North American Journal of Fisheries Management 33:994–999, 2013

ISSN: 0275-5947 print / 1548-8675 online
DOI: 10.1080/02755947.2013.822445

ARTICLE

A Comparison of Different Age Estimation Methods for the Northern Snakehead

Pang-hua Gu
College of Animal Science and Technology, Hunan Agricultural University, Changsha, Hunan 410128, China

Jian-guo Xiang*
College of Animal Science and Technology, Hunan Agricultural University, Changsha, Hunan 410128, China; and State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan, Hubei 430072, China

Yi-fang Chen, Yan-liang Li, and Jun Tang
College of Animal Science and Technology, Hunan Agricultural University, Changsha, Hunan 410128, China

Song-guang Xie
State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan, Hubei 430072, China

Yushun Chen*
Aquaculture and Fisheries Center, University of Arkansas, 1200 North University Drive, Pine Bluff, Arkansas 71601, USA

Abstract
We used otoliths, scales, pectoral fins, dorsal fins, basioccipital bones, opercular bones, interopercular bones, vertebrae, teeth, and cleithra sampled from 221 known-age Northern Snakeheads *Channa argus argus* representing five age-groups to determine and compare age estimation methods for this species. Results showed that accuracy of age estimation methods varied among structures: 85.68% of otoliths, 68.91% of scales, 49.25% of pectoral fin rays, and 27.67% of opercular bones yielded age estimates that were consistent with the actual fish ages. Ages determined from other structures, such as interopercular bones, dorsal fins, basioccipital bones, and vertebrae, had lower rates of agreement (<20%) with actual ages. The current results suggest that age estimation with otoliths, especially sagittal otoliths, is the most accurate method for use with Northern Snakeheads. Furthermore, analysis of scales can be used as a supplementary approach, especially for younger (age < 3) Northern Snakeheads.

The Northern Snakehead *Channa argus argus* is one of the most widespread benthic carnivorous fishes (Meng 1989). The life span of this fish is relatively short; Northern Snakeheads naturally reach the period of senescence after 5 years' growth, so it is usually difficult to catch fish older than age 5 in the wild (Zhang et al. 1999). In China, the Northern Snakehead has been a favorite seafood fish because of its tasty meat, and it has been used as a top predator in ecosystem biomanipulation. In the USA, the Northern Snakehead became known as "Frankenfish" when it was discovered in a Maryland pond in 2002, having

*Corresponding authors: xjg68102000@aliyun.com; ychen@uaex.edu
Received March 14, 2013; accepted June 27, 2013
Published online September 25, 2013

already destroyed the native aquatic ecosystem and biological diversity (Cunningham 2005).

It is necessary to develop an accurate age estimation method for managing Northern Snakeheads in natural water bodies since different age estimation methods may vary in precision (Campana 2001; Zymonas and McMahon 2009; Kowalewski et al. 2012). Some methods require ages from several structures to be compared for more accurate estimation (Boxrucker 1986; Welch et al. 1993; Isermann et al. 2010), and the availability of known-age reference fish is usually very helpful for these comparisons (Buckmeier 2002; Maceina et al. 2007). Moreover, the timing of first annulus formation in Northern Snakeheads has not been reported in recent years.

Scales have long been considered one of the most reliable structures for use in fish age estimation, and the precision of age estimates from scales can be similar to the precision from otoliths for some species (e.g., Kruse et al. 1993). To our knowledge, most previous studies have only used the scale method for aging Northern Snakeheads (e.g., Zhu and Liang 1999; Yu et al. 2008). However, we have found no study evaluating whether the scale method is the most accurate approach relative to other methods such as otolith analysis, which is widely used in age determination for other fishes.

Because otoliths are within the fish's body, the information contained in otoliths is resistant to disturbance by external environmental factors. This is in contrast to scales, which have direct exposure to the environment, making the rings on scales too complex to identify in some situations. Otoliths possess some advantageous characteristics, such as sustained growth and a low probability of being absorbed. For example, Hoxmeier et al. (2001) found that for Bluegills *Lepomis macrochirus* in Illinois reservoirs, ages estimated from otoliths were more precise than those estimated from scales. In the present study, we hypothesized that for Northern Snakeheads, the otolith method would provide more reliable and more accurate age estimates than scales.

