Age and Growth of Roundtail Chub in the Upper Verde River, Arizona

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Abstract.-Estimates of the age and growth of roundtail chub Gila robusta were made by analyzing thin cross sections of sagittal otoliths from 280 specimens collected in the upper Verde River, Arizona, from January 1997 to December 1999. Marginal increment analysis indicated that a single annulus completed formation between January and May. The use of otoliths to age roundtail chub was further validated using otoliths of knownage, hatchery-reared fish for which (upon examination) the number of annuli agreed 100% of the time with the true age of the fish. In addition, daily increments were observed on sagittal otoliths of age-0 roundtail chub and, when counted, agreed with the true age of fish 70% of the time; daily ring counts were within 1 and 2 d of the true age 80% and 90% of the time, respectively. Ages assigned to wild-caught fish by three independent readers resulted in a coefficient of variation (100 × SD/ mean) of 8.2. Ages of roundtail chub varied from 1 to 7, the largest fish being a 427-mm total length (TL), 836-g total weight (TW), age-7 female. The largest male was 413 mm TL, 629 g TW, and age 6. The growth of age-1-7 roundtail chub (both sexes combined) was greatest during the first year and was best described by the equation $L_t = 499.30 \left[1 - e^{-0.23(t-0.147)}\right]$, where L_t is length at time t.

The roundtail chub *Gila robusta* is one of a diverse group of relatively large-bodied cyprinids that was once widely distributed throughout the Colorado River basin of the western United States and northwestern Mexico. Although still locally common in places throughout its historic range, the roundtail chub, like many native fishes in the arid western United States, has been reduced in numbers and distribution due to altered flow regimes (Miller 1961; Minckley and Deacon 1968; Minckley and Douglas 1991), predation and competition by introduced exotic species (Meffe 1985; Miller et al. 1989; Douglas et al. 1994), and inadvertent introductions of parasites (Hoffman and Shubert 1984; Brouder and Hoffnagle 1997). Con-

Received June 7, 2004; accepted December 27, 2004 Published online June 13, 2005 sequently, roundtail chub is on state-sensitive species lists in Colorado, Utah, Arizona, and New Mexico, and is currently being reviewed for listing as a federally threatened or endangered species.

The majority of published literature on this species pertains to status, distribution, and abundance (Vanicek et al. 1970; Minckley 1973; Holden and Stalnaker 1975; Tyus et al. 1982; Bestgen and Propst 1989), taxonomy (Holden and Stalnaker 1970; Douglas et al. 1989; McElroy and Douglas 1995; Douglas et al. 1998), recruitment (Brouder 2001), culture (Muth et al. 1985), parasites and their effects (Mpoame and Rinne 1983; Brouder 1999), and other life history aspects (Vanicek and Kramer 1969; Schreiber and Minckley 1981; Bestgen et al. 1987; Kaeding et al. 1990, Karp and Tyus 1990; Barrett and Maughan 1995). However, very little detailed information exists on the age and growth of roundtail chub, especially in the lower Colorado River basin. The primary objectives of this study were to describe the age, growth, and size structure of the roundtail chub population in the upper Verde River and to validate annulus formation in the otoliths of roundtail chub.

Methods

Fish collections.—To determine age structure and growth of roundtail chub, fish were collected quarterly from February 1998 through December 1999 at five sites located within the upper 72 km of Verde River, Arizona, primarily by using a Smith-Root backpack shocker but with other techniques (including seining, hoop nets, trammel nets, and angling) as well. After capture, total length (TL, mm) and total weight (TW, g) were recorded, and fish were sacrificed for the removal of otoliths and determination of sex (based on presence of testes or ovaries) in the laboratory.

