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Otolith Microstructure Analysis Elucidates Spawning and Early Life Histories of Federally Endangered Fishes in the San Juan River

Stephani L. Clark Barkalow¹, Martinique J. Chavez¹, and Steven P. Platania¹

The short larval ontogenetic phase has a large impact on success or failure of fish populations due to naturally high mortality rates. Yet, for the federally endangered fishes Colorado Pikeminnow *Ptychocheilus lucius* and Razorback Sucker *Xyrauchen texanus*, information about this sensitive life stage is limited. We determined species-specific age-length functions, spawning periodicity, and environmental factors related to growth and the onset of spawning by these species in the San Juan River of the Colorado River basin. Daily ages were determined from lapillar otoliths, and growth rates were calculated for subsamples of San Juan River larval Colorado Pikeminnows and Razorback Suckers collected from 2009 through 2017 and six candidate age-length functions were fit to evaluate the relationship between age and growth. Spawning periodicity estimates from the best-fit-model and published function were compared to those from observed otolith ages and differences were analyzed. The responses of spawning periodicity and growth to abiotic and temporal conditions were evaluated. For both species, spawning dates produced by otolith ages and best-fit functions were significantly different from those produced by the published functions, but not from each other. Age-length functions determined in this study enable more accurate back-calculation of ages and prediction of spawning periodicity than published functions. The spawning periodicity estimate of San Juan River Colorado Pikeminnow and Razorback Sucker were both impacted by multiple parameterizations of temperature and discharge. For both species, fish age had the biggest influence on growth. Accurate knowledge of spawning periodicity and factors affecting fish growth can improve timing of management activities to maximize benefit to Colorado Pikeminnows and Razorback Suckers in the San Juan River.

LARVAL fishes serve as valuable indicators of ecological integrity of rivers (Schiemer et al., 2003) and of population dynamics (Humphries and Lake, 2000). Early life history studies are integral to the study of fishes and are important for detection of rare species (Brandenburg et al., 2019), recovery of endangered species (Snyder and Muth, 2004), and elucidation of spawning periodicity (Miller and Storck, 1984; Jolley et al., 2009; Clark Barkalow et al., 2020). Though the larval stage is temporally short, understanding processes affecting larval fishes is crucial for managing species. Natural mortality of larval fishes is high, driven by factors including starvation and predation. Environmental factors such as discharge and temperature influence growth rates (Miller et al., 1988), and transition time between ontogenetic phases (Clarkson and Childs, 2000; Bestgen, 2008) may increase or decrease exposure to factors affecting mortality.

Colorado Pikeminnow *Ptychocheilus lucius* and Razorback Sucker *Xyrauchen texanus* are federally endangered species once prevalent in the Colorado River basin, including the San Juan River (USFWS, 2002a, 2002b). Populations of both species declined due to large-scale habitat modification, potential competition with and predation by nonnative fish species, and poor water quality (USFWS, 2002a, 2002b). Predation pressures differ across ontogenetic phases of fishes with larvae being susceptible to predatory small-bodied cyprinids like Red Shiner *Cyprinella lutrensis* (Bestgen et al., 2006) or Fathead Minnow *Pimephales promelas* (Webber and Haines, 2014) that inhabit backwaters and other low velocity habitats. Monitoring of adult and larval life stages is performed in the San Juan River annually, and despite larval production by both species, a recruitment bottleneck exists. Though currently the ultimate cause of the recruitment

bottleneck is unknown, it may be influenced by predation, feeding ecology, or environmental factors (Bestgen et al., 2006; Farrington et al., 2018; Pennock et al., 2019).

As the second largest tributary of the Colorado River (SJRBRIP, 2018), the San Juan River historically supported robust populations of endemic big-river fishes including Colorado Pikeminnow and Razorback Sucker. The area between Navajo Dam and Lake Powell is designated critical habitat for both of these species (SJRBRIP, 2018). The drainage size and diversity of the ichthyofaunal community of the San Juan River make it an essential component of the upper Colorado River basin recovery effort. Recovery of Colorado Pikeminnow and Razorback Sucker in the San Juan River is an important milestone toward downlisting or delisting the species (USFWS, 2002a, 2002b). The upper San Juan River is typified by relatively unconfined floodplain with braided channels, heavy to moderate vegetation, and a moderate to steep gradient (Bliesner and Lamarra, 2000; SJRBRIP, 2018). The high prevalence of nonnative woody plants, including Russian Olive *Elaeagnus angustifolia* and Salt Cedar *Tamarix ramosissima*, contributed to the narrowing and stabilization of the channels (SJRBRIP, 2018). Lower reaches of the river below the confluence with Chinle are characterized by a dissected canyon, a slightly flatter gradient, and a higher sediment load relative to the upper section (Bliesner and Lamarra, 2000). Anthropogenic activities along the upper San Juan River include agriculture, livestock grazing, and extraction of fossil fuels.

Prior to its 1962 impoundment by the Navajo Dam, the San Juan River's hydrograph was characterized by high spring peaks with low base flows the rest of the year (Bliesner and Lamarra, 2000; Propst and Gido, 2004). The construction of Navajo Dam drastically altered the San Juan River's natural

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hydrograph resulting in reduced spring peak flows and increased base flows (Bliesner and Lamarra, 2000). With release capacity at 141.6 m³/s, spring peak flows below Farmington, New Mexico are unable to match those of the pre-dam hydrograph (Propst and Gido, 2004; SJRBRIP, 2018). Hypolimnetic releases from the dam largely alter the temperature regime of the San Juan River with cooler summer and warmer winter temperatures (SJRBRIP, 2018).

In dam-regulated systems, controlled release of water can be used to promote (Propst and Gido, 2004) or disrupt (Hill and Bestgen, 2014) spawning. Discharge in the San Juan River is partially regulated by the Navajo Reservoir, which is operated to meet water delivery obligations while also attempting to benefit Colorado Pikeminnows and Razorback Suckers (Propst and Gido, 2004). Inaccurate back-calculation of spawning dates could result in missed opportunities for water release from dams or inadvertent harm to larval fishes. A drainage-specific age-length equation is necessary for each species to accurately and efficiently back-calculate spawning dates and thus manage water usage for the benefit of the fishes.

Spawning can be difficult to detect by direct observation. However, hatch dates can be back-calculated from daily otolith growth rings and, with data on temperature dependent incubation period, used to estimate spawning dates (Hill and Bestgen, 2014; Clark Barkalow et al., 2020). Otoliths are acellular structures in the inner ear of a fish (Campana and Thorrold, 2001) that form annuli, alternating opaque and translucent rings, on daily and annual timescales. Due to the regularity of annulus deposition, otoliths are ideal structures for age determination studies. A variety of calcified structures produce growth rings—fin rays, vertebrae, cleithra, opercula, and scales—and have all been used with varying success to obtain annual age of fishes, but only otoliths may be used to obtain daily ages of larval fishes (Campana, 2001). Laboratory studies have confirmed that both Colorado Pikeminnows (Bestgen and Bundy, 1998) and Razorback Suckers (Bundy and Bestgen, 2001) form the first otolith increment on hatch date and increments are deposited daily post-hatch regardless of temperature. Otolith microstructure analysis is commonly used in both adult and larval fishes (Campana and Thorrold, 2001; Ashworth et al., 2017), including in the upper Colorado River basin, to determine population age classes (Albrecht et al., 2010), assess the relationship between somatic growth and age (Bestgen and Bundy, 1998), and back-calculate timing of processes such as hatching or spawning (Hill and Bestgen, 2014).

Aging larval otoliths is a time-intensive process requiring preservation of fishes in ethanol and dissection of larvae in the laboratory. Species-specific age-length functions can be used to expedite this process by estimating otolith age from observed larval fish size (Vilizzi and Walker, 1999; Katsanevakis and Maravelias, 2008; Ashworth et al., 2017), usually measured in standard length (SL) or total length (TL). A reliable age-length function is the foundation of calculating larval fish age from fish length. Growth rate may be impacted by the conditions in which larvae develop including temperature (Clarkson and Childs, 2000; Bestgen, 2008), food availability (Papoulias and Minckley, 1992), and water quality (McDougall et al., 2018). Differences in riverine conditions often result in species-specific age-length functions that may be inaccurate outside the river system where they were developed.

Age estimates of larval and juvenile San Juan River endangered fishes were previously determined using published functions calculated from hatchery-produced Colorado Pikeminnow larvae (Nesler et al., 1988) and Razorback Sucker larvae from the Green River (Bestgen et al., 2011). These age-length functions produce a poor fit of age to length when used for fishes in the San Juan River and generate unrealistic hatch and spawn dates (M. A. Farrington, pers. comm.). Thus, the published age-length functions misrepresent important parameters of reproductive and early life histories of San Juan River Colorado Pikeminnows and Razorback Suckers. In this study, we determined a San Juan River-specific age-length function estimating age from observed larval fish length for each species, resulting in more accurate estimates of spawning periodicity. Additionally, we evaluated effects of water temperature, discharge, and other variables on spawning periodicity and larval growth for Colorado Pikeminnows and Razorback Suckers. Accurate knowledge of spawning periodicity and factors impacting larval growth and spawning will help guide management activities to protect the sensitive life stages of both species.

MATERIALS AND METHODS

Study area.—The headwaters of the San Juan River originate in the San Juan Mountains; the basin drains approximately 98,400 km² into southwestern Colorado, northwestern New Mexico, southeastern Utah, and northeastern Arizona (SJRBRIP, 2018). The San Juan River flows about 365 km between its upstream impoundment of Navajo Dam and its terminus in Lake Powell. Perennial tributaries include the Animas, La Plata, and Mancos Rivers and McElmo Creek. The San Juan River is the downstream-most major tributary in the hydrologic area known as the upper Colorado River basin. The San Juan River is divided into eight geomorphic reaches between Navajo Dam and Lake Powell (Bliesner and Lamarra, 2000). Larval Colorado Pikeminnow and Razorback Sucker have not yet been documented in reaches 7 and 8; therefore, the study area for this project was limited to reach 6 through reach 1 (Fig. 1). The characteristics of the San Juan River in the study area were representative of the variability in the system from unconfined floodplain to dissected canyon.