Other calcified structures can also be used to estimate fish age. For example, vertebral bones were used to estimate ages of Shortfin Mako *Isurus oxyrinchus* (Ribot-Carballal et al. 2005). Polat et al. (2001) found that in comparison with otoliths and scales, vertebrae were the most reliable structure for determining the age of European Flounder *Pleuronectes flesus luscus*. Furthermore, Wang and Dai (2006) used scales, opercular bones, dorsal fin spines, vertebrae, and cleithra to estimate age in the cyprinid fish *Tor* (*Folifer*) *brevifilis brevifilis* and found that among all pairs of structures, the combination of scales and opercular bones had the highest tally rate and yielded the most reliable predictions of fish age. In addition, pectoral fins, basioccipital bones, teeth, and other hard structures can also be used for age estimation. To our knowledge, no previous study has compared different age estimation methods for use with Northern Snakeheads. Thus, in the current study, we examined otoliths, scales, pectoral fin rays, dorsal fin rays, opercular bones, interopercular bones, basioccipital bones, vertebrae, teeth, and cleithra from known-age Northern Snakeheads to compare the various age estimation methods for this species.

METHODS

Experimental fish.—Samples of known-age Northern Snakeheads were collected from farming ponds at a field research station near Yuanjiang City, Hunan Province, China (101°39′E, 28°46′N). The samples ($n = 221$ fish) were collected in March, April, May, June, and December 2009 (Northern Hemisphere Time; Table 1) and included five different age-groups: age 0, age 1, age 2, age 3, and age 4 and older (age 4+; including five age-4 fish, two age-5 fish, and one age-6 fish). We removed every age structure from each fish to observe and compare the annual rings in the laboratory. Two trained readers, who did not have any age information for each specimen, examined the aging structures twice within a 20-d interval. Unreadable structures or the results of a sample that showed an irreconcilable difference between the two readers were considered as disagreeing with the actual fish age.

Otoliths.—We followed standard methods to handle the various structures (DeVries and Frie 1996; Campana 2001). Otoliths were removed, cleaned, immersed in xylene for approximately 5 min, and examined with a dissecting microscope. Otoliths with unclear annual rings were ground with sandpaper to make the annuli more distinct for age reading (Tandon and Johal 1996). The first one or two increments deposited on the otolith were often broader and less distinct than subsequent increments (Gunn et al. 2008). An annulus is the translucent margin at the end of 1 year's growth (Xie and Watanabe 2005).

Scales.—Scales were removed from three locations on the fish's body: at the base of the pectoral fin (front area), above the lateral line but below the first dorsal fin (middle area), and above the lateral line but below the last dorsal fin (back area). Over 10 scales from each site on each fish were cleaned and dried in the laboratory. Of these, 8–10 scales were pressed on glass slides and examined under a microscope (DeVries and Frie 1996).

Other structures.—The leading pectoral or dorsal fin rays were excised as close to the body as possible by using

TABLE 1. Sampling dates and corresponding numbers of Northern Snakeheads that were sampled for a comparison of age estimation methods (fish were considered age 0 before January 1 and subsequently were considered age 1).

Date	Age	*n*
7 Mar	1	36
10 Apr	1	33
4 May	1	30
10 Jun	1	28
8 Dec	0	26
8 Dec	4+	8
11 Dec	1	24
14 Dec	2	21
15 Dec	3	15

dissecting scissors. Whole or proximal portions of fin rays were set in epoxy and sectioned (0.5–0.7 mm) on the transverse plane by using a slow-speed saw. If necessary, fin ray sections were ground wet or dry with 600-grit sandpaper (Sylvester and Berry 2006). Sections for each fin ray were pressed on microscope slides and aged under a dissecting microscope (Buckmeier et al. 2002). The opercular bones, interopercular bones, vertebrae, basioccipital bones, teeth, and cleithra were removed from fish that had been boiled in water for 2–3 min. These structures were boiled in 1% NaOH solution for 5–10 min, washed clean, dried in air, and then observed under a microscope (Hua et al. 2005).

