

## Comparison of aging structures and life history of a historical population of Roundtail Chub (Cyprinidae) in the Yampa River, Colorado

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**ABSTRACT.**—The *Gila robusta* complex includes 3 large cyprinid species native to the Colorado River drainage. All 3 members of this complex, Humpback Chub *G. cypha*, Bonytail Chub *G. elegans*, and Roundtail Chub *G. robusta*, historically occurred in the Yampa River Canyon of Colorado and Utah. Both Humpback Chub and Bonytail Chub are federally listed as endangered, and the Roundtail Chub is now considered either imperiled or vulnerable to extinction by all state management agencies in the Colorado River basin. In late June or early July of 1998, 1999, 2001, and 2002, we collected Roundtail Chub from the Yampa River in Dinosaur National Monument. Our objectives were to determine the efficacy of scales, opercular bones, and otoliths as aging structures, and to describe age, mortality, growth, and tuberculation patterns relative to reproductive stages. We obtained interpretable annuli for 111 fish by using otoliths, 91 fish by using opercles, and 111 fish by using scales. Otolith annuli showed the least measurement error; scale annuli generally agreed with otolith annuli up to approximately 7–8 years, after which scale annuli consistently underestimated otolith annuli. Opercular annuli closely agreed with otolith annuli up to 10–12 years before underestimating otolith annuli. Based on otoliths, the oldest fish was aged at 22 years, with 7 fish exceeding 15 years. Using a linearized catch-curve, Roundtail Chub had an instantaneous mortality rate of  $-0.158$ , corresponding to 85% survivorship. Growth in length was initially high and then declined at older ages. Slopes of mass versus length differed between sexes, and females had a slightly greater body mass per unit length than males for lengths above 398 mm TL. Both sexes have similar ages at first reproduction (5–6 years), and both develop tubercles. The greatest tubercle development occurred in males with maturing or running ripe testes, and in females with maturing ovaries.

**RESUMEN.**—El complejo *Gila robusta* incluye tres grandes especies de ciprínidos nativos de la cuenca del río Colorado. Los tres miembros de este complejo: *G. cypha*, *G. elegans* y *G. robusta*, históricamente se encontraban en el Cañón del Río Yampa en Colorado y Utah. Tanto *G. cypha* como *G. elegans*, están catalogados a nivel federal como en peligro de extinción, y todas las agencias estatales de gestión en la cuenca del río Colorado ahora consideran a *G. robusta* como una especie vulnerable o en peligro de extinción. A finales de junio y principios de julio de 1998, 1999, 2001 y 2002, recolectamos peces *G. robusta* del río Yampa en el Monumento Nacional Dinosaurio. Nuestro objetivo fue determinar la eficacia de las escamas, los huesos operculares y los otolitos como estructuras de envejecimiento, y describir los patrones de edad, mortalidad, crecimiento y patrones de tuberculación en relación con las etapas reproductivas. Obtuvimos anillos interpretables de: 111 peces usando otolitos, 91 peces usando opérculos y 111 peces usando escamas. Los anillos en los otolitos mostraron el menor error de medición. Los anillos de escamas generalmente coincidieron con los anillos de otolitos hasta aproximadamente siete a ocho años, después de lo cual los anillos de escamas subestimaban consistentemente los anillos de otolitos. Los anillos operculares correspondieron estrechamente con los anillos de los otolitos hasta 10 a 12 años antes de subestimar los anillos de los otolitos. Según los otolitos, el pez de mayor edad tenía 22 años, y 7 peces superaban los 15 años. Utilizando una curva de captura linealizada, *G. robusta* tuvo una tasa de mortalidad instantánea de  $-0.158$ , correspondiente a un 85% de supervivencia. El crecimiento en longitud fue alto al principio y luego disminuyó durante las edades más avanzadas. Las pendientes de masa en comparación con las de longitud difirieron entre sexos, las hembras tuvieron una masa corporal ligeramente mayor por unidad de longitud que los machos para longitudes superiores a 398 mm LT. Ambos sexos tuvieron edades similares a la primera reproducción (cinco a seis años) y ambos desarrollan tubérculos. El mayor desarrollo de tubérculos ocurrió en machos con testículos maduros o en crecimiento, y en hembras con ovarios maduros.

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The Roundtail Chub *Gila robusta* is endemic to the Colorado River basin and historically occurred in lotic systems ranging from moderate-sized tributaries to mainstem rivers (Minckley 1973, as *G. r. robusta*). In the upper Colorado River (drainages upstream from Lake Powell), Roundtail Chub formerly were sympatric with Bonytail Chub *G. elegans* and Humpback Chub *G. cypha*. Research on mtDNA determined that prior to human impacts, Roundtail Chub hybridized with Bonytail Chub, resulting in the replacement of Roundtail Chub mitochondrial DNA with Humpback Chub mtDNA. However, mtDNA is not coupled with morphological variation (Dowling and DeMarais 1993, Gerber et al. 2001).

In the lower Colorado Basin in New Mexico, Utah, and Arizona, Roundtail Chub were generally allopatric with Bonytail and Humpback chubs and do not show evidence of hybridization. However, in the lower Colorado system, the Roundtail Chub complex was considered to include 3 forms, *G. robusta*, *G. intermedia*, and *G. nigra*, with the latter two restricted to the Gila River drainage (Rinne 1976, Minckley and DeMarais 2000, Page et al. 2017). Recently, Carter et al. (2018) and Copus et al. (2018) documented widespread overlap in the morphological and genetic characters used to separate the Gila River forms, and also showed that the 3 forms were often sympatric, leading to their recommendation to consider *G. robusta* a single polymorphic species.

Although the 2 closely related congeners, Humpback and Bonytail chubs, have been listed as Endangered by the Endangered Species Act (ESA) since 1967 and 1980, respectively (USFWS 1994), the Roundtail Chub remained relatively abundant in the Yampa River within Dinosaur National Monument, at least through 2002 (Ross and Modde personal observation). Since then, especially because of predation impacts from the nonnative Smallmouth Bass *Micropterus dolomieu* and Northern Pike *Esox lucius*, the Roundtail Chub has severely declined in the upper Colorado River basin and is listed as endangered, as a species of concern, or as a species of special concern by Arizona, Colorado, New Mexico, Utah, and Wyoming (Propst 1999, Anderson and Stewart 2003, Utah DNR 2006, Bestgen et al. 2007, Johnson et al. 2008). In addition, the Gila

Chub *G. intermedia* (now considered a synonym of Roundtail Chub) has been listed as endangered under the ESA (USFWS 2005), and both Roundtail Chub and Headwater Chub *G. nigra* (now considered a synonym of Roundtail Chub) have been proposed for listing as Threatened under the ESA (USFWS 2015). This proposal has been withdrawn by the USFWS pending a species status assessment of the newly defined Roundtail Chub (Copus et al. 2018).

Despite its sensitive status, relatively little has been published on reproductive development, age, or growth of the Roundtail Chub. Although the Blue Chub *G. coerulea* has a reported maximum age of 17 years (Scopettone 1988) and the Bonytail Chub has a maximum age of 13 years (Marsh 1997), the maximum reported longevity of the Roundtail Chub, based on a combination of length frequency (Neve 1976), scale (Vanicek and Kramer 1969), and otolith (Brouder 2005) analyses, is between 5 and 7 years. However, Bestgen (1985) suggested that life span may be influenced by the size of available habitat, so that larger rivers would support older fish. In smaller tributaries of the Gila River system, such as Fossil Creek (Neve 1976) and Turkey Creek (Bestgen 1985), Roundtail Chub produced gametes at as early as 2 years old, whereas in the East Fork of the Gila River, Roundtail Chub did not mature until they were 4–7 years old (Bestgen 1985). Similarly, in the larger Verde River, Brouder et al. (2000) observed mature female chub to range between ages 4 and 7.