Sample selection.—Extensive larval fish surveys have occurred since 1998 in the San Juan River to determine annual spawning periodicity and magnitude of reproduction by Colorado Pikeminnow and Razorback Sucker. Larval fishes were collected using a fine mesh (0.8 mm) seine (about 1 × 1 m), preserved in the field, identified, and catalogued in the Division of Fishes, Museum of Southwestern Biology (MSB), University of New Mexico. Larval fishes collected from 1988 to 2008 could not be used for this study as they were preserved in a 10% solution of formalin resulting in degraded otoliths; larvae were preserved in a 95% solution of ethanol beginning in 2009. Colorado Pikeminnows and Razorback Suckers used for this study were selected from MSB larval fish collections from 2009 to 2017. Only larval ontogenetic phases were used in this study. Juvenile fishes, from either species, were not examined due to increased variability in size at age in older life stages and capture bias by larval seine gear type. Institutional abbreviations used herein follow Sabaj (2020).

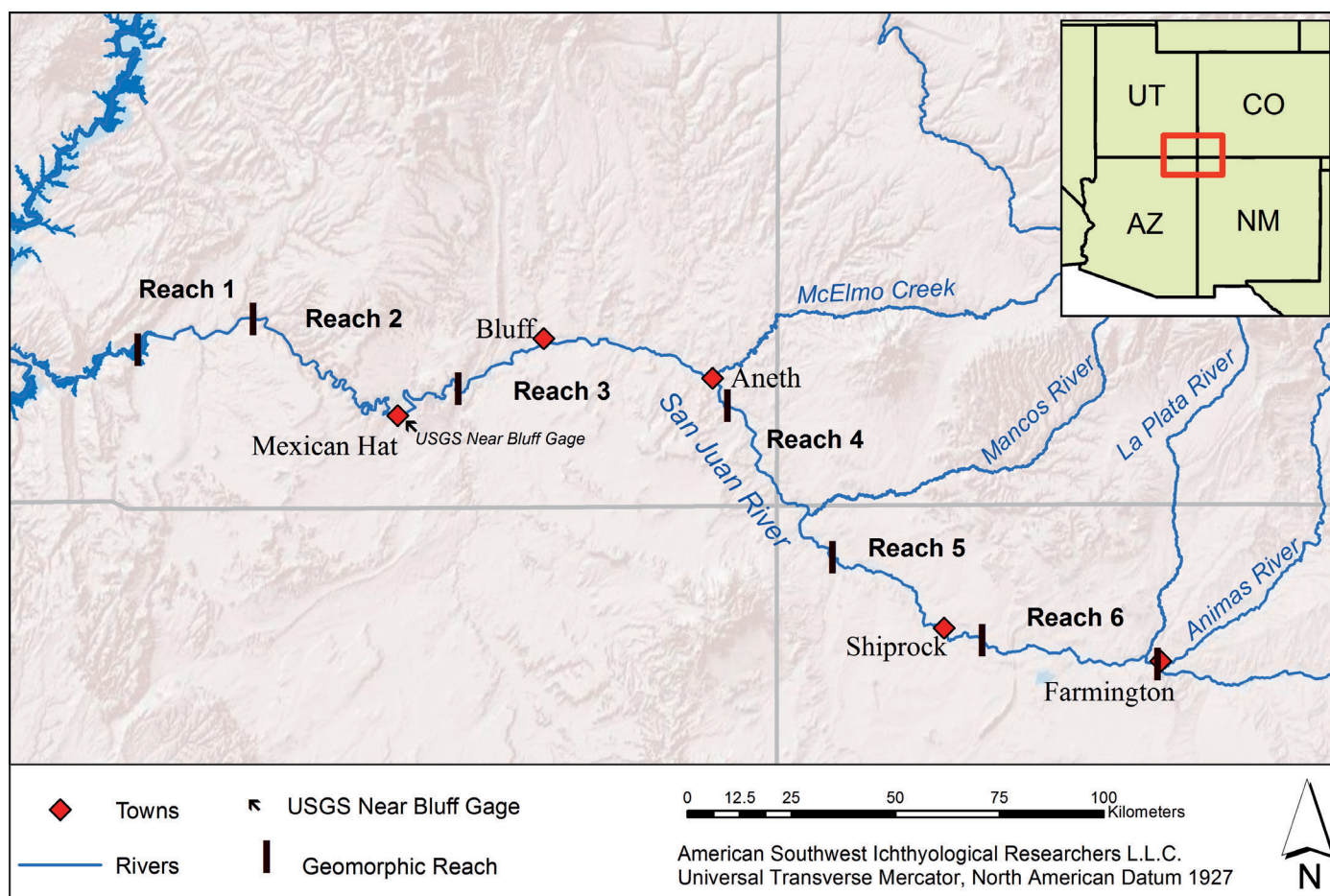


Fig. 1. Colorado Pikeminnow and Razorback Sucker larvae for this study were selected from 2009 to 2017 larval fish collections made in the San Juan River between reach 6, near Farmington, NM, and reach 1, near the terminus of the San Juan River in Lake Powell.

A total of 985 larval and juvenile (age-0) Colorado Pikeminnow (Table 1) and 7,187 Razorback Suckers (Table 2) were collected between 2009 and 2017. To prevent overutilization of fishes with certain spatial (reach, habitat) or temporal (month, ontogenetic phase) characteristics, larvae were subsampled from these collections when possible. Few Colorado Pikeminnows were collected in 2009–2013, 2015, and 2017 (Table 1), so every suitable larva collected in these years of low abundance was used in this study. However, the number of Colorado Pikeminnows taken annually in 2014 and 2016 exceeded 100, so larvae from these years were subsampled by month, habitat, reach, and ontogenetic phase. Due to the restricted distribution of larval Colorado Pikeminnows, samples were uneven in regard to month, habitat, reach, and ontogenetic phase. In total, 514 Colorado Pikeminnow larvae were selected for inclusion in this study. Sufficient numbers of Razorback Suckers were available to allow subsampling by month, habitat, reach, and ontogenetic phase, and 522 were selected for inclusion in the study.

Most larval fish monitoring occurred between reach 5 and reach 1, so the majority of larval fish selected for this project were from those reaches. Higher numbers of Colorado Pikeminnow and Razorback Sucker larvae were collected in backwaters relative to other habitat types (Farrington et al., 2018); therefore, this was the predominant mesohabitat type occupied by specimens used in the study. Larval Colorado Pikeminnows and Razorback Suckers taken in isolated pool

habitats were excluded from this study because those habitats were disconnected from the river and are not representative of the environmental conditions to which the vast majority of larval San Juan River fishes were exposed.

Otolith extraction and aging.—Sagittal and lapillar otoliths were extracted from the inner ear using a stereomicroscope fitted with a transmitted light base and polarized filter. Otoliths were embedded in Crystalbond 509 (SPI Supplies, West Chester, Pennsylvania) and mounted on a glass microscope slide between two pieces of 0.10-diameter ultra wire (Wapsi Fly UTC, Mountain Home, Arkansas) to prevent otoliths from being crushed. A glass cover slip was used to allow the otolith to be viewed with an oil immersion lens under microscopy.

Mounted otoliths were viewed under a BX53 microscope (Olympus, Tokyo, Japan) using Low Auto-Fluorescence Immersion Oil (Olympus) and oil immersion lenses under magnification of 100–1,000X. Lapillar otoliths are often preferred over other otoliths in the Cypriniformes because this structure is present at hatching and has a relatively uniform shape that enhances readability (Victor and Brothers, 1982; Hoff et al., 1997; Bestgen and Bundy, 1998). Of the three paired otoliths, lapilli were selected for use in both Colorado Pikeminnow and Razorback Sucker because sagitta became fragile in older fishes and asteriscus were not present when the fishes hatched. To reduce the introduction of

Table 1. Colorado Pikeminnow larvae collected between 2009 and 2017 in the San Juan River and subsampled for our study. Values in parentheses are length range (SL) present in each ontogenetic phase. The mesolarva category encompasses both flexion and post-flexion mesolarva ontogenetic phases.

Year	Protolarva	Mesolarva	Metalarva
2009	—	—	1 (19.2 mm)
2010	—	—	5 (16.2–21.4 mm)
2011	—	28 (8.4–11.2 mm)	4 (12.5–16.5 mm)
2012	—	—	—
2013	—	—	7 (12.3–13.5 mm)
2014	—	192 (8.0–12.1 mm)	117 (10.1–14.8 mm)
2015	1 (8.5 mm)	23 (8.2–9.1 mm)	—
2016	—	451 (8.3–12.2 mm)	92 (10.5–12.1 mm)
2017	—	39 (8.2–11.3 mm)	25 (11–17.5 mm)
Total	1	733	251

reader bias, otolith readings were conducted with all specimen collection identifiers masked and read in random order. Before the two readers examined the study specimens included, they independently aged 50 practice otoliths of each species. This process provided the readers with adequate practice to become familiar with the species-specific structure of otoliths and daily increment deposition. Two experienced readers (SLCB and MJC) independently determined age, in days, by counting daily growth rings from the primordium to the outer edge of the rostrum along the longest axis of the otolith (Sponagule, 2009). When the longest axis was not the clearest, both readers used the same alternate axis (Sponagule, 2009) to maintain consistency. The alternate axis was not set but was agreed upon by the readers based on annulus clarity. Each otolith was read twice, one time by each reader. When age counts between readers differed, otolith age was reconciled during a joint reading. While aging otoliths for this study, readers agreed 97.9% of the time for Colorado Pikeminnow and 97.1% for Razorback Sucker. Otolith age was reconciled in a joint reading for 2.1% ($n = 11$) Colorado Pikeminnow otoliths and 2.9% ($n = 15$) Razorback Sucker otoliths; all ages were reconcilable.

