mm/day) of caged and at large (pond) cichlids at stocking (day 0) and collected from day 10 through day 60.

Day/Group	*N*	TL (mm)	SE	Wt(g)	SE	Mean DG (mm)
Day 0	113	22.2	0.8	0.20	0.02	–
Cage	25	26.5	0.2	0.29	0.02	–
Pond	88	20.4	0.9	0.15	0.02	–
Day 10	14	28.4	1.1	0.68	0.07	0.58
Cage	5	28.8	2.1	0.61	0.11	0.23
Pond	9	28.2	1.4	0.71	0.08	0.78
Day 30	15	62.7	2.8	5.08	0.64	1.35
Cage	5	54.4	4.3	3.11	0.68	0.93
Pond	10	66.8	2.9	6.06	0.72	1.55
Day 60	55	105.3	2.4	26	1.58	1.39
Cage	9	75.7	3.7	8.78	1.24	0.82
Pond	46	111.1	1.8	29.36	1.42	1.51
Overall	84	–	–	–	–	1.28
Cage	19	–	–	–	–	0.69
Pond	65	–	–	–	–	1.42

of K values over similar size ranges should be valid because weight-length regression slopes were similar for cage and pond cichlids. The regression for caged cichlids was $\log_{10}Wt = -4.682 + 2.978 \log_{10}TL$ (SE(a')=0.0627; SE(b)=0.0364). Pond cichlids standard weight regression was $\log_{10} Wt = -4.674 + 2.911 \log_{10}TL$)SE(a')=0.0306; SE(b)=0.0158).

Whereas maximum densities of cichlids in cages were only about 1 fish per 14 liters, these differences between confinement groups were most likely due to a combination of prey availability and confinement stress. Whereas our cages were small (<121

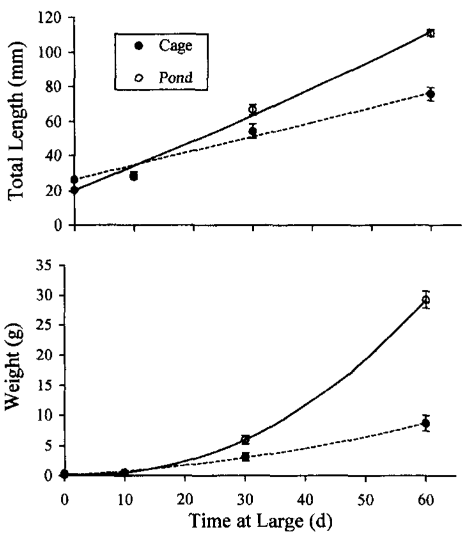

Figure 1. Growth in total length (top) and weight (bottom) of cage and pond-raised cichlids for the 60-day study period. Starting sizes are given at day 0, and mean size of harvested fish is presented for each sample period. Error bars are SE of the mean, and fitted trendlines (polynomial function) are plotted to model observed growth.

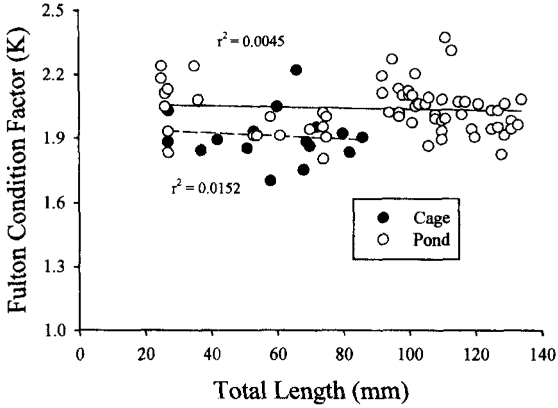

Figure 2. Condition of cage and pond-raised cichlids as measured by a Fulton-type condition factor (K). Trendlines were fitted using linear regression.

liters of available habitat) and small mesh size could have limited prey immigration, it is possible that prey resources were depressed within the cages. The ability of pond cichlids to search for areas of increased prey density within the pond might have given them a predatory advantage, as prey distribution was most likely patchy. In addition, the stress of confinement may have impacted growth and condition of caged cichlids. Many studies have shown that short-term confinement can elicit stress responses in a wide range of fish species (e.g., Carmichael et al. 1984, David and Parker 1986, McDonald et al. 1993, Alford et al. 1994, Harms et al. 1996). In addition, Strange and Cech (1992) found that confinement for as little as 15 minutes reduced swimming speeds (and hence reduced foraging ability) in 2- to 3-year-old striped bass *(Morone saxiatilis)*. Information on the effects of long-term confinement on wild fish are absent in the literature.

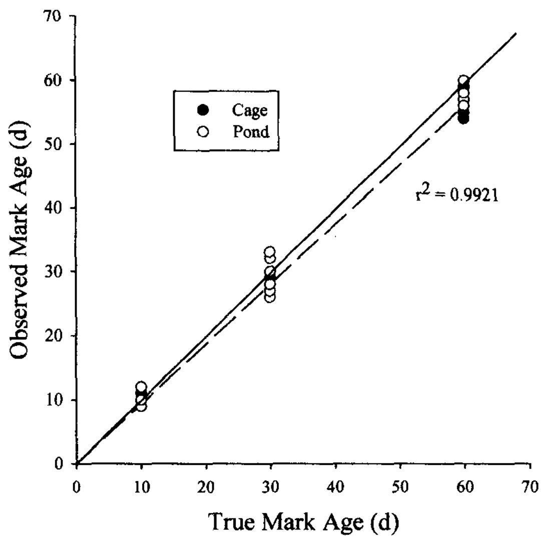

Figure 3. Observed mark age versus true mark age for otoliths of cage and pond cichlids during the 60-day study period. Solid line represents unbiased ageing, and the dashed trendline represents observed accuracy. Trendlines were fitted using linear regression, and are identical (superimposed) for both cage and pond cichlids.

Our cages were small and had inhibited water flow and substantial shade. Only about 35% of the cage walls contained screening to allow water and prey passage, which may have contributed to the observed cage effects. Larger cages with more passable surface area may experience improved performance, and further study is warranted on this issue. Although this study was not designated to test the effects of cage confinement, our data suggest that cage confinement can limit growth when the natural prey base is not supplemented. Hence, we recommend future research focusing on the biases associated with cage confinement.

We found no difference in ageing accuracy between cichlids reared in either cage or pond environments (Fig. 3). Otoliths proved to be an accurate measure of daily age for both groups combined on days 10 and 30, with mean age approximating true age, and low values of SE (0.34 and 0.67, respectively). Daily ring counts slightly underestimated mark age at day 60, giving a mean age interval of 57.4, and SE of 2.86. Enumeration of rings formed before capture and marking indicated an average of 50.9 rings between the AC ring and the otolith nucleus. Hence, these cichlids averaged 50 days old at marking, and 110 days old at the day 60 sample. Whereas other studies have found daily ring accuracy to decrease at about 100 days of age (Sweatman and Kohler 1991, DiCenzo and Bettoli 1995), we suggest a similar limit for ageing this cichlid species.

Conclusions

As with many fish species in temperate regions, daily rings are detectable and appropriate for ageing *Cichlasoma managuense* in the tropics. Observed rings on otoliths were formed on a daily basis, and appeared to be useful for age estimation up to about 100 days of age. We found no differences in daily ring formation between caged cichlids and cichlids at large within the pond, and concluded that cage studies are reliable for daily age validation. We found major differences in growth and condition of cichlids from each confinement situation, and we believe prey availability and confinement stress contributed to depressed performance of caged fish. Hence, we suggest further study of the potential impacts of cage confinement on growth and condition.

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