The majority of published studies on the life history of Roundtail Chub describe populations in the lower Colorado River basin. The scale analysis of Vanicek and Kramer (1969) is the only age and growth analysis of Roundtail Chub from the upper Colorado River basin. Given the decline of the once-abundant populations of Roundtail Chub in the mainstem Colorado and Yampa rivers, an understanding of basic life history aspects of this population is important for management decisions impacting its recovery.

Herein we compare the efficacy of 3 structures used in aging: scales, opercular bones, and otoliths. We then describe age, mortality, growth, and tuberculation patterns relative to reproductive stages of Roundtail Chub in the Yampa River, the last relatively free-flowing

TABLE 1. Stages of gonadal development based on Heins and Machado (1993) and Albanese (2000).

Gonad type	Code	Stage of development
Ovaries	LA	<i>Latent</i> : small, thin, transparent to slightly translucent
	EM	<i>Early maturing</i> : small to moderate, translucent to white
	LM	<i>Late maturing</i> : becoming enlarged, white to cream
	MA	<i>Mature</i> : moderate to greatly enlarged, mature oocytes opaque
	MR	<i>Ripening</i> : moderate to greatly enlarged, mature oocytes translucent
	RE	<i>Ripe</i> : ripe ova concentrated in lumen, chorion separated from yolk
Testes	LA	<i>Latent</i> : very small, translucent
	MA1	<i>Maturing</i> : small, cloudy white but not opaque
	MA2	<i>Mature</i> : medium to large, white
	RE	<i>Ripe</i> : large, white, discharging sperm

tributary in the upper Colorado River drainage. Data were collected between 1998 and 2002, preceding by several years the more recent decline of Roundtail Chub in the Yampa River as discussed above.

## METHODS

### Fieldwork

All fieldwork was conducted as part of ongoing native fish surveys and nonnative fish removal efforts by the USFWS. Fish were captured by angling (1998) and electrofishing (1999, 2001, 2002) from the Yampa River in Dinosaur National Monument from Deer Lodge (latitude 40.446527, longitude -108.512046) to the confluence of the Yampa River with the Green River at Echo Park (latitude 40.5205195, longitude -108.990379). Dates of collections were 9–12 July 1998, 12–14 July 1999, 25–28 June 2001, and 10–12 June 2002. Water temperatures ranged from 20.5 to 22.5 °C in 1998, 21 to 23 °C in 1999, 19 to 23 °C in 2001, and 18 °C in 2002.

Roundtail Chub were retained in live wells and then euthanized in an overdose of MS-222 before processing in the field for ageing structures, pattern and degree of tuberculation, sex determination, and gonadal staging. Because of limited space in the rafts and issues of transporting formalin, fish carcasses were not retained once they were processed.

We used scales, opercular bones, and otoliths to age Roundtail Chub. We removed approximately 5–10 scales from the left side of the body dorsal to the pectoral fin and ventral to the lateral line and placed them in paper scale envelopes. Scales were taken in 1998, 1999, and 2001. The left opercular bones were removed (occasionally the right operculum was used if the left one was damaged) and

stored dry in envelopes. Opercula were taken in all 4 years of fieldwork. We removed otoliths in the field and stored them dry in plastic vials until processing. As is true of otophysans in general (Popper and Coombs 1982), the inner ear of Roundtail Chub contains 2 pairs of otoliths of approximately equal size, the more posterior lagenar otoliths and the saccular otoliths, as well as the small utricular otoliths (lapilli). The lagenar otolith (asteriscus) is highly sculptured with a broadly serrated edge and, although slightly larger in diameter than the saccular otolith, is about half as thick. Hence, we used the saccular otolith (sagitta) for age determination. Otoliths were taken in all 4 years of the study.

Gonads were staged following Heins and Machado (1993), as modified for cyprinids by Albanese (2000) (Table 1). Our late June/early July sampling may have preceded actual spawning dates, given that we did not collect females with ovaries in ripening or ripe stages, although Kaeding et al. (1990) found female Roundtail Chub in the upper Colorado River with “expressible ova” from mid-June to late July.

Tubercles were difficult to discern in photographs taken in the field, so tuberculation patterns were recorded by sketching them on a standardized drawing of the head and dorsum of Roundtail Chub (Fig. 1). If tubercles were present, the degree of tuberculation was divided into 4 stages: (1) tubercles restricted to head and pectoral fin rays; (2) tubercles covering head, the anterior area of the nape, and pectoral fin rays; (3) tubercles covering head, nape, dorsum, and pectoral rays; and (4) tubercles densely covering head, nape, dorsum, and pectoral and pelvic rays. Because of limited sample sizes, stages 3 and 4 were combined.

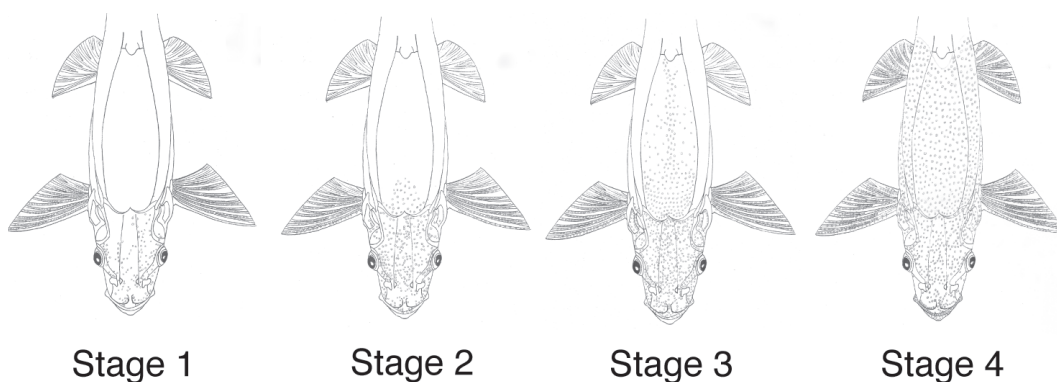


Fig. 1. Degrees of tuberculation in the Roundtail Chub *Gila robusta* (drawings by D. Ross).

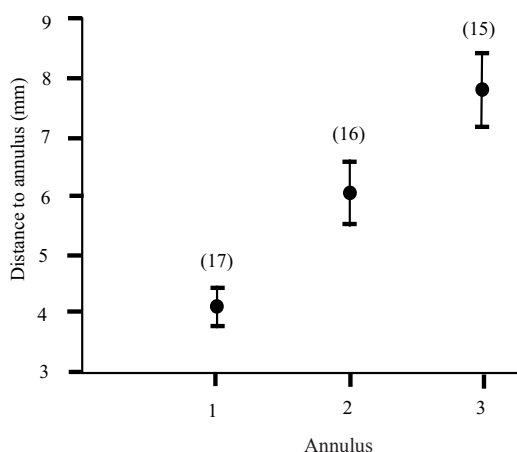


Fig. 2. Distance to the first, second, and third annuli in opercular bones of the Roundtail Chub *Gila robusta*. Bars show 95% confidence intervals; sample sizes are in parentheses.

