OTOLITH AGING OF LARVAL AND JUVENILE STRIPED BASS IN CALIFORNIA

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Stephen F. Foss and Lee W. Miller
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Technical Report 51
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Abstract

Sagittal otoliths from two age groups of known-age striped bass, *Morone saxatilis*, were examined to determine if growth increments form on a daily basis. For striped bass under 35 days after hatch, increment counts were consistent with daily formation. Our study found that otolith increment formation began at hatch, which conflicts with a previous finding that initial increment formation occurred about 2 days after hatch. Average age estimates by three different readers of fish under 35 DAH included some large differences in individual estimates. The increment count/age relationship was highly variable for older fish, 53 to 87 DAH, and reflected the difficulty in interpretation of otoliths as structural complexity increased with age.

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Introduction

Otoliths have been used widely in age determination of larval fish (Jones 1986). Brothers *et al* (1976) first applied the otolith aging technique to young field-captured striped bass, *Morone saxatilis*, and found that increment-estimated ages corresponded with known spawning season and growth rate information. As a method of aging individual larval and juvenile fish, the otolith increment technique yields estimates of age-specific growth and mortality that would facilitate assessment of how environment affects young striped bass survival.

A fundamental assumption of otolith-based aging of young fish is that increments are formed daily. An otolith increment, as seen under a light microscope, consists of adjacent bands of light and dark material, termed incremental and discontinuous zones, respectively (Campana and Neilson 1985). Jones and Brothers(1987) found that increments were deposited daily for striped bass reared under optimal conditions. However, otoliths of older fish were difficult to read, and the ages of fish older than 60 days after hatch were underestimated. Secor and Dean (1989) validated daily increment formation in striped bass of ages 10 to 51 days after hatch.

Determination of age at first increment deposition is required to correctly assign ages to field-caught fish. Houde and Morin (1990) found that temperature could influence the timing of initial deposition; as temperature increased, the day of the first increment occurred earlier. Secor and Dean(1989) observed first increments at 4-5 DAH for pond-reared striped bass, but they did not report temperatures.

The goals of our study were to:

- Validate daily increment deposition in otoliths of California larval and juvenile striped bass,
- Evaluate age at first increment formation in California striped bass, and
- Evaluate the accuracy and precision of our laboratory's age estimates for subsequent application to studies of field-caught fish.

Methods

In 1983, we used striped bass from the Department of Fish and Game's Central Valley Hatchery to validate daily increment deposition. A cohort of hatchery-reared striped bass was held at 20°C and initially sampled from aquaria 2 DAH. The remaining fish were transferred to hatchery ponds, and individuals were collected at varying intervals by dip-net or seine up to 34 DAH. In 1991, older known-age hatchery-reared fish were evaluated for daily otolith increment deposition by sampling pond-stocked individuals of ages 53-87 DAH.

Saggital otoliths, the largest and most easily readable of the three pairs of otoliths, were removed and cleaned. In 1983, all otoliths were mounted permanently using *Eukitt*, whereas in 1991 otoliths from fish <10.5mm were mounted permanently in *Cytoseal*. Otoliths from fish >10.5mm were embedded in *Epo-Mix* epoxide resin, sectioned, sanded with 400- and 600-grit sandpaper, and polished with alumina powder. Otoliths were examined using either a Zeiss or an Olympus light microscope at 400x or 1000x magnification. The Olympus, used for 1991 samples, was fitted with a video camera, monitor, and Macintosh computer with *Bony Parts* image analysis system to aid increment counting through image enhancement.

To validate daily increment deposition, we compared multiple independent readings by the same reader of known-age fish. The reader had no prior experience reading otoliths, had no knowledge of larval fish age, and received no feedback about aging results between readings. We used two groups of readings to compare readings of otoliths collected in 1983: a group of 31 otoliths, which was read three times, and a group of 199 otoliths that included the smaller group and for which only two readings were compared. Two readings were compared for older fish collected in 1991. Otoliths for which two counts varied more than 50% from the third count were judged unreadable and were excluded. We also evaluated between-reader variation, using three different readers' age estimates from a subsample of 22 otoliths of fish up to 35 DAH.

Linear regressions of increment count on days after hatch were computed (SAS 1991). Student's *t* was used to test whether growth increments were not formed daily (regression slope 1.0) and whether increment formation was not initiated at hatching (regression intercept 0). Differences were tested for significance at the 5% level.

