Precision of Age Estimates of Wyoming Walleyes from Different Calcified Structures

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ABSTRACT - The ages of 77 walleyes (*Stizostedion vitreum*) from Grayrocks Reservoir, Wyoming, were estimated using otoliths, dorsal spines, and scales, and 48 walleyes from Hawk Springs Reservoir, Wyoming, using pectoral fin rays, pelvic fin rays, and scales. Precision of age determinations among independent readings by a single reader, among three readers, and among structures was low. Otoliths provided the highest precision due to distinctness of their annuli; scales provided the least precision.

Key words: Walleye, *Stizostedion vitreum*, age determination, otoliths, Wyoming

Accurate and precise age determination methods are essential when estimating growth rates, mortality rates, age structure, longevity, and production of fish (Beamish and McFarlane 1983, Babaluk and Campbell 1987). Serious consequences can result from errors in age determinations. For example, Beamish and McFarlane (1983) pointed out that fish stocks can be overexploited when ages are underestimated. Therefore, it is advisable to evaluate age determination methods for both accuracy and precision before management decisions are made (Beamish and McFarlane 1983).

Precision is a measure of the reproducibility of age estimates, while accuracy is the proximity of age estimates to the actual values (Summerfelt and Hall 1987). Accuracy of age estimates can only be validated by the use of known-age fish or mark-recapture studies (Beamish and McFarlane 1983). We agree with the obvious importance of age validation and accuracy stressed by Beamish and McFarlane (1983, 1987), but without a high degree of aging precision, accuracy will suffer.

Many calcified structures have been used to estimate the ages of walleyes (*Stizostedion vitreum*), including scales, otoliths, dorsal spines, pelvic and pectoral fin rays, vertebrae, and opercula (Mackay et al. 1990). Only a few studies have addressed the precision of aging walleyes using these structures (Carlander 1961, Campbell and Babaluk 1979, Erickson 1983), and these studies compared the percent of determinations that were in agreement within a certain number of years. We evaluated precision of aging walleyes with otoliths, dorsal spines, pectoral and pelvic fin rays, and scales using a method that provides a better estimate of precision.

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Slow-growing walleye stocks can present a high degree of difficulty, and thus less precision, in age estimations (Campbell and Babaluk 1979, Erickson 1979, 1983). Most walleye stocks in Wyoming exhibit slow growth rates, due to high elevations and low exploitation rates (Marwitz 1994). We assessed precision of age determinations of Wyoming walleyes among readings by one reader and among readings by three different readers, as well as the precision of age estimates among different structures.

METHODS

We collected walleyes from Grayrocks Reservoir, Platte County, Wyoming, using gill nets during September 1992. The three anterior dorsal spines were removed from each fish at the point of insertion, allowed to air dry, and placed in individual envelopes. We extracted sagittal otoliths and removed 5-10 scales from below the lateral line posterior to the distal end of the pectoral fin and placed them in envelopes.

We collected walleyes from Hawk Springs Reservoir, Goshen County, Wyoming, using trap nets in April 1993. The first three fin rays from both a pectoral and a pelvic fin of each fish were removed at the point of insertion, allowed to air dry, and placed in envelopes. Scales were collected and stored as described above.

The second dorsal spine and the first pectoral and pelvic fin rays were removed from their associated tissues using a scalpel, scraped clean of dried tissue, embedded in epoxy, and allowed to dry for 12 hr. We ground the proximal end of each embedded spine or fin ray flat, using a grinding wheel, and polished each smooth with 600-grit silicon carbide sandpaper. A 0.5-1.0-mm section was cut from the proximal end and affixed with mounting medium (polished side down) on a glass microscope slide. When the mounting medium had hardened for 24 hr, we ground and polished the exposed surface of the section using 360- and 600-grit sandpaper.

Otoliths were sectioned similar to the method described by Maceina (1988). We ground otoliths from the posterior edge to the nucleus, using 360-grit silicon carbide sandpaper glued to a grinding wheel, to produced a flat surface in the transverse plane. The flat surface was polished using 600-grit sandpaper and mounted with epoxy on a glass microscope slide. After 12 hr of hardening, we ground and polished the anterior edge parallel to the surface of the slide to produce a section about 0.5 mm thick.