Otolith preparation.—Cross sections of roundtail chub otoliths were prepared by mounting whole otoliths on microscope slides using CrystalBond thermoplastic cement and grinding frontally through the nucleus using a combination of 600–2,000-grit wet–dry sandpaper. Sectioned otoliths were then viewed using an image analysis

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system (EPIX, Inc.; PIXCI software) consisting of a personal computer, video monitor, and camera equipment connected to a compound microscope. Otoliths were viewed under oil immersion at 40×10^{-5} magnification using transmitted light. Measurements were taken from the nucleus to the outer edge of each opaque band and from the nucleus to the margin or otolith radius.

Age and growth.—Three independent readers counted the number of annuli on each cross section and assigned ages accordingly. When discrepancies occurred, all readers reread the otolith and if the readings still disagreed, the otolith was discarded. Precision in age estimates among readers was determined by calculating a coefficient of variation ($CV = 100 \times SD/mean$; Chang 1982).

Length of roundtail chub at previous age was back-calculated from otolith measurements using the modified Fraser-Lee method and a biologically relevant intercept (Campana 1990) because the size of fish and otolith at swim-up were available from hatchery-reared fish used in the validation component of this study. The biological intercept method is based on the assumption of a constant linear relationship between fish and otolith length within an individual. Deviations from this assumption using the biological intercept, which uses a biologically determined intercept value, are less problematic in the calculation of annual growth than the traditional Fraser-Lee method, which uses a statistically estimated intercept value (Campana and Jones 1992). Back-calculated length at age was determined using the following equation:

$$L_a = L_c + (O_a - O_c)(L_c - L_o)/(O_c - O_o),$$

where L_a is the back-calculated length to annulus a, L_c is the length of fish at capture, L_o is the length of fish at swim-up (8.5 mm), O_a is the otolith radius at annulus a, O_c is the otolith radius at capture, and O_o is the otolith radius at swim-up (0.1243 mm). Because t-tests did not indicate significant differences between male and female lengths at age, data for both sexes were combined to back-calculate growth.

Growth rate of roundtail chub was determined by fitting the von Bertalanffy growth model to back-calculated length-at-age data. The model is

$$L_t = L_{\infty}[1 - e^{-K(t-t_0)}],$$

where L_t is the total length at time t (years), L_{∞} is the maximum theoretical attainable total length (mm), K is the growth coefficient, and t_0 is the

time (years) when length theoretically would be zero (von Bertalanffy 1957). Model parameters were estimated using the computer program FISH-PARM (Prager et al. 1989).

Otolith validation.—To validate opaque bands as annuli and to determine time of annulus formation. otoliths from roundtail chub collected monthly from January through December 1997 were examined, and mean marginal increment data (distance from the last annulus to the edge of the otolith) were plotted against month of capture. To further validate the use of otoliths to age roundtail chub, adult fish were collected from the upper Verde River in March 1997 and transported to Arizona Game and Fish Department's Bubbling Ponds Fish Hatchery in Cornville, Arizona. Broodfish were artificially spawned, progeny were raised in the laboratory, and fry were reared outdoors in an earthen pond. Approximately 50 roundtail chub were collected annually in late June to early July for 3 years using a fyke net, measured for TL (millimeters), and sacrificed. Otoliths from knownaged fish were removed and aged using the same procedures as wild-caught fish. In addition to validating annul marks on sagittal otoliths of knownage roundtail chub, daily increments on otoliths of age-0 roundtail chub were validated by comparing the age (in days) with the number of rings on their otoliths.

Results

Age and Growth

A total of 312 roundtail chub, ranging in length from 50 to 415 mm TL, were collected from the upper Verde River (of which 280 had otoliths that were legible), aged with a 96% first-time reader agreement, and measured. Among-reader precision (CV) for wild-caught roundtail chub otoliths was 8.2. Ages of roundtail chub ranged from 1 to 7 years, the oldest fish from the upper Verde River population having completed seven growing seasons. Two distinct cohorts of roundtail chub were sampled in both 1998 and 1999. The majority (44%) of fish collected in 1998 were age 5 (1993 year-class) followed by age-3 fish (36%; 1995 year-class). Similarly, roundtail chub from the 1993 and 1995 year-classes (age 6 and age 4, respectively) comprised the two highest percentages of fish collected in 1999 at 42% and 28%, respectively. The longest and heaviest fish collected was a 427-mm TL, 836-g, age-7 female; the largest male (age 6) was 413 mm TL and 629 g.

