Invalidation of Otolith Ageing Techniques for Tropical Largemouth Bass

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Abstract: The validity of otolith ageing in Puerto Rico was examined using known-age largemouth bass *(Micropterus salmoides)* in Lucchetti Reservoir. Age-0 largemouth bass were tagged with binary-coded wire microtags and stocked into the reservoir on 6 separate events between April 1992 and May 1996. Fish were collected throughout the study at ages 1–3 and otoliths and microtags were removed from tagged bass. Of 36 age-1 and older tagged bass recovered, 50% had no discernible otolith rings (read as age 0). Observed ages of all otoliths agreed with true ages only 14% of the time, and often differed by 2 years. Thus, we concluded that observed opaque bands were not annuli and this ageing technique is invalid for tropical largemouth bass. Length-at-age keys and length-frequency distributions were developed as alternative techniques for age determination, and these methods proved effective for Lucchetti Reservoir.

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Largemouth bass have been introduced into most of the reservoirs of Puerto Rico and are considered the principal target freshwater fish of the island's sport fisheries (Corujo-Flores 1989, 1990). To successfully manage this predatory sport fish, a strong scientific base of information regarding fish population dynamics and predator-prey relationships is required (Churchill et al. 1995). One component of population dynamics that is extremely important in almost every aspect of fisheries management is determination of age and growth. Accurate age determinations are paramount in assessments of year-class strengths, growth rates, recruitment, and mortality of a fish population.

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Otoliths have been used to determine individual age of many species of fish in different regions of the world (Warburton 1978, Erickson 1983, Summerfelt and Hall 1987, Crawford et al. 1989, Brothers 1990, Fowler 1990). For temperate regions, annual growth checks (annuli) in otoliths have been validated for many species, and are generally related to seasonal changes in water temperature and corresponding changes in the growth rate of the fish (Williams and Bedford 1974, Bagenal and Tesch 1978, Jearld 1983, Schramm 1989). In sub-tropical regions, such as Florida, accurate ageing of centrarchids using otoliths has been accomplished where scales have been shown to lose their validity (Hoyer et al. 1985, Crawford et al. 1989, Schramm 1989). The same is true for temperate fishes such as walleye *(Stizostedion vitreum)*, for which otoliths are preferable to scales and spines for age determination (Erickson 1983).

Ageing of tropical fish using otoliths has not enjoyed the same success as in temperate regions (Pannella 1980, Longhurst and Pauly 1987). This inadequacy is due to a lack of discernible or interpretable patterns on otoliths, possibly resulting from the lack of variation in environmental conditions affecting growth rates (Pannella 1980). However, fish ages obtained from otolith annuli have been verified in some tropical marine fish families, including Lutjanidae, Serranidae, Lethrinidae, and Sciaenidae (Fowler 1990).

Largemouth bass are not native to the tropics or marine environments, but they have been widely introduced into freshwater reservoirs in tropical regions. For age and growth studies of largemouth bass in temperate and sub-tropical regions, annuli from whole and sectioned otoliths have been verified (Miller and Storck 1982, Taubert and Tranquilli 1982, Hoyer et al. 1985, Crawford et al. 1989). Yet, there is no evidence in the literature of validation studies of ageing techniques for tropical largemouth bass. As sport fish management increases in Puerto Rico, so does the use of otoliths as an indicator of fish age without proper validation. This study was undertaken to assess the utility of otoliths for age and growth analyses of largemouth bass populations in a Puerto Rican reservoir.

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Methods

Lucchetti Reservoir is a 108-ha impoundment in the mountain region of southwestern Puerto Rico. The area receives an average of 198 cm rainfall annually and was originally tropical forest, although much of the landscape is now used for agriculture. Lucchetti Reservoir has been categorized from mesotrophic to eutrophic on the basis of nutrients, physical limnology, chlorophyll *a*, and phytoplankton biomass data (Cham and Carvajal-Zamora 1981, Tilly 1983, Puerto Rico Environ. Quality Board 1992, Perez-Santos 1994). The primary function of the reservoir is water storage for irrigation, but the creation of the Lucchetti Field Station and associated facilities has improved recreational access and increased reservoir popularity among boating anglers (Churchill et al. 1995).