Data processing.—We used percent agreement and the coefficient of variation (CV = [SD/mean] × 100) to evaluate the accuracy and precision of estimated ages within and between the two trained readers (Chang 1982; Campana et al. 1995; Hoxmeier et al. 2001; Blackwell and Kaufman 2012). Percent agreement was calculated as $(A/n) \times 100$, where A is the number of fish with an estimated age (i.e., from a given structure) that agreed with the actual age, and n is the total number of fish in the sample.

The agreement rate obtained for each age-group represented the efficiency and accuracy of a given method for estimating the age of fish at a particular life stage, whereas the mean agreement rate for each method was used to compare accuracy among the aging methods. A paired t-test was also used to compare results between the two readers.

RESULTS

Timing of Annulus Formation

Samples collected during four consecutive months (March–June 2009) showed the following pattern: in March, the edges of otoliths were in the white layer, but 40.91% of scales began to form annular rings (Figure 1). In April, the narrow dark layer emerged on the otolith edge, but the annual ring did not form on it, whereas 63.64% of scales showed the beginning of annular deposition at the edge. In May, 37.50% of otoliths and 66.67% of scales had a newly formed annular zone. In June, 97.73% of otoliths and 76.69% of scales had clear annuli. Thus, the annual ring in Northern Snakeheads began to form in April and became fully formed in June.

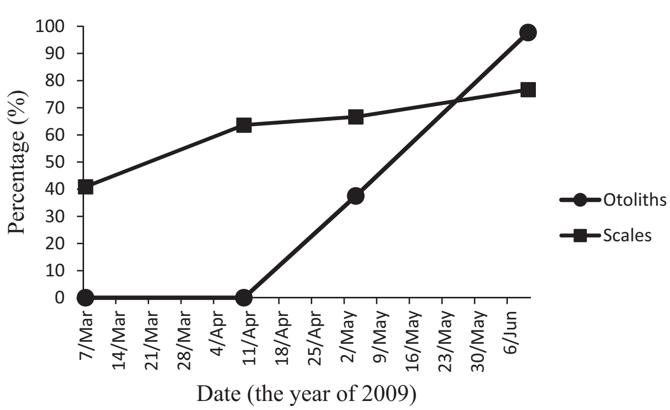

FIGURE 1. Percentage of annual ring formation on otoliths and scales in Northern Snakeheads from March to June.

Characteristics of Annual Rings on Hard Structures

Although there were rings on the pectoral fins, dorsal fins, vertebrae, basioccipital bones, opercular bones, and interopercular bones, most of the annuli were ambiguous and it was difficult to estimate age. In addition, no rings were found on the teeth or cleithra, so those structures were excluded from further analysis.

On otoliths, there were some periodic growth zones, with alternately deposited wide white layers and narrow dark layers, the latter of which were regarded as the growth rings. Although annular rings could be observed in all the three types of otolith, the ring structures on most of the asteriscus and lapillus otoliths were not continuous. Therefore, these otolith types were not considered as aging structures for Northern Snakeheads. Annuli were more pronounced on sagittal otoliths, and in young-age Northern Snakehead samples there were complete rings around the cross section of the sagittal otolith (Figure 2). All age-groups exhibited high clarity of annual rings in the depression of the lateral sagittae. The annuli on scales were also clear, appearing dispersed in the front area of the scale and transitional characteristics in the lateral areas (Figure 3), but the crumbliness was not obvious on scales from younger-age fish.

Evaluation of Results from Different Age Determination Methods

The mean agreement rates between the two readers were 92.17% for otoliths, 85.38% for scales, 63.91% for pectoral fins, and 58.61% for opercular bones (Figure 4). Paired t-tests showed that ages estimated with otoliths, scales, and pectoral fins did not significantly differ between the two readers (P = 0.20, 0.31, 0.21, respectively), whereas the ages determined

FIGURE 2. Annual ring on a sagittal otolith from an age-1 Northern Snakehead. [Figure available in color online.]