Mean length at age did not significantly differ

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TABLE 1.—Mean back-calculated total length (TL) of roundtail chub at the end of each growing season in the upper Verde River, Arizona, March 1997—December 1999.

Age (year-class)	Number of fish	Annulus						
		1	2	3	4	5	6	7
1 (1996)	79	104						
2 (1995)	70	103	184					
3 (1994)	58	100	170	220				
4 (1993)	47	102	180	245	285			
5 (1992)	16	107	186	257	298	330		
6 (1991)	8	93	184	248	302	345	382	
7 (1990)	2	103	187	246	303	345	380	412
Mean TL		101	181	243	297	340	381	412
Increment		101	80	62	54	43	41	32

between sexes (P > 0.05); however, analysis of covariance (ANCOVA) indicated a significant difference (F = 674.5; df = 311; P < 0.0001) in sexspecific length—weight relations of roundtail chub, females being heavier per unit of length than males. The length—weight relationship for males and females of the upper Verde River roundtail chub population is best described by the following equations:



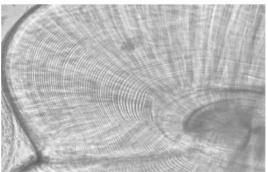


FIGURE 1.—Photograph of cross section of roundtail chub sagittal otoliths depicting annual (top) and daily (bottom) increments. The line on the top figure denotes the plane of measurement from the focus to each annulus and edge. The arrows show where the annuli were measured (outer edge of the thin opaque zone).

$$\log_{10}\text{TW} = 3.140 \log_{10}\text{TL} - 5.411$$

 $(N = 151; r^2 = 0.95)$ and $\log_{10}\text{TW} = 3.142 \log_{10}\text{TL} - 5.345$
 $(N = 56; r^2 = 0.96).$

Growth of the upper Verde River roundtail chub population (both sexes combined) was greatest during the first year of life (101 mm) and declined steadily afterward from 80 mm at age 2 to 32 mm at age 7 (Table 1). Growth of roundtail chub in the upper Verde River is best described by

$$L_t = 499.30[1 - e^{-0.23(t-0.147)}].$$

Otolith Validation

Otoliths exhibited alternating wide translucent and narrow opaque zones (Figure 1), and marginal increment analysis indicated that a single opaque band was formed once a year (Figure 2). Mean marginal increments were significantly lower (P = 0.015) in May than in any other month, further

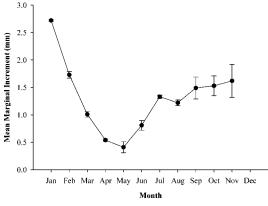


FIGURE 2.—Mean marginal increment distances by month (\pm SE) for all ages of roundtail chub collected in the Verde River. Arizona.

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suggesting that opaque bands are formed once per vear.

Otoliths were taken from 110 roundtail chub, ranging from 58 to 236 mm TL, from Bubbling Ponds Hatchery over a 3-year period to further validate annulus formation. From the examination of 99 otoliths (11 were inadvertently destroyed in preparation) from hatchery-reared, known-age fish, the number of opaque bands (annuli) agreed 100% of the time with the actual age of the fish. Daily rings were also observed on the sagittal otoliths of age-0 hatchery-reared fish (Figure 1), and ages based on daily ring counts of age-0 fish agreed 70% of the time with true age, and 80% and 90% of the time within 1 and 2 d of the true age, respectively.

Discussion

The age structure of the upper Verde River roundtail chub population was dominated by two distinct cohorts. Fish from both the 1993 and 1995 year-classes comprised the majority of the catch in both 1998 and 1999. In the late winter through early spring of both 1993 and 1995 floods had occurred, and Brouder (2001) found a significant relationship between flooding and the recruitment of roundtail chub. The presence of two strong year-classes in years when flooding occurred further supports this hypothesis.