Growth rate.—To calculate daily somatic growth rate between hatching and date of collection, the SL at hatching for Colorado Pikeminnow (Hamman, 1981; Marsh, 1985; Bestgen and Williams, 1994; Snyder et al., 2016) and Razorback Sucker (Marsh, 1985; Snyder and Muth, 2004) was subtracted from SL

on collection date and divided by the number of annuli aged from otoliths. Because variation in hatch size could not be measured in wild spawned and hatched Colorado Pikeminnows and Razorback Suckers, mean hatch length from literature values was used in growth rate calculations. Length measurements of newly hatched protolarvae Colorado Pikeminnow and Razorback Sucker include both TL and SL, but because newly hatched larvae lack caudal-fin rays and hypural plates, measurements of SL and TL generally produce the same value and can be used interchangeably for fish of the protolarval ontogenetic phase. Colorado Pikeminnow length at hatching ranged from 5.0 to 7.0 mm TL (Hamman, 1981; Marsh, 1985; Bestgen and Williams, 1994; Snyder et al., 2016) with a mean length across studies of 5.5 mm TL. Razorback Sucker length at hatching ranged from 6.5 to 8.6 mm TL (Marsh, 1985; Snyder and Muth, 2004) with a mean length across studies of 8.0 mm TL.

Development of age-length functions.—The von Bertalanffy function is frequently used *a priori* to calculate the relationship between length and age (Zweifel and Lasker, 1976; Katsanevakis and Maravelias, 2008; Sibley et al., 2015); however, other growth equations may provide a better fit of the data. In total, six age-length functions were performed for each species (Tables 3, 4). Because the goal of this study was to develop age-length functions that can predict daily age from larval fish length, age was designated as the dependent variable in every function. Three sigmoidal functions, von Bertalanffy, Gompertz, and logistic functions were performed using the FSA (Ogle et al., 2019) package in program R (R Core Team, 2016). The von Bertalanffy, Gompertz, and logistic functions were mathematically rearranged from the original formats (Ogle et al., 2019) to analyze age as the dependent variable before being fit to data. A third order polynomial equation was calculated for each species to match the format of the existing Colorado Pikeminnow growth curve (Nesler et al., 1988). Lastly, two variations of multiple linear functions were fit to age and length datasets of both Colorado Pikeminnows and Razorback Suckers. The multiple linear functions were nearly identical except that SL in one function was natural log transformed (function hereafter referred to as transformed linear) and SL of the other linear function remained untransformed.

Two variations of multiple linear functions were fit to age and length datasets of both Colorado Pikeminnows and Razorback Suckers. The linear functions were nearly identical except that SL in one function was natural log transformed (function hereafter referred to as transformed linear) and SL

Table 2. Razorback Sucker larvae collected between 2009 and 2017 in the San Juan River and subsampled for our study. Values in parentheses are length range (SL) present in each ontogenetic phase. The mesolarva category encompasses both flexion and post-flexion mesolarva ontogenetic phases.

Year	Protolarva	Mesolarva	Metalarva
2009	28 (9.3–11.5 mm)	224 (10.4–15.9 mm)	5 (14.9–16.3 mm)
2010	246 (8.5–15.4 mm)	633 (8.9–23.5 mm)	44 (9.3–16.8 mm)
2011	58 (8.2–11.5 mm)	918 (9–17.4 mm)	77 (14.1–20.4 mm)
2012	20 (5.9–10.6 mm)	987 (6.2–18.4 mm)	161 (14.8–22.0 mm)
2013	56 (9.1–11.9 mm)	776 (9.3–16.9 mm)	72 (13.1–21.9 mm)
2014	75 (8.3–11 mm)	437 (9.4–17.1 mm)	22 (13.8–19.5 mm)
2015	220 (9.2–11.8 mm)	967 (9.3–18.7 mm)	—
2016	73 (9–11.9 mm)	679 (9.2–17.2 mm)	60 (14.9–23 mm)
2017	6 (8.8–10.7 mm)	288 (9.6–16.7 mm)	46 (15.5–21.9 mm)
Total	782	5,909	487

Table 3. Colorado Pikeminnow age-length models developed in this study. Because the intention of this study is to develop a growth curve to estimate age from larval fish standard length, growth models were evaluated with age as the dependent variable.

Model name	Equation	
von Bertalanffy	$age = \frac{\ln[-(\frac{t}{L_{inf}} - 1)]}{-K} + t_0$	$L_{inf} = 349.653$ $K = 0.001$ $t_0 = -8.154$
Gompertz	$age = \frac{-\ln[-\ln(\frac{SL}{L_{inf}})]}{g_i} + t_i$	$L_{inf} = 3.176 \times 10^7$ $g_i = 2.485 \times 10^{-3}$ $t_i = 1.105 \times 10^3$
Logistic	$age = \frac{\ln(\frac{L_{inf}}{SL} - 1)}{-g_{ninf}} + t_i$	$L_{inf} = 48.626$ $g_{ninf} = 0.048$ $t_i = 44.394$
Polynomial	$age = 0.009(SL^3) - 0.382(SL^2) + 7.650(SL) - 30.853$	
Transformed linear	$age = -42.086 + 26.196(\ln SL) - 1.080e^{-03}(JulyQ) - 5.480e^{-03}(Rkm)$	
Linear	$age = -5.685 + 2.381(SL) - 0.001(JulyQ) - 0.006(Rkm)$	

of the other linear function remained untransformed. Simultaneous backward and forward stepwise AIC variable selection was used to select variables for inclusion in functions (Yamashita et al., 2007). Multicollinearity was assessed using the variance inflation factor (VIF; Kabacoff, 2011; Montgomery et al., 2012), and any variable with VIF > 5 was removed from the model (Daoud, 2017). For each species, age was analyzed as the dependent variable with individual SL or lnSL, temperature at collection date, river km (Rkm), mean discharge (Q, m³/s) in the month in which most individuals were spawned, and cumulative growing degree-days (ΣGDD) at collection date (Table 5). Cumulative growing degree-days are a thermal integral used to parameterize metabolically relevant temperature in ectothermic organisms (Neuheimer and Taggart, 2007; Venturelli et al., 2010; Murphy et al., 2011). While both ΣGDD and temperature at collection date are related to river temperature, ΣGDD was included as a covariate to include a metric of the overall warmth of the river throughout the growing season, whereas temperature at collection date was included to evaluate influence of water temperature at the end of the growing period (i.e., the day the fish was captured). Water temperature for the San Juan River was obtained from a gage toward the lower end of the study area (Near Bluff Gage, USGS 09379500; located at Mexican Hat); however, water temperature increases with increasing distance downstream

of Navajo Dam. The Rkm parameter was included both to encompass spatial attributes of the river and to approximate differences in water temperature from upstream to downstream. Mean discharge (Q) in spawning month, also obtained from the Near Bluff Gage (USGS 09379500), was analyzed for each species. Mean spawning month for each species was calculated by subtracting otolith annulus count from the collection date and subtracting incubation period and is a mean value across the nine years of the study period; spawning month was not calculated for individual fish or dependent on calculated age from the age-length function. Across years, the majority of Colorado Pikeminnows spawned in July and the majority of Razorback Suckers spawned in April, so mean July Q was applied to discharge at spawning date for all Colorado Pikeminnow larvae and mean April Q was applied to discharge at spawning for all Razorback Sucker larvae. Cumulative GDD were calculated using the equation $\Sigma GDD = T_{avg} - T_{base}$ (Neuheimer and Taggart, 2007; Murphy et al., 2011), where T_{avg} is mean daily temperature and T_{base} is the minimum temperature required for spawning to occur. For Colorado Pikeminnows, T_{base} was 18°C (USFWS, 2002a) and for Razorback Suckers, T_{base} was 14°C (USFWS, 2002b). Because ΣGDD are based on minimum temperatures required to perform physiological processes, only temperatures greater than T_{base} were included in calculations. The ΣGDD at collection date parameter was

Table 4. Razorback Sucker age-length models developed in this study. Because the intention of this study is to develop a growth curve to estimate age from larval fish standard length, growth models were evaluated with age as the dependent variable.

Model name	Equation	
von Bertalanffy	$age = \frac{\ln[-(\frac{t}{L_{inf}} - 1)]}{-K} + t_0$	$L_{inf} = 1.143 \times 10^3$ $K = 2.894 \times 10^{-4}$ $t_0 = -1.602$
Gompertz	$age = \frac{-\ln[-\ln(\frac{SL}{L_{inf}})]}{g_i} + t_i$	$L_{inf} = 2.572 \times 10^3$ $g_i = 4.600 \times 10^{-3}$ $t_i = 3.864 \times 10^2$
Logistic	$age = \frac{\ln(\frac{L_{inf}}{SL} - 1)}{-g_{ninf}} + t_i$	$L_{inf} = 44.247$ $g_{ninf} = 0.035$ $t_i = 49.078$
Polynomial	$age = -0.030(SL^3) + 1.236(SL^2) - 13.437(SL) + 55.049$	
Transformed linear	$age = -62.941 + 38.051(\ln SL) + 0.076(AprilQ) + 0.011(Rkm) - 0.988(T) + 0.035(GDD_{coll})$	
Linear	$age = -2.783 + 2.841(SL) + 0.073(AprilQ) + 0.012(Rkm) - 0.999(T_{coll}) + 0.034(GDD_{coll})$	

Table 5. Parameters used in regression analyses.

Analysis	Parameter	Parameter description
A) Age–length	JulyQ	Q in mean spawning month for Colorado Pikeminnow
	AprilQ	Q in mean spawning month for Razorback Sucker
	T _{coll}	Temperature (°C) on collection day
	GDD _{coll}	Cumulative growing degree-days on collection date
	Rkm	River kilometer
B) Spawning	lnSL or SL	Fish standard length, dependent on function
	GDD _{spawn}	Cumulative growing degree-days on spawning date
	T _{spawn}	Temperature (°C) on spawn date
	Q _{spawn}	Mean daily discharge on spawning date
	Photoperiod	Daylength (hours)
	ΔQ	Seven day change in discharge
C) Growth	ΔT	Seven day change in temperature
	Otolith age	Obtained from annulus counts (days)
	T _{coll}	Temperature (°C) on collection day
	Q _{coll}	Mean daily discharge on collection date
	GDD _{coll}	Cumulative growing degree-days on collection date
	Rkm	River kilometer

chosen for inclusion in multiple linear regression analysis because it best represents the cumulative effect of temperature across the growing period from spawning to capture.