Results

For the smaller group of otoliths from 2-34 DAH fish, the first reading tended to underage fish, the second reading tended to overage fish, and the third aged fish correctly, on average (Figure 1). The regression slopes for all three readings did not differ significantly from 1.0, and the intercepts for all three readings did not differ significantly from zero (Group 1, Table 1). For the larger group, the regression slope for the first reading was significantly less than 1.0 and, on average, fish were underaged (Figure 2A). The regression slope from the second reading did not differ significantly from 1.0 (Group 2, Table 1) and, on average, fish were aged correctly (Figure 2B).

For older fish aged in 1991, the readings, on average, resulted in over-aging fish 53 DAH by 6.6 days for reading #1 and 9.6 days for reading #2 (Figure 3). The first reading continued to overestimate age, on average, to the oldest fish in the sample. The mean overestimate was 6.3 days at 87 DAH. Reading #2 tended to underestimate the age of older fish, with a mean underestimate of 3.6 days at 87 DAH. Regression of increment counts on known age for the first reading resulted in a regression slope not significantly different from 1.0 (Table 2). The regression slope for the second reading was significantly less than 1.0.

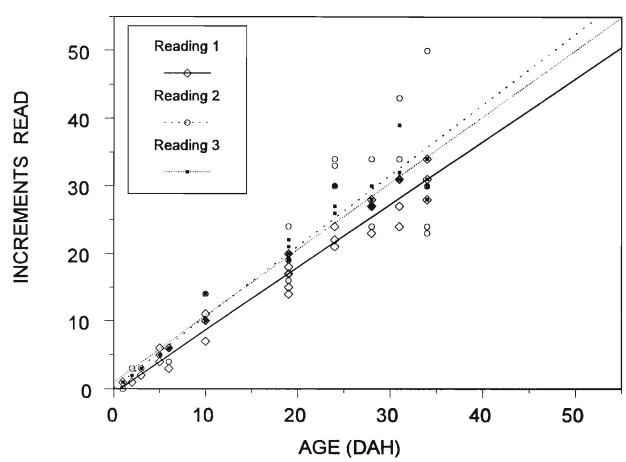

Figure 1
REGRESSIONS OF
THREE OTOLITH INCREMENT COUNTS ON
KNOWN AGE FOR 1983 STRIPED BASS

Age in Days After Hatch, N=31

Variability in age estimates increased with age. For the young fish, 34 DAH or less, the percentage of age estimates within 1 day and 3 days of the known age decreased as age increased, particularly for fish older than 10 DAH (Table 3). However, the percentage of age estimates within 3 days of the known age remained greater than 70%, except for the first reading of 19 and 31 DAH fish and the second reading of 34 DAH fish.

Table 1
SIMPLE LINEAR REGRESSION PARAMETERS OF INCREMENT COUNTS ON
DAYS AFTER HATCH OF 2- TO 34-DAY-OLD STRIPED BASS

Increment count = a + b(DAH). Ho: a=0, b= 1.0. NS = No significant difference at α=0.05. *** p<0.001.

Group	Reading	N	Slope	SE	t-test	Intercept	SE	t-test	r^2
1	First	31	0.93	0.04	NS	–0.63	0.79	NS	0.96
	Second	31	1.05	0.10	NS	0.13	2.12	NS	0.80
	Third	31	0.98	0.04	NS	0.92	0.97	NS	0.94
2	First	199	0.96	0.01	***	–0.21	0.12	NS	0.99
	Second	199	1.02	0.02	NS	–0.13	0.23	NS	0.96

Percentages within 3 days of known age for older fish were much lower than those for younger fish. For fish older than 53 DAH, the percentage of age estimates within 3 days of known age never exceeded 20% for the first reading, but ranged from 30 to 60% for the second reading.

To evaluate between-reader precision, age estimates of three different readers were compared. All readers had at least one reading with 5 or more days error (Table 4). However, all readers tended to underage fish (Figure 4), and for 16 of the 22 otoliths aged (73%), estimates between readers differed by 3 days or less.

