

Comparing aging precision of calcified structures in shovelnose sturgeon

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Summary

Age estimates for population analysis must be precise. We assessed the usefulness of pectoral fin rays, sphenoids, opercula, and dorsal scutes of shovelnose sturgeon *Scaphirhynchus platyrhynchus* ($n = 30$) as aging structures based on ease of collection, distinctness of annuli, and measures of precision both between and within readers. We also determined how age estimates from paired fin rays of individuals were related ($n = 106$). Pectoral fin rays generated the highest within-reader precision (100% within 2 years) followed by sphenoids (58%), opercula (56%), and dorsal scutes (49%). Ages estimated by the pectoral fin ray also had higher between-reader agreement (80% within 1 year) than did those from the operculum (60%), sphenoid (59%), or dorsal scute (56%). Likewise, age estimates from pectoral fin rays had the lowest mean coefficient of variation (8.2%) followed by sphenoids (9.9%), opercula (11.3%), and dorsal scutes (11.5%). Only the operculum produced biased estimates between readers. Ages from paired fin rays agreed poorly (36% exact, 30% within 1 year) although no aging bias occurred. The pectoral fin ray is typically used to age shovelnose sturgeon. Because uncertainty about accuracy and precision of age estimates from this structure remains, shovelnose sturgeon management objectives that result from age data should remain conservative.

Introduction

In recent years, interest in the shovelnose sturgeon, *Scaphirhynchus platyrhynchus*, has increased. It is one of eight North American acipenseriforms, three of which are confined to the large rivers of the interior United States. Like many of its Asian and European counterparts, the shovelnose sturgeon range and population size have declined since the late 1800s (Carlander, 1954; Birstein, 1993; Keenlyne, 1997). These declines may be due to habitat change, but commercial harvest has potentially resulted in a decline in shovelnose sturgeon numbers as well (Morrow et al., 1998; Quist et al., 2002).

The shovelnose sturgeon is harvested primarily for its eggs, which have increased in demand and value in recent years. Commercial fishers sell processed shovelnose sturgeon eggs for as much as US\$44 kg⁻¹ and retail for \$462–714 kg⁻¹, depending on market demand (Whiteman et al., 2004). Furthermore, as stocks of sturgeon overseas dwindle, continued growth of the North American caviar market is evident. In recent years, the reported sale of *S. platyrhynchus* eggs in Illinois has increased dramatically (Maher, 2001) and is likely increasing basin-wide (Morrow et al., 1998).

Due to the increased concern over North American sturgeon stocks, midwestern states are in the process of implementing a basin-wide regulation aimed at protecting the shovelnose sturgeon from over-harvest. To understand the impact of that regulation, fishery managers will monitor population parameters such as age structure, growth, and mortality based on age estimates taken from pectoral fin rays. While pectoral fin rays have been widely used in estimating ages of shovelnose sturgeon (Whiteman et al., 2004), other bony structures such as sphenoids, opercula and dorsal scutes have been used as aging structures in other sturgeon species (Brennan and Cailliet, 1989). Comparisons of shovelnose sturgeon aging structures have not been previously reported; therefore, we assessed the relative precision of pectoral fin rays, sphenoids, opercula, and dorsal scutes as aging structures. Precise estimates of age are necessary to avoid systematic errors in estimates of mortality, growth, and population trajectory (Quinn and Deriso, 1999).

Materials and methods

Shovelnose sturgeon were collected from the Middle Mississippi River (MMR), between the confluence of the Missouri River near St Louis, MO and the Ohio River at Cairo, IL during spring through summer 2000. Trammel nets used to catch sturgeon were 91.4-m long and 1.8-m deep with alternating 22.9-m panels of 2.5 cm and 5.1 cm inner mesh and outer mesh of 30.5 cm. Nets were drifted over sandbars on inside bends and between dikes where sturgeon are commonly found.

Upon capture, sturgeon were placed on ice and returned to the laboratory. All sturgeon were measured and weighed, and the left and right anterior pectoral fin rays were removed. The opercula, sphenoids and fourth dorsal scutes were removed from a subsample of 30 shovelnose sturgeon. All fin rays were placed in coin envelopes and allowed to dry. Other structures were boiled in water for approximately 30 min and scrubbed with a brush to remove excess flesh before being placed in envelopes for drying. Cross-sections of 0.6 mm were taken from fin rays, dorsal scutes and sphenoids whereas opercula were aged whole. Cross-sectioned structures were aged with a binocular microscope at 250× magnification with transmitted light. Opercula were aged with the same microscope at 60× magnification. Cross-sectioned structures were submerged in glycerin to better elucidate growth rings. However, the growth rings of opercula were better resolved when placed in water.