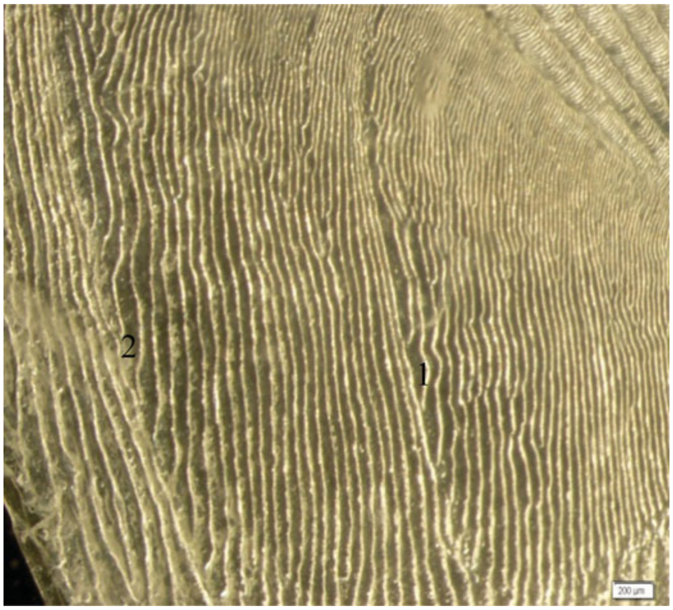

FIGURE 3. Annuli on a scale from an age-2 Northern Snakehead. [Figure available in color online.]

with opercular bones were significantly different between the readers ($P = 0.00061$). Agreement rates between the readers on ages estimated from basioccipital bones, vertebrae, interopercular bones, and dorsal fins were all below 30%, and there were also many subjective errors between readers when annual rings were counted from these structures. The mean CV in age estimates between readers was 2.83% for otoliths, 6.92% for scales, 27.04% for pectoral fin rays, and 23.83% for opercular bones.

Among all of the structures used for age determination, otoliths from age-1 Northern Snakeheads had the highest occurrence rate (100%) of annual rings (i.e., percentage of structures that had rings to count), and the average percent agreement for otoliths was 85.68% (Figure 5). The mean percent agreement for scales was 68.91%. Other structures had relatively clear annual

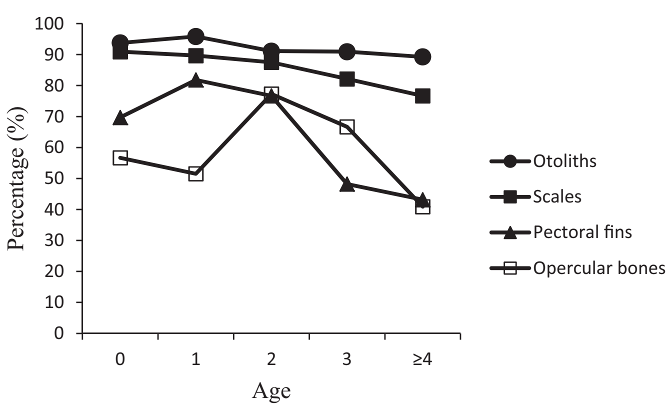

FIGURE 4. Percent agreement (%) in ages estimated by the two readers using different aging structures from Northern Snakeheads.

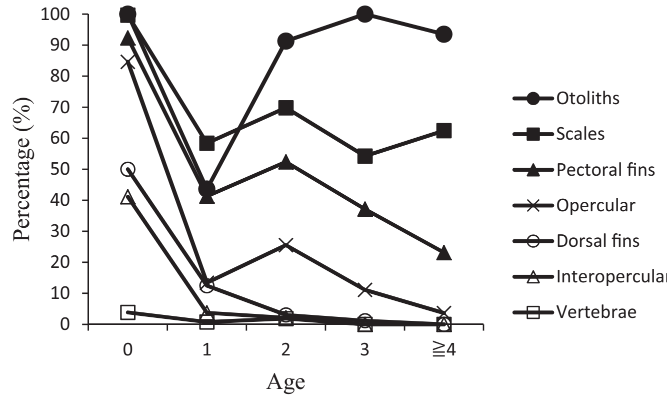

FIGURE 5. Percent agreement (%) between the estimated age (as determined from seven aging structures) and the actual age of Northern Snakeheads.

rings along with too many false rings, hindering the estimation of age. The values of percent agreement from these structures were also low: only 49.25% for pectoral fins, 27.67% for opercular bones, and less than 20% for dorsal fins, interopercular bones, vertebrae, and basioccipital bones.