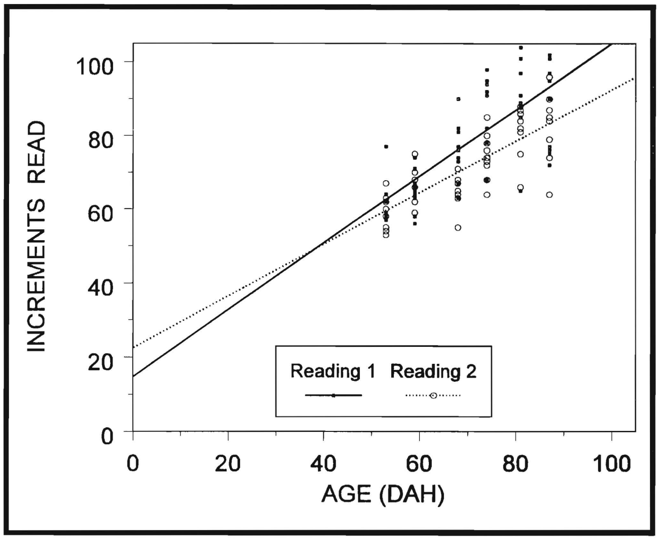

Figure 3
REGRESSIONS OF OTOLITH INCREMENT COUNTS ON KNOWN AGE FOR 1991 STRIPED BASS

Age in Days After Hatch, N=58

Table 2
SIMPLE LINEAR REGRESSION PARAMETERS OF INCREMENT COUNTS ON DAYS AFTER HATCH OF 53- TO 87-DAY-OLD STRIPED BASS

Increment count = a + b(DAH). H_0: b= 1.0.
NS = No significant difference at α=0.05. *** p<0.001.

	First Reading	Second Reading
N	58	58
Slope	0.90	0.70
SE	0.11	0.08
t-test	NS	***
r^2	0.55	0.60

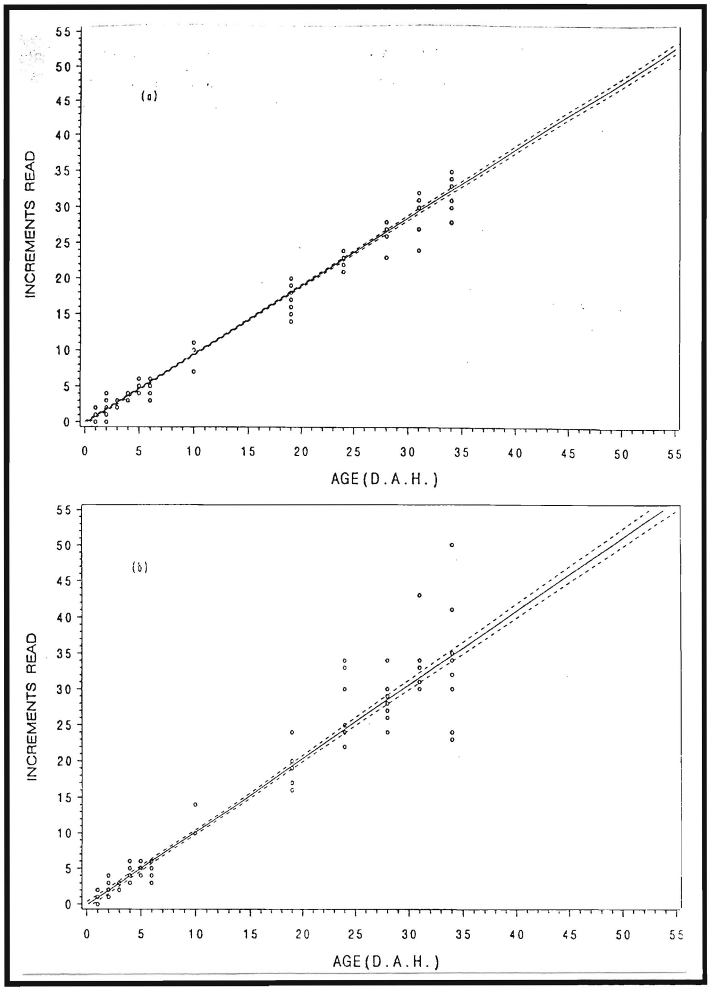

Figure 2
REGRESSIONS OF OTOLITH INCREMENT COUNTS ON KNOWN AGE AND 95% CONFIDENCE LIMITS FOR FIRST AND SECOND READINGS OF 1983 STRIPED BASS

(a) READING 1 (b) READING 2
N=199

Table 3
MEAN AGE ESTIMATES AND
PERCENTAGE OF AGE ESTIMATES WITHIN 1 DAY AND 3 DAYS OF
KNOWN AGE FOR STRIPED BASS

		Reading 1			Reading 2		
Age	N	Mean Estimate	% Within 1 Day	% Within 3 Days	Mean Estimate	% Within 1 Day	% Within 3 Days
1	20	1.2	100	100	1.4	100	100
2	39	1.9	95	100	2.1	97	100
3	17	2.6	100	100	2.9	100	100
4	16	3.7	100	100	4.5	88	100
5	19	4.6	100	100	4.7	100	100
6	23	5.1	74	100	5.0	78	100
10	6	9.7	83	100	10.7	83	83
19	15	15.8	53	67	19.0	73	93
24	13	23.2	69	100	25.2	69	77
28	11	27.9	82	91	27.6	55	73
31	9	29.2	67	67	32.9	56	89
34	11	30.0	64	82	33.7	45	55
53	8	62.3	0	0	59.0	25	38
59	10	65.1	10	20	66.1	20	30
68	10	75.6	10	10	65.9	20	60
74	10	88.7	0	0	75.5	20	40
81	10	90.0	0	0	81.7	30	40
87	10	88.0	0	10	82.0	20	50