Although the timing of annulus deposition likely varies in shovelnose sturgeon, Whiteman et al. (2004) showed that a single opaque band is laid down in the pectoral fin ray starting in May. This supports the previous assumption in most

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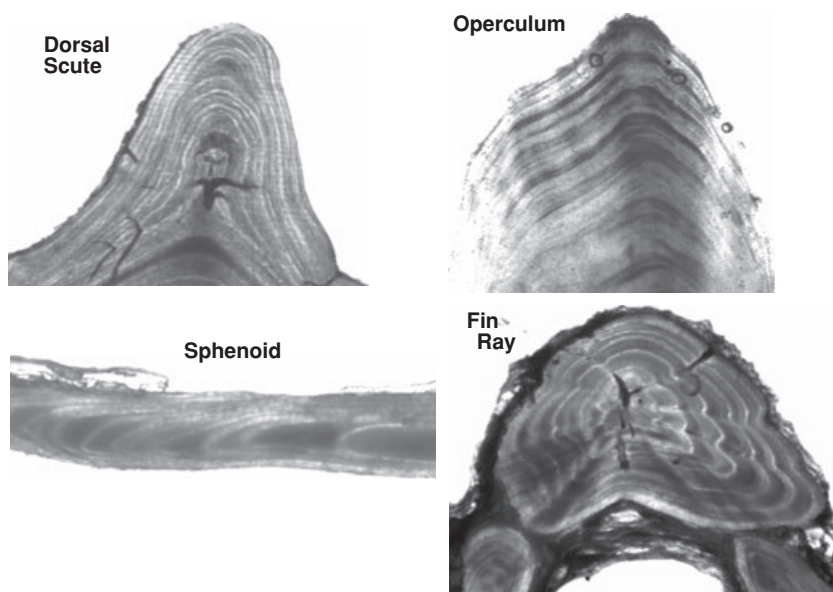


Fig. 1. Magnified images of structures used in aging shovelnose sturgeon (*Scaphirhynchus platyrhynchus*). Dorsal scutes, sphenoids, and fin rays were cross-sectioned to 0.6 mm and viewed under 250× magnification. Opercula were aged whole in water under 60× magnification

sturgeon aging studies that a pair of translucent and opaque rings is formed annually (Brennan, 1988). Under transmitted light, the translucent ring (winter growth) appears bright while the opaque ring (summer growth) is usually wider and appears dark in color (Fig. 1). Thus, the aging criteria established prior to aging were, (i) each pair of opaque and translucent rings was counted as an annulus and (ii) only distinct and continuous annuli were counted. Both readers aged each structure blindly before the identification numbers were coded and rearranged to eliminate any reader biases. The structures were then aged blindly a second time to obtain an estimate of repeatability. After aging, each structure was measured along the axis in which annuli were counted.

We assessed the usefulness of four aging structures based on ease of collection, distinctness of annuli, and measures of precision both between and within readers. Measures of precision also were calculated for paired age estimates taken from left and right pectoral fin rays. Percent agreement and the coefficient of variation (CV) were used as indices of precision (Chang, 1982). The CV is expressed as the ratio of the standard deviation of age estimates to the mean and can be written as:

$$CV_j = 100 \times \frac{\sqrt{\frac{\sum_{i=1}^R (X_{ij} - X_j)^2}{R-1}}}{X_j},$$

where X_{ij} is the i th age determination of the j th fish, X_j is the mean age of the j th fish, and R is the number of times each fish is aged. The resulting value represents the CV of the age estimates for a single fish (j th fish). We averaged individual CVs across fish to produce a mean CV for each structure. Differences in CV between structures were tested using a non-parametric Kruskal–Wallis test.

The relationship between the size of each structure and estimated age was analyzed using linear regression. Age bias plots were created by graphing the mean estimated age of one set of age estimates by each age class of the second set of age estimates. These data were analyzed using linear regression. If agreement were perfect between sets of age estimates, the slope of the regression line would equal one. We tested the null hypothesis that each slope was equal to one using a t -test. This

method is useful for assessing aging bias between readers, within readers, and between paired structures.

Results

A total of 136 shovelnose sturgeon were captured for age analysis. The shovelnose sturgeon ($n = 30$) used in age structure analysis had a mean fork length of 610 mm ($SD = 45.4$, range = 527–667) and a mean weight of 793 g ($SD = 187.9$, range = 399–1114). The mean fork length and weight of shovelnose sturgeon from which only left and right anterior pectoral fin rays were removed ($n = 106$) were 517 mm ($SD = 154$, range = 167–770) and 689 g ($SD = 368.6$, range = 160–1509), respectively. The size of each aging structure increased with increasing fork length (Table 1), but precision differed among aging methods.