### Laboratory Methods

We mounted scales between 2 microscope slides and initially examined them at 40 $\times$  magnification with transmitted light. One scale showing the clearest growth checks was selected for further processing using image analysis. Images were captured using a Nikon digital camera (Nikon Coolpix 4500) mounted on a dissecting microscope at 4 $\times$  magnification. Image enhancement (rarely needed) and measurement of scales were done using ImageJ for Mac OS X (U.S. National Institutes of Health, 1997–2018, Bethesda, MD, USA; <https://imagej.nih.gov/ij>). Scale images were imported into Adobe Illustrator CS3 (version 13.0.0) or equivalent program for reading and

were enlarged as needed using the zoom function. Presumed annular marks in scales were indicated by the cutting over of circuli, as described by Quist et al. (2012).

Prior to analysis, opercula were soaked in water containing a small amount of detergent, and then they were gently scrubbed with a toothbrush and paper towels to remove the overlying skin. Opercula were viewed at 6.3–10 $\times$  with reflected light over a dark background, and images of medial and lateral aspects (except for 2001 samples that were not digitized) were captured at magnifications of 0.63–1.0 $\times$  and processed as described above for scales. Following Le Cren (1947), Bardach (1955), Casselman (1974), and Scoppettone (1988), we located presumed annular marks in opercular bones where a narrow, translucent slow-growth zone (appearing dark with reflected light) adjoined a wide, opaque, fast-growth zone (white in reflected light). We measured the distance to presumed annuli along the medial surface of the bone from the tip of the hyomandibular socket postero-ventrally to the farthest extension of the bone. A problem with using opercular bones for aging is that early annuli located closest to the hyomandibular socket may become hidden in older fish (Scoppettone 1988, Peterson et al. 1999). For fish  $\leq 150$  mm SL (all classified as juveniles based on gonadal development; SL = standard length), we determined the average distances (mm) to the first, second, and third annuli to be 4.07 (95% CI 3.76 to 4.38,  $n = 17$ ), 6.07 (95% CI 5.57 to 6.57,  $n = 16$ ), and 7.8 (95% CI 7.17 to 8.43,  $n = 15$ ), respectively (Fig. 2). For all fish  $>150$  mm SL we recorded all visible annuli and then added the

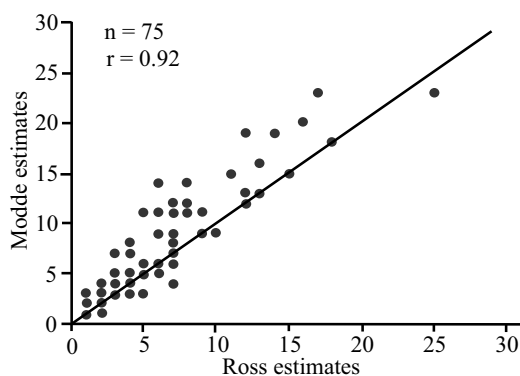


Fig. 3. Agreement between T.C. Modde and S.T. Ross on numbers of annuli in otoliths.

early annuli as appropriate based on the distance to the first annular mark. We analyzed opercular bones from all 4 years. However, opercular bones from 2001 were aged by direct viewing with a dissecting microscope rather than with digital images. Because lengths to annuli 1–3 were not available for these fish, readings were adjusted for hidden annuli by determining fish lengths at which the adjustments were made for fish collected in 1998, 1999, and 2001. No adjustments were required for fish averaging 141.4 mm SL ( $n = 19$ , upper 95% confidence limit = 161.6 mm SL); ages were adjusted by 1 for fish averaging 263.9 ( $n = 21$ ; 95% CI 252.1 to 275.7) and by 2 for fish averaging 312.7 ( $n = 6$ , lower 95% confidence limit = 279.3). To be conservative, we did not adjust opercles for fish <252 mm SL.

Transverse sections of sagittal otoliths (DeVries and Frie 1996) were prepared by mounting them with the globular anterior edge down on a glass slide, using clear thermoplastic cement, and then grinding with progressively finer grit abrasives (600–2000 grit) until the nuclear region of the otolith was visible. The otolith was then turned over and ground on the opposite side for a final thickness of 20–30 microns. The mounted slides were placed in a glass petri dish, covered with distilled water, and viewed over a black background at 40–60 $\times$ , with reflected light directed at a low angle onto the slide. Images were captured with a Diagnostics Instruments model 3.2.0 digital camera and Spot version 3.3 software or with the Nikon digital camera as described above. We enhanced images and obtained measurements of otoliths and dis-

tances between annuli by using ImageJ as described above.

Following Stevenson and Secor (1999), we identified presumed annuli in otoliths as the region including an opaque and translucent (hyaline) zone. Otolith terminology remains variable, even with respect to translucent and opaque zones (Williams and Bedford 1974). Viewed with reflected light, otoliths showed wide opaque zones and narrow translucent zones. Opaque zones, formed during periods of more rapid growth, tend to be high in calcium and protein, whereas translucent zones, formed during slow growth periods, are low in calcium and protein (Kalish 1989, Stevenson and Secor 1999).

All images of scales, opercles, and otoliths were read at least twice by S.T. Ross (STR), with intervals of months to years between readings. Images were read without reference to fish length or previous age estimates with the following protocol: Images were viewed with Adobe Illustrator so that presumed annuli could be marked directly on the image, with each aging placed in a separate layer. After the second reading, the positions of the annuli were compared and a final decision made on the number of annuli. In addition, samples of otoliths from 1998, 1999, 2001, and 2002 were read by both STR and T.C. Modde (TCM), with 63% agreement at  $\pm 1$ . Differences were greater for fish estimated to be >6 years (Fig. 3). Following this, otoliths were reprocessed to improve resolution, and a second otolith was also examined when possible. These images were then read by STR as described above. Comparison of the first and second age estimates by STR for otoliths showed an 84% agreement ( $n = 111$ ) with only a small effect of fish size on the magnitude of the difference ( $R^2 = 0.06$ ). Percent agreement between the first and second readings were lower for opercles (75%;  $n = 91$ ) and scales (70%;  $n = 111$ ), with a similarly small effect of fish size on the magnitude of difference for scales ( $R^2 = 0.05$ ), and a slightly larger effect for opercles ( $R^2 = 0.16$ ).

Because we were able to sample fish only during moderate flow periods of late June to early July, seasonal methods for verification of annular marks, such as marginal increment analysis (Beamish and McFarlane 1987, DeVries and Frie 1996), were not possible. However, Brouder (2005) verified annular marks in



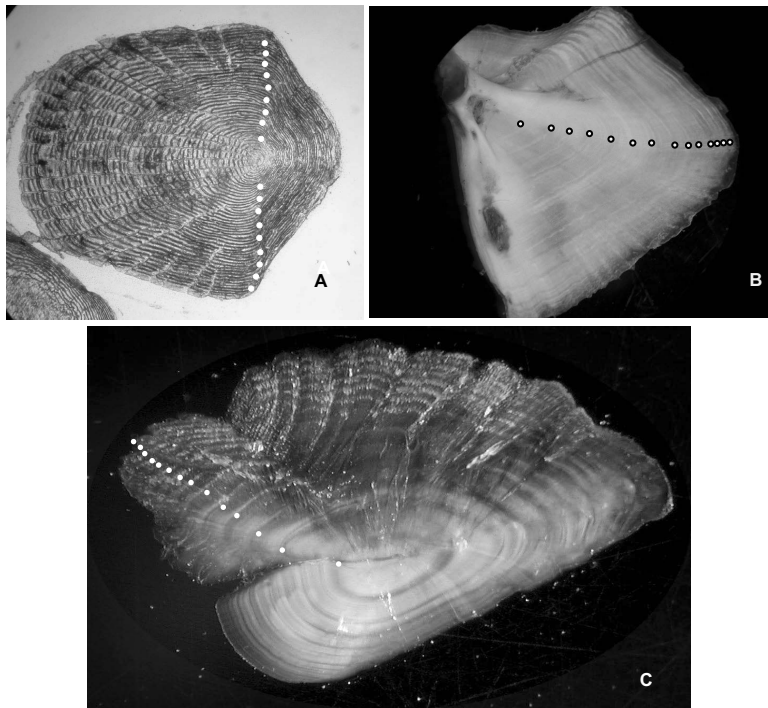


Fig. 4. Structures used in aging Roundtail Chub *Gila robusta*. **A**, Scale from a 295 mm TL female with 9 annuli. **B**, Opercle from a 380 mm TL female showing 14 annuli with an additional hidden annulus. **C**, Otolith from the same female as in **B** showing 14 annuli.