All six candidate age–length functions were ranked using Akaike information criterion (Burnham and Anderson, 2002; Symonds and Moussalli, 2011; Brewer et al., 2016). Support for each age–length function was evaluated using Akaike weights and ΔAICc. Models with ΔAICc < 2 were considered as top models.

Incubation period.—Incubation period was calculated using species-specific equations relating temperature and hatch rate. An equation for incubation period from Bestgen et al. (2011) was used for Razorback Sucker larvae

$$y = 1440.3e^{-0.109x}$$

where y = time to 50% hatch in hours and x = water temperature in °C. However, no published incubation equation existed for Colorado Pikeminnows. An equation for estimating incubation time was developed based on published data (Marsh, 1985; Bestgen and Williams, 1994) of time (h) to 50% hatch at incubation temperatures of 18–30°C. A regression line was fitted to the data and the resulting equation used to calculate incubation time in hours. Published data produced only six Colorado Pikeminnow from which to calculate temperature-dependent incubation period at temperatures of 18–30°C (Marsh, 1985; Bestgen and Williams, 1994). Incubation time of Colorado Pikeminnow was best described by the equation:

$$y = 315.42e^{-0.05x}$$

where y = time to 50% hatch in hours and x = water temperature in °C.

Comparison of spawning dates.—Back-calculated spawn dates from otolith ages, hereafter referred to as actual spawn dates, were compared with those produced by best-fit equations and published functions (Nesler et al., 1988; Bestgen et al., 2011). Spawning dates were calculated by subtracting hatch dates and incubation period from the collection date. Hatch dates were calculated by subtracting age, determined from

annulus counts or calculated from best-fit function or published functions, from collection date. Species-specific temperature-dependent incubation times were then subtracted from hatch dates to obtain spawning dates. Spawn dates across years were combined and analyzed by creating a frequency histogram with months and days as the independent variable. Actual spawn dates and spawning dates produced with best-fit equations and published functions (Nesler et al., 1988; Bestgen et al., 2011) were evaluated for normality using Anderson-Darling normality test. Spawning distributions were non-normal, so a Kruskal-Wallis test followed by *post hoc* Dunn's test was performed to evaluate differences between actual spawn dates and those produced by age–length functions.

Response of spawning periodicity and growth to abiotic and temporal variables.—Multiple linear regression was used to examine the relationship between environmental variables on spawning dates and growth (Table 5). Growth was calculated by subtracting mean hatch length from SL. To facilitate comparisons of spawning dates across years, dates were only considered as month and day. The seven-day difference in discharge (ΔQ) and seven-day difference in temperature (ΔT) were also included in the multiple linear regressions to encompass thermal or hydrograph changes that could promote spawning. To evaluate an annual effect, year was included as a covariate in the multiple linear regression. Simultaneous backward and forward stepwise AIC variable selection was used to select variables. Multicollinearity was assessed for each model using VIF, and any variable with VIF > 5 was removed.

RESULTS

Larval San Juan River Colorado Pikeminnows ($n = 514$) ranged from 7 to 34 d in age and from 7.8 to 17.5 mm SL. Mean Colorado Pikeminnow growth rate was 0.29 mm/d with a range of 0.18 to 0.54 mm/d. Growth rates were significantly different across years (ANOVA: $F = 12.07$, $df = 6$, $P < 0.0001$) with highest growth in 2010 (mean 0.38 mm/d) and lowest in 2011 (mean 0.24 mm/d). San Juan River Colorado Pikeminnow protolaryvae transitioned to flexion

Table 6. Colorado Pikeminnow age–length models were ranked using Akaike information criteria. The transformed linear model and linear model both had $\Delta\text{AICc} < 2$ and were the top performing models.

Model	K	AICc	ΔAICc	AICcWt	LogLik
Transformed linear	5	2365.89	0	0.7	−1177.89
Linear	5	2367.57	1.68	0.3	−1177.14
Logistic	4	2404.33	38.44	0	−1198.13
Gompertz	4	2405.00	39.11	0	−1198.62
Polynomial	5	2406.73	40.84	0	−1198.31
von Bertalanffy	4	2413.66	47.77	0	1202.79

mesolarvae at 8 d mean age, to postflexion mesolarvae at 11 d mean age, and to metalarvae at 16 d mean age.

San Juan River larval Razorback Suckers ($n = 522$) ranged from 9 to 51 d in age and from 8.9 to 22.0 mm SL. Mean Razorback Sucker growth rate was 0.20 mm/d with a range of 0.08 to 0.56 mm/d. Growth rates were significantly different across years (ANOVA: $F = 5.17$, $df = 8$, $P < 0.0001$) with highest growth rate in 2013 (mean 0.23 mm/d) and lowest in 2015 (mean 0.17 mm/d). San Juan River Razorback Sucker protolarvae transitioned to flexion mesolarvae at 13 d mean age, to postflexion mesolarvae at 20 d mean age, and to metalarvae at 28 d mean age.

San Juan River Colorado Pikeminnow larvae age–length functions.—The relationship between Colorado Pikeminnow age and length was best described by the transformed linear function (Table 6, Fig. 2):

$$\text{age} = -42.086 + 26.196(\ln\text{SL}) - 1.080e^{-03}(\text{JulyQ}) - 5.480e^{-03}(\text{Rkm})$$

where JulyQ = mean July discharge and Rkm = river km. Though the equation produced by the transformed linear equation ultimately had the lowest AICc value, and is here

forth presented as the top model, the linear model had $\Delta\text{AICc} < 2$ and is also a top model (Table 6).

San Juan River Razorback Sucker larvae age–length functions.—For Razorback Suckers, the relationship between age and length was best described by the linear function (Table 7, Fig. 3):

$$\text{age} = -2.783 + 2.841(\text{SL}) + 0.073(\text{AprilQ}) + 0.012(\text{Rkm}) - 0.999(\text{T}_{\text{coll}}) + 0.034(\text{GDD}_{\text{coll}})$$

where AprilQ = mean April discharge, Rkm = river km, T_{coll} = temperature in °C at collection date, and GDD_{coll} = growing-degree days at collection date. The linear model was the top model and was the only model with $\Delta\text{AICc} < 2$.

Incubation period.—Colorado Pikeminnow incubation time ranged from 3.30 to 5.49 d. Razorback Sucker incubation time calculated with the equation from Bestgen et al. (2011) ranged from 3.09 to 17.40 d.

Spawning periodicity.—Calculations from otolith ages showed that San Juan River Colorado Pikeminnows began spawning 10 June and finished spawning 18 July, with a small peak in the middle of June and a larger peak beginning in the first week of July. Spawn dates produced by back-calculating with the transformed linear equation—beginning 12 June and finishing 15 July—closely mirrored those calculated from otolith ages (Fig. 4), and more closely matched the peak spawning period than did spawn dates calculated from the published function (Nesler et al., 1988). Spawning dates produced by otolith ages and the transformed linear function were significantly different from those produced by the published function (Kruskal-Wallis: $\chi^2 = 24.095$, $df = 2$, $P < 0.001$; Dunn's test: $P < 0.001$) but not from each other (Dunn's test; $P = 0.99$). Across years, San Juan River Colorado Pikeminnows initiated spawning when

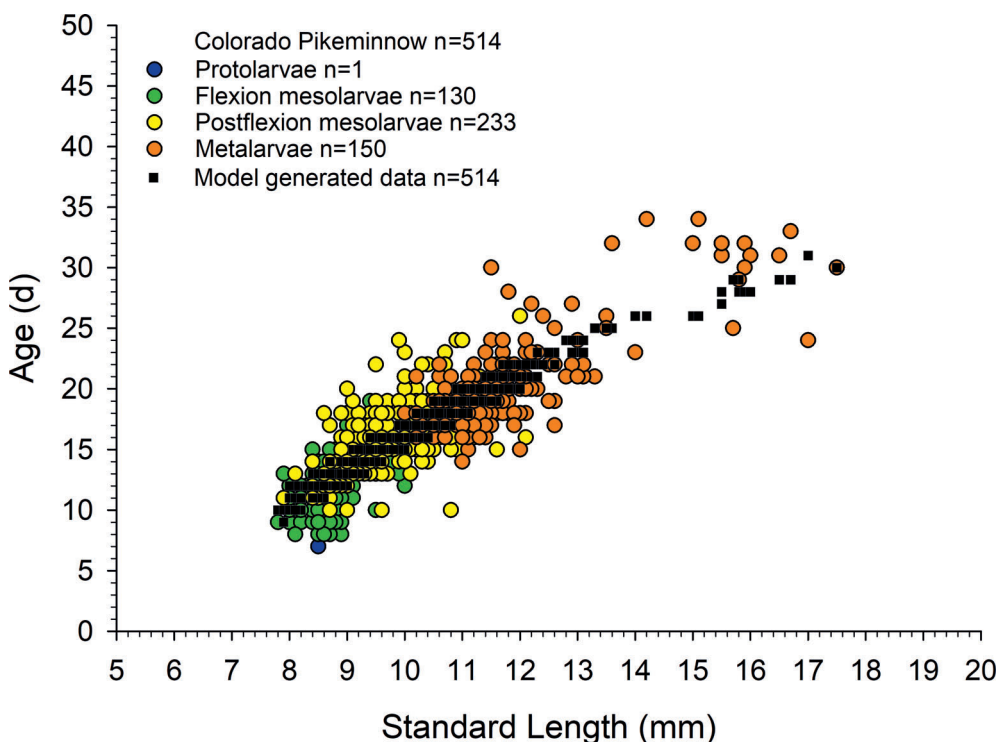


Fig. 2. Colorado Pikeminnow age as a function of length (SL). Colored dots indicate otolith ages, and black squares are predicted ages produced from the transformed linear function.

Table 7. Razorback Sucker age-length models were ranked using Akaike information criteria. Only the linear model had $\Delta\text{AICc} < 2$, so it was considered the best-fit model.