Ages ranged from 5 to 20 years. Anterior pectoral fin rays had the highest within-reader precision (100% agreement within 2 years) followed by sphenoids (58%), opercula (56%), and dorsal scutes (49%). Percent agreement between readers within 2 years ranged from 76% to 81% for all four structures. However, the pectoral fin ray had a higher percent agreement within 1 year (80%) than did the operculum (60%), sphenoid (59%), or dorsal scute (56%). There were no significant differences in CV between structures (Kruskal–Wallis, $P = 0.5138$, Fig. 2). The slope of the age bias regression for opercula was significantly different from one (Table 2). Slopes of the age bias regressions for dorsal scutes, sphenoids, and pectoral fin rays showed no difference (Table 2).

Table 1

Summary of results of linear regressions analyzing relationship between fork length and size of calcified structures used in age estimation of shovelnose sturgeon captured in Mississippi River in spring 2000

Structure	R^2	P-value	Intercept	Slope	n
Fin ray	0.28	0.01	−0.2	0.005	30
Dorsal scute	0.45	0.01	−2.4	0.05	30
Operculum	0.71	<0.0001	−10.6	0.08	30
Sphenoid	0.72	<0.0001	−0.6	0.03	30

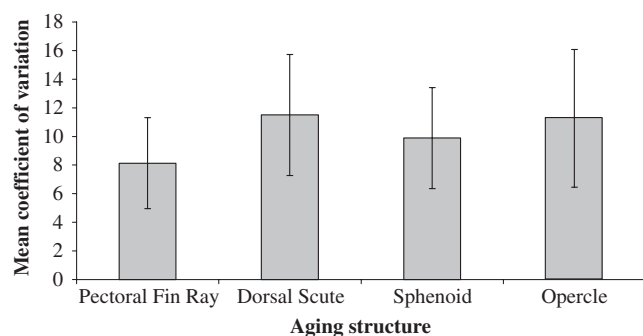


Fig. 2. Mean coefficient of variation for each structure used in aging shovelnose sturgeon *Scaphirhynchus platyrhynchus*. Error bars indicate 95% confidence intervals. No significant difference was found between structures (Kruskal–Wallis $P = 0.5138$)

Table 2

Results of linear regression for independent age estimates between readers one and two for whole opercula and sectional dorsal scutes, sphenoids, and anterior pectoral fin rays of shovelnose sturgeon captured during spring 2000 in the Mississippi River

Structure	Slope	Intercept	R ²	P slope	N
Opercula	0.43	5.71	0.52	0.01	25
Scutes	1.00	-0.22	0.82	NS*	25
Sphenoids	0.96	0.58	0.73	NS	24
Fin rays	0.85	1.52	0.89	NS	29

P-value is a result of a t-test testing the null hypothesis that the slope of each regression is equal to one. Only structures that could be aged by both readers were included in the analysis.

*NS: $P > 0.20$.

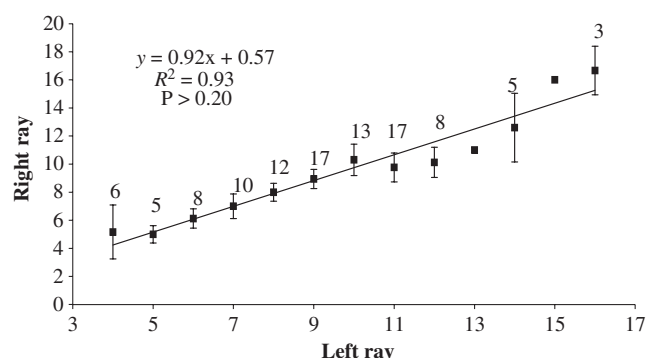


Fig. 3. Linear regression analysis of age bias between paired fin rays from 106 shovelnose sturgeon (*Scaphirhynchus platyrhynchus*) captured in Mississippi River in spring 2000. Age estimates plotted as the mean estimated age for the left fin ray for 13 age classes of the right fin ray. Error bars indicate 95% confidence intervals. Numbers above error bars indicate sample size. Those data points lacking error bars have a sample size of one. P-value result of t-test testing null hypothesis that slope equal to 1

Age estimates taken from paired fin rays ranged from 4 to 17 years. The estimates revealed an exact per cent agreement between fin rays of 36%, agreement within 1 year of 30%, and agreement within 2 years of 18%. There was little aging bias between ages taken from left and right pectoral fin rays as shown by the age bias graph (Fig. 3).