Roundtail Chub from the Verde River, Arizona, by using marginal increment analysis of seasonally collected fish and by examining otoliths of known-age fish. Using length frequency analyses for age corroboration was also ineffective because of bias against small fish in both the electrofishing and angling sampling (Quist et al. 2012).

#### Data Analysis

Regression analyses and other basic statistics were done with AcaStat software for the Mac (version 10.2.4). We estimated survivorship using a linearized catch-curve, after truncating the analysis so that no age interval had fewer than 5 fish and so that only ages that appeared to be fully recruited to the sampling gear were used (Miranda and Bettoli 2007). Because fish were collected by angling rather than electrofishing in 1998, we checked for bias in the size of fish captured for the 2 techniques. Overall, fish captured by angling (mean = 303 mm TL, SD = 66.4,  $n = 63$ ) were significantly larger than those taken by electrofishing (mean = 247 mm TL, SD =

87.0,  $n = 176$ ,  $P < 0.001$ ). However, mean lengths for ages of fish used in the catch-curve analysis did not differ between the 2 sampling gears (angling mean = 335 mm TL, SD = 63,  $n = 20$ ; electrofishing mean = 310 mm TL, SD = 60,  $n = 61$ ), so the data were combined in the analysis.

We performed an analysis of covariance (ANCOVA) to examine variation in mass/length relationships between males and females, using a combination of three R packages: *car* (Fox and Weisberg 2019), *effects* (Fox and Weisberg 2019), and *ggplot2* (Wickham 2016). We used a log transformation on the response variable (mass) and covariate (total length) to achieve linearity. After checking the assumptions of the ANCOVA model, data were fit using the *aov* function in R.

#### RESULTS

From all 4 years of data, we obtained interpretable annuli for 111 otoliths (1998,  $n = 24$ ; 1999,  $n = 23$ ; 2001,  $n = 35$ ; 2002,  $n = 29$ ), 91 opercles (1998,  $n = 7$ ; 1999,  $n = 27$ ; 2001,

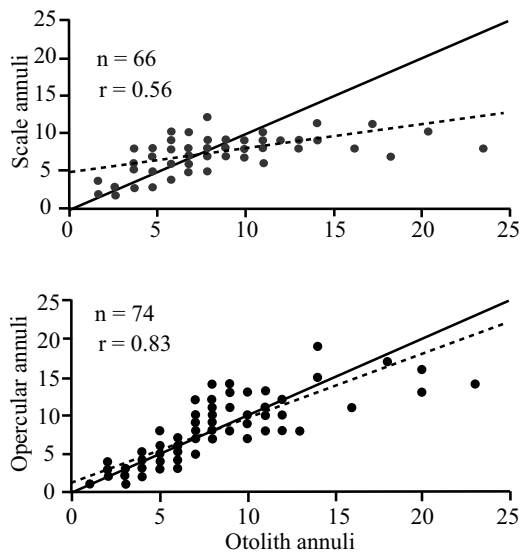


Fig. 5. Ages of Roundtail Chub *Gila robusta* based on scales (upper) and opercles (lower) versus ages based on otoliths. Solid line shows a 1:1 relationship; dashed line shows the linear regression line.

$n = 38$ ; 2002,  $n = 19$ ), and 111 scales (1998,  $n = 54$ ; 1999,  $n = 27$ ; 2001,  $n = 30$ ) (Fig. 4).

Although variable, scale annuli generally agreed with otolith annuli up to 7–8 years, after which they underestimated ages. Annuli from opercular bones showed better agreement with annuli based on otoliths up to 10–12 years before underestimating ages based on otoliths (Fig. 5). Based on the above information, and following the recommendation by Quist et al. (2007), for the remainder of the analyses, ages are based on otoliths.

For comparison with other studies, we present total length (TL). Standard length (SL) is related to total length with the following equation:

$$SL = -4.198 + 0.824(TL)$$

( $n = 239$ ,  $R^2 = 99.6$ ,  $P < 0.00001$ ). Roundtail Chub ranged from 63 mm to 446 mm TL, with an average of 262 mm TL ( $n = 239$ ); mass ranged from 6 g to 766 g, with an average of 181 g ( $n = 207$ ).

Based on the 4 sampling years, the oldest fish was a 350 mm TL male with 22 annuli; 7 fish exceeded 15 years and ranged in size from 350 mm to 446 mm TL (Fig. 6). For ages 6–14, the Roundtail Chub had an instantane-

TABLE 2. Age and length data for Roundtail Chub (*Gila robusta*). The last 7 data points were combined in the analyses: mean TL = 404.14 mm, mean age = 18.57 years.

Mean TL (mm)	Age (years)	<i>n</i>
71.30	1	1
126.10	2	4
121.28	3	8
175.49	4	7
219.00	5	10
245.00	6	14
279.10	7	10
307.36	8	14
328.71	9	7
324.50	10	8
343.63	11	8
351.40	12	5
329.33	13	3
378.20	14	5
374.00	16	1
406.00	17	2
423.00	18	1
435.00	20	2
350.00	22	1

ous mortality rate of  $-0.158$ , which is equivalent to 85% survivorship.

Growth in length (Table 2) was modeled using a von Bertalanffy equation based on a Walford plot (Ricker 1975, Quist et al. 2012). As is typical for many species, growth was initially rapid and then declined at older ages (Fig. 7). Data points for lengths of males and females versus age were broadly overlapping, so males and females, along with juvenile fish, were combined in the analysis.

As a measure of condition, we determined mass–length relationships for all fish that were weighed and measured, where

$$\log_{10}(\text{mass}) = -5.046 + (2.968) \log_{10}(\text{TL})$$

( $n = 200$ ,  $R^2 = 0.987$ ,  $P < 0.0001$ ).