Scales were soaked in warm, soapy water to remove mucus and dirt. Two or three scales from each fish were mounted (sculptured side up) between a glass microscope slide and a cover slip held together with masking tape.

We photographed prepared sections of otoliths, dorsal spines, pectoral fin rays, and pelvic fin rays using photomicroscopy equipment (31x), transmitted light, and 35-mm film (200 ASA). Scales were magnified (25x) and photocopied with a microfiche copier. Prints were numbered randomly and a record kept to identify each.

The principal reader (R1) made three independent age determinations (reader did not know results from previous readings) for each structure to determine the precision among readings by one reader. Two additional readers (R2 and R3) independently read each print once to determine variability among readers. All readers were experienced in annulus identification and followed the criteria set by Jearld (1983) and Mackay et al. (1990).

For comparisons among structures, we derived final age estimates for each structure using the first reading by R1 and the readings of R2 and R3. When at least two readers agreed, we accepted this value as the correct age. The median age was used when no two readers agreed.

The index of average percent error (APE) of Beamish and Fournier (1981) was computed to provide estimates of the repeatability of age determinations by a single reader, by different readers, and when using different structures. We used the three age determinations by R1 (single reader), the first reading by R1 and the readings of R2 and R3 (different readers), and final age estimates (different structures). Greater precision was achieved as APE was minimized.

We used Chi-square analysis to determine if significant differences in age frequencies occurred among readings (R1) and among readers (R1, R2, and R3), and to determine if significant differences in the ratio of agreements to disagreements occurred among structures. Computations were made using STATISTIX 3.1 (Analytical Software 1990) and significance determined at $P \leq 0.05$.

RESULTS

Otoliths, Dorsal Spines, and Scales (Grayrocks Reservoir)

A total of 77 walleyes were collected from Grayrocks Reservoir. Dorsal spines and scales were obtained from all fish and otoliths from all but two.

Agreement of age determinations among three independent readings by R1 (all three readings agreed) ranged from 12% with scales to 80% with otoliths (Table 1). The APE resulting from these readings ranged from 2% for otoliths to 12% for scales, indicating that precision among readings was greatest for otoliths and poorest for scales. Age-frequency distributions for the three independent readings were significantly different only for scales (Chi-square=35.02, df=16, P=0.0039).

Mean agreement of age determinations between two independent readings by R1 was 86% for otoliths, 62% for dorsal spines, and 33% for scales. Differences in agreement between readings were small for otoliths (range=84-89%) and dorsal spines (range=60-64%), but relatively large for scales (range=21-40%, Table 1).

Agreement of age determinations among three independent readers (all three readers agreed) ranged from 14% with scales to 23% with otoliths (Table 2). The APE ranged from 10% for dorsal spines to 12% for scales. The only significant difference in age-frequency distributions among readers occurred with scales (Chi-square=37.21, df=16, P=0.0020).

Table 1. Results of three independent age determinations by one reader (R1) using different structures from walleyes collected from Grayrocks and Hawk Springs reservoirs, Wyoming. (1, 2, and 3 represent the first, second, and third readings respectively; n is the number of fish from which structures were read; and APE is the average percent error).

		Percent agreement				
Structure	n	1,2,&3	1&2	1&3	2&3	APE
			Grayrocks Reservoir			
Otoliths	75	80	89	86	84	2
Dorsal spines	77	47	60	62	64	7
Scales	77	12	21	38	40	12
			Hawk Springs Reservoir			
Pectoral fin rays 47	32	55	47	51	7	
Pelvic fin rays	46	17	28	35	61	10
Scales	45	11	40	29	31	9

Mean agreement of age determinations between two independent readers was 44% for otoliths, 42% for dorsal spines, and 37% for scales. Differences in agreement between readers were large for otoliths (range=32-55%), dorsal spines (range=35-56%), and scales (range=25-43%, Table 2).