Discussion

When assessing methods for age determination in fishes, it is important to consider both accuracy and precision. Accuracy is the ability of a reader to estimate the true age of an

individual, whereas precision is the ability of a reader to reproduce age estimates in consecutive readings from an individual fish. Imprecise age estimates suggest variability in aging criteria or the presence of unclear annuli that are difficult to distinguish and count, while inaccurate age estimates bias population parameters such as growth and mortality (Quinn and Deriso, 1999). Both accuracy and precision can adversely affect our ability to estimate the age structure of a population, which can lead to mismanagement of long-lived species (Beamish and McFarlane, 1983).

Aging precision of all four structures assessed in this study varied. Anterior pectoral fin rays provided the best overall structure for estimating ages of shovelnose sturgeon compared to sphenoids, opercula and dorsal scutes. Marginal pectoral fin rays are the most practical aging structure because their removal is non-deleterious (Collins and Smith, 1996). They also were most precise in terms of within- and between-reader agreement as well as CV. In a study comparing aging structures for white sturgeon, *Acipenser transmontanus*, Brennan and Cailliet (1989) cited consistency in aging and ease of collection as reasons for choosing pectoral fin rays over five other calcified structures. They found 84%, 53%, and 74% between reader agreement within 2 years for pectoral fin sections, dorsal scutes and opercula, respectively. Although our results were similar for shovelnose sturgeon, we found that disagreements of 2 years between readers occurred less often for pectoral fin rays (1%) than for other structures (16–20%). The pectoral fin ray also showed the highest between-reader precision as indicated by the CV, likely because of the clarity and spacing of annuli. For example, annuli on pectoral fin rays had more distinct clear and opaque zones resulting in easier identification and counting of annuli. Another factor that may have contributed to these results is reader experience. Both readers had more experience aging fin ray sections than any of the other three structures.

Despite the call for more statistically robust and consistent analysis of aging data in recent years (Beamish and Fournier, 1981; Chang, 1982; Campana et al., 1994), many studies report population data (i.e. length-at-age and mortality) without mention of the precision of their age estimates. This is the first study in which shovelnose sturgeon age data has been reported using the coefficient of variation in hopes that these data will be useful in future comparisons. Unaccounted error in aging structures will translate to increased uncertainty in estimates of mortality and growth, which are used to evaluate responses to regulations.

Age bias plots revealed that there was no significant bias in age estimates taken from dorsal scutes, sphenoids and pectoral fin rays between readers. However, opercula showed extreme aging bias whereby the second reader over-aged younger fish and under-aged older fish relative to the first reader. These results may be a function of reader training such that each reader recognized annuli differently. However, Kohlhorst et al. (1980) failed to increase between-reader aging precision by setting criteria for annulus determination in white sturgeon. Thus, these results are likely a consequence of differences in the methods used to prepare structures for aging. Opercula were the only structures aged whole. The other structures were cross-sectioned, which revealed relatively distinct annuli under transmitted light. However, attempts to cross-section opercula were difficult and failed to distinguish clear annuli. Because the opercula were aged whole, transmitted light did not as effectively distinguish the annuli as in cross-sections from other structures.

Whiteman et al. (2004) found that the timing of annulus formation varies among shovelnose sturgeon and recommended mark-recapture as another way to assess the accuracy of age estimates taken from pectoral fin rays. Previous attempts to validate aging methods using this technique have had mixed results. Rien and Beamesderfer (1994) attempted to validate aging techniques for white sturgeon by determining the number of years-at-large after sturgeon were marked with OTC, released and re-captured. They found that the estimated age was equal to the known age 28–46% of the time for fish that spent from 1 to 3 years at large. In contrast, 20 marked and recaptured lake sturgeon revealed 85–100% agreement between estimated age and the number of years-at-large (Rossiter et al., 1995). In order for this age validation method to be successful, annuli must be laid down at equal intervals on both the left and right pectoral fin ray, and the correct age interpreted from the annuli. In this study, age estimates taken from paired fin rays showed no age bias, but per cent agreement between paired fin rays from shovelnose sturgeon was low (36% exact agreement, 84% within 3 years). Thus, generating accurate age estimates from paired fin rays is not promising.

While this study shows that pectoral fin rays are the overall preferred aging structure, there is still a considerable amount of error involved with this aging technique. Furthermore, we have found that the reproduction of shovelnose sturgeon age estimates is difficult for even the most experienced readers. Subtle differences in the methods used in preparing fin rays for aging may affect the reproducibility of age estimates. Thus, aging methods need to be standardized across management and research agencies; assessing the impact of uncertainty around resulting population parameters (e.g. using model sensitivity analysis; Coggins and Quinn, 1999) is advised.

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