We also determined mass–length relationships for fish identified as males ( $n = 37$ ) or females ( $n = 38$ ). Both mass and total length measures were logarithmically transformed to achieve linearity. Initial analysis of covariance and subsequent assumption testing revealed nonhomogeneity of regression slopes between males and females. Furthermore, residuals were non-normally distributed and variances among sexes were unequal. Five individual measures were outliers that significantly influenced the fit of the model. To achieve normality of residuals and homogeneity of variances, we removed the 5 outliers and repeated the

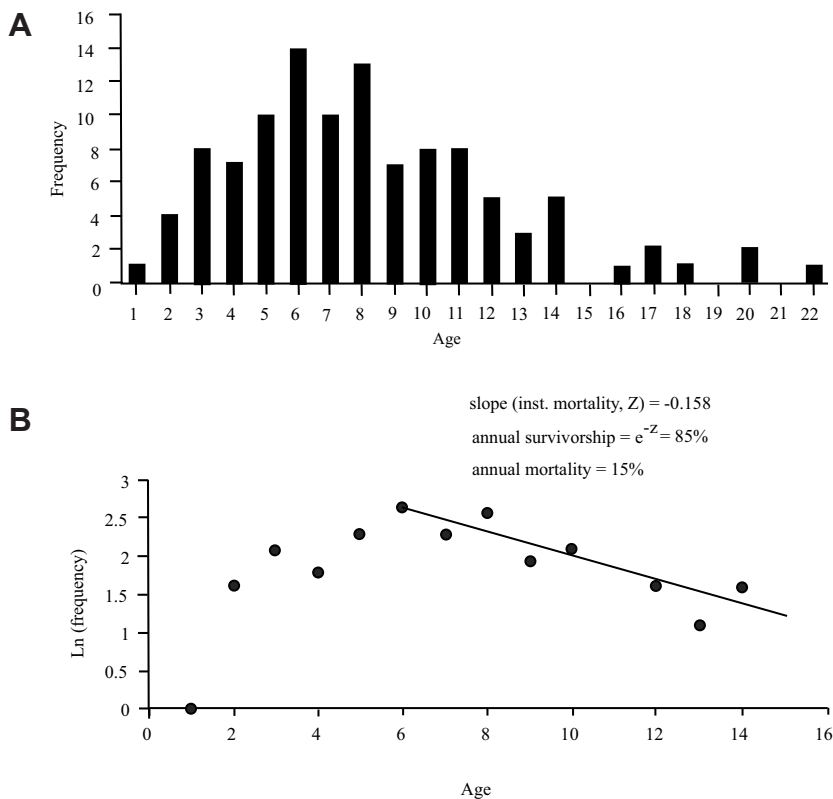


Fig. 6. **A**, Age frequency distribution based on otoliths of Roundtail Chub *Gila robusta*. **B**, Survivorship based on a catch-curve for ages 6–14.

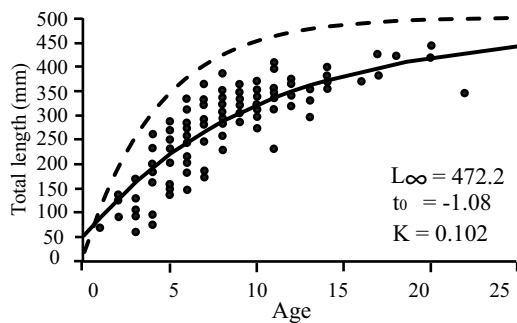


Fig. 7. Total length versus age for Roundtail Chub *Gila robusta* from the Yampa River (closed circles;  $n = 111$ ). The solid line shows the fit of a von Bertalanffy growth model. The dashed line shows a von Bertalanffy growth curve for Roundtail Chub from the Verde River, Arizona (from Brouder 2005).

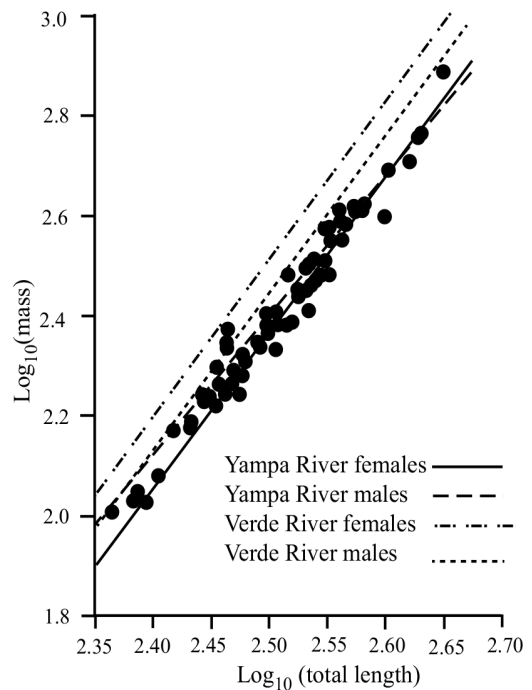


Fig. 8. (Right) Mass (g) versus total length (mm) for Roundtail Chub *Gila robusta* from the Yampa (present study) and Verde (Brouder 2005) rivers. Closed circles show data for both sexes of the Roundtail Chub from the Yampa River. Data were  $\log_{10}$  transformed for linearity.



TABLE 3. Mass–length relationships of male and female Roundtail Chub *Gila robusta* using analysis of covariance. Both fish length and mass were  $\log_{10}$  transformed.  $R^2 = 0.962$  (adjusted  $R^2 = 0.960$ ).

Source	Type III SS	df	Mean square	F	P value
Intercept	0.949	1	0.949	710.97	0.000***
Total length (TL)	2.096	1	2.096	1570.31	0.000***
Sex	0.006	1	0.006	4.37	0.041*
TL * Sex	0.006	1	0.006	4.27	0.043*
Error	0.088	66	0.001		
TOTAL	3.145	70			

\* $P \leq 0.05$   
 \*\*\* $P \leq 0.001$

TABLE 4. Estimated mean mass ( $\log_{10}$  transformed) of male and female Roundtail Chub *Gila robusta* after adjusting for the covariate total length.

Sex	Mean	SE	95% Confidence interval	
			Lower bound	Upper bound
Female	2.4	0.007	2.387	2.413
Male	2.409	0.006	2.396	2.421

analysis (33 females, 35 males; Fig. 8). After removal of outliers, the linear model still showed a significant interaction effect between the independent variable (sex) and the covariate (TL), indicating that the covariate (TL) significantly adjusts the association between predictor (sex) and response (mass) variables ( $F = 1570.31$ ,  $P < 0.0001$ ; Table 3). Overall, intercepts ( $F = 4.37$ ,  $P < 0.05$ ) and slopes ( $F = 4.27$ ,  $P < 0.05$ ) differed slightly between sexes. However, mean mass was comparable between sexes when adjusting for the covariate (Table 4). Our model included interaction between total length and sex and accounted for about 96% of the total variation ( $R^2 = 0.960$ ). Mass–length relationships for females and males, respectively, were the following:

$\log_{10}(\text{mass}) = -5.34 + 3.08(\log_{10} \text{TL})$   
 and

$\log_{10}(\text{mass}) = -4.56 + 2.78(\log_{10} \text{TL})$ .

Data are limited, but based on the presence of gonads in the early maturing or maturing stages, both sexes had similar ages at first reproduction, with females showing early maturing gonadal stages at 5 years, and later maturing or mature gonadal stages from 8 to 20 years (Table 5). Fish in these older categories likely had spawned multiple times. We did not collect females with later develop-

TABLE 5. Female gonadal stages versus age. LA = latent, EM = early maturing, LM = late maturing, MA = mature. See Table 1 for more detailed code definitions.

AGE	LA (n = 7)	EM (n = 9)	LM (n = 4)	MA (n = 16)
2	1			
4	1			
5		3		
6	3			
7		1		
8	2	1	1	3
9		2		1
10		2		2
11			1	1
12				1
≥13			2	8

mental stages of ovaries (e.g., MR to RE). Males showed testicular development beginning at 6 and 7 years, with mature or running ripe testes in fish of 6–22 years (Table 6).

Both sexes develop tubercles, with the intensity and extent of tubercles on the body related to gonadal stages. The greatest tubercle development occurred in males with maturing or running ripe testes, and in females with maturing ovaries (Table 7).