Model	K	AICc	ΔAICc	AICcWt	LogLik
Linear	7	3033.53	0	0.94	-1509.66
Transformed linear	7	3039.13	5.6	0.06	-1510.65
Polynomial	5	3085.48	51.95	0	-1537.68
Logistic	4	3106.23	72.7	0	-1549.08
von Bertalanffy	4	3106.54	73.01	0	-1549.23
Gompertz	4	3107.42	73.89	0	-1549.67

temperature was 15.9–24.2°C. Mean temperature on spawn date was 21.8°C.

San Juan River Razorback Sucker began spawning 20 March and ended 5 July, according to calculations from otolith ages and temperature-dependent incubation equation. A large peak in spawning occurred in the second half of April. Spawn dates produced by back-calculated ages from the linear equation closely mirrored those calculated from otolith ages (Fig. 5) with initiation on 15 March and cessation 28 June and a peak overlapping the peak produced by otolith ages. Conversely, spawn dates calculated by the published function (Bestgen et al., 2011) showed earlier initiation of spawning (26 February), later cessation of spawning (12 July), and earlier peak spawning (Fig. 5). Spawning dates produced by otolith ages and the linear function were significantly different from those produced by the published function (Kruskal-Wallis: $\chi^2 = 121.14$, $\text{df} = 2$, $P < 0.001$; Dunn's test: $P < 0.001$) but not from each other (Dunn's test: $P = 0.92$). Razorback Sucker initiated spawning when temperature was 11.2–16.1°C. Mean temperature on spawn date was 15.2°C.

Response of spawning periodicity and growth to abiotic and temporal variables.—A multiple linear regression was calcu-

lated to evaluate influence of abiotic and temporal variables on San Juan River Colorado Pikeminnow spawning periodicity and growth. Following an initial multiple linear regression temperature at spawn date (T_{spawn}) was omitted from further analyses due to $\text{VIF} > 5$. Spawning periodicity was explained by $\text{GDD}_{\text{spawn}}$, ΔT , ΔQ , photoperiod, and year (Table 8; MLR: $R^2 = 0.897$, $F_{5,508} = 883.9$, $P < 0.001$). Growth of larval San Juan River Colorado Pikeminnow was primarily by age and river km (Table 8; MLR: $R^2 = 0.719$, $F_{2,511} = 652.2$, $P < 0.001$).

Likewise, a multiple linear regression was performed to evaluate influence of abiotic and temporal variables on San Juan River Razorback Sucker spawning periodicity and growth. Spawning periodicity (MLR: $R^2 = 0.986$, $F_{4,517} = 8851$, $P < 0.001$) was influenced by $\Sigma\text{GDD } T_{\text{spawn}}$, Q_{spawn} , photoperiod, ΔT , ΔQ , and year (Table 9). Larval San Juan River Razorback Sucker growth was influenced by river km, T_{coll} , age, and Q_{coll} (MLR: $R^2 = 0.765$, $F_{4,517} = 421.1$, $P < 0.001$).

DISCUSSION

Larval Colorado Pikeminnows and Razorback Suckers in the San Juan River experience different abiotic conditions, resulting in differences in growth and developmental rates. Mean San Juan River Razorback Sucker growth rate was lower than that of San Juan River Colorado Pikeminnow. Rapid growth enhances survival because of improved ability to feed and to avoid predators (Bestgen et al., 2006; Bestgen, 2008). Both species become less susceptible to gape-limited predatory cyprinids around 25 mm TL (Bestgen et al., 2006; Bestgen, 2008), so early-season larvae with slower growth rates (Bestgen et al., 2006) would be more susceptible to predation for longer than late-season larvae. In laboratory studies, Razorback Suckers exceeded 25 mm TL in 30–41 d depending on temperature (Bestgen, 2008) and transitioned to the juvenile ontogenetic phase at 27–30 mm TL (Snyder

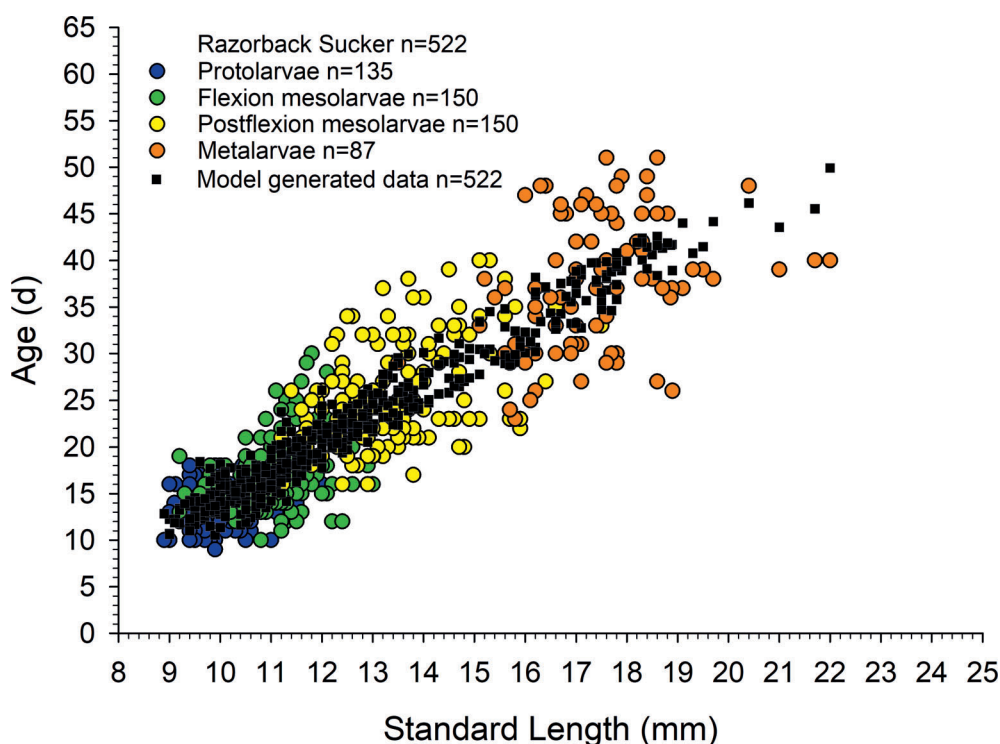


Fig. 3. Razorback Sucker age as a function of length (SL). Colored dots indicate otolith ages, and black squares are predicted ages produced from the linear growth function.

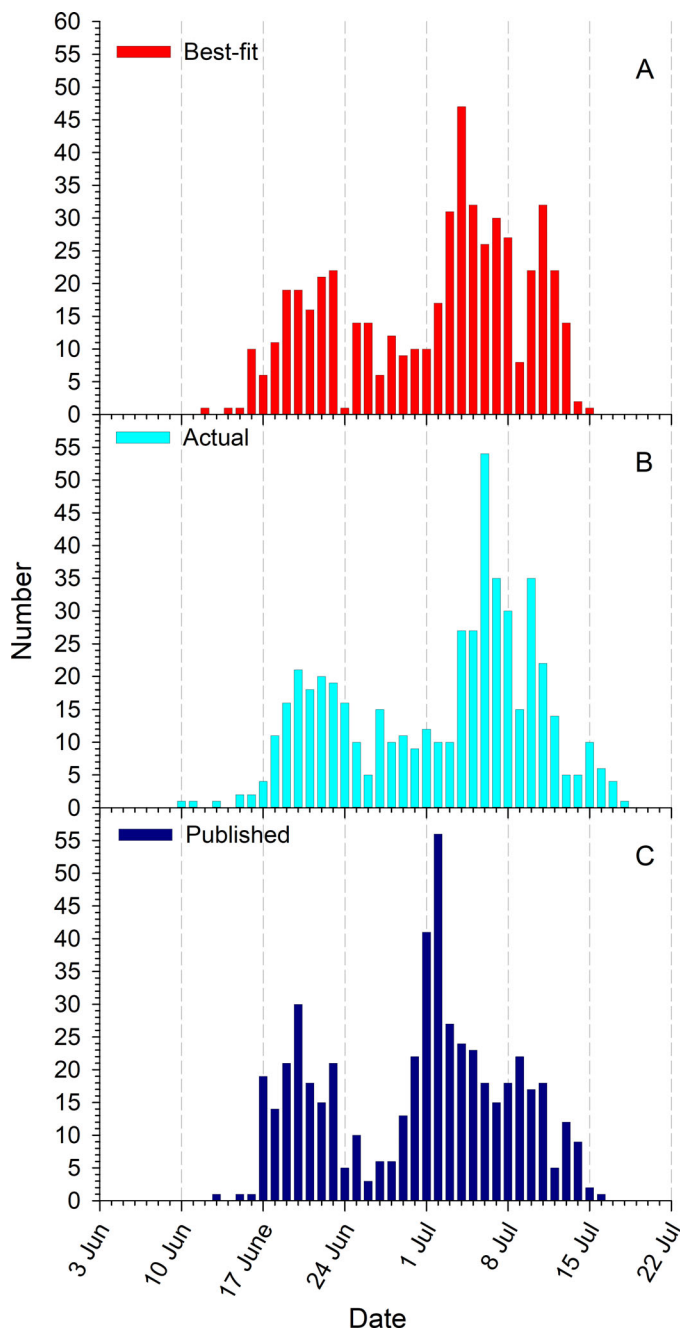


Fig. 4. Back-calculated spawn dates of Colorado Pikeminnows produced from otolith ages (Plot B: Actual), from the transformed linear equations (Plot A: Best-fit), and from the published function (Nesler et al., 1988; Plot C: Published). Data are from larvae collected from 2010 to 2017 in the San Juan River. Dashed gray vertical bars denote weekly intervals.

and Muth, 2004). Of the 113 Razorback Sucker larvae older than 30 d in the current study, most were well below 25 mm TL, and only five were >25 mm TL. Colorado Pikeminnows failed to reach 25 mm TL, though due to the rapidity with which they transitioned to larval phases, it is likely that individuals above 25 mm TL would have been in the juvenile phase (Snyder et al., 2016) and were thus excluded from this study. The largest Colorado Pikeminnow in this study, a metalarva, obtained 22.4 mm TL in just 30 d; however, many older individuals present in the study were smaller. Snyder et al. (2016) reported larval Colorado Pikeminnow transitioned to juvenile ontogenetic phases at approximately 25 mm TL.