Agreement between age estimates was highest for comparisons between otoliths and scales (57%). Agreement was 33% between otoliths and dorsal spines, and 32% between dorsal spines and scales. The APE ranged from 6% between

Table 2. Results of independent age determinations by three readers using different structures from walleye collected from Grayrocks and Hawk Springs reservoirs, Wyoming. (R1, R2, and R3 represent readers 1, 2, and 3, respectively; n is the number of fish from which structures were read; and APE is the average percent error).

		Percent agreement				
Structure	n	R1,R2,&R3	R1&R2	R1&R3	R2&R3	APE
			Grayrocks Reservoir			
Otoliths	75	23	44	55	32	11
Dorsal spines	77	21	35	56	36	10
Scales	77	14	43	25	42	12
			Hawk Springs Reservoir			
Pectoral fin rays 47	15	47	34	30	9	
Pelvic fin rays	46	22	56	41	30	7
Scales	45	9	31	18	22	13

otoliths and scales to 11% between dorsal spines and scales and between dorsal spines and otoliths. A 2x3 Chi-square contingency test of the ratio of agreements to disagreements in age estimates among the three comparisons yielded a significant difference (Chi-square=16.61, df=2, P=0.0002). When the same test was performed without the comparison of otoliths and scales, no significant difference was observed.

Differences in age estimates among structures were evident (Fig. 1). When otoliths were compared to dorsal spines, otoliths yielded lower ages for fish up to four years and generally higher ages for older fish (Fig. 1a). Ages estimated from scales were generally less than ages estimated from otoliths and dorsal spines (Fig. 1b, 1c).

Figure 1. Comparisons of walleye age estimates (years) between otoliths, dorsal spines, and scales from Grayrocks Reservoir, Wyoming. Points are individual fish except where indicated by a number. Points fall on diagonal line when age estimates were equal.

Fin Rays and Scales (Hawk Springs Reservoir)

Forty-eight walleyes were collected from Hawk Springs Reservoir. From these, 47 pectoral fin rays, 46 pelvic fin rays, and 45 samples of scales were obtained.

Agreement among three independent age determinations by R1 ranged from 11% with scales to 32% with pectoral fin rays (Table 1). The APE ranged from 7% for pectoral fin rays (highest degree of precision) to 10% for pelvic fin rays. However, comparisons of age-frequency distributions, utilizing Chi-square contingency tables, yielded no significant differences among the three readings for any structure.

Mean agreement of age determinations between two independent readings by R1 was 51% for pectoral fin rays, 41% for pelvic fin rays, and 33% for scales. Differences in agreement between readings were large for pelvic fin rays (range=28-61%), but only moderate for pectoral fin rays (range=47-55%) and scales (range=29-40%, Table 1).

Agreement of age determinations among independent readings by three readers ranged from 9% with scales to 22% with pelvic fin rays (Table 2). The APE ranged from 7% for pelvic fin rays to 13% for scales. The only significant difference among independent readers was observed when age-frequency distributions for scales were compared (Chi-square=44.71, df=14, $P<0.0001$). Pelvic fin rays produced the highest degree of precision among readers and scales produced the lowest.

Mean agreement of age determinations between two independent readers was 42% for pelvic fin rays, 37% for pectoral fin rays, and 24% for scales. Differences in agreement between readers were large for pelvic fin rays (range=30-56%), pectoral fin rays (range=30-47%), and scales (range=18-31%, Table 2).

Agreement between age estimates was highest for comparisons between the pelvic and pectoral fin rays (33%). Agreement was 30% between pelvic fin rays and scales and 18% between pectoral fin rays and scales. The APE ranged from 7% between pelvic fin rays and scales, and between pelvic fin rays and pectoral fin rays, to 9% for pectoral fin rays and scales. A 2x3 Chi-square contingency test of the ratio of agreements to disagreements in age estimates yielded no significant difference among the three comparisons.