## DISCUSSION

Scales and otoliths have been widely used as aging structures for North American freshwater fishes, while opercular bones are used less often (Maceina et al. 2007). When compared with Roundtail Chub otolith annuli, scale and opercular annuli generated similar age estimates for young fish but underestimated ages for older fish. In a comparison of scales and otoliths as aging structures in Utah Chub *Gila atraria*, ages generally agreed between scales and opercle bones, but use of the latter gave a maximum age of 11 compared to 8 (Neuhold 1957). Studies of other

TABLE 6. Male gonadal stages versus age. LA = latent, MA1 = maturing, MA2 = mature, RE = ripe. See Table 1 for more detailed code definitions.

AGE	LA (n = 9)	MA1, 2 (n = 13)	RE (n = 12)
6	4	2	
7		2	3
8	4	1	
9	1	2	1
10			3
11		2	3
12		2	1
≥13		2	1

cypriniform species have shown that scale annuli underestimate opercle annuli in the catostomid Cui-ui *Chasmistes cujus* for fish >6 years, but there was close agreement between opercle and otolith annuli for fish up to 13 years (Scoppettone 1988). Opercular bones rather than scales were also recommended for aging Common Carp *Cyprinus carpio* in Utah (McConnell 1952). Similarly, the maximum age reported for White Sucker based on scales was 9–10 years, but the maximum age was more than doubled when fin ray sections were used for aging (Beamish and McFarlane 1983).

Although otoliths are a superior aging structure for long-lived fishes such as Roundtail Chub, they also require more processing time than opercles or scales, and use of otoliths and opercle bones for aging is lethal to the fish. In instances where the focus is on younger age groups, scales would be an appropriate aging structure. In addition, the increased precision and accuracy obtained from otoliths or opercle bones may need to be weighed against the use of lethal techniques, especially for studies of threatened or endangered fishes. In cypriniform fishes, sectioned pectoral rays have also shown promise as nonlethal aging structures, with age estimates based on pectoral rays only slightly lower than ages from otoliths in White Sucker *Catostomus commersonii* older than 6 years (Sylvester and Berry 2006). Similarly, ages of Common Carp based on pectoral rays only had a 1.2 percent error relative to otoliths (Phelps et al. 2007).

The largest fish we examined was a 446 mm TL female (366 mm SL) and the oldest was a 22-year-old male. In contrast, the largest and oldest Roundtail Chub collected by Vanicek and Kramer (1969) in the Green River was a 366 mm TL, 7-year-old female. The

TABLE 7. The relationship of tubercle and gonadal stages for males and females. EM = early maturing, MA = mature, LA = latent, RE = ripe. See Table 1 for more detailed code definitions.

Sex	Tubercle stage	Gonadal stage		
		EM	MA	
Female	1	1	1	
	2	—	4	
	3	—	3	
Male		LA	MA	RE
	1	1	—	—
	2	—	4	—
	3	—	2	4
	4	—	1	2

discrepancy in age is likely due in part to their use of scales as an aging structure. Based on various aging structures, including otoliths, Roundtail Chub from a small headwater stream in the upper Colorado River basin in Wyoming did not exceed 5 years, and one reader, but not another, showed close agreement between aging structures (scales and otoliths) for these young fish (Quist et al. 2007). In the Verde River, part of the Gila River watershed, the longest and oldest fish (based on otoliths) reported by Brouder (2005) were a 427 mm TL, age-7 female and a 413 mm TL, age-6 male. In the Gila River in New Mexico, the largest fish was 405 mm TL and the oldest, based on scales, was 7, but fish from a smaller tributary of the Gila River had a maximum size of only 253 mm TL and a maximum age of 5. However, a preliminary analysis of opercle bones from some of the largest Gila River fish resulted in age estimates of up to 13 years (Bestgen 1985). The Utah Chub is also long lived, with a maximum reported age of 29 years based on otoliths, and a maximum size of approximately 350 mm SL (Johnson and Belk 1999).

Our estimate of survivorship for ages 6–14 was 85%, based on a linearized catch-curve. Assumptions for the use of catch-curves include basing the analysis on age groups that are fully recruited to the sampling gear, having a random sample, and constant recruitment over time (Miranda and Bettoli 2007, Smith et al. 2012). The first 2 assumptions are likely satisfied, given the focus on sizes fully recruited to the gear and the attempt in the field to capture all fish observed during electrofishing. Size bias for fish ≥6 years caught by angling

versus electrofishing was not apparent from our data. Constant recruitment over time is not likely in most fish populations, especially for large western fishes, but as suggested by Miranda and Bettoli (2007), moderate variations in recruitment will not substantially alter the shape of a catch-curve, especially if catch data are pooled over several years as done here.

The 85% survivorship of adult fish was comparable to other large-river western fishes including Colorado Pikeminnow *Ptychocheilus lucius* and Razorback Sucker *Xyrauchen texanus*. For Colorado Pikeminnow, estimated survival rates for subadult and adult fish were 65%–82% for 2000–2003 (Bestgen et al. 2007) and 85% for adult fish for 1991–1999 (Osmundson et al. 1997). For Razorback Sucker, adult survival ranged from 69% (Bestgen et al. 2002) to 71% (Modde et al. 1996).

Growth in length, as expressed with the von Bertalanffy equation, showed a theoretical maximum size ( $L_{\infty}$ ) of 472.2 mm TL, which is similar to the value of 499.3 for Roundtail Chub from the Verde River (Fig. 7; Brouder 2005). However, Roundtail Chub from the Verde River had a maximum age of 7 years, in contrast to fish exceeding 15 years in the Yampa River, indicating that Verde River fish grew more rapidly than those in the Yampa River. Roundtail Chub from the Verde River also showed greater body mass per unit length compared to fish from the Yampa River (Fig. 8). The mass–length relationship for the Yampa River fish was essentially identical to the relationship for Green River Roundtail Chub determined by Vanicek and Kramer (1969):

$$\text{Log}_{10}(\text{Mass}) = -5.2462 + (3.086) \log_{10}(\text{TL}),$$

indicating that body condition did not show a meaningful change over the approximately 30-year interval between the 2 studies.

Reported ages at first reproduction of Roundtail Chub vary among different habitats. In the Yampa River, both sexes are mature by 5–7 years (present study). Similarly, fish from the Gila River matured at 3–7 years, with an average age for females of 5.2 years. But Roundtail Chub from the smaller tributary of the Gila River were mature at ages 2–3 (Bestgen 1985). Even though age at first reproduction is similar for the Gila and Yampa river populations, Yampa River fish, with their

greater longevity, may have a reproductive life span of 9–14 years, although Bestgen's preliminary aging data from opercle bones suggests that the 2 populations might be more similar. In comparison, Utah Chub had reproductive life spans of 6–9 years, with the greater life span occurring in lakes containing predaceous Cutthroat Trout *Oncorhynchus clarkii* (Johnson and Belk 1999).

Both sexes of Roundtail Chub develop tubercles, as also reported by Bestgen (1985), and development of tubercles is related to the degree of gonadal maturation. Although based on limited data, the more intense tuberculation occurs in males.