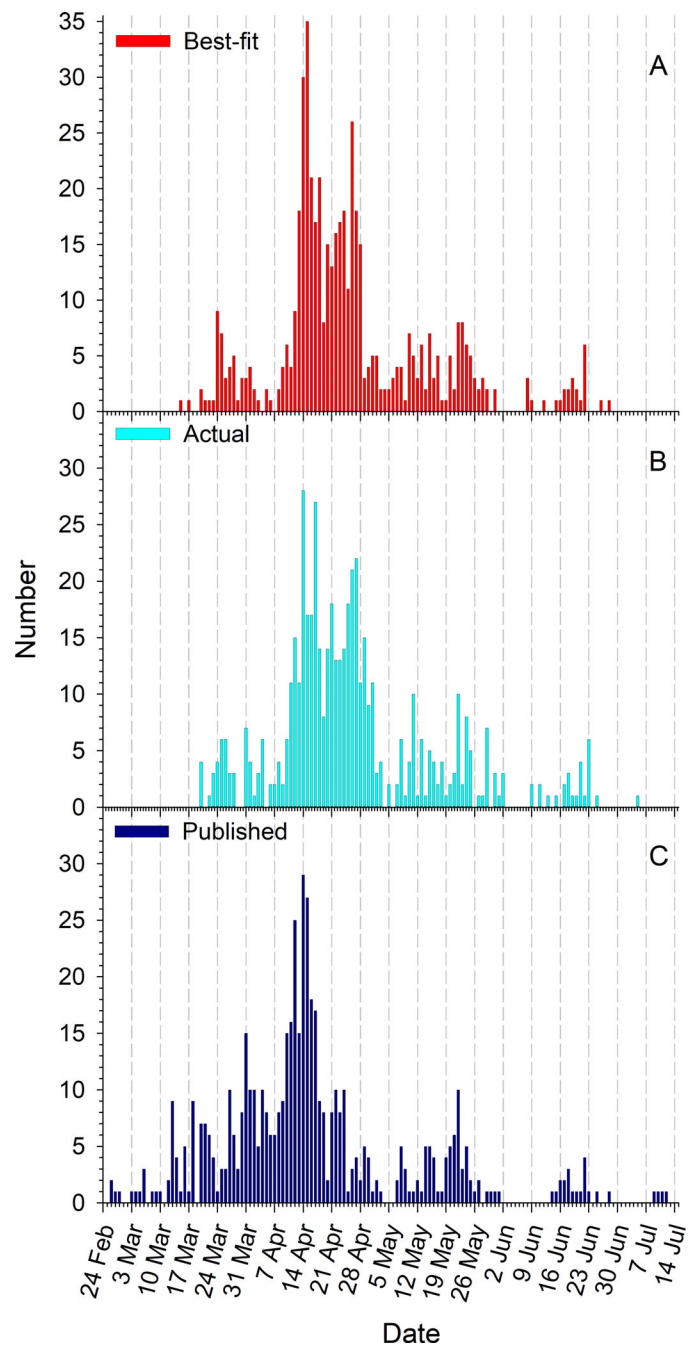


Fig. 5. Back-calculated spawn dates of Razorback Suckers produced from otolith ages (Plot B: Actual), from the linear equation (Plot A: Best-fit), and from the published function (Bestgen et al., 2011; Plot C: Published). Data are from larvae collected from 2009 to 2017 in the San Juan River. Dashed gray vertical bars denote weekly intervals.

The rate at which larvae transition to later phases is influenced by temperature and may be delayed at cooler temperatures (Clarkson and Childs, 2000) or amplified in warmer temperatures. In Colorado Pikeminnows, rapidly transitioning protolarvae may remain in the interstitial spaces in which they hatched for several days (Nesler et al., 1988) before emerging and drifting downstream. Nesler et al. (1988) reported mostly protolarvae in drift, yet from 2009 to 2017 only one protolarva Colorado Pikeminnow was collected (Table 1). This suggests that either Colorado Pikeminnows drift as protolarvae and enter nursery habitat as flexion

Table 8. Results of multiple linear regressions analyzing impacts of abiotic and temporal variables on spawning (A) and growth (B) in larval Colorado Pikeminnows in the San Juan River.

Analysis	Covariates	β	SE	P-value
A) Spawning	Intercept	1.177×10^3	2.779×10^2	<0.001
	GDD _{spawn}	-3.143×10^{-2}	5.328×10^{-3}	<0.001
	ΔT	2.177×10^{-1}	5.682×10^{-2}	0.002
	ΔQ	3.348×10^{-2}	8.104×10^{-3}	<0.001
	Photoperiod	-1.815	4.375×10^{-2}	<0.001
	Year	2.984×10^{-1}	1.267×10^{-1}	0.019
B) Growth	Intercept	-2.570×10^{-1}	1.588×10^{-1}	0.106
	Rkm	2.547×10^{-3}	6.156×10^{-4}	<0.001
	Age	2.933×10^{-1}	8.103×10^{-3}	<0.001

mesolarvae or transition almost immediately upon arrival in low-velocity nursery habitat such that the protolarval period is too brief to capture. Conversely, relatively fewer Razorback Suckers in the metalarval ontogenetic phase were available for aging. Razorback Suckers spawn earlier in the season and larvae are exposed to cooler temperatures than Colorado Pikeminnow. Warm temperature may accelerate transitioning in Colorado Pikeminnows (Clarkson and Childs, 2000), whereas cool temperature likely has an adverse effect in Razorback Suckers leading to slower transitioning. Though temperature likely affects transitioning time in both species, Colorado Pikeminnows and Razorback Suckers have different life histories, so transitioning time may be affected by other factors as well.

Six candidate age-length functions were developed for San Juan River Colorado Pikeminnows and Razorback Suckers producing a wide range of back-calculated ages. Ultimately, the transformed linear and linear equations were the top ranked models for describing the relationship between age and length in Colorado Pikeminnow, and a linear equation was top ranked for describing the relationship in Razorback Suckers. Unlike the published equations (Nesler et al., 1988; Bestgen et al., 2011) that were previously used for Colorado Pikeminnow and Razorback Suckers in the San Juan River, the best-fit equations developed in the present study included environmental variables, thus subsequent age calculations should be resilient environmental conditions that influence larval growth. Both the transformed linear equation for Colorado Pikeminnow and the linear equation

for Razorback Suckers incorporated discharge in the mean spawning month (JulyQ or AprilQ) and Rkm indicating a thermal influence to the age-length relationship.

Relatively few San Juan River Colorado Pikeminnow larvae were collected between 2009 and 2017, so samples from a subset of years—2014, 2016, and 2017—numerically dominated the dataset used in this study. In total, 90% ($n = 464$) of Colorado Pikeminnows were collected in just three years of the nine-year sample period. These years did represent diverse abiotic conditions, and June and July (spawning months) of 2016 and 2017 had significantly lower temperature (ANOVA: $F = 11.65$, $df = 8$, $P < 0.001$) and higher discharge (ANOVA: $F = 33.49$, $df = 8$, $P < 0.001$) than did 2014. Regardless of diverse thermal and discharge conditions, if Colorado Pikeminnows spawn under different abiotic conditions than were represented in this study, supplemental evaluation of larval growth may be required.

Though incubation period is temperature dependent (Marsh, 1985; Bestgen and Williams, 1994), a constant value of 5 d has previously been applied to Colorado Pikeminnows collected in the San Juan River (Farrington et al., 2018). Despite small sample size, a temperature-dependent incubation equation was calculated for Colorado Pikeminnows and produced incubation periods of 3.30–5.49 d (mean 4.1 d). These calculated incubation periods were congruent with those observed in hatchery-produced Colorado Pikeminnows, 3.50–6.00 d (Hamman, 1981). Hamman (1981) reported Colorado Pikeminnow embryos were incubated at 20.0–24.0°C, whereas in the San Juan River, for the period of this study, embryos were incubated 15.9–26.6°C with the majority ($n = 442$ of 515) incubated at 20°C or greater. Incubation periods produced by the equation were very similar to the historical value of 5 d, so likely did not affect back-calculated spawning dates.

Otolith ages from this study have redefined spawning periodicity of both Colorado Pikeminnow and Razorback Sucker in the San Juan River; spawning in the San Juan River was consistent with timing and temperature of reproduction recorded in other drainages. In this study, San Juan River Razorback Suckers spawned earlier in the season than San Juan River Colorado Pikeminnows, at temperatures as low as 11.2°C, and spawned for a longer period, 107 d compared to 38 d for Colorado Pikeminnows. Colorado Pikeminnow typically begin spawning at temperatures 16.0–18.0°C and

Table 9. Results of multiple linear regressions analyzing impacts of abiotic and temporal variables on spawning (A) and growth (B) in larval Razorback Suckers in the San Juan River.

Analysis	Covariates	β	SE	P-value
A) Spawning	Intercept	-3.983×10^2	9.014×10^1	<0.001
	GDD _{spawn}	1.102×10^{-1}	5.198×10^{-3}	<0.001
	T _{spawn}	-1.899×10^{-1}	8.745×10^{-2}	0.030
	Q _{spawn}	5.971×10^{-2}	3.496×10^{-3}	<0.001
	Photoperiod	4.121×10^{-1}	5.920×10^{-3}	<0.001
	ΔT	2.685×10^{-1}	5.962×10^{-2}	<0.001
	ΔQ	-4.693×10^{-2}	6.115×10^{-3}	<0.001
	Year	8.829×10^{-2}	4.393×10^{-2}	0.045
	Intercept	-1.963	4.735×10^{-1}	3.96×10^{-5}
B) Growth	Rkm	-3.306×10^{-3}	7.772×10^{-4}	2.50×10^{-5}
	T _{coll}	7.883×10^{-2}	2.342×10^{-2}	8.21×10^{-4}
	Age	2.363×10^{-1}	6.970×10^{-3}	<0.001
	Q _{coll}	2.919×10^{-2}	1.163×10^{-3}	1.24×10^{-2}

from June to August (USFWS, 2002a). Spawning initiated at similar temperatures in this study, though the spawning period was slightly shorter in the San Juan River than in other rivers. Razorback Suckers are capable of spawning in riverine, lacustrine, and tributary habitats and under a range of thermal conditions (USFWS, 2002b; Bestgen et al., 2006; Albrecht et al., 2018; Cathcart et al., 2019). In riverine habitats, spawning has been recorded from April through June at a mean temperature of 15.0°C (USFWS, 2002b). Similar mean spawning temperature was recorded in the San Juan River in this study, 15.2°C, though an extended spawning period, March to July, occurred relative to other rivers.