The percent agreement of annual rings on scales in the five age-groups of Northern Snakeheads was variable: 99.66% for age-0 fish, 58.40% for age-1 fish, 69.82% for age-2 fish, 54.23% for age-3 fish, and 62.44% for age-4+ fish (Figure 6). We also found that scales removed from the base of the pectoral fin had the highest percent agreement and the highest occurrence rate of annual rings (average agreement rate = 74.06%) in all age-groups; the average agreement rates were 68.47% for scales from the middle area and 64.20% for scales from the back area. In addition, the agreement rates between pairs of structures were 94.72% for otoliths and scales, 63.35% for otoliths and pectoral fins, and 60.84% for scales and pectoral fins.

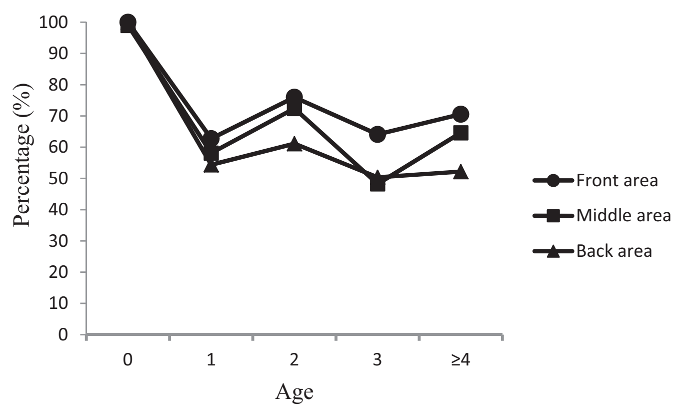

FIGURE 6. Percent agreement (%) in Northern Snakehead ages from annulus counts on scales taken from three locations: the front area (base of the pectoral fin), middle area (above the lateral line but below the first dorsal fin), and back area (above the lateral line but below the last dorsal fin).

DISCUSSION

We found that annulus formation was generally earlier on scales than on otoliths. The annuli on scales were formed from March to June, similar to the results of a previous study of Northern Snakeheads in Poyang Lake, central Yangtze River basin (Yu et al. 2008). We therefore conclude that the annulus formation period in scales is longer than that of otoliths but is relatively scattered. The rings on the otoliths were formed between April and June. The period of annulus formation on otoliths was shorter and centralized. The first annual ring on calcified structures did not form in age-1 Northern Snakeheads until June, so the two readers may have underestimated the age of the specimens that were collected from March to June. We suggest that when using hard structures to estimate the age of Northern Snakeheads, the sampling date must be considered, as it may be better to sample the fish sometime after June to avoid underestimation of fish age to some extent.

Results from the present study showed that the sagittal otolith was the best structure for use in Northern Snakehead age estimation. Buckmeier et al. (2002) also validated the otolith and pectoral spine methods and recommended the use of otoliths for estimating the age of Channel Catfish *Ictalurus punctatus*. Use of sagittal otoliths as aging structures for Northern Snakeheads is preferable not only because of the clear rings, high accuracy, and repeatability but also because the annuli could be distinguished easily (even on the otolith edge) in Northern Snakeheads older than 4 years. Although this method involves killing the fish, the use of sagittal otoliths seems to be the most reliable approach for determination of age in Northern Snakeheads.

Generally, scales can be obtained easily without sacrificing the fish and also can be observed directly, but the use of scales as aging structures has some drawbacks, including the presence of false annuli, difficulties in locating the first annulus, and crowding of annuli along the outer scale margin in older specimens. Since the 1970s, researchers have noted that the use of scales to estimate fish age has limitations (e.g., Jessop 1972; Beamish 1981), and some studies have shown that the scale aging method is not appropriate for certain species (e.g., Schill et al. 2010). In the present study, especially for Northern Snakehead specimens over age 3, the number of annual rings on the scales was less than the actual fish age, as further evidenced by the lower agreement rate for these age-classes. There are several possible reasons for the inaccuracy of scale-based age estimates. First, some of the scales we collected might have been regenerated ones; the potential for obtaining regenerated scales is higher when fish of greater ages are sampled, and the number of rings on such scales would always be limited. Second, as the fish grow, the rings on their scales may become increasingly crowded, possibly causing two rings to be read as one ring or causing a ring to be considered discontinuous by the reader. Our results did suggest that there were many regenerated and secondary scales on Northern Snakeheads, as 23.53% of the scale samples had no rings. Therefore, we should consider these problems when using the scale approach for aging older fish. Our results are similar to those from previous studies emphasizing that the scale aging method is appropriate for younger, but not older, fish (Borkholder and Edwards 2001; Maceina and Sammons 2006; Sylvester and Berry 2006; Gunn et al. 2008).