The variation in estimated longevity in different populations of Roundtail Chub may be due in part to the structures used in aging, but it may also be a function of habitat size. Bestgen (1985) indicated for Roundtail Chub in the Gila River drainage that larger streams may support older fish and that fish in small streams had earlier ages at first reproduction compared to fish in larger streams. On a broader scale, Smith (1981) documented that fishes in arid environments, including Utah Chub and Tui Chub *Siphateles bicolor*, increased in body length and maximum age as a function of habitat size, with Utah Chub living to 11 years (from Neuhold 1957, based on opercular annuli) in Bear Lake, Utah and Idaho, but Utah Chub in small habitats were smaller, shorter lived, and matured earlier than those in large habitats. In a later study of Utah Chub based on otoliths, the maximum ages in lakes with predators, including Bear Lake, were 13–29 years (Johnson and Belk 1999). Smith (1981) considered that the key variable controlling for body size, maturity, and longevity was adult mortality, with low adult mortality leading to selection for large body size and iteroparity. In contrast, high adult mortality would select for small body size and early maturation. Similarly, Utah Chub populations exposed to Cutthroat Trout predation had faster juvenile growth rates and delayed age at maturity, larger body size at maturity, and lower female reproductive effort than populations without predators, indicating that energy resources were initially directed toward outgrowing the window of predation from gape-limited Cutthroat Trout (Johnson and Belk 1999).

In terms of the 3-endpoint life history model of Winemiller and Rose (1992), the

historic population of Roundtail Chub in the Yampa River, as well as Utah Chub in lakes with predators, would be characterized as periodic species (larger body size, longer life span, longer generation time) versus populations of Roundtail Chub in smaller habitats such as tributaries of the Gila River (e.g., Turkey Creek; Bestgen 1985) or Utah Chub in lakes without predators, which would be characterized more as opportunistic species (smaller body size, shorter life span, shorter generation time; Johnson and Belk 1999, Mims et al. 2010, Ross 2013).

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#### LITERATURE CITED

- ALBANESE, B. 2000. Life-history of the Flagfin Shiner, *Pteronotropis signipinnis*, from a blackwater stream in southeastern Mississippi. *Ecology of Freshwater Fish* 9:219–228.
- ANDERSON, R.M., AND G. STEWART. 2003. Riverine fish flow investigations. Federal Aid in Fisheries and Wildlife Restoration. Federal Aid Project F-289-R6. Colorado Division of Wildlife, Denver, CO. 91 pp.
- BARDACH, J.E. 1955. The opercular bone of the yellow perch, *Perca flavescens*, as a tool for age and growth studies. *Copeia* 1955:107–109.
- BEAMISH, R.J., AND G.A. MCFARLANE. 1983. The forgotten requirement for age validation in fisheries biology. *Transactions of the American Fisheries Society* 112: 735–743.
- BEAMISH, R.J., AND G.A. MCFARLANE. 1987. Current trends in age determination methodology. Pages 35–42 in P.C. Summerfelt and G.E. Hall, editors, *The age and growth of fish*. Iowa State University Press, Ames, IA.
- BESTGEN, K.R. 1985. Distribution, biology, and status of the Roundtail Chub, *Gila robusta*, in the Gila River basin, New Mexico. Master's thesis, Colorado State University, Fort Collins, CO. 104 pp.
- BESTGEN, K.R., G.B. HAINES, R. BRUNSON, T. CHART, M. TRAMMELL, R.T. MUTH, G. BIRCHELL, K. CHRISTOPHERSON, AND J.M. BUNDY. 2002. Status of wild Razorback Sucker in the Green River basin, Utah and Colorado, determined from basinwide monitoring and other sampling programs. Final Report, Colorado River Recovery Implementation Program Project Number 22D, Denver, CO. 73 pp.
- BESTGEN, K.R., J.A. HAWKINS, G.C. WHITE, K.D. CHRISTOPHERSON, J.M. HUDSON, M.H. FULLER, D.C. KITCH- EYAN, R. BRUNSON, P. BADAME, G.B. HAINES, ET AL. 2007. Population status of Colorado Pikeminnow in the Green River basin, Utah, and Colorado. *Transactions of the American Fisheries Society* 136: 1356–1380.
- BROUDER, M.J. 2005. Age and growth of Roundtail Chub in the Upper Verde River, Arizona. *Transactions of the American Fisheries Society* 134:866–871.
- BROUDER, M.J., D.D. ROGERS, AND L.D. AVENETTI. 2000. Life history and ecology of the Roundtail Chub *Gila robusta*, from two streams in the Verde River basin. Technical Bulletin 3, Arizona Game and Fish Department, Phoenix, AZ. 17 pp.
- CARTER, J.M., M.J. CLEMENT, A.S. MAKINSTER, C.D. CROWDER, AND B.T. HICKERSON. 2018. Classification success of species within the *Gila robusta* complex using morphometric and meristic characters—a reexamination. *Copeia* 106:279–291.
- CASSELMAN, J.M. 1974. Analysis of hard tissue of pike *Esox lucius* L. with special reference to age and growth. Pages 13–27 in T.B. Bagenal, editor, *Ageing of fish*. Unwin Brothers, Limited, Surrey, U.K.
- COPUS, J.M., W.L. MONTGOMERY, Z.H. FORSMAN, B.W. BOWEN, AND R.J. TOONEN. 2018. Geopolitical species revisited: genomic and morphological data indicate that the Roundtail Chub *Gila robusta* species complex (Teleostei, Cyprinidae) is a single species. *PeerJ* 6:e5605. 32 pp.
- DEVRIES, D.R., AND R.V. FRIE. 1996. Determination of age and growth. Pages 483–512 in B.R. Murphy and D.W. Willis, editors, *Fisheries techniques*. 2nd edition. American Fisheries Society, Bethesda, MD.
- DOWLING, T.E., AND B.D. DEMARAIS. 1993. Evolutionary significance of introgressive hybridization in cyprinid fishes. *Nature* 362:444–446.
- FOX, J., AND S. WEISBERG. 2019. An R companion to applied regression. 3rd edition. SAGE Publishing, Thousand Oaks, CA. 472 pp.
- GERBER, A.S., C.A. TIBBETS, AND T.E. DOWLING. 2001. The role of introgressive hybridization in the evolution of the *Gila robusta* complex (Teleostei: Cyprinidae). *Evolution* 55:2028–2039.
- HEINS, D.C., AND M.D. MACHADO. 1993. Spawning season, clutch characteristics, sexual dimorphism and