Supplemental stocking of juvenile largemouth bass into Lucchetti Reservoir was conducted 6 times from April 1992 to November 1995. Largemouth bass fingerlings were provided by Puerto Rico DNER at the Maricao Fish Hatchery. Individuals from each cohort were batch-tagged with cohort-specific, binary-coded wire microtags at the hatchery and transported to the reservoir for stocking. Fish were collected periodically from electrofishing and gillnetting samples and from Lucchetti Field Station creel studies. All microtagged fish were measured (total length mm), weighed (g), and sacrificed for saggital otolith and microtag removal.

Various methods of otolith examination were attempted, and best resolution was achieved by sectioning and polishing the otoliths. A jeweler's saw was used to obtain a transverse section that included the nucleus. Otolith sections were coated with immersion oil to enhance clarity between the translucent (hyaline) and opaque bands and read on a dissection microscope. Magnification was varied 12X–60X and reflected light was used. Otoliths were examined independently by 2 readers without reference to fish size; disagreements among age estimates were resolved by mutual examination and discussion of otolith features. Opaque bands were considered to be annuli. There was no evidence available to distinguish any rings as false annuli, so all rings were counted for age estimation. This method provided the observed age of each bass and was compared to the true age given by microtag identification. Average ageing error was determined from the absolute values of the differences between true and observed ages.

In addition, the utility of other methods for age determination was evaluated as an alternative in the event that otoliths proved unreliable. A length-at-age key was created using the same known-age bass (including age-0 bass) collected throughout the study. Initial sizes at day 1 were the sizes at introduction for all 6 cohorts. Also, qualitative length-frequency analysis was performed on largemouth bass data collected during a mark-recapture study in January–February 1997. Ages were assigned to individually distinguishable length-modes.

Results

We collected 36 adult largemouth bass (≥age 1) from April 1994 to February 1997 that contained the binary-coded wire microtags. Only 18 (50%) of the sectioned otoliths had discernible rings, many of which were faint and barely recognizable. Otoliths that did not exhibit rings were similar in size to the otoliths with growth rings, and there was no visually obvious relationship between ageing error and fish length.

Since all bass were ≥age 1, otoliths containing no discernible rings contributed significantly to ageing error, producing a negative bias (Fig. 1). Observed age agreed with true age in only 5 of the 18 bass otoliths that displayed growth checks (28%).

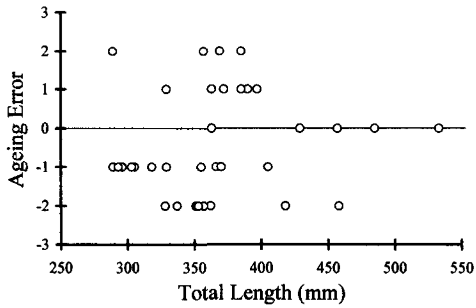

Figure 1. Difference between observed age (in years) obtained from otoliths and true age (in years) obtained from microtags (ageing error) for 36 largemouth bass, Lucchetti Reservoir, Puerto Rico, 1994–1997.

For the entire sample, only 14% (5 of 36) of the otoliths exhibited opaque bands that correctly identified largemouth bass age. Although true age only spanned 3 years, observed ages from otoliths often missed the true age by 2 years, and average ageing error was 1.25 years.

Discussion

The lack of accuracy displayed by our results demonstrates that the growth checks observed were not annuli. By definition, an annulus is any zone, mark, or pattern of growth that forms once a year (Ricker 1968). Opaque bands are associated with periods of slow growth and translucent bands are associated with periods of fast growth (Schramm 1989) regardless of whether these periods occur annually or more or less frequently.