Differences in age estimates among the three structures from Hawk Springs walleyes were less clearly defined (Fig. 2), probably due to the smaller sample size and narrower range of ages. Pectoral fin rays had a tendency to yield older ages for older fish and younger ages for younger fish than pelvic fin rays and scales (Fig. 2a, 2b). Pelvic fin rays also tended to yield older ages than scales for fish of older age groups (Fig. 2c).

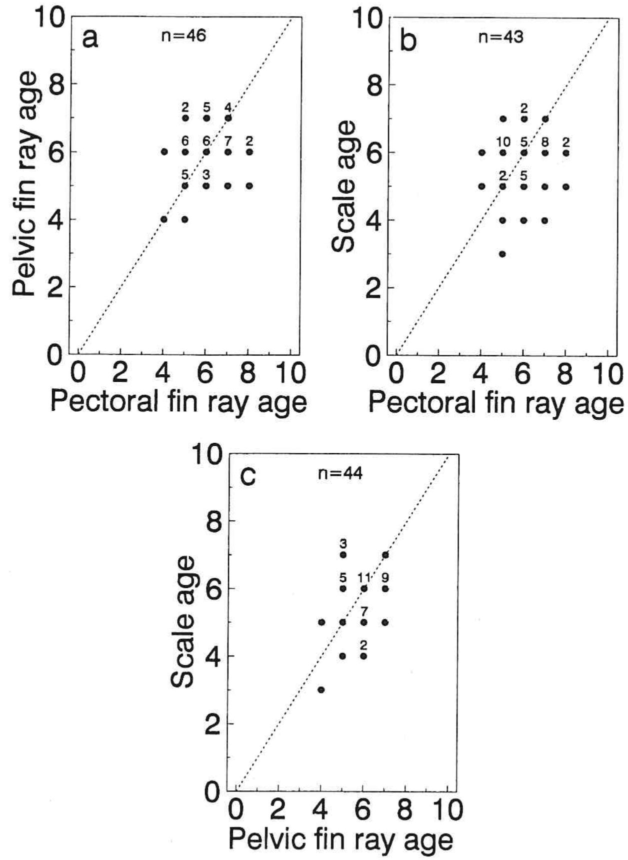

Figure 2. Comparisons of walleye age estimates (years) between pectoral fin rays, pelvic fin rays, and scales from Hawk Springs Reservoir, Wyoming. Points are individual fish except where indicated by a number. Points fall on diagonal line when age estimates were equal.

DISCUSSION

Variability in age estimates of individual fish was common. Of the three aging structures used from Grayrocks Reservoir, otoliths yielded the highest level of precision. This finding is similar to Campbell and Babaluk (1979) and Erickson (1983). Otolith extraction, preparation, and sectioning was time-consuming, but annuli were distinct. Some dorsal spine sections seemed to have false annuli present, identifiable by their faintness and close proximity to distinct annuli (Mackay et al. 1990). Scales exhibited crowding of annuli near the periphery and indistinct annuli throughout. We found scales to be unreliable aging structures, as indicated by the low percent agreement and high APE among readers and readings.

When fin rays of walleyes from Hawk Springs Reservoir were examined, we found that the first annulus was often difficult to identify (Beamish 1973), contributing to the variability in age determinations. Pelvic fin rays, although yielding the highest percent agreement and lowest APE among three independent readers, yielded a lower percent agreement and higher APE among three readings by a single reader than pectoral fin rays.

Belanger and Hogler (1982) pointed out that many factors contribute to the decision of which aging structure to use. The extraction of otoliths, because it necessitates sacrificing fish, may not be acceptable for some recreational fisheries. In this situation, dorsal spines may be preferable over scales or fin rays due to the former's higher precision among independent readers and readings by one reader. Additionally, only a subsample of fish may need to be sacrificed to obtain otoliths as a check to verify ages obtained with dorsal spines.

High precision in age determination was not obtained for any structure studied. Scales yielded the least precision and should be avoided as a primary aging structure for walleyes. Otoliths had distinct annuli and provided the highest level of precision.

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