Previous studies have demonstrated the effectiveness of using other structures in fish age determination (e.g., Logsdon 2007; Quist et al. 2007; Colombo et al. 2010). Very early in the current study, we found that teeth and cleithra did not have any useful rings with which to estimate fish age, so these structures were excluded from further analysis. Although for pectoral fins the total agreement rate between the two readers was 63.91% ($P > 0.05$), the agreement with actual age was only 49.25%, which is relatively low. The use of opercular bones is not recommended, as this method involves killing the fish and has lower accuracy and repeatability than the pectoral fin method. Other structures, such as dorsal fin rays, interopercular bones, basioccipital bones, and vertebrae, had similar defects: too many incomplete or false rings on these structures, a low accuracy rate, poor repeatability, and the need to sacrifice fish to collect the structures. Therefore, we suggest that these other structures should only serve to supplement the use of otoliths and scales in estimating the age of Northern Snakeheads.

ACKNOWLEDGMENTS

The current study was financially supported by grants from the State Key Laboratory of Freshwater Ecology and Technology (2010FB19), the Hunan Department of Education (09A032), and the Research Program of Hunan Province, China (2009NK3095). We appreciate the two journal reviewers who helped us to improve the manuscript.

REFERENCES

Beamish, R. J. 1981. Use of fin-ray sections to age Walleye Pollock, Pacific Cod, and Albacore, and the importance of this method. Transactions of the American Fisheries Society 110:287–299.

Blackwell, B. G., and T. M. Kaufman. 2012. Timing of Yellow Perch otolith annulus formation and relationship between fish and otolith lengths. North American Journal of Fisheries Management 32:239–248.

Borkholder, B. D., and A. J. Edwards. 2001. Comparing the use of dorsal fin spines with scales to back-calculate length-at-age estimates in Walleyes. North American Journal of Fisheries Management 21:935–942.

Boxrucker, J. 1986. A comparison of the otolith and scale methods for aging White Crappies in Oklahoma. North American Journal of Fisheries Management 6:122–125.

Buckmeier, D. L. 2002. Assessment of reader accuracy and recommendations to reduce subjectivity in age estimation. Fisheries 27(11):10–14.

Buckmeier, D. L., E. R. Irwin, R. K. Betsill, and J. A. Prentice. 2002. Validity of otoliths and pectoral spines for estimating ages of Channel Catfish. North American Journal of Fisheries Management 22:934–942.

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., M. C. Annand, and J. I. McMillan. 1995. Graphical and statistical methods for determining the consistency of age determinations. Transactions of the American Fisheries Society 124:131–138.

Chang, W. Y. B. 1982. A statistical method for evaluating the reproducibility of age determination. Canadian Journal of Fisheries and Aquatic Sciences 39:1208–1210.

Colombo, R. E., Q. E. Phelps, C. M. Miller, J. E. Garvey, R. C. Heidinger, and N. S. Richards. 2010. Comparison of Channel Catfish age estimates and resulting population demographics using two common structures. North American Journal of Fisheries Management 30:305–308.

Cunningham, A. 2005. Snaking its way in: feared snakehead fish sets up house in the U.S. Scientific American 292(January 1):16.

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, Maryland.

Gunn, J. S., N. P. Clear, T. I. Carter, A. J. Rees, C. A. Stanley, J. H. Farley, and J. M. Kalish. 2008. Age and growth in Southern Bluefin Tuna, *Thunnus maccoyii* (Castelnau): direct estimation from otoliths, scales and vertebrae. Fisheries Research 92:207–220.

Hoxmeier, R. J. H., D. D. Aday, and D. H. Wahl. 2001. Factors influencing precision of age estimation from scales and otoliths of Bluegills in Illinois reservoirs. North American Journal of Fisheries Management 21:374–380.