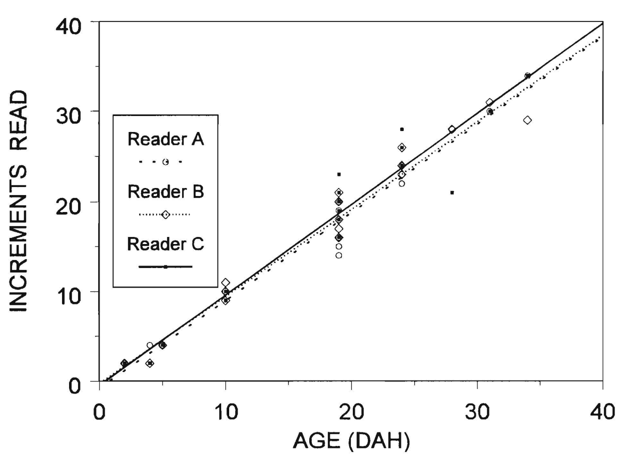

Figure 4
REGRESSIONS OF OTOLITH INCREMENT COUNTS
BY THREE READERS ON KNOWN AGE FOR
1983 STRIPED BASS

Age in days after hatch, N=22.

Table 4
DIFFERENCES OF
INCREMENT COUNTS FROM
KNOWN AGE OF STRIPED BASS
FOR
THREE READERS

(N=22)

	Reader		
Age	A	B	C
2	0	0	0
4	0	2	-2
5	-1	-1	-1
10	0	0	0
10	0	1	-1
10	0	-1	-1
19	-4	-2	-3
19	-3	-1	0
19	-1	2	-1
19	1	-1	-1
19	-5	-2	-1
19	-1	1	1
19	-4	-2	4
19	0	-2	2
19	-4	-3	-1
24	-1	2	0
24	-2	-1	4
24	0	0	2
24	0	-1	0
28	0	0	-7
31	-1	0	-1
34	0	-5	0
Total Days Under-Read	-27	-24	-20
Total Days Over-Read	1	6	13
Total Days Deviation	28	30	33
Total Correct	10	5	6
Percent Correct	45%	23%	27%

Discussion

Our study has attempted to validate daily increment deposition in two age groups of striped bass: 2-34 DAH and 53-87 DAH. For younger fish, growth increment counts were consistent with daily deposition, confirming previous results (Jones and Brothers 1987; Secor and Dean 1989). The first reading for the younger group (n=199) did depart from the expected relationship; however, it is our supposition that the second reading should be more accurate than the first due to the accumulation of knowledge and skill by the reader. Although individual readings deviated from actual ages by as much as 16 days, average readings based on the regressions (Figure 2) were reasonably accurate up to 34 DAH. The effects of randomly distributed errors in otolith aging may be minimized by grouping fish into age classes or cohorts, instead of assigning a specific age to individual fish. However, the greater variability with increased age indicates that caution should be used when applying otolith-estimated ages to striped bass. Although individual age estimates between readers exhibited some large differences (Table 4), average estimates agreed (Figure 3), indicating high statistical precision in aging young striped bass in this age range.

Age estimation was more difficult for older fish because complexity of otolith structure increased with otolith size. In larger otoliths, growth axis shifts result from development of peripheral foci for subsequent increment formation. Growth around these accessory primordia (Campana and Neilson 1985) results in increments of varied width and appear as overlapping rings, making selection of continuous counting transects difficult (Jones and Brothers 1987). High variability in increment counts, over the range of ages exceeding 52 DAH, reflects low aging precision. Thus, the usefulness of otolith aging for striped bass older than about 50 days, is limited. Since we sampled no fish between ages 35-52 DAH, we do not have a clear picture of how precise aging would be for this interval. Since variability increased with age over the range 1-34 DAH, we would expect less precision than for the younger ages.

Larvae 1 and 2 DAH had, on average, otolith increments consistent with increment formation beginning at hatch (Figure 2), which is inconsistent with Houde and Morin's (1990) finding that hatching and rearing temperatures of 18-20°C delayed initial increment formation by about 2 days. We cannot explain this discrepancy. Temperature does not appear to be the cause, since our fish were initially reared at 20°C. Increment formation at hatch did not occur in Houde and Morin's study, conducted over 12-21°C.

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