The impact of temperature and discharge on spawning, incubation, and hatching in Colorado Pikeminnows and Razorback Suckers (Marsh, 1985; Clarkson and Childs, 2000; USFWS, 2002a, 2002b) is well known. In this study, multiple parameterizations of temperature and discharge were significant predictors of spawning dates. Colorado Pikeminnow are potadromous, historically participating in spawning migrations (Durst and Franssen, 2014; Cathcart et al., 2018) in response to cues from temperature, discharge, and photoperiod (Irving and Modde, 2000). Our study supported these findings with photoperiod having a large influence on spawning date ($\beta = -1.815$) relative to the other parameters included in the model. Two different temperature covariates, GDD_{spawn} and ΔT were included in the model indicating thermal cues for spawning. The year variable was selected for inclusion in the model by AIC indicating an annual effect in spawning periodicity. Likewise, temperature and discharge are the main documented spawning cues for San Juan River Razorback Suckers (USFWS, 2002b) with photoperiod suggested as a cue (Modde and Irving, 1998). The β coefficients for the environmental covariates included in the Razorback Sucker spawning model were less different than those of Colorado Pikeminnow making it difficult to identify a primary spawning cue. Razorback Suckers have been thought to spawn on the ascending limb of the hydrograph (USFWS, 2002b), and spawning was at least in part related to changes in the hydrograph (ΔQ and Q_{spawn}), but other thermal cues (GDD_{spawn} , T_{spawn} , and ΔT) were influential as well. As with Colorado Pikeminnow, the year covariate was included in the spawning model indicating significant variation across years.

Considerable variation in growth rates was documented in both species and could not be incorporated into the functions. Though temperature typically positively influences growth of both Colorado Pikeminnows and Razorback Suckers (Clarkson and Childs, 2000; Bestgen, 2008), in this study it was responsible for relatively little of the variation in growth, and age was the dominant variable affecting growth for both species. Food availability is positively related to growth of larval riverine fishes (Schiemer et al., 2003), including Colorado Pikeminnows (Bestgen et al., 2006) and Razorback Suckers (Papoulias and Minckley, 1992), but biotic variables such as genetics and food availability are difficult to measure in wild specimens. Abiotic variables like dissolved oxygen, conductivity, or pH may influence growth rates (McDougall et al., 2018) but were not included in our analyses of growth due to challenges related to obtaining accurate point measurements. Additionally, the location at which temperature and discharge were measured likely

influenced both the effects on growth rate and the ability of the function to encompass variation in growth rate. For consistency, temperature was measured at a single point in the river, the USGS San Juan River Near Bluff Gage, necessarily ignoring longitudinal differences in temperature. Moreover, larval Colorado Pikeminnows and Razorback Suckers (Carter et al., 1986; Nesler et al., 1988) rely on a drifting stage after emergence to be moved from spawning grounds into low-velocity nursery habitat (Tyus et al., 2000; Lechner et al., 2016). This drifting life stage exposes larvae to a wide range of main stem San Juan River conditions before they move into backwater and other low-velocity habitat. It is also unlikely that a fish remains in the same low-velocity habitat for the entirety of its larval phase, so larvae from the same cohort may experience very different conditions. This life history variation makes it difficult to encompass all growth rate variation in individual larvae. Lastly, because exact hatch length of each wild larva is unknown, a mean hatch length was used for both species. A range of hatch lengths has been reported in the literature, and deviations from the mean have the potential to alter growth rate calculations.

The inaccuracy of ages and spawn dates calculated with the published functions, growth functions from this study, and otolith ages highlights the need for drainage-specific equations. Mean growth rates of San Juan River Colorado Pikeminnows (0.29 mm/d) and Razorback Suckers (0.20 mm/d) in our study were lower than published rates of 0.51 mm/d (Hamman, 1981) and 0.30 mm/d (Bestgen et al., 2011), respectively. Conversely, length-at-age of Colorado Pikeminnows was higher in San Juan River fish than in fish of the same age from other drainages in the upper Colorado River basin (Durst and Franssen, 2014). Previous functions were formulated with hatchery-produced Colorado Pikeminnow larvae and with Razorback Sucker larvae from the Green River and use of growth equations developed with hatchery-produced larvae or larvae from another basin has resulted in inaccurate estimates of ages (M. A. Farrington, pers. comm.). These growth functions had produced a close approximation of spawning periodicity in San Juan River larvae but misrepresented the period of peak spawning in both species. This inaccuracy propagates across back-calculated spawning dates and has the potential to erroneously influence management decisions regarding water release to benefit spawning fish.

Colorado Pikeminnows and Razorback Suckers in the San Juan River exist in different conditions than those in which they evolved; altered thermal and hydrologic regimes, habitat modification, and interactions with nonnative species may impede recovery efforts. Although it is difficult to disentangle and rectify threats impeding recovery, improving survival of early life stage Colorado Pikeminnow and Razorback Sucker may reduce or eliminate the survival bottleneck between larval and juvenile life stages and promote recovery of these species. Discharge in the San Juan River is largely regulated by discharge from Navajo Reservoir, which can be timed for when it is most beneficial to embryos and larvae. Improved estimates of spawning periodicity coupled with understanding of environmental drivers of spawning and growth provide resource managers with the information needed to make informed decisions for the conservation of these species.

DATA ACCESSIBILITY

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LITERATURE CITED

- Albrecht, B. A., P. B. Holden, R. B. Kegerries, and M. E. Golden. 2010. Razorback Sucker recruitment in Lake Mead, Nevada–Arizona, why here? *Lake and Reservoir Management* 26:336–344.
- Albrecht, A., H. E. Mohn, R. Kegerries, M. C. McKinstry, R. Rogers, T. Francis, B. Hines, J. Stolberg, D. Ryden, D. Elverud, B. Schleicher, K. Creighton, B. Healy, and B. Senger. 2018. Use of inflow areas in two Colorado River basin reservoirs by the endangered Razorback Sucker (*Xyrauchen texanus*). *Western North American Naturalist* 77:500–514.
- Ashworth, E. C., S. A. Hesp, and N. G. Hall. 2017. A new proportionality-based back-calculation approach, which employs traditional forms of growth equations, improves estimations of length at age. *Canadian Journal of Fisheries and Aquatic Sciences* 74:1088–1099.
- Bestgen, K. R. 2008. Effects of water temperature on growth of Razorback Sucker larvae. *Western North American Naturalist* 68:15–20.
- Bestgen, K. R., D. W. Beyers, J. A. Rice, and G. B. Haines. 2006. Factors affecting recruitment of young Colorado Pikeminnow: synthesis of predation experiments, field studies, and individual-based modeling. *Transactions of the American Fisheries Society* 135:1722–1742.
- Bestgen, K. R., and J. M. Bundy. 1998. Environmental factors affect daily increment deposition and otolith growth in young Colorado Squawfish. *Transactions of the American Fisheries Society* 127:105–117.
- Bestgen, K. R., G. B. Haines, and A. A. Hill. 2011. Synthesis of flood plain wetland information: timing of Razorback Sucker reproduction in the Green River, Utah, related to stream flow, temperature, and flood plain wetland availability. Final Report to the Upper Colorado River Endangered Fish Recovery Program, Denver. Larval Fish Laboratory Contribution 163.
- Bestgen, K. R., and M. A. Williams. 1994. Effects of fluctuating and constant temperatures on early development and survival of Colorado Squawfish. *Transactions of the American Fisheries Society* 123:574–579.
- Bliesner, R., and V. Lamarra. 2000. Hydrology, Geomorphology, and Habitat Studies. San Juan River Basin Recovery Implementation Program, USFWS, Albuquerque, New Mexico.
- Brandenburg, W. H., T. A. Francis, D. E. Snyder, K. R. Bestgen, B. A. Hines, W. D. Wilson, S. Bohn, A. S. Harrison, and S. L. Clark Barkalow. 2019. Discovery of Grass Carp larvae in the Colorado River arm of Lake Powell. *North American Journal of Fisheries Management* 39:166–171.
- Brewer, M. J., A. Butler, and S. L. Cooksley. 2016. The relative performance of AIC, AICc and BIC in the presence of unobserved heterogeneity. *Methods in Ecology and Evolution* 7:679–692.
- Bundy, J. M., and K. R. Bestgen. 2001. Confirmation of daily increment deposition in otoliths of young Razorback Suckers. *The Southwestern Naturalist* 46:34–40.
- Burnham, K. P., and D. R. Anderson. 2002. Model Selection and Multimodal Inference: A Practical Information-Theoretic Approach. Second edition. Springer, New York.
- Campana, S. E. 2001. Accuracy, precision and quality control in age determination, including a review of the use and abuse of age validation methods. *Journal of Fish Biology* 59:197–242.
- Campana, S. E., and S. R. Thorrold. 2001. Otoliths, increments, and elements: keys to a comprehensive understanding of fish populations? *Canadian Journal of Fisheries Aquatic Sciences* 58:30–38.
- Carter, J. G., V. A. Lamarra, and R. J. Ryel. 1986. Drift of larval fishes in the Upper Colorado River. *Journal of Freshwater Ecology* 3:567–577.
- Cathcart, C. N., K. B. Gido, M. C. McKinstry, and P. D. MacKinnon. 2018. Patterns of fish movement at a desert fish confluence. *Ecology of Freshwater Fish* 27:492–505.
- Cathcart, C. N., M. C. McKinstry, P. D. MacKinnon, and C. M. Ruffing. 2019. A tribute to tributaries: endangered fish distributions within critical habitat of the San Juan River, USA. *North American Journal of Fisheries Management* 39:1015–1025.
- Clark Barkalow, S. L., M. A. Brandenburg, and S. P. Platania. 2020. Otoliths reveal spawning ecology and early life history of sympatric catostomids. *North American Journal of Fisheries Management* 40:415–426.
- Clarkson, R. W., and M. R. Childs. 2000. Temperature effects of hypolimnial-release dams on early life stages of Colorado River basin big-river fishes. *Copeia* 2000:402–412.
- Daoud, J. I. 2017. Multicollinearity and regression analysis. *Journal of Physics Conference Series* 949:012009.
- Durst, S. L., and N. R. Franssen. 2014. Movement and growth of juvenile Colorado Pikeminnows in the San Juan