Hua, Y. Y., L. J. Shi, H. Y. Li, and X. F. Zhang. 2005. Age discrimination of *Takifugu obscurus* Abe. Acta Hydrobiologica Sinica 29:279–284.

Isermann, D. A., M. H. Wolter, and J. J. Breeggemann. 2010. Estimating Black Crappie age: an assessment of dorsal spines and scales as nonlethal alternatives to otoliths. North American Journal of Fisheries Management 30:1591–1598.

Jessop, B. M. 1972. Aging Round Whitefish (*Prosopium cylindraceum*) of the Leaf River, Ungava, Quebec, by otoliths. Journal of the Fisheries Research Board of Canada 29:452–454.

Kowalewski, L. K., A. P. Maple, M. A. Pegg, and K. L. Pope. 2012. Latitudinal influence on age estimates derived from scales and otoliths for Bluegills. North American Journal of Fisheries Management 32:1175–1179.

Kruse, C. G., C. S. Guy, and D. W. Willis. 1993. Comparison of otolith and scale age characteristics for Black Crappies collected from South Dakota waters. North American Journal of Fisheries Management 13:856–858.

Logsdon, D. E. 2007. Use of unsectioned dorsal spines for estimating Walleye ages. North American Journal of Fisheries Management 27:1112–1118.

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.

Maceina, M. J., and S. M. Sammons. 2006. An evaluation of different structures to age freshwater fish from a northeastern US river. Fisheries Management and Ecology 13:237–242.

Meng, Q. W. 1989. Ichthyology: morphology and classification. Shanghai Science and Technology Press, Shanghai, China.

Polat, N., D. Bostanci, and S. Yılmaz. 2001. Comparable age determination in different bony structures of *Pleuronectes flesus luscus* Pallas, 1811 inhabiting the Black Sea. Turkish Journal of Zoology 25:441–446.

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.

Ribot-Carballal, M. C., F. Galván-Magaña, and C. Quiñónez-Velázquez. 2005. Age and growth of the Shortfin Mako shark, *Isurus oxyrinchus,* from the western coast of Baja California Sur, Mexico. Fisheries Research 76:14–21.

Schill, D. J., E. R. J. M. Mamer, and G. W. LaBar. 2010. Validation of scales and otoliths for estimating age of Redband Trout in high desert streams of Idaho. Environmental Biology of Fishes 89:319–332.

Sylvester, R. M., and C. R. Berry Jr. 2006. Comparison of White Sucker age estimates from scales, pectoral fin rays, and otoliths. North American Journal of Fisheries Management 26:24–31.

Tandon, K. K., and M. S. Johal. 1996. Age and growth in Indian freshwater fishes. Narendra Publishing House, New Delhi.

Wang, X. H., and Y. G. Dai. 2006. Annulus characteristics and age diagnosis of *Tor* (*Folifer*) *brevifilis brevifilis* Peters. Journal of Shanghai Fisheries University 15:247–251.

Welch, T. J., M. J. Van Den Avyle, R. K. Betsill, and E. M. Driebe. 1993. Precision and relative accuracy of Striped Bass age estimates from otoliths, scales, and anal fin rays and spines. North American Journal of Fisheries Management 13:616–620.

Xie, S., and Y. Watanabe. 2005. Hatch date-dependent differences in early growth and development recorded in the otolith microstructure of *Trachurus japonicus*. Journal of Fish Biology 66:1720–1734.

Yu, H. Y., S. Ouyang, X. P. Wu, Y. G. Dai, and S. Y. Zou. 2008. Studies on age and growth of *Ophiocephalus argus* in Poyang Lake. Life Science Research 12:91–94.

Zhang, X. P., S. Y. Gong, J. Liu, X. G. He, and J. H. Gao. 1999. Studies on age and growth of *Channa argus* in the Wanghu Lake. Acta Hydrobiologica Sinica 23:600–603.

Zhu, B. K., and Y. J. Liang. 1999. Studies on age and growth of snakehead fish in Jinsha River reservoir. Reservoir Fisheries 19:36–38.

Zymonas, N. D., and T. E. McMahon. 2009. Comparison of pelvic fin rays, scales and otoliths for estimating age and growth of Bull Trout, *Salvelinus confluentus*. Fisheries Management and Ecology 16:155–164.