Although water temperature in Lucchetti Reservoir followed the same general cyclic pattern seen in temperate reservoirs, the seasonal variation never exceeded 6 C during the study period (Churchill et al. 1995). Fluctuations of 5 C were shown to be sufficient to result in opaque zone formation in lake trout *(Salvelinus namaycush)*, but the temperature changes were much more rapid (Brothers 1990). In bluegill *(Lepomis macrochirus)*, opaque bands form in response to changes in water temperature, but relatively rapid temperature changes induce their formation more effectively (Schramm 1989). The bluegill study also revealed that only translucent bands formed when water temperature exceeded 22 C. In Lucchetti Reservoir, the lowest recorded temperature during the study period was 22.3 C at the deep, anoxic bottom (Churchill et al. 1995); warmer temperatures prevailed in available bass habitat. If band formation is similar in largemouth bass to that in bluegills, no opaque bands would result from temperature fluctuation in Lucchetti Reservoir.

Wet and dry seasons have been shown to affect growth ring formation in tropical fishes (Warburton 1978), and Lucchetti Reservoir experienced extreme annual fluctuations in water level throughout the study (Churchill et al. 1995). Although these fluctuations occurred on an annual cycle, our results demonstrate that they did not affect growth rates sufficiently to produce identifiable annuli. Whereas 50% of our sample otoliths contained no discernible rings, there was no environmental factor sufficient to produce populational growth checks. Therefore, opaque bands observed on individual otoliths must have been the result of conditions affecting largemouth bass individually. For example, Gran (1995) found that largemouth bass in Lucchetti Reservoir demonstrated an extended spawning season with multiple spawns per individual. Other conditions could have included behavioral changes or differential predatory success with a changing forage base as described by Alicea (1995).

Regardless of the reasons for opaque band formation, this research has shown that otoliths are not a reliable structure for largemouth bass ageing in Puerto Rico. Thus, other techniques must be considered for determining largemouth bass age. In tropical systems such as Lucchetti Reservoir, bass growth can be extremely rapid due to the continuous growing season, and longevity tends to be reduced (Churchill et al. 1995). For example, Lilyestrom et al. (1994) found that <1% of telemetered largemouth bass reached age 3 in La Plata Reservoir, Puerto Rico. Limited age classes combined with rapid growth allows fish ageing to be accomplished using more traditional methods. For instance, length-at-age keys (Fig. 2) from known-age fish can be used to estimate age from fish length. This technique requires large numbers of tagged fish of different ages to be identified, however, and is only useful within a single system during a limited time frame since growth rates may change. A more practical approach given the rapid growth and truncated age structure would be the use of length-frequency analyses (Fig. 3). This method can be used to separate age-groups visually or statistically (DeVries and Frie 1996).

The opaque bands we found in transverse thin sections of largemouth bass otoliths were not annuli, and therefore were not reliable indicators of fish age. We

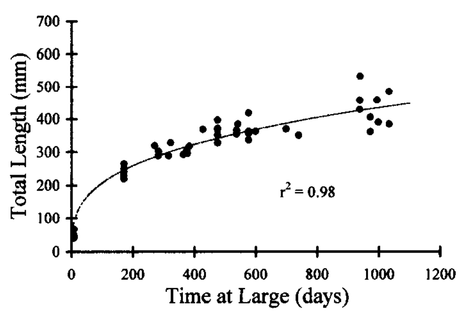

Figure 2. Length-at-age for all (including age-0 bass) stocked largemouth bass collected in Lucchetti Reservoir, Puerto Rico. Mean total length at stocking for each cohort (Apr 1992–May 1996) is given at day 1. Fitted trendline (power function) is plotted to model observed growth.

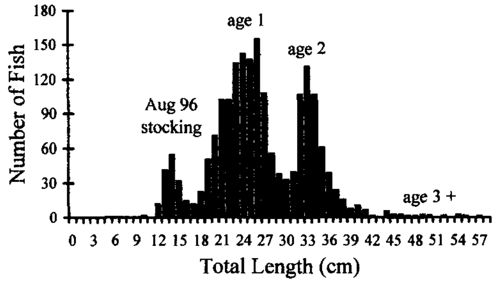

Figure 3. Length-frequency distribution of largemouth bass in Lucchetti Reservoir, Puerto Rico, January–February 1997. The first small mode is from the stocking in August 1996, and ages are assigned to length groups beginning at age 1.

suggest that future studies on age and growth of largemouth bass in Puerto Rico utilize length-frequency analyses for age determination. Also, we would caution researchers working in other tropical systems to perform similar validation studies before attempting to use otoliths in studies of age and growth.

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