- sex ratio in the Redfin Darter *Etheostoma whipplei*. American Midland Naturalist 129:161–171.
- JOHNSON, B.M., P.J. MARTINEZ, J.A. HAWKINS, AND K.R. BESTGEN. 2008. Ranking predatory threats by non-native fishes in the Yampa River, Colorado, via bioenergetics modeling. North American Journal of Fisheries Management 28:1941–1953.
- JOHNSON, J.B., AND M.C. BELK. 1999. Effects of predation on life-history evolution in Utah Chub (*Gila atraria*). Copeia 1999:948–957.
- KAEDING, L.R., B.D. BURDICK, P.A. SCHRADER, AND C.W. MCADA. 1990. Temporal and spatial relations between the spawning of Humpback Chub and Roundtail Chub in the Upper Colorado River. Transactions of the American Fisheries Society 119:135–144.
- KALISH, J.M. 1989. Otolith microchemistry: validation of the effects of physiology, age and environment on otolith composition. Journal of Experimental Marine Biology and Ecology 132:151–178.
- LE CREN, E.D. 1947. The determination of the age and growth of the Perch (*Perca fluviatilis*) from the opercular bone. Journal of Animal Ecology 16:188–204.
- MACEINA, M.J., J. BOXRUCKER, D.L. BUCKMEIER, R.S. GANGL, D.O. LUCCHESI, D.A. ISERMANN, J.R. JACKSON, AND P.J. MARTINEZ. 2007. Current status and review of freshwater fish aging procedures used by state and provincial fisheries agencies with recommendations for future directions. Fisheries 32:329–340.
- MARSH, P.C. 1997. Age estimation from otoliths of Bonytail Chub *Gila elegans* from Lake Mohave, Arizona and Nevada. U.S. Fish and Wildlife Service, Parker, AZ.
- MCCONNELL, W.J. 1952. The opercular bone as an indicator of age and growth of the Carp, *Cyprinus carpio* Linnaeus. Transactions of the American Fisheries Society 81:138–149.
- MIMS, M.C., J.D. OLDEN, Z.R. SHATTUCK, AND N.L. POFF. 2010. Life history trait diversity of native freshwater fishes in North America. Ecology of Freshwater Fish 19:390–400.
- MINCKLEY, W.L. 1973. Fishes of Arizona. Arizona Fish and Game Department, Phoenix, AZ. 293 pp.
- MINCKLEY, W.L., AND B.D. DEMARAIS. 2000. Taxonomy of chubs (Teleostei, Cyprinidae, genus *Gila*) in the American Southwest with comments on conservation. Copeia 2000:251–256.
- MIRANDA, L.E., AND P.W. BETTOLI. 2007. Mortality. Pages 229–277 in C.S. Guy and M.L. Brown, editors, Analysis and interpretation of freshwater fisheries data. American Fisheries Society, Bethesda, MD.
- MODDE, T., K.P. BURNHAM, AND E.J. WICK. 1996. Population status of the Razorback Sucker in the middle Green River (USA). Conservation Biology 10:110–119.
- NEUHOLD, J.M. 1957. Age and growth of the Utah Chub, *Gila atraria* (Girard), in Panguitch Lake and Navajo Lake, Utah, from scales and opercular bones. Transactions of the American Fisheries Society 85:217–233.
- NEVE, L.L. 1976. Life history of the Roundtail Chub *Gila robusta grahami*, at Fossil Creek, Arizona. Master's thesis, Northern Arizona University, Flagstaff, AZ.
- OSMUNDSON, D.B., R.J. RYEL, AND T.E. MOURNING. 1997. Growth and survival of Colorado Squawfish in the Upper Colorado River. Transactions of the American Fisheries Society 126:687–698.
- PAGE, L.M., C.C. BALDWIN, H. ESPINOSA-PÉREZ, L.T. FINDLEY, C.R. GILBERT, K.E. HARTEL, R.N. LEA, N.E. MANDRAK, J.J. SCHMITTER-SOTO, AND H.J. WALKER. 2017. Taxonomy of *Gila* in the Lower Colorado River basin of Arizona and New Mexico. Fisheries 42:456–460.
- PETERSON, M.S., L.C. NICHOLSON, D.J. SNYDER, AND G.L. FULLING. 1999. Growth, spawning preparedness, and diet of *Cycleptus meridionalis* (Catostomidae). Transactions of the American Fisheries Society 128: 900–908.
- PHELPS, Q.E., K.R. EDWARDS, AND D.W. WILLIS. 2007. Precision of five structures for estimating age of Common Carp. North American Journal of Fisheries Management 27:103–105.
- POPPER, A.N., AND S. COOMBS. 1982. The morphology and evolution of the ear in Actinopterygian fishes. American Zoologist 22:311–328.
- PROBST, D.L. 1999. Threatened and endangered fishes of New Mexico, Technical Report No. 1. New Mexico Department of Game and Fish, Santa Fe, NM. 84 pp.
- QUIST, M.C., Z.J. JACKSON, M.R. BOWER, AND W.A. HUBERT. 2007. Precision of hard structures used to estimate age of riverine catostomids and cyprinids in the Upper Colorado River basin. North American Journal of Fisheries Management 27:643–649.
- QUIST, M.C., M.A. PEGG, AND D.R. DEVRIES. 2012. Chapter 15: age and growth. Pages 677–731 in A.V. Zale and T.M. Sutton, editors, Fisheries techniques. 3rd edition. American Fisheries Society, Bethesda, MD.
- RICKER, W.E. 1975. Computation and interpretation of biological statistics of fish populations. Department of the Environment, Fisheries and Marine Service, Ottawa, Canada. 382 pp.
- RINNE, J.N. 1976. Cyprinid fishes of the genus *Gila* from the Lower Colorado River basin. The Wasmann Journal of Biology 34:65–107.
- ROSS, S.T. 2013. Ecology of North American freshwater fishes. University of California Press, Berkeley, CA. 460 pp.
- SCOPPETTONE, G.G. 1988. Growth and longevity of the Cui-ui and longevity of other catostomids and cyprinids in western North America. Transactions of the American Fisheries Society 117:301–307.
- SMITH, G.R. 1981. Effects of habitat size on species richness and adult body sizes of desert fishes. Pages 125–171 in R.J. Naiman and D.L. Soltz, editors, Fishes in North American deserts. John Wiley & Sons, New York, NY.
- SMITH, M.W., A.Y. THEN, C. WOR, G. RALPH, K.H. POLLOCK, AND J.M. HOENIG. 2012. Recommendations for catch-curve analysis. North American Journal of Fisheries Management 32:956–967.
- STEVENSON, J.T., AND D.H. SECOR. 1999. Age determination and growth of Hudson River Atlantic Sturgeon, *Acipenser oxyrinchus*. Fisheries Bulletin 98:153–166.
- SYLVESTER, R.M., AND C.R. BERRY. 2006. Comparison of White Sucker age estimates from scales, pectoral fin rays, and otoliths. North American Journal of Fisheries Management 26:24–31.
- [USFWS] UNITED STATES FISH AND WILDLIFE SERVICE. 1994. Endangered and Threatened wildlife and plants; determination of critical habitat for the Colorado River Endangered fishes: Razorback Sucker, Colorado Squawfish, Humpback Chub, and Bonytail Chub. Federal Register 59(54):13374–13400.
- [USFWS] UNITED STATES FISH AND WILDLIFE SERVICE. 2005. Endangered and Threatened wildlife and plants; listing *Gila* Chub as Endangered with critical



- habitat. 50 CFR Part 17. Federal Register 70: 66664–66721.
- [USFWS] UNITED STATES FISH AND WILDLIFE SERVICE. 2015. Endangered and Threatened wildlife and plants: Threatened species status for the Headwater Chub and a distinct population segment of the Roundtail Chub. Federal Register 80:60754–60783.
- [UTAH DNR] UTAH DEPARTMENT OF NATURAL RESOURCES. 2006. Range-wide conservation agreement and strategy for Roundtail Chub *Gila robusta*, Bluehead Sucker *Catostomus discobolus*, and Flannelmouth Sucker *Catostomus latipinnis*. Publication Number 06-18. Utah Department of Natural Resources, Salt Lake City, UT. 59 pp.
- VANICEK, C.D., AND R.H. KRAMER. 1969. Life history of the Colorado Squawfish, *Ptychocheilus lucius*, and the Colorado Chub, *Gila robusta*, in the Green River in Dinosaur National Monument 1964–1966. Transactions of the American Fisheries Society 98:193–208.
- WICKHAM, H. 2016. Ggplot2: elegant graphics for data analysis. Springer Publishing, New York, NY.
- WILLIAMS, T., AND B.C. BEDFORD. 1974. The use of otoliths for age determination. Pages 114–123 in T.B. Bagenal, editor, Ageing of fish. Unwin Brothers, Limited, Surrey, United Kingdom.
- WINEMILLER, K.O., AND K.A. ROSE. 1992. Patterns of life-history diversification in North American fishes: implications for population regulation. Canadian Journal of Fisheries and Aquatic Sciences 49:2196–2218.

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