- River, Colorado, New Mexico, and Utah. Transactions of the American Fisheries Society 143:519–527.
- Farrington, M. A., R. K. Dudley, J. L. Kennedy, S. P. Platania, and G. C. White.** 2018. Colorado Pikeminnow and Razorback Sucker larval fish survey in the San Juan River during 2017. San Juan River Basin Recovery Implementation Program, USFWS, Albuquerque, New Mexico.
- Hamman, R. L.** 1981. Spawning and culture of Colorado Squawfish in raceways. Progressive Fish Culturist 43:173–177.
- Hill, A. A., and K. R. Bestgen.** 2014. Otolith daily increment deposition in age-0 Smallmouth Bass reared in constant and fluctuating water. North American Journal of Fisheries Management 34:774–779.
- Hoff, G. R., D. J. Logan, and D. F. Markle.** 1997. Otolith morphology and increment validation in young Lost River and Shortnose Suckers. Transactions of the American Fisheries Society 126:488–494.
- Humphries, P., and P. S. Lake.** 2000. Fish larvae and the management of regulated rivers. River Research and Applications 16:421–432.
- Irving, D. B., and T. Modde.** 2000. Home-range fidelity and use of historic habitat by adult Colorado Pikeminnow (*Ptychocheilus lucius*) in the White River, Colorado and Utah. Western North American Naturalist 60:16–25.
- Jolley, J. C., K. R. Edwards, and D. W. Willis.** 2009. Bluegill (*Lepomis macrochirus*) spawning periodicity and hatching duration in the northern Great Plains, USA. Journal of Freshwater Ecology 24:29–38.
- Kabacoff, R. I.** 2011. R in action, data analysis and graphics with R. Manning Publications Co., Shelter Island, New York.
- Katsanevakis, S., and D. Maravelias.** 2008. Modeling fish growth: multi model inference as a better alternative to *a priori* using von Bertalanffy equation. Fish and Fisheries 9: 178–187.
- Lechner, A., H. Keckeis, and P. Humphries.** 2016. Patterns and processes in the drift of early developmental stages of fish in rivers: a review. Reviews in Fish Biology and Fisheries 26:471–489.
- Marsh, P. C.** 1985. Effect of incubation temperature on survival of embryos of native Colorado River fishes. The Southwestern Naturalist 30:129–140.
- McDougall, C. A., P. A. Nelson, and C. C. Barth.** 2018. Extrinsic factors influencing somatic growth of Lake Sturgeon. Transactions of the American Fisheries Society 147:459–479.
- Miller, S. J., and R. Storck.** 1984. Temporal spawning distribution of Largemouth Bass and young-of-the-year growth, determined from daily otolith rings. Transactions of the American Fisheries Society 113:571–578.
- Miller, T. J., L. B. Crowder, J. A. Rice, and E. A. Marschall.** 1988. Larval size and recruitment mechanisms in fishes: toward a conceptual framework. Canadian Journal of Fisheries and Aquatic Sciences 45:1657–1670.
- Modde, T., and D. B. Irving.** 1998. Use of multiple spawning sites and seasonal movement by Razorback Suckers in the Middle Green River, Utah. North American Journal of Fisheries Management 18:318–326.
- Montgomery, D. C., E. A. Peck, and G. G. Vining.** 2012. Introduction to Linear Regression Analysis. Fifth edition. Wiley, New York.
- Murphy, S. C., N. C. Collins, and S. E. Doka.** 2011. Thermal habitat characteristics for warmwater fishes in coastal embayments of Lake Ontario. Journal of Great Lakes Research 37:111–123.
- Nesler, T. P., R. T. Muth, and A. Wasowicz.** 1988. Evidence for baseline flow spikes as spawning cues for Colorado Squawfish in the Yampa River, Colorado, p. 68–79. *In*: 11th Annual Larval Fish Conference. R. D. Hoyt (ed.). American Fisheries Society, Symposium 5, Bethesda, Maryland.
- Neuheimer, A. B., and C. T. Taggart.** 2007. The growing degree-day and fish size-at-age: the overlooked metric. Canadian Journal of Fisheries and Aquatic Sciences 64: 375–385.
- Ogle, D. H., P. Wheeler, and A. Dinno.** 2019. FSA: Fisheries Stock Analysis. R package version 0.8.26. <https://github.com/droglenc/FSA>
- Papoulias, D., and W. L. Minckley.** 1992. Effects of food availability on survival and growth of larval Razorback Sucker ponds. Transactions of the American Fisheries Society 121:340–355.
- Pennock, C. A., M. A. Farrington, and K. B. Gido.** 2019. Feeding ecology of early life stage Razorback Sucker relative to other sucker species in the San Juan River, Utah. Transactions of the American Fisheries Society 148:938–951.
- Propst, D. L., and K. B. Gido.** 2004. Responses of native and nonnative fishes to natural flow regime mimicry in the San Juan River. Transactions of the American Fisheries Society 133:922–931.
- R Core Team.** 2016. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>
- Sabaj, M. H.** 2020. Codes for natural history collections in ichthyology and herpetology. Copeia 108:593–669.
- Schiemer, F., H. Keckeis, and E. Kamler.** 2003. The early life history stages of riverine fish: ecophysiological and environmental bottlenecks. Comparative Biochemistry and Physiology Part A 133:439–449.
- Sibley, R. M., J. Baker, J. M. Grady, S. M. Luna, A. Kodric-Brown, C. Venditti, and J. H. Brown.** 2015. Fundamental insights into ontogenetic growth from theory and fish. Proceedings of the National Academy of Sciences of the United States of America 112:13934–13939.
- SJRBRIP (San Juan River Basin Recovery Implementation Program).** 2018. Final Program Document. San Juan River Basin Recovery Implementation Program, USFWS, Albuquerque, New Mexico.
- Snyder, D. E., and R. T. Muth.** 2004. Catostomid fish larvae and early juveniles of the upper Colorado River basin—morphological descriptions, comparisons, and computer-interactive key. Colorado Parks and Wildlife Technical Publication 38.
- Snyder, D. E., S. C. Seal, J. A. Charles, and C. L. Bjork.** 2016. Cyprinid fish larvae and early juveniles of the upper Colorado River basin—morphological descriptions, comparisons, and computer-interactive key. Colorado Parks and Wildlife Technical Publication 47.
- Sponagule, S.** 2009. Daily otolith increments in early stages of tropical fish, p. 93–132. *In*: Tropical Fish Otoliths: Information for Assessment, Management and Ecology. B. S. Green, B. D. Mapstone, G. Carlos, G. A. Begg, and J. L. Nielsen (eds.). Springer, New York.

- Symonds, M. R., and A. Moussalli.** 2011. A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behavioral Ecology and Sociobiology* 65:13–21.
- Tyus, H. M., C. W. Brown, and J. F. Saunders, III.** 2000. Movements of young Colorado Pikeminnow and Razorback Sucker in response to water flow and light level. *Journal of Freshwater Ecology* 15:525–535.
- UFR (Use of Fishes in Research) Committee.** 2013. Guidelines for the use of fishes in research. American Fisheries Society, Bethesda, Maryland.
- USFWS (U.S. Fish and Wildlife Service).** 2002a. Colorado Pikeminnow (*Ptychocheilus lucius*) Recovery Goals: Amendment and Supplement to the Colorado Squawfish Recovery Plan. U.S. Fish and Wildlife Service, Mountain-Prairie Region (6), Denver, Colorado.
- USFWS (U.S. Fish and Wildlife Service).** 2002b. Razorback Sucker (*Xyrauchen texanus*) Recovery Goals: Amendment and Supplement to the Razorback Sucker Recovery Plan. U.S. Fish and Wildlife Service, Mountain-Prairie Region (6), Denver, Colorado.
- Venturelli, P. A., N. P. Lesterm T. R. Marshall, and B. J. Shuter.** 2010. Consistent patterns of maturity and density-dependent growth among populations of walleye (*Sander vitreus*): application of the growing degree-day metric. *Canadian Journal of Fisheries and Aquatic Sciences* 67: 1057–1067.
- Victor, B. C., and E. B. Brothers.** 1982. Age and growth of the Fallfish *Semotilus corporalis* with daily otolith increments as a method of annulus verification. *Canadian Journal of Zoology* 60:2543–2550.
- Vilizzi, L., and K. F. Walker.** 1999. Age and growth of the Common Carp, *Cyprinus carpio*, in the River Murray, Australia: validation, consistency of age interpretation, and growth models. *Environmental Biology of Fishes* 54: 77–106.
- Webber, P. A., and G. B. Haines.** 2014. Survival and growth rates of stocked Razorback Suckers (*Xyrauchen texanus*) in a wetland of the Green River, Utah. *The Southwestern Naturalist* 59:459–464.
- Yamashita, T., K. Yamashita, and R. Kamimura.** 2007. A stepwise AIC method for variable selection in linear regression. *Communications in Statistics – Theory and Methods* 36:2395–2403.
- Zweifel, J. R., and R. Lasker.** 1976. Prehatch and posthatch growth of fishes—a general model. *Fishery Bulletin* 74: 609–621.