The maximum observed age (age 7) of roundtail chub from the upper Verde River population was similar to that reported for roundtail chub in the Gila and Green rivers (Bestgen 1985; Vanicek and Kramer 1969). However, mean lengths at age of roundtail chub in this study were larger than those reported for roundtail chub in two streams in the Gila River basin, New Mexico (Bestgen 1985), Fossil Creek, Arizona (Neve 1976), and the Green River, Utah (Vanicek and Kramer 1969). In addition, growth rates were more rapid than those reported by other researchers but agreed with the observation that growth is greatest during the first year. Fast growth rates of roundtail chub from the upper Verde River during this study may be explained by the various impacts of the 1993 and 1995 floods. In the Southwest, flooding benefits native fishes by disproportionately displacing nonnative fishes (Meffe 1984; Minckley and Meffe 1987; Rinne and Stefferud 1997) that prey upon and compete for resources with native fishes. Displacement of nonnative fishes likely alters resource utilization patterns of native fishes, which may ultimately affect or, as in this case, accelerate individual growth rates (Werner and Hall 1977).

The apparent discrepancies in the length at age and growth of roundtail chub in this study and those reported elsewhere may also be attributed to the use of scales in previous studies and otoliths in this study. Both Bestgen (1985) and Neve (1976) concluded that due to the difficulty in discerning distinct annuli and therefore ages, the use of scales may not be an accurate method for aging roundtail chub and, therefore, their length-at-age data should be viewed as tentative. Furthermore, the difficulty in discerning distinct annuli in roundtail chub scales that both Bestgen (1985) and Neve (1976) experienced may have introduced a bias toward overestimating the ages of roundtail chub, which would result in, and explain, the smaller lengths at age of roundtail chub in their studies than that of the fish in this study.

Frontally sectioned sagittal otoliths of roundtail chub contain easily discernible annual marks. The high degree of precision among three independent readers of wild roundtail chub confirms the relative ease in counting annuli in sectioned otoliths. Marginal increment analysis suggests that a single annul mark is deposited in a period extending from January to May. This period of opaque band formation seems reasonable because factors affecting the deposition of opaque bands (i.e., reduced winter growth and spawning) occur during this same time frame. In addition, the number of opaque bands on known-age, hatchery-raised roundtail chub was in exact agreement (100%) with the actual age of fish, further validating the use of sagittal otoliths to age roundtail chub. Because the roundtail chub used to validate the use of otoliths in this study were, at most, 3-years-old, it is possible that true annuli may not be deposited on the otoliths of older fish, thus possibly making the aging of older roundtail chub more difficult (Campana 2001). However, because the relative age of roundtail chub in this study was age 7 or younger, this potential problem is likely minimal. Of special note was the observation of daily increments on the sagittal otoliths of age-0 roundtail chub. Daily rings on otoliths can be used to gather important information on hatching times, growth, and mortality of fishes (Miller and Storck 1982; Essig and Cole 1986; Isley and Noble 1987).

Otolith cross sections are a viable technique for determining ages of roundtail chub in the upper Verde River, Arizona, and should be throughout the species' range. The use of otoliths gives biologists a reliable means of determining the age of roundtail chub, which allows for a better understanding of age structure, growth rates, recruit-

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ment, and mortality. Unfortunately, the need to sacrifice fish to extract otoliths is an obvious drawback of this method, especially when working with a species of special concern like roundtail chub.

Acknowledgments

I would like to thank Jody Walters for his valuable input into study design, Mike Childs for spawning and rearing fish, and Diana Rogers and Lorraine Avenetti for their valuable field and laboratory assistance. I would also like to thank Scott Bonar, Scott Bryan, Bill Persons, and three anonymous reviewers for providing suggestions that improved the manuscript. Funding was provided by the Sportfish Restoration fund administered through the Arizona Game and Fish Department (Project F-4-R). Reference to trade names does not imply endorsement by the U.S. Government.

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